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B Marambe, UR Sangakkara, WAJM De Costa, ASK Abeysekara

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Message from the President
Asian Pacific Weed Science Society (2005-2007)

Welcome to Sri Lanka!, the pearl of the Indian Ocean, where the 21st conference of the Asian Pacific Weed Science Society (APWSS) is held. The APWSS, at its 19th conference held in Philippines in 2003, selected Sri Lanka to be the host country for the 21st Conference. We are proud to be the host nation.

The 21st APWSS Conference is held at a time where the APWSS celebrates its 40th Anniversary. Established in 1967, the society has made a significant contribution to the development of the field of weed science, providing a forum for the weed scientists, not only from the region but also from various parts of the world, to discuss major issues of current importance, identify challenges to be met, and propose remedies based on the research findings and experiences. This year's conference comprises of 30 Technical sessions where more than 135 research papers are presented. Commemorating the 40 years of service to the field of weed science, the APWSS will also launch a special publication giving the history of the society. Hence, this will be a landmark event of the conferences organized by the APWSS.

The papers presented at the conference were not reviewed, but edited and formatted to improve the clarity. Hence, the responsibility of the scientific content lies with the authors. All in all, the papers submitted are of high quality and have addressed the major issues in the field of weed science.

The Faculty of Agriculture of the University of Peradeniya, Sri Lanka, in collaboration with the National Plant Protection Committee of the Council for Agricultural Research Policy (CARP) of Sri Lanka, helped the APWSS in organizing the 21st Conference. On behalf of the organizing committee, I thank all the individuals and many other organizations who contributed significantly, to make this event a reality. The international membership of the society has always being helpful in providing necessary guidance to ensure the organization of the conference is done maintaining its scientific quality and standards. At this juncture, it is my duty to extend my heartfelt gratitude to all the members of the Organizing Committee of the 21st APWSS Conference for their dedication and active participation in the related activities, and also to the paper presenters whose untiring efforts have placed this conference at a higher level on scientific merit.

I am optimistic that the 21st APWSS Conference, which will be held from 2-6 October 2007, in Colombo, Sri Lanka, will be a success, and be a memorable event for the participants both in terms of scientific quality and Sri Lankan hospitality. I wish all participants a pleasant stay in Sri Lanka, and every success in their endeavours.

Professor Buddhi Marambe
President/APWSS 2005-2007, & Chairman/National Plant Protection Committee of the Council for Agricultural Research Policy (CARP) of Sri Lanka

2nd October 2007

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EFFECTS OF THE SEED ESTABLISHMENT METHOD OF RICE (*Oryza sativa* L.) ON WEED GROWTH AND YIELD OF RICE

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Abstract: A field experiment was conducted at the Rice Research and Development Institute, Batalagoda, Sri Lanka, during he major cultivating season in 2005/06 to determine the influence of seed establishment method on the weed growth, and yield of rice. Three seed establishment methods, namely (a) 24 hrs of soaking followed by 48 hrs of incubation and broadcasting of pre-germinated seeds on drained puddle soil (standard method), (b) 48 hrs soaking followed by 24 hrs incubation and broadcasting of pre-germinated seeds on 3-5 cm flooded soil (water-seeding), and (c) 24 hrs of soaking followed by 48 hrs of incubation and broadcasting of pre-germinated seeds in rows using a plastic drum seeder in drained puddle soil (row-seeding), were tested under weeded (herbicide-treated) and un-weeded situations in a Randomized Complete Block Design. The un-weeded plots recorded a higher weed count and lower grain yields, while that of in row-seeded plots (treatment c) were significantly higher than the standard and water-seeded plots. The lowest weed count and dry weights were observed in water-seeded plots (treatment b). In water-seeded plots, the emergence and growth of grass weeds namely, *Echinochloa crus-galli* and *Leptochloa chinensis* and dominant sedges were delayed when compared to that of *Ischaemum rugosum*. *Monochoria vaginalis* was the dominant weed in the water-seeded plots. The grain yield was significantly higher (5.67 t/ha) in row-seeded plots than the rest. Row-seeding with effective weed management was found to be the best seed establishment method among those tested, to obtain higher rice yields. Water-seeding gave superior results under water-logged situations.

Key words: Broadcast seeding, row seeding, rice, water seeding, weeds

Introduction

A multitude of social cultural and economic factors have influenced more than 90% of Sri Lankan farmers to resort to direct sowing of pre-germinated seeds, manually, on moisture-saturated puddled soil (Weerakoon *et al.* 2000). As a result of rapid expansion of broadcast sowing, farmers face serious issues in maintaining good crop stands with effective weed management. Cultural practices including tillage, fertilizer management, and seed establishment method are some of the important operations that determine the intensity of weed infestation in rice fields (Abeysekara, 1999). Herbicide application has become the predominantly adopted cost effective option among 90% farmers (Marambe and Amarasinghe, 2002). In Sri Lanka, efforts are being made to identify and improve crop establishment methods, which would facilitate effective weed management in rice fields. A sound knowledge on biology, ecology and competition of weeds under different crop establishment methods are essential to determine the best method that farmers can follow. Therefore, this study was conducted to determine the effect of three seed establishment methods on weed growth and yield of rice.

Materials and Methods

Experiment was conducted at the Rice Research and Development Institute, Sri Lanka. The soil of the experimental site was a low Humic glay, sandy loam in texture with a pH 5.8, organic matter 1.6%, total N 0.15%, Olsen P 3.8 ppm, and exchangeable K 0.06 meq/100g. Three different seed establishment methods were compared with weeded (herbicide-treated) and un-weeded situations namely, (a) the standard method – seeds soaked in water for 24 hrs followed by 48 hrs of incubation, and broadcasting of pre-germinated seeds on well drained puddle soil, (b) the water-seeding – seeds soaked in water for 48 hrs followed by 24 hrs of incubation, and broadcasting of pre-germinated seeds on 3-5 cm flooded soil and (c) the row-seeding – seeds soaked in water for 24 hrs followed by 48 hrs incubation, and seeding pre-germinated seeds in a row using a plastic drum seeder on drained puddle soil. In the weeded treatments, bispyribac sodium (Nominee® 10 SC) was applied at 100 ml/ha at 7 days after sowing (DAS). These six treatments were replicated 4 times in a Randomized Complete Block Design. The plot size was 6 x 3 m^2. Each plot had an independent inlet and outlet, for irrigation and drainage. The experiment was conducted in 2005/06 major season using rice (*Oryza sativa*) Bg 357, a 105-day old rice variety. All the other cultural practices were carried out according to the recommendation of the Department of Agriculture (2000). The weed population densities (grasses, broadleaves and sedges) and dry weight were recorded at 42 (DAS). Dominant weed species were counted in each treatment. Rice yield and yield components were recorded at maturity. The treatment means were compared using the Duncan's New Multiple Range Test (p = 0.05).

Results and Discussion

In the experimental plots, *Echinochloa crus-galli*, *Leptochloa chinensis*, *Isachne globosa*, and *Paspalum distichum* were the dominant grass weeds, *Cyperus iria, C. difformis* and *Fimbristylis miliacea* were the dominant sedges and *Monocharia vaginalis, Murdannia nudiflora*, and *Ludwigia perennis* were the major broad leafy weeds (Table 1).

Table 1. Population density (number/m²) of dominant weed flora at 42 DAS in un-weeded plots established under different seeding methods.

Treatments	Grasses				Broadleaves			Sedges		
	*E.c**	*L.ch*	*I.ru*	*P.d*	*M.v*	*M.n*	*L.p*	*C.i*	*C.d*	*F.m*
Standard-seeding & un-weeded	13.8	14.2	8.4	6.5	3.5	7.8	1.5	4.5	3.7	6.4
Water-seeding & un-weeded	6.3	7.8	6.3	3.2	12.3	6.2	2.6	3.6	3.4	4.1
Row-seeding & un-weeded	11.1	19.3	7.5	7.4	6.4	3.4	3.2	4.3	3.8	4.10

**E.c=E. crus-galli, L.ch = L. chinensis, I.ru = I. rugosum, P.d= P. distichum, M.v= M. vaginalis, M.n= M. nudiflora, L.p= L. perennis, C.ir = C. iria,* C.d = *C. difformis, F.mi= F. miliacea*

Weed growth

The highest weed population density (582.6/m^2) and weed dry weight (342.2 g/m^2) were recorded in row-seeded plots with no weeding. This could be attributed to available space between rows as well as fertilizer N for weed growth. The water-seeded plots had nearly 50% less weeds than the rest (Table 2)

Table 2. Weed population densities and weed dry weight at 42 DAS in un-weeded plots with different seed establishment methods

Treatments	Weed count (number/m^2)			Weed dry weight (g/m^2)		
	Grasses	Broadleaves	Sedges	Grasses	Broadleaves	Sedges
Standard-seeding	306.4 b	63.2 a	124.3 b	106.5 b	65.3 a	88.3 ab
Water-seeding	161.3 a	68.4 a	46.5 a	67.3 a	112.4 b	32.3 a
Row-seeding	357.8 b	96.5 b	128.3 b	132.4 c	108.5 b	101.3 b

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05)

Water-seeding affected emergence, establishment, seedling height and weight of *E. crus-galli* and *L. chinensis* (data not shown), but not in *I. rugosum*, which was the dominant weed in the irrigated rice eco-systems. Moody and De Datta (1982) reported that flooding delays seedling emergence and population densities of weeds in rice fields. Rao and Moody (1995) reported that the buoyant nature of the *I. rugosum* seeds and its ability to germinate on water surface and establish from shallow water depths during intermittent periods of flooding.

Grain yield

Seed establishment methods of rice resulted in a significant difference ($p< 0.01$) in rice grain yield. Herbicide-treated row-seeded plots produced the highest grain yield (5.67 t/ha) (Table 3). This may be due to the reduced intra-specific competition in rice with the availability of a wider space and reduced rice-weed competition due to effective weed management.

Table 3. Effect of different seed establishment on yield and yield component of rice.

Treatment	No. of panicles/ (m^2)	No. of grains/ Panicle	1000 grain weight (g)	Yield (t/ha)
Standard-seeding and un-weeded	378 b	78 b	27.6	1.46 d
Water-seeding and un-weeded	308 b	92 ab	27.0	2.12 cd
Row-seeding and un-weeded	474 a	104 a	28.5	3.67 c
Standard-seeding and herbicide-treated	501 a	112 a	27.7	5.23 b
Water-seeding and herbicide-treated	505 a	92 a	28.2	5.13 b
Row-seeding and herbicide-treated	543 a	121 a	26.8	5.67 a

Within a column, means followed by the same letter are not significantly different by the DMRT ($p=0.05$)

The yield loss in un-weeded plots when compared to the herbicide-treated plots were 35%, 58% and 72% in row seeded, water-seeded and standard plots, respectively. The grain yield in water-seeded and standard plots was not significantly different. Row-seeding with effective weed management produced a higher grain yield when compared to standard- and water-seeded methods. Water-seeding reduced the emergence and growth of grasses and sedges and resulted in a similar rice yield to that of the standard crop establishment method. The results showed that water-seeding can be easily practiced in water-logged rice lands in Sri Lanka without a significant reduction in rice yields.

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SEQUENCE VARIATION IN THE GENE AFFECTING THE GRAIN SHATTERING TRAIT IN WEEDY RICE (*Oryza sativa* L. f. *spontanea*) IN ASIA

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Abstract: Weedy rice (*Oryza sativa* L. f. *spontanea*) is one of the most harmful weeds invading paddy fields resulting in loss of income for farmers. This is caused by their undesirable traits, easily shattered seeds at maturation, seed dormancy, and other traits. Its resemblance to cultivated rice in plant architecture results in its higher survival rate in the rice field. Therefore, understanding the evolutionary mechanism of weedy traits is important for sustainable crop management. This study focuses on sequence variation of the *sh4* gene controlling the seed shattering trait among weedy rice accessions collected from Japan, Malaysia, and Thailand. The sequence results indicated no difference in the particular regions analyzed among the weedy and cultivated rice, suggesting a portion of *sh4* gene may not be a major control factor in the shattering trait in weedy rice. Weedy rice strains from different sources, one from the direct seeded rice paddies and the other from marginal regions of rice fields in contact with wild rice, share similar sequences. This may be explained by the shattering trait of weedy rice which may not be the outcome of evolution of the same genetic site at domestication.

Key words: Seed shattering, *sh4* gene, weedy rice

Introduction

Rice (*Oryza sativa* L.), the world's primary food, was domesticated from the wild perennial species, *O. rufipogon* Griff. A major difference between domesticated rice and wild rice is that the former has the ability to retain mature seeds until harvesting, while the latter naturally sheds its seeds immediately after maturity, and the seeds then wait for the next season by dormancy in the form of soil-buried seeds (Cai and Morishima, 2000). The difference is caused by unconscious and conscious human selections under cultivation, suggesting the increase of non-shattering allele frequency, and eventual replacement of the shattering alleles during domestication. However, during the last decade, the shattering trait has been found to be present again in direct seeded paddy rice fields (Baki *et al*. 2000; Kim, 1995; Suh *et al*. 1997; Tang and Morishima, 1997). A significant amount of genetic research for unwanted traits found in weedy rice has been conducted by scientists, including major allele(s) controlling the shattering trait.

There are four previously mapped loci governing the shattering habit, *sh1* on chromosome 11 (Nagao and Takahashi, 1963), *sh2* on chromosome 1 (Oba *et al*. 1990), *Sh3* on chromosome 4 (Eiguchi and Sano, 1990; Nagai *et al*. 2002), and *Sh4* on chromosome 3 (Fukuta *et al*. 1994; Fukuta and Yagi, 1998). Additionally, several other QTL for shattering have been reported on chromosomes 1, 3, 4, 7, 8, and 11 (Xiong *et al*. 1999; Cai and Morishima, 2000; Bres-Patry *et al*. 2001; Thompson *et al*. 2003). Recently, one locus on the rice chromosome No. 4 has been found to account for most of the phenotypic differences of grain shattering between wild and cultivated rice, and sequence variation has been found between wild and cultivated rice in this genomic region (Li *et al*. 2006). In this study, we compare a partial sequence of the *sh4* gene, and the resulting genetics framework will be used for characterizing the sequence variation of the seed shattering gene in Asian weedy rice.

Materials and Methods

Weedy rice samples were collected from direct seeded paddy fields in Okayama Prefecture (Japan) and southeast Asian countries during the harvesting season. Material samples of cultivated rice, (*O. sativa* subspp. *indica* and *japonica*), weedy rice (*O. sativa* L.), and wild rice (*O. rufipogon*) were selected in this study. The total genomic DNA was extracted by FTA Classics Card (Whatman Inc.) during leaf sample collection outside Japan, or by CTAB method with modification of Doyle and Doyle (1987) in Japan. Primers for amplification of a 1.1 kb portion of the *sh4* gene (Figure 1a) were designed based on the conserved region from GeneBank (accessions nos. AL606619, DQ383373, DQ383371, DQ383399 and DQ383411). PCR reactions were carried out using an Astec Thermal Cycler (Astec model PC-802, Japan) with the following protocol: 95ºC for 5 min; 35 cycles of 95ºC for 1 min; 58ºC for 1.5 min; 72ºC for 2 min; and a final extension of 72ºC for 5 min. PCR products were checked by 1% agarose gel electrophoresis, and purified using Viogene, PCR-M™ clean up system following the manufacturer's protocol. Cycle sequencing was conducted using the amplification primers and BigDye version 3 reagents (Applied Biosystems, USA), with an ABI 3100 Prism automated DNA sequencer (Applied Biosystems). The region was aligned using the CLUSTAL option in the multiple alignment program MEGA V3.1 (Kumar *et al*. 2004), with minor manual adjustment.

Results and Discussion

Field observation - seed shattering in weedy rice

The degree of shattering of all weedy rice during the harvesting time was very high, and grain shed with the slightest touch on the panicle's tip or even being blown by wind, while the cultivated *japonica* rice was non-shattering, and cultivated *indica* rice showed a very low degree of shattering. However, the wild rice also indicated a very high degree of grain shedding.

Sequence comparison of sh4 gene

The DNA sequencing of a 1.1 kb portion of the *sh4* gene revealed low variation among weedy, wild, and cultivated rice. Li *et al*. (2006) compared the 1.7 kb of *sh4* gene among weedy, wild, and cultivated rice, and indicated two major changes in genomic constituents: the first is a nucleotide substitution (t/g), resulting in amino acid changes between Asparagine (N) in all cultivated rice accessions and Lysine (K) in all wild rice accessions (Figure 1b) at amino acid position No. 79. Second, there are 2 sites of indel at amino acid position No. 152 and a stretch 5 amino acids from No. 157 until position No. 162. However, the single nucleotide substitution (N/K) has been reported as critical for shattering trait, which may alter the DNA binding or sequence recognition properties of the protein (Lin *et al*. 2007). Based on the shattering trait shown by weedy rice, it was speculated that the genetic structure of its *sh4* gene could be similar to wild rice (wild sequence) (Li *et al*. 2006). However, our results indicated that weedy rice shared the same sequence with all cultivated rice accessions at these sites (domesticate sequence), but it had the shattering phenotype. A part of the *sh*4 non-coding genomic region was also accessed in weedy rice, but still no variation was found between weedy and cultivated rice.

Previous studies indicated that the shattering trait in rice can be controlled by different genes on different chromosomes (Bres-Patry *et al*. 2001; Cai and Morishima, 2000; Eiguchi and Sano, 1990; Fukuta *et al*. 1994; Fukuta and Yagi, 1998; Nagai *et al*. 2002; Nagao and Takahashi, 1963; Oba *et al*. 1990; Thompson *et al*. 2003; Xiong *et al*. 1999).

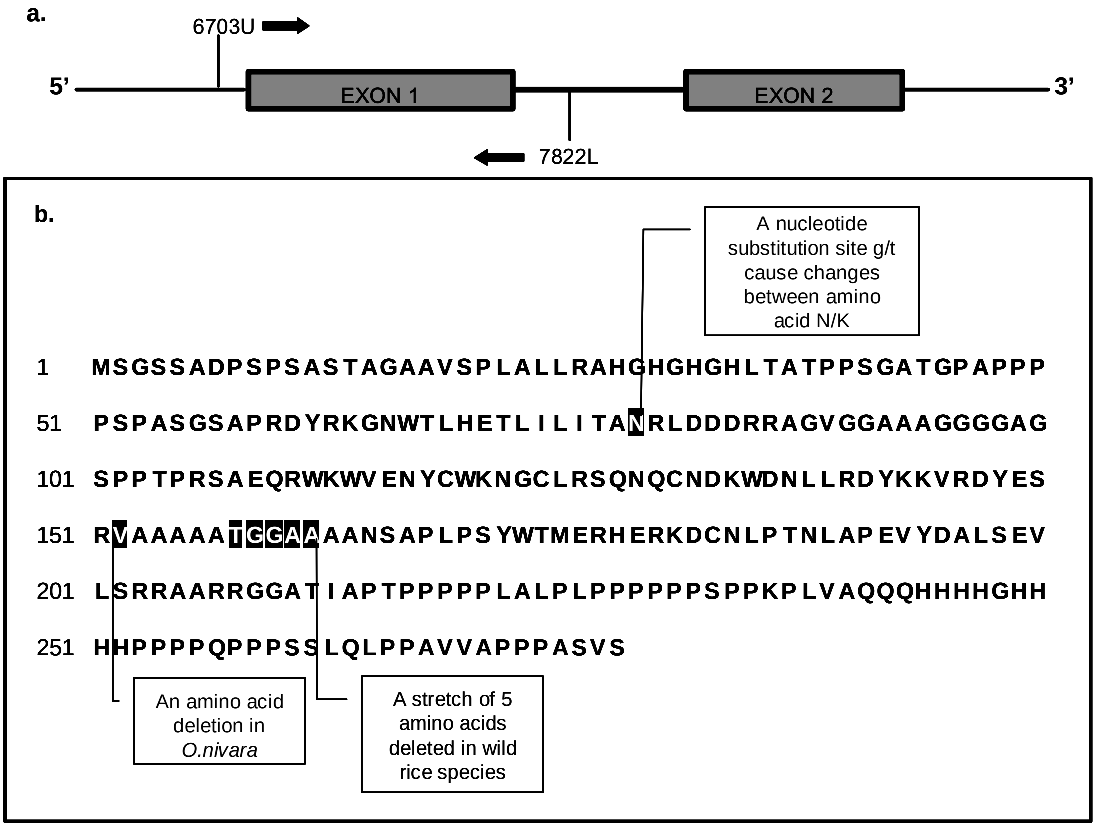

Figure 1. (a) The genomic region of the *sh4* gene in all of the cultivated and weedy rice accessions. 6703U and 7822L are the primers used for sequencing of the 1.1 kb *sh*4 genomic regions in weedy rice. Solid bars indicated the coding region. (b) Protein sequence of the first exon of *sh4* gene N/K is the nucleotide substitution site at position No. 273, while V, and TGGAA are the 2 sites of indel. Deletion of these regions has been reported in *O. nivara* (Li *et al.*, 2006).

Our results indicated a non-shared similar sequence at nucleotide +273 and deletion of 6 amino acids between weedy rice and wild species, which is in contrast to reports of Li *et al*. (2006). These results explained that the partial sequence of the *sh4* gene examined in this study does not appear to control the shattering trait in weedy rice. In other words, different sites of genes may be selected for the development of the shattering phenotype during the diversification of weedy rice. However, better sequencing of different genes and molecular markers with a larger sample size of weedy rice is needed to examine and confirm this result. On the other hand, the appearance of weedy rice is not only limited to a place where cultivation is marginal to wild rice population, but to everywhere in rice-growing regions. Even though weedy rice occurred extensively in Japan before the 1950s, seed replacement was practiced during every 5 year period, and transplanting rice cultivation helped to diminish the weedy rice from fields. However, recently, weedy rice has re-appeared in direct seeded rice fields, probably caused by lack of erasing the degenerated seeds in the field, or a mixture of weedy rice seeds and cultivated rice seeds being sown together in the field.

In short, in an evolutionary context, accumulation of gene mutations such as the grain shattering trait, pericarp coloring, long awned trait, and seed dormancy effectively resulted in an increased diversification of weedy rice. Therefore, extensive collections of weedy rice genetic resources, in particular at the population level, and comparison of different loci among populations will help promote better understanding of the diversification process of weedy rice which will be the next step of our research.

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INTERACTION OF CROPS, WEEDS AND THE ENVIRONMENT: 3D ARCHITECTURAL MODELING AND VISUALIZATION FOR IMPROVED INTEGRATED WEED MANAGEMENT

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Abstract. The University of Queensland has been involved in the development of a number of interactive three dimensional (3D) plant architectural models for weed science research and education. The overall approach makes use of the Lindenmayer systems (L-systems) formalism using L-studio software. Such models can explore and simulate the dynamic competitive relationships between crops and weeds in 3D on the computer screen. A combination of real-world and virtual experiments are used to investigate the influence of a number of biotic or abiotic factors on weed performance. Results from further real-world studies can then be used to refine and validate the various models. These models can be useful in helping to refine IWM packages both in terms of directing future research work and in the arena of extension and education. As examples, these models cane be used to simulate the inter- and intra-specific response of a crop to a weed at different crop and weed densities and under different levels of soil moisture, simulate the application of a pesticide to weed and crop canopies, simulate the shading effect of a crop on a developing weed, and used to help select crop cultivars with traits that promote weed growth suppression.

Key words: architecture, competition, education, model, software, L-systems

Introduction

A number of software tools for weed science research and education have been developed with the involvement of the University of Queensland, Australia. In one such example, a series of interactive simulation models of weed and crop growth are possible. To create the three-dimensional plant growth (virtual plants) a combination of mathematical equations, computer science and biological expertise are used. Using such systems it is possible to model the canopy interaction of a crop and a weed or the fate of a pesticide application on to plants and off target locations such as the soil surface. It is also possible to model insect damage on plants at different stages of growth, for example, by a biological control agent.

Simulation models

The growth of plants and their responses to different internal and external stimuli can now be simulated in computers. To do this, scientists at the University of Queensland have harnessed technology to measure, in three dimensions, the structural development of weeds and crops. These measurements are transformed into models that are interpreted by software, developed by collaborators at the University of Calgary in Canada (Prusinkiewcz *et al*. 2000), to generate virtual plants: computational representations of crop and weed structure that can be displayed on computer screens (Hanan and Room, 1997; Prusinkiewicz, 2004). This approach is revolutionizing the way weed scientists can study plant interactions with one another. Some expensive field trials, which take many months to complete, will be replaced by virtual experiments that provide answers in minutes or hours, allowing the researcher to only undertake key field experiments. As well as simulating crop and weed growth and development, the simulations can incorporate the movement of insects over a plant, and the movement of metabolites, plant growth regulators or systemic pathogens and pesticides within a plant. Simulating insect movement and behavior, for example, can allow scientists to predict the amount of damage a weed will sustain when subjected to certain kinds of biological control agents at different stages of growth and in different environments. It is also possible to simulate how herbicide is sprayed on to weeds, to see where it is deposited on leaves and how much is lost into the environment. Different kinds of application can be simulated under a wide range of environmental conditions

L-Systems

To create plant growth simulations or virtual plants a combination of mathematics, computer science and biological expertise are used. In their simplest, empirical form, the models can provide an animated representation of field measurements that would normally appear as numbers in tables. To convert these numbers into structures that can be visualised, a set of 'growth rules' expressed in a notation called an 'L-System', after its inventor, Lindenmayer (1968), are used.

An L-system takes advantage of the modular nature of plants, for instance dividing the structure into repeating units consisting of a segment of stem, a leaf, an apical bud and an axillary bud. Each apical bud grows to produce another whole unit; its development can be represented by the growth rule, where A = apical bud, → = "produces", I = internode, L = leaf, B = axillary bud and [] represents the start and end of a branch (Room *et al.* 1996). Computer software applies the rules for the time period to be simulated, then 'interprets' the resulting strings of symbols graphically. For example, applying the rule A → IL[B]A in a series of 'time steps' generates a series of growth stages which appear as a growing, virtual plant (see Figure 1 and http://www.cpai.uq.edu.au/virtualplants.html). The simulation can focus on a whole plant or on a part of a plant. Images may also be viewed from any angle and can be displayed in sequence to give the impression of growth.

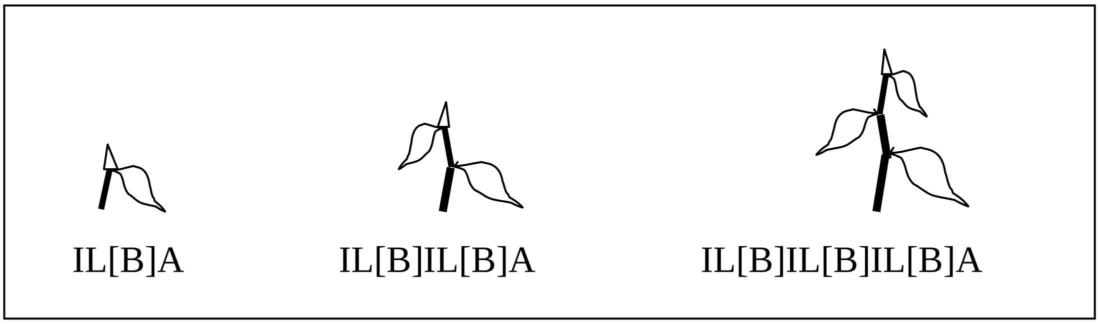
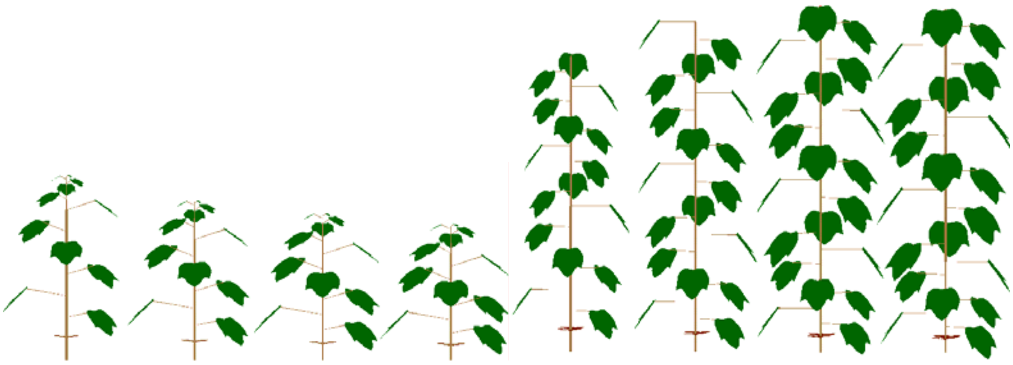


Figure 1. The first three stages in the development of the growth rule A → IL[B]A. The strings below are the computer representations, which are interpreted to create the visualizations above.

Example one: Crop-weed interactions at different densities and soil moisture levels.

In this interactive simulation model it is possible to grow a crop (Figure 2) and modify the planting and row spacings of that crop (Figure 3) and the density of the weeds around it and see the effect on canopy architecture of both. These simulations can be run under different soil moisture contents.

Figure 2. Two stages in the development of a virtual cotton plant.



First Reproductive Node Stage At Plant Harvest Stage

Figure 3. Virtual cotton models showing plant height at first reproductive node stage (left hand four plants) and at harvest (right hand four plants). The spacings from left to right, in each half of the figure, are 5, 10, 20 and 120 cm.

Example two: Pesticide fate at the canopy interface

In this simulation model it is possible to apply a pesticide and, using knowledge of the dynamics of spray droplet movement, see the fate of droplets both on a simulated plant and onto the soil below (Figure 4). This is achieved by combining the models of plant architecture, different boom sprayer configurations and the simulation of individual pesticide droplet movements in the atmosphere.

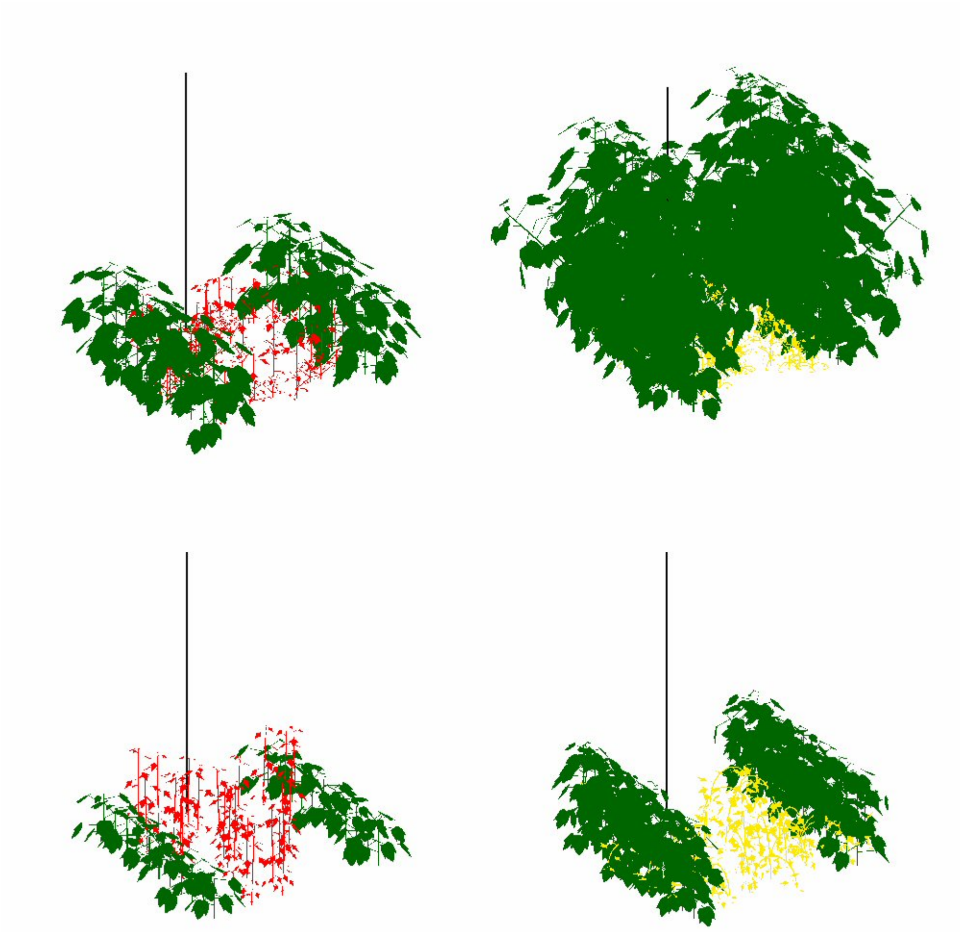


Figure 4. Interaction of Cotton and Bladder ketmia, top row no water stress, bottom row with water stress at ages 60 (left) and 120 (right) days. Green - cotton, red – immature bladder ketmia, yellow senesced bladder ketmia.

The equations used allow for the effects of gravity and air drag, but not air turbulence. The model allows for particular nozzle types to simulate pesticide application from different configurations of overhead and dropper boom sprays. Routines can be run to detect when droplet trajectories intersect plant parts, to calculate the relative amounts of pesticide reaching different plant parts, and to show the amounts of pesticide in visualizations reaching the crop/weed or soil surface (Figures 5 and 6).

Example three: Canopy-based plant-plant interactions

Plate 1 illustrates the use of a light simulation model (Cieslak *et al*. 2007) to assess the shading ability of a clump of a riverine weed *Arundo donax* (Thornby *et al.* 2007). In the simulation, an array of light sensors is positioned below the modeled plant with white cells representing full sun, and darker cells those with varying levels of shade In the left image, the light is evaluated at noon, and in the right image it is evaluated over the course of a day. A similar approach could be used to evaluate the effect of different cultivars of a crop upon weed growth and reproductive capacity.

Figure 5. Simulation of the movement of spray droplets and deposition on a cotton plant

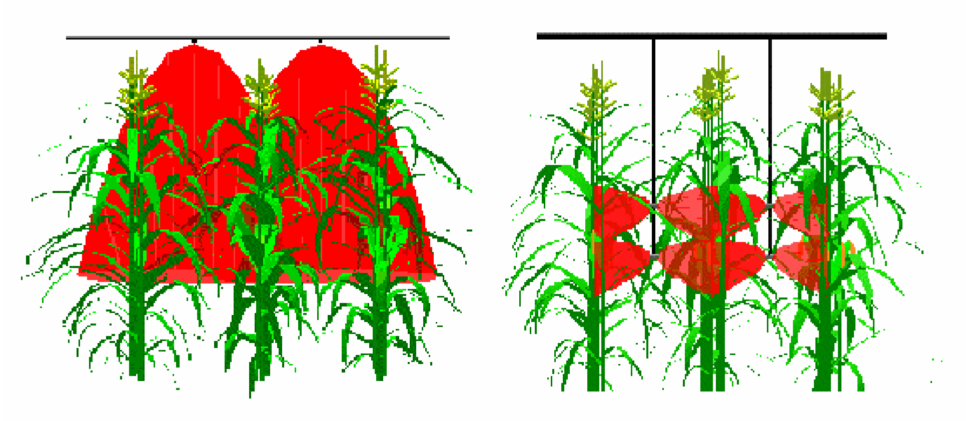


Figure 6. Views along furrows during a simulated pass of a spray boom with overhead nozzles (left) and with droppers having 2 nozzles each side (right).

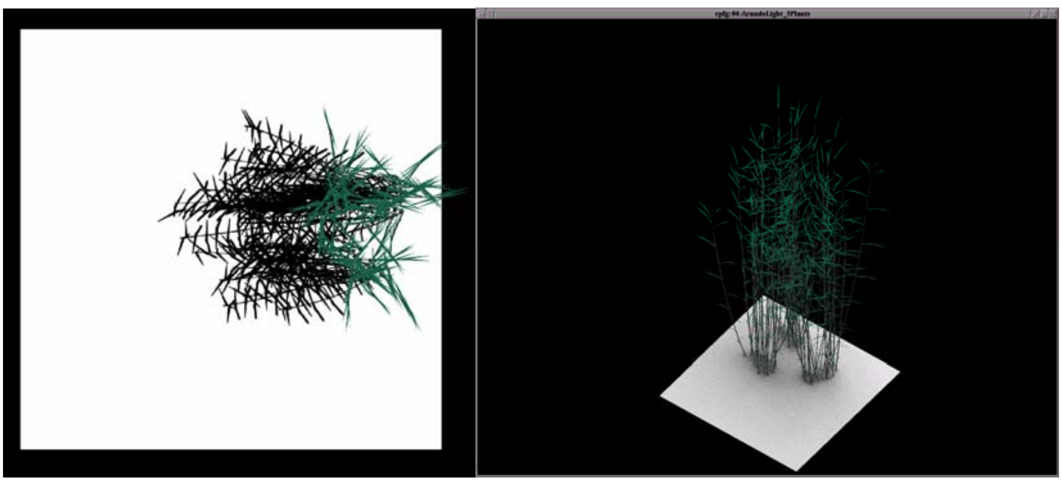


Plate 1. *Arundo donax* with light sensor array. Overhead view showing shadows created by noon sun (left) and diagonal view showing light integrated over an entire day (right).

Conclusion

We have developed a number of interactive software tools for weed science research and education. In one such example, a series of interactive simulation models of pesticide application based on the dynamics of spray droplet movement and the architectural development of crop plants has been developed.

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ALLELOPATHIC EFFECTS OF *Parthenium hysterophorus* (L.) EXTRACTS ON SEED GERMINATION AND GROWTH OF WHEAT AND ASSOCIATED WEEDS

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Abstract: *Parthenium hysterophorus* (L.) being a declared invasive weed is threatening the biodiversity of Pakistan. A laboratory based study was undertaken during January 2007 in Weed Research Laboratory, Department of Weed Science, NWFP Agricultural University Peshawar, Pakistan to investigate the allelopathic potential of aqueous extracts of *P. hysterophorus* (L.) against wheat (*Triticum aestivum*), *Avena fatua* and *Lepidium* sp. The whole plants of *P. hysterophorus* were dried in shade and ground to a fine powder, which was dissolved at 10, 20 and 30 g l^{-1}. Ten seed of each test species were placed in Petri dishes and extracts were applied when needed. A control was also included for comparison. Increasing the concentration of *P. hysterophorus* reduced the germination percentage, seedling length and seedling weight of all the three species tested significantly. The tolerance order of the species against the extract concentration of *P. hysterophorus* were *Triticum aestivum* > *Avena fatua* > *Lepidium* sp. The present study suggests that *P. hysterophorus* (L.) affects the agro ecosystem and needs careful attention. However, these extracts can be used as a viable weed management technique, but further studies are needed to explore the full potential of Parthenium as a bioherbicide.

Key words: Parthenium, allelopathy, wheat, weeds.

Introduction

The concept of plants affecting neighboring plants by releasing chemicals in the environment has been known since 370 BC (Willis, 1997). Allelopathy is a chemical process that a plant uses to keep other plants out of its vicinity. It is a natural and environment friendly technique, which may prove to be a unique tool for weed management and thereby increase crop yields. Chemicals with allelopathic potentials are present in virtually all plants and their tissue, including, leaves, stems, flowers, roots seeds and buds. Under appropriate conditions these chemicals may be released into environment (generally the rhizosphere) in sufficient quantities to affect the neighboring plants (Khan *et al.* 2005).

Allelopathic crops may affect the germination of subsequent crops; therefore those crops should be included in rotation with tolerant species. One potential technique for exploiting allelopathy in weed management is the transfer of allelopathic characters from wild species into commercial crops cultivars *i.e*. germplasm selection. These allelochemicals offers great potential as biopesticides because they are free from problems associated with the present chemical pesticides. In the past two decades much more work has been done on plant derived compounds as environmentally safe alternatives to herbicides (Duke *et al*. 2002). These chemicals could be used for weed management directly or their chemistry could be used to develop new herbicides. Parthenium (*Parthenium hysterophorus* L.) is an aggressive weed native to Southern North America, Central America, the West Indies and Central South America (Picman and Picman 1984). It has allelopathic effects and drastically retards the growth of many species (Tefera 2002). The inhibitory effects of *Parthenium hysterophorus* (L.) on germination of many crops have been reported (Narwal, 1994). Similarly, Tefera (2002) concluded that with the increasing concentration of Parthenium extracts, seed germination and growth of *Eragrostis tef* decreased significantly. Recently Ko *et al.* (2005) reported the inhibitory effects of the husk extracts of seven rice varieties on growth of barnyard grass [*Echinochola crus-galli* (L.) Beauv.]. Similar adverse effects of water extracts of different *Brassica* sp. against germination and growth of cut leaf ground cherry weed

(*Phyllis angulated* L.) have been reported by Uremis *et al*. (2005). Sundramoorthy *et al*. (1995) concluded that *Prosopis juliflora* significantly inhibited the seed germination in Pearl millet. Ibrahim *et al.* (1999) reported that *Eucalyptus camaldulensis* has allelopathic effects on crops.

Parthenium has spread rapidly in Pakistan over the last 20 years and is now a serious weed of wastelands and grazing lands, especially in rainfed areas (Javaid and Anjum, 2006). Presently Parthenium can be found along the roadsides and even in agricultural crops such as maize in NWFP, Pakistan. Therefore a detailed study of this weed will lead towards a better management approach and its possible use as a bioherbicide. Keeping in view the importance of the allelopathic potential of *P. hysterophorus* (L.), this experiment was conducted under the laboratory conditions to investigate the allelopathic status of *P. hysterophorus* (L.) and to quantify the response of *Triticum aestivum* and its associated weeds *i.e. Avena fatua* and *Lepidium* sp. to different concentrations of *P. hysterophorus L.* extracts.

Materials and Methods

A laboratory experiment was conducted in the Weed Research Laboratory, Department of Weed Science, NWFP Agricultural University Peshawar, Pakistan, during January 2007 to investigate the allelopathic effects of *Parthenium hysterophorus* (L.) on seed germination, growth and fresh weight of wheat and its associated weeds *i.e. Avena fatua* and, *Lepidium* sp. The seeds of crop and weed species were collected in April 2006 and stored at room temperature. *Parthenium hysterophorus* (L.) plants (grown naturally in Islamabad) were collected at the flowering stage in November 2006 and dried under shade. The whole plants were ground and the dry powder form of *P. hysterophorus* (L.) was soaked for 24 hrs in tap water at room temperature. The concentrations of the powder used were 10, 20, and 30 g l^{-1}. Ten seeds of each crop and weeds species were placed in Petri dishes on blotting paper, which were soaked with the respective Parthenium solutions at 0% (control), 10, 20, and 30 g l^{-1}, at the inception and at regular intervals. All treatments were maintained at 20^{o}C, with three replicates in a Completely Randomized Design. After ten days, the data on seed germination percentage, and seedling length (cm) and seedling weight (mg) per plant were recorded, using an electronic balance and a graduated scale. The data collected was subjected to analysis of variance using MSTATC computer software (Russel, 1991). Means were separated using the least significance difference test at P<5% (Steel and Torrie, 1980).

Results and Discussion

Germination percentage

The different concentrations of Parthenium significantly affected the germination of all test species (Table1). Species means indicated that maximum (98.33%) germination was recorded in wheat followed by *Avena fatua* with 67.5% and *Lepidium* sp. (31.67%) germination. Similarly, the concentration means indicated that the germination in the control treatment (81.11%) was at par with the Parthenium extract at 10 g L^{-1} (81.11%) also a marginal inhibitory effect of Parthenium on the wheat seed germination while inhibitory effect on the other species was greater. With the increasing concentrations of Parthenium, the seed germination of *A. fatua* and *Lepidium* sp. decreased drastically. The present findings suggest that weed seeds are more sensitive to Parthenium extracts when compared to wheat seeds. Hence Parthenium can be used as a tool for weed management in the future research programs. Tefera (2002) reported that *P. hysterophorus* (L.) extracts significantly inhibited the seed germination of *Eragrostis spp.* Different kinds of allelopathic chemicals have been identified in Parthenium leaves (Stephen and Sowerby, 1996). Seed germination of *Lepidium* sp. was more prone to higher concentration of Parthenium extracts, and there was no germination at Parthenium concentration of 30g l^{-1}.

Table 1. Effect of different concentrations of Parthenium extracts on seed germination of the test species.

Species tested	Concentration of Parthenium extracts				Means
	0 g l^{-1}	10 g $^{-1}$	20 g $^{-1}$	30 g l^{-1}	
Wheat	100 a	100 a	96.6 ab	96.6 ab	98 A
Avena fatua	80 cd	86 bc	73.3 de	30.0 g	67 B
Lepidium sp.	63 ef	57 f	6.6 h	0.00 h	32 C
Means	81 A	81 A	59 B	42.22 C	

LSD (p=0.05) for concentration means = 0.6659, LSD (p=0.05) for species means = 0.5767, LSD (p=0.05) for interaction = 1.153. Values followed by same letter are not significantly different at p=0.05.

Seedling Length per plant

The different concentrations of Parthenium had a significant effect on seedling length of wheat and associated weeds (Table 2). Among the species, the highest seedling length (8.29 cm) was recorded for *A. fatua*. However, this value was similar to that of wheat (7.59 cm). The lowest value (0.33 cm) was recorded for *Lepidium* sp. Similarly, the seedling length plant^{-1} in the control (7.96 cm) and Parthenium extract at 10 g l^{-1} were similar and significantly decreased with increasing concentrations.

Table 2. Effect of different concentrations of Parthenium extracts on seedling length (cm) per plant of test species.

Species tested	Concentration of Parthenium extracts				Means
	0 g l^{-1}	10 g l^{-1}	20 g l^{-1}	30 g $^{-1}$	
Wheat	10.08 a	12.03 a	5.55 b	7.72 bcde	7.59 A
Avena fatua	12.97 a	12.77 a	3.44 bcd	4.00 bc	8.29 A
Lepidium sp.	0.82 cde	0.45 de	0.05 e	0.00 e	0.33 B
Means	7.96 A	8.41 A	3.01 B	2.24 B	

LSD (p=0.05) for concentration means = 1.916. LSD (p=0.05) for species means = 1.659. LSD (p=0.05) for interaction = 3.319. Values followed by the same letter are not significantly different at p=0.05.

Seedling length of all the test species (wheat, *A. fatua* and *Lepidium* sp.) significantly decreased with the increase in the concentration of Parthenium. Thus, it can be concluded that the inhibitory effect of Parthenium is greater at higher concentrations. Heavy infestation of Parthenium in an area may accumulate allelochemicals in higher amounts in the soil, adversely affecting crops as well as the weeds. However, further studies of Parthenium extracts are needed to support their use for weed management in agricultural crops. It can be concluded that higher concentrations of Parthenium extracts retard the growth of plants. This could be due to the inhibition of cell division, as allelopathic chemicals have been found to inhibit gibberellin and indole-acetic acid activities (Tomaszewski and Thimann, 1966).

Seedling Weight per plant

The different concentrations had significant effects on the seedling weight of the test species (Table 3). Among the species, the maximum seedling weight per plant was recorded in *A.*

fatua and the minimum in *Lepidium* sp. The seedling weight of the test species among 0, 10, and 20 g l^{-1} concentrations of the extracts were not significantly different ($p>0.05$). However, the highest concentration of the extracts used in the present study (30 gl^{-1}) resulted in the lowest seedling weight per plant, which was significantly higher ($p<0.05$) than the rest of the treatments.

Table 3. Effect of different concentrations of Parthenium extracts on seedling weight (mg) per plant of test species

Species tested	Concentration of Parthenium extracts				Means
	0 g l^{-1}	10 g l^{-1}	20 g l^{-1}	30 g l^{-1}	
Wheat	49.96 abc	78.23 a	37.02 bcd	23.64 cde	47.21 A
Avena fatua	75.95 a	75.15 a	67.22 ab	14.53 de	58.21 A
Lepidium sp.	1.33 e	0.87 e	0.10 e	0.00 e	0.57 B
Means	42.43 A	51.42 A	34.78 A	12.72 B	

LSD (p=0.05) for concentration means = 18.27. LSD (p=0.05) for species means = 15.82. LSD (p=0.05) for interaction = 31.64. Values followed by the same letter are not significantly different at p=0.05.

Seedling weights of all the species tested were significantly affected by increasing concentrations of Parthenium. *Lepidium* sp. was more sensitive to the inhibitory property of Parthenium, than the rest of the test species. Thus, it can be concluded compounds found in the tissues of Parthenium may cause phytotoxic effects on agricultural crops and weeds. The study suggests that Parthenium plants should be prevented from growing and spreading to non-infested areas due to its strong allelopathic nature.

Conclusions

Based on the results of the experiment, it can be concluded that *Parthenium hysterophorus* has the potential to be used as a bioherbicide for different crop, however further extensive study is needed. Infestation of *P. hysterophorus* should be discouraged in croplands and rangelands, to avoid the accumulation of allelochemicals in the soil. Further intensive studies are needed to explore the concept of allelopathy of such invasive weeds to reduce the use of expensive chemical herbicides.

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BIOACTIVE TERPENOID FROM *Helianthus annuus* (L.) AGAINST WEEDS COMMON IN WHEAT

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Abstract: Increasing environmental and health hazards have increased interest in biological control strategies. Allelochemicals can provide a good alternative for chemical herbicides. Isolated and purified naturally occurring compounds can be identified using different techniques, most of which are based on spectral characteristics of the substance. Nuclear Magnetic Resonance (NMR) spectroscopy is now the most powerful tool for providing information about the structure of molecules in solution. So far this facility has revealed a wealth of bioactive compounds from diverse sources. In a study carried out in search of natural herbicides we evaluated the potential of various sunflower varieties against five most problematic weeds of wheat. The extract found effective was then scanned for its chemical profile. Chemicals isolated and collected through several runs of High Performance Liquid Chromatography (HPLC) were rechecked against selected weeds and those found most efficient were purified for their structural elucidation. The ^{1}H and ^{13}C spectral studies revealed the structure of a new bioactive compound, Annuionone H.

Key words: *Helianthus annuus*, allelochemicals, weeds, Annuionone.

Introduction

In agriculture, weeds are of concern because they compete with cultivated crop plants for growth factors. Modern agriculture relays on synthetic chemicals to get rid of these unwanted plants. Due to increased awareness about the risks involved in use of synthetic chemicals, much attention is being focused on the alternative methods of weed control. In past two decades, much work has been done on plant-derived compounds as environment friendly alternatives to synthetic herbicides for weed control. Allelochemicals are plant secondary metabolites that are submitted to biological and toxicological screens to identify their potential as natural pesticides. Contemporary research in allelopathy focuses on isolating, identifying and quantifying specific active allelochemicals. Once these substances are identified and characterized they can be used either as natural herbicides or as models for developing new and environment friendly herbicides.

Sunflower is well known for its allelopathic compounds. Several phenols and terpenes have been reported in various cultivars of sunflower (Spring *et al*. 1992; Macias *et al.* 2002). This study was designed to evaluate the allelopathic potential of three sunflower cultivars against most problematic weeds wheat.

Materials and Methods

Plant material
Helianthus annuus var. Suncross42, Gulshan93 and Supper25, were grown and collected during the third plant developmental stage (plants 1m tall with flowers, 1 month before harvest).

Extraction
For crude aqueous extract bioassays, fresh roots, stem and leaves of *H annuus* were extracted in water (50 g/100 ml) for 24 hrs at room temperature. For fractionation guided bioassays, roots, stem and leaves were extracted in water (1:3) after 24 hrs soaking at room temperature. The crude aqueous extract was then partitioned by dichloromethane (DCM). The organic

layer was removed by reduced pressure evaporation and residue was dissolved in water for assays or in 1-2 ml of methanol for HPLC analysis. The leaf DCM fraction was further fractionated through repeated HPLC and ten fractions were isolated and bioassayed with selected weeds.

Bioassay

Seeds of problematic weeds found in wheat, namely *Chenopodium album* L., *Coronopis didymus* (L.) Sm., *Medicago polymorpha* L., *Rumex dentatus* L. and *Phalaris minor* Retz., were sown in 9 cm diameter Petri dishes moistened with 5ml of various extracts and fractions, while control received distilled water in equal amount. Petri dishes were placed in the dark at 20± 1°C for 10 days. Each treatment was replicated thrice. Dry weights were recorded at the end. Data was statistically analyzed using Duncan's Multiple Range Test.

General procedure

A Waters system consisting of a 600E pump, 717 autosampler and 996-photodiode-array detector with detection span from 200 to 700 nm was used for HPLC. To determine the chemical profiles of extracts, analytical HPLC was carried out on a Merck LiChrospher100 RP-18e column (250 mm x 4 mm i.d.) with a 5 µm particle size, and at flow rate of 1ml min^{-1}. A linear solvent gradient of MeOH:H_2O 25:75 to MeOH:H_2O 100:0 over 40 min was used for separation. Positive ion-first-order MS were recorded using LC-MS (Thermo Finnigan LCQ) with an electrospray ionization (ESI) source. Atmospheric pressure chemical ionization mass spectra (negAPCI-MS) were acquired using a micromass LCT mass spectrometer calibrated with a PEG calibration solution (acetonitrile: water 50:50). ^{1}H NMR and ^{13}C NMR spectra were recorded at 399.92 MHz and 100.6127 MHz, respectively, on a Bruker AM-400 NMR spectrometer at 30°C. Samples were dissolved in $CDCl_3$ and TMS used as a primary reference.

Results

Allelopathic effect of sunflower crude aqueous extract on dry weight of weeds

Various concentrations of sunflower roots, stem and leaf extracts resulted in significant losses, on dry weight basis, of selected weeds. The effect was found directly proportional to the extract concentration.

The 10% root extracts enhanced the weed dry weight, the 30% aqueous extracts significantly reduced ($p<0.05$) the weed dry weight, and this impact increased with increasing concentration. The 50% extract of the Suncross42 incurred the maximum losses of 86.6% in *Rumex dentatus*, followed by *Chenopodium album* (83.8%). The least effect was observed in *Coronopis didyma* that depicted 27.1% dry weight losses (Figure 1a).

A significant decline in the weed dry weights was observed when the concentration of sunflower stem extracts was 30% and higher (Figure 1b). In the case of aqueous extracts of the sunflower stem, the maximum dry weight reduction of weeds by the highest concentration used was observed in *R. dentatus* (92.9%), followed by 84% in *C. album* and *Phalaris minor*. The least effect (55.1%) was observed in *Medicago polymorpha*.

The leaf extract was found to be effective even at a low concentration of 10%. The maximum effect of Suncross42 was observed in the case of *R. dentatus* (98.4%), followed by *C. album* (96.7%) and *P. minor* (91.6%). The least effect was recorded in *M. polymorpha* that resulted in 43.8% losses in dry weight as compared to the control (Figure 1c). The interaction analyzed between weed species and concentrations of sunflower varieties used, proved that the variety Suncross42 is the most allelopathic among the tested, that followed by Gulshan93 and Supper25.

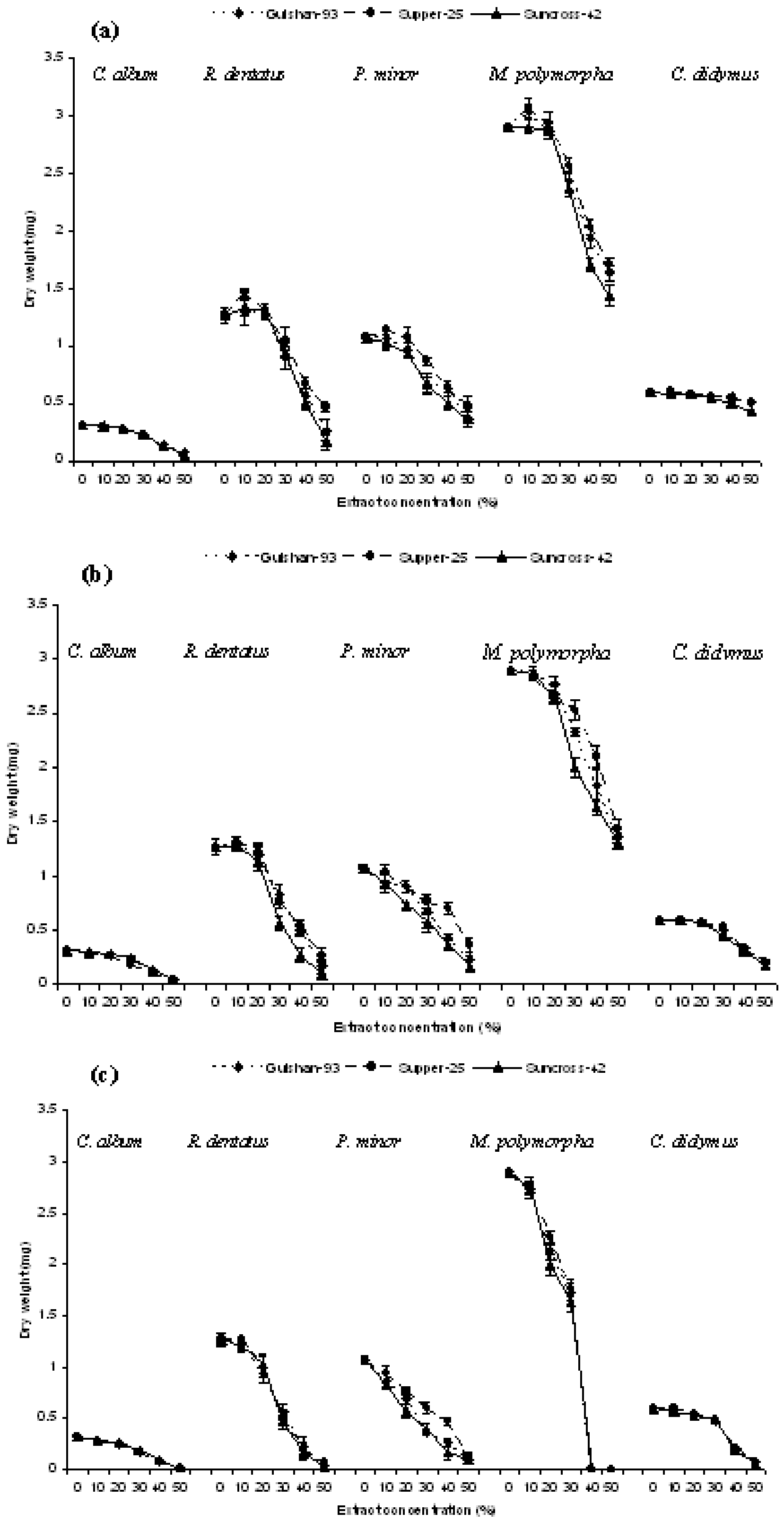

Figure 1. Effect of crude water extract of roots (a), stem (b), and leaves (c) of sunflower varieties at different concentrations.

Fractionation guided bioassays

On the bases of the above results, Suncross42 was selected for further analyses. Leaf DCM fraction of Suncross42 was found to be most efficient in reducing the dry weights of the weeds. This was followed by the stem and root fractions. The largest reduction of 75.37% in dry weight was observed in *R. dentatus*, followed by .*C album* (Figure 2). The DCM fraction of leaves was selected for further fractionation-guided bioassays. The leaf DCM fraction was sub-fractionated into nine fractions using HPLC and the fractions were then checked against *R. dentatus* and *C. album* through aqueous extract bioassays. The DCM combination was found most effective against selected weeds when compared to the tested fractions. Dry weight data analysis attested the equal efficiency of the Fr1, Fr2, Fr5, Fr6, Fr7, Fr8, and Fr9 on both the tested weeds. The remaining two fractions *i.e*. Fr3 and Fr4, were not found significantly effective in reducing the biomass of weeds (Figure 3).

Structural elucidation

The possible effective compounds from above fractions were subjected to the structural elucidation, their mass spectra were acquired, and compounds from Fr2 and Fr3 were also analyzed through NMR. These spectra revealed the presence of sesquiterpene lactones (Anjum *et al.* 2005).

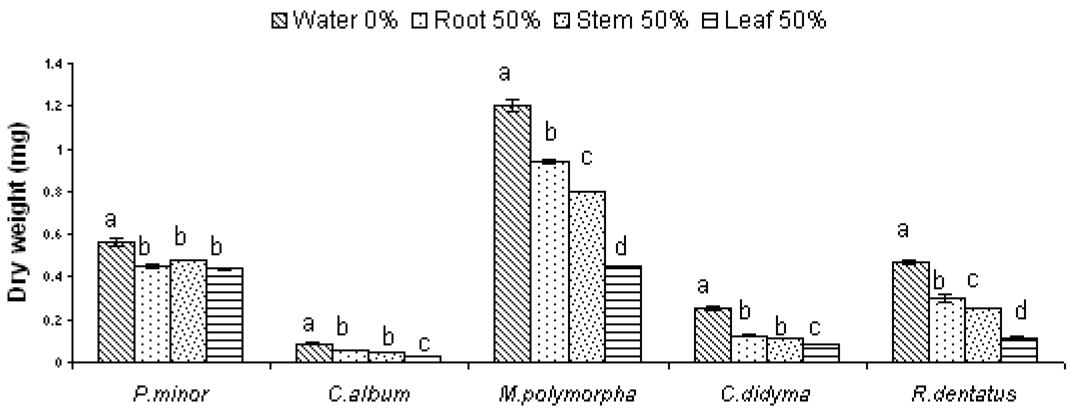

Figure 2. Effect of DCM fraction of root, stem and leaves of Suncross42 on Dry weight of selected weeds.

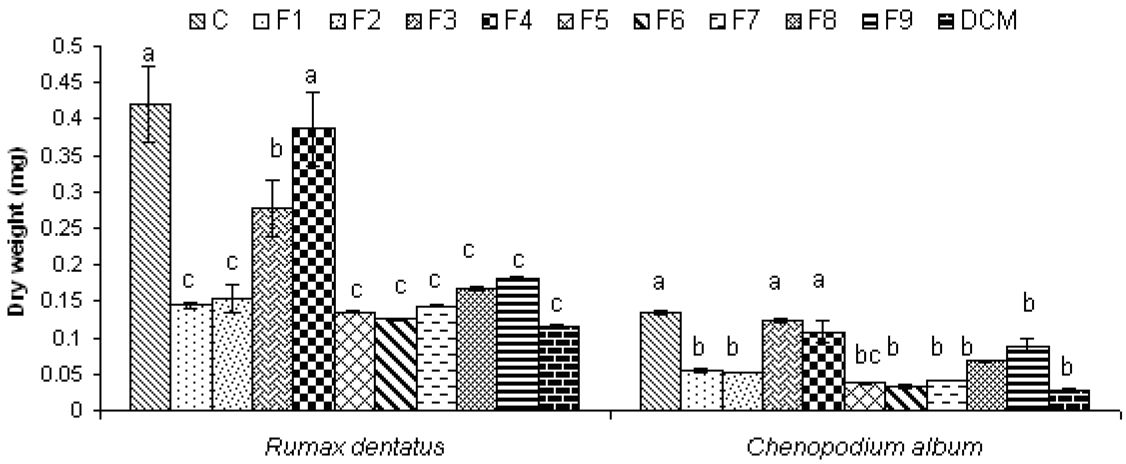

Figure 3. Effect of sub-fractions of leaf DCM fraction of Suncross42 on Dry weight of *R. dentatus* and *C. album*.

Discussion

Analysis of data acquired in crude aqueous extract bioassays showed strong allelopathic potential against selected weeds with slight variation. The reduction in dry biomass clearly indicates the phytotoxic effects of sunflower allelochemicals. Genetic variability was observed among the selected sunflower varieties in showing phytotoxic effect against weeds. This observation was found parallel to previously documented studies (Wu *et al,* 2000). Dry weight was least reduced in *Phalaris minor.* A larger effect was observed on broad-leaved weeds consistent with previous studies on rice hull (Kuk *et al.* 2001). In present study, among the broad-leaved weeds *Rumex dentatus* and *Chenopodium album* was affected the most in which dry weight reduction was recorded up to 98%. The fractionation guided bioassays also proved leaves as most effective source of allelochemicals to be used as natural herbicides. The MS and NMR spectra are in the process of interpretation for the structural elucidation of possible allelochemicals. Strong clues are there about the presence of sesquiterpene lactone in fractions analyzed. ^{1}H NMR and ^{13}C NMR data showed that analyzed compound contains a similar basic cyclohexanone ring of Annuionone, the apocarotenoids reported earlier by Macias *et al* (2004) with modified structures.

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WEED MANAGEMENT IN A RICE-BASED CROPPING SYSTEM

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Abstract: Field experiments were conducted in two different locations in late samba rice (September 2002 – January 2003) and rice-fallow-blackgram (January – April 2003) with the cultivars of CO 43 (rice, *Oryza sativa* L)) and ADT 3 (blackgram, *Vigna mungo* L) to study the influence of off-season land management in combination with various rice weed control measures on the weed competition and yield of rice-blackgram sequence. The experiments were taken up in a split plot design with the main treatments comprising off-season land management practices *viz*., ploughing the land twice at an interval of 45 days in off-season and raising a green manure in the off-season and ploughing *in situ* during land preparation, which were compared with an untreated fallow during off-season. Weed control and cultural measured taken up during the rice cropping period *viz*., hand weeding twice (at 20 and 40 days after transplanting), butachlor (1.25 kg ha^{-1}), pendimethalin (1.5 kg ha^{-1}), butachlor (1.25 kg ha^{-1}) plus 2,4-D (1.0 kg ha^{-1}), pendimethalin (1.5 kg ha^{-1}) plus 2,4-D (1.0 kg ha^{-1}) as pre-emergent application were compared with un-weeded control as sub treatments. Among the off-season land management practices, off-season ploughing (twice at an interval of 45days) excelled the other treatments by recording least weed biomass (445.57 kg ha^{-1} in location 1 and 384.03 kg ha^{-1} in Location 2) and higher weed control index (67.88% in location 1 and 70.24% in location 2) favouring higher yield attributes and grain yield (4.38 t ha^{-1} in location 1 and 4.47 t ha^{-1} in location 2), which was comparable with raising green manure in the preceding off-season and incorporating it just before preparing the land. Leaving the land fallow during the preceding off-season encouraged the highest weed dry matter production (DMP) and recorded lowest weed control index resulting in poor grain yield. Among the rice weed control measures, hand weeding twice at 20 and 40 DAT recorded the lowest weed biomass (530.78 kg ha^{-1} in location 1 and 444.2 kg ha^{-1} in location 2) and the highest weed control index (61.73% in location 1 and 64.97% in location 2) favouring higher yield attributes and grain yield (5.20 t ha^{-1} in location 1 and 5.27 t ha^{-1} in location 2). This was on par with pre-emergence application of butachlor at 1.25 kg ha^{-1} plus 2,4-D at 1.0 kg ha^{-1}. These treatments were significantly superior than the rest of the treatments in reducing the weed infestation and ultimately increased grain yield. The un-weeded control recorded the highest weed count and weed biomass resulting in the lowest grain yield. Significant interaction effects between the main treatments and sub treatments were also observed. The weed control effect of off-season land management as well as weed control practices during the late samba rice crop were shown to be carried over to the blackgram in sequence. However, the carryover effect of off-season land management practices were more pronounced in rice - fallow blackgram.

Key words: Rice-based cropping system, blackgram, integrated weed management

Introduction

India is the world's second largest producer and consumer of rice and low rice productivity in India is attributed to infestation of weeds, insect pests, diseases, poor water quality and fertility besides low yield potential of varieties. Annually, 15 million tonnes of rice is reported to be lost due to weeds in India (Chatterjee and Maiti, 1981). In the state of Tamil Nadu, a leading state in respect of rice productivity in India, transplanted rice during the monsoon season followed by mungbean or blackgram in the summer, is the traditional cropping pattern followed. As mungbean is raised as a relay crop, without any of the agro-inputs such as manures, plant-protection chemicals etc., exclusively dependency on residual soils moisture, weed competition poses severe threat and management practices need to be programmed for the whole cropping sequence. Dependence on herbicides solely, in the rice-blackgram cropping system, resulted in a shift towards perennial weeds like nut sedge, where as hand weeding alone lead to the dominance of grassy weeds and exclusive un-weeded rice culture caused the preponderance of broadleaved weeds (Kathiresan, 2002). The complexity of this situation has resulted in a need to develop a wholistic weed control programme throughout the farming period that is sustainable in terms of enhanced productivity without eroding the resource base.

Materials and Methods

Field experiments were conducted simultaneously in two locations during 2002-2003 to evolve a wholistic weed management for rice-blackgram sequence. Location 1 was the Annamalai University experimental farm situated at 11°24' N latitude, 79°41'E longitude and location 2 was the Paradhur Village, which is situated 25 km away to the west of location 1. The off-season land management practices such as ploughing the land twice during summer raising a leguminous green manure crop during summer and ploughing it *in situ* at the time of land preparation were compared with leaving the land fallow during summer, as main treatments of a split plot design. These treatments were taken up individually in strips of size 5 m x 25 m. In off-season ploughing the field was ploughed first and second ploughing was done after 45 days of first ploughing. In green manuring treatment green manure (*Indigofera tinctoria*) seeds were sown in respective strips and incorporated 12 days before transplanting of rice and in off-season fallow treatment the field strips were left fallow without any disturbance. In the ensuing rice crop, each strip that received a particular off-season treatment was superimposed with crop weed control measures in individual plots of size 5 m x 4 m. These treatments compared as sub-treatments were hand weeding twice, butachlor at 1.25 kg ha^{-1}, pendimethalin at 1.5 kg ha^{-1}, butachlor at 1.25 kg ha^{-1} + 2, 4 – D at 1.0 kg ha^{-1}, pendimethalin at 1.5 kg ha^{-1} + 2, 4-D at 1.0 kg ha^{-1} and an un-weeded control. Herbicides were applied in respective treatment plots as pre-emergence spray on 3 days after transplanting (DAT) with 500 liters $ha\text{-}^{1}$ of water using knapsack sprayer fitted with flood jet nozzle. After harvesting the rice crop in both locations, blackgram was sown in all the plots without any other agro-input and the carry over effect of rice weed control measures were studied in both the locations. All the data were statistically analysed and the critical difference was worked out at 0p=0.05 as suggested by Panse and Sukhatme (1978). The original values recorded in weed control index were subjected to Arc sin transformation before analysis.

Results and Discussion

The weed flora in both the experimental sites was dominated by *Cyperus rotundus, C. difformis, Fimbristylis littoratlis* and *Sphenoclea zeylanica*. However, weeds that occurred sporadically with lesser densities varied and they were *Leptochloa chinensis, Marsilea quadrifolia, Echinochloa sp., Bergia capensis* and *Eclipta alba* in Annamalai University experimental farm and *L. chinensis, M. quadrifolia, Echinochloa sp., Scirpus sp.* and *B. capensis* in Paradhur. Among the off-season land management practices compared ploughing twice at an interval of 45 days recorder the least weed DMP and highest weed control indices and grain yield. Raising green manure in the preceding off-season was on par with off-season ploughing. Leaving the land fallow during the off-season recorded the highest weed dry matter production and least weed control indices (Tables 1 and 2) summer ploughing exposed tubers and rhizomes of the weeds in the hot summer months with an ultimate exhaustion of their food reserves and perennation potential. Ploughing the soil at the first instance helped the dormant seed to break their dormancy and sprout, whose seedling or established aerial growth were destroyed during the second ploughing. Because of this multitude destruction of the weed vegetation and its soil seed bank, off–season ploughing surpassed the other off-season land management practices in respect of weed suppression in the succeeding crops. These findings are in line with the earlier reports of Tewari and Singh (1991) and Gnanavel and Kathiresan (2002).

Table 1. Effect of off-season land management and weed control options on weeds in rice.

Treatment	Weed dry matter production on 60 DAT (kg ha^{-1})		Weed Control Index (%)	
	AU farm	Paradhur	AU farm	Paradhur
Main treatments				
Raising green manure in off-season	494.40	430.34	53.36 (64.36)	54.69 (66.59)
Off-season ploughing	445.57	384.03	55.48 (67.88)	56.94 (70.24)
Off-season fallow	1129.70	1004.27	-	-
CD (p=0.05)	60.35	59.26	3.02	3.01
Sub treatments				
Un-weeded control	925.75	822.21	-	-
Two hand weeding (20 & 40 DAT)	530.78	444.26	51.78 (61.73)	53.71 (64.97)
Butachlor at 1.25 kg ha^{-1}	740.73	700.19	43.05 (46.10)	43.62 (47.60)
Pendimethalin at 1.5 kg ha^{-1}	803.10	764.40	40.45 (42.10)	42.64 (45.89)
Butachlor at 1.25 kg ha^{-1} + 2,4-D at 1.0 kg ha^{-1}	548.01	473.92	51.06 (60.49)	53.32 (62.63)
Pendimethalin at 1.5 kg ha^{-1} + 2,4-D at 1.0 kg ha^{-1}	609.93	532.32	55.93 (48.41)	49.61 (58.02)
CD (p=0.05)	76.87	64.31	3.43	3.45

(Figures in parenthesis are original values)

Table 2. Effect of off-season land management and weed control treatment on rice grain yield.

Treatment	Grain yield (t ha^{-1})	
	AU farm	Paradhur
Main treatments		
Raising green manure in off-season	4.22	4.29
Off-season ploughing	4.38	4.47
Off-season fallow	3.49	3.58
CD (p=0.05)	0.27	0.29
Sub treatment		
Un-weeded control	2.50	2.60
Two hand weeding (20 & 40 DAT)	5.20	5.27
Butachlor at 1.25 kg ha^{-1}	3.67	3.79
Pendimethalin at 1.5 kg ha^{-1}	3.42	3.50
Butachlor at 1.25 kg ha^{-1} + 2,4-D at 1.0 kg ha^{-1}	4.89	4.96
Pendimethalin at 1.5 kg ha^{-1} + 2,4-D at 1.0 kg ha^{-1}	4.50	4.56
CD (p=0.05)	0.38	0.39

Raising a green manure crop during the off-season and ploughing it *in situ*, just before land preparation also proved to suppress the weeds in the succeeding crops and was comparable with that of off-season ploughing. This could be due to (1) the smothering effect of standing green manure (*Indigofera tinctoria*) crop on the emergence and growth of weeds in the off-season contributing for the weed seed bank in the soil and (2) the improved soil health due to addition of organic matter and improved fertility status of the soil with an ultimate enhanced competitive ability of the crop. This finding is in line with the result of Gracy Mathew and Alexander (1995).

Among the weed control options compared as sub-treatments, twice hand weeding recorded the least weed DMP, highest weed control indices and grain yield. The superior performance could be attributed to manual removal of existing vegetation of all the weeds without sparing any one group or individual weeds. This finding is in line with the earlier reports of Navarez and Moody (1989). However, the performance of tank mix spray of butachlor 1.25 kg ha^{-1} plus 2,4-D 1.0 kg ha^{-1} was comparable with twice hand weeding and these treatments were significantly superior than the rest of the treatments. This may be due to reduced crop-weed competition particularly at early growth stages of the crop growth and enlarged spectrum of activity of the mixture due to inclusion of 2,4-D. These results are in conformity with the reports of Malik and Samunder singh (1996) and Gogoi *et al*. (2000).

The carry over effect of weed control on subsequent crop of rice-fallow blackgram was more pronounced with off-season land management. Accordingly, the highest weed control indices were recorded with off-season ploughing (Table 3). Although the carry over effects of weed control measures proved its significance, they were not well pronounced as all of them were on par.

Table 3. Carry over effect of off-season land management and weed control options in rice-fallow blackgram.

Treatment	Weed control index (%)	
	AU farm	Paradhur
Raising green manure in off-season	34.96 (32.83)	35.00 (32.90)
Off-season ploughing	35.59 (33.87)	35.46 (33.65)
Off-season fallow	-	-
CD (p=0.05)	1.42	1.44
Unweeded control	-	-
Two hand weeding (20 & 40 DAT)	29.67 (24.50)	30.13 (25.21)
Butachlor at 1.25 kg ha^{-1}	28.27 (22.43)	28.62 (22.94)
Pendimethalin at 1.5 kg ha^{-1}	27.78 (21.73)	28.11 (22.20)
Butachlor at 1.25 kg ha^{-1} + 2,4-D at 1.0 kg ha^{-1}	29.23 (23.85)	29.79 (24.68)
Pendimethalin at 1.5 kg ha^{-1} + 2,4-D at 1.0 kg ha^{-1}	28.86 (23.29)	29.32 (23.99)
CD (p=0.05)	NS	NS

Figures in parenthesis are original values, NS – Non-significant

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WILD OAT (*Avena ludoviciana* L.) INTERFERENCE IN WHEAT (*Triticum aestivum* L.)

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Abstract: Wild oat is a major weed of field crops in Iran. The effects various densities of wild oat and wheat on the growth and reproductive output of wheat and growth of wild oat were investigated in a field study in 2004. The experiments were established as a factorial combination of wheat varieties (Rooshan and Niknejad), wheat density (recommended, recommended + 25% and recommended + 50%) and wild oat (0, 25, 50, and 75 plants m^{-2}) densities. The effect of wild oat density on wheat yield loss and the effect of wheat density on wild oat biomass were described with a rectangular hyperbola model. The presence of wild oat in wheat reduced grain yield of wheat, number of tiller, number of spikes m^{-2}, aboveground dry matter and leaf-area index (LAI), and the magnitude of this reduction was dependent on weed density. Increasing the density of wheat significantly reduced the unpleasant effects of wild oat on wheat. Wild oat dry matter and LAI decreased and height increased, as crop density increased. The maximum grain yield of Niknejad and Rooshan were achieved at its recommended+25% and recommended wheat density, respectively and increased density due to increased inter specific competition decreased the grain yield of two cultivars. As the wild oat density increased, the number of tillers per plant, spikes m^{-2} and seeds per spike decreased. The results indicated that higher densities of wild oat had a negative effect on wheat yield and higher wheat densities were able to suppress biomass and LAI of this weed species.

Key words: Competition, yield loss, wild oat, wheat, plant density

Introduction

A major component of integrated weed management is the use of more competitive crops (Lemerle *et al.* 2001a; Menana and Zandstra, 2005). If a crop cultivar can tolerate weeds, it may reduce the need for synthetic herbicide, allow the use of less costly and more environmentally sound herbicides, decrease the number of cultivations, or improve yield stability in weedy fields (Lemerle *et al.* 2001a). Large differences exist in the competitive ability (CA) of field crops. Baghestani and Zand (2005) found that Niknejad had more CA than Rooshan. CA on wild oat for Niknejad and Rooshan was 1.7 and 0.56, respectively. Integrated weed management practices, such as using more competitive wheat cultivars and increased plant density, potentially improve weed management (Holman *et al.* 2004). Wheat yield loss was often lower and economic threshold values were higher at the higher wheat-plant densities (O'Donovan *et al.* 2007). Eslami *et al.* (2006) reported that increasing the density of wheat substantially reduced the adverse effects of wild radish on wheat. As crop density increased, wild radish dry matter, LAI and seed production per unit area decreased.

Considerable research has been conducted to examine the effects of wild oat interference on crop yield (O'Donovan *et al.*, 2000; Ponce and Santin, 2001; Baghestani and Zand, 2005). In Canada, 150 wild oat plants m^{-2} emerging 6 days before the crop reduced barley yield by 42% (O'Donovan *et al.* 2000). Yet, few studies compare competitive interactions of weeds with different crops. The goal of this research was to quantify the effects of *A. ludoviciana* interference on more and less competitive wheat cultivar yield and yield components over a range of crop and weed densities. We hypothesized, that increasing the plant density of wheat, would decrease the yield impact of *A. ludoviciana* on more and less competitive wheat cultivars.

Material and Methods

A field experiment was conducted in the 2004-2005 growing season at Plant, Pest and Disease Institute in Karj, Iran. The factorial set of treatments was arranged within a randomized complete block design with four replicates. Individual plot size was 2.4 m wide by 6 m long. Treatments consisted of two cultivars Niknejad and Rooshan (as more and less competitive, respectively,) 3 plant densities (*i.e.* their recommended density; recommended density + 25% and recommended density + 50%) and 4 wild oat densities (0, 25, 50 and 75 plants/m^2). Wheat cultivars were sown on 10th November 2004 by hand in rows 30 cm apart. The seeds of wild oat were broadcast by hand on the soil surface between wheat rows immediately after sowing wheat. Wild oat seedling density was determined at the 1 to 2-leaf stage of wild oat in two 0.25 m^{-2} quadrats positioned approximately 1 m from the front and back of each plot.

Grain yields were determined by harvesting a 1 m x 1 m area in each plot. The area outside the harvest plot was used for sampling of above-ground biomass. Yield components were taken as the average of 30 counts of ears in 0.1 m^2 rings randomly placed in each plot. The harvest index was calculated as the ratio of dry matter grain yield to wheat above-ground dry matter. The yield and yield component and biomass data were subjected to analyses of variance using the ANOVA procedure of the SAS statistical analysis system (SAS Institute, 1996).

Regression analyses

Competitive index (CI) was calculated on the basis of the following equation,

$$CI = \frac{V_i}{V_{mean}} \times \frac{W_i}{W_{mean}} \qquad \text{(Equation 1)}$$

where, V_i is the yield of each weed-infested cultivar, V_{mean} the mean wheat yield in all weed-free plots, W_i the biomass of wild oat in each weed-infested plot and W_{mean} the mean biomass of wild oat in all weed-infested plots.

Effect of the crop on the weed was evaluated by analysing the relationship between *A. ludoviciana* biomass and *A. ludoviciana* plant density for each plant density of wheat separately. The model selected to fit the data was the rectangular hyperbolic equation proposed by Cousens (1985),

$$Y = \frac{I * D}{1 + \frac{I * D}{A}} \qquad \text{(Equation 2)}$$

where, *y* is the biomass in g m^{-2} of *A. ludoviciana*, *d* is the weed density in plants m^{-2}, *i* is the weed biomass per unit weed density as *d* approaches zero and *a* is the maximum weed biomass at infinite weed density.

Effect of the weed on the crop was evaluated by analysing the relationship between weed density and wheat grain yield separately for each plant density, using the rectangular hyperbola described by Cousens (1985),

$$Y = Ywf \left[1 - \frac{I * D}{100 \left[1 + \frac{I * D}{A} \right]} \right] \qquad \text{(Equation 3)}$$

where, *y* is the yield of wheat, *Ywf* is the weed-free yield, *i* is the yield as weed density approaches zero, *a* is the maximum yield at infinite weed density and *d* is the weed density in plants m^{-2}.

Results and Discussion

In both weed-free and weed-infested conditions, Niknejad had greater grain yield as compared to the Rooshan (Figure 1). High genetic potential, high harvest index and having more ear number per square meter caused more yield in Niknejad. Zand and Rahimian Mashhadi (2003) also reported in both weed-free and weed-infested conditions, new cultivars (Ghods, Alamot and Alvand) had greater grain yield compared to the older cultivars (Omid, Bezostaya and Azadi). Niknejad also had higher Competitive Index than Rooshan (Table 1). Increase in the competitive index was mainly due to higher wheat grain yield, Leaf Area Index (LAI) and dry matter accumulation under weed-infested conditions (data not shown). Increase in the competitive index was mainly due to higher wheat grain yield and lower weed dry matter in new wheat cultivars compared to old cultivars under weed-infested conditions (Zand and Rahimian Mashhadi, 2003).

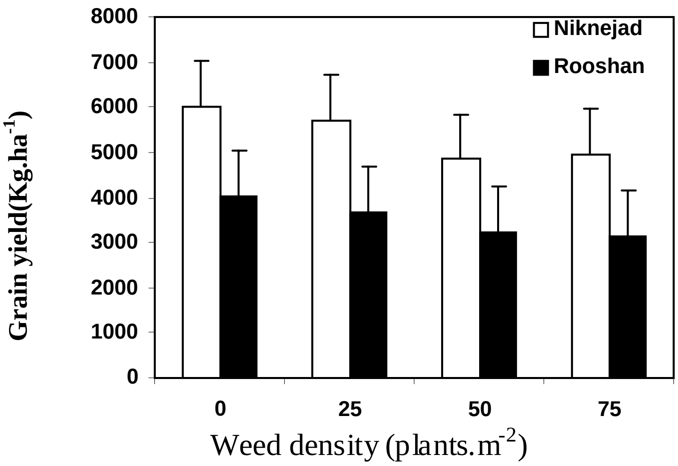

Figure 1. Effect of increasing wild oat density on grain yield in two cultivars. Standard error bars represent standard errors of means.

The wheat density had significant effects ($P<0.01$) on all growth and seed production attributes of both wheat and *A. ludoviciana* (Table 1). Competition index was increased with increasing of wheat density. The highest competitive ability was observed in the highest wheat density (Table 1). O'Donovan *et al.* (2000) reported that increasing the seeding rate improved the competitiveness of different barley varieties, but the yield was higher only in some situations. Increasing wheat density to recommended+25% increased grain yield in Niknejad but further increases in wheat density reduced grain yield. Rooshan grain yield did not increase with an increase in wheat density (Figure 2). Increasing inter- and intra-specific competition, loading and infertile tillers probably resulted in grain yield reduction in this variety. This result indicated that increasing plant density in tall cultivars is not useful. Menana and Zandstra (2005) also found that wheat yield increased in all cultivars with an increase in the seeding rate, from 150 to 200 kg ha^{-1}, but yield did not increase significantly at the seed rate of 250 kg ha^{-1}

Table 1. Effects of cultivar, plant density and weed density on some wheat traits and wild oat biomass.

Treatments	Grain yield (kg ha^{-1})	CI	No. of tillers (plant^{-1})	No. of Spike (plant^{-1})	Wild oat biomass (g m^{-2})
Cultivar					
Niknejad	5442.5	1.173762	2.525	644.0286	121.9333
Rooshan	3408.06	0.490065	1.991667	349.6538	114.4417
LSD (p=0.05)	149.21	0.033	0.133	49.71	2.05
Plant Density					
Recommended	4446.87	0.797401	2.525	464.1363	130
Recommended + 25%	4586.56	0.832316	2.3125	495.2544	116.2875
Recommended + 50%	4242.40	0.866023	1.9375	531.1329	108.275
LSD (p=0.05)	182.75	0.041	0.163	60.88	2.52
Weed Density					
0	5046.542	0	2.616667	634.4357	0
25	4540	1.094451	2.35	549.0813	98.9375
50	4107.083	1.10358	2.083333	445.6813	103.0333
75	4007.5	1.129622	1.983333	358.1664	106.6167
LSD (p=0.05)	211.02	0.047	0.188	70.32	2.91
CV (%)	8.2	9.92	14.49	24.57	4.27

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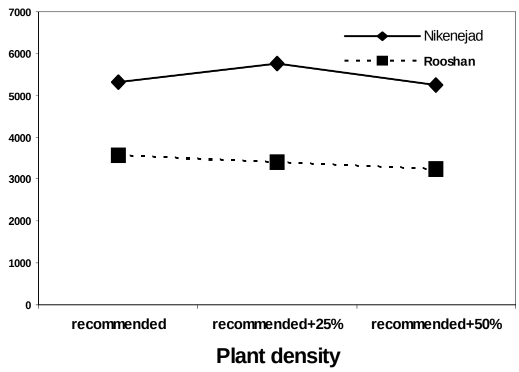

Figure 2. Effect of increasing plant density on grain yield in two wheat cultivars

The components of yield which showed a significant response to plant density are presented in Table 1. Number of tillers and number of spikes m^{-2} were the components which were affected by plant density. The maximum tillers were observed at recommended density and increasing plant density by 50% decreased the number of tillers by 23%. Decrease in fertile tillers with increasing plant density is related to lower supply of essential resources for growth for one plant. The maximum spikes m^{-2} was achieved at recommended+50% density. More plants at high plant density increased spikes m^{-2}.

Weed biomass showed significant responses to increasing of wheat density (Table 1). The relation between *A. ludoviciana* biomass and weed density was hyperbolic in shape for the two cultivars with R^2 being 0.79 or higher, indicating that *A. ludoviciana* biomass depends on plant and weed densities (Figure 3). Izguierdo *et al.* (2003) also reported that *L. rigidum* biomass was related to weed and plant density. Low crop densities allowed individual weed plants to become larger, but the high intra-specific competition which occurred at high *L. rigidum* densities caused this relationship to reach a plateau. The *A* parameter (maximum biomass achieved by the weeds) ranged from 310.18 to 255.69 g m^{-2} in Niknejad and from 297.12 to 244.68 g m^{-2} in Rooshan at recommended and recommended+50% plant densities.

The *I* parameter ranged from 9.84 to 7.69 in Niknejad and from 8.12 to 7.49 in Rooshan in recommended and recommended+50% plant densities (Table 2). As expected, the lowest *I* values were always found in the highest crop densities in two cultivars, supporting the concept that high crop stands tend to reduce the aggressiveness of the weed.

Table 2. Estimates of parameters for the hyperbolic regressions (Eqn 2) of yield (kg ha^{-1}) against weed density (plants m^{-2}). Standard errors of estimates are given in parentheses

Cultivar	Plant density	I	A	Adj R^2	Estimated reduction (%)
Niknejad	400	9.84(3.65)	310.18(70.38)	0.97	-
	500	8.09(2.52)	279.53(57.70)	0.98	9.88
	600	7.69(1.81)	255.69(38.60)	0.99	17.56
Rooshan	300	8.12(2.90)	297.12(73.50)	0.97	-
	375	7.62(1.69)	271.52(40.93)	0.99	8.61
	450	7.49(1.39)	244.68(28.69)	0.99	17.64

Wheat grain yield was affected by different densities of *A. ludoviciana* in two cultivars (Table 1). The highest grain yield was found in weed free condition in two cultivars (Figure 1). Grain yield reduction was 10.12, 18.80 and 19.95% in 25, 50 and 75 plant m^{-2} weed density. In both cultivars as *A. ludoviciana* density increased, the wheat yield decreased substantially.

There was a hyperbolic relationship between *A. ludoviciana* density and wheat yield loss. The maximum grain yield estimated by the Y_{wf} parameter of the model declined with increasing wheat density (except at recommended + 25% in Niknejad) from 6.31 to 5.80 t ha^{-1} (8.08%) in Niknejad and from 4.20 to 3.63 (13.73%) in Rooshan. The parameter *I*, which represents yield loss per *A. ludoviciana* plant, showed a steady decline with increasing wheat density in the two cultivars (Table 3). However, the relative change in *I* was considerably greater in Niknejad. The maximum yield loss (parameter *A*) also showed a consistent reduction with increasing crop density. Carlson and Hill (1985) examined the effect of wild oat and wheat density on wheat yield. Yield reductions resulting from wild oat competition were much greater at lower seeding rates. Wheat sown to achieve a plant density of 100 plants m^{-2} suffered a yield reduction of 20% from an infestation of 5.5 wild oat plants m^{-2}. However, with a wheat density of 700 plants m^{-2}, 38 wild oat plants m^{-2} would have to be present in order to cause similar yield loss. Yield loss was almost 25% when 70 wild oat plants m^{-2} were established.

Table 3. Estimates of parameters for the hyperbolic regressions (Eqn. 3) of yield (t.ha^{-1}) against weed density (plants m^{-2}). Standard errors of estimates are given in parentheses.

Cultivar	Plant density	Y_{wf} (t.ha-1)	I	A	Adj R^2
Niknejad	400	6.31 (0.30)	0.87 (0.75)	44.19 (30.06)	0.57
	500	6.40 (0.11)	0.52 (0.25)	31.47 (14.52)	0.79
	600	5.80 (0.14)	0.46 (0.34)	30.70 (21.57)	0.64
Rooshan	300	4.20 (0.09)	0.63 (0.23)	60.91 (31.63)	0.86
	375	3.96 (0.18)	0.61 (0.56)	49.94 (53.07)	0.52

The *A. ludoviciana* had significant effects ($p<0.01$) on number of tillers and number of spike m^{-2}. The maximum number of tillers and spikes m^{-2} was achieved in weed free condition. As *A. ludoviciana* density increased to 75 plants m^{-2}, number of tillers and spikes m^{-2} decreased by 24.13 and 43.54% respectively (Table 2). Izquierdo *et al.* (2003) found that density of ears was the component which was affected by competition with *L. rigidum*. This component had the highest losses due to *L. rigidum* competition, with estimated reductions of 30%-60% depending on location. As expected, increasing *A. ludoviciana* density corresponded with increase of *A. ludoviciana* biomass. *A. ludoviciana* biomass exhibited a significant linear increase with increasing *A. ludoviciana* density (Table 2).

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DISTRIBUTION, SPREAD AND PRESENT STATUS OF *Mimosa pigra* L. (MIMOSACEAE) IN PENINSULA MALAYSIA: 1980-2004

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Abstract: *Mimosa pigra* L. (Mimosaceae), a noxious semi-aquatic woody plant and once considered to be an important invasive species in the early 1980s has now spread through out Peninsula Malaysia. This happened despite an effort to control the spread through the introduction of two biological control agents, *Carmenta mimosa* and *Acanthoscelides puniceus* between 1992 and 1994. To assess the present status of Mimosa spread, a survey was conducted in 2004, which covered six habitat types namely a construction site, roadside, riverbank, reservoir, plantation and forest reserve. A total of 53 from 80 sites visited have been recorded with Mimosa populations. Five habitat types were more prone to be invaded by Mimosa whilst forest reserves were found to be non-susceptible to Mimosa invasion. Three conclusions can be drawn from this survey; (1) Soil contaminated with Mimosa seeds used in construction industries is likely to be the main dispersal agent, (2) Mimosa infestation has never been dealt successfully at its earliest establishment phase, and (3) the invasion is no longer restricted to wetland habitat but has spread into various types of landscapes and terrestrial ecosystems.

Key words: *Mimosa pigra*, distribution, Peninsula Malaysia

Introduction

Mimosa pigra L., Fabaceae (henceforth Mimosa) is a fast spreading thorny shrub, growing frequently up to 4 m but occasionally to 5-6 m tall (Forno *et al.* 1989). With annual seed rain of c. 9000 seeds m^{-2} (Lonsdale 1992; Lonsdale *et al.* 1985), Mimosa infestation can easily spread through soil contaminated with it seeds. The introductions of Mimosa outside its native range have caused great concern as it become highly invasive and important weed of the tropics (Lonsdale, *et al.* 1985). Serious environmental threats associated with Mimosa invasion are including loss of important native flora (Braithwaite *et al.* 1989; Triet *et al.* 2004a) and fauna, (Lonsdale 1992), restricted of accessible water body both to human and livestock, safety problem, and reducing land value and its productivity (Heard *et al.* 2005; Marambe *et al.* 2001; Marambe, 2001; Miller and Pickering, 1983; Napompeth, 1983; Robert, 1982; Thamasara, 1982). Wetland ecosystems face a constant threat by Mimosa invasion, where it can forms dense and impenetrable monospecific stands (Lonsdale 1992) which affecting agricultural activity and conservation effort. Similar condition already faced by some Southeast Asia countries (e.g. Thailand, Vietnam and Cambodia) which bordering the Mekong River basin (Winotai *et al.* 1992; Samouth 2004; Son *et al.* 2004;).

The earliest record of Mimosa in Peninsula Malaysia was in 1980 from a survey conducted by the Department of Agriculture (DOA) although farmers in the state of Kelantan claimed the species has spread as early as 1970s (Anwar and Sivapragasam, 1999). Its population in Kelantan was assumed to be originated from Thailand where propagules might have been brought across the border by man-related activities and/or by water along the Golok River (Anwar and Sivapragasam, 1999; Napompeth, 1983). Mimosa was gazetted as “an A2 pest” based on the 4th Schedule of the Agriculture Pest and Noxious Plants (Import/Export) Regulation in March 1982 (Mislamah *et al.* 1991) where it was considered as already introduced and spreading in Malaysia. This has been further confirmed by a DOA survey in 1991 which revealed ten of 11 states surveyed were recorded with Mimosa populations, compared to only three states in 1981 (Anwar and Sivapragasam, 1999; see also Chan *et al.* 1981 and Mansor, 1987). To control further spread of Mimosa, Malaysian government through the Malaysian Agriculture and Research Development Institute (MARDI) has initiated collaboration on biological control program with neighbouring countries and CSIRO, Australia in the 1980s. As a result, two agents; *Carmenta mimosa* and *Acanthoscelides puniceus* were introduced between 1992 and 1994.

After 24 years, Mimosa has been reclassified as weed of waste land (Othman and Abu-Hashim 2003) and less attention has been given to monitor its spread and impact. However, Mimosa invasion into wetland habitats remains a threat. Based on this, a general survey has been conducted from March to May 2004 with objectives to review current Mimosa infestation especially in Peninsula Malaysia and to monitor Mimosa population and its distribution especially in wetland habitats.

Materials and Methods

Mimosa populations were surveyed in all states within Peninsula Malaysia, divided geographically into four groups as follow; (a) northern states: Perlis, Kedah, Penang, (b) east coast: Kelantan, Terengganu and Pahang, (c) southern states: Johor, Negeri Sembilan and Melaka, and (d) west coast: Perak and Selangor. Six habitat types (Table 1) were determined prior to the survey. The survey was conducted in such way that every habitat type is represented in each geographical group. Survey was carried out by; (a) driving along the main trunk road (federal road) and for every 100 km, a 10 m x 10 m plot was established to asses the mimosa population wherever the geographical condition permitted (data for this is to be published elsewhere), (b) assessment of Mimosa population for wetland habitats type and forest reserve were carried out within the riparian zone and forest fringe, respectively.

Results and Discussion

The survey has shown that Mimosa has spread to all 13 states in Peninsula Malaysia (Figure 1). Generally infestations are recorded at more than 60% of the area surveyed in each habitat type. Results also suggested that construction sites, roadsides and riparian zones are highly susceptible to Mimosa. However, forest reserves were free of Mimosa (Table 1). Major infestation was marked in Figure 1, along with eight forest reserves where no record of Mimosa was made.

Table1: Major site surveyed recorded with Mimosa infestation.

Habitat Type	No. of sites surveyed	No. of sites with infestation	%
Construction site	20	15	75.0
Roadside	20	17	85.0
Riverbank	12	9	75.0
Reservoir	8	5	62.5
Plantation	10	7	70.0
Forest reserve	10	0	0.0
Total	80	53	

Construction sites and roadsides

Comparatively, roadside is more highly invasible by Mimosa than are other habitat types (Table 1). Soil contaminated with its seeds is most likely the source of propagule. The movement of seed banks is enhanced by transporting soil from one construction site to another (Benyasut and Pitt, 1992; Miller and Pickering, 1983). This may also explain its successful establishment along the East-West Highway. Since this highway is one of the main routes linking East and the West coast of Peninsula Malaysia, it is likely that Mimosa seeds are being moved unintentionally, in greater distance. However, populations observed in developing zones with active construction projects are most probably only to survive during the construction period.

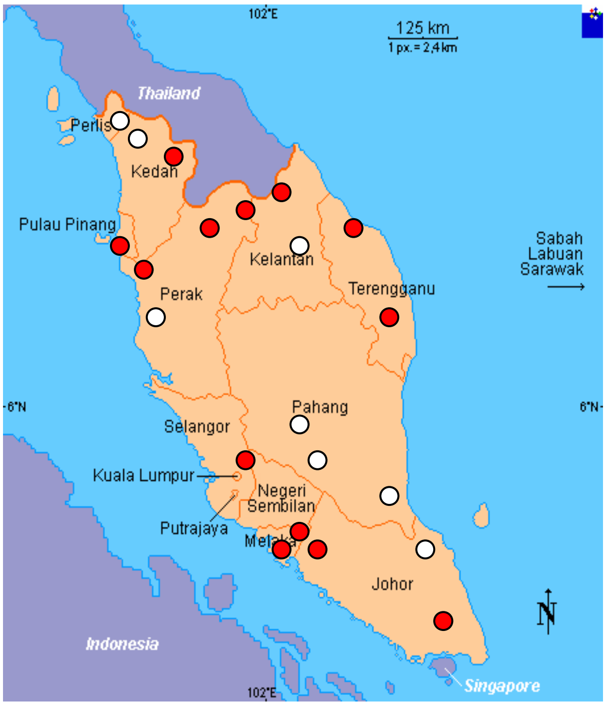

Figure1. Some of major infestation of Mimosa (●) and forest reserves (only eight of ten forest reserves shown here) visited were without Mimosa (○) during the survey

Riverbanks and reservoirs
Wetland habitats especially river corridors, flood plains, lakes and reservoirs face extreme possibility of being invaded by Mimosa as also observed in other tropical countries (Samouth, 2004; Triet, *et al.* 2004b). Five reservoirs have been observed to be infested with Mimosa, namely Pedu (Kedah), Temenggor and Bukit Merah (Perak), Pergau (Kelantan) and Kenyir (Terengganu). Infestations in all these sites are likely due to soil containing Mimosa seeds brought into those areas during construction. The spread of its population however is limited to open flood plain and has not been observed within the forest edge bordering the dams. Newly constructed water canals and riverbanks used as soil dumping sites are also highly prone to Mimosa infestation, which conform to studies by Napompeth (1983) and Mansor (1987). The survey also revealed that waterways located in vicinity of newly developed roads and highways are likely to be infested with Mimosa. Although Mimosa infestation in reservoirs may has not reached an alarming state, as faced by other Southeast Asian countries, e.g. Cambodia (Samouth, 2004) and Vietnam (Triet *et al.* 2004b), preventing possible outbreak must be included as part of the management program.

Agricultural lands and forest reserves
Plantations recorded with Mimosa were young oil-palm plantations and coconut orchards. Plantation areas under constant disturbance (e.g. weed management program) are relatively free of Mimosa. However, as the above crops are much stronger competitor in their later stage, Mimosa infestation is presumably negligible.

Intact forest areas are non-susceptible to Mimosa infestation. However, Mimosa might be able to establish at the forest edges due to large canopy opening and with substantial supply of propagules, although it is very unlikely to survive within the intact forest.

Conclusions

Generally the spread of Mimosa into many parts of Peninsula Malaysia is due to movement of soil with its seed from an infested areas to another as also reported in Australia (Miller, 1983) and Thailand (Napompeth, 1983). In addition, biological control program of Mimosa was most probably failed to materialise although scattered population of seed bruchids can be found especially in Penang and Selangor (personal observation, 2004). The reclassification of Mimosa as a common weed doesn’t reduce its potential of being invasive. As shown, Mimosa is capable to invade successfully into terrestrial ecosystem and no longer restricted to aquatic and semi-aquatic habitats. Thus preventing new invasion should be the first priority for land owners and respective government agencies. The integrated pest management (IPM) which combine several control methods (Paynter and Flanagan, 2004) should be taken into consideration for better result in managing Mimosa infestation.

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WEED CONTROL EFFICACY OF BISPYRIBAC SODIUM + FENOXAPROP-P-ETHYL IN LOWLAND RICE

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Abstract: A field trial was conducted at the Rice Research and Development Institute, Batalagoda, Sri Lanka during dry season 2006 to evaluate the weed control efficacy of bispyribac-sodium (Nominee® T-32 SC) and four herbicide mixtures containing bispyribac-sodium (Nominee® 100 g l^{-1} SC or Nominee® T-32 SC) and fenoxaprop-p-ethyl (Whip super® 7.5 g l^{-1} EW), in lowland rice cultivation. These treatments were compared with un-weeded and hand-weeded controls. Emphasis was given to determine the effectiveness of the above mixtures on *Leptochloa chinensis* and *Isachne globosa*, which could not be controlled with Nominee 100 SC alone. The Nominee® 100 SC + Whip super® 7.5 EW (300 ml + 300 ml ha^{-1}) and Nominee® T-32 SC + Whip super® 7.5 EW (300 ml + 300 ml ha^{-1}) found to be more effective in controlling weeds (grasses, broadleaves and sedges) than the lower concentrations of the above formulations. In addition, these herbicide mixtures effectively controlled *Isachne globosa* and *Leptochloa chinensis.* Plots treated with Nominee® 100 SC + Whip super® 7.5 EW (300 ml + 300 ml ha^{-1}) recorded higher grain yield than those treated with other mixtures. None of the above herbicide mixtures showed any phytotoxicity effect on rice.

Key words: Bispyribac Sodium, fenoxaprop-p-ethyl, tank mixtures, weed control

Introduction

Over the past 4-5 decades, more than 20 herbicides have been recommended for the control of weeds in wet-seeded rice in Sri Lanka. Among these, bispyribac-sodium (Nominee® 100 g l^{-1} SC) became the most widely used early-post emergent one-shot herbicides in rice since late 1990s due to its effectiveness on local weed composition and agro-climate conditions (Abeysekera, 1999; 2000a; 2000b). However, grass weed such as *Leptochloa chinensis* and *Isachne globosa* could not be controlled by the application of Nominee® alone.

Hence, this research was conducted to evaluate the efficacy of herbicide mixtures, namely bispyribac sodium (Nominee® 100 g l^{-1} SC) + fenoxaprop-p-ethyl (Whip super® 7.5 g l^{-1} EW) at ratio 1:1 for the control of grasses, broad leaves, sedges with special emphasis on *L. chinensis* and *I. globosa*, and on growth, and yield of direct-seeded rice.

Materials and Methods

Experimental site

The Experiment was conducted at the Rice Research Development and Institute, (RRDI), Batalagada, Ibbagamuwa, Sri Lanka during 2006 dry season (May - August). The soil type is red yellow podsolic, with a pH of 5.9. The selected experimental site was ploughed two times followed by a single harrowing. Each plot was puddled and leveled separately and demarcated by 30 cm wide bunds to prevent mixing of irrigation water. Recommended doses of nutrients were supplied to the plots. The experimental was conducted in a Randomized Complete Block Design (RCBD) with three replicates. The plot size was 6 m x 3 m. Sprouted seed paddy of variety Bg 300 (90 day-old variety) was broadcasted uniformly at the rate at 140 kg ha^{-1}.

Treatmets

A new formulation of bispyribac sodium (Nominee® T-32 SC) and four mixtures containing different concentrations of bispyribac-sodium (Nominee® 100 g l^{-1} SC or Nominee® T-32 SC) and fenoxaprop-p-ethyl (Whip super® 7.5 g l^{-1} EW) were tested (Table 1). These treatments were compared with un-weeded and hand-weeded controls.

Table 1. Herbicide mixtures selected for testing against weeds in rice

Herbicides	Dosage (formulation, ml ha^{-1})	Dose (g a.i ha^{-1})
Nominee® T – 32 SC	250	25
Nominee® T – 32 SC	300	30
Nominee® T – 32 SC + Whip super® 7.5 EW	250 + 250	25 + 18.75
Nominee® T – 32 SC + Whip super® 7.5 EW	300 + 300	30 + 22.5
Nominee® 100 SC + Whip super® 7.5 EW	250 + 250	25 + 18.75
Nominee® 100 SC + Whip super® 7.5 EW	300 + 300	30 + 22.5
Whip super® 7.5 EW	300	22.5
Hand weeding		-
No wedding		-

Fields were irrigated daily during first 8 days after sowing (DAS) and allowed to drain before the application of herbicides at 12 DAS. The plots were irrigated 3 days after herbicide application. The weed population density and dry weight (grasses, broad leaves and sedges separately) were collected in 30 cm x 30 cm areas in plots at 4th and 6th weeks after sowing (WAS). In addition the phytotoxicity effects of the herbicides were observed visually. At maturity the yield components and yield of rice were collected.

Results and Discussion

Common weeds in the experimental site

The weed flora in the experimental location composed of 43% grasses, 21% sedges and 36% broad leaves (Table 2**).** *Leptochloa chinensis* (15% of grasses), *Cyperus difformis* (6% of sedges) and *Monochoria vaginalis,* (9% of broad leaves) were the most dominant weeds observed in the experimental location.

Table 2. Percentage of initial weed count in the experimental site

Grasses	%	Sedges	%	broad leaves	%
Leptochloa chinensis	14.5	*Cyperus difformis*	6.0	*Monochoria vaginalis*	9.2
Echinochloa crus-galli	7.2	*Cyperus irria*	5.8	*Commalina diffusa*	7.9
Isachne globosa	6.8	*Fimbristylis miliacea*	4.3	*Eclipta alba*	5.9
Eragrostis tremula	4.2	*Fimbritilyis dichotama*	2.4	*Murdannia nudiflora*	4.4
Paspalum distichum	4.3	*Cyperus rotundus*	1.2	*Marselia quadrifolia*	3.4
Ischaemum rugosum	3.9			*Ludwigia octavalvis*	2.2
Echninochloa colona	0.9	Others	1.1	*Aeschynomene indica*	2.1
Others	1.2			Others	1.1
Total	43	Total	20.8	Total	36.2

Effect of herbicides on weed density

At 4 WAS, the weed count showed a significant differences among treatments (Table 3). The un-weeded control recorded the highest weed density ($p<0.05$). The plots treated with

Nominee® T-32 SC or Nominee® 100 SC + Whip super® 7.5 EW at 300 ml + 300 ml ha^{-1} gave the best weed control. The lower rates of Nominee® 100 SC and Whip super® 7.5 EW and the plots treated only with Nominee® T-32 SC or Whip super® gave relatively lower weed control.

Table 3. Weed density (number per m^2) in plots treated with different herbicides and in the control plots at 4 and 6 weeks after sowing

Herbicides	Weed density 4th WAS*	Weed density 6th WAS
Nominee® T - 32 SC 250	93.4 b	60.5 c
Nominee® T - 32 SC 300	44.4 dc	24.0 d
Nominee® T - 32 SC + Whip super® 7.5 EW (250 + 250)	40.1 d	25.7 d
Nominee® T - 32 SC + Whip super® 7.5 EW (300 + 300)	16.0 fe	14.5 de
Nominee® 100 SC + Whip super® 7.5 EW (250 + 250)	25.0 de	12.0 de
Nominee® 100 SC + Whip super® 7.5 EW (300 + 300)	6.6 f	6.7 e
Whip super® 7.5 EW 300	75.1 bc	107.1 b
Hand weeding	0.01 g	0.03 f
No weeding	266.7 a	209.3 a
CV%	39.8	17.5

* WAS – weeks after sowing. Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05). Refer to Table 1 for dosages.

Effect of herbicides on grasses, broad leaves and sedges

At 6 WAS, the density and dry weight of grasses, broad leaves and sedges in plots treated with different herbicides found to be significantly different (Table 4 and 5). The un-weeded control recorded the highest weed count ($p<0.05$) than the treated plots. The plots treated with Nominee® T-32 SC + Whip super® 7.5 EW (300 ml + 300 ml ha^{-1}) and Nominee® 100 SC + Whip super® 7.5 EW (300 ml + 300 ml ha^{-1}) gave the best control of grasses.

Table 4. Density (number per m^2) of grasses, broad leaves and sedges in plots treated with different herbicides and in the control plots at 6 weeks after sowing.

Herbicides	Grasses	Sedges	Broad leaves
Nominee® T - 32 SC 250	29.8 b	10.1 b	13.2 ab
Nominee® T - 32 SC 300	09.0 cbd	05.9 c	08.6 bc
Nominee® T - 32 SC + Whip super® 7.5 EW (250 + 250)	11.4 bc	03.6 cd	09.7 b
Nominee® T - 32 SC + Whip super® 7.5 EW (300 + 300)	04.9 dc	02.9 d	04.8 bcd
Nominee® 100 SC + Whip super® 7.5 EW (250 + 250)	07.0 cbd	02.6 d	02.3 cde
Nominee® 100 SC + Whip super® 7.5 EW (300 + 300)	04.4 dc	0.8 e	0.8 de
Whip super 7.5 EW 300	25.7 bc	22.2 a	25.5 a
Hand weeding	0.01 d	0.01 e	0.01 e
No weeding	161.1 a	20.7 a	26.1 a
CV%	43.9	17. 4	31.9

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05). Refer to Table 1 for dosages.

Effect of herbicides on Isachne globosa and Leptochloa chinensis

At 6 WAS, the number of *Isachne globosa* plants showed a significant difference among treatments (Table 6). The plot treated only with Whip super® 7.5 EW and Nominee® T-32 SC at the rate of 250 ml ha^{-1} had more *Leptochloa chinensis* and *I. globosa*. The un-weeded control recorded the highest *I. globosa* count ($p<0.05$) and hand weeded plot recorded highest weed control efficacy (100%).

Table 5. Dry weight (g per m^2) of grasses, broad leaves and sedges in plots treated with different herbicides and in the control plots at 6 weeks after sowing.

Herbicides	Grasses	Sedges	Broad leaves
Nominee® T - 32 SC 250	10.8 bc	1.27 b	1.38 ab
Nominee® T - 32 SC 300	04.5 dc	0.79 bc	0.82 bc
Nominee® T - 32 SC + Whip super® 7.5 EW (250 + 250)	03.8 dc	0.39 c	0.85 bc
Nominee® T - 32 SC + Whip super® 7.5 EW (300 + 300)	20.8 d	0.27 c	0.27 c
Nominee® 100 SC + Whip super® 7.5 EW (250 + 250)	05.5 bcd	0.33 c	0.01 c
Nominee® 100 SC + Whip super® 7.5 EW (300 + 300)	01.7 d	0.14 c	0.07 c
Whip super® 7.5 EW 300	13.1 b	2.63 a	1.49 ab
Hand weeding	0.01 d	0.01 c	0.01 c
No weeding	38.5 a	2.98 a	2.19 d
CV%	40.6	39.1	36.8

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05). Refer to Table 1 for dosages.

Table 6. Density (number per m^2) of *Isachne globosa* and *Leptochloa chinensis* in plots treated with different herbicides and in the control plots, at 6 weeks after sowing

Herbicides	*Isachne globosa*	*Leptochloa chinensis*
Nominee® T - 32 SC 250	5.90 b	3.2 bc
Nominee® T - 32 SC 300	3.20 b	2.99 bc
Nominee® T - 32 SC + Whip super® 7.5 EW (250 + 250)	3.61 b	2.31 bc
Nominee® T - 32 SC + Whip super® 7.5 EW (300 + 300)	3.20 b	1.17 dc
Nominee® 100 SC + Whip super® 7.5 EW (250 + 250)	3.61 b	2.16 bc
Nominee® 100 SC + Whip super® 7.5 EW (300 + 300)	0.29 c	0.41 dc
Whip super® 7.5 EW 300	5.50 b	6.51 b
Hand weeding	0.01 c	0.01 d
No weeding	16.32 a	26.52 a
CV%	20.14	40.56

Within a column, means followed by the same letter are not significantly different by the DMRT at p=0.05. Refer to Table 1 for dosages.

The plots treated with Nominee® 100 SC 300 ml + Whip super® 7.5 EW 300 ml ha^{-1} (WCE 98 %), Nominee® T-32 SC 300 ml ha^{-1} +Whip super® 7.5EW 300 ml ha^{-1} (WCE 80 %) and Nominee® T-32 SC 300 ml ha^{-1} (WCE 80%) recorded good control of *I. globosa*. At 6 WAS the number of *L. chinensis* showed a statistically significant difference among the treatments (Table 6). The un-weeded control recorded the highest *L. chinensis* count ($p<0.05$) and hand-weeded plot recorded highest weed control efficacy (100%). The plots treated with Nominee® 100SC 100 ml ha^{-1} + Whip super® 7.5EW (WCE 98 %), Nominee® T-32 SC 300 ml ha^{-1} + Whip super® 7.5 EW (WCE 96%) recorded good control of *L. chinensis*. Increasing the dosage of the tank mixture enhanced the efficiency of control of *L. chinensis*. Plots treated only with Whip super® 7.5 EW had higher number of *L. chinensis* and recorded a lower weed control efficacy (77%). Plots received only Nominee® T-32 SC 250 ml ha^{-1} did not give a satisfactory control of weeds, including *L. chinensis*. It is important to note that the selection of *L. chinensis* in rice fields treated with Nominee® 100 SC has been observed in the recent past in different part of Sri Lanka, giving arise to the dominance of this weed in rice fields that are continuously treated with the herbicide.

Influence of herbicides on grain yield

The grain yield was significantly different ($p<0.05$) among treatments (Table 7). The un-weeded control recorded the lowest yield while the highest was recorded in hand weeded control. Plots treated with Nominee® T-32 SC + Whip super® 7.5 EW (300 ml + 300 ml ha^{-1}) and Nominee® 100SC + Whip super® 7.5EW (300 ml + 300 ml ha^{-1}) gave a comparatively higher yield.

Table 7. Estimated rice yield (t/ha) in plots treated with different herbicides and in the control plots at 6 weeks after sowing

Herbicides	Yield (t/ha)
Nominee® T - 32 SC 250	4.12 b
Nominee® T - 32 SC 300	4.35 b
Nominee® T - 32 SC + Whip super® 7.5 EW (250 + 250)	4.25 b
Nominee® T - 32 SC + Whip super® 7.5 EW (300 + 300)	4.50 ab
Nominee® 100 SC + Whip super® 7.5 EW (250 + 250)	4.14 b
Nominee® 100 SC + Whip super® 7.5 EW (300 + 300)	4.84 ab
Whip super® 7.5 EW 300	4.35 b
Hand weeding	5.30 a
No weeding	4.35 b
CV%	11.61

Means followed by the same letter are not significantly different by the DMRT at p=0.05. Refer to Table 1 for dosages.

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EVALUATION OF ERIJAN® 400 EW, A NEW HERBICIDE MOLECULE, AGAINST GRASSES AND SEDGES IN TRANSPLANTED *KHARIF* RICE

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Abstract: The loss of rice yield occurs from 25 to 30 % due to unchecked weed growth in transplanted paddy. Keeping this in view, a field experiment was conducted in Gangetic New Alluvial soil with a pH of 6.8 and sandy loam in texture of the ‘C’ – Block Farm, Bidhan Chandra Krishi Viswavidyalaya, Nadia ,West Bengal, India during *Kharif*-2006 with the objective of determining the effective dose and suitable method of application of herbicide Erijan 400 EW along with the bio-efficacy and phytotoxicity of the herbicide. The experiment was laid out in a Randomised Complete Block Design having ten treatments and replicated thrice. Herbicide Pretilachlor (Erijan® 40 EW) was applied at three different doses (1250, 1500 and 1850 ml ha^{-1}) at 3 days after transplanting (DAT) through spraying by knapsack sprayer with floodjet deflector WFN 040 nozzle using spray volume at 500 l ha^{-1} and through splash bottle with 6 l ha^{-1} water volumes. The standard herbicides Pretilachlor (Rifit® 50 EC) and Butachlor (Machete® 50 EC) were applied at 3 DAT as spray application, and Oxydiargyl (Topstar 80® WP) was applied at 3 DAT as a splash application under drained conditions. The herbicide treated plots were re-flooded at 2 days after application (DAA) of the herbicide and water was maintained thereafter. The results revealed that Erijan® at 1875 ml ha^{-1} as splash application and Erijan® at 1500 and 1875 ml ha^{-1} as spray application were most effective against the annual grass and sedges in transplanted rice. Spray application of the testing herbicide Erijan performed better as compared to the splash application and this did not cause any phytotoxicity to the rice plant.

Key words: New herbicide molecule, pretilachlor (Erijan® 400 EW), rice, weeds

Introduction

Rice (*Oryza sativa*) is the most important and staple food crop in India. The slogan ‘Rice is life’, is most appropriate for India as this crop plays a vital role in food security and is a means of livelihood for millions of rural households. Weeds no doubt, are considered as the major pest, which stands in the way of increasing rice production (Labrada, 1996). Weeds compete with the rice crop for the same resources, namely mineral nutrients, water, space, light and sometimes serve as an alternate host of many diseases and insect pests and thereby reduce the yield of rice. The extent of yield reduction due to weeds alone is estimated to be around from 25 to 30 % in transplanted paddy (Upadhyay and Gogoi, 1993). The most critical period of crop-weed competition in rice is between four and six weeks after transplanting (Gill and Kolar, 1980). So timely weed management is very important for getting a higher yield. Application of herbicide is easy, rapid and more effective for controlling weeds over cultural and mechanical methods in rice culture (Bharadwaj and Verma, 1969). Continuous application of the same herbicides to the same field develops resistance in weeds to these herbicides. So, different types of new molecules and formulations are needed for the efficient weed control as well as to sustain production.

Materials and Methods

The field experiment was conducted in the Gangetic *Inceptisol* having a sandy loam texture, a pH 6.8 and moderate fertility status of the ‘C’ – Block Farm, Bidhan Chandra Krishi Viswavidyalaya, West Bengal, India during Kharif-2006 to evaluate Pretilachlor (Erijan® 400 EW), a new herbicide molecule, against grasses and sedges in transplanted *kharif* rice. During the experiment, ten treatments were randomly allocated in Randomised Complete Block Design and replicated thrice. The treatments were untreated control, Pretilachlor (Erijan® 40 EW) at three different doses (1250, 1500 and 1850 ml ha^{-1}) applied at 3 days after transplanting (DAT) through spraying and through splash bottle, Pretilachlor (Rifit® 50 EC) and Butachlor (Machete® 50 EC) as standard through spray application and Oxydiargyl (Topstar® 80 WP) at 3 DAT as splash application .The spraying was done by knapsack sprayer with floodjet deflector WFN 040 nozzle using a spray volume at 500 l ha^{-1} and through splash bottle with 6 litre ha^{-1} water volume. Twenty one day old seedlings of rice cv. *Khandagiri* (IET 10397) were transplanted with a plot size of 5 m x 4 m having a spacing of 20 cm x 15 cm and were fertilized with one fourth of the Nitrogen @ 60 kg ha^{-1} through Urea along with full Phosphorus at 30 kg ha^{-1} through Single Super Phosphate and full doses of Potash @ 30 kg ha^{-1} through Muriate of Potash were applied as basal during final land preparation and he remaining Nitrogen was top dressed in 2 splits *i.e.* half of N at active tillering and the rest one fourth of N at panicle initiation stage of the crop. All the recommended improved package of practices was followed uniformly to raise the crop.

Visual observations on the efficacy of the applied herbicides were taken at 7, 15, 30 and 45 DAA (days after application). The density and dry weight of grasses and sedges were recorded separately at 7, 15, 30 and 45 DAA of the herbicide with the help of a 50 cm x 50 cm quadrat and were converted to m^{-2}. Phytotoxicity scoring was done on the basis of a rating scale (PRS) for all the herbicides. The parameters on phytotoxicity were taken as necrosis of leaf tips and margins, wilting, vein clearing, necrosis, epinasty and hyponasty.

Results and Discussion

Weed flora

The predominant weed flora present in the experimental field were, grasse weeds such as *Echinochloa colona*, *E. crus-galli*, *E. formosensis*, *Eleusine indica*, *Leersia hexandra*, sedges such as *Cyperus difformis, C. iria*, *Frimbristylis dichotoma* and broadleaves such as *Alternanthera philoxeroides, Blainvillea latifolia, Eclipta alba, Ludwigia parviflora* , *Stellaria media*, *Lindernia ciliate*, *Marselia quadrifolia* and *Oldenlandia corymbosa*. Sahu and Mandal (1966) have made similar observations.

Effect on weeds

All treatments reduced both the population and dry weight of weeds over the untreated control (Tables 1 and 2). All the herbicidal treatments reduced the population and dry weight of grasses significantly than the untreated control in all dates of observation except for Erijan® at 1250 ml ha^{-1} at 7 DAA. Spray application of Rifit® at 1250 ml ha^{-1}, Erijan® at 1875 ml ha^{-1} and Machete® at 2500 ml ha^{-1} recorded minimum grass weed populations throughout the observation period .This efficiency of machete was supported by Bhanu Rekha *et al.* (2002). There was a gradual decrease in dry weight of grass weeds when the dose of Erijan increased from 1250 to 1875 ml ha^{-1}. Splash application of Erijan® was less effective than the same as spray application. Erijan® at 1250 ml ha^{-1} as splash application and Machete as spray application was not effective against the sedge weeds. Erijan® at 1500 and 1875 ml ha^{-1} along with the Rifit® at 1250 ml ha^{-1}, all as spray application produced the minimum population and dry weight of sedges closely followed by Topstar® at 125 g ha^{-1} as splash application. Erijan® 400 EW performed better when applied as a spray application as compared to a splash application and the effectiveness of Erijan® 400 EW increased with doses against the grass weeds.

Table 1. Effect of different treatments on population and densities of total grassy weeds at different times after application

Treatment	Dose (a.i. ha^{-1})	Method	Population (number) per m^2				Dry weight (g) per m^2			
			7 DAA	15 DAA	30 DAA	45 DAA	7 DAA	15 DAA	30 DAA	45 DAA
Check (Untreated control)	-	-	4.00	36.00	90.67	104.7	0.12	2.04	8.77	9.75
Pretilachlor (Erijan® 400 EW)	500	Splash	2.00	25.33	51.33	57.33	0.14	1.36	5.41	6.91
Pretilachlor (Erijan® 400 EW)	600	Splash	2.33	24.67	29.33	35.67	0.07	1.04	2.69	3.55
Pretilachlor (Erijan® 400 EW)	750	Splash	0.00	20.33	34.67	42.67	0.00	1.88	4.50	4.13
Pretilachlor (Erijan® 400 EW)	500	Spray	0.00	12.67	38.67	50.67	0.00	0.72	5.01	5.40
Pretilachlor (Erijan® 400 EW)	600	Spray	1.00	13.67	42.67	54.00	0.02	0.48	3.27	4.05
Pretilachlor (Erijan® 400 EW)	750	Spray	0.00	16.67	29.33	30.33	0.00	0.80	2.71	2.94
Pretilachlor (Rifit® 50 EW)	625	Spray	1.33	15.00	24.67	26.00	0.04	0.84	2.62	3.07
Butachlor (Machete® 50 EC)	1250	Spray	0.00	22.67	30.33	30.33	0.00	1.04	3.35	6.56
Oxydiargyl (Topstar® 80 WP)	100	Splash	2.00	30.00	48.00	35.67	0.10	0.56	3.76	6.35
S. Em (±)			0.443	3.350	5.678	5.282	0.032	0.182	0.449	0.517
C.D. (p=0.05)			1.316	9.953	16.87	15.69	0.095	0.542	1.333	1.536

Table 2. Effect of different treatments on population and densities of total sedge weeds at different times after application

Treatment	Dose (a.i. ha^{-1})	Method	Population (number) per m^2				Dry weight (g) per m^2			
			7 DAA	15 DAA	30 DAA	45 DAA	7 DAA	15 DAA	30 DAA	45 DAA
Check (Untreated control)	-	-	9.33	80.33	240.67	276.00	1.23	10.44	19.84	31.76
Check (Untreated control)	500	Splash	10.33	76.33	187.67	180.00	1.14	7.72	16.76	20.37
Pretilachlor (Erijan® 400 EW)	600	Splash	4.67	56.00	124.33	215.67	0.56	8.44	17.02	19.87
Pretilachlor (Erijan® 400 EW)	750	Splash	0.00	60.33	114	169.33	0.00	6.68	16.56	18.65
Pretilachlor (Erijan® 400 EW)	500	Spray	2.00	56.33	140.67	163.00	0.23	6.24	14.32	19.31
Pretilachlor (Erijan® 400 EW)	600	Spray	0.00	38.67	112.67	128.67	0.00	4.96	11.74	15.09
Pretilachlor (Erijan® 400 EW)	750	Spray	1.33	34.67	56.00	97.67	0.32	4.44	5.95	9.63
Pretilachlor (Erijan® 400 EW)	625	Spray	2.00	26.33	64.33	90.67	0.35	2.78	4.02	8.93
Pretilachlor (Rifit® 50 EW)	1250	Spray	5.67	54.00	96.33	161.33	0.86	9.16	14.34	17.56
Butachlor (Machete® 50 EC)	100	Splash	0.00	32.67	68.00	106.33	0.00	2.34	5.36	12.11
Oxydiargyl (Topstar® 80 WP)			1.550	5.935	15.129	20.812	0.145	0.753	2.078	2.168
S. Em (±)			4.604	17.63	44.945	61.829	0.431	2.236	6.174	6.441

Effects of treatments on crops

The testing herbicide did not show any phytotoxicity symptoms on leaf tips, leaf wilting, vein clearing, epinasty and hyponasty symptoms of paddy leaves. The minimum grain yield of 2567 kg ha^{-1} was recorded against the untreated control, which was 23.3% lower than the average yield of the herbicidal weed management (Table 3). The maximum yield of 3288 kg ha^{-1} was recorded in Erijan® at 1875 ml ha^{-1} which was significantly higher (28.1%) than the untreated control. The data on straw yield also recorded a similar trend as grain yield.

From the above findings, it may be concluded that Erijan® at 1875 ml ha^{-1} as splash application, Erijan® at 1500 and 1875 ml ha^{-1} as spray application were most effective against the annual grass and sedges in transplanted rice. Spray application of the testing herbicide Erijan performed better as compared to the splash application. The testing herbicide Erijan® did not cause any phytotoxicity to the rice plant.

Table 3 Effect of treatments on grain and straw yield (kg ha^{-1}) of transplanted rice

Treatment	Dose (a.i. ha^{-1})	Method	Grain yield	Straw yield
Check (Untreated control)		-	2567	2960
Pretilachlor (Erijan 400 EW)	500	Splash	2950	3583
Pretilachlor (Erijan 400 EW)	600	Splash	3146	4207
Pretilachlor (Erijan 400 EW)	750	Splash	3212	4133
Pretilachlor (Erijan 400 EW)	500	Spray	3128	4033
Pretilachlor (Erijan 400 EW)	600	Spray	3099	4238
Pretilachlor (Erijan 400 EW)	750	Spray	3288	4677
Pretilachlor (Rifit 50 EW)	625	Spray	3263	4473
Butachlor (Machete 50 EC)	1250	Spray	3213	3997
Oxydiargyl (Topstar 80 WP)	100	Splash	3190	3727
S. Em (±)			133.7	245.6
C.D. (p=0.05)			397.3	729.6

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STUDIES ON THE DISSIPATION OF CYHALOFOP- BUTYL, A NEW RICE HERBICIDE IN PADDY UNDER WEST BENGAL CLIMATIC CONDITIONS

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Abstract: Cyhalofop-butyl (a member of aryloxyphenoxy propionate group) is a post-emergent herbicide newly-introduced in India by Dow Agro Sciences Pvt. Ltd., Mumbai. It selectively controls barnyard grasses and silver top grasses in transplanted paddy. It is highly effective in the seed bed of paddy. A long-term, three season residue study of Clincher (Cyhalofop-butyl, 10 EC) was conducted from 2003 to 2004 at BCKV, Kalyani. Clincher was applied at 40 g a.i./ha (T_1), 80 g a.i./ha (T_2) and 160 g a.i./ha (T_3) along with untreated control, each treatment being replicated thrice. The plot size was 25 m^2. Soil samples were collected at 0, 15, 30 and 60 days after herbicide application and at harvest. Grain and straw samples were collected at harvest. The samples were extracted with aqueous acetone (9:1) followed by partitioning with dichloromethane and the organic fraction after collection was dried in rotary vacuum evaporator at 40^0C. The residue was finally reconstituted in methanol for GC analysis using ECD with the following operating parameters: Column DB - 5, 30 mt x 0.53 mm i.d., 5 μm thickness; oven temperature 230°C, injector temperature 250^{o}C, detector temperature 280^{o}C; carrier gas flow 40 ml/min. The Limit of Detection (LOD) and Limit of Quantification (LOQ) were found to be 0.01 ppm and 0.05 ppm, respectively. The study revealed that no herbicide residue was detected in harvested grain, straw and soil samples irrespective of treatment doses and seasons. From the study it was evident that the dissipation pattern of Cyhalofop-butyl in soil follows first order kinetics. The half-life in soil varies from 7.92-21.50 days irrespective of treatment doses and seasons. From the above study, it may be concluded that Cyhalofop-butyl may not pose any residue problem in harvested paddy.

Key words: Cyhalofop-butyl, residue, paddy.

Introduction

Rice (*Oryza sativa*) is the main crop cultivated in Asian countries, especially in India. India has the highest area under rice and it is known that weeds are one of the main causes of yield loss of rice due to their competition for nutrients, water light etc. *Echinochloa crus-galli is* the most common grass weed in rice (Scott, 2002). Among 40 million ha of rice fields infected with pests, only 2 million ha of rice fields are treated with herbicides and only 30% of cropped area is under agrochemical usage (Majumdar, 1994). Among the different groups of synthetic pesticides, herbicides are the most widely used and constitute 55% of the world's pesticide market (Rao, 2000). Cyhalofop-butyl is a new member of aryloxyphenoxy propionate group, first introduced to India by Dow Agro Sciences Ltd. Cyhalofop-butyl is highly effective post-emergent herbicide in controlling paddy weeds, especially barnyard grasses and silver top grasses in transplanted paddy without causing phytotoxicity to rice. It has an excellent herbicidal activity and is required at a very low dose. Aims of the present experiment were to evaluate the dissipation pattern of Cyhalofop-butyl in paddy soil under waterlogged condition and to determine the residue in paddy straw, grain and husk at harvest under West Bengal conditions.

Materials and Methods

Cyhalofop- butyl (10% EC formulation, Clincher and 99.5% pure analytical standard) was supplied by M/S Dow Agro Sciences Ltd, Mumbai. All solvents used in this experiment were of analytical grade and purchased from E. Merck, Qualigens and Spectrochem. The experiment was conducted on the high yielding variety Khitish (IR 4074) in the Experimental Research field of BCKV for three consecutive seasons during 2003-04. Cyhalofop-butyl was applied at 40 g a.i./ha (T_1), 80 g a.i./ha (T_2) and 160 g a.i./ha (T_3) as post emergence. Fertilizers were applied as per recommended dose. Each treatment was replicated thrice in a randomized block design.

Soil samples were collected from each treated replicated plot after 0 (2 h), 15, 30 and 60 days after application and also at the time of harvest. Harvested straw, grain and husk samples were also taken for residual analysis. Soil samples (100 g) from each replicated plot along with the untreated control were taken in 250 ml conical flasks and 100 ml of acetone:water (9:1) was added and kept overnight. Then the extracts were shaken in mechanical shaker for 1 h. The soil extract was filtered through a Buchner funnel and washed with 50 ml (2 x 25 ml) of the same extracting solvent. The filtrate was then concentrated in a rotary vacuum evaporator after addition of 150 ml of saturated aqueous NaCl partitioned with 200 ml (100+50+50) of dichloromethane. The dichloromethane layer was then evaporated to dryness in a rotary vacuum evaporator at 40^{o}C and then the residue was reconstituted in distilled methanol for GLC analysis. The residue from grain (100 g), straw (25 g) or husk (25 g) samples were extracted by the Soxhlet apparatus with 300 ml of acetone for 6 h. The acetone extract was dried in the rotary vacuum evaporator at 40^{o}C. Saturated aqueous NaCl (150 ml) solution was added to the dried extract and was partitioned in a separatory funnel with 200 ml (100+50+50) of dichloromethane. Then the same procedure was followed as mentioned for the soil samples. Final volume was made up with distilled methanol for GLC analysis.

Cyhalofop- butyl residue was estimated with the GLC (Model Chemito 1000) coupled with ECD detector using Chemito 5000 data processor with the following operational parameters: column DB- 5, 30 mt x 0.53 mm id, film thickness 5 µm, Oven temperature 230^{o}C, Injector temperature 250^{o}C and Detector temperature 280^{o}C. Carrier gas flow was 40 ml/min. A stock solution of Cyhalofop-butyl (100 ppm) was prepared in acetonitrile as the standard. The retention time of Cyhalofop-butyl was found to be 5.75 min. The retention time of Cyhalofop-butyl occurring in soil, grain, straw and husk were compared with that of external standard, and the data were recorded in a Chemito 5000 data processor.

The efficiency of extraction and clean up method was verified by fortifying different substrates with known quantities of the analytical standard to give final concentrations of 1, 5 and 10 ppm. The areas under GLC peaks were plotted against different concentrations of the herbicide to draw the linearity curve. The limit of detection was found to be 0.01 ppm and limit of quantification as 0.05 ppm. The average recovery was found to be in the range of 83-92% for different substrates.

Results and Discussion

More than 60% of the initial deposit of Cyhalofop- butyl was dissipated within 15 days of application. For soil, the calculated half-life varied from 7.92 – 21.50 days (Table 1). In paddy field soil, the initial deposit varied from 0.71 to 1.08 ppm for T_1, 1.25 to 1.81 ppm for T_2 and 1.93 to 2.54 ppm for T_3 irrespective of any season and no residues were detected in the soil at 60 days or at harvest. Post-harvest residue was not detected in paddy straw, grain or husk samples irrespective of treatment doses and season of experiment. Thus, from the results it was evident that the dissipation of Cyhalofop-butyl residue follows first order kinetics in paddy field soil and appeared to be fairly rapid.

The faster dissipation of Cyhalofop-butyl (Table 2) suggested a minimum chance of ground water contamination. The initial deposit and half-life values varied in the three seasons. The higher deposition of the residue in paddy field soil in season II (Boro) may be due to less rainfall as compared to Kharif seasons (season I and III). Since no residue was detected in the harvested soil samples it can be concluded from the study that the application of Cyhalofop- butyl will not result in built up of Cyhalofop- butyl residue in the soil. As no residue was detected in the harvested grain, straw or husk samples of all three seasons, it can be safely stated that the use of Cyhalofop-butyl may not pose any residual problem in rice ecosystems under West Bengal conditions.

Table 1. Dissipation of Cyhalofop-butyl in paddy soil

Treatment	Season	Days					
		0	5	10	15	30	60
		Residue (ppm) (% dissipation)					
T_1	Season I	0.95±0.04 (-)	0.79±0.09 (16.84)	0.72±0.10 (24.21)	0.55±0.12 (42.10)	0.06±0.04 (93.68)	BDL (-)
	Season II	0.89±0.05 (-)	0.81±0.06 (8.99)	0.54±0.09 (39.33)	0.25±0.04 (71.91)	0.07±0.03 (92.35)	BDL (-)
	Season III	0.79±0.06 (-)	0.68±0.06 (13.92)	0.51±0.10 (35.44)	0.22±0.04 (72.15)	0.06±0.03 (92.41)	BDL (-)
T_2	Season I	1.79±0.10 (-)	1.59±0.11 (11.17)	1.18±0.25 (34.08)	0.54±0.10 (69.83)	0.11±0.05 (93.85)	BDL (-)
	Season II	1.90±0.12 (-)	1.58±0.09 (16.84)	1.14±0.10 (34.74)	0.57±0.09 (70.00)	0.15±0.06 (92.10)	BDL (-)
	Season III	1.99±0.13 (-)	1.49±0.13 (25.13)	1.05±0.06 (47.24)	0.52±0.06 (73.87)	0.23±0.04 (73.87)	BDL (-)

BDL = Below Detection Limit

Table 2. Regression equation and half-life of cyhalofop-butyl in paddy soil.

Treatment	Season	Regression equation	Half life ($T_{1/2}$) (days)
T_1	Season I	Y = 3.139 – 0.041X	7.34
	Season II	Y = 3.039 – 0.039X	7.72
	Season III	Y = 2.987 – 0.040X	7.52
T_2	Season I	Y = 3.061 – 0.028X	10.75
	Season II	Y = 3.243 – 0.030X	10.03
	Season III	Y = 3.262 – 0.016X	18.81
T_3	Season I	Y = 3.246 – 0.024X	12.54
	Season II	Y = 3.403 – 0.026X	11.58
	Season III	Y = 3.371– 0.014X	21.50

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BEHAVIOR OF DITHIOPYR IN SOIL AND ITS EFFECTS ON ESTABLISHMENT, GROWTH, AND DEVELOPMENT OF TURFGRASSES

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Abstract: Dithiopyr, a pyridine class of herbicide, is used for selective control of weeds in established cool-season and warm-season turfgrasses. It enters to the plant system through developing roots or shoots, and can be absorbed by the plants any time between germination and seedling emergence. Dithiopyr stops plant cells from dividing and elongating in the meristematic regions. Its activity is most visible at the root tips of the susceptible plants. Dithiopyr is strongly absorbed to soil colloids and has low tendency for leaching. The potential movement of dithiopyr in runoff water is also low due to low water solubility (1.38 mg/kg) and adsorption (K_d = 3.97 to 5.78 in alluvial soil) to soil colloids and turfgrass. Dithiopyr has a short to moderate persistence (4 to 49 days half lives in turfgrasses, and 11.9 to 12.9 days in alluvial soil), and is degraded primarily by soil microorganisms. Chemical degradation of dithiopyr is also common in natural systems. Other losses can occur through volatility and to a lesser extent by photodegradation (UV light). Dithiopyr may cause initial shoot injury to turfgrass in the form of thinning, yellowing of leaf tip, leaf die-back, and stunting of growth. It can reduce initial rooting of newly seeded turfgrass or installed sod of both cool-season and warm-season turfgrass species. Proper timing and rate of dithiopyr application helps in better establishment of turfgrass from seed by reducing weed competition, particularly from *Digitaria* species. Dithiopyr can safely be used for weed control in established turfgrass environments.

Key words: Turfgrass, persistence, soil adsorption, microbial degradation

Introduction

Dithiopyr (S,S´-dimethyl 2-difluoromethyl -4- isobutyl -6- trifluoromethyl pyridine-3,5-dicarbothioate) is a selective herbicide, recommended for annual grass and broadleaf weed control in turfgrass and golf courses, landscape plants, horticulture and ornamental plants, agricultural crops like wheat, barley, and rice (Gupta and Gajbhiye 2002; Johnson 1997; Bhowmik and Bhanson, 1989). The herbicide is effective when applied as pre- or early post-emergence against major grasses and small seeded broadleaf weeds (Saikia and Kulshrestha 2002). In United States, dithiopyr is registered for use in the control of crabgrass (*Digitaria* spp.) in turfgrass (including greens) (Hong and Smith 1996). Due to the increasing concern for the environmental fate of herbicides and their residues in turfgrass system, it is important to understand the overall soil behavior of this herbicide, and its effect on establishment and growth of turfgrass. The goal of this presentation is to provide a comprehensive review of the behavior and persistence of dithiopyr in soil, along with its influence on turfgrass establishment.

Mode of Action

Dithiopyr is absorbed through the crowns, roots and shoots of plants. It is readily absorbed by roots and to a lesser degree by shoots. Dithiopyr accumulates in meristematic regions, but does not translocate. The major sites of physiological activity of dithiopyr are meristems of roots and shoots of susceptible plants. Dithiopyr inhibits cell division, particularly mitosis. It may interact with a microtubule-associated protein and/or microtubule-organizing centers, altering microtubule polymerization and stability rather than interacting with the dimeric protein tubulin.

Dithiopyr inhibited root elongation and promoted swelling of meristematic zones. With extended exposure, dividing cells were arrested in prometaphase, and reduced numbers of spindle microtubules were found in treated wheat (*Triticum aestivum* L.) root tip cells (Molin *et al*. 1988; Armbruster *et a*l. 1988). Armbruster *et al*. (1991) studied effects of dithiopyr on cell division in wheat root tips. They found that mitotic index of root tips increased from 2.5 to 12% with mitotic cells arresting in late prometaphase. Their studies suggested that dithiopyr did not alter accumulation of cells in prometaphase. The application of dithiopyr elicited morphological changes similar to phytotoxic reactions to dinitroanilines herbicides (pendimethalin, and prodiamine) and pronamide (amide herbicide).

Soil Behavior and Persistence

Volatilization and photodegradation

Many herbicides are susceptible to volatilization and photodegradation. Volatilization and photodecomposition losses can decrease herbicide efficacy. Dithiopyr has vapor pressure of 5.3×10^{-7} kPa. Pesticides with vapor pressures $\geq 5.2 \times 10^{-7}$ kPa at 25˚C have been classified as moderately to highly volatile in field (Schleicher et al. 1995). Herbicide broke down or degraded by sunlight, specifically, the ultraviolet portion of sunlight is responsible for losses of herbicides due to photodecomposition. Hong and Smith (1996) studied the effects of UV radiation on dithiopyr degradation in rooting media (RM) leachate collected from established golf course greens. They found greater degradation of dithiopyr in RM leachate under UV radiation than in dark. Their studies revealed half-lives for dithiopyr in RM leachate ranged from 515 days (sterile RM leachate, dark, 6.8 pH, 20˚C) to 0.8 days (non-sterile RM leachate, UV light, 6.8 pH, 20˚C). Mueth and Cowell (1990) also observed that dithiopyr residues declined rapidly during the first 3 days following application to turfgrass. Higgins (1991) suggested photolysis as a primary route of dithiopyr degradation. However, later studies showed that dithiopyr losses could rarely occur through volatilization and to a lesser extent by photodegradation.

Sorption

Sorption behavior is combination of both adsorption as well as absorption. In general, sorption capacity of an herbicide molecule increases with increasing amount of soil organic matter. The low water solubility (1.38 mg/kg), high octanol-water partition coefficient (K_{ow} = 56,250), and organic carbon partition coefficient (K_{oc} = 1920) suggest a high potential of dithiopyr retention within the thatch, mat, and surface soil (Schleicher *et al*. 1995). Sorption and mobility of dithiopyr in golf course greens rooting medium (RM) were studied by Hong and Smith (2001). The sorption increased from 20 to 27˚C at 24 hours after treatment. The equilibrium of the sorption of dithiopyr was reached 48 hrs after treatment at 20˚C. The golf course greens rooting medium was composed of 80% sand, and 20% sphagnum peat moss (v/v) (96.83:3.17 w/w), which is specific for 'Tifdwarf' Bermudagrass [*Cynodon dactylon (L.) Pers.x C. transvalensis Burtt-Davy*].

Hong and Smith (2001) studied the sorption of dithiopyr on peat moss, RM and sand. The N value ranged from 1.42 to 1.48 for peat moss and RM and 1.038 to 1.385 for sand. The sorption capacity decreased in the following order peat moss > RM > sand. The fraction of dithiopyr sorbed to sand was much lower than peat moss and RM. These results infer that peat moss should be an integral part of the RM to avoid leaching loss of herbicides. They found that the sorption capacity decreased with increased in pH of the system. This may be due to the fact that at low pH the peat moss and sand materials are in a hydrogenated form, which favors lipophilic organic materials to sorb. In sand, dithiopyr sorption had not varied much with concentrations of 4 and 40 μg/L. For RM, dithiopyr sorption capacity was not changed with change in concentration from 40 to 401 μg/L, but sorption capacity in 4 μg/L concentration varied with pH. They also found that dithiopyr sorption increased with increasing temperature. This may be due to higher amount of diffusion starting from the liquid phase to the solid phase and also along the surface of the liquid phase. Agitation of the solution also decreased the sorption of dithiopyr, which implies sorption of dithiopyr occurred through diffusion.

A study conducted by Gupta and Gajbhiye (2002) showed that 87 to 93% of the dithiopyr was recovered during adsorption-desorption studies. They observed strong adsorption of dithiopyr in the alluvial soil with K_d (distribution coefficient) ranging from 3.97 to 5.78, and Freundlich capacity factor K_f value of 2.41. The strong adsorption of dithiopyr was evident from the hysteresis effect observed during desorption. The H value (H= amount sorbed/amount desorbed) ranged from 0.17 to 0.40. The persistence study was carried out under field capacity moisture and submerged condition. This study indicated that dithiopyr persisted beyond 90 days with half-life varying from 11.5 to12.9 days under different moisture regimes.

Leaching characteristics

Generally, dithiopyr is immobile in soil. Hong and Smith (2001) examined the leaching loss of dithiopyr in the rooting medium of a golf course green. The RM leachate was collected from RM lysimeters located under the well-established *C. dactylon* sod maintained similarly to the golf course greens in the southwestern parts of United States. The pH of the leachate obtained was 5.5. The lysimeter study indicated that more than 90% of the added dithiopyr remained in the top 10 cm of the lysimeter and very little amount of dithiopyr was present at the depths below 35 cm. The concentration of dithiopyr in lysimeter effluent was less than 3.5 μg/L. The fraction of sorbed dithiopyr on the RM from the aqueous phase was greater than 89% after 6 min. Results from these experiments indicated that dithiopyr is quite immobile in golf course greens RM, and has minimal potential for movement into surface water, drainage or ground water.

Another study on the leaching potential of dithiopyr in the alluvial soils of India was conducted by Gupta and Gajbhiye (2002). The leaching experiment was carried out in a packed column with analytical grade, and EC formulation of dithiopyr and 1000 μg of dithiopyr was applied. About 98 to 98.6% dithiopyr was recovered from the soil in the column and leachate. The remaining 1.4 to 2.0% was lost during the experiment. The analysis of soil column and the leachate indicated that major portion of dithiopyr remained in the soil. The distribution of dithiopyr in soil column at different depths showed that dithiopyr is very immobile even after passage of 2.5 liter of water (equivalent to 65 cm of rainfall). From the above results, it is quite evident that the mobility of dithiopyr through the soil profile is very minimal. The mobility will decrease even further with increasing amount of organic matter present in the top layer of soil profile.

Degradation and dissipation in soil

Microbial degradation of dithiopyr by bacteria and fungus has been known to occur. Saikia and Kulshrestha (2002) observed that soil microflora imparts a major role in the degradation of dithiopyr. Degradation by both bacterial and fungal communities followed a first order kinetics and bacterial degradation ($t_{1/2}$=5.8) was faster than the fungal degradation ($t_{1/2}$=6.8). Hong and Smith (1996) also found similar results in rooting medium of golf course greens. They observed a short dithiopyr half-life at pH 6.8 and 20 or 30˚C, which indicated biological degradation. They concluded that degradation of dithiopyr by chemical and biological systems appears to be equally as important at the pH of 6.8.

The dissipation of dithiopyr in the soil after pre- and post-emergence application (at 100 and 200 g ai ha^{-1}) to *T. aestivum* was studied by Saikia and Kulshrestha (2002). They observed a 33% decline in the dithiopyr concentration in upper soil horizon in 10 days. The rates of dissipation were very similar for both the application rates during the initial stage of application (about 43% losses at 15 days), but relatively higher for the higher application rate. The dissipation was 91.2% for the lower rate and 96% for the higher rate after 150 days. Persistence of the herbicide in the soil may be due to the sorption on soil colloids, which reduces dissipation properties. They found a similar trend in the post-emergence treatments of dithiopyr. These studies revealed that the decay of dithiopyr from the soil follows a first order kinetics for first two months in case of preemergence application and for the entire study period for the post-emergence application. The half-life of dithiopyr ranged between 17.3 and 25 days.

Dissipation of dithiopyr in perennial ryegrass (*Lolium perenne* L.) was monitored by Schleicher *et al*. (1995). They estimated the half-life for dithiopyr in soil under turfgrass at Mead, Nebraska, to be 35 days. More herbicide was present in upper level of turfgrass profile (verdure + thatch + mat) than in underlying soil (0 to 5cm + 5 to 10cm). They observed that verdure and thatch intercepted much herbicide during application. Adams and Cowell (1990) reported a range of half-lives for dithiopyr, in soil under turfgrass observed over a range of locations across the United States, to be from 4 to 49 days. Hong and Smith (1996) calculated half-lives for dithiopyr ranging from 68.8 days (20˚C) to 39.2 days (35˚C) in golf course greens RM in sealed containers in the dark.

Effects on Turfgrass Establishment, Growth and Development

Effects on root growth

Dithiopyr can be applied to large number of warm- and cool-season turfgrasses for the control of *Digitaria* spp., goosegrass (*Elusine indica* L.), and other annual grasses. Many studies reported the effects of herbicides on rooting of different turfgrass species (Fishel and Coats, 1994; Mitra and Bhowmik 2005; Bhowmik 2005). Landschoot *et al*. (1993) studied the influence of preemergence herbicides, including dithiopyr on rooting of Kentucky bluegrass (*Poa pratensis* L.) and tall fescue (*Festuca arundinacea* Schreb.). Dithiopyr at 0.4 kg ha^{-1} inhibited root growth when cores, 10 cm in diameter and 2.5 cm deep, of herbicide treated *P. pratensis* were placed into pots of perlite. *Festuca arundinacea* rooting was unaffected by dithiopyr.

Bhowmik *et al*. (1993) found comparatively lower root strength of *P. pratensis* at 12 week after treatment of dithiopyr as compared to the untreated turfgrass. No harmful effects were observed with root length, rhizome number, or root dry weight of *P. pratensis*. Bhowmik *et al*. (1995) also observed slight reduction of root length and root weight of *A. palustris* with dithiopyr up to 6 WAT and 4 WAT respectively, but recovery of both characteristics was also observed (Figure 1). Application of dithiopyr to turfgrass under abiotic or biotic stress had much more deleterious effects on rooting. Although, many studies revealed same effects of dithiopyr on turfgrass rooting, but with proper timing and application rate majority of turfgrasses do not show a significant reduction in root growth. Reductions in root mass of bentgrass (*Agrostis palustris* L.) were observed with dithiopyr at 0.4 or 0.8 kg ha^{-1} during fall (Hart *et al*. 2004).

Severe effects of dithiopyr on rooting of *C. dactylon* sod were noticed (Fishel and Coats, 1994). Root fresh weight was reduced with dithiopyr (0.56 and 1.1 kg ai ha^{-1}). Significant numbers of malformed roots were observed for greater periods of time, where dithiopyr had been applied. McCarty *et al*. (1995) noticed 12 to 24% unrooted stolons of St. Augustingrass [*Stenotaphrum secundatum* (Walt.) Kuntze] even 4 to 8 months after the dithiopyr application (0.8 kg ai ha^{-1}). They did not find any significant reduction of *S. secundatum* root weight with dithiopyr. Dithiopyr applied at 0.84 and 2.5 kg ha^{-1} reduced *C. dactylon* rooting by 59 and 70% at 60 days after initial treatment (Ferrell *et al*. 2003). However, dithiopyr applied sequentially (0.42 kg ha^{-1}) did not reduce *C. dactylon* root strength.

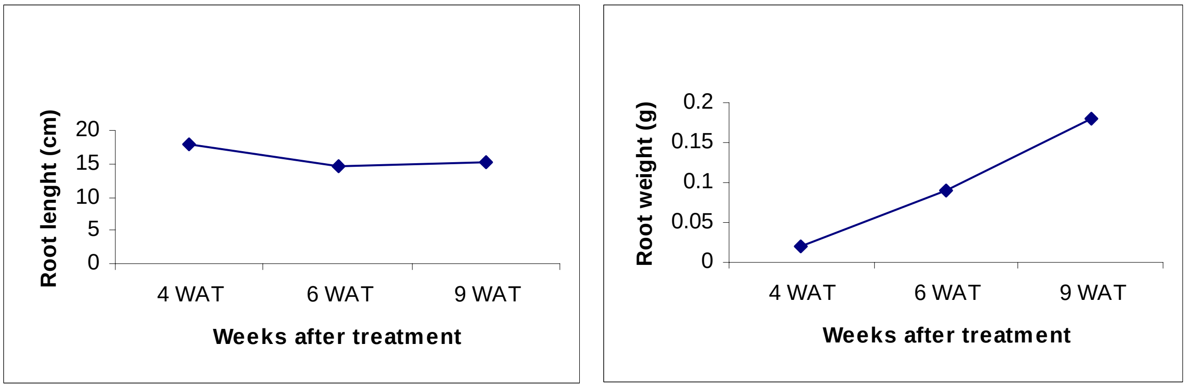

Figure 1. Effects of dithiopyr on root length and root weight of *Agrostis palustris.*

Effects on shoot growth

Dithiopyr on shoot growth of turfgrasses are less injurious compared to the effects on rooting. Dithiopyr is a preemergence herbicide with post-emergence activity on smooth crabgrass [*Digitaria ischaemum* (Schreb. *ex* Schweig)] up to the three-leaf stage and may be applied after turfgrass seedling emergence (Bhowmik and Bahnson 1989). Wetzel and Dernoenden (1993) examined the tolerance of *Lolium perenne* seedlings to dithiopyr applications at various times. Dithiopyr applied 2, 4, and 6 weeks after emergence caused initial injury to *L. perenne* in the form of thinning, leaf die back, and leaf yellowing. The emulsifiable concentrate (EC) formulation was more injurious than granular (G) formulation regardless of application timing, but all plants recovered, and did not show any injury three months after treatment. Bhowmik *et al*. (1993) did not find any harmful effects of dithiopyr (0.4 kg ha^{-1}) on *P. pratensis*, 4 and 9 weeks after treatment. Bhowmik *et al*. (1995) observed a slight reduction of clipping fresh weight, and number of tillers of *A. palustris* up to 6 weeks after dithiopyr application (Figure 2). Rapid recovery of shoot growth of *A. palustris*, 6 weeks after treatment was also noticed in same experiment.

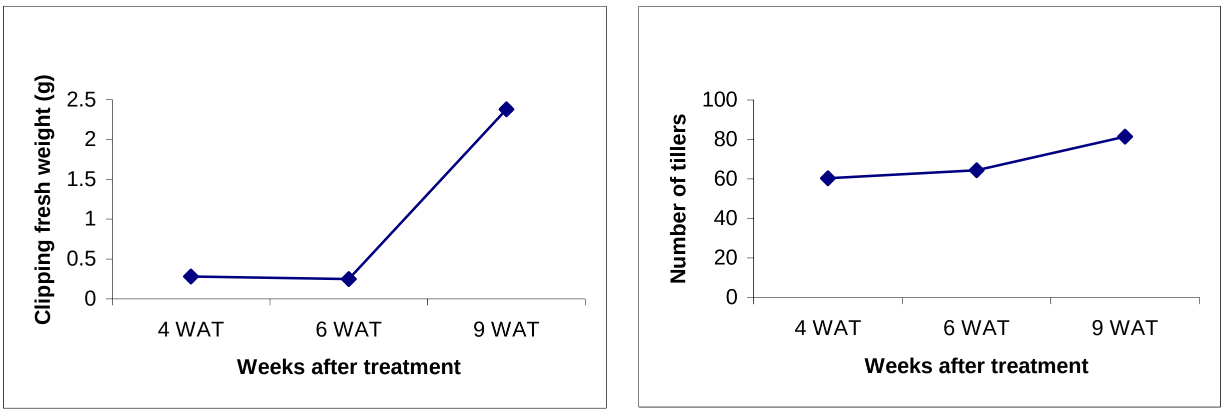

Figure 2. Effects of dithiopyr on clipping weight, and number of tillers of *Agrostis palustris.*

Bevard and Watschke (1999) studied 'Penneagle' *A. palustris* seedling tolerance to dithiopyr. They applied both EC and G formulations of dithiopyr to seedlings 2, 4, and 8 weeks after emergence (WAE). They found unacceptable seedling injury regardless of formulations and rates (0.14, 0.21, 0.28, 0.42 kg ha^{-1}) when applied 2 WAE. Shoot injury was characterized by stunting of plants, leaf tip die back, yellowing of older leaves, and a visible decline in quality. Dithiopyr applied 4 WAE caused less injury, and within the acceptable range, while applied 8 WAE only slightly injured *A. palustris* at the high rate for both formulations.

Tolerance of Zoysiagrass (*Zoysia japonica* Steud) cultivars to preemergence herbicides, including dithiopyr was studied by Johnson and Carrow (1999). During summer months (May, June and July), significant injury to *Z. japonica* cultivars was noted with the recommended rate (0.8 kg ha^{-1}) and three times the recommended rate (2.5 kg ha^{-1}) of dithiopyr. This rate of dithiopyr during June application reduced the quality of 'Diamond' cultivar by 16%. The quality of 'Palisade' cultivar was 76% in May when treated with three times the rate of dithiopyr, and 82% in July. Johnson (1995) observed that dithiopyr at 0.8 kg ha^{-1} suppressed the quality of four *C. dactylon* cultivars. When dithiopyr was applied at 2.5 kg ha^{-1}, the quality of all cultivars was reduced severely (34 to 39%). Many other studies showed that there was no significant shoot injury of turfgrass with dithiopyr applications (Yelverton and McCarty 2001; Reicher *et al*. 2000).

Effects on turfgrass establishment from seed

Turfgrass establishment from seed requires a significant weed free period for growth and development. Overall performance of turfgrass is very much dependent on its initial establishment. Applying preemergence herbicides close to seeding may injure and delay establishment of turfgrasses (Dernoeden 1990; Bhowmik, 2005). However, herbicides are often applied preemergence for weed control in spring or fall-seeded grasses. Johnson and Bundschuh (1993) found significant interactions between dithiopyr rates, formulations, intervals between application and seeding on establishment of *F. arundinacea*, *L. perenne*, and *A. palustris*. *F. arundinacea. L. perenne* tolerated a closer interval between dithiopyr EC at 0.56 kg ha^{-1} (2 weeks before seeding-WBS) treatment and seeding date than did *A. palustris* (12WBS). However, they found that when the rate of the EC formulation was increased to 0.84 kg ha^{-1}, an 8 weeks interval was needed to prevent stand reduction in *L. perenne* cover. This study revealed that the optimum interval between applications of granular formulation of dithiopyr (0.56 kg ha^{-1}) and seeding was 8 weeks for *F. arundinacea* and *L. perenne*. In contrast, ground cover of *A. palustris* was reduced when treated within 12 weeks of seeding.

Keeley and Zhou (2005) reported that 6-wk interval was required between dithiopyr application and *P. pratensis* seeding in case of broadcast seeding, where 11-wk interval was needed for safe establishment of slit-seeded *P. pratensis*. In another study, dithiopyr applied as early as 3 days after emergence (DAE) improved cover of *P. pratensis* by reducing competition from large crabgrass (Reicher *et al*., 2000). They found that, when seeding is done early in the spring before *Digitaria spp*. germination, dithiopyr could be applied at 0.28 kg ha^{-1} 10 DAE or at 0.56 kg ha^{-1} 14 DAE without any turfgrass injury. Reicher *et al*. (1999) also observed similar trend in *P. pratensis* and *L. perenne*. They found that dithiopyr applied PPI or 0 WAE severely damaged germination and cover of both turfgrasses. But dithiopyr applied 2 WAE or later had no negative effects on *P. pratensis* cover and increased *P. pratensis* cover. Johnson (1994) reported that single (0.6 to 1.1 kg ha^{-1}) and multiple (0.3 + 0.3 kg ha^{-1}) applications did not have any influence on transition from over seeded *L. perenne* back to *C. dactylon*. Generally, application of dithiopyr at proper time and with proper rate can enhance turfgrass establishment from seed by reducing the early competition from *Digitaria* spp. The rate and timing of dithiopyr applications largely depends on turfgrass species and cultivars.

Patton *et al*. (2004) found that *C. dactylon* coverage 3 weeks after emergence (WAE) was reduced when dithiopyr was applied PRE or 0 WAE, while by 7 WAE only PRE treatment of dithiopyr reduced *C. dactylon* coverage. Their study revealed that dithiopyr could be safely applied to *C. dactylon* seedling 1 WAE or later for better establishment.

Effects on turfgrass establishment from sod

One of the greatest concerns with winter-installed sod is weed infestations. Winter annuals and perennials can be controlled with many herbicides. However, many of the commonly used herbicides can potentially affect the growth, development and survival of newly installed sod. Ferrell *et al*. (2003) found that dithiopyr applied at 2.5 kg ha^{-1} reduced root strength of *F. arundinacea* sod 60 days after initial treatment (DAIT). However, by 120 DAIT the same rate of dithiopyr did not affect *F. arundinacea* root development. They also found severe reduction of root development of *C. dactylon* sod even up to 120 DAIT, when applied as PRE at 0.84 kg ha^{-1}, while dithiopyr applied sequentially (0.42 fb 0.42 kg ha^{-1}) did not reduce root strength. Fishel and Coats (1994) observed that dithiopyr reduced root growth of *C. dactylon* sod as much as 60% when applied PRE at 0.56 kg ha^{-1}. They also noted that dithiopyr caused abnormal rooting in *C. dactylon* sod and produced ≤ 21 abnormal roots per pot 2 and 4 weeks after treatment.

Pre-emergence herbicides may affect rooting of newly installed sod. Bhowmik *et al*. (1993) reported lower root strength of *P. pratensis* at 12 weeks after treatment of dithiopyr. Sod regrowth of *S. secundatum* was studied by McCarty *et al*. (1995). They found that dithiopyr caused unrooted or poorly rooted stolon on harvested *S. secundatum* after preemergence herbicide applications, including dithiopyr. However, they concluded that as phytotoxicity ratings were not reduced below the minimum acceptable level of 6.5, dithiopyr could be used safely on *S. secundatum*, remaining after sod harvest. Fagerness *et al*. (2002) observed suppression of 'Tifway' *C. dactylon* establishment as much as 25% with fall application of dithiopyr, but recovery was evident by the end of the growing season. They found that *Z. japonica* establishment was less affected by dithiopyr compared to prodiamine. Their results suggested that reduced rates of dithiopyr may be warranted in areas prone to winter injury. In general, dithiopyr can be applied safely to healthy sod for better weed control, resulting into proper establishment of turfgrass.

In summary, dithiopyr can safely be used for weed control in cool-season as well as warm-season turfgrass environments. However, the rate and timing of application, regardless of turfgrass species, influence its effects on turfgrass species.

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INTEGRATED MANAGEMENT OF ALLIGATOR WEED [*Alternanthera philoxeroides* (Martt.) Griseb] AT BOTANY WETLANDS, SYDNEY- A CASE STUDY

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Abstract: Sydney Water provides drinking water and wastewater services to over four million people in Sydney. This case study addresses the difficulties and costs associated with the control of Alligator weed at Botany Wetlands, which is a key natural resource owned by the Corporation. This pond system is ecologically significant, and historically important, as they served as a drinking water supply for early settlers in Sydney. First noted as occasional patches in the ponds in 1985, Alligator weed infestations became extensive by 1996, posing a significant threat to the wetlands. Since 1996, Sydney Water has implemented an integrated weed management (IWM) plan to manage Alligator weed. This combines (a) systematic monitoring, (b) regular application of herbicides (™Biactive glyphosate and metsulfuron-methyl), (c) manual removal of floating masses, (d) lowering of water levels to allow better control, (e) controlled burning of sedges in infested areas, (f) large scale planting of native macrophytes in treated areas, and (g) exploiting the feeding by bio-control agents. Minimising the potential impact of chemicals, herbicides are not applied on patches where Alligator Weed flea beetles *Agasicles hygrophila* are most active. Herbicides are applied as spot-treatments at monthly intervals during cooler months, but more frequently in warmer months. New infestations are treated with herbicides or are manually removed, preventing downstream spread. Disposal of collected material is by burial in sand, after desiccation and treatment with herbicides. It is estimated that, on average, 90–95% of the previous Alligator Weed infestation has been controlled over the past 10 years. The costs of control and amounts of chemicals used have declined. Although this is a relatively successful result, the current gains made on Alligator Weed management at Botany Wetlands have been achieved only through diligent action, continual assessment and persistent management intervention, on a yearly basis.

Key words: Alligator weed, management

Introduction

Alligator Weed [*Alternanthera philoxeroides* (Mart.) Griseb.] is one of twenty Australian Weeds of National Significance (WONS), and is a declared noxious weed in New South Wales (NSW). Alligator Weed was accidentally introduced to Australia from ship's ballast into Newcastle harbour and Botany Bay around 1944. The weed has since spread in many areas of NSW, and is also present in south-east Queensland and Victoria. It has the potential to spread much further and threaten agriculture, wetlands, waterways and properties over much of Australia, with potential losses of many millions of dollars to agricultural, tourism and extractive industries (NWSEC 2000; Chandrasena *et al*. 2004).

The Botany Wetlands are the largest coastal freshwater lakes in the Sydney Region. The Wetlands comprise of 11 interconnected ponds and dams (Figure 1), which stretch over a 4 km corridor in Sydney's eastern suburbs, north of Botany Bay. The ponds are an integrated surface and ground water system, fed by the Botany sand aquifer and surface runoff from 20 km^2 of urban catchment. The Wetlands are ecologically valuable because they constitute an extensive aquatic habitat assemblage, supporting aquatic biota in a highly urban area. They are historically significant, because the ponds served as a drinking water supply to early Sydney settlers during 1858 to 1886 (Chandrasena *et al*. 2002). Sydney Water Corporation owns and manages Botany Wetlands.

Minor infestations of Alligator weed at this historic location were first noted around 1985; limited control action commenced in early 1990s, but the infestations increased in size and spread over the next few years. By 1996, infestations were extensive, posing a significant threat to the Wetlands. Fragmentation of patches, leading to spread and entry of new fragments from the catchment through stormwater in major storm events was a problem that was identified early. The purpose of this article is to discuss the results of an integrated Alligator Weed management program at Botany Wetlands.

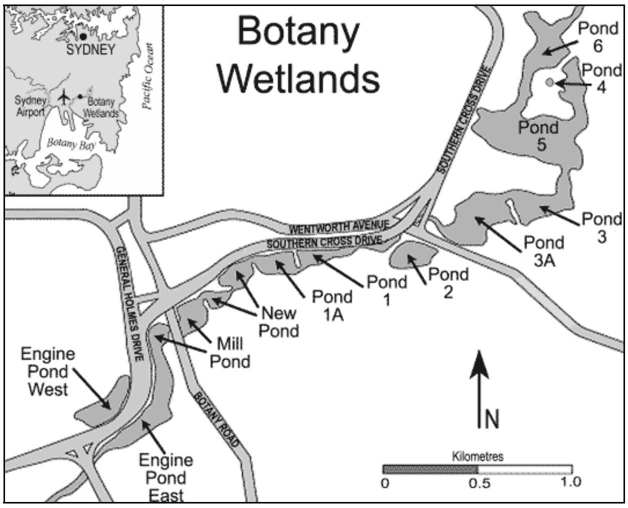

Figure 1. The Botany Wetlands, Sydney, NSW, Australia

Integrated weed management

Broadly, integrated management involves the integration of a number of weed control options to achieve the most favourable economic (cost-benefit ratio), sociological and ecological outcomes for managing an invasive organism like Alligator Weed. IWM is based on a logical approach, system understanding, planning and implementing a control program, which is often 'site-specific'. The broader goal would be to arrest the spread of an invading weed, and reduce or eliminate the threat posed by the invader. Understanding the various factors that influence weed invasion of a site is a key to planning and successfully implementing IWM (Figure 2). IWM also includes monitoring of performance against nominated criteria and critical analysis of success.

Planning, including R&D

- System understanding
- Knowledge of target weed's biology, ecology, physiology
- Control options- physical, chemical, biological, cultural
- Costs and impacts of control
- Level of control & Strategies- Site-specific plans;
- Link to Local, Regional, National Strategic Plans

→

Program Implementation

- Apply methods & monitor performance
- Assessment of success and impacts
- Continuous improvement

→

Program Management

Assess program success-
- Cost-effectiveness
- Stakeholder expectations
- Environmental goals

Figure 2. Components of an integrated weed management (IWM) program

IWM Methodology

The IWM approach applied to manage Alligator weed infestations in Botany Wetlands has the following elements: (1) setting up of specific sites and zones for monitoring the weed's spread and treatment effectiveness; (2) establishment of performance criteria (nil, minor or low-level occurrence, within the pond system) (3) systematic monitoring and recording, to detect and treat new infestations; (4) scouting to check potential upstream sources of new inoculum from the catchment; (5) regular spot applications of herbicides- ™Bi-Active glyphosate, 1% v/v, on aquatic infestations; metsulfuron-Methyl (™Brush-Off) on terrestrial infestations; frequency of treatments, on average, was once per month during cooler months, and twice per month from September to May each year; (6) water level management, to better access to persistent patches for herbicide treatments; (7) manual removal of floating masses or loosely attached plants, caught up in snags and debris, treatment with Glyphosate, before disposal by burial in sand; (8) controlled burning of weed infested pond shorelines occupied by macrophytes; (9) adjustment of treatment regimes to allow bio-control agent Flea Beetle (*Agasicles hygrophila* Selman & Vogt) to do the maximum damage in January-February, so that the overall herbicide loads applied could be reduced; (10) large-scale planting of native macrophytes in riparian zones, cleared of entrenched Alligator weed; and (11) annual review and critical assessment against the performance criteria.

Results and Discussion

When the IWM program commenced in 1997, Alligator weed (total estimated area ≈ 5000 m^2) was largely restricted to upstream Ponds 4, 5 and 6 (Figure 1). With intensive herbicide spot treatments (total herbicide load applied- 24 kg a.i in 1996-97), this area was reduced to less than half within one year (Figure 3). However, treatments were often only partially effective, because of access difficulties in slushy mud for spraying. Inadequate uptake of herbicide by a few leaves of the weed exposed above water was a probable cause of ineffective treatments. Inadequate uptake was also probably due to the lack of a surfactant in ™Bi-Active Glyphosate, which caused poor retention of spray on leaves. Incorporation of a biodegradable additive, even a simple vegetable oil like Synertrol®, increased the effectiveness of this Glyphosate formulation on several species (Sharma *et al*. 2004). Such an additive may prevent sub-lethal treatments from causing fragmentation, and consequently, reduce the spread. Metsulfuron-Methyl is highly effective on 'terrestrial' Alligator Weed, but this herbicide has not yet been tested on infestations 'in water'.

The ability to lower the water level in the largest water body- Pond 5 was important, as this allowed more effective treatments with Glyphosate. Slushy mud and snags in the ponds were major obstacles, as they prevented boat access. Where access was possible by boat and by shoreline walking, removing any loosely attached clumps was essential to reduce downstream spread of the weed. A high inspection and spot treatment frequency included time spent on scouting to find new patches, both within Botany Wetlands and in upstream channels that drain stormwater into the Wetlands. Figure 3 shows how the overall herbicide load applied decreased over the first few years of IWM implementation, to ≈12 kg a.i. by 1999-2001.

Preventing the entry of new 'inoculum' from upstream sources is essential to manage Alligator Weed in Botany Wetlands, within its large urban catchment. Scouting to find potential upstream sources, and treatment to eradicate those, was an integral part of IWM. Successful control in some ponds can also be attributed to burning of sedges and rushes, such as *Bolboschoenus fluviatilis* (Torrey) Soják, and *Typha orientalis* Presl, carried out several times during their winter-dieback. The clearing after burning allowed re-emerging, persistent

Alligator Weed patches to be effectively treated. Macrophytes grew back after burning, because their rhizomes and bulbs were unaffected by fire.

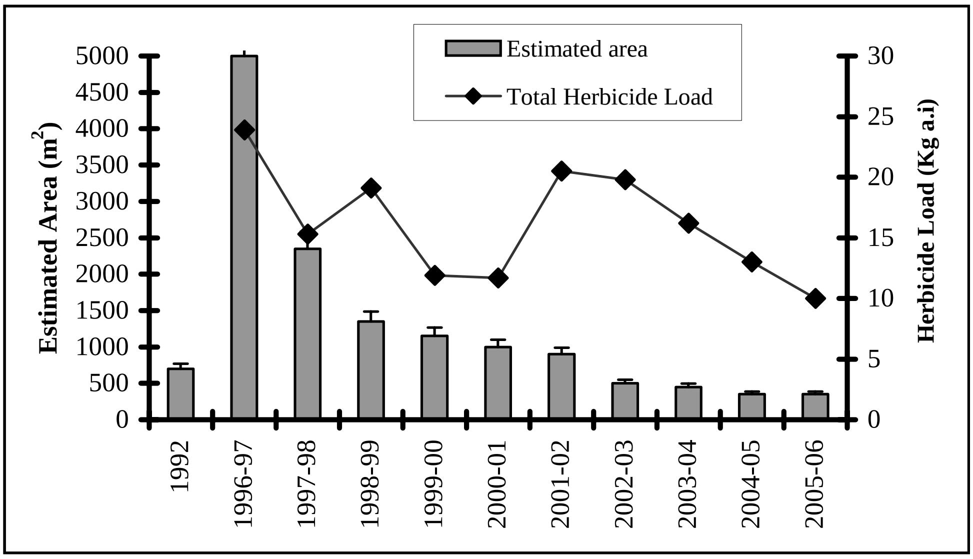

Figure 3. Estimated extents of Alligator weed infestations and annual herbicide loads used to control weed spread in Botany Wetlands, Sydney, 1996-2006.

Annually, the Alligator weed flea beetle (*Agasicles hygrophila* Selman & Vogt; Coleoptera: Chrysomelidae) becomes active during January to April; its foraging strips the shoot system of leaves, sometimes as much as by 90%. We avoided treating these patches during this period, allowing the flea-beetle to do as much damage as possible. However, the control achieved by the flea-beetle appears limited in the Sydney basin, because adults do not survive or breed well under temperatures below 13 ^{0}C (Stewart *et al*. 1999), which are common during the winter in Sydney. Like sub-lethal glyphosate treatments, flea-beetle foraging may cause disintegration and spread of the weed.

The control of vast infestations of Primrose Willow [*Ludwigia peruviana* (L.) Hara] in the ponds during 1996-2000 (Chandrasena *et al*. 2002) was followed by large-scale revegetation of shorelines with wetland plants, which were once dominant in Botany Wetlands. Shoreline revegetation did not directly prevent these areas from being re-infested by Alligator weed, as entangled weed fragments grow among macrophytes. However, the benefits of revegetation are indirect- possibly through the stabilisation of degraded, muddy and peaty shorelines and general environmental improvements.

Removal of Primrose Willow infestations in downstream ponds increased the water flow through the system (Chandrasena *et al*. 2002). This caused Alligator weed to spread downstream in major storm events. Significant patches were found growing in downstream Pond 3 and 3A in 1999-2000 and in Ponds 1 and 1A in 2001-02. Preventing wider spread became a high priority. Inspections were increased, as well as spot treatments. During 2001-03, scouting to detect new patches increased; spot-treatment frequency, and overall herbicide load used (Figure 3) also increased, mainly to arrest this spread. During the drought years (2002-06), water levels in all ponds decreased, allowing more shoreline access for treatments. Exposed 'terrestrial' Alligator weed could also be treated with the selective herbicide-Metsulfuron-Methyl. The overall reduction of Alligator weed at Botany Wetlands is about 90% of what existed prior to 1996 (Figure 3). Reduction of upstream infestations in Ponds 4, 5 and 6 is >95%. The decline in overall management cost, over time, is shown in Figure 4.

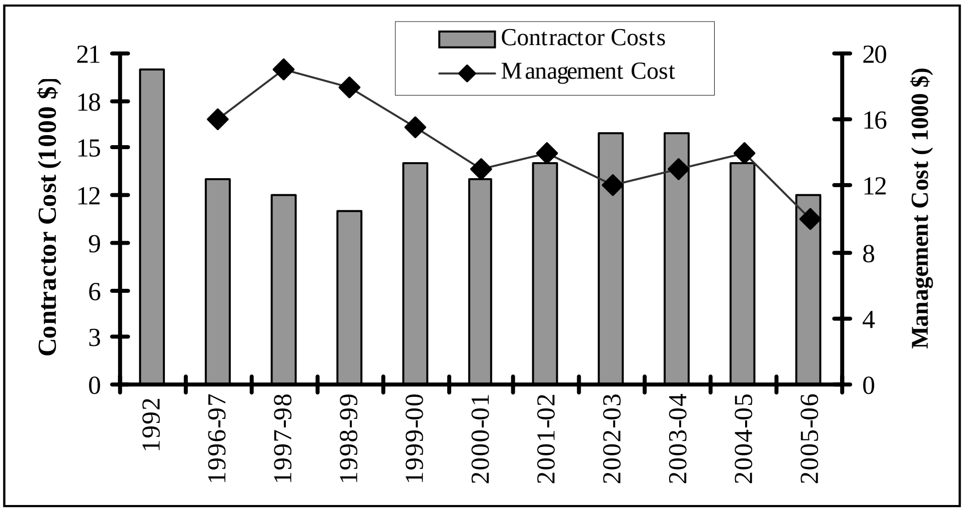

Figure 4. Contractor costs and management costs for the IWM program, 1996-2006.

Conclusions

The inability to eliminate Alligator weed from Botany Wetlands reflects its resilience to current control measures in a nutrient-enriched environment. Our experience of the past 10 years of implementing IWM is that without more effective herbicide treatments or new biological control agent(s), complete eradication of Alligator Weed from the Wetlands may not be achieved. The chances of some minor amount of weed existing within the nutrient-enriched substrata in the macrophyte-dominated system are high. However, the Case Study shows that by implementing a well-managed IWM program, with dedicated funding and resources, backed up by strong project management, it is not impossible to reduce its threat to a low level. Sydney Water's commitment to the above has protected the integrity of natural ecosystems in this historic wetland system, where uncontrolled growth of Alligator Weed could have had serious repercussions. The lessons learnt from Botany Wetlands are replicable elsewhere.

An interesting research study would be to determine whether partial killing of the weed by sub-lethal Glyphosate treatments increase the palatability of the leaves to flea-beetle foraging. Another study that could improve the overall management effort is to investigate the possibility of incorporating a commercially available hormone (such as Na salt of 2,4-D at 500 ppm; 1-Naphthyl Acetic Acid, NAA or Amino ethoxy vinyl glycine, AVG at 100-250 ppb) into herbicide treatments, to prevent disintegration of patches. Given that the flea-beetle's capacity to damage the weed is limited, it is also necessary to develop other bio-control agents, for integration into future management.

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WEED CONTROL BY IMIDAZOLINONE HERBICIDES IN "CLEARFIELD™" PADDY GROWN WITH AN IMI-TOLERANT INDICA RICE VARIETY

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Abstract: Experiments were carried out at the Experimental Farm of the Cuulong Delta Rice Research Institute (CLRRI) in the Mekong Delta in Vietnam with two types of rice culture, dry- seeded and wet- seeded rice. The tested variety was OM 5749-5, indica rice developed by plant breeders at CLRRI by crossing between promising Vietnamese indica rice with IMI-tolerant japonica rice from Louisiana State University, USA. The tested herbicides were : imazapic, imazapyr, imazapic+ imazapyr and imazethapyr+imazapyr. Common weeds observed in the experimental field including *Echinochloa crus- galli*, *Leptochloa chinensis, Cyperus iria, Cyperus difformis, Ludwigia octovalvis* and especially weedy rice (*Oryza sativa*) were controlled successfully by the herbicides . The density and dry weight of weeds were reduced significantly when compared to those in the untreated check. In dry seeded rice, the average rice grain yield of six imidazolinone treatments was 1,83 t/ha , 101,1% higher than check (0,91 t/ha). The corresponding data in wet -seeded rice were 2.15 t/ha and 0.88 t/ha with a 143.9% increment. Rice quality was also improved by the treatments. The number of contaminated weedy rice seeds in rice products, seeds dropped on the soil surface, the percentage of red grains in milled rice were also reduced under herbicide treatments when compared to that of check statistically.

Key words: Imidazolinone, herbicide tolerant variety, weedy rice control, Clearfield ™ paddy.

Introduction

Weedy rice, commonly considered to be ecotypes of *Oryza sativa*, is a new pest in rice growing countries in the world including Vietnam. In tropical areas, weedy rice develop as progenies of crosses between wild rice and cultivated rice or come from degradation of cultivated rice varieties. The major characteristic of weedy rice is easy shattering . Other characteristics are taller plants, fewer tillers, and high percentage of red rice in milled rice (Chin *et al*. 2000). Weedy rice competes with cultivated rice for sunlight, water and nutrients resulting in reductions in rice yield. The quality of milled rice is reduced due to contaminated red rice. Weedy rice infestation in rice fields is also dangerous because the soil seed bank increase over time with self regeneration and there is no effective selective herbicide for controlling weedy rice. Recently, a new option for controlling weedy rice and also common weeds in rice fields has been initiated by exploring the integration of imidazolinone herbicides and tolerant trait containing variety (called CLEARFIELD™ rice). Imidazolinone herbicides controls weeds by inhibiting the plant specific enzyme acetohydroxyacid synthase (AHAS), which is involved in the biosynthesis pathway of the branched-chain amino acids – valine, leucine and isoleucine. This inhibition causes a disruption of protein synthesis which in turn, interferes with DNA synthesis and cell growth (Shanner and Connor, 1991). CLEARFIELD™ rice has been developed by Louisiana State University Agricultural Center breeders through a combination of mutagenesis and conventional plant breeding using a variety tolerant to imidazolinone herbicides. This is characterized as a non-GMO variety. In Vietnam, the plant breeders at the Cuulong Delta Rice Research Institute have developed indica rice varieties, which are tolerant to imidazolinone herbicides. The objective of this research was to determine whether the integration of imidazolinone herbicides and the tolerant trait containing variety can be used for controlling weedy rice and common weeds in rice fields

Materials and Methods

Two experiments were conducted in a clay soil in lowland fields with a good water supply in the Spring-Summer season with dry seeded rice and Summer-Autumn of 2006 with wet seeded rice at the Experimental farm of the Cuulong Delta Rice Research Institute (CLRRI). The rice variety was OM 5749-5, an indica rice type with IMI-tolerance trait PWC bred by plant breeders at the CLRRI. In dry seeded rice, 200 kg of cultivated rice seeds were mixed with 100 kg of weedy rice seeds and broadcasted randomly before incorporation into the soil surface. In wet seeded rice, 100 kg of pre-germinated seeds were sown in lines by a drum seeder. Pre-germinated seeds of weedy rice at the rate of 100 kg /ha were broadcast randomly one day later. The field trials with plot size of 20 m^2 were established in a randomized complete block design with four replications and 7 treatments. Three active ingredients of imazapic, imazapyr and imazethapyr were used as solo and ready-mix formulations at various rates from 100 to 120 g a.i./ha. Herbicides were sprayed 12 days after emergence (DAE) in dry seeded rice and 10 days after sowing (DAS) in wet seeded rice. Crop oil was added at 0.5% water volume as a non-ionic surfactant.

Results and Discussion

Herbicidal activity

The populations of weedy rice, *Echinochloa crus-galli*, *Leptochloa chinensis*, *Cyperus iria* affected by treatments of imidazolinone herbicides have been presented in Table 1. All herbicide treatments reduced weedy rice densities significantly as compared to that of the untreated check (182.7 plants/m^2). All treatments were very effective in controlling *Echinochloa crus-galli* except in treatment T6 [imazethapyr+imazapyr] applied at 120 g a.i/ha. There was no difference between this treatment with the weedy check. The density of *Leptochloa chinensis* remained at 22 plants/m^2 in the control plot while it was completely killed in all herbicide treatments. The same trend was observed in the case of *Cyperus iria* . The density of *Cyperus iria* was lower in all treatments except in T6 [imazethapyr+imazapyr] at 120 g a.i/ha. In this treatment the weed populations were 3.3 plants /m^2.

Table 1. Weed and weedy rice density (No. of plants/m^2) at 70 days after emergence (DAE) as affected by treatments at CLRRI, Spring-Summer 2006.

Treatments		Weedy rice*	ECHCG	LEPCH	CYPIR
T1	Untreated check	182.7	12.0	22.0	17.3
T2	[Imazapic+imazapyr] at 100 g a.i/ha	0.0	0.0	0.0	0.0
T3	[Imazapic+imazapyr]at 110 g a.i/ha	0.0	0.0	0.0	0.0
T4	[Imazapic+imazapyr]at 120 g a.i/ha	0.0	0.0	0.0	0.0
T5	Imazapyr at 120 g a.i/ha	12.7	0.0	0.0	0.0
T6	[Imazethapyr+imazapyr] at 120 g a.i/ha	0.0	14.0	0.0	3.3
T7	Imazapic at 120 g a.i/ha	0.0	0.0	0.0	0.0
	LSD (p=0.05)	81.4	6.1	12.8	14.1

* Weedy rice (*Oryza sativa*); ECHCG = *Echinochloa crus-galli*; LEPCH = *Leptochloa chinensis*; CYPIR= *Cyperus iria*.

All imidazolinone herbicide treatments caused reduced weedy rice and weed dry weights when as compared to that of the untreated check (269 g/m^2) (Table 2). However, the treatment T5 [Imazapyr] at 120 g a.i/ha could not control weedy rice completely, resulting in the presence of weedy rice with a dry weight of 21.7 g/m^2. *Echinochloa crus-galli* also was controlled successfully by all tested herbicides. However, some plants of this weed remained in the treatment with T6 [imazethapyr+imazapyr] at 120 g a.i/ha resulting in the accumulation of 8.0 g/m^2 of dry matter. *Leptochloa chinensis* species was controlled completely by all treatments. Dry weights of *Cyperus iria* were reduced significantly in all treatments when compared to that of the untreated plot (26.6 g/m^2) but the weed dry weight remained at 12 g/m^2 in the treatment T6 [imazethapyr+imazapyr] at 120 g a.i/ha.

Table 2. Weed and weedy rice dry weight (g/m^2) at 70DAE as affected by treatments at CLRRI, Summer-Autumn 2006.

Treatments		Weedy rice *	ECHCG	LEPCH	CYPIR
T1	Untreated check	269.7	35.9	7.7	26.6
T2	[Imazapic+imazapyr] at 100 g a.i/ha	0.0	0.0	0.0	0.0
T3	[Imazapic+imazapyr]at 110 g a.i/ha	0.0	0.0	0.0	0.0
T4	[Imazapic+imazapyr]at 120 g a.i/ha	0.0	0.0	0.0	0.0
T5	Imazapyr at 120 g a.i/ha	21.7	0.0	0.0	0.0
T6	[Imazethapyr+imazapyr] at 120 g a.i/ha	0.0	8.0	0.0	12.0
T7	Imazapic at 120 g a.i/ha	0.0	0.0	0.0	0.0
	LSD (p=0.05)	78.2	25.6	3.5	3.1

* Weedy rice (*Oryza sativa*); ECHCG = *Echinochloa crus-galli*; LEPCH = *Leptochloa chinensis*; CYPIR= *Cyperus iria*.

Yield components and grain yield.

The imidazolinone herbicides successfully controlled weedy rice and common weeds in rice fields and therefore the competition of weeds on rice plants was minimized and rice plants could grow better when compared to that of untreated plots (Table 3).

Table 3. Yield components and yield of dry seeded rice as affected by treatments at CLRRI, Spring-Summer 2006.

Treatments		No. of panicles per m^2	No. of filled grains per panicle	1000-grain wt (g)	Yield (t/ha)
T1	Untreated check	286	51.1	26.6	0.9
T2	[Imazapic+imazapyr] at 100 g a.i/ha	311	63.5	26.4	1.7
T3	[Imazapic+imazapyr]at 110 g a.i/ha	323	62.1	26.7	1.9
T4	[Imazapic+imazapyr]at 120 g a.i/ha	325	63.2	26.7	1.8
T5	Imazapyr at 120 g a.i/ha	341	63.3	26.3	1.8
T6	[Imazethapyr+imazapyr] at 120 g a.i/ha	345	62.6	26.4	1.8
T7	Imazapic at 120 g a.i/ha	350	67.2	26.5	2.1
	LSD (p=0.05)	48	4.2	0.9	0.5

Three treatments of [imazapic+imazapyr] at 100; 110 and 120 g a.i/ha marginally increased the numbers of panicles/m^2. The tiller numbers/m^2 of plants in the three treatments of T5 [Imazapyr] at 120 g a.i/ha; T6 [imazethapyr+imazapyr] at 120 g a.i/ha and T7 [Imazapic] at 120 g a.i/ha were superior to that of untreated check . All herbicide treatments increased the numbers of filled grain per panicle significantly when compared to values of the untreated check (51.1 grains/panicle). There was no significant difference in 1000-grain weights amongst treatments. Rice yields in treated plots were significantly greater when compared to that of untreated check (0.9 t/ha). However grain yield in all herbicide applied plots were similar. On an average, application of all herbicides increased rice yield by 106% when compared with that of the untreated check.

Research results in wet-seeded rice

One wet-seeded experiment was conducted with the same design and treatments was during the Summer-Autumn season of 2006 .

Table 4. Density (No. plants /m^2) of weedy rice at 28; 56 and 84 days after sowing (DAS) as affected by treatments , CLRRI, Summer-Autumn, 2006.

Treatments		Weedy rice density (No. of plants/m^2)		
		28 DAS*	56 DAS	84 DAS
T1	Untreated check	69.3	245	246
T2	[Imazapic+imazapyr] at 100 g a.i/ha	0.3	0	0
T3	[Imazapic+imazapyr]at 110 g a.i/ha	5.0	0	0
T4	[Imazapic+imazapyr]at 120 g a.i/ha	0.4	0	0
T5	Imazapyr at 120 g a.i/ha	10.3	0	0
T6	[Imazethapyr+imazapyr] at 120 g a.i/ha	1.8	0	0
T7	Imazapic at 120 g a.i/ha	0.0	0	0
	LSD (p=0.05)	11.4	28	28

DAS – days after sowing

At 28 DAS, the density of weedy rice was 69.3 plants/m^2 in untreated check (Table 4). All herbicide treatments reduced the population of weedy rice at 28 DAS significantly when compared to that of the control plots. However, some plants of weedy rice were observed at this stage because the remaining weedy rice showed the symptoms of injury but were not killed completely. At 56 and 84 DAS, all weedy rice plants were susceptible and completely killed. When imidazolinone herbicides were not sprayed, weedy rice grew normally and reached a density of 245-246 plants/m^2.

Table 5. Yield components and grain yield as affected by treatments, CLRRI, Summer-Autumn 2006.

Treatments		No. of panicles per m^2	No. of filled grains per panicle	1000-grain wt (g)	Yield (t/ha)
T1	Untreated check	142	57.4	26.5	0.88
T2	[Imazapic+imazapyr] at 100 g a.i/ha	301	62.6	26.5	2.22
T3	[Imazapic+imazapyr]at 110 g a.i/ha	339	76.7	27.2	2.25
T4	[Imazapic+imazapyr]at 120 g a.i/ha	317	66.6	25.7	2.15
T5	Imazapyr at 120 g a.i/ha	292	71.7	26.0	2.05
T6	[Imazethapyr+imazapyr] at 120 g a.i/ha	316	66.5	27.1	2.13
T7	Imazapic at 120 g a.i/ha	294	70.2	26.5	2.08
	LSD (p=0.05)	46	16.7	2.1	0.48

All the imidazolinone herbicide treatments increased number of panicles/m^2 significantly when compared with the untreated check (142 panicles /m^2) (Table 5). The highest number of panicles/m^2 was in treatment T3 [imazapic+imazapyr] at 110 a.i/ha (339 panicles /m^2), which was greater than that of T5 [imazapyr] at 120 a.i/ha (292 panicles /m^2). However, panicle numbers in treatment T3 [imazapic+imazapyr] at 110 a.i/ha, similar to other herbicide treatments. The average data of 6 treatments was 310 panicles /m^2 and increased by 118% when compared to the untreated check. The 1000-grain weights were similar in all treatments including the check. Rice grain yields under all herbicide treatments were significantly greater than that of untreated check (0.88 t/ha) There was no significant difference amongst herbicide treatments in terms of yields. The average yield of 6 treated plots was 2.15 t/ha which was an increment of 144 % as when compared to that of the untreated check (0.88 t/ha).

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ALLELOPATHIC EFFECTS OF *Lantana camara* L. ON GERMINATION AND SEEDLING GROWTH OF *Ischaemum rugosum* Salisb., *Vernonia cinerea* (L.) Less. AND *Eleusine coracana* (L.) Gaertn.

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Abstract: Allelopathic effect of *Lantana camara* L. (Family: Verbenaceae) on algae, antimicrobial effects, germination and seedling establishment of some agricultural crops have been recognized. The present study was conducted to identify the effect of *L. camara* on the germination and seedling growth of some weeds such as *Ischaemum rugosum*, *Vernonia cinerea* and *Eleusine coracana* in the presence of Lantana allelochemicals. Laboratory tests were conducted with boiled and un-boiled Lantana extracts [1:5 and 1:10 v/v (extract:water)] prepared using dried leaves. Weed seeds on filter paper in Petri dishes were moistened with extracts where distilled water was used in the control. Treatments were replicated twenty times and arranged in Complete Randomized Design (CRD). The final germination percentage (FGP), and the lengths of shoots and roots were measured and number of seedlings with expanded leaves/cotyledons were recorded on the 19th day after germination. Glasshouse experiments were conducted using *L. camara* residues weighing 1, 2 and 4 g, each mixed with 85 g of herbicide free paddy soil. Lantana was not added to the control. Treatments were replicated ten times and arranged in a CRD. The number of germinated seedlings was recorded. The FGP, shoot and root development and expansion of leaves/ cotyledons for all of the weeds were significantly affected when exposed to high concentrations of *L. camara* extracts. Boiled extracts had a significantly greater effect on *V. cinerea* and *E. coracana* than the un-boiled extract. Glasshouse experiments showed a significant increase in germination of *E. coracana* when Lantana residue was added. Inhibition of germination and seedling growth of the above weeds under laboratory conditions highlighted the potential allelopathic effect of *L. camara* on selected weeds.

Key words: Allelopathy, Lantana, weeds

Introduction

Lantana camara L. (Family: Verbenaceae) is considered as an invasive woody shrub found throughout Sri Lanka. It's toxic properties against cattle, small ruminants (Ghisalberti, 2000), spores of liverwort *Asterella angusta* (Kothari and Chaudhary, 2001), cyanophyte *Microcystis aeruginosa* (Kong *et al.* 2006), antimicrobial activity on *Bacillus subtillis* and *Staphylococcus aureus* etc. have been well documented (Ioset *et al*. 2000; Misra and Laatsch, 2000). Further, the toxic effects brought out by allelopathy of Lantana on root growth suppression of agricultural crops such as soya bean and wheat have also been reported (Oudhia, 2000, Oudhia and Tripathi, 2000). On the other hand, some beneficial effects of *L. camara* have also been recognized. It is used, either alone or together with other plants such as *Croton lacciferus*, as mulch in paddy fields in the conventional agricultural practices to get rid of insect pests. Allelochemicals of Lantana contain Lantadene A and Lantadene B, volatile oils such as caryophyllene, cineol and pinene (Kong *et al*. 2006, Abdel-Hady *et al*. 2005). However, its' effect on weeds have received poor attention by researchers.

The present study was undertaken to reveal the alleopathic activity of *Lantana camara* on paddy field weeds. The study focuses on some important weeds such as *Ischaemum rugosum, Vernonia cinerea* and minor paddy field weed *Eleusine coracana*, which is found in the wet zone paddy fields but an important crop species in drier parts of Sri Lanka. The aim of the study was to identify the impact on germination and seedling growth of these weeds in the presence of Lantana allelochemicals.

Materials and Methods

Laboratory experiments on germination and seedling emergence
Lantana camara leaves were collected from various locations of the island and air dried for 2 weeks. Dried leaves were ground using a domestic grinder and passed through a 2 mm sieve. Two hundred g of Lantana residue was soaked in 1000 ml of distilled water and kept at 10ºC. On the 7th day, the mixture was filtered through four layers of muslin cloth to obtain an extract of concentration in the ratio of 1:5 (w/v) (extract:water). A portion of this was diluted to half the original concentration to obtain a 1:10 (v/v) extract. Part of each solution was boiled. Laboratory trials were conducted with boiled and un-boiled extracts. More than thousand seeds of a test weed species were subjected to a seed wash by placing the seeds on a layer of muslin cloth in a funnel and running tap water over it for approximately twenty minutes. The seeds were then blotted and dried on filter paper for half an hour. Ten of the washed seeds were arranged on dry Whatmann No. 1 filter paper in a 9 cm diameter glass Petri dish. Twenty of these Petri dishes were prepared and moistened with 1:10 boiled extract. Another 20 Petri dishes were treated with 1:10 un-boiled extract. The same procedure was carried out with the 1:5 boiled and un-boiled extracts using 20 Petri dishes for each treatment. Distilled water was used as a control. The replicated treatments were arranged in a Complete Randomized Design (CRD) at room temperature. The filter paper was moistened daily with the appropriate solutions. After four days germination was determined by counting the number of seeds germinated (radicle length of ≥2 mm). The results were recorded separately for each Petri dish.

The same methodology was used to asses the germination of three weed species *i.e.* *Ischaemum rugosum*, *Vernonia cinerea* and *Eleusine coracana*. Lengths of shoots and roots of the germinated seedlings were measured and the number of seedlings with expanded leaves/cotyledons was recorded on the 19th day after germination.

Glasshouse experiments on germination and seedling emergence
Germination of weed seeds was tested under glasshouse conditions using paddy soil containing different quantities of *L. camara* residues. Lantana residue weighing 1, 2 and 4 g were each mixed with 85 g of pesticide free paddy soil in plastic cups (65 mm in diameter and 35 mm in height) to obtain four different soil residue combinations. Control treatment contained paddy soil without Lantana residues. Ten seeds of each weed species were sown in separate cup, and treatments were replicated ten times. The cups were kept moist, and were arranged in CRD. Germination of seeds was monitored after 4 days until 22 days by recording the number of seedlings in each cup separately. Recorded seedlings were removed from each cup.

Data Analysis
Data analysis of the above two experiments were conducted using Analysis of Variance (ANOVA) using SPSS software (version 10) and the significance was tested using the Least Significant Difference (LSD) at $p=0.05$.

Results

Laboratory experiments on germination and seedling emergence of paddy field weeds.
The experiments showed that germination and seedling establishment of tested weed species was affected by *L. camara* (Table 1). A significant difference ($p<0.05$) was observed between the control and the two concentrations and/or the boiled, un-boiled nature of the extract. The most effective solution was the 1:5 w/v (high) concentration of aqueous Lantana extract.

Germination of *I. rugosum* was affected by the presence of the *L. camara* extract, but the strength of the extract or boiling had no significant effect on this species. A significant suppression in FGP was observed in seeds of *V. cinerea* and *E. coracana* species due to both the factors *i.e*. strength and boiling of the extract.

Table 1. Laboratory experiments on Final Germination Percentage of *I. rugosum V. cinerea* and *E. coracana* at high (1:5 v/v) and low (1: 10 v/v) concentrations of *L. camara* extract. [R = unboiled extract, B = boiled extract. ^{p,q,r} show significance between concentrations and ^{x,y,z} show significance between the boiled un-boiled nature of extract. Standard errors are given in parentheses at p = 0.05 level.]. Mean root, shoot and leaf numbers MRL = mean root length, MSL = mean shoot length, MLN = mean leaf number.

Conc. of Extracts	*I. rugosum*				*V. cinerea*				*E. coracana*			
	FGP (%)	MRL (%)	MSL (%)	MLN (%)	FGP (%)	MRL (%)	MSL (%)	MLN (%)	FGP (%)	MRL (%)	MSL (%)	MLN (%)
Control	$78.00^{x,p}$	$49.36^{x,p}$	$36.69^{x,p}$	$13.34^{x,p}$	$59.50^{x,p}$	$34.23^{x,p}$	$5.78^{x,p}$	$18.09^{x,p}$	$72.50^{x,p}$	$37.28^{x,p}$	$24.70^{x,p}$	$17.55^{x,p}$
	(±0.36)	(±0.33)	(±0.16)	(±0.11)	(±0.47)	(±0.29)	(±0.04)	(±0.11)	(±0.34)	(±0.13)	(±0.03)	(±0.04)
1: 10R	$36.00^{y,q}$	$17.18^{y,q}$	$32.36^{y,q}$	$9.64^{y,q}$	$21.50^{y,q}$	$3.60^{y,q}$	$4.07^{y,q}$	$2.00^{x,p}$	$78.00^{y,q}$	$51.24^{x,q}$	$25.30^{x,q}$	$14.86^{y,q}$
	(±0.52)	(±0.03)	(±0.32)	(±0.09)	(±0.32)	(±0.07)	(±0.07)	(±0.00)	(±0.30)	(±0.24)	(±0.06)	(±0.06)
1: 10B	$44.0^{y,q}$	$3.28^{y,r}$	$24.23^{y,r}$	$7.89^{y,r}$	$5.00^{y,r}$	$2.33^{y,q}$	$2.53^{y,q}$	$1.00^{x,q}$	$44.00^{y,r}$	$18.06^{x,r}$	$21.17^{x,r}$	$14.29^{y,r}$
	(±0.48)	(±0.04)	(±0.24)	(±0.10)	(±0.15)	(±0.04)	(±0.06)	(±0.23)	(±0.47)	(±0.15)	(±0.12)	(±0.09)
1: 5R	$38.50^{y,q}$	$2.32^{z,q}$	$22.84^{z,q}$	$6.16^{z,q}$	$0^{z,q}$	$0.10^{y,q}$	$0.00^{z,q}$	$0.00^{y,p}$	$18.00^{z,q}$	$6.21^{y,q}$	$12.86^{y,q}$	$9.14^{z,q}$
	(±0.39)	(±0.03)	(±0.23)	(±0.09)		(±0.01)	(±0.00)	(±0.00)	(±0.31)	(±0.12)	(±0.19)	(±0.17)
1: 5B	$23.00^{y,q}$	$0.25^{z,r}$	$6.81^{z,r}$	$1.07^{z,r}$	$0^{z,r}$	$0.15^{y,q}$	$0.00^{z,q}$	$0.00^{y,q}$	$0.50^{z,r}$	$0.00^{y,r}$	$0.05^{y,r}$	$0.00^{z,r}$
	(±0.32)	(±0.00)	(±0.26)	(±0.06)		(±0.01)	(±0.00)	(±0.00)	(±0.05)	(±0.00)	(±0.00)	(±0.00)

The root development in *I. rugosum* was significantly affected by both the concentration and boiling of extracts. Root development in *V. cinerea* though affected by the presence of extract was not affected by boiling of the extract, while that of *E. coracana* was affected by the higher concentration boiling of the extract. Shoot development in *I. rugosum* and *V. cinerea* were both affected by the strength of the Lantana extract, where higher concentration showed a greater effect. The shoot development of *E. coracana* was only affected by the higher concentration of extract. In the case of *V. cinerea,* boiling of the extract had no effect on shoot development, while the other two species were affected. The leaf development in all three species was affected by boiled extracts. In *V. cinerea* however there was no significance in the leaf number between seedlings exposed to the control and lower concentration of the Lantana extract though root and shoot development displayed a significant difference ($p<0.05$). Leaf development in *I. rugosum* and *E. coracana* species were affected by the strength of the Lantana extract.

Glasshouse experiments on germination and seedling emergence of paddy field weeds.
From the three weed species tested *E. coracana* showed a significant increase in germination in the presence of Lantana residues added to paddy soil. There was no effect of Lantana residues on the other two weed species (Table 2).

Discussion

Germination behavior of weeds differed depending on the experimental conditions *i.e.* when exposed to Lantana aqueous extracts on filter paper and when allowed to germinate with residues added to paddy soil. However, inhibition of germination and seedling growth of the above weeds in the presence of Lantana extract highlighted the potential allelopathic effect on some weeds.

Table 2. Glasshouse experiments on final percentage germination of *E. coracana*, *I. rugosum* and *V. cinerea* seeds exposed to different weights of lantana residue in soil. x, y, z show significant difference. Standard errors are given in parentheses at P = 0.05 level.

Weight of Lantana residue	Final germination %		
	I. rugosum	*V. cinerea*	*E. coracana*
Control	82.00^x (±0.36)	43.00^x (±0.42)	17.00^x (±0.54)
1 g	92.00^x (±0.25)	43.00^x (±0.68)	47.00^y (±0.56)
2 g	81.00^x (±0.31)	64.00^x (±0.73)	40.00^y (±0.67)
4 g	85.00^x (±0.40)	51.00^x (±0.64)	44.00^y (±0.60)

In contrast, the seed germination of *E. coracana*, which is also considered a minor crop in Sri Lanka, was susceptible to the allelopathic effects of Lantana.

Acknowledgements

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WEEDY RICE IN SOUTHERN SRI LANKA: ABUNDANCE, DIVERSITY AND FARMER AWARENESS

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Abstract: This study was carried out in southern Sri Lanka in the dry season 2007 to assess the abundance and diversity of the weedy rice and to evaluate farmer awareness regarding the situation. The weedy rice in the area showed a very high morphological diversity and eleven distinct types were identified based on the grain morphology. The yield losses caused by weedy rice is apparent and in highly infested fields the density of weedy rice plants found to be three times higher than the density of cultivated rice plants. Ninety five per cent of the farmers were aware of weedy rice and their negative effects on the yield but did not know how to manage it effectively.

Key words: Diversity, farmer awareness, Sri Lanka, weedy rice, yield loss

Introduction

Weedy rice, believed to be originated from crosses between wild and cultivated forms of rice, is becoming an immense problem in many rice growing countries in Asia and Africa. During the last two decades weedy rice has been recorded from several regions in Sri Lanka (Marambe and Amarasinghe, 2000). To date weedy rice has been reported from: Vavuniya, Batticaloa, Ampara, Kurunegala, Puttalam, Kuliyapitiya, Chilaw and Madampe. Recent observation indicated that weedy rice is spreading rapidly in lowland rice lands in Matara district (< 300m MSL; annual rain fall 1,200 – 2,400 mm) in the Southern Province of Sri Lanka. The nature of high grain shattering makes weedy rice control extremely difficult and challenging. Hence, this study was undertaken to evaluate the current situation of weedy rice in Matara district in the southern province of Sri Lanka, emphasizing its abundance, diversity and farmers' awareness and to help design a strategy to control this problem.

Materials and Methods

Five rice growing villages in the Matara district in the southern province of Sri Lanka, namely Kamburupitiya, Kapudoowa, Thihagoda, Hathamuna and Yatiyana were selected for sampling and field observations. Twenty leading farmers in these villages were interviewed to obtain their views about weedy rice. Abundant and prominent weedy rice plants were uprooted for laboratory observations and characterization. Quadrate sampling was made to assess the abundance and diversity of the weedy rice in selected rice lands, and rice yield was estimated under different weedy rice densities.

Results and Discussion

Presence of weedy and wild rices with cultivated rice
The rice lands in the area are irrigated through perennial streams and direct seeding of sprouted seeds was the main crop establishment method. More than 90% of the rice lands were grown to Bg 379-2, a 120 day-old improved variety. The weedy rice types observed in Matara were morphologically closer to cultivated rice unlike those present in other areas of the country. Although some of the wild rice species like *Oryza nivara, Oryza rufipogen* and *Oryza rhizomatis* was found to occur in association with weedy rice in other regions of the country (Abeysekera, 2006), the authors did not observe wild rice species in paddy fields in

the Matara district. However, *O. nivara* was found to grow in irrigation channels nearby some paddy fields in our study areas.

Agro-Morphological features of weedy rice

In general, the weedy rice infested fields were distinctly visible by the taller plant stratum of weedy type above the cultivated varieties (about 90cm). These weedy rice types showed a very high degree of morphological variations. The height at maturity ranged from < 90cm to 160cm, the flag leaf length from 24 cm to 68 cm and the flag leaf angle from erect to horizontal. Panicle exertion varied from enclosed to well-exerted.

Morphological variation in panicle characteristics

The panicle length of the above weedy rice types ranged from 20 cm to 45 cm. The variations of the grain characters were remarkably high. Based on these characters 11 distinct and abundant weedy rice types were identified from the study area (Table 1). The grain shape ranged from long to intermediate and the lemma and palea colour from black to straw (data not shown). Among the weedy rice types there were both awned and non-awned grains. The awn lengths ranged from 7cm to 0.1cm and the colour from straw to dark brown. Furthermore, there was a high variation of the awn length within a panicle of the same weedy rice type (Plate 1).

Table 1. Diversity of grain morphological features of the selected weedy rice types

Type	Awn	Apiculus colour	Lemma and palea colour
1	Long	Purple	Black
2	Long	Purple	Brown
3	Long	Brown	Brown spots on straw
4	Long	Straw	Straw
5	Short	Purple	Straw
6	Short	Brown	Brown
7	Short	Straw	Straw
8	None	Purple	Black
9	None	Purple	Brown
10	None	Purple	Straw
11	None	Brown	Brown

A very high degree of unevenness was seen in the grain maturity within a panicle. As a result some grains of the panicle remained green while the rest ripened and shattered. More prominently, these weedy rice types showed moderate to high grain shattering as compared to cultivated varieties. Since the mature grains shattered by wind panicles having a few un-ripened grains is a characteristic appearance in the field.

Plant density and yield loss

Almost all the rice lands observed in the study were infested with weedy rice with a variable density from field to field and the density of the weedy rice plants varied from 12 to 360 plants per m^2 in the infested fields. In highly infested fields the density of weedy rice plants was three times higher than the number of cultivated rice plants. Farmers had to abandon the crop in some rice fields that are heavily infested by weedy rice. The effect of the presence of weedy rice on the rice yield is apparent (R^2= 0.71) and the estimated paddy yield under different densities of weedy rice is shown in the Figure 1.

Plate 1. Variation of awn length within a panicle

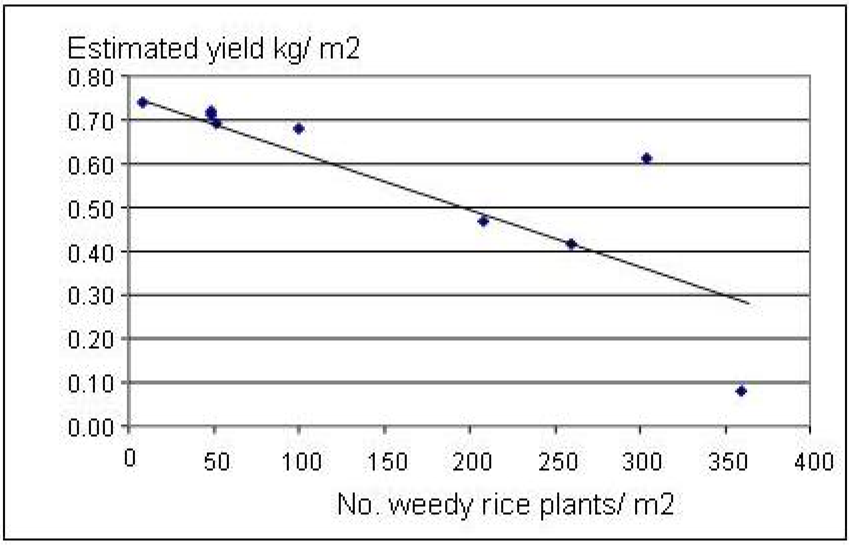

Figure 1. The estimated paddy yield under different densities of weedy rice

Farmer awareness

In Matara district, weedy rice is commonly known as '*Wal Goyam*' (weedy rice) or as '*Kalawam Wee*' (mixed rice). Ninety five per cent of the farmers were aware of weedy rice and their negative effect on rice yields. Some farmers have given up rice cultivation as they could not control the weedy rice. Farmers were not aware of any effective management strategy for weedy rice. Sixty three per cent of the farmers uprooted taller plants at the vegetative stage or/and slashed the upper portion of the plants with the panicle before maturity. These practices are labour intensive, expensive and time consuming and hence impracticable for large scale operations. Therefore, farmers resolved to seek immediate government intervention to solve this problem.

Future scope

The occurrence, distribution and diversity of weedy rice in cultivated rice ecosystems are still left behind to be revealed. The genetics of these rice types and their inter-relationship with the cultivated varieties and wild rice species are not known. The diversity of the weedy rice in southern Sri Lanka should be studied in detail using both morphological and molecular techniques. The features of weedy rice, which would be important in handling them as competitive weed plants, such as seed viability, longevity, dormancy, shattering pattern and soil seed bank should be studied. Furthermore, the attention of the relevant authorities must be immediately focused to develop a sustainable strategy to eliminate weedy rice in order to secure paddy cultivation in Sri Lanka.

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EFFECTS OF DIFFERENT WEED CONTROL METHODS IN CINNAMON (*Cinnamomum verum* Presl.)

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Abstract: Improper weed control is one of the major factors that affects the bark yield and bark quality of cinnamon. Field experiments were conducted to compare the effect of chemical and manual weeding on subsequent weed growth in cinnamon fields at the Cinnamon Research Station in the southern province of Sri Lanka. In the first experiment, weed biomass was measured once in every two month intervals after treatments for a period of one year. From the eighth month onwards, weed biomass increased rapidly and by the end of the year, no significant difference in weed biomass was observed among the treatments. The second experiment was laid out in the Cinnamon Research Station to determine the effects of glyphosate (Roundup®) and paraquat (Gramoxone®), with the use of different nozzles for herbicide application namely open nozzle and guarded nozzle, together with manual weeding using mammoty, on bark yield, yield parameters and time taken for peeling of the bark. Treatments were applied twice a year. A significantly higher dry bark yield of 664 kg ha^{-1} yr^{-1} was recorded with manual weeding when compared to chemical weeding and the control. The lowest dry bark yield (340 kg ha^{-1} yr^{-1}) and higher time requirement for peeling of one kg of bark (111.5 min) were observed with the herbicide paraquat applied using an open nozzle. The highest number of harvestable stems per ha (15,064), was recorded with manual weeding and the lowest amount of 8,532 stems per ha was recorded from paraquat applied with an open nozzle. A significantly higher number of new shoots was observed in the manually weeded plots. Stem diameter and stem length were comparatively higher in manual weeding than other weed control methods. Based on the results and consideration of the environmental impacts, manual weeding twice a year using a mammoty is recommended as the most suitable weeding practice for cinnamon.

Key words: Biomass, cinnamon, glyphosate, paraquat, mammoty

Introduction

A weed is a plant but considered undesirable for various reasons mostly connected with decreased economic return from the land. They compete with cinnamon plants for moisture, light and nutrients and hinder field operations such as harvesting, pruning and fertilizer application. Cinnamon (*Cinnamomum verum* Persl. 2n=24) is an indigenous spice crop in Sri Lanka and it is one of the major spice crops in export economy. Currently farmers are facing many difficulties with weed growth in the fields of cinnamon cultivation. Their presence in and around cinnamon lands, directly reduces crop yield and also indirectly reduces bark quality of cinnamon. When the cinnamon crop is grown despite the presence of weeds, field operations including harvesting are difficult.

Weeds become a serious problem when cinnamon plants are at the younger stage. Plant growth is decreased significantly because of competition with weeds. Weeds usually absorb mineral nutrients and moisture faster than young cinnamon plants. On the other hand, young cinnamon plants are badly affected by the shading effects of weeds.

Therefore, cinnamon farmers should be trained to practice weed control methods. In mature cinnamon cultivation, weed problems can be minimized with the recommended management practices. Among the techniques used, the commonly used herbicides in cinnamon lands are Roundup® (glyphosate) and Gramoxone® (paraquat). However, being major export commodity of Sri Lanka, the presence of residues of these chemicals in cinnamon is a major concern as three an increasing demand for chemical-free products in the world market. Hence, the present study was carried out to evaluate different weed management strategies in cinnamon to recommend judicious and environmentally friendly practices to the growers.

Materials and Methods

Experiment 1

Field was selected in Cinnamon Research Station and plot size was arranged as 5 m x 5 m with three replicates. Data was recorded once every two months over a one year period. Weed biomass was measured in separate plots and weed species were counted separately and preserved for identification.

Experiment 2

The second experiment was laid out in a spilt plot design with different control measures of weeds. The treatments were replicated thrice. Plot size was the same as in the first experiment. In this experiment, two herbicides namely, glyphosate (Roundup®) and paraquat (Gramoxone®) were applied using two nozzle types namely, open nozzle and guarded nozzle for weed control, which were compared with manual weeding methods and an un-weeded prior to applying treatments, and twice a year thereafter. The average bark yield, time requirement for peeling of one kg of bark, amount of harvestable stems, number of new shoot penetration, stem diameter and stem length were measured during the last three years during the period where following treatment combinations were applied, namely T_1 - glyphosate/open nozzle, T_2 - glyphosate/guarded nozzle, T_3 - paraquat/open nozzle, T_4 - paraquat/guarded nozzle, T_5 - control (No weeding) and T_6 - manual weeding with mammoty.

Results and Discussion

Experiment 1

The weed flora in the experimental site is given in Table 1. Up to six months from application of treatments, weed biomass (wet basis) was less for manual weeding when compared to chemical weeding (Table 2).

Table 2. Common weed flora in cinnamon plantation

Common Name	Scientific Name	Common Name	Scientific Name
Wathupalu	*Micania cordata*	Kadupahara	*Emilia sonchifolia*
Fox tail	*Pennisetum polystachion*	Heen Undupiyaliya	*Desmodium triflorum*
Podisingchomaran	*Chromalaena odorata*	Thuththiri	*Chrysopogen aciculatus*
Patta apela	*Urena lobata*	Balathana	*Eleusine indica*
Monarakudumbiya	*Vernonia cinerea*	Gotukola	*Centella asiatica*
Maduwel	*Merrima tridentate*	Wal penala	*Cardiospermum halcacabum*
		Kuppameniya	*Acalypha indica*
		Balu-nakuta	*Stachytapheta urticaefolia*

Thereafter, the weed biomass increased rapidly under both manual and chemical weeding, thus resulting in a reduction in cinnamon bark yield. Competition for nutrient, water and light, mainly from creepers is the main factor responsible for the low crop yield. Quality of the cinnamon bark was also reduced due to weeds.

Experiment 2

The highest dry bark yield (664 kg ha^{-1} yr^{-1}) was recorded with manual weeding (Figure 1), which was significantly higher ($p<0.05$) from the rest of the weed control methods. The lowest dry bark yield (340 kg ha^{-1} yr^{-1}) was recorded with application of paraquat using an open nozzle. Moreover, it took more time to peel off the bark (Figure 2). Number of stems per hectare and the number of shoots per hectare were significantly higher with mammoty weeding when compared to chemical weeding and control plots (Figures 3 and 4). The stem diameter (Figure 5) and stem length (Figure 6) were also comparatively higher in manual weeding.

Table 2. Weed biomass (kg/ha) under different weed control methods

Treatments	Fresh weight of weed biomass after treatment application					
	2 Months	4 Months	6 Months	8 Months	10 Months	12Months
Manual	8011 c	8715 c	8936 c	23319 b	33473 b	39853 a
Chemical	8374 c	8495 c	9731 c	21569 b	37823 b	38475 a
Control	33378 a	47185 a	39435 a	42204 a	43748 a	42895 a

Within a column, means followed by the same letter are not significantly different at p=0.05.

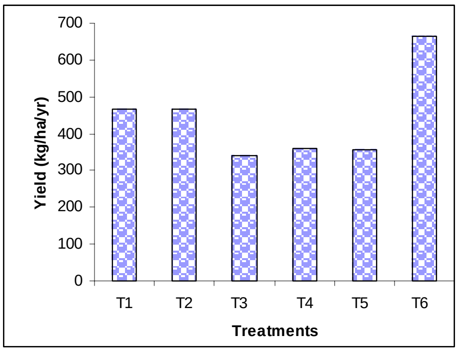

Figure 1. Dry bark yield with different control methods

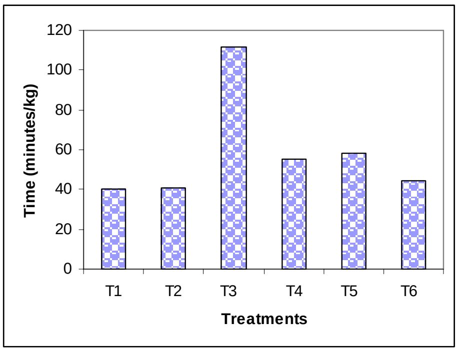

Figure 2. Time requirement for peel off the one kg of wet bark

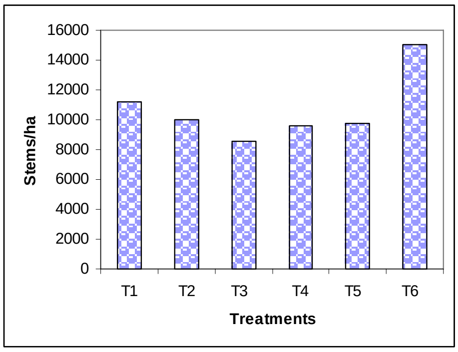

Figure 3. Amount of harvestable stems with different weed control methods

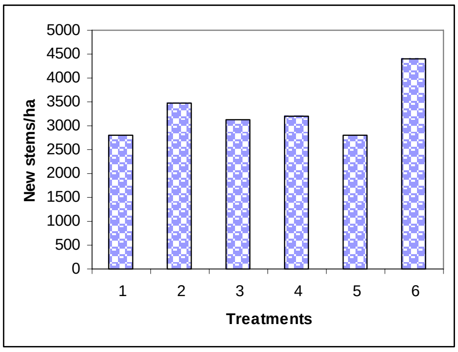

Figure 4. Number of new shoots penetration from bushes

The results suggested that the most effective weed control method and frequency was manual weeding twice a year. Chemical weeding badly affects the formation of new shoots and also the ability of peeling.

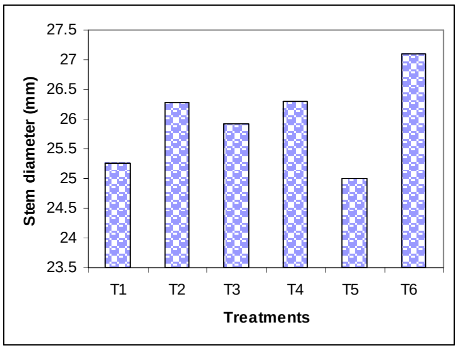

Figure 5. Diameter of harvestable stems

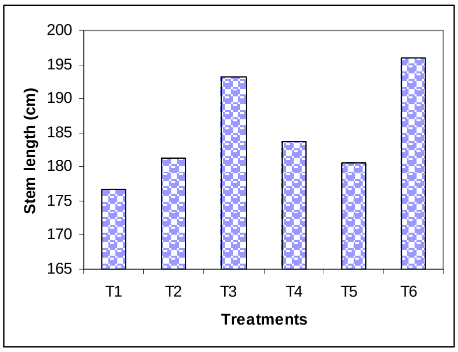

Figure 6. Length of harvestable stems

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OPTIMISATION OF HERBICIDE DEPOSITION AND RETENTION USING COMPUTER MODELLING VALIDATED THROUGH WIND TUNNEL STUDIES

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Abstract: Herbicides are widely used, to control weeds in most agricultural systems world wide. In recent years, there has been increasing interest in the use of air induction nozzles rather than traditional or low-drift (pre-orifice) flat fan nozzles for the application of herbicides. This paper reports on a project measuring spray deposition and herbicide efficacy on a range of weeds species sprayed using air induction and extended range flat fan nozzles. Three dimensional plant architectural models are being developed for these weed species using the Lindenmayer systems (L-systems) formalism and programmed using L-studio software. A spray droplet trajectory model has been incorporated with the plant architecture model and linked to an environmental program to determine the interception of spray droplets by the various vegetative elements. Algorithms have also been introduced to investigate the influence of droplet splash and bounce on spray deposition. Results from wind tunnel and field trials are being used to refine and validate the combined plant architecture and spray model.

Key words: Herbicide, model, spray

Introduction

Herbicides play an important role in the control of weeds in most agricultural systems world wide. They are generally applied as a spray to cover the weed or soil surface with pesticide-laden droplets. Spray may however, be lost to non-target areas within a crop through deposition on to non-target plant surfaces or on the soil. The plant architecture of the crop and weed species can influence the distribution of the spray droplets, which can in turn influence the efficacy of the herbicide. The action of wind may also result in spray moving away from the spray area.

In recent years, there has been an increasing interest in the use of air induction nozzles rather than traditional or low-drift (pre-orifice) flat fan nozzles for the application of herbicides. These nozzles use a venturi action to draw in air and can produce a spray of droplets containing air pockets which are commonly termed as 'air inclusions' (Buttler-Ellis *et al.*, 2002). The larger droplet sizes that can be generated by air induction nozzles have been shown to reduce drift compared to conventional flat fan nozzles. However, there have been concerns that the larger droplet size may reduce spray coverage and hence herbicide efficacy.

By selecting and using spray equipment and techniques that maximise deposition of pesticides onto the target, it is possible to both maximise the effectiveness of the pesticide application and reduce the amount of off-target deposition and damage. In this paper a droplet movement (spray) model has been combined with a three-dimensional plant architecture model to give a probabilistic model of turbulence-related spray transport around various plant architectures(Dorr *et al.*, 2005; Dorr *et al.*, 2006b). Measurements of pesticide droplet interactions with the crop canopy from wind tunnel and field studies are being used to refine and validate the combined spray and plant architecture model.

Materials and Methods

Spray Deposition

A variety of weed species including Sow thistle (*Sonchus oleraceus*), Fleabane (*Conyza* spp.), Barnyardgrass (*Echinochloa* spp.), Wild oats (*Avena fatua*) and Bindweed (*Convolvulus arvensis*) plants were grown in 150 mm pots. A three-dimensional sonic digitizer (Freepoint XL2) was used to map the plants at the time of spraying to assist in the development of the plant architectural models.

Table 1 shows the weed species, stage of growth and herbicides used. Both an extended range flat fan nozzle and an air induction nozzle were used for each treatment (Table 2). A fluorescent tracer (pyranine) was added to the spray mix at a rate of 0.3g/l.

Table 1. Weeds sprayed and method of application

Treat No.	Weed	Stage of Growth When Sprayed	Method Sprayed	Herbicide	Herbicide Rate (l/ha)	Total volume (l/ha)
1	Sowthistle	Advanced Rosette	Wind Tunnel	Roundup CT [1]	2.0	57
2	Sowthistle	Advanced Rosette	Wind Tunnel	Roundup CT [1]	2.0	86
3	Sowthistle	Rosette	Field	Roundup CT [1]	1.0	57
4	Fleabane	Rosette	Field	Roundup CT [1]	1.0	57
5	Bindweed	3-4 Leaf	Wind Tunnel	Starane 200 [2]	0.75	57
6	Barnyardgrass	Tillering to Seeding	Wind Tunnel	Roundup CT [1]	2.0	86
7	Wild Oats	2 Leaf	Field	Wildcat 110EC [3]	0.175	57

[1] 450 g/l glyphosate [2] 200g/l fluroxypyr [3] 110g/l fenoxaprop-p-ethyl

The weeds were sprayed either in the low-speed working section of the wind tunnel research facility at the University of Queensland Gatton Campus or in the field by a vehicle-towed plot sprayer. The wind tunnel has an electronically controlled traversing mechanism to move the spray boom at a constant velocity along the length of the working section. For each treatment about ten pots were positioned in a single row along the sprayer direction and sprayed with one nozzle type and then another set of plants positioned and sprayed with the other nozzle type.

Table 2. Nozzle and operating parameters used to apply the herbicide

Treat No*	Extended Range Flat Fan Nozzle			Air Induction Nozzle		
	Nozzle	Pressure (bar)	Speed (km/h)	Nozzle	Pressure (bar)	Speed (km/h)
1	XR110015	2.75	11.4	TDCFFC110015	5.0	16
2	XR110015	2.75	7.4	TDCFFC110015	5.0	10.8
3	XR11002	2.5	15	TDCFFC11002	5.0	21.2
4	XR11002	2.5	15	TDCFFC11002	5.0	21.2
5	XR110015	2.75	11.4	TDCFFC110015	5.0	16
6	XR110015	2.75	7.4	TDCFFC110015	5.0	10.8
7	XR11002	2.5	15	TDCFFC11002	5.0	21.2

*Refer to Table 1 for treatments.

After spraying, half the pots were left to grow under field conditions to assess spray efficacy.The remainder were used to assess the amount of dye retained on the plant surface. All the above ground parts of the weeds were placed in a plastic bag, 60 ml of deionised water was added to the sample and the bag shaken. Concentration of dye was measured by a calibrated fluorometer (Turner-Sequia model 450).

After the dye was removed from the sample the sow thistle leaves were removed, arranged flat on a stand, a photograph was taken and image analysis software (Image Pro v 5.0) was used to measure the surface area of the sample. The amount of dye per unit area was expressed as a percentage of the application rate to enable the two different rates to be compared.

Spray efficacy

Spray efficacy was initially assessed as the percent mortality resulting from the herbicide application. In general a low herbicide rate was used in an effort to determine difference in efficacy for the two nozzle types. In those treatments where there was no mortality other methods were used to give an indication of plant vigor. For the fleabane (Treatment 4), the weeds were dried in an oven and dry weight measured. For sowthistle, the number of capitula per plant was counted and for wild oats the number of seed per plant was counted.

Plant architectural model

The spray model in this paper (Dorr *et al.*, 2006a), used on a combined ballistic and random walk approach, was similar to that used by Mokeba *et al* (1997) and Cox *et al* (2000). The trajectory of each droplet is followed as it moves through the atmosphere by dividing it into a large number of small discrete time steps during which the velocity components (u, v, w) of the particle are kept constant. A meaningful estimate of dispersal statistics can be obtained by following a large number of trajectories (Hashem and Parkin, 1991). The model includes algorithms to account for droplet evaporation (Cox et al., 2000), entrained air and movement of air around the spray (Ghosh and Hunt, 1998), droplet splash and retention. The spray input parameters used in the model were based on experimental measurements undertaken by the Centre for Pesticide Application and Safety (data not shown).

The plant architecture model utilises a set of growth rules expressed in the Lindenmayer systems (L-systems) formalism (Prusinkiewiez *et al.*, 2000). Existing models of plants, for example sowthistle (Cici, 2007) have been sourced where possible and inserted into the spray model as sub L-systems (Dorr *et al.*, 2006a). The spray and plant architecture program is linked to an environment program that determines when droplets are intercepted by specified plant components such as leaflets. The droplets are stopped at the point of impact, and the leaflets coloured to visualise the amount of spray captured.

Results and Discussion

Spray Deposition

Average spray deposit for each treatments are shown in Figure 1. The error bars indicates the standard deviation of the measurements. A two-sample T-Test was conducted to compare the nozzle types for each treatment and nozzle type was only significantly different ($p=0.003$) for fleabane (Treatment 4).

Herbicide efficacy

Table 3 shows the environmental conditions at the time of spraying and the herbicide efficacy. There was no significant difference due to nozzle type on herbicide efficacy.

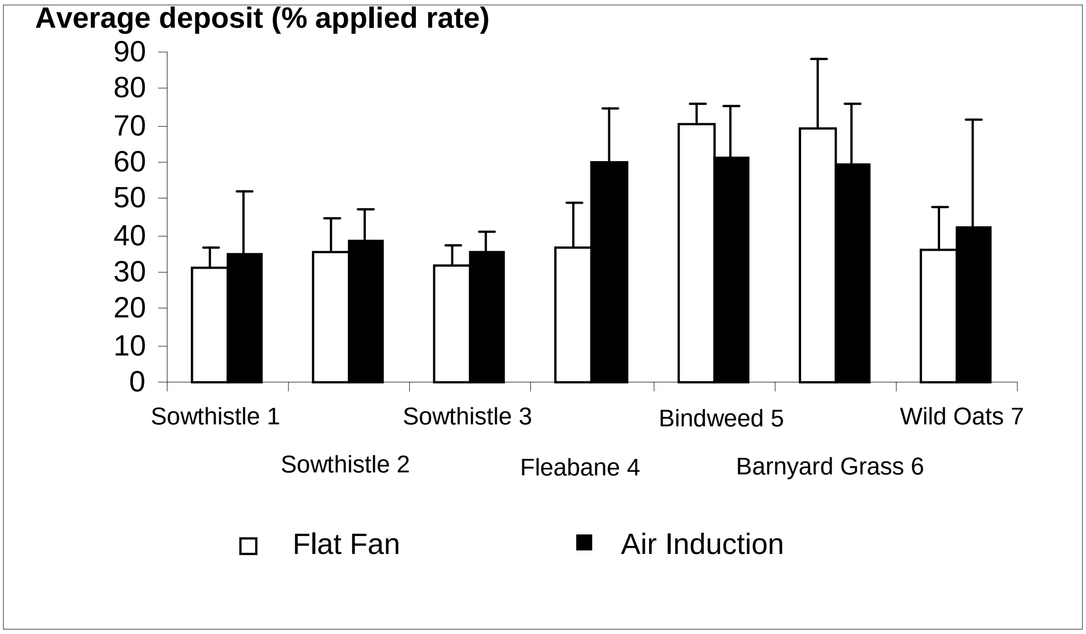

Figure 1. Comparison of measured spray deposition on weeds sprayed with extended range flat fan and air induction nozzles. Error bars show standard deviation.

Plant architectural model

The plant architectural models for fleabane and grass weeds are still under development so only the results from the sow thistle treatments are presented. Since the model has a random walk component a different result is obtained each time the model is run. The model was therefore run five times for each treatment (with approximately 10,000 droplets tracked per run) and the average deposit and standard error of the results also shown in Figure 2. The average deposit from the model agrees well with the experimental measurements. Figure 3 shows an animation from the model.

Table 3. Environmental conditions and herbicide efficacy for each treatment

Treatment No.*	Date Sprayed	Wind Speed (m/s)	Temperature (°C)	Relative Humidity (%)	Efficacy (% mortality) XR	TD
1	27/07/2006	2	23	59	100	100
2	27/07/2006	2	23	59	100	100
3	9/01/2007	3.6	23	69	0 (26)[1]	0 (29)[1]
4	22/12/2006	2.2	24	65	0 (2.8)[2]	0 (2.8)[2]
5	28/07/2006	2	19	89	100	100
6	27/07/2006	2	23	59	100	100
7	22/12/2006	2.2	25	64	0 (83)[3]	0 (30)[3]

[1]number of capitula per plant; [2]plant dry weight (g); [3]number of seeds per plant; *Refer Table 1 for the treatments.

For the weed species and growth stages tested there was no difference between extended range nozzles and air induction nozzles on herbicide efficacy, measured amount of spray deposited on weeds or modelled spray deposit on weeds. In future research, the resulting spray models will be extended to explore optimization of herbicide deposition onto weed surfaces.

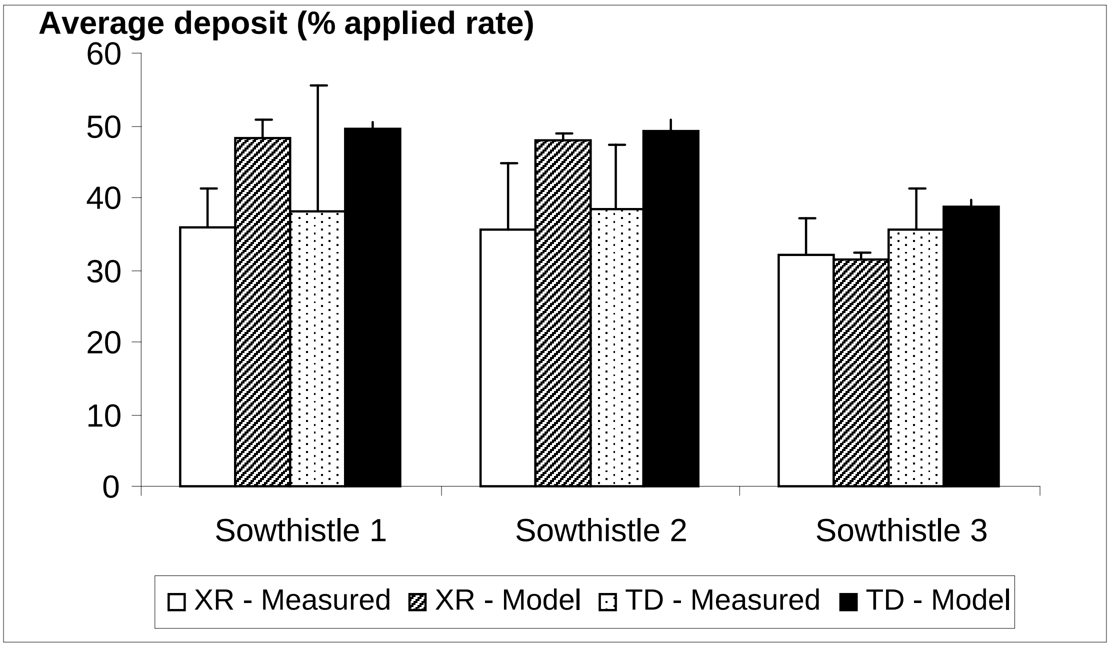

Figure 2. Comparison of measured and modeled spray deposition on sow thistle sprayed with extended range flat fan (XR) and air inductions (TD) nozzles. Error bars show the standard deviation of the results.

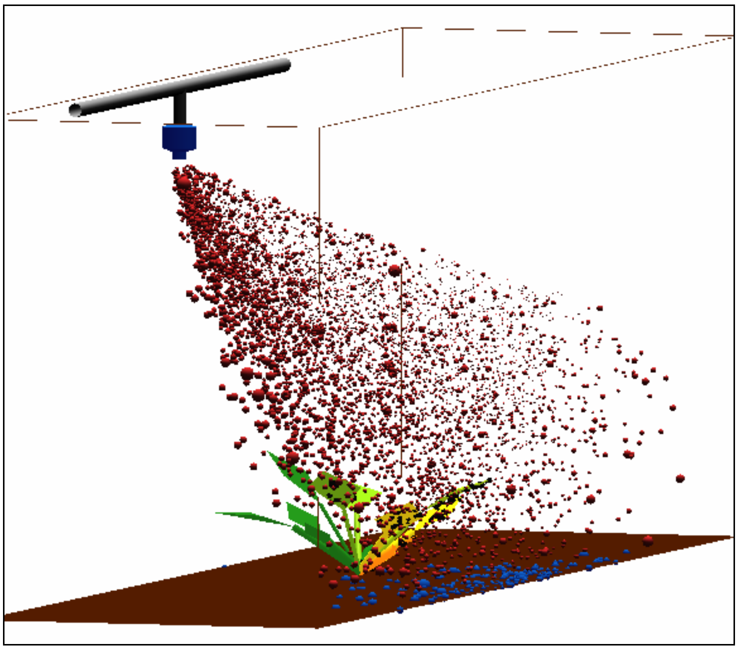

Figure 3. L-Studio simulation of sow thistle plant being sprayed in the wind tunnel.

Acknowledgements

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MODELLING THE EFFECTS OF FARM MANAGEMENT ON THE SPREAD OF HERBICIDE RESISTANCE

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Abstract: Herbicide resistance is an increasing problem in Australian cropping systems, but little is known about how resistance spreads and how farmers can manage their paddocks to minimise its spread. This paper presents a modelling framework for predicting biological processes in agriculture, with particular emphasis on the spatial and temporal spread of herbicide resistance. It includes a model of the population dynamics of weeds growing in competition with crops, a polygenic model of the development of herbicide resistance and gene transfer by means of seed and pollen movement. The modelling framework is used to predict the long-term spread of resistant weeds given different integrated weed management choices combining tillage and herbicide treatments, as well as new technologies such as seed capture at harvest and precision planting systems that allow different treatments for inter and intra-rows. The model's predictions are used to devise management options that minimise the spread of herbicide resistant weeds.

Key words: Resistance spread, modelling, gene transfer.

Introduction

The evolution of herbicide resistance depends on the intensity of selection, the frequency of resistance alleles in natural (unselected) populations, the mode of inheritance of resistance, fitness penalties associated with resistance and the spread of resistance by gene flow within and between populations (Diggle and Neve, 2001). Modelling approaches that simulate the evolutionary processes that result in resistance can be used to assess the impacts of weed management technologies including crop rotation, spray strategies, precision farming and wide-row (tram-line) systems that allow different treatments for inter and intra-rows. By simulating the population dynamics of weeds growing in crops, the long-term development and spread of resistance under different farm management practices are predicted and the model's predictions used to devise strategies to minimise spread.

This paper presents a modelling framework for predicting spread of herbicide resistance. It considers the most widespread and problematic weed in Australian agriculture, annual ryegrass (*Lolium rigidum*). Annual ryegrass is known to be resistant to herbicides with nine different modes of action in Australia, including glyphosate. Glyphosate is one of the most valuable herbicides in AustraliaN agricultural systems. This paper uses the spread modelling framework to predict the spread of glyphosate resistance in ryegrass by pollen and seed movement.

Data Types

The modelling framework is implemented as a *Mathematica* Version 5.2 package. The data types have been designed as a cascading sequence. Described from the top-level to the bottom, a map is a sparse data representation that is composed of a list of cells and a reference state. The map only lists cells that are of interest. That is, cells that contain resistance. Each cell contains information about its current state and location. The map's reference state describes the situation in all locations that are not yet of interest. The state of a cell is composed of a community of species populations and information about the soil and any herbicide residues that are present in the soil at a particular time. Each population is separated into four components: the seed bank, plants, seed on plants and pollen. Each of these components comprises of a set of cohorts. A cohort is identified by the time of its creation. For example, plant cohorts are identified by their germination time, seed on plant cohorts are identified by the time of seed set and seed cohorts are identified by the time of seed rain. At the lowest level, each cohort is composed of a set of genotypes. Genotypes are stored in sparse sets. This allows any number of alleles to be included in the model. For example, consider the case where there are six alleles that may confer resistance. If the resistance alleles are rare, then many of the possible combinations of alleles will not be present in any cohort. The sparse set representation stores only a list of those combinations that are present in the cohort, and a count of the number of times it occurs.

Overview of the Dynamic Model

The dynamic life-cycle model simulates the population biology of a crop and several weed species growing together. Its general structure (Figure 1) is similar to existing weed life-cycle models such as those used by Colbach *et al.* (2001), Diggle *et al.* (2002), and Doyle (1991). Holst *et al.* (2007) provide a recent review of dynamic life-cycle models.

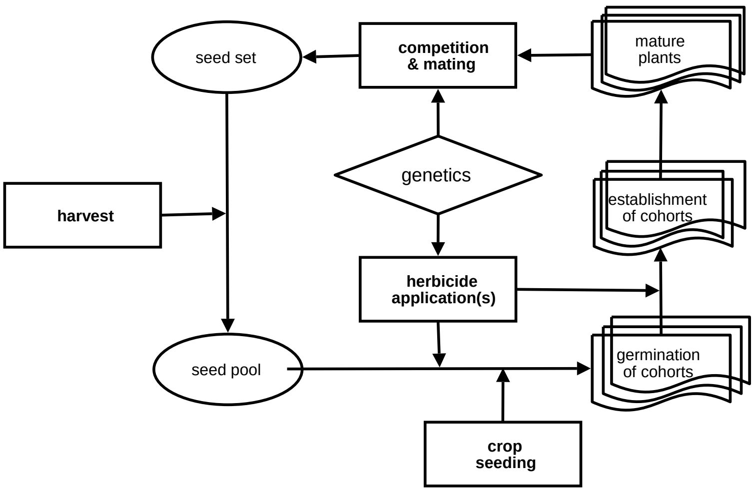


Figure 1. Overview of the dynamic life-cycle model

The starting point for all simulations is the initial seed bank. Crop seeds are added to the seed bank. Seeds germinate and seedling cohorts become established. Herbicides may be applied at various times, affecting seedling germination and survival. Some seedlings survive to become mature plants that flower and set seed. The number of seed set is affected by the amount of competition from other plants. Crop seed is harvested and weed seed fall to replenish the seed bank. All annual plants die at the end of each year and the seed bank declines over time due to natural causes. The genetics of individual plants affect their response to herbicide applications, and are considered explicitly when plants mate to produce new seed. After the first year of simulation, the seed bank is also separated into cohorts. This allows the future inclusion of modifications to the model to allow seed viability to vary with age.

The model is specified by the number of years to be simulated and a given crop rotation (for example, wheat – lupin – wheat – canola). For each crop, a sequence of events is specified that includes both life-cycle and management events. The life-cycle events are the same for each crop, but the management events may differ. Events are specified to occur at particular times for each year with respect to an ‘average’ season. The parameters corresponding to different events (Table 1) depend on the species, herbicides applied and time of year.

Table 1. Annual life-cycle and management events

Event	Parameters
germinate	Probability of germination and survival.
spray	Probability of killed (adjusted for resistance alleles).
germinate	Probability of germination and survival.
seed	Number seeded per square metre.
germinate	Probability of germination and survival.
spray	Probability of killed (adjusted for resistance alleles).
germinate	Probability of germination and survival.
seed bank survival	Probability of post-winter seed survival.
seed set & mating	Calculated using a competition sub-model.
harvest	Probability that seed are collected by the harvester; Probability that seed are exported with straw; Probability that seed are exported as chaff; Probability that chaff is collected (eg. by chaff cart).
seed rain	All remaining seed on plants are added to the seed bank.
senescence	All annual plants die at the end of the year.
seed pool survival	Probability of post-summer survival.

The modelling framework is fully stochastic. That is, if an event requires the selection of individuals (seed, grains of pollen, plants or seed set on plants) with a given rate or probability *p*, then the number selected, n_t, is assumed to have a Binomial distribution,

$$n_t = Bin\,(n_{t-1}, p)$$

where, n_{t-1} is the number the event selects. In most dynamic life-cycle models, a deterministic approach is taken. That is, n_t is set to be the mean of the Binomial distribution, $n_t p$. In contrast, the stochastic approach sets n_t to be a random sample generated from the Binomial distribution. Where the deterministic approach always gives the mean result, the stochastic approach gives different values for n_t each time the model is run. By performing multiple simulations of the model, the stochastic approach enables the simulation of rare (but possibly important) events.

Competition, seed set and mating sub-model

The numbers of seed set by each plant cohort is calculated using the age-structured hyperbolic competition model of Neve *et al.* (2003). Following Diggle *et al.* (2002), ovules and pollen are produced in direct proportion to predicted seed yields and all gamete haplotypes have an equal chance of reproductive success. The pollen and ovule haplotypes recombine at random to produce diploid zygotes that develop into mature seed.

Pollen spread by wind

Because of the importance of rare long-distance dispersals of both seed and pollen, dispersal models must be able to describe and predict these occurrences. While the scale and fine details of dispersal curves vary considerably amongst species, their general shapes remain similar: at one end, there is an abundance of relatively short dispersal distances whereas at the other end, there is a scarcity of relatively long-distance dispersals (Nathan *et al.* 2003). However, there is little data available for quantifying long-distance seed dispersal, and model-fitting is often driven by the large number of short-distance dispersal events, so that the tail of the dispersal curve may be poorly estimated.

Given the scarcity of dispersal data, we used a mechanistic model to generate a large set of dispersal distances, and then tested statistical approaches for both describing the generated data and for simulating dispersal distances. We implemented the Jongejans and Schippers (1999) model in *Mathematica* and used to generate 10^6 seed flights. The terminal velocity and vegetation height used correspond to those of annual ryegrass. Of the models tested (exponential, normal, lognormal, normal mixture, exponential mixture, Weibull, Cauchy and log-lognormal), the Cauchy distribution provided the best balance between goodness of fit and capacity for predicting long-distance dispersals. To simulate pollen spread, we therefore use a product distribution of a circular uniform distribution for direction and a Cauchy distribution for dispersal distance. To allow for the effect of prevailing wind directions, a product distribution using a Von Mises distribution for wind direction is also implemented (Figure 2).

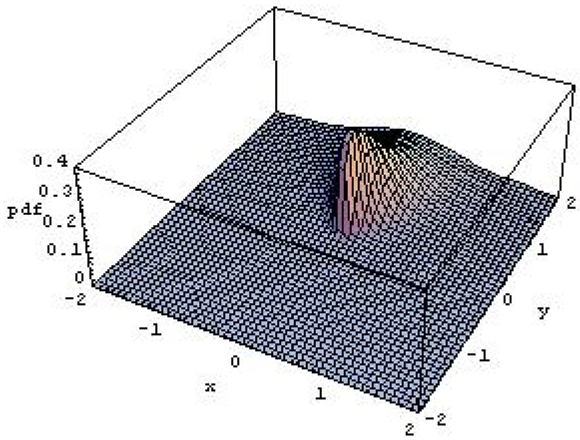


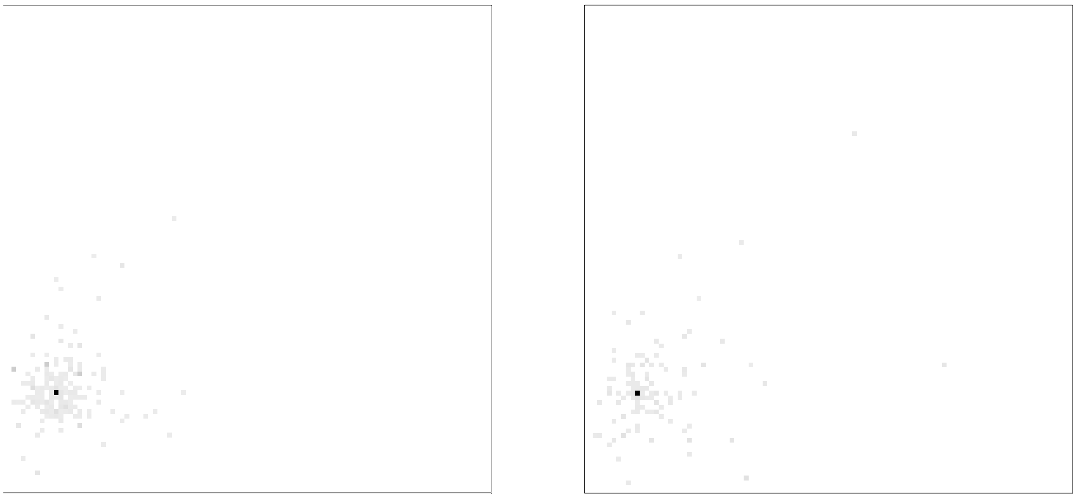
Figure 2. Modelling dispersal given prevailing wind direction

Spread of glyphosate resistance by pollen movement

This section presents the results of modelling spread of glyphosate resistance in annual ryegrass, that has been modelled under two management scenarios. The first applies an annual pre-sowing application of glyphosate as a knockdown herbicide. The second replaces the annual glyphosate application with simazine. In each case, a wheat – lupin rotation is adopted, and 20 year simulations were performed. A single major gene is assumed, so that the presence of a resistance allele confers full resistance to glyphosate. Each model was initialized with a single patch (one 10 square metre cell) that contained resistant seeds at a rate of one in ten. The background non-selected population was assumed to contain resistance alleles at a rate of one in one million as per Neve *et al.* (2003). A one square kilometre area was modeled.

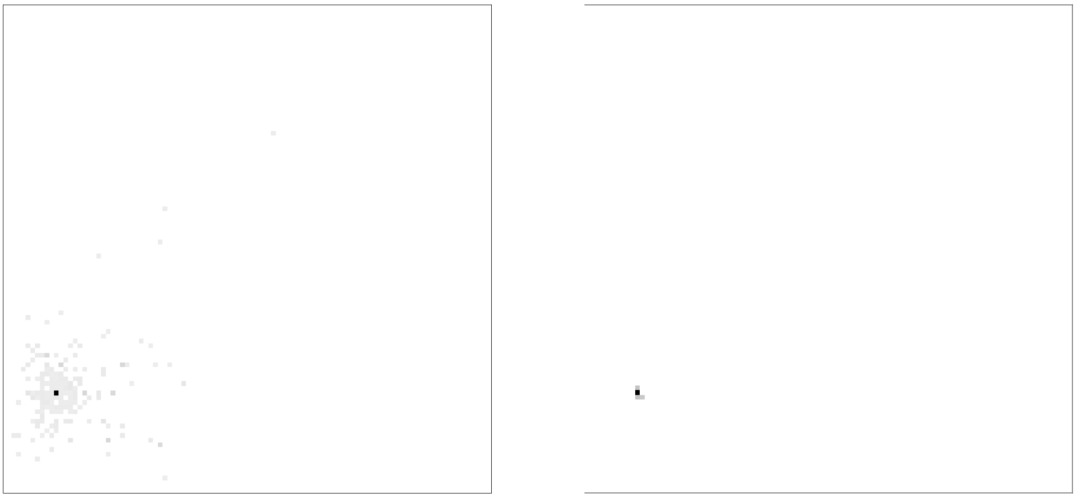
The median distance for pollen spread was set to 0.3 m. In the case where glyphosate was applied annually, the flow of resistance genes occurred rapidly. Within five years, around one third of the paddock contained some degree of resistance (Figure 3a). After 20 years, the distribution of resistance is largely unchanged (Figure 3b). When simazine was used to replace glyphosate, resistance spread similarly within five years (Figure 4a). However, repeated application of simazine controlled the resistance in all but the originating cell and its neighbours (Figure 4b).

This simple application of the spread modelling framework shows how a small change in farm management can have a significant effect on the long-term spread of resistance.



a. Glyphosate knockdown (5 years). **b. Glyphosate knockdown (20 years)**.

Figure 3. Resistance spread by pollen movement: no resistance is shown in white, grey-scale values show log-scaled percentage of the seeds with resistance



a. Simazine knockdown (5 years). **b. Simazine knockdown (20 years).**

Figure 4. Resistance spread by pollen movement: no resistance is shown in white, grey-scale values show log-scaled percentage of the seeds with resistance

Acknowledgements

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SEED GERMINATION, GROWTH PATTERNS AND PHENOLOGY OF A WILD MALAYSIAN BRASSICA [*Brassica juncea* (L.) Czern. *var.* Ensabi]

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Abstract: The wild species of Malaysian brassica (*Brassica juncea* L. Czern. *var.* Ensabi) with its distinct pungent aroma and bitter taste is now widely cultivated as a local vegetable by the Malays and natives of Sabah and Sarawak. The species is also used as a popular kimchi by the locals. Hence, seed germination, general growth patterns and phenology of *B. juncea* was evaluated under different light and temperature regimes. *Brassica juncea* did not display seed dormancy, but seed germination was positively photoblastic, and the rate of germination was temperature-dependent with measurably higher percentages at 25^{o}C *vis-a-vis* other temperatures. No significant differences were noted in germination percentages among seeds exposed to light (PAR = 625 µmole photon m^{-2} s^{-1}) and those in darkness. Seedlings subjected to darkness in the growth chamber had greater shoot and root lengths when compared with their fully exposed counterparts. Seeds exposed to full sunlight in the open (midday PAR = 622 µmole photon m^{-2} s^{-1}) displayed higher percentages of germination compared their counterparts growing in the shade under the insect-proof house (60% sunlight at midday PAR = 384 µmole photon m^{-2} s^{-1}). Distinct manifestation of a lag phase in seedling growth was recorded between Stage 1 and Stage 2, otherwise plant growth was rapid. Plants growing under shade registered 23% bigger stem diameter, greater mean leaf number/plant with bigger leaf size, plant height was *ca.* 106 cm, but lower number of flowers/plant and pods/plant *vis-à-vis* those subjected to full sunlight. The number of seeds/pod was similar irrespective of the light regimes that the plants were exposed to.

Key words: *Brassica juncea*, seed germination, growth

Introduction

Brassicaceae is a large family with over 3,200 species in about 375 genera. The species are herbaceous annuals, biennuals, and perennials. Many weed species in agriculture such as *Brassica nigra* (L.) Koch, *B. rapa* L., *Cardaria draba* (L.) Desv. *Raphanus raphanistrum* L., and *Sinapis arvensis* L., belong to this family. The crop species *viz. B. napus* L., *B. rapa* L., and *B juncea* (Czern.) Coss.), are among the most important crop species in this family. The wild species of Malaysian brassica (*Brassica juncea* L. Czern. var. Ensabi, with its distinct pungent aroma and bitter taste and a member of the sub tribe *Brassicinae* of the tribe *Brassiceae* is widely domesticated as a vegetable among the Malays and natives in the Malaysian states of Sabah and Sarawak. The species is also used as a popular kimchi by the locals.

Seed germination is a complex physiological process that is responsive to many environmental signals including temperature, water potential, light and other factors (Bewley and Black, 1994; Baskin and Baskin, 1998). Seed germination can be divided into three phases, imbibition, increased metabolic activity, and initiation of growth, which loosely parallel the triphasic water uptake of dry mature seeds. By definition, germination incorporates those events that start with the uptake of water by the quiescent dry seed and terminate with the protrusion of the radicle and the elongation of the embryonic axis. Morphologically, initiation of growth corresponds to radicle emergence; subsequent growth is generally defined as seedling growth. Temperature has a primary influence on germination, affecting the capacity for germination by regulating dormancy and the rate or speed of germination in non-dormant seeds. Temperatures for germination are generally related to the environmental range of adaptation of a given species and serve to match germination timing to favorable conditions for subsequent seedling growth and development (Alvardo and

Bradford, 2002). Light is considered as the most important environmental factor regulating growth and development of plants, and Smith (1982) reported the light requirement of seeds is also dependent on the temperature. Seeds of many plant species, mostly those that inhabit open or frequently disturbed habitats, require light to germinate. Environmentally induced photosensitivity of seeds is often interpreted as an adaptation ensuring that seeds will germinate in sites in which the probability of seedling establishment is high (Labouriau and Agudo 1987).This paper described the results of our study on the effects of light, temperature and different media on seed germination of *Brassica juncea.* Growth patterns and phenology of *B. juncea* as influenced by light and temperature regimes are also assessed.

Materials and Methods

Experiment 1. Effects of light and temperature regimes, and different media on seed germination in Brassica juncea var. Ensabi in the laboratory

Germination experiments were conducted in the laboratories of the Institute of Biological Science, University of Malaya. For seed germination the fresh seeds of *B. juncea* were dried in an oven at 35°C for a week and stored in a refrigerator at 5°C until use. Fifty seeds were placed in each petri-dish for each treatment, previously lined with 9 cm diameter Whatman No.2 filter paper. The filters were moistened with 6 ml of water or other solutions of the chemical media KNO_3, H_2O_2 and HNO_3. Seeds were treated in the following manner: untreated seeds (H_2O, control pH=7); seeds soaked H_2O + 0.2 M KNO_3 (pH=2.5); seeds soaked in H_2O + 5% H_2O_2 (pH=2.8); and seeds soaked in H_2O + 0.01 M HNO_3 (pH=2.5). The Petri dishes were placed in growth chambers with temperature regimes of 15, 20, 25, 30 and 35°C under a photoperiod of two light levels (12:12 hrs light: dark period, and 24-h dark environment). For treatments in the dark environment but with similar sets of chemical media and temperature regimes, another set of Petri dishes were incubated in a double layer aluminum foil. Treatments under similar temperature and light regimes and chemical media were used for oven-dried seeds. Germination were recorded and removed daily and the germination percentages were noted at 10 days after sowing. Standard procedures for testing seed viability and growth recommended by ISTA (1985) were employed for seeds failing to germinate. For the purpose of this study, germination was considered as being complete when the radicle emerged from the seed. Germinated seeds were counted and removed every day until germination stopped and length of radicles and plumule was measured. The rate of germination was estimated from the reciprocal of the time taken to reach 50% of the final cumulative germination. An arcsine transformation was performed to the seed germination percentage data before statistical analysis to ensure homogeneity of variance. The effects of light, temperature and seed treatments with chemical media on the germination and rate of germination were examined using ANOVA, with Tukey's tests made on difference between means where appropriate (Zar, 1999).

Experiment 2. Growth patterns of Brassica juncea var. Ensabi as influenced by light regimes

Assessments on the growth patterns and structural demography of *Brassica juncea* var. Ensabi in relation to different light regimes were made in a greenhouse experiment at Rimba Ilmu, University of Malaya, Kuala Lumpur, Malaysia. The experiment was done in greenhouse (with light regime at midday of PAR= 384 μmole photon m^{-2} s^{-1}) with clay pots measuring 35 cm in diameter, and 40 cm high, previously filled with garden soil of the Malacca series. Prior analysis of the soil was conducted to establish its physico-chemical status (Faravani and Baki, 2007). Six uniform seedlings previously thinned from those sown in clay pots were maintained. The plants were watered from above twice daily with a fine rose. A similar set of six pots of *B. juncea* plants were maintained with full exposure to sunlight in the open (with light regime at midday of PAR = 622 µmole photon m^{-2} s^{-1}) until harvest. Growth parameters, *viz*. plant height, stem diameter (at harvest) number of leaf modules/plant/every three days, branch number/plant/every three days, number of flowers/plant/every three days after budding, number of immature and mature pods/plant/every three days after starting of the pod stage, seed number/ pod/ plant/mature plant, seed weight/pod/plant/mature plant were determined.

Results and Discussion

Temperature has a major impact on seed germination (Benvenuti and Miele, 2001; Teketay 2002). There were significant temperature-mediated differences in the rate of seed germination of *B. juncea*, principally between (15-20ºC) and (25-30°C), reaching the optimum at 30°C (Table 1). Over 90% germination occurred at 25-30°C, thereafter germination rates decreased. Fresh and dry seeds of *B. juncea* subjected to all chemical media, KNO_3, H_2O_2 and HNO_3, irrespective of temperature regimes in darkness and in light failed to germinate which different from wrinkle grass, the latter is positively photoblastic (Baki and Nabi, 2003). The 2, 3, 5-triphenyl-tetrazolium chloride test confirmed that seed embryos of the non-germinated seeds were killed by chemical media. It is indicative that the temperature thresholds for seed germination of *B. juncea* were lower than 15°C and higher than 35°C, respectively.

Table 1. Seed germination and seedling growth of *Brassica juncea var*. Ensabi as influenced by temperature regimes in the laboratory

Temperature (°C)	3 Days	5 Days	7 Days	Germination (%)	Plumule length(mm)	Root length (mm)
15	17.9 C	61.0 B	79.7 A	79.8 d	14.95 d	9.98 d
20	29.8 C	69.7 B	84.2 A	84.2 c	17.5 c	11.27 c
25	39.2 C	78.3 B	95.5 A	95.5 a	22.15 a	15.21 a
30	37.7C	89.0B	94.6 A	94.6 a	21.71 ab	12.82 b
35	41.3 C	85.6 B	86.7 A	86.7 b	21.26 b	11.98 bc

Within a column, means followed by the same lowercase letters, and within a row the means followed by the same upper case letters are not significantly different at p=0.05 (HSD tests).

The radicle and hypocotyl lengths of *Brassica juncea* seeds were significantly (*p*=0.05) affected by temperature and light regimes when seeds were soaked with distilled water (Figures 1 and 2).

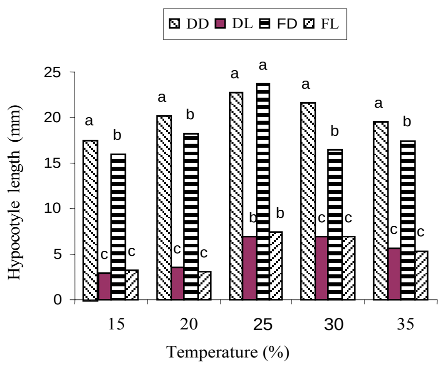

Figure 1. Effects of seed treatments and temperatures on hypocotyl lengths of *Brassica juncea var*. Ensabi under different light regimes soaked in distilled water. Bars with same letters are not significantly different at p=0.05 (Tukey's test).DD: dry seed in dark; DL: dry seed in light; FD: fresh seed in dark; FL: fresh seed in light

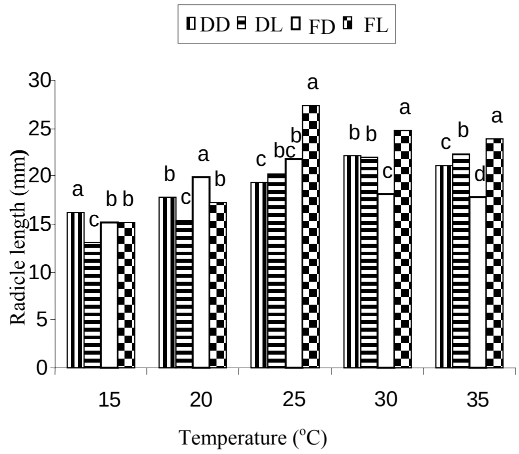

Figure 2. Effects of seed treatments and temperature regimes on radicle lengths of *Brassica juncea var.* Ensabi under different light regimes. Bars with the same letters are not significantly different at p=0.05.

The hypocotyl indicated the optimum length growth to be 22.8 (mm) at 25°C on dry seeds under total darkness and for fresh seeds in the same condition was 23.7 (mm) at 25°C. The optimum hypocotyls lengths under light were 6.93 (mm) and 7.39 (mm) at 25°C for dry and fresh seeds respectively. But the optimum linear growth of radicle measured 22.1 (mm) at 30°C and 21.7 (mm) at 25°C under total darkness for dry and fresh seeds, respectively. The optimum root seedling length in light measured 22.3 (mm) and 27.3 (mm) at 35° and 25°C for dry and fresh seeds respectively. Characteristics of the light that affect germination include duration, quality and photon irradiance of the light reaching the seed (Casal and Sanchez, 1998). The growth of the hypocotyl was affected in light and total darkness, although the radicle remained unaffected in both the conditions. It was concluded that the hypocotyl grew around five times more in total darkness as compared to the growth in light condition, whereas the growth of the radicle in light condition and darkness did not indicate same difference. The growth of the hypocotyl in total darkness also depended on a suitable temperature. Temperature has influenced to regulate the growth of hypocotyl in total darkness.

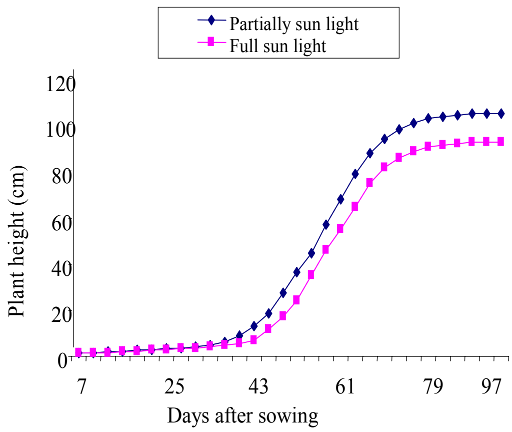

Figure 3. Effects of different light regimes on plant height of *Brassica juncea var.* Ensabi

Result of study on phenology and general growth pattern of *B. juncea* showed that germination in *B. juncea* was rapid, but between Stages 1 and 2, plant growth was very slow.

Two light regimes: (1) full sunlight in an open (at midday, mean photosynthetically active radiation, PAR = 622 µmole photon m^{-2} $s^{-1)}$ and (2) 60% of full sunlight within the greenhouse (at midday, PAR = 384 µmole photon m^{-2} s^{-1}) (the light readings were taken using a LICOR Radiometer) affected growth. Partial sun light enhanced plant height (Figure 3) at later stages. The total growth parameters of the plant at different light regimes are also shown in Table 2. Full light significantly affected flowers/plant flowers, pods/plant and pods on primary branches Full light also affected seed weight and stem diameter. There were no significantly differences in leaves/plant, leaves on primary branches, seeds/pod and number of nodes/plant.

Table 2. Growth characters of matured *Brassica juncea* (L.) Czern. var. Ensabi at harvest in different light regimes.

Growth parameter	Mean values in full light ± SD	Mean values in partial light ± SD
Plant height (cm)	97.6 ± 2.9	104.7 ± 2.5
No. of leaves/plant	89.7 ± 3.5	186.6 ± 3.6
No. of nodes/plant	10.6 ± 0.9	10.0 ± 1.2
No. of leaves on primary branch No. 1	8.3 ± 0.9	7.5 ± 0.9
No. of leaves on primary branch No. 2	17.0 ± 1.2	16.3 ± 2.2
No. of leaves on primary branch No. 3	22.2 ± 1.9	22.0 ± 1.8
No. of pods/plant	287.1 ± 4.2	234.0 ± 2.5
No. of pods in primary branch No. 1	34.0 ± 1.0	29.0 ± 2.0
No. of pods in primary branch No. 2	45.0 ± 1.7	32.0 ± 1.6
No. of pods in primary branch No. 3	48.0 ± 2.6	36.0 ± 2.8
No. of seeds/pod	11.6 ± 1.5	11.1 ± 1.5
No. of seeds/plant	3286.0 ± 52.3	2531.0 ± 41.9
No. of primary branches/plant	10.6 ± 1.2	10.1 ± 0.9
No. of secondary branches/plant	16.0± 1.7	14.0 ± 1.7
Plant stem diameter (mm)	4.2 ± 0.2	3.3 ± 0.2

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HOST INDUCED RESISTANCE AS A CONTROL METHOD FOR PARASITIC WEEDS

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Abstract: Parasitic weeds restrict crop production in many countries. Plant induced resistance is against not only pathogens and herbivores, but also parasitic weeds. Induced resistance against parasitic weeds has been demonstrated in the associations of sunflower-*Orobanche cumana*, tobacco/hemp-*O. ramosa* and pea-*O. crenata* under greenhouse conditions and/or in the field. Plant activators include biotic and abiotic agents. Host plants underlay systemic acquired resistance (SAR) or induced systemic resistance (ISR) pathway against parasitic weeds. Induced resistance should be integrated in the parasitic weed management systems. This paper reviews the history and development, pathway and mechanism, as well as prospect and outlook of plant induced resistance on the control of parasitic weeds.

Key words: Induced resistance, parasitic weed control, systemic acquired resistance (SAR), induced systemic resistance (ISR)

Introduction

Parasitic weed is a plant that has to associate with its host-plant and absorb water, minerals and assimilates through a haustorium from the host. It cannot complete its life cycle alone and causes damage to the host. Parasitic weeds include holoparasites (*e.g. Cuscuta* and *Orobanche*) and hemiparasites (*e.g. Striga* and *Loranthus*) or root parasites (*e.g. Orobanche* and *Striga*) and shoot parasites (*e.g. Cuscuta* and *Loranthus*). It is very difficult to control parasitic weeds once they have attacked their host because of the close association between host and –parasite. Hence parasitic weeds are becoming an increasing threat to agriculture and forestry. The genera of *Cuscuta*, *Orobanche* and *Striga* are listed as quarantine weeds by many countries (Parker and Riches, 1993).

Plant induced resistance is activation of its natural immune system to defend against invasions and/or stresses through inducing agents. Inducing agents or plant activators include biotic or abiotic factors, such as microorganisms and natural or synthetic compounds. Plant induced resistance is generally systemic, known as systemic acquired resistance (SAR) or induced systemic resistance (ISR), but sometime local, known as localized acquired resistance (LAR). It controls not only pathogens and herbivores, but also parasitic weeds (Durrant and Dong, 2004; van Loon *et al*. 1998; Sauerborn *et al*. 2002). Induced resistance by acibenzolar-S-methyl (ASM) against *O. cumana* in sunflower (*Helianthus annuus*) was first reported by Sauerborn *et al.* (2002). In this paper, the history and development, pathway and mechanism, as well as prospect and outlook of plant induced resistance on control of parasitic weeds were summarized.

History and development of plant induced resistance against parasitic weeds

Plant induced resistance has a century history against pathogens (Beauverie, 1901; Ray, 1901) and more than three decades against herbivores (Green and Ryan, 1972; Haukioja and Hakala, 1975), but only very recently against parasitic weeds. Bhargava (1991) and Hassan *et al*. (1991) reported that exogenous application of ascorbic acid and chlormequat inhanced tomato (*Lycopersicon esculentum*) resistance to *O. cernua* and *O. ramosa*, respectively. Joel (2000) reported that soil application of uniconazole reduced infection of *O. cumana* in sunflower.

However, he only considered that uniconazole inhibited seed germination of the parasite and neglected that uniconazole could induce host resistance (Zhou and Leul, 1999).

The hypothesis of induced resistance as a strategy for parasitic weeds control was first formally proposed by Jorrin and co-workers in 2001. The milestone of induced resistance against parasitic weed was first established in the sunflower-*O. cumana* association by Sauerborn and co-workers in 2002. Sunflower defended oneself against *O. cumana* after the seed or seedling were treated with ASM (Sauerborn *et al*. 2002). Hereafter, Fan *et al.* (2005, 2007a,b) reported on the efficiency of different application type, time and frequency of ASM and its combination with a biocontrol fungus — *Fusarium oxysporum* f. sp. *orthoceras* against *O. cumana* in sunflower. They also found out a new chemical -- prohexadione-Ca for inducing sunflower resistance to the parasite. Buschmann *et al.* (2005) and Müller-Stöever *et al.* (2005) have also reported on this field, respectively. Pérez-de-Luque *et al.* (2004) reported pea (*Pisum sativum*) resistance to *O. crenata* induced by ASM. Mabrouk *et al.* (2007) demonstrated that pea roots inoculated with *Rhizobium leguminosarum* strains led to a reduced root infection by *O. crenata*, resulting from an enhanced peroxidase activity and constantly high phenylalanine ammonia lyase activity in pea roots. Gonsior *et al.* (2004) proved that tobacco (*Nicotiana tabacum*) and hemp (*Cannabis sativa*) treated with ASM, extracts of algae (*Ascophyllum nodosum*) and *Pseudomonas* spp. acquired resistance to *O. ramosa*. Thus, the prelude of induced resistance against parasitic weeds has been undrawn and the research is in the ascendant.

Pathway and mechanism of induced resistance against parasitic weeds

The mechanism of sunflower against *O. cumana* induced by ASM underlies SAR pathway, which depend on salicylic acid (SA), accumulate phenolic compounds and phytoalexins, as well as synthesize pathogenesis-related proteins. Lignification is accompanied in sunflower resistance cultivar infested by *O. cumana*, but not in susceptible cultivar induced by ASM (Labrousse *et al*. 2001; Sauerborn *et al*. 2002; Buschmann and Sauerborn, 2002; Fan, 2005). This indicates that sunflower susceptible cultivar directly produces defense-related compounds against the parasite rather than constitutively modifies its tissues when resistance is induced by ASM.

The ISR pathway might be involved in tobacco and hemp against *O. ramosa* induced by rhizobacteria (Gonsior *et al*. 2004). This needs further investigation. Plant growth retardants, such as chlormequat, uniconazole and prohexadione, strengthen host plant resistance to parasites through inhibition of gibberellin biosynthesis, alteration of host metabolism and accumulation of defenses (Rademacher, 2000). This pathway, similar to SAR (Bubán *et al*. 2004; Rademacher, 2004) or another, is still in discussion.

Prospect and outlook of induced resistance against parasitic weeds

Parasitic weeds impair crop production in many countries. Economically important genera of parasitic weeds in agriculture and forestry are *Cuscuta*, *Orobanche*, *Striga*, *Cassytha*, *Dendrophthoe* and *Taxillus*, etc. The genus of *Cuscuta*, nearly distributing worldwide, mainly damage soybean (*Glycine max*), groundnut (*Arachis hypogaea*), tobacco, potato (*Solanum tuberosum*), alfalfa (*Medicago sativa*), clover (*Trifolium* ssp.), flax (*Linum usitatissimum*), citrus (*Citrus* ssp.), peach (*Prunus persica*), litchi (*Litchi chinensis*), mulberry (*Morus alba*), tea (*Camellia sinensis*), coffee (*Coffea spp*.) and numerous forest trees. The genus *Orobanche*, occurs in the temperate as well as in the subtropical ecozones of the globe, and mainly attacks melon (*Cucumis* spp. or *Citrulus* spp.), tobacco, sunflower, tomato and hemp. The genus *Striga*, is found in tropical regions, and parasitises mainly cereal crops, such as sorghum (*Sorghum bicolor*), maize (*Zea mays*) and rice (*Oryza sativa*), and few leguminous crops, such as cowpea (*Vigna unguiculata*). The genera of *Dendrophthoe* and *Taxillus* are harmful to fruit and forest trees, such as citrus, litchi, and rubber (*Hevea brasiliensis*). The technique of parasitic weed control employs mainly traditional plant protection procedures, including quarantine, physical, cultural, biological, chemical and host resistance breeding (Parker and Riches, 1993). It seems accepted that no single method can combat satisfactorily the parasites, hence integrated management is promising (Dhanapal *et al*. 1996).

Preventive crop protection which includes plant induced resistance, is playing a more and more important role in cropping systems. It controls many crop diseases and pest insects in the greenhouse and the field (Vallad and Goodman, 2004). So far, plant induced resistance has been demonstrated against the root parasites of the genus *Orobanche*. First experiments to induce resistance in cereals to control *Striga* were not successful. Nevertheless there are a lot of investigations in this field of research, such as screening for new inducing agents. Plant induced resistance should soon be integrated in parasitic weed management in crop production systems.

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SEED GERMINATION, GROWTH PATTERN AND PHENOLOGY OF RHODODENDRON (*Melastoma malabathricum* L.) IN MALAYSIA

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Abstract: Seed germination and general growth patterns of *M. malabathricum* under different light, media and temperature regimes were studied in the laboratory and insect-proof house conditions at the University of Malaya. Germination was very sensitive to chemical media. The highest rate of germination were observed at 25-30°C in seeds dried at 35°C and immersed in distilled H_2O. No seed germination observed in darkness, or when exposed to chemical media, and at temperature regimes less than 20°C. Uniformly grown, two-month seedlings were exposed to two light regimes, *viz.* full sunlight (FSP) in an open (at midday, mean photosynthetically active radiation, PAR = 622 µmole photon $m^{-2} s^{-1}$) or 20% of full sunlight within the greenhouse (Partial exposed plants)(PEP) (at midday, PAR = 125 µ mole photon $m^{-2} s^{-1}$). The light regime strongly influenced both vegetative and reproductive growth of *M. malabathricum* The FSP displayed higher rate of growth in terms of number of berries, seeds, 2^{ry} branches leaves /2^{ry} branch, leaves /1^{ry} branch, berries/2^{ry} branch and total number of leaves/plant .Exposure to full sunlight led to higher reproductive and vegetative growth with higher number of leaves/plant and number of 1^{ry} branches/plant, number of flowers/plant, number of seeds/plant for the FSP plants, but the PEP had more secondary and tertiary branches and internodal length. The regression analysis generated model for plant height data in relation to days after planting (x) were y = 56.2Ln(x) - 209.3, r = 0.96 for the PEP plants, and y = 15.99Ln(x) - 2.65, r = 0.92 for the FEP plants.

Key words: *Melastoma malabathricum*, growth, phenology.

Introduction

Melastoma malabathricum is a serious weed in many crops, on derelict and abandoned farmlands, and arable lands in Malaysia (Ridley 1922; Maxwell 1989, Baki 2004; 2006; Faravani and Baki 2007), and else where in the tropics and subtropics (Renner & Meyer 2001; Clausing and Renner 2001). The weed has a propensity to become invasive with adaptive life strategies including robust vegetative and reproductive growth coupled with efficient seed dispersal, often aided by ants and birds, and are attracted by copious production of fruits. This opportunity rarely arises in the native habitat of a species as there tends to be a higher rate of competition from other plants. They are primary colonizers of disturbed habitats, pastures, roadsides, landslides, light gaps and rivers. This species is fast growing, shade tolerant, devoid of natural pests, and sets an abundance seeds with a high rate of germination leading to monospecific stands – easily out-competing native flora, putting them at great risk (Penneys, 2004).

Plants are influenced by sunlight reduction in a different ways. The main limitation of leaf net photosynthetic carbon assimilation at high photon flux density is the concentration of CO_2. When photon flux density decreases to approximately 40% of that a full sunlight, carbon assimilation become light-limited (Cohen *et al.* 2005). However, plants have considerable ability to acclimatize to different light regimes through changes in leaf properties, as well as canopy structure (Syvertsen and Smith, 1984). Plant responses to light include a variety of adaptations at physiological and biochemical levels such as alteration of growth rate and plant architecture and finally on morphological characteristics and distribution. The architecture of plant canopy influences the interception, absorption and scattering of solar radiation (Christopher *et al.* 1998; Nasrullahzadeh *et al.* 2007). These photo-morphological responses are mediated by the phytochrome system under dense canopy. Generally branching is more inhibited by vegetation shade than by natural shade (Wan and Ronald.1998). Shade imposes a limitation to biological productivity in plant, although the extent of the limitation varies with shade tolerance level of the species and the nitrogen supply (Wong, 1991). Knowledge on the effects of light for some pioneer photoblastic species like *M. malabathricum* would have a practical value in designing the degree of canopy opening to stimulate the establishment of these seedlings. Interestingly, variation in specific leaf mass, the trait considered to be important in adaptive shade-avoidance responses, was only partially attributable to ontogenetic plasticity (environmentally-induced adjustments of ontogeny).

This paper describes some results of a study on the response of *M. malabathricum* to environmental factors, and their interactions on seed germination, and vegetative and reproductive growth, and phenology of the weed.

Materials and Methods

Experiment 1: Effects of chemical media, light and temperature regimes on seed germination and seedling growth of Melastoma malabathricum.

Mature seeds of *M. malabathricum* were stored with silica gel in darkness at 4 ºC before use. A series of germination experiments and growth studies on the weed was conducted in the laboratories of Institute of Biological Sciences, University of Malaya. Fifty fresh seeds were placed in each Petri-dish, previously lined with 9 cm diameter Whatman No.2 filter paper were treated with distilled water (pH = 7) for 24 h as control; seeds soaked in distilled H_2O in 24 h + 0.2 M KNO_3 (pH=2.5); seeds soaked in distilled H_2O in 24 h + 5% H_2O_2 (pH=2.8); seeds soaked in distilled H_2O in 24 h + 0.01 M HNO_3 (pH=2.5); and seeds soaked in distilled H_2O in 24 h + 0.01 M HNO_3 (pH=2.5). Four replications were allocated for each treatment. A similar set of treatments were replicated but with seeds of *M. malabathricum* previously oven-dried at 35^{o}C for a week and stored in a refrigerator at 4^{o}C until use. The Petri-dishes were placed in growth chamber with different temperature regimes of 15, 25, 30 and 35^{o}C, and exposed to fluorescent light with intensity of 630 Em^{-2} s^{-1}. Another set of Petri-dishes, wrapped in a double layer of aluminum foil and with the same treatment combination media and temperature regimes, were also maintained to assess seed germination in darkness. Seed germination was recorded at three-day intervals and the germination percentages were determined 18 days after sowing. All petri-dishes in the light treatment were augmented with 6 ml of deionized water at 3 day- intervals in order to maintain moisture. Seedlings and dead seeds were counted and removed. Germinated seeds were counted and removed every 3 days until no germination were recorded, root and shoot lengths of seedlings were recorded. The rate of germination was estimated from the reciprocal of the time taken to reach 50% of the final cumulative germination. Differences in germination were subjected to analyses of variance. Data transformations were done prior to achieve homogeneity of variances.

Experiment 2. Effects of light regimes on clonal and reproductive growths of two biotypes of Melastoma malabathriucm

Six young uniform seedlings of the pink- and white-flower biotypes) of *M. malabathricum* were selected from a commercial nursery in Genting Highlands, Pahang, Malaysia (3^o 8’ N; 101^o 42’ E) in December 2005. These were transplanted into clay pots (35 cm in diameter, and 40 cm high), previously filled with garden soils of Malacca series. The physico-chemical charateristics of the soil have been presented elsewhere (Faravani and Baki 2007). The plants were watered in the morning and in the evening from aboveusing a watering can and rose. The plants were divided in two groups - outdoor (FEP = full exposed to sunlight) (mean midday radiation of 622 μ mole photon m^{-2} s^{-1}) and inside in insect-proof house (PEP = partially exposed to sunlight) with 12 hr of natural sunlight (mean midday radiation of 125 μ mole photon $m^{-2} s^{-1}$) and mean ambient temperatures of 33 ^{o}C (day) and 25 ^{o}C (night) at Rimba Ilmu, University of Malaya, Kuala Lumpur. Growth parameters, i.e. plant height; number and lengths of primary, secondary, and tertiary branches, leaf numbers in each category of branches; and phenological traits (time and duration of flowering, number of flowers/branch or number of flowers/plant) were recorded. The growth data were analyzed with ANOVA and regression analyses where appropriate. The process of finding the best fit was done by CurveExpert 1.3 by comparing the data to each model. The XY data was modeled using a toolbox of linear regression models, nonlinear regression models, interpolation, or-splines.

Results and Discussion

Germination and Seedling Growth

Germination of *M. malabathricum* seeds was significantly (p =0.05) affected by temperature but was not manifested in fresh and dry seed treatments (Table1). No seed germination prevailed in darkness, or when exposed to chemical media, and at temperature regimes less than 20^{o}C. The seed viability tests with 2,3,5-triphenyltetra-zolium chloride test showed that seed embryos were killed by chemical media. Seed germination of *M. malabathricum* was positively photoblastic. The highest rate of germination of 37 -40 % was observed at 25-30^{o}C for oven dried seeds in distilled H_2O.

Light regime strongly influenced both clonal and reproductive growth of *M. malabathricum.* Germination of the light-mediated *M. malabathricum* seeds is expected to take place near the soil surface. This is beneficial for the survival of the species as germinated seeding prevailing too deeply in the soil where it may not be able to reach the soil surface (Egley 1995).Germination capacity declined at 35ºC and thereafter, revealing that 28ºC as the optimum germination temperature for this seed lot. The length of radicle was affected by different temperatures (Table 1) and root and radicle lengths were increasing in high temperatures, but longer shoot lengths were observed at low temperatures. The root/shoot ratio was significantly affected by different temperatures and it was increasing with high temperatures (Table 1).

Table 1. Comparison of means of seed germination of *Melastoma malabathricum* under different temperatures and seed treatments.

Group		% Germination	Shoot (mm)	Root (mm)	R:S
Temperature ^{0}C	35	26.3 b	2.36 b	8.45 a	3.718 a
	30	37.3 a	2.66 ab	5.31 b	1.813 b
	25	39.8 a	2.89 a	3.23 c	1.119 c
	15	0.00 c	0.00 c	0.00 d	0.00 d
Seed treatment	Fresh seed	1.82 bc	3.83 b	1.63	1.82 bc
	Fresh + 24 h distilled water	1.74 c	3.29 b	1.49	1.74 c
	Oven-dried seed	2.28 a	5.96 a	2.06	2.28 a
	Oven-dried seed + 24h distilled water	2.06 ab	3.91 b	1.464	2.06 ab

Within a row, values followed by the same letter are not significantly different at p= 0.05 by the LSD test.

General growth and branching patterns

Table 2 displays the regression models representing the best fitted models for different plant characters as a function of days after planting in PEP and FEP plants. These models describe the growth for different parameters such as plant height, branch number, leaf number/plant or leaf number/branch, and flowers/branch or flowers/plant. The regression analysis generated respective models for plant height data in relation to days after planting (x) were y = 56.2Ln(x) - 209.3, r = 0.96 for the PEP plants, and y= 15.99Ln(x) - 2.65, r = 0.92 for the FEP counterparts (Figure 1).

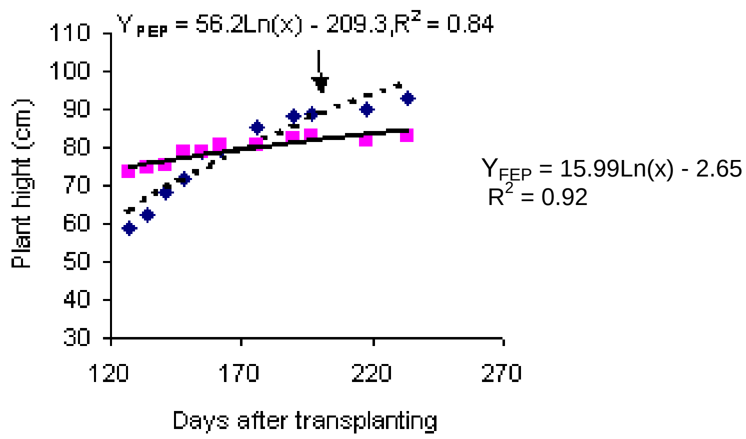

Figure 1. Plant height (cm) of *Melastoma malabatricum* as influenced by light regimes.

It can be argued that some death of old branches remained within the PEP and FEP canopies, especially at the bottom and inside due to the reduction in substrate production capacity and light availability. Holeman *et al.* (1990) reported that the average number of flushes on branch shoots decreased with crown depth in *Pinus taeda* trees. Rook *et al.* (1987) showed the same results for *Pinus radiata* and reported that the number of flushes on a branch is controlled by hormones, and the environmental conditions or substrate availability is largely unknown. The equations for other discrete variables like as the number of leaves, branches are presented in Table 2.

Table 2. Model summaries and parameter estimates of the regression relationships between selected growth parameters of *Melastoma malabathricum* as the function of time after transplanting exposed to full and partial sunlight

Dependent variables	Partial sunlight		Full sunlight	
	Regression model	r	Regression model	r
1. Number of 2° branch/plant	y = -13.2x/(-79.7+ x)	0.99	y =2.2+0.3*cos(0.1x-3.3)	0.90
2. Number of leaves /2° branch/ plant	y = 0.2357x - 20.372	0.99	y = 0.51x - 55.3	0.92
3. Number of leaves /1° branch/ plant	y = 0.2424x - 5.8708	0.99	y = 0.3881x - 20.243	0.90
4. The length of 1° branch/plant	$y = -50.5+1.08x-0.002x^2$	0.99	$y =-31.9+1.03x-0.003x^2$	0.99
5. Plant height (cm)	y = 56.2Ln(x) - 209.3	0.96	y = 15.99Ln(x) - 2.6491	0.92
6. Number of leaves/plant	y =12.4+0.5*cos (0.2x-5.02)	0.80	$y =(-12995.9*80.569+197.6 * x^{2.2})/(80.6+x^{2.2})$	0.79
7. Number of 3° branch/plant	$y = 0.42-0.007x-0.0003x^2$	0.89	No analysis done, 3° absent	-
8. Number of leaves /3° branch/ plant	$y = (-700072+6005x)/(1+13368.775x-53.2x^2)$	0.98	No analysis done, 3° absent	-
9. Number of 1°branch/plant	constant equal 4.3	-	y =1/(-0.001x+0.2761)	0.96

The FEP displayed higher rate of growth in terms of number of berries, seeds, 2^{ry} branch, leaves /2^{ry} branch, leaves /1^{ry} branch, berries/ 2^{ry} branch and total leaves in each plant type (Figure 2).Two biotypes (pink flower and white flower) showed different rates of biomass accumulation and plant height .Sunlight caused a higher rate of seed production ,number of leaves and 1^{ry} branches and flower but the PEP had more second & third branches and internodal length.

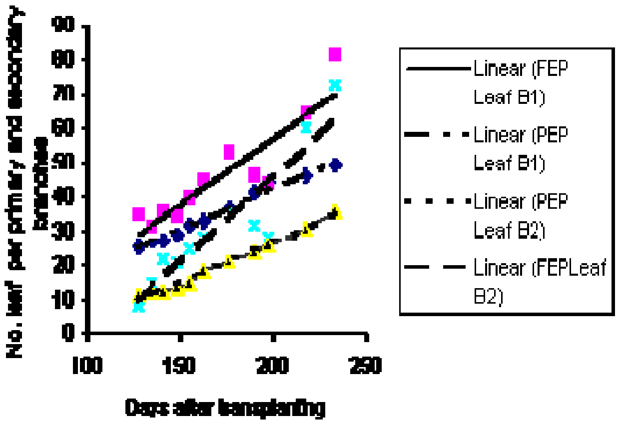

Figure 2. Leaf number of primary and secondary branch per plant of *Melastoma malabathricum* as influenced by light regimes. FEP Leaf B1, number of leaves on branch #1; PEP Leaf B1, number of leaves on branch #1; PEP Leaf B2, number of leaves on branch #2; FEP Leaf B2, number of leaves on branch #2. The regression equations # 2 and 3 representing the relationships are shown in Table 2.

The regression analysis generated these models for plant height and number of leaves 1^{ry} and 2^{ry} branch per plant in relation to days after planting are shown in Figures 1 and 2. The lower and upper temperature thresholds for germination of *M. malabathricum* were not encountered in this study, but no germination prevailed at temperatures lower than 20^{o}C, and germination declined at temperatures higher than 3^{o}C. However, effect of low and high temperatures among seeds in a population or in germination sensitivity were observed by others (Orozco-Segovia *et al.* 1996; Kebreab and Murdoch, 2000; Grundy *et al.* 2000), and changes in both the upper and lower temperature limits for germination are often associated with the imposition and release of dormancy (Kruk and Benech, 2000). Strong dormancy was not observed in *M. malabathricum* in this study. The dormancy level of seeds can change on a presumably continuous scale, for example in dark conditions. Seeds of many species can react to the environment and adjust the level of dormancy by deepening or weakening it (Vleeshouwers *et al.* 1995; Baskin and Baskin, 1998).

Knowledge on seed dormancy and germination helps to explain the occurrence of weeds, and can be important when developing, or foreseeing the effect of various weed-control methods. We observed early death of some branches within the plant canopy which could be due to light and internal growth competition between different plant parts and numbers of leaves. In the present study, the phenological variations of *Melastoma malabathricum* and the environmental conditions that the plants have been subjected to have largely been empirical and for a predictive understanding growth strategies and carbon allocation in relation to light regimes, more information on the mechanisms of phenology is needed.

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CHEMICAL WEED CONTROL IN NON-CROPPED AREAS

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Abstract: A field experiment was conducted during *rabi* 2005-06 and 2006–07 to find out the effect of herbicides on weed control in non-cropped areas. The experiment was carried out in non-cropped areas of the Viswavidyalaya Farm, Bidhan Chandra Krishi Viswavidyalaya, Kalyani, West Bengal, India in Randomized Complete Block Design with seven treatments and three replications. The treatments comprised two doses of the standard market available glyphosate 41 % SL *viz*. Glycel (5 and 10 ml l^{-1} water), four doses of the testing herbicide XL-COMBI-SG (3, 5, 7 and 10 g l^{-1} water) and a weedy check. The herbicides were applied on third week of October in both the years with a knapsack sprayer fitted with flat-fan nozzle with a spray volume of 500 l ha^{-1}. The major weed flora were *Axonopus compressus, Oplismenus compositus, Imperata cylindrica, Eleusine indica, Cynodon dactylon* and *Paspalidium flavidum* among grasses, *Cyperus rotundus, C. esculentus* and *C. aromaticus* among sedges. *Acalypha indica, Scoparia dulsis, Heliotropium indicum, Croton sparsiflorus, Bidens pilosa, Vitis trifolia, Tridax procumbans, Solanum khasianum, Mikania micrantha, Sida carpinifolia, Coccinea grandis, Ocimum sanctum, Ageratum conyzoides* and *Leucas linifolia* among broadleaved weeds. The maximum control of grasses, sedges and broadleaved weeds was obtained from XL-COMBI-SG 10 g l^{-1} water followed by XL-COMBI-SG 7 g l^{-1} water and glyphosate 41 % SL 10 ml l^{-1}. The gradual decrease in Weed Control Efficiency after 30 DAA was mainly due to the resurgence of perennial weeds having stolon or sucker (grasses), nut (sedges) and corm, tuber and rhizome (broadleaved). Therefore, from this experiment it can be concluded that XL-COMBI-SG 10 g l^{-1} water and XL-COMBI-SG 7 g l^{-1} water as well as glyphosate 41 % SL 10 ml l^{-1} water can be recommended for controlling the weeds in non-cropped areas or in fallow lands in between the two main crops.

Key words: glyphosate, herbicide, non-crop areas, weed

Introduction

Weeds, in the non-cropped areas, also affect our environment *e.g.* they reduce the aesthetic value of land, exhaust soil moisture through transpiration, deteriorate soil fertility, cause harm to livelihoods. Adverse effect of parthenium is a clear example of the above. Therefore, these weeds should be controlled either by mechanical or chemical means. Chemical methods are superior to mechanical methods because of their effectiveness in terms of time, cost and labour. Glyphosate and paraquat are commonly used to control the weeds in non-cropped areas. The XL-COMBI-SG is a readymix of glyphosate ammonium salt 35% and 2, 4-D ammonium salt 35%. Hence, an attempt was made to evaluate the effect of herbicides *viz.* market available glyphosate and the new product, XL-COMBI-SG on weeds in non-cropped areas.

Materials and Methods

The field experiment was conducted during *rabi* 2005-06 and 2006-07 to evaluate the efficacy of herbicides including market available glyphosate and the new herbicide, XL-COMBI-SG (glyphosate ammonium salt 35% + 2, 4-D ammonium salt 35%) on weed control in non-cropped areas. The experiment was carried out at the Viswavidyalaya Farm, Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal, India on sandy loam soil of this Inceptisol having a pH of 6.8 with good drainage facility. The experiment was laid out in a randomized block design (RBD) with seven treatments and three replications. The treatments comprised of market available glyphosate at 5 and 10 ml l^{-1} water as standard, five doses of XL-COMBI-SG (3, 5, 7 and 10 g l^{-1} water) and a weedy check. The herbicides were applied during the 3rd week of October with a knapsack sprayer fitted with a nozzle WFN 0.40. The observations on weeds were taken at initial, 7, 15, 21, 30 and 60 days after application (DAA) by using a quadrat of 0.5 m *x* 0.5 m.

Results and Discussion

The major weed flora were *Axonopus compressus*, *Oplismenus compositus*, *Imperata cylindrica*, *Eleusine indica*, *Cynodon dactylon* and *Paspalidium flavidum* among grasses, *Cyperus rotundus*, *C. esculentus* and *Cyperus aromaticus* among sedges. *Acalypha indica*, *Scoparia dulsis*, *Heliotropium indicum*, *Croton sparsiflorus*, *Bidens pilosa*, *Vitis trifolia*, *Tridax procumbans*, *Solanum khasianum*, *Mikania micrantha*, *Sida carpinifolia*, *Coccinea grandis*, *Ocimum sanctum*, *Ageratum conyzoides* and *Leucas linifolia* among broadleaved weeds. The dry weight of weeds decreased with an increasing dose of the standard market available glyphosate from 5 to 10 ml l^{-1} water and XL-COMBI-SG (glyphosate ammonium salt 35% + 2, 4-D ammonium salt 35%) from 3 to 10 g l^{-1} water. Among the herbicides, the minimum and maximum dry weight of weeds was recorded under XL-COMBI-SG at 10 g l^{-1} water and market available glyphosate at 5 ml l^{-1} water (Tables 1, 2 and 3). The XL-COMBI-SG at 10 g l^{-1} was closely followed by XL-COMBI-SG at 7 gl^{-1} water and market available glyphosate at 10 ml l^{-1} water. These herbicide treatments allowed significantly lower dry weight of weeds of all categories than other the other three herbicide treatments, *viz.* market available glyphosate at 5 ml l^{-1} water, XL-COMBI-SG at 3 and 5 g l^{-1} water.

Table 1. Effects of different herbicide treatments on dry weight (g m^{-2}) of grasses (av. of two years).

Treatments	Dose (l^{-1} water)	Initial	7 DAA	15 DAA	21 DAA	30 DAA	60 DAA
Glyphosate (market available)	5 ml	26.8	20.8	17.64	11.4	10.20	20.20
Glyphosate (market available)	10 ml	23.9	16.4	10.34	5.3	3.06	8.48
XL – COMBI - SG	3 g	29.9	18.7	14.88	11.2	9.80	20.68
XL – COMBI - SG	5 g	22.3	14.8	10.96	9.5	8.88	13.40
XL – COMBI - SG	7 g	32.4	17.2	8.07	4.7	3.24	9.08
XL – COMBI - SG	10 g	31.9	13.8	6.72	3.6	2.80	6.56
Weedy check	-	28.9	20.1	23.60	34.05	45.88	81.52
S.Em (±)		1.373	0.724	0.723	0.76	0.791	1.354
CD (p=0.05)		NS	2.231	2.228	2.341	2.437	4.172

Table 2. Effects of different herbicide treatments on dry weight (g m^{-2}) of sedges (av. of two years)

Treatments	Dose (l^{-1} water)	Initial	7 DAA	15 DAA	21 DAA	30 DAA	60 DAA
Glyphosate (market available)	5 ml	27.4	24.3	21.68	9.04	3.40	9.00
Glyphosate (market available)	10 ml	21.2	12.4	7.76	3.72	1.44	2.15
XL – COMBI - SG	3 g	18.4	15.3	14.32	8.52	5.76	8.70
XL – COMBI - SG	5 g	22.9	15.4	10.56	7.40	3.08	5.95
XL – COMBI - SG	7 g	29.4	11.3	7.44	2.94	1.60	2.10
XL – COMBI - SG	10 g	26.9	8.5	5.87	1.44	0.60	1.75
Weedy check	-	19.8	23.5	34.80	39.28	45.12	63.10
S.Em (±)		1.481	0.890	0.666	0.748	0.44	0.645
CD (p=0.05)		NS	2.743	2.053	2.305	1.355	1.986

Table 3. Effects of different herbicide treatments on dry weight (g m^{-2}) of broad leaved weeds (average of two years).

Treatments	Dose (l^{-1} water)	Initial	7 DAA	15 DAA	21 DAA	30 DAA	60 DAA
Glyphosate (market available)	5 ml	27.8	21.4	12.07	9.51	4.72	7.40
Glyphosate (market available)	10 ml	25.4	9.3	3.09	2.20	0.88	1.36
XL – COMBI - SG	3 g	29.0	16.4	12.60	10.84	5.69	8.92
XL – COMBI - SG	5 g	22.9	9.3	8.32	4.88	2.32	5.75
XL – COMBI - SG	7 g	29.5	6.8	4.52	2.12	1.04	1.46
XL – COMBI - SG	10 g	28.6	4.1	1.36	0.72	0.40	1.02
Weedy check	-	28.4	39.6	49.84	63.08	77.28	98.50
S.Em (±)		0.841	0.481	0.619	0.451	0.490	0.965
CD (p=0.05)		NS	1.483	1.907	1.390	1.508	2.973

The maximum control of grasses, sedges and broadleaved weeds was obtained under XL-COMBI-SG at 10 gl^{-1} water followed by XL-COMBI-SG at 7 gl^{-1} water as well as market available glyphosate at 10 ml l^{-1} water (Tables 4, 5 and 6). The gradual decline in weed controlling ability of the herbicides after 30 DAA was mainly due to the resurgence of perennial weeds having stolons, suckers, nuts, tubers, rhizomes or corms. Ghosh *et al.* (2004) also found similar observations.

Table 4. Effects of different herbicide treatments on percent control of grasses over the weedy check (average of two years)

Treatments	Dose (l^{-1} water)	Initial	7 DAA	15 DAA	21 DAA	30 DAA	60 DAA
Glyphosate (market available)	5 ml	0	5	20	60	56	14
Glyphosate(market available)	10 ml	0	20	52	89	74	36
XL – COMBI - SG	3 g	0	16	40	70	65	23
XL – COMBI - SG	5 g	0	21	48	82	71	28
XL – COMBI - SG	7 g	0	32	60	92	76	43
XL – COMBI - SG	10 g	0	37	68	96	85	47
Weedy check	-	0	0	0	0	0	0

Table 5. Effects of different herbicide treatments on percent control of sedges over the weedy check (average of two years)

Treatments	Dose (l^{-1} water)	Initial	7 DAA	15 DAA	21 DAA	30 DAA	60 DAA
Glyphosate (market available)	5 ml	0	19	45	62	40	17
Glyphosate (market available)	10 ml	0	38	67	86	74	41
XL – COMBI - SG	3 g	0	21	42	57	36	16
XL – COMBI - SG	5 g	0	33	59	65	49	28
XL – COMBI - SG	7 g	0	41	81	87	77	43
XL – COMBI - SG	10 g	0	46	86	95	86	50
Weedy check	-	0	0	0	0	0	0

Therefore, from this experiment, it can be concluded that XL-COMBI-SG at 7 and 10 g l^{-1} water and market available glyphosate at 10 ml l^{-1} water can be recommended safely for controlling the weeds in non-cropped areas or in the fallowed crop fields in between the two main crops.

Table 6. Effects of different herbicide treatments on percent control of broadleaved weeds over the weedy check (average of two years).

Treatments	Dose (l^{-1} water)	Initial	7 DAA	15 DAA	21 DAA	30 DAA	60 DAA
Glyphosate (market available)	5 ml	0	23	35	56	27	15
Glyphosate (market available)	10 ml	0	42	67	89	73	48
XL – COMBI - SG	3 g	0	24	33	51	30	18
XL – COMBI - SG	5 g	0	36	56	70	57	31
XL – COMBI - SG	7 g	0	45	73	91	68	52
XL – COMBI - SG	10 g	0	49	88	97	78	59
Weedy check	-	0	0	0	0	0	0

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IMPACT ON ENVIRONMENT AND WEED DYNAMICS OF CHEMICAL WEED MANAGEMENT IN GANGETIC FIELD CROPS

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Abstract: Field Experiments conducted during 2002-05 at the Viswavidyalaya farm and also in the local farmers' field revealed that out of 12 herbicides tested on (a) rice, sulfonyl urea as POE and acid aniline group as PE, (b) in wheat, Isoguard or Clodinafop as POE, (c) in oilseed crops fluchloralin, pendimethalin or trifluralin as PPI or PE, (d) in pulses Quizalofop as POE, (e) in potato, metribuzin as PE, (f) in jute, fenoxaprop-p-ethyl, (g) in onion, oxyfuorfen as POE and (g) in maize atrazine + surfactant as POE, recorded better weed control efficacy and 12-47% higher production. The weed dynamics showed the dominance of the perennial sedge, *Cyperus rotundus*, in up medium land while grasses in the lowlands. All these chemicals showed an initial decrease followed by gradual recovering of beneficial micro-flora populations within a month after application of the chemicals. The pH, soil texture, bulk density, moisture and nutrient content in soil did not show any major changes when these chemicals were applied at the recommended doses within the critical crop-weed competition period of the respective crops.

Key words: Chemical weed management, environment effect, Entisol field crops

Introduction

Gangetic India suffers more than 25% production losses in field crops due to weeds. As a result of 120 cm rainfall during *kharif* (July – October) and high humidity, the grassy and semi-aquatic broadleaf weeds are dominant while in short winter (November – January) and hot summer (February - June) coupled with 50-60 cm scattered rainfall favours the broadleaf weeds to grow profusely in *rabi* and pre *kharif* seasons. The *kharif* rice, linseed/lentil/khesari as paira, summer rice in lowland and jute/soybean/groundnut/summer rice/sesame/green or blackgram/ maize/vegetables, *kharif* rice/groundnut/vegetables, and potato/rapeseed-mustard/wheat/onion/ sunflower/safflower in up medium lands are the dominant crop sequences in Gangetic Entisol. Lack of timely labour availability and gradual rise of labour wages enforce the farmers for alternate weed management than to its traditional hand weeding. Herbicides due to their effectiveness and easiness in application have become the major control measure of weeds in most Asian rice production system (Kim, 2004) and in India also the reliance on herbicides for managing weeds in rice based cropping system have been increasing sharply. In view of the above the field studies were conducted on rice, wheat, maize, rapeseed-mustard, sesame, potato and onion to find out the efficacy of herbicides on controlling weed flora and effects of herbicides on soil physico-chemical and the status of micro-flora in the rhizosphere soil of *Inceptisol*.

Materials and Methods

Field experiments were conducted during 2002-05 at the Viswavidyalaya farm and also in the local farmers' fields on different field crops. All the recommended package of practices was adopted to grow these crops except weed management. Viswavidyalaya farm situated at 89°E longitudes, 23.5°N latitude with average altitude of 9.75 m above msl. Soil is sandy clay loam in texture with pH 6.83, organic carbon – 0.069%, total nitrogen 0.067%, available phosphorus 26 and available potash 135 kg ha^{-1}. The climatic condition in this areas revealed that the temperature begins to rise from May and reach the maximum in July. It starts dropping from middle of October and gradually attains the minimum in January. The mean monthly rainfall is the highest in July and lowest in January. The average rainfall is 1700 mm per annum of which around 75% rainfall occurs during June to September. The lowest relative humidity is observed in the month of December – January while the maximum observed in July – August. All the experiments were designed as Randomized Complete Block Design (RCBD) or Split Plot Design with three replicates with a plot size of 5 m x 4 m. Soil physico-chemical properties were analyzed by using the standard available methods. The microbial population study on total bacteria, fungi and actinomycetes were analyzed by Serial dilution technique and Pour plate method by using Thornton's agar medium, Martin' Rose Bengal Streptomycin agar medium, and Jensen's agar medium, respectively.

Results and Discussion

The dominated weed flora observed in the rainy season in lowland rice fields was *Echinochloa* sp. (*E. crus-galli*, *E. colona,* and *E. formosensis), Paspalum distichum, Brachiaria platyphylla, Leersia hexandra, Cyperus difformis, Fimbristylis littoraolis C. iria, Eclipta alba, Ammania multiflora, Sphenoclea zeylanica, Lindernia ciliata, Marsilea quadrifolia and Stellaria media.* In up medium summer and winter crops *Chenopodium album, Checorium intybus, Melilotus alba, Solanum nigram, Digera arvensis, Gnaphalium luteoalbum, Physalis minima* and *Portulaca oleracea* were observed besides the grasses *Eleusine indica*, *Digitaria sanguinalis, E. colona*. The most important among all the weed flora was the purple nutsedge (*Cyperus rotundus*). The critical crop-weed competition lies between 20-30 days after sowing (DAS), transplanting (DAT), and planting (DAP) in most of the crops grown in this Inceptisol.

In direct seeded rice application of azimsulfuron at 50 g a.i. ha^{-1} combining with metsulfuron methyl at 4 g a.i. ha^{-1}, recorded a grain yield of rice in par as that obtained in hand weeding (HW) at 20 and 40 DAS due to the efficient control of weed flora. This herbicide treatment did not show any detrimental effects on the population of the total bacteria or actinomycetes at harvest (Table 1).

Table 1. Effect of weed management treatments on Weed Control Efficiency (%), grain yield, population of micro flora of direct seeded rainy season rice during 2005.

Treatments	Dose (g ha^{-1})	Weed Control Efficiency (%)	Grain yield (t ha^{-1})	Total bacteria (CFU x 10^6 g^{-1})	Actinomycetes (CFU x 10^5 g^{-1})
Azimsulfuron + Metsulfuron methyl +0.2 % Surf	50+4	61.06	3.42	35.17	66.23
Azimsulfuron + Metsulfuron methyl +0.2 % Surf	60+4	78.04	2.57	35.77	69.80
Azimsulfuron + 0.2 % Surf	50	44.82	2.73	35.07	68.88
Azimsulfuron + 0.2 % Surf	60	59.22	2.76	35.88	70.17
Metsulfuron methyl + 0.2 % Surf	4	43.42	2.80	35.93	70.18
Pyrazosulfuron-ethyl	30	68.85	3.53	41.40	77.27
Pretilachlor (Sofit)	500	62.21	3.16	37.69	66.26
Hand weeding at 20 and 40 DAS		88.10	3.87	44.22	64.34
Untreated control		-	1.97	35.92	68.29
CD (p=0.05)			0.53	1.35	4.53

In *kharif* transplanted rice, ethoxysulfuron (Sunrice® 60 WG) when applied at 20 and 40 a.i. g ha^{-1} was the most effective against the annual sedges and broadleaf weeds and recorded a yield in par with that obtained in HW plots at 20 and 40 DAT. The minimum grain yield of 3.84 t ha^{-1} was recorded against the untreated control, which was 17.7% lower than the average yield of the herbicidal weed management (Table 2).

Table 2. Yield parameter and yield of transplanted rice during 2005

Treatments	No. of effective tillers m^{-2}	Grain yield (t ha^{-1})	Straw yield (t ha^{-1})
Sunrice® at 15 g ha^{-1}	321.67	4.08	5.61
Sunrice® at 17.5 g ha^{-1}	334.33	4.16	5.65
Sunrice® at 20 g ha^{-1}	371.00	4.52	6.82
Sunrice® at 18.75 g ha^{-1}	366.33	4.48	6.58
Sunrice® at 40 g ha^{-1}	374.67	4.68	6.89
Almix® at 4 g ha^{-1}	389.33	5.01	7.22
2,4-DEE at 400 g ha^{-1}	332.00	4.15	6.15
HW at 20 and 40 DAT	391.00	5.08	7.14
Untreated control	266.00	3.84	5.57
S Em (±)	19.797	0.261	0.374
C.D. (p=0.05)	59.345	0.783	1.120

HW – and weeding. S.Em – standard error of the means. DAT – days after transplanting

The maximum yield of 5.08 t ha^{-1} was recorded in plots that received hand weeding twice, which was closely followed by metsulfuron methyl + chlorimuron ethyl (Almix®) and Ethoxysulfuron (Sunrice® 60 WG) at 20 and 40 g ha^{-1}. The data on straw yield also recorded similar trend as grain yield. This chemical also did not show any phytotoxicity against rice plants. In the follow up crops (Table 3), wheat and lentil, this herbicide did not show phytotoxicity on crop plants or on their production efficiency.

Table 3 Effect of the herbicides on yield of follow-up crop

Treatment	Wheat			Lentil		
	Grain yield (kg ha^{-1})			Seed yield (kg ha^{-1})		
	R_1	R_2	R_3	R_1	R_2	R_3
Sunrice® at 20 g ha^{-1}	2462	2422	2369	889	852	831
Sunrice® at 40 g ha^{-1}	2046	2188	2070	774	802	812
Untreated control	1842	1823	1940	647	658	703

In summer rice application of imazosulfuron, either alone at 40 g ha^{-1} or in combination with anilophos at 300 g ha^{-1} or pretilachlor at 550 g ha^{-1} showed effective weed control and increase in rice yield by 1.0, 0.99 and 1.42 t ha^{-1}, respectively, over the weedy check. Pretilachlor at 750 g ha^{-1} and anilophos at 400 g ha^{-1} also showed 0.67 and 0.56 t ha^{-1} higher grain yield of rice, respectively, over the control treatment. This increase in yield is mainly due to the effective weed control of the chemical herbicides. These herbicides also did not show any harmful effect in the population of the beneficial micro flora at harvest and have no phytotoxicity on rice plants in their respective doses used in this experiment (Table 4). Rice grown in a different culture followed by zero tilled rapeseed experiment weed flora dynamics indicated that winter crops after transplanted rice suffered lesser problem than to direct seeded rice (Table 5). Among the different weed flora the population of *Cyperus rotundus* was the minimum after transplanted rice may be due to the reason of stagnant water helped to manage the rhizome growth and development in a better way. More grassy weeds were observed in the succeeding crops of direct seeded rice but broadleaves were dominated in the winter crops grown after puddle rice (Table 5).

Table 4. Effect of weed management treatments on grain yield, straw yield and dry weight of weeds of summer season rice during 2003.

Treatment	Dry weight of weeds ($g\ m^{-2}$) at 42 DAA			Grain yield ($t\ ha^{-1}$)
	Grasses	Sedges	Broadleaves	
Imazosulfuron 20 $g\ ha^{-1}$	8.33	4.43	8.84	4.81
Imazosulfuron 30 $g\ ha^{-1}$	6.74	3.52	6.58	5.03
Imazosulfuron 40 $g\ ha^{-1}$	3.80	2.41	4.88	5.44
Imazosulfuron 20 $g\ ha^{-1}$ + Anilophos 250 $g\ ha^{-1}$	2.64	2.06	4.40	5.28
Imazosulfuron 30 $g\ ha^{-1}$ + Anilophos 300 $g\ ha^{-1}$	3.32	2.18	3.94	5.36
Imazosulfuron 40 $g\ ha^{-1}$ + Anilophos 300 $g\ ha^{-1}$	2.64	1.56	2.79	5.43
Imazosulfuron 20 $g\ ha^{-1}$ + Pretilachlor 337.5 $g\ ha^{-1}$	2.24	1.97	3.39	5.74
Imazosulfuron 30 $g\ ha^{-1}$ + Pretilachlor 550 $g\ ha^{-1}$	3.05	2.01	3.00	5.80
Imazosulfuron 40 $g\ ha^{-1}$ + Pretilachlor 550 $g\ ha^{-1}$	2.27	1.42	2.31	5.86
Pretilachlor 750 $g\ ha^{-1}$	3.22	2.94	5.38	5.11
Anilophos 400 $g\ ha^{-1}$	3.90	3.12	7.14	5.00
Weedy check	12.04	5.38	11.41	4.44
S.Em. (±)	0.323	0.144	0.612	0.331
CD (p=0.05)	0.946	0.423	1.797	0.997

S.Em – standard error of the mean.

Table 5. Effect of weed management treatments on grain yield and dry weight of weeds in rice-rapeseed crop sequence (pooled over two years) during 2004-05

Treatment	Paddy yield ($t\ ha^{-1}$)	Zero till Rapeseed yield ($t\ ha^{-1}$)	Dry weight of weeds in paddy at 60 DAS ($g\ m^{-2}$)		
			Grasses	Sedges	Broadleaves
Direct sown rice + Ambica paddy weeder	2.364	0.689	2.76 (7.15)	2.28 (4.75)	1.67 (2.30)
Beushaning rice	2.622	0.629	2.51 (5.86)	1.67 (2.41)	1.65 (2.23)
Puddled direct seeded rice	3.242	0.688	1.13 (0.79)	0.74 (0.06)	1.50 (1.76)
Transplanted rice	3.621	0.675	0.81 (0.16)	0.75 (0.07)	1.28 (1.16)
CD (p=0.05)	0.278	0.028	0.122	0.184	0.112

Figures in the parentheses indicates the original values

In the experiment on wheat the zero tilled proved better than the conventional tillage. Among the herbicides Sulfosulfuron at 25 $g\ ha^{-1}$ and the Isoguard at 1000 $g\ ha^{-1}$ recorded statistically at par grain yieldas that obtained in weed free treatments. Therefore in this Gangetic Entisol zero till wheat treated with either Sulfosulfuron or a mixture of Isoproteuron and 2,4-D showed better efficacy in increasing weed control that reflected in higher grain yield over the conventional tillage practices (Table 6). In potato, metribuzin at 600 g/ha, hand weeding twice, and Pendimethalin at 560 $g\ ha^{-1}$ recorded 83.2, 85.5 and 69% higher tuber yield, respectively, over the weedy check proving the efficacy of the herbicides (Table 7).

Table 6. Effect of weed management treatments on grain yield and dry weight of weeds of wheat (pooled over two years) during 2003-04.

Treatment	Grain yield ($t\ ha^{-1}$)	Weed index	Dry weight of weeds at 45 DAS ($g\ m^{-2}$)		
			Grasses	Sedges	Broadleaves
Tillage					
Zero tillage	3.12	13.69	6.87	15.88	3.59
Conventional tillage	2.75	15.90	7.30	8.45	10.73
CD at p=0.05	0.152		0.334	1.586	1.001
Weed Management					
Untreated control	2.35	31.69	33.03	52.43	36.29
Weed free	3.44		0.00	0.00	0.00
Isoguard at 750 $g\ ha^{-1}$	2.71	21.22	4.10	7.10	1.73
Isoguard at 1000 $g\ ha^{-1}$	3.07	10.76	2.22	4.07	1.26
Sulfosulfuron at 25 $g\ ha^{-1}$	3.18	7.56	2.23	2.50	1.29
Clodinafop at 60 $g\ ha^{-1}$	2.84	17.44	0.94	6.89	12.90
CD at p=0.05	0.057		0.603	1.326	1.432

Table 7. Weed management treatments on grain and straw yield, population of micro flora and WCE of potato during 2003.

Treatments	Dose (g/ha)	Weed control efficiency (%) at 90 DAP	Tuber yield ($t\ ha^{-1}$)	Net production value	NS-N fixing bacteria (CFU x $10^5\ g^{-1}$)	P-solubilizing microorganism (CFU x $10^5\ g^1$)
Un-weeded Control		-	13.71	0.74	231.65	84.80
Hand weeding	15 and 30 DAP	92.59	25.43	1.85	239.39	82.00
Linuron	375	47.52	19.57	1.40	233.61	74.36
Metribuzin	450	61.06	20.04	1.44	206.85	62.79
Alachlor	750	64.94	21.32	1.74	188.15	54.95
Metribuzin	600	84.39	25.11	2.04	172.19	47.03
Pendimethalin	560	79.41	23.21	1.72	227.41	48.13
CD at p=0.05	--	-	0.402	0.00	9.28	9.36

However, the herbicide treatments reduced the micro-flora populations initially. In the case of metribuzin - NS N-fixing and P-solubilizing bacteria were recorded at 25.7% and 44.5% less populations, respectively, than the weedy check at 30 days after planting, but the populations recovered quickly.

The pre-emergence application of pendimethalin when combined with the soil solarization or pre-plant incorporation of trifluralin provided higher weed control efficacy and higher yield (Table 8). The total bacteria population was found quite lower in case of pre-plant incorporation of trifluralin as compared to Soil solarization with transparent polyethylene sheet but the reverse was found in case of actinomycetes and fungi population through out the growth period (Table 9). The experiment conducted on the evaluation of the bio–efficacy of fenoxaprop–p-ethyl (Whip super® 9% EC) in jute in this Inceptisol it revealed that the higher two doses of the Fenoxaprop-p-ethyl at 56.25 $g\ ha^{-1}$ and 67.5 $g\ ha^{-1}$ because of its higher grassy weed control ability recorded a fibre and stick yield in par with the standard quizalofop-ethyl at 62.5 $g\ ha^{-1}$

or hand weeding at 20 and 40 DAS (Table 10). Furthermore, this herbicide did not show any phytotoxicity in jute plants.

Table 8. Effect of treatment on dry weight of weeds (g m^{-2}) at 40 DAS in groundnut (pooled) during 2004 and 2005.

Treatments	Pod yield (t ha^{-1})	Weed index	Dry weight of weeds at 45 DAS (g m^{-2})		
			Grasses	Sedges	Broadleaves
Soil solarization	3.25	14.94	2.27	8.21	1.75
Pre-plant incorporation of Trifluralin	2.99	12.89	3.52	13.75	2.69
CD at p=0.05	0.043	-	0.298	0.820	0.243
Weed Management					
Untreated control	2.57	27.75	7.67	26.42	5.64
Hand weeding at 15 and 30 DAS	3.31	6.36	0.86	2.24	0.53
Weed free	3.53	-	0.00	0.00	0.00
Propazizafop at 100 g ha^{-1}	2.99	15.61	0.57	19.10	3.46
Alachlor at 2500 g ha^{-1}	3.07	13.29	3.70	10.31	1.17
Pendimethalin at 1000 g ha^{-1}	3.26	7.80	2.96	2.83	0.52
CD at p=0.05	0.065	-	1.287	1.151	0.310

Table 9. Effect of weed management treatment on microbial population in groundnut (pooled) during 2004 and 2005

Treatment	Total bacteria (CFU x 10^6 g^{-1})		Actinomycetes (CFU x 10^5 g^{-1})		Fungi (CFU x 10^4 g^{-1})	
	15 DAS	At Harvest	15 DAS	At Harvest	15 DAS	At Harvest
Soil solarization	31.03	71.06	13.42	36.89	18.92	53.50
Pre-plant incorporation of Trifluralin	23.42	49.22	57.70	74.75	51.06	127.31
CD at p=0.05	1.484	2.909	3.862	5.550	1.586	9.158
Weed Management						
Untreated control	23.50	49.75	37.50	65.00	34.25	164.00
Hand weeding at 15 and 30 DAS	28.50	75.92	36.00	74.84	34.50	93.75
Weed free	23.84	81.83	39.75	79.00	31.00	62.17
Propazizafop at 100 g ha^{-1}	30.58	64.58	30.50	59.67	36.92	146.83
Alachlor at 2500 g ha^{-1}	26.67	37.58	35.67	25.67	35.17	43.25
Pendimethalin at 1000 g ha^{-1}	30.00	51.17	33.92	30.75	38.08	32.42
CD at p=0.05	1.478	3.902	4.495	5.769	2.672	8.802

These findings corroborate the results obtained from the experiments conducted by Shaik Mohammad (2001) and Datta *et al.* (2005) in rice, Singh and Mishra (2003) in rice–wheat crop sequence, Patel and Barevadia (2003), and Ghosh *et al.* (2004) on vegetables and their succeeding crops, Taab and Alizadeh (2004) on lentils, etc. The pH, soil texture, bulk density, moisture and nutrient content in soil did not show any major changes when these chemicals were applied in recommended doses within the critical crop weed competition period of the respective crops.

From the findings of these experiments conducted in the Inceptisol of India it can be concluded that eco-safety chemicals could be able to replace the traditional costly physical method of weed management without affecting much to the environment as well as the soil physico-chemical and microbial properties.

Table 10. Fibre and stick yield of jute during 2005

Treatments	Fibre yield (q ha^{-1})	Stock yield (q ha^{-1})
Fenoxaprop-p-ethyl at 45 g ha^{-1}	27.80	70.00
Fenoxaprop-p-ethyl at 56.25 g ha^{-1}	31.93	83.13
Fenoxaprop-p-ethyl at 67.5 g ha^{-1}	33.03	85.00
Quizalofop-ethyl at 62.5 g ha^{-1}	32.15	82.13
Pendimethalin at 600 g ha^{-1}	23.25	64.25
Hand Weeding 20 and 40 DAS	34.58	90.00
Untreated Control	20.20	58.50
S Em ±	1.351	2.634
C.D. (p=0.05)	4.015	7.825

S.Em – standard error of the mean.

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FIELD EVALUATION OF LEAF EXTRACTS OF *Calotropis gigantea* UNDER *INCEPTISOL* OF WEST BENGAL

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Abstract: A field investigation was carried out in Kalyani 'C' Block farm of Bidhan Chandra Krishi Viswavidyalaya, during summer season of 2004 and 2005 to evaluate the efficacy of *Calotropis gigantea* leaf extract in soybean under *Inceptisol* of West Bengal. The results revealed that pre-emergence application *Calotropis* leaf extract and pendimethalin significantly reduced the grass weed population at early stages of crop growth. Pre-emergence application of *Calotropis gigantea* leaf extract was effective to keep the weeds under check during initial period. But thereafter, its activity gradually reduced. All the weed management treatments have ensured lowest weed pressure when they were integrated with narrow row spacing. Post emergence application of quizalofop-ethyl, *Polygonum* mulching followed by hoeing and pre-emergence application of *Calotropis* leaf extract also resulted significantly higher yield attributes and yield of soybean according to their merit of weed controlling ability.

Key words: *Calotropis* leaf extracts, bioherbicide, soybean, weed management

Introduction

Soybean is grown in all three cultivating seasons in India under conducive environments for weed infestation. The losses due to weeds vary from 20 to 77 per cent depending upon the type and intensity of infestation (Tiwari and Kurchania, 1990). Therefore, weed management is the most important aspect that plays a crucial role in exploiting the yield potential of soybean, provided other inputs are not limiting (Chhokar *et al.* 1996). The nature of yield response to weed management and economic analysis determines the feasibility of adoption of technology by the growers. Hoeing does not control the weeds within rows and thus allows much more weed competition to crop. On the other hand, use of safer herbicides offers selective and economic weed control (Gill *et al.* 1984) right from the beginning, giving the crop a good start and competitive edge. Selective safer herbicide may control certain species and group of weeds and may not be effective on other weed species. In such a situation, while one group of weeds is effectively eliminated, the other groups take over and offer severe competition to the crop. High dose of herbicides may leave residues in soil to injure next crop to fallow and also create the pollution problem (Chhokar *et al.* 1996). Besides, repeated herbicide use may result in a weed flora shift towards more persistent perennials, build up of herbicide residues in the soil and consumable products (Kathiresan, 2001). Hence, a field investigation was carried out to study the efficacy of bioherbicides such as *Calotropis gigantea* leaf extracts for weed management in soybean with varied row spacing under *Inceptisol* of West Bengal, India.

Materials and Methods

A field experiment was carried out in Kalyani 'C' Block farm of Bidhan Chandra Krishi Viswavidyalaya during summer season of 2004 and 2005 under medium land situation in sandy loam soil with a soil pH of 6.74, available N, P_2O_5 and K_2O of 0.054%, 26.29 kg ha^{-1} and 148.72 kg ha^{-1}, respectively. The soybean (cv. PK-327) was grown with all the recommended package of practices except row spacing and weed management. Three different row spacings of 20 (S_1), 30 (S_2) and 40 (S_3) cm (main plot treatment) and six weed management methods as subplot treatments (*Polygonum* mulching followed by hoeing at 30 DAS = T_1, pre-emergence application of *Calotropis gigantea* aqueous leaf extract (5% w/v) =

T_2, pre-emergence application of pendimethalin (1.0 kg ha^{-1}) = T_3, post emergence application of quizalofop-ethyl (50 g ha^{-1}) = T_4, farmers' practice with hand weeding at 20 and 35 DAS = T_5 and weedy check = T_6) were adopted with three replications in split plot design to evaluate the efficacy of weed management methods in soybean. All the herbicides were applied with knapsack sprayer fitted with flood jet nozzle using the spray volume of 500 l ha^{-1}. The data on weed dry weight and weed count were recorded by using 0.25 m^2 quadrate and expressed in per sq. meter basis. The data on weed count and weed dry matter accumulation showed considerable variation and hence was subjected to $\sqrt{(X+ 0.5)}$ transformation before statistical scrutiny.

Results and Discussion

Effect on weeds

The results related to dry weight of weeds have clearly indicated that the highest dry weight (179.14 g m^{-2}) of weeds was recorded under wider row spacing (40 cm) and the gradual reduction was followed with reduced row spacing (Table1).

Table 1. Effect of weed management on of grass and total weeds in soybean (pooled).

Treatment	Dry weight (g m^{-2}) of grass weeds				Dry weight (g m^{-2}) of total weeds			
	At 15 DAS	At 30 DAS	At 45 DAS	At harvest	At 15 DAS	At 30 DAS	At 45 DAS	At harvest
Row spacing								
S_1	1.65 (2.46)*	3.42 (14.85)	4.11 (23.14)	6.70 (53.39)	2.69 (7.15)	4.65 (25.04)	7.20 (59.90)	11.85 (146.01)
S_2	1.66 (2.47)	3.34 (14.20)	4.20 (23.86)	6.80 (54.49)	2.73 (7.34)	4.64 (24.71)	7.40 (62.97)	12.10 (151.98)
S_3	1.67 (2.49)	3.74 (17.86)	4.68 (29.54)	7.47 (65.17)	2.77 (7.55)	5.12 (30.18)	8.19 (77.37)	13.17 (179.14)
S. Em (±)	0.020	0.059	0.071	0.064	0.028	0.055	0.079	0.107
C.D. (p=0.05)	NS	0.192	0.232	0.209	NS	0.179	0.258	0.349
Weed management methods								
T_1	1.39 (1.44)	4.60 (21.24)	3.12 (9.43)	7.68 (58.70)	2.30 (4.84)	5.74 (32.79)	4.91 (23.74)	11.61 (134.94)
T_2	1.22 (0.99)	3.78 (13.94)	6.73 (45.08)	9.40 (88.62)	2.18 (4.29)	5.03 (24.96)	10.07 (101.32)	14.98 (224.91)
T_3	1.13 (0.78)	2.67 (6.81)	4.43 (19.39)	7.10 (50.09)	2.03 (3.66)	3.84 (14.35)	7.41 (54.80)	10.90 (118.33)
T_4	2.08 (3.86)	1.53 (1.91)	1.43 (1.58)	1.53 (1.90)	3.29 (10.43)	3.98 (15.44)	8.18 (66.89)	11.34 (128.45)
T_5	2.08 (3.86)	1.49 (1.77)	1.55 (1.94)	5.29 (27.82)	3.29 (10.40)	1.91 (3.23)	2.95 (8.31)	9.21 (84.89)
T_6	2.09 (3.91)	6.93 (48.15)	8.71 (75.68)	10.92 (118.96)	3.30 (10.47)	8.32 (69.08)	12.06 (145.62)	16.20 (262.23)
S. Em (±)	0.027	0.065	0.078	0.079	0.042	0.090	0.083	0.098
C.D. (p=0.05)	0.076	0.184	0.221	0.223	0.121	0.255	0.235	0.277

*Data in parentheses indicate the original values. DAS – days after sowing. S.Em – standard error of the mean

Low weed pressure and formation of close canopy at narrow row spacing helped the crop plant in smothering the weeds. These findings are in conformity with the findings of Singh and Bhan (2000). The different weed management approaches significantly reduced dry matter accumulation by the weeds throughout the stages of crop growth (Table 1).

In all the cases weedy check recorded the maximum dry weight of grass weeds as well as total weeds as expected, whereas pre-emergence application of pendimethalin and *Calotropis* leaf extract recorded significantly lower value at 15 DAS. Post emergence application of quizalofop-ethyl showed maximum controlling ability of grass weeds during 45 DAS and at harvest stage of soybean. Pre-emergence application of pendimethalin was found next best treatment after farmers' practice in respect of total weed controlling ability which provided lower weed pressure since crop establishment till harvest. On the other hand, pre-emergence application of *Calotropis gigantea* leaf extract was effective to keep the weeds under check during the initial period. However, its activity gradually decreased thereafter. This may be due to that, being a plant material it was decomposed by the soil microflora and sunlight very quickly. This plant product mainly inhibited the seed germination and physiological metabolism of grass weed category and to some extent on sedge but not the broadleaf weeds (Table 1). Similar germination inhibition was recorded by Oudhia and Tripathi (1997) and Swain *et al.* (2005). The higher weed control efficiency of *Calotropis gigantea* leaf in transplanted rice was also observed by Mandal and De (2001) under red and lateritic soil of West Bengal.

All the weed management treatments have ensured lowest weed pressure when they were integrated with narrow row spacing (Table 2). Initial weed controlling ability of different weed management approaches according to the order of merit, have created an environment of lesser crop - weed competition during initial crop growth period that resulted higher weed control efficiency.

Table 2 Interaction effect of weed management on dry weight (g m^{-2}) of grasses and total weeds in soybean (pooled).

Treatment	Dry weight (g m^{-2}) of grass weeds						Dry weight (g m^{-2}) of total weeds					
	Weed management methods											
	T_1	T_2	T_3	T_4	T_5	T_6	T_1	T_2	T_3	T_4	T_5	T_6
Row spacing	**At 15 DAS**											
S_1	1.35 (1.35)*	1.19 (0.93)	1.11 (0.75)	2.09 (3.93)	2.09 (3.91)	2.09 (3.90)	2.18 (4.33)	2.15 (4.18)	2.00 (3.54)	3.26 (10.24)	3.27 (10.29)	3.28 (10.34)
S_2	1.40 (1.46)	1.23 (1.01)	1.13 (0.79)	2.07 (3.85)	2.06 (3.79)	2.09 (3.93)	2.30 (4.80)	2.19 (4.34)	2.03 (3.65)	3.29 (10.42)	3.29 (10.40)	3.29 (10.42)
S_3	1.42 (1.51)	1.24 (1.04)	1.13 (0.79)	2.06 (3.81)	2.08 (3.87)	2.08 (3.89)	2.43 (5.40)	2.19 (4.34)	2.06 (3.78)	3.32 (10.62)	3.31 (10.50)	3.33 (10.65)
		S × T			T × S			S × T			T × S	
S. Em (±)		0.048			0.066			0.073			0.102	
C.D.(p=0.05)		NS			NS			NS			NS	
Row spacing	**At 30 DAS**											
S_1	4.54 (20.15)	3.62 (12.72)	2.54 (6.06)	15.0 (1.83)	1.45 (1.60)	6.86 (46.74)	5.59 (30.85)	4.82 (22.77)	3.67 (13.04)	3.86 (14.44)	1.78 (2.70)	8.18 (66.43)
S_2	4.22 (18.49)	3.72 (13.37)	2.59 (6.30)	1.51 (1.87)	1.45 (1.64)	6.55 (43.52)	5.40 (29.26)	4.94 (23.93)	3.75 (13.68)	3.96 (15.29)	1.85 (2.95)	7.93 (63.13)
S_3	5.04 (25.07)	4.00 (15.74)	2.89 (8.08)	1.58 (2.02)	1.58 (2.08)	7.38 (54.17)	6.22 (38.25)	5.34 (28.18)	4.09 (16.34)	4.13 (16.58)	2.11 (4.05)	8.84 (77.68)
		S × T			T × S			S × T			T × S	
S. Em (±)		0.113			0.162			0.157			0.216	
C.D.(p=0.05)		0.320			0.470			NS			NS	
Row spacing	**At 45 DAS**											
S_1	2.81 (7.51)	6.48 (41.52)	4.13 (16.80)	1.33 (1.31)	1.50 (1.80)	8.38 (69.91)	4.55 (20.26)	9.77 (92.97)	6.89 (47.11)	7.82 (60.79)	2.82 (7.52)	11.45 (130.73)
S_2	2.98 (8.49)	6.50 (41.85)	4.27 (17.93)	1.42 (1.54)	1.52 (1.84)	8.48 (71.54)	4.80 (22.57)	9.79 (95.32)	7.27 (52.42)	7.97 (63.04)	2.86 (7.77)	11.71 (136.72)
S_3	3.57 (12.29)	7.21 (51.87)	4.89 (23.43)	1.54 (1.89)	1.63 (2.19)	9.27 (85.59)	5.37 (28.40)	10.77 (115.67)	8.08 (64.89)	8.76 (76.25)	3.16 (9.63)	13.03 (169.39)
		S × T			T × S			S × T			T × S	
S. Em (±)		0.127			0.160			0.143			0.193	
C.D.(p=0.05)		0.359			0.465			0.404			0.552	
Row spacing	**At harvest**											
S_1	7.40 (54.24)	8.93 (79.72)	6.88 (46.97)	1.37 (1.40)	4.95 (24.10)	10.69 (113.89)	11.04 (121.55)	14.41 (207.47)	10.37 (107.14)	10.88 (117.94)	8.64 (74.18)	15.76 (247.81)
S_2	7.45 (54.97)	9.04 (81.83)	6.99 (48.46)	1.53 (1.87)	5.06 (25.19)	10.73 (114.63)	11.27 (126.82)	14.55 (211.79)	10.74 (115.14)	11.28 (127.02)	8.84 (77.85)	15.93 (253.27)
S_3	8.20 (66.88)	10.23 (104.32)	7.43 (54.84)	1.70 (2.44)	5.87 (34.17)	11.35 (128.37)	12.52 (156.47)	15.99 (255.46)	11.58 (134.22)	11.87 (140.41)	10.15 (102.66)	16.91 (285.62)
		S × T			T × S			S × T			T × S	
S. Em (±)		0.137			0.199			0.180			0.269	
C.D.(p=0.05)		0.387			0.579			0.509			0.789	

*Data in parentheses indicate the original values. S.Em – standard error of the mean.

Effect on yield attributes and yield

Weedy check recorded 57.3 and 25.9% lower number of pods plant^{-1} and number of seeds per pod, respectively than that of farmers' practice, which was ultimately reflected 59.7% lower seed yield due to the maximum weed pressure since crop establishment till harvest of the crop (Table 3). Similar type of yield reduction have also reported by several workers under different condition Tiwari *et al.* (1996) and Kurchania *et al.* (2001).The maximum value of yield attributes was recorded from farmers' practice with hand weeding at 20 and 45 DAS, mainly due to better aeration and minimum crop - weed competition throughout the crop growth stage. Similarly, pendimethalin, the next best treatment recorded 114.8% and 30.1% higher number of pods per plant and number of seeds per pod, respectively over weedy check (Table 3) due to its higher weed controlling ability that offered conducive environment for crop growth and more competitiveness to crop.

The post emergence application of quizalofop-ethyl, *Polygonum* mulching followed by hoeing and pre-emergence application of *Calotropis* leaf extract also resulted in a significantly higher ($p<0.05$) yield attributes and yield of soybean according to their merit of weed controlling ability (Table 3).

Table 3. Effect of weed management on yield parameters and seed yield of soybean (pooled).

Treatment	Number of pods plant^{-1}	Number of seeds pod^{-1}	Seed yield (t ha^{-1})
Row spacing			
S_1	29.37	2.40	1.733
S_2	31.72	2.63	1.557
S_3	31.82	2.53	1.120
S. Em (±)	0.303	0.031	0.0349
C.D. (p=0.05)	0.988	0.101	0.1138
Weed management methods			
T_1	31.46	2.50	1.541
T_2	22.38	2.29	1.030
T_3	37.68	2.72	1.811
T_4	35.64	2.66	1.750
T_5	41.11	2.82	1.915
T_6	17.54	2.09	0.771
S. Em (±)	0.410	0.036	0.0267
C.D. (p=0.05)	1.160	0.102	0.0755

S.Em – standard error of the mean.

An overall assessment of interaction effect between different row spacings and weed management methods gave a clear indication that higher yield attributes and yield were recorded from the weed management treatments under different row spacing according to their weed control efficiency as well as intra-plant competition for growth resources (Table 4). Performance of *Polygonum* mulching followed by hoeing at 30 DAS, pre-emergence application of pendimethalin and *Calotropis* leaf extract and post emergence application of quizalofop-ethyl was better under narrow row spacing (20 cm and 30 cm) due to lower weed pressure as compared to wider row spacing of 40 cm.

Table 4. Interaction effect of weed management on yield parameters and seed yield of soybean (pooled).

Treatment	Weed management					
	T_1	T_2	T_3	T_4	T_5	T_6
Row spacing			Number of pods plant^{-1}			
S_1	30.58	22.68	34.97	32.10	38.58	17.28
S_2	33.15	22.73	38.10	36.13	42.13	18.08
S_3	30.65	21.73	39.97	38.70	42.62	17.25
		S × T			T × S	
S. Em (±)		0.711			1.013	
C.D. (p=0.05)		2.011			2.944	
Row spacing			Number of pods plant^{-1}			
S_1	2.45	2.29	2.51	2.44	2.58	2.10
S_2	2.68	2.54	2.78	2.64	2.86	2.27
S_3	2.38	2.03	2.86	2.92	3.02	1.93
		S × T			T × S	
S. Em (±)		0.056			0.086	
C.D. (p=0.05)		0.158			0.254	
Row spacing			Seed yield (t ha^{-1})			
S_1	1.914	1.385	2.084	1.944	2.124	0.947
S_2	1.738	1.101	1.913	1.813	1.973	0.802
S_3	0.970	0.604	1.436	1.495	1.649	0.566
		S × T			T × S	
S. Em (±)		0.0463			0.0778	
C.D. (p=0.05)		0.1310			0.2337	

S.Em –standard error of the mean.

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A STUDY ON THE LEACHING OF FLUROXYPYR IN THE SOIL OF AN OIL PALM AGROECOSYSTEM

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Abstract: A study on the leaching of fluroxypyr in the soil of an oil palm agroecosystem was conducted. The method for determination of fluroxypyr in the soil involved two steps namely; the liquid-solid extraction followed by ten times dilution with fluroxypyr-free water, before concentration onto the sorbent and injection into the HPLC-DAD. The efficiency of the method was tested by adding known amounts of fluroxypyr to the soil known to be free from the fluroxypyr residue. The recovery of fluroxypyr, analyzed by high performance liquid chromatography (HPLC) with the diode array detector (DAD) was determined by analyzing soil samples spiked with fluroxypyr standard solution at different levels namely 1, 4, 5, 10, 25, and 50 μg/kg. It was found that the recovery of fluroxypyr ranged from 82% to 107% with the coefficient of variation ranging from 0.50% to 4.7%. The results above showed that the analytical methods used for determination of fluroxypyr in the soil were effective. Therefore, these methods were adopted for the determination of the fluroxypyr content in the soil from the field trial. The trial for determining the leaching of fluroxypyr at an oil palm agroecosystem in Sepang was conducted at the Kuala Lumpur International Oil Palm Plantation. This oil palm estate is situated centrally in the state of Selangor in Peninsular Malaysia, at an altitude 70 m to 100 m above sea level. The study plot covered 6 hectares, and consisted of 9 subplots of 0.66 hectare each. The 9 subplots received three treatments namely; the manufacturer's recommended dosage, double the manufacturer's recommended dosage, and the control without herbicide treatment. Each treatment was done in triplicate and each subplot was separated from the next by a 2 m buffer zone. The herbicide fluroxypyr was sprayed using a knapsack sprayer. The herbicide residue in the soil was monitored at 0, 5, 7, 14, 21, 30, 60 and 90 days after treatment. Soil samples were collected from each replicated plot at different depths (0 – 10, 10 – 20, 20 – 30, 30 – 40 and 40 – 50 cm). Soil samples from the same depth in each plot were bulked together. It was found that at the recommended and the double the recommended dosage of the herbicide, leaching of fluroxypyr in soil at the experimental plots was detected only up to day 7 and 14 after treatment respectively.

Key words: Fluroxypyr, soil, solid phase extraction.

Introduction

Fluroxypyr belongs to the chemical family pyridinoxy acid with the chemical abstract service (CAS) number of 69377-81-7 (Kidd and James, 1991). The chemical structure of fluroxypyr is shown in Figure 1.

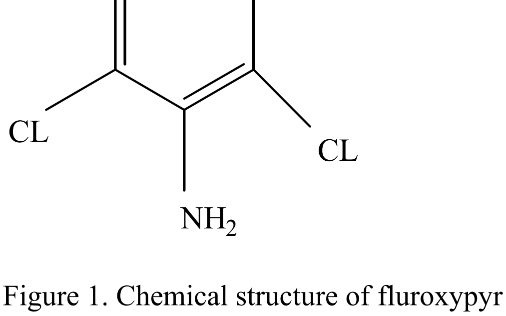

Figure 1. Chemical structure of fluroxypyr

The environmental fate of pesticides has recently caught the attention of researchers because of their pollution potential to the environment. Lake and river water can undoubtedly be contaminated with the runoff water from adjacent agricultural fields if pesticides are not managed properly. The movement of a pesticide from its site of application is significant with regard to the chemical's bioefficacy, availability, degradation, microbial toxicity, phytotoxicity and leaching pattern. Although the mobility of herbicides in the soil has been studied in detail, little is known about the mobility of herbicide metabolites or other groups of chemicals such as insecticides. Some pesticide metabolites that have been associated with insecticidal activity (Chapman and Harris, 1980), enhanced microbial degradation of the parent compound (Somasundaram *et al*. 1989), phytotoxicity (Kaufman and Kearny, 1970) and groundwater contamination (Cohen *et al*. 1984). Therefore, it is imperative to understand pesticide movement and their fate in the soil.

Heavy usage of pesticides in agricultural activities may cause adverse effects on the environment and consequently on human health. The leaching of pesticides into groundwater is of major concern with regard to environmental pollution because it affects the quality of underground water (Lehmann *et al*. 1993). The number of different pesticides in ground and surface water has been found to be increasing steadily (Lehmann and Miller, 1989). Therefore, an understanding of adsorption, desorption and mobility of pesticides in the soil needs immediate attention.

There is a steady increase in the use of pesticides in developing countries although integrated pest management practices are concurrently being promoted and implemented. This increased usage of pesticides has become a growing concern because of the possibility of their presence in food and the danger posed by residues in the environment. Pesticides used in tropical countries could adversely affect environmental quality (Tanabe *et al*. 1990). It was reported that pesticide pollution of ground and surface water caused problems in tropical countries, with heavy rainfall, high humidity and high temperature (Cheah *et al*. 2000; Ismail *et al*. 2004; Khakural *et al*. 1995). There is scarce information on the impact of pesticides on tropical environments such as those found in Malaysia. Although extensive research has been carried out in other countries, the results are not totally applicable in Malaysia because of climatic differences.

Materials and Methods

Experimental design

The study was conducted at the Kuala Lumpur International Airport (KLIA) Oil Palm Plantation owned by the Malaysian Agricultural Horticultural Sdn. Bhd. (MAAH). The study plot was a hilly land of slope 45^o with soil predominantly clayey in texture. The soil had a high content of clay (52.2%), a CEC (cation exchange capacity) of 6.9%, 15% coarse sand, 27% fine sand and 5.8% silt. The soil was classified as clay soil. All soil data are reported on dry weight basis.

Starane® 200 (fluroxypyr, 1-methylheptyl ester, 29.6% w/w, EC, Dow Elanco Ltd.) was applied as an aqueous spray using a knapsack sprayer (nozzle 5/64) at a spraying volume of 250 l/ha. The herbicide was applied at 72.05 g a.i/ha and 144.10 g a.i/ha, which represented the manufacturer's recommended and double the recommended dosage, respectively. The control plot was not treated with herbicide.

Method of extraction for soil samples

The soil samples were thawed before extraction. The method involved extraction of the analyte using liquid-solid extraction, followed by cleaning up using the solid phase extraction. Each 20 g soil sample was mixed with 80 ml of 5% water in acetone. It was then shaken using the ultrasonic bath for 15 min and centrifuged at 3000 rpm for 20 min. Ten ml of the extract were pipetted out and transferred into a 10 ml vial. The extract was then dried in vacuum and re-dissolved in 1 ml of acetonitrile prior to analysis by the HPLC equipped with a diode array detector (DAD) at wavelength of 254 nm.

Results and Discussion

Figure 2 shows the amount of fluroxypyr residue in the soil (μg/kg dry soil) when sprayed at the recommended dosage during the dry season (June to September 2002). It was observed that the fluroxypyr residue in the soil at the recommended dosage had a lower residue as compared to that obtained from soil treated at double the recommended dosage.

Concentration of fluroxypyr (μg/kg)

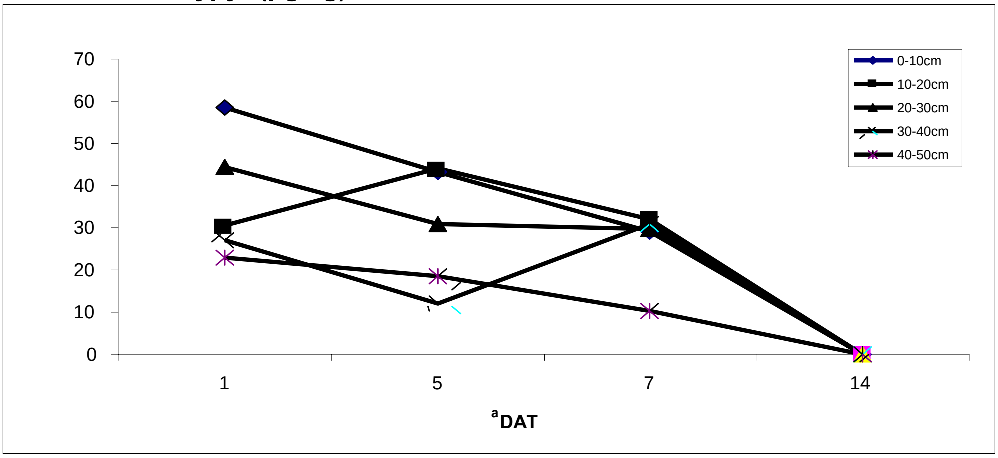

Figure 2. Concentration of fluroxypyr residue in the soil profile when applied at the manufacturer's recommended dosage

At the recommended dosage, on 1 day after treatment (DAT) the total amount of fluroxypyr found in the soil profile (0-50 cm) was 162.9 μg/kg with the highest amount deposited in the top 0-10 cm layer, followed by the next highest deposit in the third layer (20-30 cm). On 5 DAT, the total amount of fluroxypyr was reduced to 148.9 μg/kg. The total amount of fluroxypyr was further reduced to 132.1 μg/kg on day 7 after spraying. Eventually on day 14, fluroxypyr was not detected at all depths.

At double the recommended dosage, on day 1 after treatment, fluroxypyr residue was detected at all depths (248.5μg/kg) with the highest residue at the depth of 0-10 cm (Figure 3). On 1 DAT, the amount of fluroxypyr detected was 248.5 μg/kg. The total amount of fluroxypyr was reduced to 85.5% on 5 DAT. On 7 DAT, the total amount of fluroxypyr was further reduced to 70.9%. On 14 DAT, there was a further reduction of fluroxypyr to 42.0%. Finally, fluroxypyr was not detected from day 21 onwards.

In studies done by Bergstrom et al. (1990), a lysimeter was used to study the mobility and decomposition of fluroxypyr in Swedish soils under field conditions. Fluroxypyr levels above the detection limit (5 μg/kg dry soil) were never found below the top (0.2 m) of the clay soil, while in the sandy soil, concentrations just above the detection limit were found occasionally in the deeper soil layers. Concentrations were reduced to undetectable or very low levels within three months after spraying. However, in this study, results showed that fluroxypyr was reduced to undetectable levels (1 μg/kg dry soil) within 21 DAT.

Concentration of fluroxypyr (µg/kg)

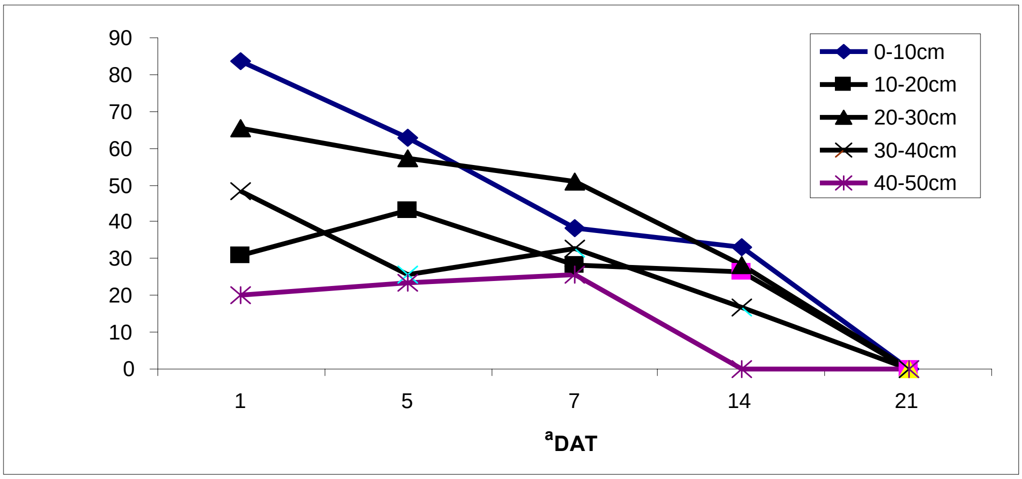

Figure 3. Concentration of fluroxypyr residue in the soil profile when applied at double the manufacturer's recommended dosage

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STUDIES ON GENETIC DIVERSITY AT DNA LEVEL AMONG VARIOUS BIOTYPES OF WILD OATS (*Avena* species)

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Abstract: Weeds cause considerable losses (25-30%) in common wheat. Wild oat is a highly competitive grassy weed of Rabi crops, especially, when associated with the wheat crop and causes approximately 60% out of total weed losses. In the present study, the genetic diversity of 10 biotypes of wild oats (*Avena* species) were analyzed using 9 Randomly Amplified Polymorphic DNA (RAPD) and 2 Simple Sequence Repeat (SSR) primer sets. The RAPD and SSR primers revealed a high level of genetic polymorphism among the various biotypes of wild oats. The RAPD primers GL-A03, GL-A04, GL-A12, GL-B07, GL-B19, GL-C07. GL-D17, GL-D18, GL-E05 and SSR primer sets gdm 86-2B and gdm 87-2D amplified 2.1, 4, 2.4. 3.8, 3.7, 4.78, 2.33, 4, 2.58, 1.2 and 3.1 loci per biotype, respectively. The Range of genetic distances estimated by using individual primers was from 0 – r100%, indicating that these biotypes were genetically very diverse and possess significant polymorphism. Preliminary PCR amplification results suggest that the two wheat chromosome specific SSR markers (gdm 86-2B and gdm 87-2D) could be used successfully to investigate genetic diversity in wild oats. Wild oat biotypes showing the maximum genetic distances (D.I. Khan white biotype and Swat biotype) should be used in future breeding programs for improving the genetic base of wild oat germplasm, while using it as a food or feed crop.

Key words: Wild oats, polymorphism, genetic diversity

Introduction

Wild oats (*Avena* species) are one of the most troublesome weed species in grain fields (Muur, 1999). Wheat is damaged by a number of biotic and abiotic stresses which cause great losses to the quality and quantity of produce (McIntosh, 1998). Wild oats (*Avena species*) is highly competitive with wheat and causes severe reductions (25-30%) in yield. A 68% control increased wheat yield by 13.54% (Samunder *et al.* 1994). It is difficult to differentiate these weed from wheat at early stages as the seedlings of the two species are similar. Life cycle of wild oats also coincides with that of the wheat crop. The problem in identification at early stage impedes in the manual control, hence herbicides application becomes inevitable.

As a result of the intensive and continuous use of herbicides over the last five decades, weed populations change in genetic composition such that the frequency of resistant alleles and resistance individuals increase (Jasieniuk *et al*. 1996). The development and evolution of weeds resistant to the herbicides is a result of these changes, thus management of weeds is becoming increasingly more difficult and complex. Further pollen movement of resistant alleles within populations occurs in several weed species (Maxwell, 1992; Stallings *et al*. 1993). Biotypes of various weeds show both cross resistance (Hall *et al.* 1994) and multiple resistance (Gill, 1992), this acquired resistance is very challenging for weed scientists. These biotypes could be controlled at much lower rates as compared to those recommended and specified in the label , but accurate information regarding the genetic tolerance of biotypes of these weeds in Pakistan is lacking.

The most important causal factor for failures observed in weed control is the biodiversity (Dekker, 1997a). Harper (1977) defined five levels of diversity that a plant may meet among its neighbors. An individual weed experiences diversity in its neighbors *viz:* genetic variants within a species; somatic polymorphism of plant parts; habitat microsite diversity; age-state diversity within the community and diversity at groupings at a higher level than species. Weed diversity is the adaptive response to these selection pressures.

Biodiversity is most typically seen as genetic polymorphism; the heterogeneity among and within weed species. Understanding the inherent taxonomic diversity of the weed species, species-group, and sub specific variant in weeds is a primary need in weed management. This morphological diversity is utilized to guide weed control as well as provide insight about weed introduction and spread (Dekker, 1997b). Studies of genetic diversity that rely on molecular polymorphism provide new insights into the heterogeneity within weedy populations (Dekker, 1991). New weed genotypes / biotypes are generated in a number of ways, including mutation, recombination, gene flow, and segregation distortion (Solbrig and Solbrig, 1981). This genotypic variation often leads to unpredictable phenotypic biodiversity (Dekker, 1997b). A single genetic mutation leads to several unexpected pleiotropic effects in the herbicide resistant phenotype (Dekker, 1993; LeBaron and Gressel, 1982).

The Random Amplified Polymorphic DNA (RAPD) analysis is a recently developed technique used to determine genetic variation and biosystematic relationships between and within plant populations, classification and characterization of closely related genotypes, identification of crops varieties and genetic diversity within a plant species (Weining and Landridge, 1991). Recently, different biotypes of weeds were identified by using RAPD marker (Mitra *et al*. 1998). RAPD also requires very low amount of total genomic DNA, thus it is easier and faster for generating data (Nissen *et al*. 1995). Another commonly used marker system is simple sequence repeats (SSRs). The distribution of simple sequence repeats (SSRs) show that SSRs are far more common in plant genomes than previously estimated (Cardle *et al.,* 2000). DNA marker data could be used as selection criteria for biocontrol by providing an estimate of a plant's genetic diversity relative to other potential target species (Nissen *et al*. 1995). Genetically heterogeneous, sexually reproducing weed populations may require a diverse array of biological events, while less variable, asexually reproducing species may require fewer agents (Nissen *et al*, 1995). Thus, weed species that rely on asexual forms of reproduction have a significantly greater chance of being controlled biologically than sexually reproducing species. The DNA marker systems offer a more efficient and cost-effective method of identifying compatible bioagent-weed relationship.

Studies on genetic diversity that rely on molecular polymorphism provide new insights into the heterogeneity within weedy populations (Dekker, 1991). Molecular analysis of the genetic diversity in weedy populations has been conducted using several different techniques (Nissen *et al*., 1992; Warwick, 1990). Unfortunately, no such work has been documented in Pakistan previously.

Materials and Methods

Plant materials

Ten biotypes of wild oats (*Avena* species) supplied by Dr. Gul Hassan, the Principal Investigator of the project on “Biology, Ecology and Physiology of Wild Aats”, were used in present study. Table1 shows list of collected biotypes along with their collection sites. All materials were planted in the green house at the Institute of Biotechnology and Genetic Engineering (IBGE), N.W.F.P. Agricultural University, Peshawar, Pakistan.

DNA Extraction

DNA was extracted from all the ten biotypes of wild oats as described by Weining and Langridge (1991). To remove RNA, DNA was treated with 40 micro-gram RNAse-A (0.2 micro liter of commercially supplied RNAse-A purchased from Gene link , USA) at 37^0C for 1 hour, thereafter, the DNA samples were run on 0.8% Agarose gel to check the quality and quantity of DNA and stored at 4°C. A 1:5 dilution of DNA was made in doubled distilled, deionized and autoclaved water for use in Polymerase Chain Reaction (PCR).

Table 1. Different biotypes of Wild oats (*Avena species*) collected from various districts of N.W.F.P. and Islamabad/Rawalpindi.

Name of Biotypes	Collection Site
Dera Ismail Khan White	Tatta Balochan , D.I. Khan
Dera Ismail Khan Black	Rakshah Koat , D.I. Khan
Karak White	Lakki Ghundakki , Karak
Karak Black	Babul Khel , Karak
Peshawar	Malkandher Farm , Peshawar
Charsadda	Mvliano Kala , Charsadda
Mardan	Lundkhawar , Mardan
Malakand	Dargai , Malakand
Swat	Batkhela , Swat
Islamabad / Rawalpindi	NARC , Islamabad

Polymerase Chain Reaction

The PCR were carried out using protocol as described by Devos and Gale, 1992 with modifications. All amplification reactions were carried out using a GeneAmp PCR System 2700 (Applied Biosystem) programmable thermocycler. Simple Sequence Repeats (SSRs) primer sets specific for wheat chromosomes (Roder *et al*. 1995; Liu *et al*. 1999) were also used for) DNA analysis of *Avena* species (wild oats) in order to find any homology of DNA sequences between the two crops and to check preliminary results that SSR primer sets specific for wheat chromosomes can be used for wild oats or not. The amplification products of RAPDs primers were separated on 1.5 % and SSRs primer sets on 3% agarose/TBE Gel and visualized by staining with Ethidium Bromide under ultra-violet (U.V) light and photographed using gel documentation system “Uvitec”.The information regarding nine Decamer Randomly Amplified Polymorphic DNA (RAPD) primers and two SSRs primer sets are given in Tables 2 and 3 respectively.

Table 2. Name, sequence, size, molecular weight and %GC content of 9 RAPD primers used to study genetic diversity among 10 biotypes of wild oats.

Name	Sequence	Size (bp)	Mol. Wt.	% GC
GLA-03	AGTCAGCCAC	10	2996.98	60
GLA-04	AATCGGGCTG	10	3068.02	60
GLA-12	TCGGCGATAG	10	3068.02	60
GLB-05	TGCGCCCTTC	10	2954.97	70
GLB-19	ACCCCCGAAG	10	2981.97	70
GLC-07	GTCCCGACGA	10	3012.99	70
GLD-17	TTCCCCCCAG	10	2923.95	70
GLD-18	GAGAGCCAAC	10	3046.00	60
GLE-05	TCAGGGAGGT	10	3108.04	60

Statistical analysis

For statistical analysis, each band was considered as single locus / allele. Only scorable bands were included in the analysis. Bands were scored as present (1) / absent (0) and bi-variate (1-0) data matrix was generated. Genetic distances were calculated using Unweighted Pair Group of Arithmetic Means (UPGMA) procedure as suggested by Nei and Li (1979).

Genotypes were clustered using computer programme "Popgene32" version 1.31 (http://www.ualberta.ca./~fyeh/fyeh). Based on these analysis dendrograms were generated for RAPD primers to estimate diversity among 10 biotypes of wild oats.

Table 3. Name, sequence, repeats and melting temperatures of 2 SSR primers sets used to study genetic diversity among 10 biotypes of wild oats.

Locus	Sequence (Forward primer)	Sequence (Reverse Primer)	Repeats	T_m
Xgdm86-2B	GGTCACCCTCTCCCATCC	GGCGCTCCATTCAATCTG	(TG)13	60
Xgdm87-2D	AATAATGTGGCAGACAG TCTTGG	CCAAGCCCCAATCTCTCT CTCT	(GT)14	60

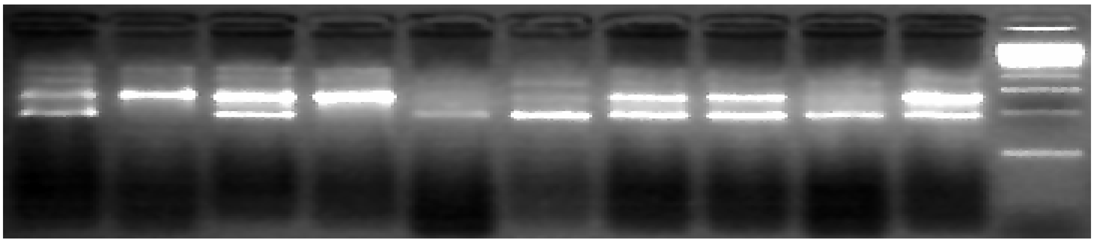
T_m = Melting Temperature, (Annealing temperature = T_m – 5°C)

Results

Genetic diversity analysis based upon randomly amplified polymorphic DNA primers (RAPD)
In crops of agronomic importance such as rye, maize, brassica and wheat, PCR-based assays have been extensively used to study genetic polymorphism. PCR based assays are easy, cheaper, faster and do not require any sequence information on the targeted genome and lower quantities of DNA are required. Randomly Amplified Polymorphic DNA (RAPDs) (Williams *et al.* 1990) is a PCR based technique which has been used for detecting genetic diversity at DNA level among varieties/biotypes /germplasm accessions in may crops of commercial importance including common wheat (Fennimore and Foley, 1998; Heun *et al*.,1994). Nine RAPD primers were used during present study to estimate genetic diversity among 10 biotypes of wild oats (*Avena* species).

The banding pattern obtained by using RAPD primer Genelink-A03, the 10 biotypes of wild oats showed various levels of genetic polymorphism. A total of 21 alleles (bands) were amplified in 10 biotypes giving an average of 2.1 alleles per biotype. The size of amplified fragments, estimated by using 1 kb DNA ladder, ranged from >250bp to >1000bp. Range of genetic distances among biotypes was 0 to 75%. Maximum genetic distance (75%) was estimated for 4 comparisons *viz* Dera Ismail Khan (black) - Dera Ismail Khan (white), Dera Ismail Khan (black) - Peshawar, Dera Ismail Khan (black) - Islamabad and Peshawar - Swat, while 4 comparisons showed homozygosity for the loci amplified by primer, GL-A03.
The PCR amplification profile using Genelink-A04 primer showed various levels of genetic polymorphism for the loci amplified. Total genomic DNA of 2 biotypes (Malakand and Islamabad) was not amplified by using GL-A04 and the results were not included in analysis. A total of 32 alleles were observed in 8 biotypes giving an average of 4 alleles per biotype. The size of amplified fragments ranged from < 500bp to >1000bp. Range of genetic distances estimated was 28 to 100%. Maximum genetic distance (100%) was observed for 3 comparisons, Dera Ismail Khan (white) - Swat, Karak (black) - Swat and Mardan - Swat, while one comparison (Dera Ismail Khan black - Karak white) showed 72% homozygosity for the amplified loci.

The PCR amplification profile of 10 biotypes using Genelink-A12 primer (Figure 1) yielded a total of 24 alleles giving an average of 2.4 alleles per biotype. The size of amplified fragments varied from 500bp to 1000bp. Genetic distances estimated for all the possible combinations ranged from 0 to 100%. Four pairs of biotypes Dera Ismail Khan (black) - Peshawar, Dera Ismail Khan (black) - Swat, Karak (black) - Peshawar and Karak (black) - Swat showed maximum genetic distance (100 %), while three comparisons Dera Ismail Khan (black) - Karak (black), Charsadda - Islamabad and Mardan - Malakand revealed no difference (GD = 0%).



Figrue 1. PCR Amplification profile of 10 wild oats biotypes using RAPD primer GL-A12. From left to right: 1 = Dera Ismail Khan White, 2 = Dera Ismail Khan Black, 3 = Karak White, 4 = Karak Black, 5 = Peshawar, 6 = Charsadda, 7 = Mardan, 8 = Malakand, 9 = Swat and 10 = Islamabad and M = Molecular size of I kb ladder DNA (size of fragments are indicated in bp on right).

The PCR amplification profile of 10 biotypes of wild oats using GL-B07 primer showed various levels of genetic polymorphism. A total of 38 alleles (bands) were amplified in 10 biotypes giving an average of 3.8 alleles per biotype. The size of amplified fragments ranged from 500bp to 1000bp. Range of genetic distances observed using RAPD GL-BO7 was 0 to 60%. Six comparisons showed 60 % genetic dissimilarity while 9 comparisons showed no genetic difference using RAPD primer GL-B07.

A total of 37 alleles were amplified using RAPD primer Genelink-B19 in 10 biotypes giving an average of 3.7 alleles per biotype. The size of amplified fragments ranged from >250bp to 1000bp. Genetic distances (GD) among the 10 biotypes ranged from 0% to 60 %. Maximum genetic distance (60 %) was estimated for 2 comparisons (Dera Ismail Khan white – Peshawar and Karak black - Peshawar), while 4 comparisons *viz* Dera Ismail Khan (black) - Karak (White), Mardan - Malakand, Mardan - Islamabad and Malakand - Islamabad showed homozygosity for the loci amplified using GL-BO7.

A total of 43 alleles (bands) were amplified in 9 biotypes giving an average of 4.8 alleles per biotype. Total genomic DNA of one biotype (Islamabad) was not amplified using GL-C07 and the result is not included in this analysis. The size of amplified fragments ranged from >500bp to > 1000bp. Genetic distances among biotypes (in percentage). Range of genetic distances estimated was 14 to 86%. Maximum genetic distance (86%) was observed for one comparison (Peshawar - Swat), while two comparisons (Dera Ismail Khan White - Peshawar and Dera Ismail Khan Black - Peshawar) were only 14% heterozygous for the detected loci.

The banding pattern of wild oats biotypes obtained using Genelink-D17 primer showed various levels of genetic polymorphism. Total genomic DNA of one biotype (Swat) was not amplified using GL-D17 and the result is not included in this analysis. A total of 21 alleles (bands) were amplified using 9 biotypes giving an average of 2.3 alleles per biotype. The size of amplified fragments varied from 500bp to approximately 1000bp. Genetic distances estimated for all the possible combinations ranged from 0 to 75%. Two pairs of biotypes (Dera Ismail Khan (white) - Mardan and Charsadda - Mardan showed maximum genetic distance (75%), while four pairs (Dera Ismail Khan (white) - Charsadda, Karak (black) - Malakand, Karak (black) - Islamabad and Malakand - Islamabad) revealed no difference (GD = 0%) in the loci detected using GL-D17 primer.

The amplification PCR profile of 10 biotypes of wild oats (*Avena* species) using GL-D18 primer amplified a total of 40 alleles in 10 biotypes giving an average of 4 alleles per biotype. The size of amplified fragments varied from 500bp to > 1000bp. Range of genetic distances observed using RAPD GL-D18 primer was 0 to 100%. One comparison (Malakand - Swat) showed 100 % genetic dissimilarity, while 3 comparisons (Dera Ismail Khan white -

Dera Ismail Khan black, Dera Ismail Khan white - Karak white and Dera Ismail Khan black - Karak white) showed no genetic difference using RAPD primer GL-D18.

A total of 16 alleles (bands) were observed in 7 biotypes giving an average of 2.6 alleles per biotype using Genelink-E05 primer. The total genomic DNA of 3 biotypes (Charsadda, Swat and Islamabad) was not amplified using GL-E05 and the results are not included in analysis. Total genomic DNA of 3 biotypes (Charsadda, Swat and Islamabad) were not amplified using GL-E05 and so the result are not included in the analysis The size of amplified fragments varied from > 250bp to 1000bp. Range of genetic distances estimated was 0 to 33 %. Maximum genetic distance (33 %) was observed for 10 comparisons, while 11 comparisons were completely homozygous for the detected loci.

Average genetic distances of 10 biotypes of wild oats using 9 RAPD primers ranged from 24 to 64.14% (Table 4). Maximum average genetic distance (64.14%) were noted for one comparison (Dera Ismail Khan Black – Swat), closely followed (63.14%) by (Peshawar – Swat), while minimum average genetic distance (24.67 %) was recorded for one comparison (Dera Ismail Khan white – Islamabad), closely followed (27.50%) by one comparison (Mardan and Malakand). Generally average genetic distances were in the range of 30 - 50%.

Table 4. Average genetic distances (in percentage) among 10 *Avena* Species (wild oats) based on 9 RAPD Primers.

Biotyes	D.I.K (W)	D.I.K (B)	KRK (W)	KRK (B)	PESH	CHRD	MRD	MKD	SWT
D.I.K. (W)	-								
D.I.K.(B)	36.67	-							
KRK(W)	31.67	28.00	-						
KRK(B)	33.67	25.89	39.33	-					
PESH	47.11	49.44	54.78	43.78	-				
CHRD	29.5	48.38	45.75	43.38	46.25	-			
MRD	56.67	45.78	44.33	48.78	34.56	45.75	-		
MKD	42.88	40.88	42.50	39.00	31.50	29.71	27.50	-	
SWT	56.57	64.14	61.57	54.71	63.14	56.28	61.57	62.00	-
ISLM	24.67	40.16	42.83	31.67	34.50	28.00	43.16	26.16	39.20

D.I. K (W) = Dera Ismail Khan White, D.I. K (B) = Dera Ismail Khan Black, KRK (W) = Karak White, KRK (B) = Karak Black, PESH = Peshawar, CHRD = Charsadda, MRD = Mardan, MKD = Malakand, SWT = Swat, ISLM = Islamabad/Rawalpindi.

Genetic Diversity Analysis Based Upon Simple Sequence Repeat (SSR)

Simple Sequence Repeats (also termed as microsatellites) are small (2-6 bp) DNA motifs, which are very conserved and distributed among the genomes of all the higher eukaryotes. Microsatellites recently have been extensively used for designing primers which are not only highly polymorphic but also species specific (Roder *et al.* 1995; Plashke *et al*. 1995; Pestova *et al*. 2000). During present study wheat chromosome specific SSR primers sets were used mainly to detect and estimate genetic polymorphism based on co-dominant marker system (as contrast to RAPD, which are mainly dominant marker system) and to find out preliminary results that that wheat chromosome specific microsatelllites (SSRs) can work for wild oats. The PCR amplification profile of 10 biotypes of wild oats (*Avena* species) using SSR primer set gdm 86-2B SSR showed various levels of genetic polymorphism among the biotypes at DNA level. Total genomic DNA of 3 biotypes (Dera Ismail Khan White, Mardan and Swat) was not amplified using SSR 86-2B and so the results are not included in analysis. A total of 12 alleles were amplified in 7 biotypes giving an average of 1.2 alleles per biotype. The size of amplified fragments were in the range of < 250bp. Estimates of genetic distances ranged from 0-50 %. Eleven comparisons showed no differences for the loci amplified using gdm 86-2B, while 10 comparisons revealed maximum (50%) genetic dissimilarities.

Ten biotypes of wild oats (*Avena* species) yielded a total of 31 alleles, giving an average of 3.1 alleles per biotype using SSR primer set gdm-87-2D. The size range for the amplified bands was from < 250 - 750bp. The estimated genetic distance among the comparisons ranged from 0-100%. Maximum genetic distance (100%) was observed for 7 comparisons while one comparison (Karak black - Islamabad) showed homozygosity for the loci amplified by using SSR primer gdm 87-2B.

Cluster Analysis

The dissimilarity coefficient matrix of 10 biotypes of wild oats is based on the data of 9 RAPD primers (Figure 2). The biotypes were grouped in two main groups (“A” and “B”). Group A was the largest (comprising of 7 biotypes) and was subdivided into three subgroups “C” (D.I.Khan white,Charshada) “D” (Malakand and Islamabad) and “E” (D.I.Khan Black, Karak White and Karak Black). Group “B” was smallest and comprised of 3 biotypes (Peshawar, Mardan and Swat). Based on the dendrogram analysis, Dera Ismail Khan white biotype (representing semi-arid region) and Swat biotype (representing humid region) were most distantly related to each other. This further supported the hypothesis that cluster analysis can reliably be used for identification of diverse biotypes of weeds which then can be utilized in weed management and breeding programs.

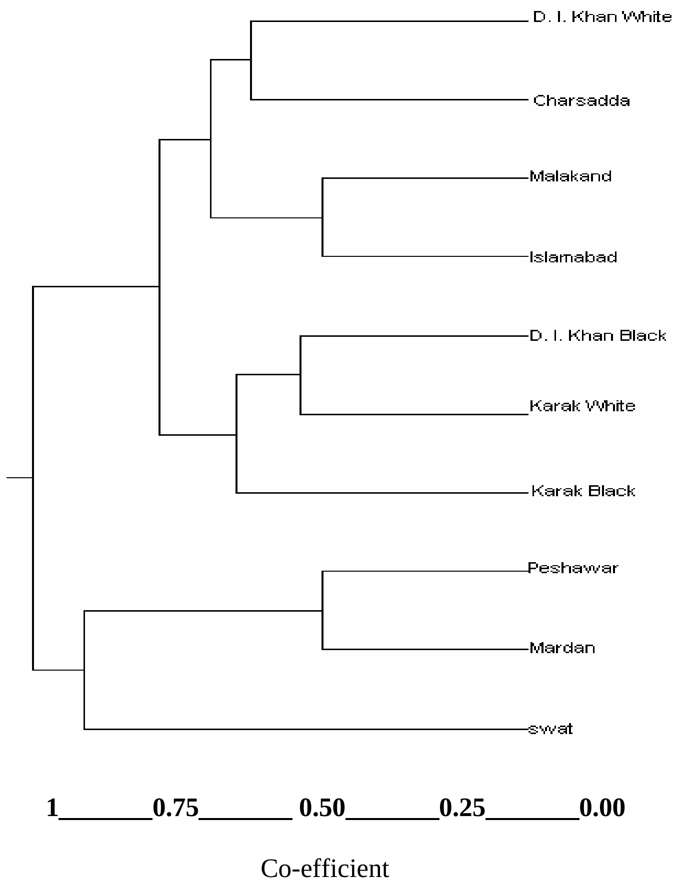


Figure 2. Dendrogram of 9 biotypes of Wild oats constructed by using data obtained from 9 RAPD primers.

Discussion

Average production of wheat in Pakistan (2 tons ha^{-1}) is much less than that of developed countries (6 tonns ha^{-1}). One of the important reasons of lower yield of wheat is weeds. The weeds cause considerable losses to the major crops and cause 25-30% losses in the most important staple food crop, wheat. Wild oat is a serious grassy weed for Rabi crops, and is especially associated with the wheat crop. It is a highly competitive weed with wheat and causes approximately 60 % of total weed losses. Due to crop resemblance with wheat at seedling stage, it is difficult to control manually, therefore, herbicides application becomes inevitable. Since different biotypes of wild oats belonging to different agro-climatic regions are showing different level of herbicide resistance, it is important to recommend herbicide dosages in accordance with diversity of each biotype for each locality, because most important cause of weed control failure is weed biodiversity (Dekker, 1997a).

Genetic diversity is of prime importance for the survival, adaptation to certain agro-climatic conditions, success and improvement of any crop species. Unless there is insufficient genetic diversity in the germplasm, it is practically impossible to increase the yield and other desirable characters of the crop, because selection for the improved genotypes depends on the availability of genetic variability within the breeding material.

There is a need to characterize the indigenous germplasm of wild oats (*Avena* species) using available methods. Phenotypic/morphological characterization (previously used) is easy and cheaper way to characterize the germplasm, but it has its own limitation, because very few morphological characters can be utilized for characterization. Similarly biochemical characterization (Koebner and Shepherd, 1986; Islam and Shepherd, 1992), though successful in many cases but because of limited number of biochemical marker loci (Hart, 1987), could not be used on wider scale. Recent development of molecular genetic techniques, especially RFLP (Paterson *et al*. 1991), Simple Sequence Repeat (Roder *et al*. 1998) and RAPD (Williams *et al*. 1990) have transformed the opportunities of utilizing molecular biology for characterization of crop species. These molecular biology techniques have been used to characterize germplasm of various crop species including barley, wheat and maize , although such work has not been done in Pakistan to characterize weeds of agricultural significance.

For genetic diversity studies, 9 Randomly Amplified Polymorphic DNA (RAPD) primers were used. The RAPD primers produced different levels of genetic polymorphism. For individual RAPD primes, higher level of genetic polymorphism was found in case of RAPD primer GL-A04, where higher levels of genetic variability were observed among different comparisons, while lower levels of genetic polymorphism was found in case of RAPD primer GL-EO5, where lower levels of genetic variability were observed among different comparisons. Over all genetic distances using Randomly Amplified Polymorphic DNA primers ranged from 0 to 100 %. The size of amplified fragments using RAPD primers ranged from approximately 250 to 1000 bp. In general, RAPD primer GLC-07 amplified bands of higher molecular weight (>1000bp), while RAPD primers GLA-03, GL-B19 and GL-E05 amplified fragments of lower molecular size (>250 bp). Average genetic distances were generally in the range of 24.6 to 64% for all RAPD primers used in this study. In comparisons highest average genetic difference (64.14%) was noted for Mardan and Malakand, while lowest average genetic difference (24.67%) was observed for Dera Ismail Khan (white) - Islamabad considering all RAPD primers used in this study.

Randomly Amplified Polymorphic DNA primers (RAPD) were successfully used to study the genetic polymorphism at DNA level and for the estimation of genetic distance among the various biotypes of wild oats. Present study also strengthened earlier reports that RAPDs can be used for estimation of genetic polymorphism in crop improvement programs

(Sun *et al*. 1998; Hamza *et al*. 2004; Heun *et al*. 1994). During recent years, RAPDs are being extensively used in studying genetic variability in a number of crops.

Two sets of Simple Sequence Repeat (SSR) primers were used for dual purpose: I) to find out homology between DNA sequences of wild oat and wheat crop ii) to estimate genetic distances (based on co-dominant marker system). The SSR primer sets produced different levels of genetic polymorphism. For individual SSR primer set, higher level of genetic polymorphism was found in case of SSR primer gdm 87-2D, where higher levels of genetic variability were observed among different comparisons, while lower level of genetic polymorphism was found in case of SSR primer gdm 86-2B, where lower levels of genetic variability were observed among different comparisons. Over all genetic distances ranged from 0 to 100 %. The size of amplified fragments for the two SSR primer sets used ranged from < 250 to 750 bp. Maximum size (750bp) of amplified fragments for SSR primer set was observed in gdm 87-2D, While minimum size (250bp) was observed in gdm 86-2B . Average genetic distances are generally in the range of 30 to 60% for the two SSR primer sets used in the present study. In comparisons highest average genetic difference (100%) was noted for six comparisons, while lowest average genetic difference (0% = no difference) was observed for Karak (black) and Islamabad considering the two SSR primer sets used in present study. Satchel *et al*. (2000) used microsatellites to differentiate different biotypes/varieties. Further, using two wheat chromosome specific SSR markers, homology was found at DNA sequences level (Table 3) between wild oats and wheat, therefore, these two SSR markers (gdm 86-2B and gdm 87-2D) are suggested for investigating genetic diversity in wild oats.

Overall range of scorable bands was from < 250 bp to > 1000 bp based on the data obtained 9 RAPD primers and two SSR primer sets used in present study, similarly the range of genetic dissimilarity (estimated as genetic distances) was from 0 to 100 % indicating that these biotypes were genetically very diverse and possess a high amount of polymorphism. It also indicates the potential of PCR based techniques for estimating genetic diversity among various biotypes (genotypes) of wild oats.

Alternately, if wild oat is used as a fodder or feeder crop, then the integration of present molecular output data will enhance breeding efficiency and add to the strength of marker-assisted selection via user-friendly techniques. In view of the extensive information about the high genetic diversity residing in biotypes of wild oats, it is suggested that mission oriented breeding programs with the help of DNA fingerprinting technology will be helpful to utilize these biotypes to produce distinct cultivars/genotypes by crossing them with one another. Considering dendrogram analysis, D.I.Khan White and Swat biotype are recommended to be crossed with one another in order to improve the genetic base of these biotypes.

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INFLUENCE OF WEED MORPHOLOGY AND CHEMICAL COMPOSITION ON ANTIXENOSIS AND ANTIBIOSIS MECHANISM AGAINST *Cnaphalocrocis medinalis* IN RICE ECOSYSTEM

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Abstract: Weeds in rice fields may affect crop production by acting as reservoirs for rice pests survival and multiplication while they display several physical and chemical defensive adaptations against insect attack. Antibiosis resistance involves biophysical and biochemical plant defenses, as well as nutritional factors. This resistance mechanism affects the growth and development of the insect. It was found that biophysical factors like morphological features of weed leaves and trichomes exert antixenosis mechanism, where weeds like *Sphenoclea zeylanica* and *Cyperus rotundus* were found to be highly preferred hosts of *Cnaphalocrocis medinalis* as against *Leersia hexandra* and *Echinochloa crus-galli*. Similarly qualitative and quantitative differences in biochemical constituents resulting in antibiosis mechanism, which was confirmed by estimation of various biochemical constituents in 13 selected weeds. Experiments were carried out in a Randomized Complete Block Design with five replicates. It was estimated that the most preferred hosts of *C. medinalis*, such as *L. hexandra, E. crusgalli* and *E. indica* showed higher content of nitrogen, sugars, and amino acids, but lesser contents of potash, silica and phenols, whereas, a reverse trend was noted with unsuitable weeds. The various weed hosts of *C. medinalis* studied, can be arranged from the least preferred ones to highly preferred ones as: *S. zeylanica* > *C. rotundus*> *C. benghalensis* > *D. aegyptium* > *P. conjugatum* > *P. repens* > *D. ciliaris* > *B. mutica* > *E. colona* > *E. indica* > *L. chinensis* > *E. crusgalli* > *L. hexandra.*

Key words: Rice leaf folder, antixenosis, antibiosis

Introduction

Weeds are important alternate hosts for many rice pests. Very few studies have been conducted on important of weeds as alternate hosts for rice pests in general and leaf folder, *Cnaphalocrocis medinalis* in particular, which is a serious pest in many southern states of India.

Weeds present in the rice fields affect the crop production directly by competing for abiotic factors required for growth, and indirectly by acting as reservoirs for pest incidence and multiplication. In the case of rice leaf folder, a number of weeds act as alternate hosts which would help the insects to hide over in the lean seasons. A new dimension of insect feeding behaviour known as ‘dietary self selection by insects’ deals with the insect’s interaction with their host plants and near by weeds. The host plant resistance interactions between rice leaf folder and weeds mainly involves antixenosis and antibiosis mechanisms are involved which depends upon biophysical and biochemical factors.

Materials and Methods

Bionomics of *C. medinalis* was studied with the selected weed host plants (Table 1) with a view to study the extent of preference shown by the leaf folder in the field and for off-season carryover. Different types of grasses and weeds present in the rice fields of Annamalai Nagar were collected and identified (Reissig *et al.* 1985). They were graded and finally 13 weeds were selected for the study on the basis of their occurrence and infestation by leaf folders. Young weed plants were transplanted directly from the field into the clay pots were used in the experiment and larvae were released on the above weed species and data on their bionomics were gathered to study the extent of preference shown by the leaf folder in the field and an off season carryover. In the antixenosis studies thickness of weed leaves and their trichome density were studied, where as in antibiosis studies, biochemical constituents such as nitrogen, potash, total sugars, and total phenols were estimated. Each treatment was replicated five times and all the experiments were carried out in Randomized Complete Block Design.

Results and Discussion

Orientation preference of the third instar of *C. medinalis* larvae on selected weed hosts revealed that minimum number of larvae (1.20) preferred *S. zeylanica* (Table 1) followed by 2.10 in *C. rotundus* and the maximum number of larvae 16.80, 15.40 were found on *L. hexandra* and *E. crus-galli* respectively. Fourth instar *C. medinalis* larvae showed positive feeding orientation to few weed plants. The minimum leaf area consumed was 0.133 cm^2 in the case of *S. zeylanica* and the highest was recorded on *L. hexandra* (1.527 cm^2).

Table 1. Orientation preference and extent of leaf feeding by *C.medinalis* on selected weed hosts

Weed hosts	Orientational preference	Leaf area consumed (sq. cm)
Brachiaria mutica	6.1 c	0.387 g
Commelina benghalensis	3.0 def	0.663 f
Cyperus rotundus	2.1 ef	0.2 i
Digitaria ciliaris	5.1 cd	1.197 de
Dactyloctenium aegyptium	4.4 cde	1.28 c
Echinochloa colona	10.2 b	1.25 cd
Echinochloa crus-galli	15.4 a	1.303 c
Eleusine indica	11.63 b	1.46 b
Leptochloa chinensis	14.6 a	1.15 e
Leersia hexandra	16.8 a	1.527 a
Panicum repens	3.4 def	0.32 h
Paspalum conjugatum	4.2 cde	1.16 e
Sphenoclea zeylanica	1.2 f	0.133 j
C.D. (p=0.05)	0.481	0.076

Values presented are means of five replicates. Within a column, means followed by the same letter are not significantly different (p=0.05).

The ovipositional preference of the leaf folder on different weed hosts, studied in free-choice test and no-choice test, showed a significant difference among weeds (Table 2). A significant difference ($p<0.05$) in the larval survival was found among the host weeds. The highest percentage of larval survival was observed on *L. hexandra* (71%) and the lowest was observed on *C. rotundus* (30%). The lowest larval period was observed on *L. hexandra* (22%) and *L. chinensis* (23%). However, a longer larval period was recorded in *C. rotundus* (33 days) and *S. zeylanica* (31 days).

A significant difference ($p<0.05$) in pupation of *C. medinalis* on various weed plants was found to vary from 18.9% in *S. zeylanica* to 70.4% in *L. hexandra*. *Cnaphalocrocis medinalis* reared on *L. hexandra* was significantly ($p<0.05$) than those reared on *E. crus-galli* (18.2 mg) and *E. colona* (17.6 mg), whereas it was just reverse on less preferred hosts such as *C. rotundus* (9.3 mg) and *S. zeylanica* (9.9 mg).

The suitability of different weed plants as hosts to *C. medinalis* revealed a significant variation in growth indices. Growth index on *L. hexandra* (3.2) was the maximum and it was the minimum on *S. zeylanica* (0.6).

Table 2. Bionomics of *C.medinalis* on selected weed hosts

Host plants	Average number of eggs laid	Larval survival (%) (S)	Pupation (%) (T)	Growth Index (S/T)
Brachiaria mutica	37 ef	44.39 f (41.78)	43.52 e (41.28)	1.6 e
Commelina benghalensis	24 hi	36.71 i (37.29)	29.04 i (32.61)	1.03 g
Cyperus rotundus	18 i	30.25 j (33.36)	23.38 j (28.91)	0.7 h
Digitaria ciliaris	33 efg	43.16 f (41.07)	42.25 e (40.54)	1.57 e
Dactyloctenium aegyptium	28 gh	38.42 h (38.30)	33.38 h (35.29)	1.33 f
Echinochloa colona	64 d	54.63 d (47.66)	51.04 d (45.59)	2.08 d
Echinochloa crus-galli	92 b	62.35 b (52.15)	61.17 b (51.46)	2.68 b
Eleusine indica	81 c	59.34 c (50.39)	57.36 c (49.23)	2.39 c
Leptochloa chinensis	105 a	52.05 e (46.18)	59.59 c (50.53)	2.58 c
Leersia hexandra	102 a	71.24 a (57.60)	70.40 a (57.06)	3.20 a
Panicum repens	31 fg	40.62 g (39.59)	38.51 f (38.36)	1.47 f
Paspalum conjugatum	39 e	38.47 h (38.32)	36.56 g (37.20)	1.53 e
Sphenoclea zeylanica	21 i	28.71 k (32.39)	18.90 k (25.77)	0.6 h
C.D. (p=0.05)		1.225	1.187	0.188

Values presented are means of five replications. Within a column, means followed by the same letter are not significantly different (p=0.05).

Studies on orientation and setting by leaf folder on selected weed host *C. medinalis* indicate that maximum number of larvae gathered and consumed larger areas of leaves on *L. hexandra* and the minimum was recorded on *C. rotundus*. This may be due to the presence of attractants and repellents which influenced orientation, settling and feeding behaviour of *C. medinalis* on different weed hosts.

The number of eggs laid on various weeds showed that in no choice tests, the least number of eggs was laid on *C. rotundus* followed by *S. zeylanica*, whereas the highest number of eggs was laid on *L. chinensis*. A similar trend was observed in free choice tests. Joshi *et al.* (1987) and Khan (1996) reported a similar difference in oviposition behaviour of *C. medinalis* on rice varieties and weeds.

The leaf area consumed by *C. medinalis* larvae on weed plants differed significantly. It was the maximum on *E. crus-galli* and the minimum on *S. zeylancia*. Furthermore, a lower fecundity, larval survival, pupal weight and growth indices on certain weed plants confirmed their status as less suitable. A similar trend was observed by Joshi *et al.* (1987) and Khan *et al.* (1996).

In the antixenosis studies, the biophysical factors such as weed leaves showed that *C. rotundus* have the maximum thickness (38.2 μ) (Table 3) followed by *B. mutica* (37.7 μ) whereas the minimum was in the case of *L. hexandra* (15.04μ). A significant negative correlation was observed between leaf thickness and larval survival of the leaf folder.

The length, breadth and density of trichome on various weeds were significantly differed. Maximum of 167.00 trichomes per sp.cm were present in the case of *E.colona* (167 cm^2) and it was on part with *B.mutica* (165 cm^2). Minimum trichomes were seen in *L.chinensis* (7 cm^2) while trichomes were absent on *C. rotundus* and *S. zeylanica*. The trichome density showed a negative correlation with larval survival.

In the antibiosis studies, analysis of certain biochemical constituents such as N, K, sugars, phenols and silica were compared with 13 weed hosts (Table 4). The highest content of nitrogen was found in *E. indica* (2.89 mg/g) and the minimum in *P. repens* (1.25 mg/g). The potassium content was the maximum in *E. colona* (1.9 mg/g) and the lowest in *E. crusgall* (0.77 mg/g). The highest content of total sugars was found in *L. lexandra* (9.72 mg/g) and the minimum in *B. mutica* (6.13 mg/g). The total phenols showed a significant difference ($p<0.05$) in content among the weed plants. The maximum total phenol content of (0.689

mg/g) was recorded in *P. conjugation* and the minimum was recorded in *D. aegyptium* (13.97 mg/g). The analytical data revealed a significant negative correlation with leaf area consumed, larval survival fecundity and the growth index. Similar observations were noted by Khan *et al.* (1996).

Table 3. Antixenosis studies on leaf parameter and trichomes on selected weeds

Host plants	Leaf length (cm)	Leaf thickness (μ)	Trichome length (μ)	Trichome density per cm²
Brachiaria mutica	24.75 e	37.7 a	276 c	165 a
Commelina benghalensis	5.24 k	31.5 c	230 d	0.003 i
Cyperus rotundus	20.02 f	38.2 a	0.0 i	96 d
Digitaria ciliaris	14.01 h	27.5 f	45 g	45 f
Dactyloctenium aegyptium	15.03 gh	38.4 a	295 b	167 a
Echinochloa colona	20.3 f	28.5 e	90 e	115 c
Echinochloa crus-galli	34.5 a	18.2 j	30 h	139 b
Eleusine indica	26.2 d	30.5 d	43.3 g	7 i
Leptochloa chinensis	12.5 i	22.5 i	28 c	31 g
Leersia hexandra	15.8 g	15.0 h	40 g	68 e
Panicum repens	30.4 b	26.1 g	80 f	44 f
Paspalum conjugatum	28.3 c	25.0 k	312 a	0.003 i
Sphenoclea zeylanica	6.52 a	36.2 b	0.0 i	9.240
C.D. (p=0.05)	0.144	0.172	0.112	

Values presented are means of five replications. Within a column, means followed by the same letter are not significantly different (p=0.05).

Table 4. Chemical constituents of selected weed hosts to study antibiosis mechanism of selected weed hosts

Host plants	Nitrogen (mg/g)	Potassium (mg/g)	Total sugars (mg/g)	Total phenols (mg/g)	Silica (mg/g)
Brachiaria mutica	1.73 e	1.17 f	6.13 j	0.689 a	11.59 gh
Commelina benghalensis	2.63 b	1.91 a	9.45 b	0.220 i	18.73 a
Cyperus rotundus	1.99 d	1.24 ef	6.84 g	0.511 i	13.03 de
Digitaria ciliaris	2.28 c	1.45 cd	7.79 f	0.216 j	10.95 h
Dactyloctenium aegyptium	1.68 e	1.36 de	8.15 d	0.205 j	13.97 bc
Echinochloa colona	2.77 ab	1.9 a	8.53 c	0.203 j	13.51 cd
Echinochloa crus-galli	2.41 c	0.77 g	9.70 a	0.450 d	7.55 i
Eleusine indica	2.89 a	0.79 g	6.27 i	0.310 g	12.51 ef
Leptochloa chinensis	2.34 c	1.56 bc	6.42 h	0.280 h	16.90 b
Leersia hexandra	2.72 ab	0.87 g	9.72 a	0.330 f	12.06 fg
Panicum repens	1.25 f	1.39 de	7.95 e	0.523 c	4.46 j
Paspalum conjugatum	2.78 ab	1.2 ef	6.93 g	0.571 b	11.18 h
Sphenoclea zeylanica	1.42 f	1.65 b	6.38 hi	0.430 e	12.83 e
C.D. (p=0.05)	0.800	0.633	0.316	0.193	0.786

Values presented are means of five replications. Within a column, means followed by the same letter are not significantly different (p=0.05).

The various weed hosts studies can be arranged from the least preferred host of *Cnaphalocrocis medinalis* to highly preferred ones as: *S. zeylancia* > *C. rotundus* > *C. benghalensis* > *D. aegyptium* > *P. conjugatum* > *P. repens* > *D. ciliaris* > *B. mutica* > *E. colona* > *E. indica* > *L. chinensis* > *E. crus-galli* > *L. hexandra*.

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HYDROGEL® FOR MANAGEMENT OF AQUATIC WEEDS: POSSIBILITIES AND CONSTRAINTS

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Abstract: A number of submerged aquatic weeds have colonized Australian and New Zealand waterways, assisted mainly by human activities. Three of these species- Lagarosiphon [*Lagarosiphon major* (Ridley) Moss], Egeria (*Egeria densa* Planch.) and Hornwort (*Ceratophyllum demersum* L.) prefer clear water, where they form dense stands, out-competing native vegetation and causing problems for recreation. Eradication of these species is considered impossible. The problem in controlling these plants relates to their mode of spread; the smallest viable vegetative fragment can re-establish a population. Left uncontained, these aquatic weeds are likely to fully colonize all available habitats, within a short period of time. Weed control techniques used for managing submerged aquatic weeds, such as Lagarosiphon, Egeria and Hornwort, in Australia and New Zealand's waterways are discussed, along with their strengths, weaknesses and costs. Most non-chemical methods are not cost effective, and are of limited value in controlling these species. In contrast, aquatic herbicides offer more promise for cost-effective control, although eradication is still not an option, due to inadequate plant exposure and uptake. A new technique for applying the aquatic herbicide, diquat, for the control of submerged aquatics has been developed in New Zealand. This method involves the use of guar gum, and formulating a Diquat gel (Hydrogel®), which can then be applied to water. Several case studies are discussed, in which effective control of Lagarosiphon, Egeria and Hornwort has been cost-effectively achieved in New Zealand and Australia without undue environmental impacts. Results indicate that there is further scope for significant expansion of using Hydrogel® to control submerged aquatic weeds.

Key words**:** Aquatic weeds, diquat, hydrogel

Introduction

A number of submerged exotic aquatic weed species have colonized Australia and New Zealand's waterways, assisted mainly by human activities. The main species are: Hydrilla (*Hydrilla verticillata* (L. f.) Royle), Hornwort (*Ceratophyllum demersum* L.), Lagarosiphon (*Lagarosiphon major* (Ridley) Moss, Pond weed (*Potamogeton crispus* L.), Elodea (*Elodea canadensis* Michx.), Egeria (*Egeria densa* Planch.) and Cabomba (*Cabomba caroliniana* Gray). Alligator Weed (*Alternanthera philoxeroides* (Mart.) Griseb.) is also well established in Eastern Australia and in North Island of New Zealand, and has the potential to spread further. There have been considerable efforts in both countries to keep waterways weed-free. Over the past 45 years many techniques have been tried for aquatic weed eradication, or to manage the adverse effects of these weeds on aesthetic, recreational and economic values of waterways. The objectives of this paper are to: (a) Review how aquatic weeds have been managed in Australian and New Zealand's waterways, and (b) Discuss the potential of a relatively new technique of applying aquatic herbicides for aquatic weed control.

Review of Aquatic Weed Management Options

We carried out an extensive review of aquatic plant management in both countries gathering information on methods, up-to-date costing and logistics issues. Information was collected from aquatic weed control contractors, scientists and field managers, as well as from published literature. Table 1 is a summary of non-chemical methods commonly used, costs, applicability and disadvantages.

Aquatic Herbicides

Aquatic weeds can be controlled effectively and cheaply by aquatic registered herbicides, when compared to mechanical methods, but the time and method of herbicide application varies with the type of weed and the habitat in which they are to be controlled. The herbicides most widely used in Australia and New Zealand in underwater treatments are diquat and endothal. Both have sound environmental profiles and concentrations required for control of aquatic weeds, they are relatively safe for humans, fish and other aquatic fauna at. They are not persistent chemicals. However, when applied correctly, they have a high degree of phytotoxicity to kill aquatic weeds fast and rapidly degrade in the water after the action on weeds. Technology should be available for their application in static or flowing water systems.

Diquat dibromide (Reglone® and Reward®) has been used for over 40 years in New Zealand and Australia for the control of submerged species. Diquat does little harm to non-nuisance native species, such as charophytes, and native potamogetons and milfoils (Wells and Clayton 2005). Endothal (Aquathol® and Aquathol Super K®) has recently been registered for use in New Zealand, but significant restrictions remain on its use. Endothal is superior to Diquat for controlling *Hydrilla* (Hofstra and Clayton, 2001, Hofstra *et al.* (2001). Biactive glyphosate, with high degree of aquatic safety, is also widely used for controlling a variety of emergent aquatic weeds, including Alligator Weed and Milfoil, but is not directly discharged into water.

The mode of delivery of herbicides is very important for the effectiveness of aquatic weed control. Various gel adjuvants have been mixed with Diquat, such as alginate gum (Torpedo®), guar gum (Aquagel®) and methocel (hydroxypropyl methylcellulose, marketed as Depth Charge®). All are formulated to mix with Diquat, and applied at 60 -80 l ha $^{-1}$. When applied as a steady stream, the mixtures sink and attach onto submerged weeds and Diquat is released into surrounding water, causing desiccation of aquatic weeds. Aquathol Super K contains an additive, which performs a similar function to the Diquat adjuvants. The most widely used gel adjuvant is Aquagel, marketed as Hydrogel® in Australia.

Hydrogel is made of guar gum, a non-toxic polysaccharide starch, which can be mixed on site to any desired viscosity (LINZ, 2003). It is superior to alginate gum, as it retains a consistent viscosity at any temperature. If viscosity varies with temperature, the delivery equipment requires recalibration throughout the day. The relatively heavy nature of the gel carrier prevents diquat from being dispersed, as it sinks in the water column and lands on target foliage. Diquat directly acts on the plants with its toxic action, but does not leave a residue in the sediments; nor is it bio-accumulated in animal tissue (LINZ, 2005). The starchy polymer is non-toxic to the environment and is dispersed in water. Hydrogel® can be applied into water from a knapsack, gun and hose, boat-mounted boom or helicopter-mounted boom. Aerial spray drift is reduced to near zero; and water dispersal and drift is also significantly reduced.

Several case studies are presented from New Zealand and Australia, which demonstrate the excellent possibilities of controlling submerged aquatic weeds with Hydrogel®.

Table 1. Non-chemical Aquatic plant management methods commonly used in Australia and New Zealand

Method	Application	Main disadvantages	Cost/ha ($ NZ)
Hand Weeding	Useful for controlling small (<1 m^2), localised, sporadic and patchy infestations	Labour intensive; Not an option for larger infestations	$ 7000-10,000
Mechanical digger	Artificial canals, shallow canals, lake shoreline areas	Loss of benthic fauna and fish; high turbidity, anoxia; widen/deepen drains; spreading weeds	$ 1000-3500 ($ 2500-5000)
Roto-tiller	Can uproot weeds in water depths between 1.5-4 m; Deeper tilling provides longer control (1-2 years vs. 6 months).	Lakebed obstacles prevent effective use. Regrowth can be increased; roto-tilling is like a plough, creating more habitats for rooted aquatic weeds.	$ 2000 (shallow) $ 5000 (deeper, up to 5 cm)
Mechanical weed cutter/harvester	Can target a specified area and cut to a nominated depth; Costs depend on density of weeds and distance to disposal site.	Quick re-growth could occur; requires repeated cutting (2-3 times in a growing season); potential to spread weeds, as fragments inevitably escape.	$ 2,000 - 4,000
Suction dredging	Use of suction pump to uproot aquatic weeds and collection in a mesh bag; Can give effective control up to 3 years.	Re-establishment can be as short as two months for hornwort. It is also ineffective in hard-bottomed or rocky substrates.	$ 15000- 20000
Nutrient control	Nutrient reduction through riparian buffers or by nutrient removal by flocculation using products like Phoslock®	Costly; depending on the amount of phosphate to be removed and the nature of the lakebed substrate.	$ 6000 – 10000 (Phoslock)
Shading and bottom lining	Dyes (Aqua shade®, Nigrosine) to suppress light and plant growth; Polyethylene, PVC, or fibreglass covers, as bottom linings.	Use is limited to smaller water bodies; adverse long-term impacts are largely unknown, although unlikely to be high.	$ 5000 - 15000
Water level manipulation	Lake draw-down is widely practiced in lakes with controlled outlets; often in hydro-power generating systems.	Re-growth can be rapid when lake refills; also, high cost (through lost hydro-generation potential) and adverse impacts (erosion, slumping).	Varies
Chinese Grass Carp *Ctynopharyngodon idella* (Valenc.)	Widely used in NZ, but not in Australia; feeds non-selectively on a range of submerged or floating soft plant tissues. Unlikely to breed in NZ waterways.	Unknown impact on some native aquatics; limited success in larger lakes, because of fish losses through escape and predation. In Lake Hood, ≈ 30 fish/ha provided required weed control.	$ 750 ($25 per fish)

Case study 1: Hornwort control in Moutere Stream, Nelson, South Island, NZ
Hornwort was not previously known in the South Island, and the target was to eradicate it from the infested location. Aquagel treatments to control Hornwort were first made in March 2002 (LINZ, 2005). Aquagel was applied in strips (60 cm wide), over about 800 m of stream; 195 l of Hydrogel covered 0.7 ha. The cost of this treatment was NZ $ 4500. After 6 weeks, all Hornwort had collapsed, and was no longer noticeable in the stream. Spot treatments were conducted 12 months later. Monitoring of the stream in November 2003 and February 2004 found no Hornwort in the treated area.

Case Study 2: Lake Benmore, South Island, NZ
Lagarosiphon invaded Lake Benmore in South Island in March 2002 and large patches were found over a 100 ha area of lakebed from 1-4 m depth. While eradication was not considered possible, suppression of the weed was necessary to prevent downstream spread throughout the catchment (LINZ, 2003). Aquagel was applied from a helicopter over the infested 100 ha site in March 2003. Helicopter spraying is cost-effective and accurate treatments can be made to specific areas of a large lake in a very short time. The cost of treating the Lake was NZ $ 1425/ha. Monitoring found that the spraying was highly effective. While immediate eradication of Lagarosiphon from the Lake is unlikely, the rate of spread has slowed, largely as a result of Aquagel treatments.

Case Study 3: Hornwort and Cabomba infestations, Botany Wetlands, Sydney
Botany Wetlands (Longitude 151°10’-151°15’; Latitude 33°55’-33°58’), in Sydney, are a series of freshwater ponds. After a sustained 5-year programme of removing European Carp (*Cyprinus carpio*), a dense Hornwort infestation covered the largest of the ponds-Pond 5. Carp, as bottom-feeders, kept submerged aquatic plant growth in check, but a large reduction in adult Carp coincided with the Hornwort infestation. A single Hydrogel® treatment, over a 600 m^2 infested area (≈ 4 l at a cost of Aus $ 250) was effective in achieving clear water within 4 weeks.

In other areas of Botany Wetlands, trials are in progress, testing Hydrogel® effectiveness on Cabomba. Initial results are that multiple treatments have reduced Cabomba in trial plots by about 50%. Optimisation of a treatment regime is envisaged in the near future.

Case Study 4: Egeria infestation, Sutherland Shire, NSW
Hydrogel® application was trialled to eradicate a 500 m^2 Egeria infestation in Sutherland Shire, NSW, Australia. One treatment of 3 l Hydrogel® completely eradicated the infestation within 2 months (Plate 1). The cost of this treatment was Aus $ 275.

Case Study 5: Egeria infestation, Georges River, Liverpool, NSW
Similar Hydrogel® application trails were conducted at an Egeria infested reach of the Georges River, Sydney. The area treated in January 2007 was 2500 m^2. One treatment of 15 l Hydrogel® completely eradicated the infestation within 2 months (Plate 2). The cost of this treatment was Aus $ 600.

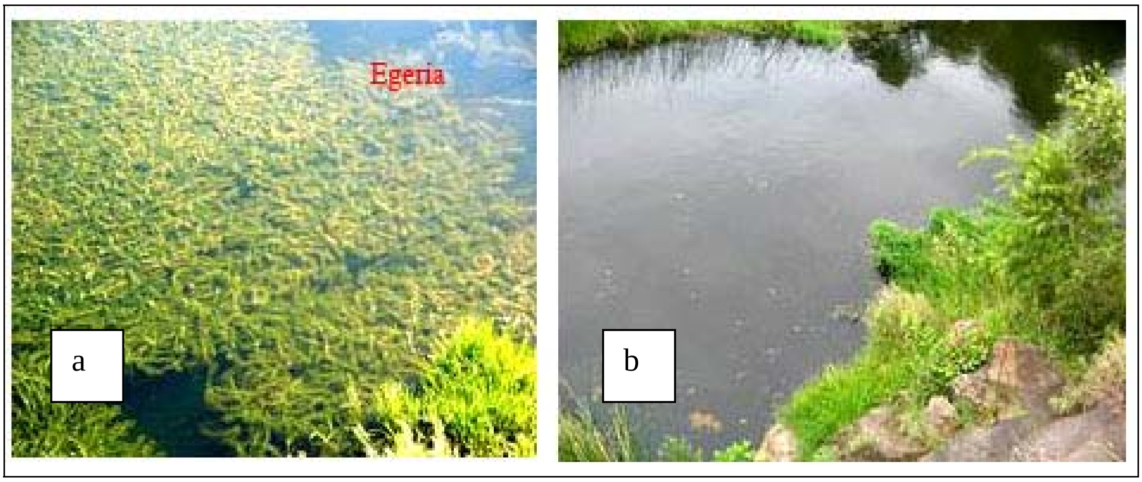

Plate 1. (a) Egeria infestation in Sutherland Shire (b) Control achieved by Hydrogel®.

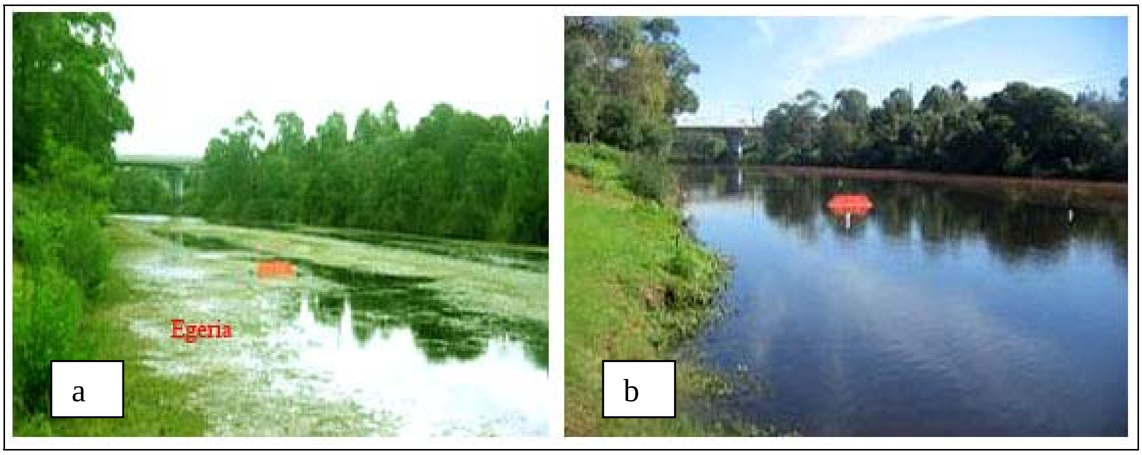

Plate 2. (a) Egeria infestation in a section of Georges River, Sydney, NSW; (b) Control achieved by Hydrogel® 2 months later.

Discussion

In our view, there is a significant body of evidence from New Zealand and increasing evidence from Australia that the effectiveness of aquatic herbicides can be improved, to suppress extensive areas of critical aquatic weeds infestations quickly at a relatively low cost. Use of smart delivery systems, such as Hydrogel®, allows for this, particularly to accurately deliver the required dosage over a treatment area, without wasting chemicals. Hydrogel® treatments make the control significantly more cost-effective than control by other methods. Additional advantages are that Hydrogel® treatments do not generate unsightly piles of Lagarosiphon, Egeria or Hornwort on shorelines and applications require a much smaller suitable weather window, because of the speed of application and action, and the result is often long lasting. The differential response in submerged plants (*i.e.* reduced effectiveness on Cabomba) could be related to less retention of Hydrogel® on the fan-like Cabomba leaves.

The use of diquat/gel (Hydrogel®) for aquatic weed control is now widespread throughout New Zealand's waterways. Its social acceptance is rapidly improving, as evidenced by most territorial authorities allowing its use as a permitted activity (*i.e*. no Council discharge permit required). In Australia, experiments are in progress, still under a trial permit, and success is spectacular in some cases. Although herbicides are the most cost-effective method of aquatic weed control, there is an understandable general community aversion for using chemicals in water. This aversion can often prevent the use of herbicides over large areas. In this situation, Hydrogel® is useful because it allows less number of treatments and specific targeting, reducing herbicide loads and offsite drift. The development of new techniques for aquatic weed control needs to continue, despite the relatively small market in this field. The potential environmental impacts and monetary costs of many of the other control methods mean that more attention is needed for aquatic herbicides and smart delivery systems to achieve superior results.

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EFFECT OF PRETILACHLOR + PYRIBENZOXIM 320 EC ON WEED CONTROL IN WET-SEEDED RICE IN SRI LANKA

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Abstract: A field experiment was conducted at the Rice Research and Development Institute, Batalagoda, Sri Lanka during the major cultivation season (October to February) in 2005 to evaluate the weed control efficacy and crop selectivity of the co-formulation, pretilachlor + pyribenzoxim (300g a.i/ha + 20 g a.i/ha EC) at three rates 1, 1.25 and 1.5 l/ha. These treatments were compared with the commonly used herbicides such as bispyribac sodium 100 SC at 100 ml/ha; and the combination of herbicide Propanil 36% EC (7.5 l/ha) followed by (*fb*) MCPA 40% SL (2.5 l/ha), un-weeded and hand weeded controls. Pretilachlor + pyribenzoxim effectively controlled grasses, broad leaves and sedges when applied at the rates of 1.25 l/ha and 1.5 l/ha at 10 days after sowing (DAS). The growth characters and harvested yield reflected and corresponded to the level of weed control of each herbicide treatment as the yield from each treatment was significantly higher ($p<0.05$) than the unwedded treatment. No detrimental effects were observed on growth and yield of rice in the herbicide treated plots. The results indicated that pretilachlor + pyribenzoxim at the rate of 1.25 - 1.5 l/ha effectively controlled of wide range of weeds with excellent selectivity in broadcasted rice in Sri Lanka.

Key words: Co-formulation, pretilachlor + pyribenzoxim, rice, weed control

Introduction

Broadcasting is the popular method of establishment of rice (93%) in Sri Lanka. Weeds are the major biotic stress in rice cultivation (Herath Banda *et al.*, 1998, Abeysekara, 1999). As a result, usage of herbicide in rice has increased over the past 4-5 decades (Amarasinghe and Marambe, 1998; Abeysekara, 1999). The combination of propanil followed by (*fb*) MCPA, and bispyribac sodium are the widely used herbicides in Sri Lanka under lowland rice cultivation to control weeds. Continuous applications of these herbicides have led to the emergence of resistant biotypes of *Echinochloa crus-galli* in some areas of the country (Abeysekara, 1999; Marambe and Amarasinghe, 2002). Hence, introduction of herbicides with new mode of action is a timely need. The, co-formulation, pretilachlor + pyribenzoxim is an early post emergent herbicide having two modes of action. Preliminary studies in other countries have indicated that the mixture it is effective against a wide range of weeds with a good crop safety in rice. A study was thus conducted with a view to make a recommendation in rice in Sri Lanka, by evaluating the weed control efficacy of the herbicide mixture and to ascertain the phytotoxicity on growth and yield of rice.

Materials and Methods

The experiment was carried out at the Rice Research and Development Institute (RRDI), Batalagoda, Sri Lanka, during the major rice growing season (October-February) 2005. The soil type was red yellow podzolic with a PH of 5.9. Rice (*Oryza sativa* L) var. Bg 300, a 90-day old rice variety, was broadcasted in plots at the size of 3 m x 6 m at a rate of 100 kg/ha. The recommended doses of nutrients were supplied to the plots. The experiment was conducted in a Randomized Complete Block Design (RCBD) with three replicates.

The herbicide co-formulation pretilachlor + pyribenzoxim (300 g a.i/l + 20 g a.i/l; Solito® 320 EC) was tested at three rates namely, (a) 1 l/ha, (b) 1.25 l/ha and (c) 1.5 l/ha, applied at 10 days after sowing (DAS) in rice. Commonly used herbicides; propanil (36% EC at 7.5 l/ha) at 7 DAS followed by MCPA (40% SL at 2.5 l/ha) at 21 DAS and bispyribac sodium (100 SC at 100 ml/ha) at 7 DAS were used as standard treatments together with an un-weeded and hand-weeded plots. The field plots were maintained with saturated moisture at the time of herbicide application and irrigated 3 days after herbicide application. All the other cultural practices were carried out according to the recommendation of the Department of Agriculture (2000).

The weed density and dry weight of grasses, broadleaves and sedges were recorded separately at 4 and 6 WAS. In addition the visual phytotoxicity and growth characters of rice plants were recorded. At maturity, the yield components and yield were measured.

Results and Discussion

Common weeds in the experimental site

The dominant grass weeds (43%) in the experimental plots were *Echinochloa crus-galli*, *Leptochloa chinensis*, *Ischaemum rugosum*, *Paspalum distichum* and *Isachne globosa*. Among the broad-leaved weeds (36.2%) *Monochoria vaginalis*, *Murdannia nudiflora* and *Ludwigia* spp were the dominant species, while *Cyperus iria*, *C. difformis*, *Fimbristylis miliacea* and *Scripus supinus* were the dominant sedges (20.8%).

Germination and plant count of rice

At 2 WAS, the rice seedling count did not show any significant difference among the treatments (data not shown) indicating that a uniform rice stand count was maintained at the rice field, before spraying herbicides. A similar trend was observed from 4 and 6 WAS in same treatments. However, a significantly lower ($p<0.05$) rice plant count was observed in the un-weeded plots at 4 and 6 WAS, which is probably due to he due to the presence of higher densities of weeds.

Weed control efficacy

A significantly different ($p<0.05$) weed count and weight were observed among the treatments (Table 1).

Table1: Effect of pretilachlor + pyribenzoxim on weed count and dry weight of 6WAS

Treatment	Weed count (No/m^2)			Weed weight (g/m^2)		
	Grasses	Broad leaves	sedges	Grasses	broadle aves	Sedges
Pretilachlor + Pyribenzoxim (1 l/ha)	57.0 b	7.3 a	2.0 c	20.6 b	5.4 b	3.1 b
Pretilachlor + Pyribenzoxim (1.25 l/ha)	29.0 bc	3.6 b	4.6 bc	17.6 b	1.8 c	4.2 b
Pretilachlor + Pyribenzoxim (1.5 l/ha)	21.3 bc	1.0 b	4.6 bc	14.6 b	0.6 c	2.0 b
Bispyribac sodium 100 SC	58.6 b	1.0 b	1.0 c	27.2 b	1.0 c	1.6 b
3.4 DPA *fb* MCPA	49.0 b	14.6 a	6.3 bc	23.6 b	3.4 bc	2.5 b
No weeding	372.3 a	6.0 a	27.6 a	128.9 a	20.7 a	16.2 a
Hand weeding	-	-	-	-	-	-

Within a column, means followed by the same letter are not significantly different by the Duncan's New Multiple Range Test (p=0.05)

The highest grass, broadleaf and sedge count and weight (at 4 and 6WAS) were found in the un-weeded treatment. All the herbicide treatments showed an effective weed control. The weed count/weight among the herbicide treatments were similar, but a lower weed count and weight were observed in the plots treated with the pretilachlor + pyribenzoxim 320 EC at the rate of 1.25 and 1.5 l/ha. Poor control of *Paspalum spp*. and *Isachne globosa* were observed with all the herbicide treated plots. In general, the results showed that the co-formulation of the herbicides effectively controlled the dominant grasses, sedges and broadleaves in wet-seeded rice fields when applied at the rate of 1.25 -1.5 l/ha at 10 DAS (Table 2).

Table.2: Effect of different herbicide treatment on specific weeds at 6 WAS in wet seeded rice

Treatment	% weed control efficacy									
	EC	LC	IR	PD	MV	LO	MN	CD	CI	FM
Pretilachlor + Pyribenzoxim (1 l/ha)	80	73	65	58	40	84	58	95	88	92
Pretilachlor + Pyribenzoxim (1.25 l/ha)	100	94	70	64	71	89	60	100	98	86
Pretilachlor + Pyribenzoxim (1.5 l/ha)	100	89	81	76	86	92	54	100	90	91
Bispyriac sodium 100 SC	92	52	85	45	90	84	50	80	90	84
3.4 DPA *fb* MCPA	97	71	90	60	86	84	62	64	70	31
No weeding	-	-	-	-	-	-	-	-	-	-
Hand weeding	-	-	-	-	-	-	-	-	-	-

EC=*Echinochloa crus-galli,* LC = *Leptochloa chinensis,* IR = *Ischaemum rugosum,* PD = *Paspalum distichum,* MV = *Monochoria vaginalis,* LO = *Ludwigia octavalvis,* MN = *Murdannia nudiflora,* CD = *Cyperus difformis,* CI = *Cyperus iria,* FM = *Fimbristylis miliacea*

Growth and development of rice plant

No detrimental effects on growth and development of rice pants were observed in the herbicide-treated plots (Table3). A significantly lower plant count and dry weight were obtained in the no-weeding treatment, which may be due to the competition due to weeds

Table.3: Effect of pretilachlor + pyribenzoxim on growth and development of rice

Treatment	Plant height (cm)	Plan count (No./m²)	Dry weight (g/ m²)
Pretilachlor + Pyribenzoxim (1 l/ha)	65.25	691	522.4 a
Pretilachlor + Pyribenzoxim (1.25 l/ha)	60.1	702	541.0 a
Pretilachlor + Pyribenzoxim (1.5 l/ha)	68.33	730	617.4 a
Bispyribac sodium 100 SC	62.43	786	585.4 a
3.4 DPA *fb* MCPA	64.37	737	605.3 a
No weeding	65.6	688	516.3 a
Hand weeding	65.13	518	408.2 b

Within a column, means followed by the same letter are not significantly different by the Duncan's New Multiple Range Test (p=0.05)

Yield and yield component of rice

Significantly higher rice yields ($p<0.05$) were observed between the herbicide treatments and the un-weeded treatment (Table 4).

The yield of rice was the highest in pretilachlor + pyribenzoxim (300 g a.i./l + 20 g a.i./l) applied at 1.5 l/ha (5.88 t/ha) but it was not significantly different to that other herbicide-treated plots and hand weeded plot (5.59 t/ha).The lowest yield was obtained from un weeded plot due to severe weed competition.

Table 4. Effect of pretilachlor + pyribenzoxim on grain yield and yield component of rice

Treatment	Yield (mt/ha)	Panicles per m²	Grains per panicle	1000-grain wt (g)
Pretilachlor + Pyribenzoxim (1 l/ha)	5.22 a	502 a	103 a	27.61 a
Pretilachlor + Pyribenzoxim (1.25 l/ha)	5.64 a	533 a	103 a	27.06 a
Pretilachlor + Pyribenzoxim (1.5 l/ha)	5.88 a	505 a	121 a	27.43 a
Bispyribac sodium 100 SC	5.18 a	442 a	102 a	27.04 a
3.4 DPA *fb* MCPA	5.45 a	503 a	92a b	27.27 a
Hand weeded	5.59 a	513 a	103 a	27.24 a
un weeded	1.46 b	374 b	88 b	26.83 a

Within a column, means followed by the same letter are not significantly different by the Duncan's New Multiple Range Test (p=0.05)

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A PRELIMINARY STUDY ON COMPETITION BETWEEN DAYFLOWER (*Commelina communis* L.) AND SOYBEAN [(*Glycine max* (L.) Merr.]

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Abstract: The nature of competition between soybean and different densities of dayflower was evaluated in plots which dayflower seeds were allowed to regenerate, thereby developing different populations of the weed. With increasing populations of the weed, interspecfic competition in dayflower and competition between soybean and the weed increased. When dayflower density increased from 5 to 60 plants/m^2 plant height of dayflower increased, but weight per plant decreased. When dayflower density increased from 0 to 60 plants/m^2, plant height of soybean increased; its stem thickness declined; pods and grains of single plant were reduced. Although the weight of one hundred grains of soybean increased gradually, the yield of soybean decreased. There was a positive correlation between soybean yield loss and dayflower density. Comparing the regression coefficient of linear, logarithmic, exponential and power function, the best regression equation was $y=1.5446x^{0.6089}$, r=0.8721**.

Key words: Dayflower, soybean, competition

Introduction

Competition between soybeans and weeds has been extensively researched. Research at the Plant Protection Research Institute of Heilongjiang Academy of Agricultural Science (1989) showed that the main period of the competition of weed with soybean was 6-9 weeks after planting and the best period of weeding was 5-6 weeks after crop establishment. The competition between soybean and Common lambsquarters (*Chenopodium album* L.) or Green foxtail (*Setaria viridis* P. Beauv.) caused a reduction in leaf area, dry weight, plant pods and grains of soybean. The yield of soybean reduced gradually along with the increasing density of Common lambsquarters and Green Foxtail (Fu *et al.* 1992a,b). Dekker and Meggitl (1983), Cordes and Bauman (1984), and Wyse *et al.* (1986), reported the competition between Velvetleaf (*Abutilon theophrasti* Medic), Ivyleaf morningglory (*Ipomoea hederacea* (L.) Jacq.), Jerusalem artichoke (*Helianthus tuberosus* L.) and soybean (*Glycine max* (Linn.)Merr.). There are no reports on the competition between dayflower and soybean.

Dayflower (*Commelina communis* L.) is an annual weed and emerges in the first ten-day of May in the farmland of Heilongjiang province. It branches from 4-6 leaves and has the characteristic of extensive branching and regenerating roots on nodes and hence has a very strange regeneration ability. There are many flowers on one plant and seeds mature in ten days after flowering and drop when mature. The average number of seeds is 164.1 grains/plant. The emergence of dayflower plants occurs at different times and there is an obvious peak period. The growing speed of dayflower plants is different where those emerging in the peak period grow faster. The resistance of Dayflower to stress is low before 4 leaves emerge and plants become stronger after 4 leaves. Dayflower of 7-10 leaves were left in the sun for 3 days and when transplanted, the survival rate was 100%, after sunning for 7 days, the survive rate was over 50%. These biological characteristic show that dayflower has very stronger adversity-resistant and adaptability to environment. This is one of reasons that it becomes a malignant weeds (Huang *et al.* 2002; You *et al.* 2002). The problem of dayflower has been increasing in recent years in the Heilongjiang province and is becoming difficult to control. A study in soybean fields in Sanjiang plain of Heilongjiang province showed that the rate of occurrence of dayflower was 66.4%, the field density 2.99, and the field even rate 26.5%.

The objective of this experiment is to determine the influence of the different densities of dayflower to its growth itself and to soybean growth and the yields. The experiment also evaluated the relationship between the density of dayflower and soybean yield loss, and established the regression equations

Materials and Methods

The experiment was located at the Hejiang Institute of Agricultural Science of Heilongjiang Academy of Agricultural Sciences, in the Jiamusi city of Heilongjiang province. The soil was a hardpan meadow soil with a smooth terrain. The organic matter content of the soil was 2.56; pH 6.49, total N 0.14%, total P 0.05%, and total K 1.46%. Soil preparation and ridging was carried out in autumn,

The experimental treatments were the six densities of Dayflower plants, namely 0, 5, 10, 20, 40 and 60 plants/m^2 arranged within a Randomized Complete Block Design with 3 replicates. The individual plots were 5 m x 0.7 m with four ridges, each 5m long and spaced at 70 cm. The densities of the dayflower plants were maintained by thinning out the extra plants. All other weeds were removed to avoid interactive effects.

Soybeans were planted in the prepared plots at a density of 357000 plants per ha, by establishing 125 healthy uniform seeds in a 5m row. The measurements made were soybean plant height on two occasions (summer and early autumn). In addition, soybean and dayflower plants within two 0.1m^2 quadrats were removed prior to the final harvest. The thickness of the main stem in soybean plants, pods per plant, 100 seed weight, seed yield per unit area and fresh and dry weights of both soybean and dayflower plants were determined.

Results and Discussion

The different densities influenced the growth of dayflower. Increasing densities of dayflower reduced both fresh and dry weights of individual plants of this species (Figure 1). When dayflower density increased from 5 plants/m^2 to 60 plants/m^2, dayflower plant fresh weight decreased from 49.7 g/plant to 10.6 g/plant (a reduction of 78.67%), the dry weight decreased respectively from 12.4 g/plant to 2.9 g/plant (a reduction of 76.61).

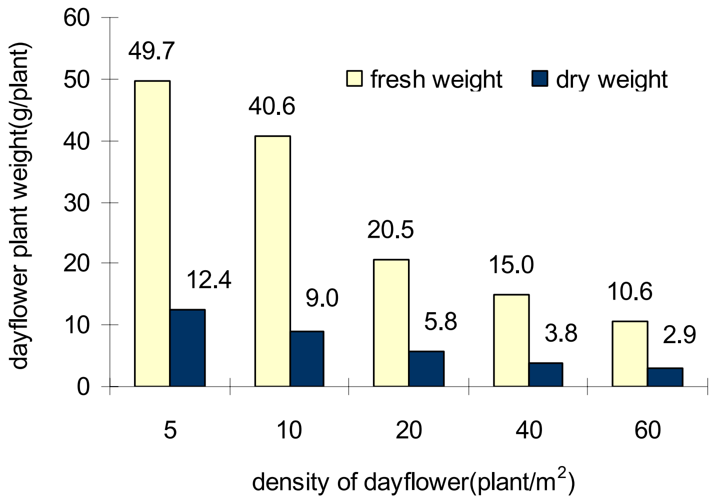


Figure1. Influence of dayflower density to its plant weight

Competitive effects between soybean and dayflower was not evident at the early stages of growth as shown by plant heights of both species at the first sampling (Figure 2), although here were different densities of the weed. At the second sampling competitive effects were clearly visible (Figure 2). When dayflower density was increased from 5 to 60 plants/m^2, the plant height of dayflower increased from 81.9 cm to 89.0 cm (increase of 8.67%). When dayflower density rising from 0 to 60 plants/m^2, the adult plant height of soybean increased from 73.9 cm to76.0 cm (an increase of 2.76%).

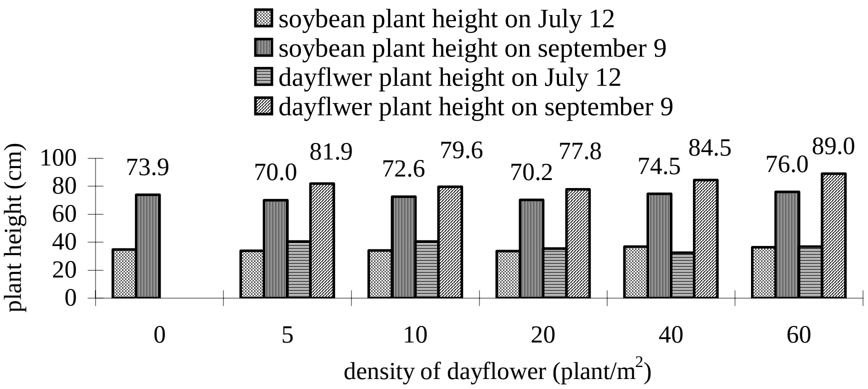


Figure 2. Influence of dayflower density to plant height of soybean and dayflower

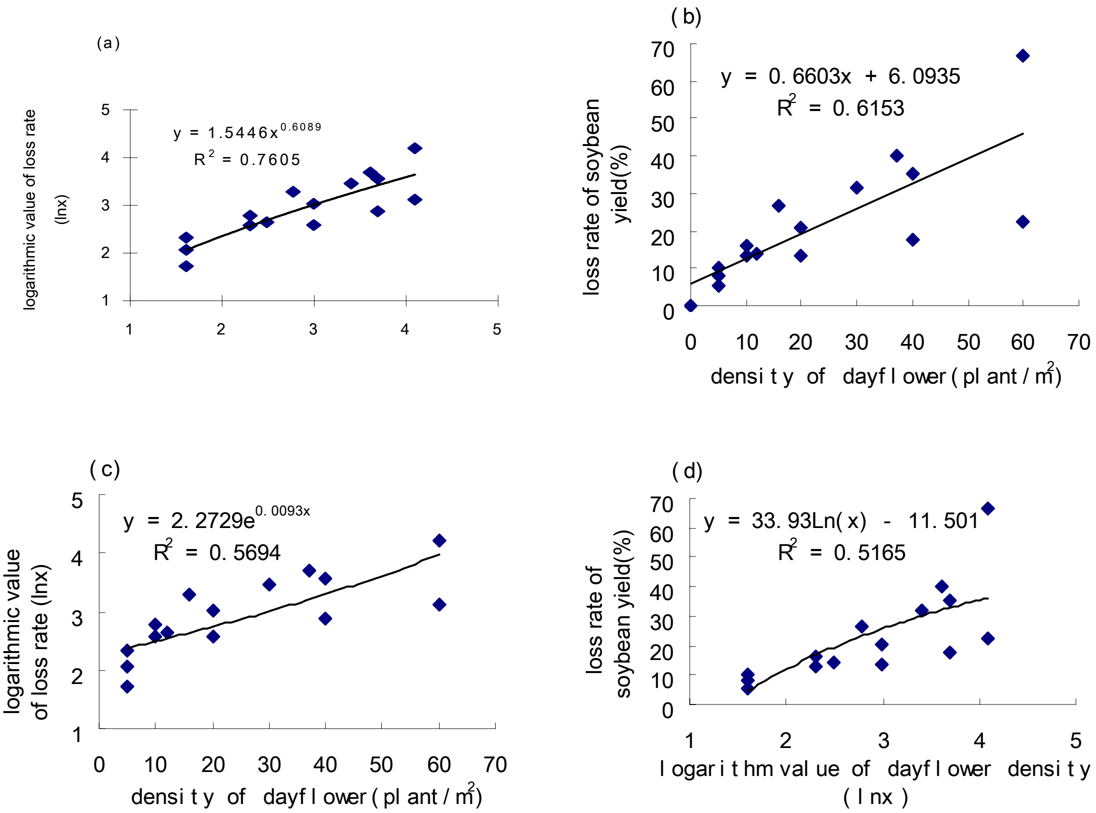
With increasing dayflower density, the diameter of soybean stems were reduced significantly, pods per plant and grain numbers reduced, resulting in a significant decline in yields (Table 1). When dayflower density increased from 0 to 60 plants/m^2, the soybean stem thickness decreased from 0.564 cm to 0.457 cm, the pods of single plant reduced from 22.1 to 10.2 pods/plant, and the grains of single plant reduced from 52.9 seeds/plant to 33.8 seeds/plant. However, the weight of one hundred grains increased marginally from 21.1g to 22.4 g, which could be due to compensatory grain filling, caused by the lower numbers of seeds per plant. The final seed yield however declined with increasing density and the decline was from 2643.0 kg/hm^2 to 1481.3 kg/hm^2 in the experiment (Table 1).

Table 1 Influence of dayflower density to soybean growth and yield

Dayflower density (plant/m^2)	Stem thickness (cm)	Pods/plant	Grains/ plant	100 grain weight (g)	Soybean yield (kg/hm^2)	Rate of yield loss (%)
0	0.564	22.1	52.9	21.1	2643.0 F	0
5	0.525	21.2	48.3	21.2	2433.8 E	7.92
10	0.520	20.4	47.1	21.7	2254.5 D	14.70
20	0.507	20.4	47.0	21.5	2193.8 C	17.00
40	0.502	17.5	43.7	22.0	1947.0 B	26.33
60	0.457	10.2	33.8	22.4	1481.3 A	43.95

The data are means of three replicates

Regression analysis using linear, logarithmic, exponential and power functions present the relationship between dayflower density and seed yield of soybean. There was a positive correlation between soybean yield loss and dayflower density, the correlation coefficient of all four functions were significant. Comparing the correlation coefficients of four functions, the best regression equation was power function, $y=1.5446x^{0.6089}$, r=0.8721**, the second was a linear function, the third an exponential function and the last was the logarithmic function (Figure 3).



(a) Regression of power function, (b) Regression of linear function,
(c) Regression of exponential function, (d) Regression of logarithmic function

As with all weed species, the competitive ability of dayflower strengthened with increasing density. This had a negative impact on soybean growth and development. The final outcome of the study was the clear evidence of the impact of increasing densities of dayflower on the growth of the same plant species and also on soybeans, resulting in a significant yield loss.

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INTEGRATED WEED MANAGEMENT IN WHEAT (*Triticum aestivum* L.) UNDER RAINFED CONDITIONS

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Abstract: A field experiment was conducted in NWFP-Pakistan during 2004-2005, where two sowing methods (Factor A), line and broadcast sowing, and two crop densities (Factor B), 100 and 150 kg seed ha^{-1}, were arranged in the main plots and four herbicides (Factor C) along with weedy check in the subplots in a split plot design. The herbicides were Puma super® 75 EW (fenoxaprop-p-ethyl) at 0.75 kg, Isoproturon® 50 WP (isoproturon) at 1.0 kg, Topik® 15 WP (clodinafop-propargyl) at 0.04 kg, Buctril super® 60 EC (bromoxynil+MCPA) at 0.45 kg a.i. ha^{-1}. The lowest weed biomass (111 kg ha^{-1}) was observed when bromoxynil+MCPA was applied to wheat sown in lines at the density of 150 kg ha^{-1} followed by isoproturon (155 kg ha^{-1}) applied to wheat sown in lines at the density of 150 kg ha^{-1} when compared to of the in the weedy check that (2188 kg ha^{-1}). Similarly, number of tillers (462 m^{-2}), grain yield (2667 kg ha^{-1}), thousand grains weight (46.4 g) and harvest index (26.5%) were the maximum in bromoxynil+MCPA followed by isoproturon (394 m^{-2}), (2294 kg ha^{-1}), (43.3 g) and (22.6%) as compared to the weedy check (248 m^{-2}), (1561 kg ha^{-1}), (38.0 g) and (15.1%), respectively. The herbicides bromoxynil+MCPA at 0.45 and isoproturon at 1 kg a.i. ha^{-1} applied post emergence in wheat sown in lines at the density of 150 kg ha^{-1} produced the best weed control and grain yield under rain-fed conditions.

Key words: Wheat, seed rates, densities, sowing methods and herbicides

Introduction

During 2004-05, the area under wheat cultivation in Pakistan was 8.358 million ha, with a production of 21.6123 million tons while in the province NWFP, this area was about 0.7486 million ha. One third of this area in the NWFP is irrigated, while two third is rain fed giving a total production of 1.091 million tons at the rate of 1458 kg ha^{-1} (Minfal, 2005). The growth rate of population is around 3% annually and there is continuous and gradual increase in per capita use of wheat and its products. Thus, rainfed agriculture must be given due consideration for enhancing productivity per unit area. Drought conditions reduce critical growth processes such as photosynthesis, cell enlargement and cell division (Hsaia, 1973, Tarner and Karmer, 1980). However, the response of different varieties to environmental conditions differs under stress, and some varieties tolerate the drought environment to a greater extent. The low yields of wheat in the rainfed area are probably due to improper soil and water management practices, soils low in organic matter content, inefficient and imbalance use of fertilizers, loss of added fertilizer due to adsorption or leaching, high pH of the soils, calcareous nature of the soils, poor soil structure and weed infestation.

There is increasing pressure to reduce applications of agrochemicals for both cost and environmental reasons. The recommended dose of an herbicide is set by the manufacturer to ensure that the product works well under a range of circumstances. In order to maximize the efficacy of an herbicide and choose an appropriate dose, an understanding of herbicide response to weather factors is important (Kudsk and Kristensen, 1992). The annual losses to wheat crop in Pakistan due to weeds on a monetary basis amount to Rs. 34 billions, while in NWFP it amounts to Rs. 2 billion (Marwat, 2002). These figures warrant an efficient control of weeds. It has been estimated that crop losses due to weeds competition throughout the world as a whole are greater than those resulting from combined effect of insect pests and diseases. At present, when the nation is facing an abrupt population increase, it is a challenge for the scientists to divert the resources towards wheat from the weeds. Conventional methods of weed control are weather dependent, laborious and costly. Thus, chemical weed control has been proved to be relatively efficient, and economical in controlling the weeds (Taj *et al*. 1986, Majid and Hussain, 1983). With rising costs of labor and power, the optimum use of herbicides will be the only acceptable method of weed control in the future. Thus, an experiment was conducted with the objectives of evaluating different herbicides for weed control in wheat (variety KT-2000) under rainfed conditions and study different sowing methods and seed rates and to select the best herbicide-seed rate-sowing method combination that may give the best yield of wheat in the rain-fed areas.

Materials and Methods

The experiment was conducted at Barani Agriculture Research Station, Kohat (NWFP-Pakistan) during October to May 2004-2005 and was laid out in a Randomized Complete Block Design with a split plot arrangement and three replications. Two seed rates (Factor A) and two sowing methods (Factor B) were assigned to main plots and four post-emergence herbicides (Factor C) along with a weedy check were assigned to subplots. The size of each experimental unit was kept 5 m x 1.8 m with 6 rows, each 30 cm apart. The data were recorded on weed control efficiency (%), fresh weed biomass (kg ha^{-1}), number of tillers m^{-2}, 1000-grain weight (g), biological yield (kg ha^{-1}), and grain yield (kg ha^{-1}). The weed control efficiency was determined by the formula,

$$\text{Weed Control Efficicency (\%)} = \frac{(\text{Weed density m}^{-2}\text{ before} - \text{Weed density m}^{-2}\text{ after}) \times 100}{\text{Weed density m}^{-2}\text{ before herbicide application}}$$

The main plots were two sowing methods namely, line sowing and broadcast sowing, and two seed rates namely, 100 kg ha-1 and 150 kg ha-1. The sub plots were herbicide treatments including Buctril super® 60 EC (bromoxynil+MCPA) at 0.45 kg, Topik® 15 WP (clodinafop-propargyl) at 0.04 kg, Puma super® 75 EW (fenoxaprop-p-ethyl) at 0.75 kg, and Isoproturon® 50 WP (isoproturon) at 1.0 kg a.i. ha^{-1} and a weedy check. The data recorded for each parameter was individually subjected to the ANOVA technique by using MSTATC computer software and means were separated by using Fisher's Protected LSD test (Steel and Torrie, 1980).

Results and Discussion

Weed control efficiency (%)

The data revealed that the differences due to herbicides were significant while among the sowing methods and seed rates were non-significant. The maximum weed control efficiency (86.2%) was recorded with Buctril super® 60 EC, followed by Isoproturon® 50 WP (75.0%) (Table 1), which may be due to better weed control. These results agree with those of Salarzai *et* al. (1999) who stated that herbicides significantly control the weed population in wheat.

Fresh weed biomass (kg ha^{-1})

In terms of weed biomass, differences among the herbicides, sowing methods and seed rates were significant. Among the herbicides, minimum weed biomass (111 kg ha^{-1}) was observed in Buctril super® 60 EC which was similar to that obtained with Isoproturon® 50 WP (155 kg ha^{-1}) while the maximum weed biomass (2188 kg ha^{-1}) was recorded in the weedy check (Table 2). Khan *et al*. (2002) also reported that herbicides application decreased the weed biomass as compared to weedy check. These findings are also in conformity with those of Shahid (1994), and Tunio *et al.* (2004), who reported that herbicides significantly reduced weed density and fresh weed biomass.

Table 1. Weed control efficiency (%) as affected by different treatments in wheat.

Herbicides	Line sowing		Broadcast sowing		Herbicide Means
	100 kg ha^{-1}	150 kg ha^{-1}	100 kg ha^{-1}	150 kg ha^{-1}	
Topik® 15 WP	57.3	36.4	24.7	25.0	35.8 c
Isoproturon® 50 WP	70.7	71.4	71.7	86.4	75.0 ab
Puma super® 75EW	28.8	30.0	31.8	44.0	33.6 cd
Buctril super® 60 EC	89.6	86.8	87.8	80.9	86.2 a
Weedy check	0.0	0.0	0.0	0.0	0.0 d
Seed rate means	44.9	49.3	43.1	47.2	
Sowing method means	47.1		45.2		

$LSD_{0.05}$ for herbicides = 21.0; Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Table 2. Fresh weed biomass (kg ha^{-1}) as affected by different treatments in wheat

Herbicides	Line sowing		Broadcast Sowing		Herbicide Means
	100 kg ha^{-1}	150 kg ha^{-1}	100 kg ha^{-1}	150 kg ha^{-1}	
Topik® 15 WP	252	308	434	292	323 b
Isoproturon® 50 WP	157	107	193	166	155 c
Puma super® 75EW	449	435	439	365	422 b
Buctril super® 60 EC	133	90	109	111	111 c
Weedy check	2387	1829	2526	2012	2188 a
Seed rate means	675 a	554 b	740 a	589 b	
Sowing method means	614 b		665 a		

$LSD_{0.05}$ for herbicides = 143; Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Number of tillers m^{-2}

The number of tillers m^{-2} in wheat was significantly affected by the herbicides. The maximum tillers (462 m^{-2}) were recorded with Buctril super® 60 EC followed by Isoproturon® 50 WP (394 m^{-2}) and the minimum number of tillers (248 m^{-2}) was in the weedy check. The differences among the seed rates and sowing methods were non-significant. Baldha, *et al*. (1998) reported similar results who reported that application of herbicides significantly influenced the number of tillers in wheat. Weed control has resulted in higher yield in wheat by increasing number of tillers (Borghain *et al*. 1985).

Thousand grain weight (g)

The 1000-grain weight was significantly affected by different herbicidal treatments, sowing methods and seed rates (Table 4). Among the herbicide treatments, the maximum (46.4 g) 1000-grain weight was recorded with Buctril super® 60 EC while the minimum (38 g) was recorded in the weedy check. Among the sowing methods, the maximum 1000 grain weight (43.7g) was recorded in line sown wheat and among the seed rates, the maximum thousand grain weight was recorded with 100 kg ha^{-1}. As 1000 grain weight is a very important yield component in every crop, an increase in this parameter will automatically increase the total grain yield of the crop. Similar results were reported by Hassan *et al*. (2003) who reported that herbicides application increased the 1000 grain weight of wheat significantly when compared with the weedy check.

Table 3. Number of tillers m^{-2} as affected by different treatments in wheat

Herbicides	Line sowing		Broadcast sowing		Herbicide Means
	100 kg ha^{-1}	150 kg ha^{-1}	100 kg ha^{-1}	150 kg ha^{-1}	
Topik® 15 WP	374	285	284	319	315 c
Isoproturon® 50 WP	443	348	377	410	394 ab
Puma super® 75EW	417	291	336	365	352 bc
Buctril super® 60 EC	490	408	459	474	462 a
Weedy check	227	241	241	284	248 d
Seed rate means	394	314	339	370	
Sowing method means	354		355		

$LSD_{0.05}$ for herbicides = 65; Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Table 4. Thousand grains weight (g) as affected by different treatments in wheat.

Herbicides	Line sowing		Broadcast sowing		Herbicide Means
	100 kg ha^{-1}	150 kg ha^{-1}	100 kg ha^{-1}	150 Kg ha^{-1}	
Topik® 15 WP	43.2	43.6	40.0	38.4	41.3 bc
Isoproturon® 50 WP	45.8	44.2	42.1	41.3	43.3 b
Puma super® 75EW	42.9	41.9	42.8	39.3	41.7 bc
Buctril super® 60 EC	48.5	48.3	45.1	43.9	46.4 a
Weedy check	39.6	39.1	37.2	36.2 l	38.0 c
Seed rate means	44.0 a	43.4 b	41.4 a	39.8 b	
Sowing method means	43.7 a		40.6 b		

$LSD_{0.05}$ for herbicides = 2.6; Within a column, means followed by the same letter are not significantly different by the (p=0.05).

Biological yield (kg ha^{-1})

The differences among the herbicides and sowing methods were significant whereas the differences between the seed rates were non-significant (Table 5).

Table 5. Biological yield (kg ha^{-1}) as affected by different treatments in wheat.

Herbicides	Line sowing		Broadcast Sowing		Herbicide means
	100 kg ha^{-1}	150 kg ha^{-1}	100 kg ha^{-1}	150 kg ha^{-1}	
Topik® 15 WP	10400	10230	9944	10570	10286 bc
Isoproturon® 50 WP	10040	10830	9333	10420	10158 ab
Puma super® 75EW	10800	10500	9333	10110	10186 bc
Buctril super® 60 EC	11170	11000	9778	10280	10555 a
Weedy check	10330	10730	9666	10480	10302 bc
Seed rate means	10548	10660	9611	10331	
Sowing method means	10604 a		9991 b		

$LSD_{0.05}$ for herbicides = 1399; Within a column (herbicide means) and within a row (sowing method means), respectively, means followed by the same letter are not significantly different by the LSD (p=0.05).

Among the herbicides, the maximum biological yield (10555 kg ha^{-1}) was recorded with Buctril super® 60 EC. Similarly among the sowing methods, maximum (10604 kg ha^{-1}) biological yield was recorded in line sown wheat. The increase in biological yield was due to the best weed control and optimum seed rate. Hassan, *et al*. (2003) also reported analogous results.

Grain yield (kg ha^{-1})

The differences among herbicides and sowing methods were significant. Among the herbicides, maximum (2676 kg ha^{-1}) grain yield was recorded with Buctril super® 60 EC due to its good weed control while minimum (1561 kg ha^{-1}) was recorded in weedy check (Table 6).

Table 6. Grain yield (kg ha^{-1}) as affected by different treatments in wheat in Kohat.

Herbicides	Line sowing		Broadcast Sowing		Herbicide means
	100 kg ha^{-1}	150 Kg ha^{-1}	100 kg ha^{-1}	150 kg ha^{-1}	
Topik® 15 WP	1933	1900	1844	1833	1877 c
Isoproturon® 50 WP	2200	2444	2267	2267	2294 b
Puma super® 75EW	2156	2100	2033	2133	2105 b
Buctril super® 60 EC	2900	2622	2618	2567	2676 a
Weedy check	1467	1511	1700	1567	1561 c
Seed rate means	2131 a	2115 b	2092 a	2073 b	
Sowing method means	2123 a		2082 b		

$LSD_{0.05}$ for herbicides = 215; Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Among the seed rates, the maximum yield (2111.5 kg ha^{-1}), the average of 2131 and 2092, was recorded with 100 kg seeds ha^{-1} which may be due the absence of intra-specific competition. Among the sowing methods, the highest grain yield (2123 kg ha^{-1}) was recorded in line sown wheat due to the optimum spacing. These results are in conformity with those reported by Hassan *et al*. (2003). They reported that herbicidal treatments significantly increased grain yield in wheat.

Conclusions

After subjecting the four herbicides plus weedy control, the two seed rates and the two sowing methods to experimentation, it was concluded that Buctril super® 60 EC at 0.45 kg a.i. ha^{-1} as post emergence herbicide was the best among all the herbicidal treatments. Among the sowing methods, wheat grain yield was maximum in line sown treatments and among the seed rates maximum grain yield was obtained with 100 kg ha^{-1}. The best seed rate and sowing method should be integrated with best performing herbicides to gain higher yields in wheat under rain-fed conditions.

Acknowledgements

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CONTROL OF SULFONYLUREA-RESISTANT WEEDS IN PADDY FIELDS OF KOREA

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Abstract: This study was conducted to determine the impact of several sulfonylurea-resistant weeds discovered in paddy fields in Korea. Non-sulfonylurea herbicides, such as oxadiazon (480 g a.i. ha^{-1}), butachlor (1,320 g a.i. ha^{-1}), pretilachlor (560 g a.i. ha^{-1}) and fentrazamide (950 g a.i. ha^{-1}) applied at pre-emergence stage controlled resistant biotypes of *Monochoria vaginalis*, *Lindernia dubia*, *Scirpus* spp. and *Cyperus difformis*, but in *Sagittaria pigmaea* the control was 90% when oxadiazon was applied. The control effect at one-leaf-stage of *M. vaginalis*, *L. dubia*, *Scirpus juncoides*, *C. difformis* and *Sagittaria pigmaea* was over 90% with the application of pretilachlor (600 g a.i. ha^{-1}), pyrazolate+butachlor (1,800+1,050g a.i. ha^{-1}), carfentrazone-ethyl+thiobencarb (75+2,100 g a.i. ha^{-1}), carfentrazone-ethyl+clomazone (75+63 g a.i. ha^{-1}) or carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl (75+30+21 g a.i. ha^{-1}). The two-leaf stage of these weeds was controlled by over 90% with the application of carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl. At the four-leaf stage of *M. vaginalis*, *L. dubia*, *S. juncoides*, the control effects were over 90% with the application of pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, and carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl. The control of sulfonylurea-herbicide-resistant-weeds by the application of thiobencarb+simetryn (2,100+300 g a.i. ha^{-1}) onto the surface water showed a high degree of success in *M. vaginalis*, *L. dubia* but it was very low in *S. pigmaea*. The resistant biotype of *S. juncoides* and *C. difformis*, which survived 30~35 days, could effectively be controlled only by foliar application of bentazone (1600 g a.i. ha^{-1}), whereas *M. vaginalis*, *L. dubia* and *S. pigmaea* could be controlled by application of mixtures of either bentazone and 2, 4-D (1200+140g a.i. ha^{-1}) or bentazone+MCPA (930+138g a.i. ha^{-1}).

Key words: Weed control, herbicide resistant weed, Korea, sulfonylurea

Introduction

Herbicide resistant weeds in paddy fields of Korea was first reported in *Monochoria kosakowii* discovered in Seoasan of Chungnam province (Park *et al*. 1999), followed by *Monochoria vaginalis* (Kwon *et al*. 2000), *Lindernia dubia* (Park *et al*. 2001) and *Rotala indica* (Kuk *et al*. 2003) in Chunnam province, *Scirpus juncoides* (Im *et al*. 2003a) and *Cyperus difformis* (Im *et al*. 2003b) in Chunbuk province, *Sagittaria pigmaea* (Im *et al*. 2005) in Chungnam province. Due to increases in herbicide resistant weeds in paddy fields, research has been conducted on their control. Sulfonylurea herbicide resistant *Monochoria vaginalis* in paddy fields decreased to 70% in directed seeded paddy fields, while the reduction was 44% in transplanted paddy fields (Kwon *et al*. 2002). Kwon *et al*. (2002) also reported that Sulfonylurea herbicide resistant *Monochoria vaginalis* must be controlled no later then 31 days after seeding in water-direct-seeding-paddy and up to 45 days after transplanting in transplanting-paddy in order to maintain rice-yield. Due to the lack of proper guidelines to control herbicide resistant weeds, this study was conducted to determine effective control methods of herbicides-resistant-weeds in paddy fields in Korea

Materials and Methods

Control of Sulfonylurea herbicide resistant weeds

Sulfonylurea-resistant weeds used in the experiments were five species namely *Monochoria vaginalis*, *Lindernia dubia*, *Scirpus juncoides*, *Cyperus difformis* and *Sagittaria pigmaea*.

Non-sulfonylurea herbicides, such as oxadiazon (480 g a.i. ha^{-1}), butachlor (1,320 g a.i. ha^{-1}), pretilachlor (560 g a.i. ha^{-1}) and fentrazamide (950 g a.i. ha^{-1}) were applied separately at the pre-emergence stage of weeds after leveling the rice field for seeding to control sulfonylurea-herbicide-resistant-weeds. Herbicides such as butachlor (1,500 g a.i. ha^{-1}), pretilachlor (600 g a.i. ha^{-1}), thiobencarb (2,100 g a.i. ha^{-1}), esprocarb (1,500 g a.i. ha^{-1}), mefenacet (1,050 g a.i. ha^{-1}), fentrazamide (300 g a.i. ha^{-1}), molinate (2,100 g a.i. ha^{-1}), pyriminobac-methyl (30 g a.i. ha^{-1}), cyhalofop-butyl (180 g a.i. ha^{-1}), dimepiperate (2,100 g a.i. ha^{-1}), pyrazolate+butachlor (1,800+1,050 g a.i. ha^{-1}), carfentrazone-ethyl+thiobencarb (75+2,100 g a.i. ha^{-1}), carfentrazone-ethyl+clomazone (75+63 g a.i. ha^{-1}), carfentrazone-ethyl+pyriminobac-methyl +pyrazosulfuron-ethyl (75+30+21 g a.i. ha^{-1}), thiobencarb+simetryn (2,100+300g a.i. ha^{-1}) and pyrazosulfuron-ethyl+molinate+simetryne (21+1,500+90 g a.i. ha^{-1}) were applied at 1, 2, and 4 leaf stages of weeds to control sulfonylurea-herbicide-resistant-species. The water level in the paddy field was maintained at a 3 cm in depth after the application of herbicide, for 20 days.

The experiments were also conducted in a green house using plastic pots of 18 cm x 26 cm x 10 cm size having 2,500 ml of silt sandy loam soil. The experiment carried out using a Randomized Complete Block Design with three replicates. Soil used in the experiment was sterilized for 20 minute in an autoclave at 120°C. Herbicides for leaf-stem application, such as bentazone (1600g a.i. ha^{-1}), 2,4-D (280g a.i. ha^{-1}), bentazone+2,4-D (1200+140g a.i. ha^{-1}) and bentazone+MCPA (930+138g a.i. ha^{-1}) were sprayed at the 30~35 days growth stage to control middle or late season weeds that are sulfonylurea-herbicide-resistant-weeds in paddy fields. Weeding efficacy was estimated at 30 days after application of the herbicides.

Results and Discussion

Pre-emergence control of Sulfonylurea herbicide resistant weeds

Non-sulfonylurea herbicides, such as oxadiazon (480 g a.i. ha^{-1}), butachlor (1,320 g a.i. ha^{-1}), pretilachlor (560 g a.i. ha^{-1}) and fentrazamide (950 g a.i. ha^{-1}) applied at pre-emergence stage controlled the resistant biotypes of *M. vaginalis, L. dubia, S. juncoides, C. difformis,* and *S. pigmaea* (Table 1).

Table 1. Control effects of sulfonylurea herbicide resistant several weeds by herbicides applied onto soil at the pre-emergence stage of weeds.

Herbicide	*Monochoria vaginalis*	*Lindernia dubia*	*Scirpus juncoides*	*Cyperus difformis*	*Sagittaria pigmaea*
Oxadiazon	100	100	100	100	90
Butachlor	100	100	100	100	73
Pretilachlor	100	100	100	100	74
Fentrazamide	100	100	100	100	45

Application of oxadiazon controlled over 90% of weeds but the effect was below 74% with the application of butachlor, pretilachlor and fentrazamide. *Monochoria vaginalis* (Kwon *et al*. 2000) and sulfonylurea-resistant *L. dubia, S. juncoides* and *C. difformis* (Im *et al*. 2003a) have been reported to have controlled by application of oxadiazon at the time of land leveling.

Control at one-leaf-growth-stage in sulfonylurea herbicide resistant weeds

The application of pretilachlor, thiobencarb, mefenacet, fentrazamide, pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl +pyrazosulfuron-ethyl+pyriminobac-methyl, pyrazosulfuron-ethyl+molinate+simetryn and thiobencarb+simetryn totally controlled *M. vaginalis,* but the application of molinate, dimepiperate, pyriminobac-methyl and cyhalofop-butyl showed a low weed control efficacy (Table 2).

Table 2. Weeding efficacy of non-sulfonylurea herbicides applied at one-leaf-growth-stage on sulfonylurea-herbicide-resistant-weeds.

Herbicides	Weed control efficacy (%)				
	Monochoria vaginalis	*Lindernia dubia*	*Scirpus juncoides*	*Cyperus difformis*	*Sagittaria pigmaea*
Butachlor	87	100	100	100	79
Pretilachlor	100	100	100	100	94
Thiobencarb	100	100	97	100	9
Esprocarb	99	100	100	100	54
Mefenacet	100	91	100	100	63
Fentrazamide	100	96	68	100	21
Molinate	4	6	1	12	3
Pyriminobac-methyl	6	7	13	48	-
Cyhalofop-butyl	2	9	5	45	-
Dimepiperate	2	4	1	45	-
Pyrazolate+butachlor	100	100	100	100	93
Carfentrazone-ethyl+thiobencarb	100	100	100	-	100
Carfentrazone-ethyl+clomazon	100	100	100	-	100
Carfentrazone-ethyl+ pyrazosulfuron-ethyl+ pyriminobac-methyl	100	100	99	-	100
Thiobencarb+simetryn	100	100	100	-	70
Pyrazosulfuron-ethyl+molinate+simetryn	100	97	99	-	39

The applications of butachlor, pretilachlor, thiobencarb, esprocarb, pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl or thiobencarb+simetryn for *Lindernia dubia,* and applications of either butachlor, pretilachlor, esprocarb, mefenacet, pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl, thiobencarb+simetryn or pyrazosulfuron-ethyl+molinate+simetryn for *S. juncoides*, the applications of either butachlor, pretilachlor, thiobencarb, esprocarb, mefenacet, fentrazamide, or pyrazolate +butachlor for *C. difformis* showed a relatively high weed control efficacy. The *S. pigmaea,* which is sulfonylurea resistant, was controlled effectively by the application of either carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone or carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl, but by the application of simetryn herbicides did not produce similar effects.

Control of sulfonylurea herbicide resistant weeds at the two leaf stage

The application of either mefenacet, pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl or thiobencarb+simetryn for *M. vaginalis,* the application of either butachlor, pretilachlor, thiobencarb, esprocarb, pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl or thiobencarb+simetryn for *L. dubia*, the application of either butachlor, pretilachlor, esprocarb, pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl, thiobencarb+simetryn or pyrazosulfuron-ethyl+molinate+simetryn for *Scirpus juncoides*, and the application of either butachlor, pretilachlor, thiobencarb, esprocarb, mefenacet, fentrazamide or pyrazolate+butachlor for *C. difformis*, controlled all tested weeds by over 90% (Table 3).

Table 3. Weeding efficacy on sulfonylurea-herbicide-resistant-weeds applied with non-sulfonylurea herbicides at two-leaf-growth-stage.

Herbicides	Weed control efficacy (%)				
	Monochoria vaginalis	*Lindernia dubia*	*Scirpus juncoides*	*Cyperus difformis*	*Sagittaria pigmaea*
Butachlor	47	96	91	100	74
Pretilachlor	77	94	94	100	72
Thiobencarb	89	98	71	100	0
Esprocarb	55	91	100	100	75
Mefenacet	96	78	89	100	53
Fentrazamide	65	89	9	100	15
Molinate	1	1	0	4	0
Pyriminobac-methyl	8	4	6	3	-
Cyhalofop-butyl	4	9	4	3	-
Dimepiperate	5	2	0	4	-
Pyrazolate+butachlor	94	99	97	100	80
Carfentrazone-ethyl+thiobencarb	100	100	100	-	98
Carfentrazone-ethyl+clomazon	100	100	100	-	100
Carfentrazone-ethyl+ pyrazosulfuron-ethyl+ pyriminobac-methyl	100	100	99	-	96
Thiobencarb+simetryn	100	100	98	-	62
Pyrazosulfuron-ethyl+molinate+simetryn	79	89	95	-	33

The applications of either carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone or carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl for *S. pigmaea* showed high weeding efficacy over 90 per cent, but the applications of thiobencarb+simetryn appeared to showed a lower weed control efficacy.

Control of sulfonylurea-resistant weeds at the four leaf stage

Several herbicides were applied to control sulfonylurea resistant weeds at the four-leaf stage (Table 4). The applications of either pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl or thiobencarb+simetryn, for *Monochoria vaginalis*, the application of either pretilachlor, pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone, carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl or thiobencarb+simetryn, for *L. dubia*, the application of either pyrazolate+butachlor, carfentrazone-ethyl+thiobencarb, carfentrazone-ethyl+clomazone or carfentrazone-ethyl+pyrazosulfuron-ethyl+pyriminobac-methyl for *S. juncoides*, and the application of either thiobencarb, esprocarb, mefenacet or pyrazolate+butachlor for *C. difformis* all controlled all sulfonylurea-herbicide-resistant-weeds by over 90 per cent.

Table 4. Weeding efficacy of non-sulfonylurea herbicides on sulfonylurea-herbicide-resistant-weeds applied with at four-leaf-growth-stage.

Herbicides	Weed control efficacy (%)			
	Monochoria vaginalis	*Lindernia dubia*	*Scirpus juncoides*	*Cyperus difformis*
Butachlor	33	89	32	70
Pretilachlor	53	92	85	86
Thiobencarb	55	89	0	97
Esprocarb	5	88	88	96
Mefenacet	74	77	48	94
Fentrazamide	44	87	1	70
Molinate	1	4	0	9
Pyriminobac-methyl	8	1	1	0
Cyhalofop-butyl	1	7	0	4
Dimepiperate	2	4	1	0
Pyrazolate+butachlor	90	95	91	98
Carfentrazone-ethyl+thiobencarb	98	98	96	-
Carfentrazone-ethyl+clomazon	100	98	95	-
Carfentrazone-ethyl+ pyrazosulfuron-ethyl+ pyriminobac-methyl	100	100	92	-
Thiobencarb+simetryn	100	100	9	-
Pyrazosulfuron-ethyl+molinate+simetryn	68	87	73	-

Controlling sulfonylurea-herbicide-resistant-weeds at the middle growth

The resistant biotypes of *S. juncoides* and *C. difformis* survived up to the 30~35 days-growth-stage could effectively be controlled by foliar application of bentazone (1600g a.i. ha^{-1}) only. *M. vaginalis*, *L. dubia* and *S. pigmaea* surviving to the same growth stage could be controlled by the application of the mixtures of bentazone and 2, 4-D (1200+140g a.i. ha^{-1}) or bentazone+MCPA (930+138g a.i. ha^{-1}).

Table 5. Weeding efficacy of foliar-application herbicides applied with at 30-35 days after seeding on sulfonylurea herbicide resistant weeds.

Herbicides	Weed control efficacy (%)				
	Monochoria vaginalis	*Lindernia dubia*	*Scirpus juncoides*	*Cyperus difformis*	*Sagittaria pigmaea*
Bentazone	90	95	100	100	98
2,4-D	100	85	89	81	90
Bentazone+2,4-D	99	100	100	100	-
Bentazone+MCPA	100	100	-	-	100

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MECHANISMS OF COMPETITION BETWEEN BARNYARDGRASS (*Echinochloa crus-galli*) AND REDROOT PIGWEED (*Amaranthus retroflexus*) WITH DRY BEAN (*Phaseolus vulgaris*) USING GROWTH INDICES

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Abstract: An additive experiment was conducted study the mechanisms of competition between barnyardgrass and redroot pigweed with dry bean. The treatments included three levels of redroot pigweed densities (4, 8, 12 plant/m^2) and three levels of barnyardgrass (10, 20, 30 plants/m^2) planted in a dry bean crop with a density of 20 plants/m^2. A weed free control was included in each block. Due to competition by the weeds, total dry matter (TDM), growth rate, height and leaf area of the bean crop decreased significantly. Redroot pigweed had a stronger competitive effect on dry bean than barnyardgrass for all of the above parameters. Except early in the season, season redroot pigweed growth index curves exceeded that of beans. In barnyardgrass, except for the end of the season (during the physiological ripening period of bean) growth index curves were lower than that of beans. Greater height and a better leaf position of redroot pigweed in the mixed canopy from early season resulted in redroot pigweed being more competitive for light than beans. Barnyardgrass had a much weaker competitive ability for light, mainly due to a lower height and an unfavorable leaf position. Apparently, barnyardgrass imposed its competitive effect mainly through root competition, whereas redroot pigweed imposed its competitive advantages primarily through its foliage.

Key words: Barnyard grass, dry bean, red root pigweed

Introduction

Understanding the mechanisms of competition between species can increase the knowledge base about the outcome of competition. This understanding is useful for weed management and can be applied in crop breeding for improved weed suppression (Fonteyn and Mahall, 1981). The success of weeds in farming systems depends on their growth strategy including a high growth rate, strong and efficient root system and good canopy structure (Maun, 1977). For example the species that have greater height, higher leaf area and more horizontal leaves are more successful in absorbing radiation than other species (Holt, 1995). This study was conducted to investigate the mechanisms of competition of barnyardgrass and red root pigweed with dry bean.

Materials and Methods

An experiment was conducted in a Randomized Complete Block Design with three replicates at Mashhad Experimental Station, Ferdowsi University, Iran, in 2001. Treatments included three levels of redroot pigweed densities (4, 8, and 12 plants/m^2) and three levels of barnyardgrass densities (10, 20, and 30 plants/m^2) planted separately in beans (20 plants/m^2). A weed-free control crop was added to each block. Destructive sampling was done weekly and height, leaf area, dry matter and crop growth rate were measured for all species. Leaf area density in three canopy layers (0-25, 25-50, and 50 cm up wards) was determined after canopy closure. The CROP SYS model (Nassiri and Kropff, 1999) was used to study competition for light.

Results and Discussion

Both weeds significantly ($p< 0.01$) reduced total dry matter, growth rate, height and leaf area of beans. Redroot pigweed had a stronger effect on bean than barnyardgrass for all of the above parameters. Barnyardgrass growth index curves were lower than bean except at the end of the season (physiological ripening period of bean). Redroot pigweed growth index curves significantly exceeded that of bean and barnyardgrass except during the early growth period (data not shown). Redroot pigweed established 70 percent of its leaf area above the bean canopy throughout the season. Only at the end of the season, barnyardgrass established 40 percent of its leaf area above the bean canopy (Figure 1).

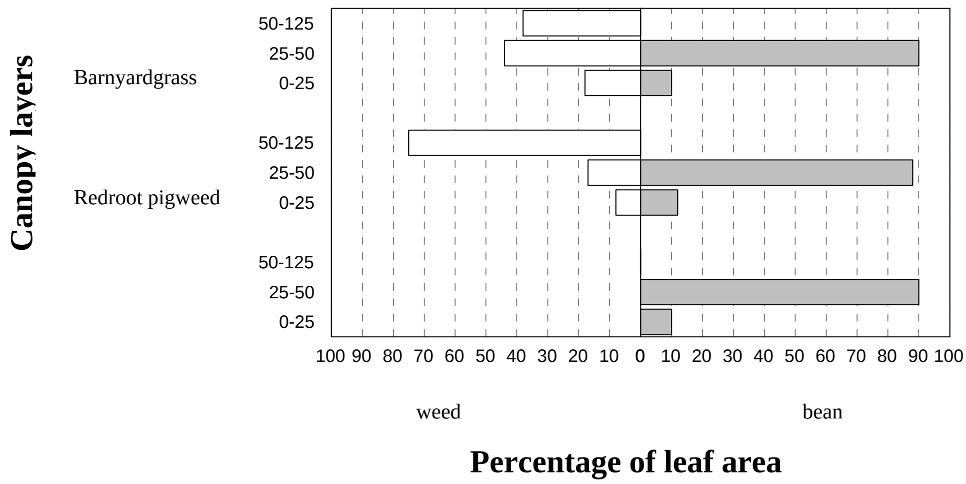

Figure1. Distribution of leaf area in three canopy layers for each species after canopy closure

This indicates that the shoots of barnyardgrass had a lower adverse effect on beans . Thus it may be assumed that barnyardgrass imposed its competitive effects via partitioning of assimilates to the roots (Maun.1977). In redroot pigweed light was an important aspect in its competition with dry bean (Figure 2), which was due to its greater height, its better leaf area position (above the bean canopy) and its horizontal leaves (Holt, 1995).

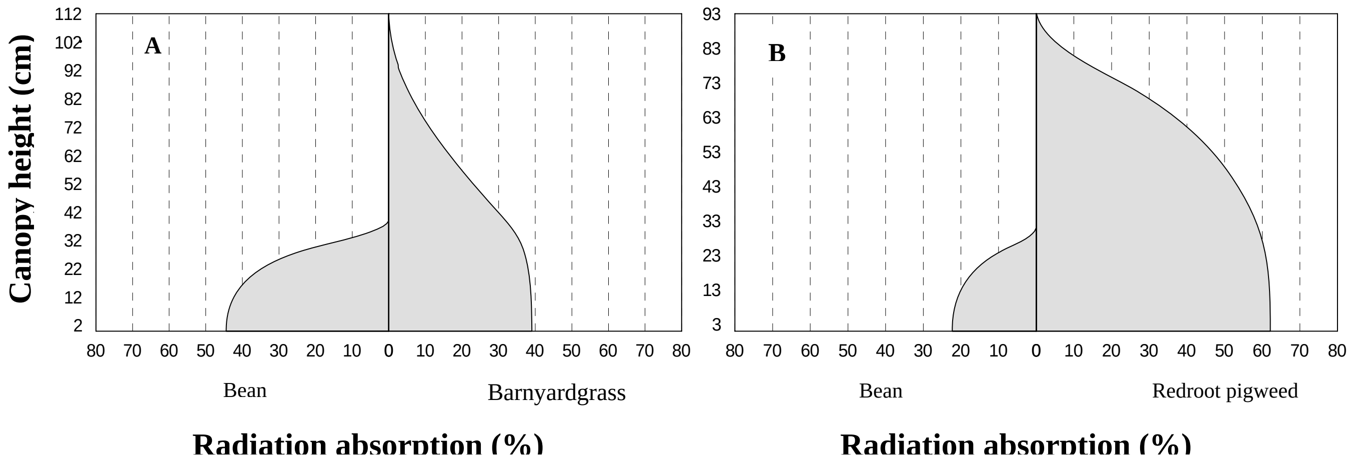

Figure 2. Cumulative radiation absorption in mixed canopies of bean, barnyardgrass (A), and redroot pigweed (B), for each of the species after canopies closures

Results from the CROPSYS model also indicated that redroot pigweed throughout the season absorbed 60 %of the radiation, while that absorbed by dry bean was only 20%. At the end of the season barnyardgrass was able to absorb 45 %of the total radiation.

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COMPOSITION OF THE WEED SEED BANK UNDER INTENSIVE IRRIGATED RICE CULTIVATION: OBSERVATIONS AFTER TWELVE CONSECUTIVE RICE CROPS

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Abstract: A study was conducted of the community structure of the weed seed bank in soil after irrigated rice had been continuously grown for seven-years. The seed bank was examined from plots where wet direct-seeded rice or transplanted rice had been grown and where these had been either kept unweeded or intensively weeded by treatment with herbicide (pretilachlor + fenclorim) and follow up hand weeding. Soil samples taken from plots were separately maintained both in a moist, aerobic condition and a saturated one in a screen house for two years. Germinating weeds were counted as soon as they were identifiable and soil periodically stirred. Data were pooled for analysis. Observation prior to the start of the experiment indicated twenty-one weed species present in rice, *Sphenoclea zeylanica*, *Cyperus difformis*, *C. iria*, *Fimbristylis miliacea*, and *Lindernia anagallis* being dominant. After 12 crops of rice, 31 weed species were recorded. Total cumulative seedling densities did not differ among treatments. *Cyperus difformis*, *C. iria, Echinochloa crus-galli*, *Echinochloa glabrescens*, *F. miliacea, Leptochloa chinensis, Ludwigia hyssopifolia* and *S. zeylanica* were the dominant weeds regardless of crop establishment method and weed control method. Significant differences ($p<0.05$) in seedling densities of *E. glabrescens*, *C. difformis* and *S. zeylanica* were found in soil from direct-seeded and transplanted rice plots where these were intensively weeded, indicative of selective seed loss from the seed bank due to establishment method.

Key words: Seed bank, crop establishment, germination

Introduction

Changes to rice crop management are well known to cause shifts in weed populations (Rao *et al.* 2007). Inter-specific selection of weed species may happen because of interrelated factors associated with tillage practices, fertilizer use, water management, cropping sequence and herbicide use that influence the survival and reproduction of weed populations. Increased herbicide use and changes in crop establishment methods have resulted in changes in weed community composition of rice and the emergence of new weed problems with an increase in grasses in particular (Moody, 1995). Janiya *et al*. (1999) reported that cropping practices resulted in rapid changes in rice weed communities over three successive seasons. *Echinochloa glabrescens* Munro ex Hook. f., was strongly selected by two seasons of transplanted rice, whereas there was a tendency for *Leptochloa chinensis* (L.) Nees, *Fimbristylis miliacea* (L.) Vahl., and *Ludwigia octovalvis* (Jacq.) Raven to be favored by wet direct-seeding. In another study (Janiya and Moody, 1989), after five seasons *E. glabrescens* was dominant in unweeded plots of both transplanted and wet direct-seeded rice, while *Monochoria vaginalis* (Burm. f.) Presl. dominated the hand weeded plots. In a three-year study of irrigated rice in India, Singh *et al*. (2003) recorded that *Ischaemum rugosum* Salisb., *L. chinensis* and *C. rotundus* L. were more abundant in dry direct-seeded rice than in transplanted crops. Contrastingly, in rainfed rice, Mazid *et al*. (2003) noted an increase in abundance of broadleaved species *Alternanthera sessilis* (L.) R.Br. Ex Roem & Schult, *Eclipta prostrata* (L.) L., *Lindernia ciliata* (Colsm.) Pennell, and *Ludwigia octovalvis* (Jacq.) Raven and the sedges *Cyperus difformis* L. and *F. miliacea* in dry-seeded rice while abundance of *M. vaginalis* decreased over a three-year cropping period.

A number of authors have discussed the relationship between the weed community composition in a crop and that of the buried seed bank. In rice, it is well known that the size of the weed seed bank may be considerable (Sahid *et al*. 1995) and that, in farm practice, thorough land preparation by tillage and puddling can reduce the density of weeds emerging during and after crop establishment. Gallandt (2006) argued that because cultivation and herbicide efficacy is generally density-dependent, seedling density following these weed control practices will be proportional to the density of germinable seeds in the seed bank. From a 12 year study of contrasting crop management practices in a corn-wheat-soybean cropping sequence, Davis *et al*. (2005) found that the structure of the weed community of the seed bank was positively correlated with weed biomass in the reduced input (reduced rates of N fertilizer and herbicide) and organic systems, but there was no relationship when conventional and no till systems were examined. Furthermore, they commented that because there was generally low correspondence between below- and above-ground weed communities, the size of the seed bank alone had limited value in predicting long term weed community dynamics. The relative contribution to the weed seedling population from seeds persisting in the seed bank is unknown for most weed species of rice. This is in part because of fragmentary knowledge of seed longevity and factors influencing the flux of seed dormancy. Whilst the water profile at the time of rice crop establishment plays a major role in inter-specific selection of weeds at establishment (Rao *et al.* 2007), the role of cultivation in determining the dynamics of the composition of the weed seed bank in rice fields is poorly researched.

In this paper, we examine the effect of two establishment methods for irrigated rice on weed species abundance and diversity of the seed bank. We hypothesized that the community composition of the weed seed bank would be governed by two processes: a) the effect of cultivation practices in determining differential rates of survival amongst species persisting in the seed bank and b) the relative species contributions to the seed bank through seed reproduction in successive seasons. A long term field trial compared the impact of differing rice establishment methods and enabled these hypotheses to be investigated by quantifying the relative abundance of species in the weed seed bank after 12 cropping seasons in the absence and the presence of the seed return to the soil.

Materials and Methods

As described in Janiya *et al*. (1999), a long term experiment was initiated in June 1997 at the International Rice Research Institute. Previously uncultivated land which from prior survey exhibited 159 plant species was divided into plots and each sown with a mixture of weed species before rice cropping. Species sown were *Cyperus difformis*, *C. iria*, *Echinochloa colona* (L.) Link, *E. crus-galli* (L.) Beauv., *E. glabrescens*, *Fimbristylis miliacea*, *Ischaemum rugosum*, *Leptochloa chinensis*, *Ludwigia hyssopifolia (G. Don) Exell*, and *Sphenoclea zeylanica* Gaertn. In the first season, all plots were uniformly planted to wet direct-seeded rice. Cumulative counts of all weed species that emerged were made by census at 15, 21, 35 and 60 days after crop establishment in two fixed, randomly placed, 50 cm x 50 cm quadrats.

After the harvest of the initial crop, a series of management patterns were experimentally maintained. These included annually either two crops of wet direct-seeded rice (WSR) or transplanted rice (TPR) as main plots with two subplots of weed management treatment, unweeded or intensively weeded (herbicide - pretilachlor + fenclorim applied at 0.45 kg a.i. ha^{-1} 3 DAS followed by handweeding 21 days after transplanting or sowing) (Janiya *et al*. 1999). Land preparation for TPR and WSR plots was similar. The plots were ploughed once, then flooded and harrowed twice and leveled with a power tiller over a period of 14 days. Subsequent flooding regimes differed in that standing water (5-8 cm) was maintained in TPR plots from 4 days after planting, whereas in DSR plots flush irrigation was done in the first 4 days and flooding was applied gradually in relation to growth of rice seedlings, with a similar flooding depth to TPR typically being achieved 10 – 14 DAS. Cropping patterns were maintained for six years being terminated after the 12th cropping season (2 crops per year). Four replicates of each treatment combination were conducted in a Randomized Complete Block Design.

After the harvest of the final crop in 2003, four sampling points were selected at random in each plot after the field had dried. From each, a soil sample was taken using a 20 cm x 20 cm galvanized steel core sampler. Samples were divided into 2 layers (0-2 cm and 2-10 cm). For each sampling point, samples from the same layer of soil were then bulked. Soil was then processed to obtain a relatively fine tilth and samples divided into four replicates per layer and placed in plastic containers for germination. The total amount of soil placed in each container was 355 cm^3. In containers, the soil was 2 cm deep with a surface area of 178 cm^2. The soil in two replicate samples in each layer was maintained in a saturated condition and the other pair in a moist aerobic condition, by careful watering.

Seedling were counted by species and removed from each tray. Once a germination flush had occurred and the seedlings were removed, the soil was stirred, and the appropriate moisture regime was established. Counting resumed when emerged weeds were identifiable and the cycle was repeated. Seedling census started in January 2003 and ended in June 2004. The data collected were pooled across layers for analysis.

Results and Discussion

In total during the first cropping season, twenty-two weed species were recorded in DSR and TPR plots combined. Figure 1 shows variation in the cumulative count of each species recorded in the first season, *S. zeylanica*, *C. difformis*, *C. iria*, *F. miliacea*, and *Lindernia anagallis* (Burm.f.) Pennell being the most frequently recorded species (comprising >10% of total abundance). These data do not reflect the flux in populations during the cropping season and therefore are inaccurate in estimating the relative abundance of each species at any point in the cropping season. These however provide an indication of the common species present.

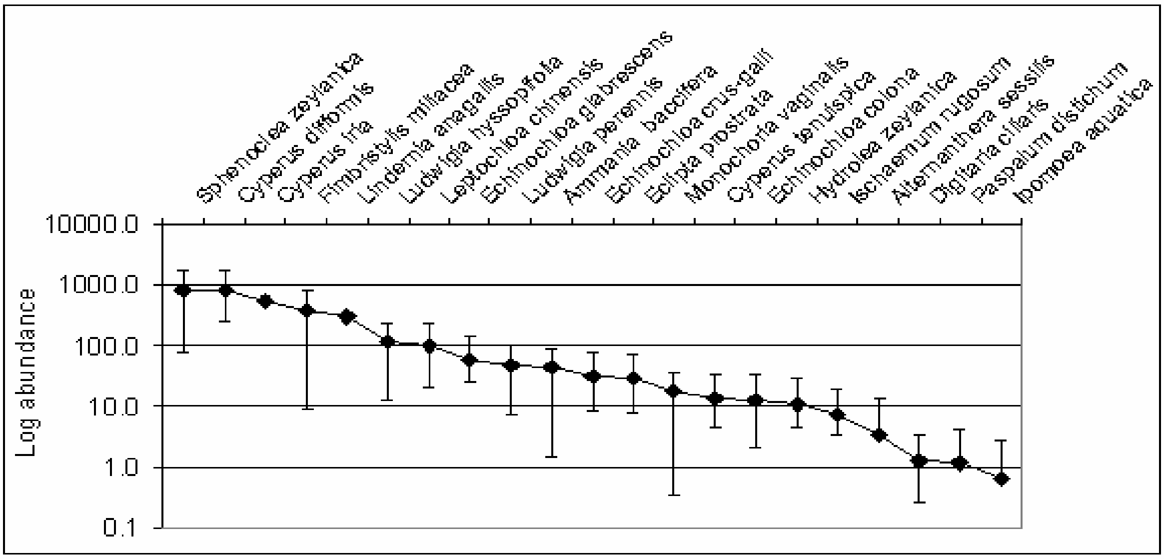

Figure 1. The abundance of weed species before the imposition of cropping sequence and weed control treatments in 1997. Data are the mean cumulative total density of each species, with bars indicating the minimum and maximum records from four replicate plots.

In total, 31 species (19 broadleaved weeds, 8 grasses and 4 sedges) were recorded as seedling emerging from the soil samples, when counts were combined for both aerobic and saturated samples. Total cumulative seedling densities did not differ amongst treatments, the mean total seedling density being $8.4 \pm 0.35\ 10^6\ m^{-3}$. The highest densities were observed for *C. difformis*, *C. iria, E. crus-galli*, *E. glabrescens, F. miliacea*, *L. chinensis, L. hyssopifolia* and *S. zeylanica.* Significant differences ($p \leq 0.05$) in seedling densities from soil from DSR and TPR plots where seed return was prohibited were found for *E. glabrescens*, *C. difformis* and *S. zeylanica.* No significant differences were detected between the treatments for any species in plots where no weed control was practiced and seed return was allowed.

Previous authors (*e.g.* Buhler *at al*. 1997; Cardina and Sparrow, 1996) have pointed out that a challenge remains in utilizing knowledge of the composition of the seed bank in long term strategies of weed management. Whilst there is often a correlation between the abundance of weed seeds returned to the upper surface of soils in one season and the seedling densities in the following season (particularly in zero and minimum tillage systems), this is not always found. Estimates of the size of the seed bank by soil sampling and subsequent seedling census may have low diagnostic power because of spatial heterogeneity in the soil seed bank and the sampling intensity needed to account for this. Moreover the absolute number of seedlings and the rate of seedling emergence relate to flux amongst dormancy states within the seed population of a species and will be environmentally dependent (Thompson and Grime, 1979). In the present study, there were differences between the numbers of seedlings emerging under aerobic and saturated conditions (data not shown) and census counts were pooled on the assumption that seed polymorphisms in germination behavior were present. In terms of crop establishment, the major differences between TPR and DSR cropping systems were in the flooding regimes post seeding or transplanting and the speed of canopy closure. Significantly more seedlings of *E. glabrescens* and *C. difformis* were recovered from TPR as opposed to DSR plots whereas the reverse was true for *S. zeylanica* (Figure 2a, intensively weeded plots).

The inference from this is that water regimes associated with DSR selectively increases the rate of seed loss from the seed bank by germination in *E. glabrescens* and *C. difformis,* immediate flooding in TPR promoting greater loss in *S. zeylanica*. Janiya *et al*. (2004) noted that in unweeded rice plots *E. glabrescens* plant populations increased in abundance under DSR over time whereas *C. difformis* and *S. zeylanica* declined in abundance under DSR over time. No detectable differences in the sizes of seed bank under the two cropping regimes were detected for any species where weeds were not controlled and seed return allowed (Figure 2b), despite noticeable differences in above ground weed community composition (Janiya, 2004). This supports the observation, above, of the lack of linkage between the size of the buried seed bank and the above ground community with the implication that it is the previous season's seed return that contributes most to the subsequent season's weed community.

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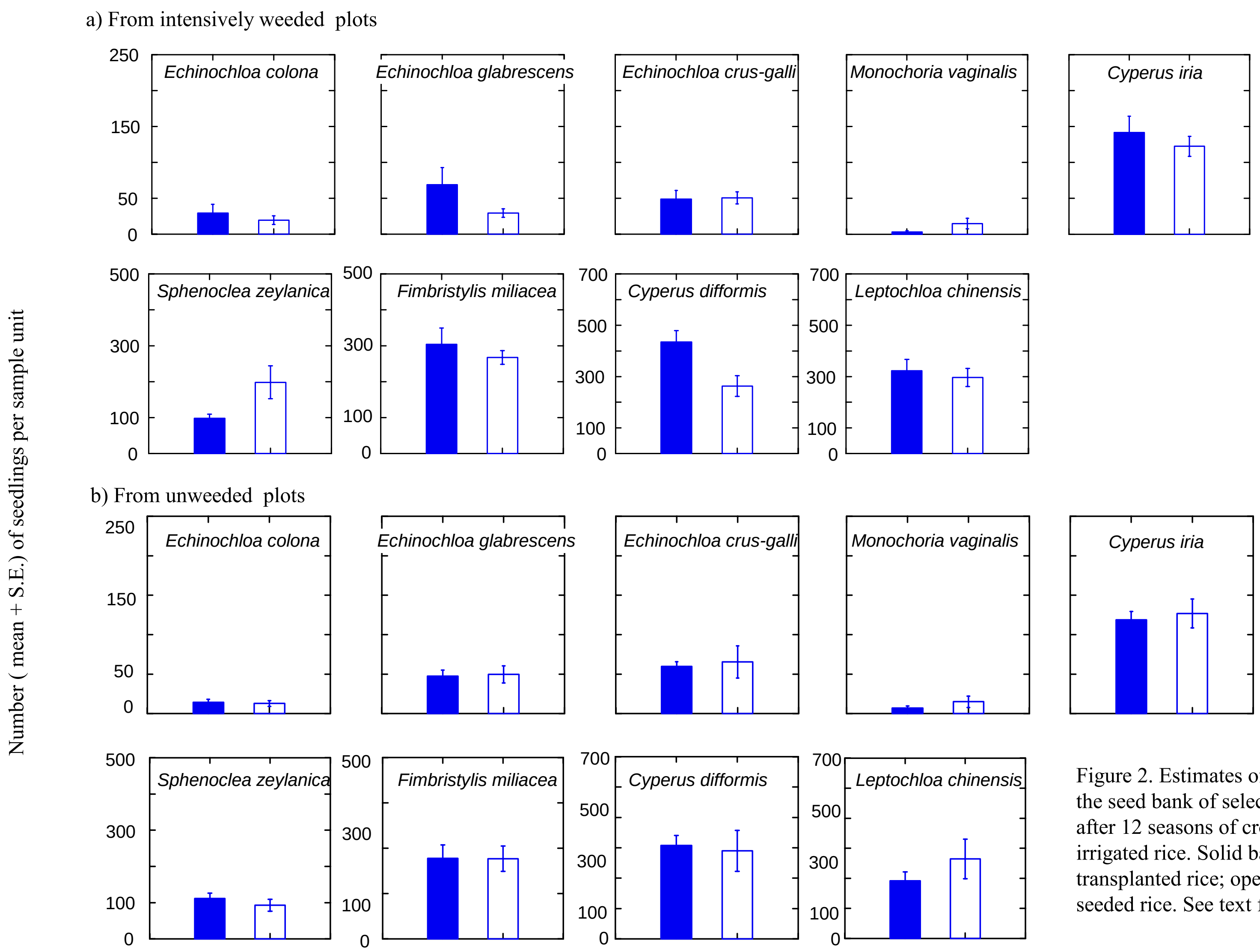

Figure 2. Estimates of the size of the seed bank of selected species after 12 seasons of cropping of irrigated rice. Solid bars, transplanted rice; open bars, wet seeded rice. See text for details.

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CONTROL OF PURPLE NUTSEDGE (*Cyperus rotundus* L.) IN COTTON (*Gossypium hirsutum* L.) WITH COMBINED APPLICATION OF ALLELOPATHIC-SORGAAB (SORGHUM WATER EXTRACT) AND LOWER DOSES OF A PRE-EMERGENT HERBICIDE

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Abstract: Allelopathy can be effective in weed management systems in different ways. One such aspect may be its utilization for reducing the use of synthetic herbicides. Considering its importance as a natural weed control approach an investigation using two doses of Sorgaab (*Sorghum bicolor* L. water extract) 12 and 15 l ha^{-1} combined with lower doses of herbicide (S. metolachlor) *i.e*., ½ and 1/3rd (1075, 717 g a.i. ha^{-1}) of the label dose as a pre-emergent spray against purple nutsedge was undertaken in cotton (*Gossypium hirsutum* L.) under irrigated conditions in central Punjab, Pakistan. S. metolachlor at 2.15 kg a.i ha^{-1} as standard treatment and an untreated treatment were maintained for comparison. Results of two years of field trials revealed that purple nutsedge control was achieved from 62% to 92% when Sorgaab was combined with reduced doses of herbicide. Similarly, purple nutsedge dry weight was reduced from 75 to 88 % with respect to control indicating that Sorgaab with lower S. metolachlor doses was quite effective in suppressing purple nutsedge.

Key words: Cotton, purple nutsedge, S. metolachlor, sorgaab.

Introduction

Allelopathy is a natural phenomenon and may be employed as an alternative weed control technique. It is environmentally safe, can conserve the available resources and may also mitigate the problems raised by synthetic chemicals (Rizvi and Rizvi, 1992; Duke *et al*. 2001). Sorghum (Sorghum *bicolor* L.) is a potent allelopathic crop containing a number of allelochemicals of which fourteen have been reported by Mahmood (2003). These chemicals are highly water soluble, can be released through root exudation, leaching from plants by rain, or decomposition of residues (Rice, 1984). The effects of these allelochemicals depend upon species; concentration (Cheema and Ahmad, 1992), their movement, fate and persistence in soil (Inderjit, 2001). Similar to synthetic herbicides, allelochemicals are also selective (Weston, 1996).

Allelopathic crops may be used in different ways to influence weeds such as surface mulch (Cheema *et al*. 2000), incorporation into the soil (Ahmad *et al*. 1995; Sati *et al*. 2004), spray of aqueous extracts (Cheema *et al*. 2002), rotation (Narwal, 2000), smothering (Narwal and Sarmah, 1996; Singh *et al*. 2003) or mixed cropping or intercropping (Kondap *et al*. 1990; Hatcher and Melander, 2003). In Sorgaab (sorghum *cv*. JS-263 water extract) seven allelochemicals (gallic acid, protocateuic acid, syringic acid, vanillic acid, p-hydroxybenzoic acid, p-coumaric acid, and benzoic acid were identified (unpublished data). Parveen (2000) found caffeic, ferulic, chlorogenic, syringic and vanillic acid from sorghum plant (leaf and stem) water extracts through thin layer chromatography. Haskins and Gorz (1985) identified dhurrin and *p*-hydroxybenzaldehyde in sorghum dry tissues with water.

Pakistan is, by and large, a mono-crop economy. Cotton contributes nearly 10.5% of value addition in agriculture and about 2.4% to GDP and is a significant source of foreign exchange earnings (Government of Pakistan, 2005). In the recent years, for controlling cotton pests the use of synthetic pesticides has made it an expensive crop. Among the pests, weeds inflict heavy losses on cotton yield (13-41 %). The indirect adverse effects of weeds on cotton due to enhanced disease and insect pest problems are significantly higher than for any other crop grown in the country. Purple nutsedge (*Cyperus rotundus* L.) is considered as one of the worst weeds of the world, widely distributed throughout the tropics and subtropics in 52 different crops and in 92 countries (Rao, 2000) and is very common throughout South East Asia (Merita and Moody, 1999). In Pakistan, it is among the most common weeds found throughout the Indus valley during summer season in major field crops such as cotton, sugarcane and maize. Purple nutsedge is highly competitive and causes seed cotton yield reduction by 62-85% as compared with no purple nutsedge control treatments (Bryson *et al*., 2003). It can harbor cotton pests and diseases and reduce irrigation efficiency (Rao, 2000). Usual weed control methods; manual or mechanical, kill only the top growth with little effect on tubers. Very few selective herbicides for purple nutsedge are available. Furthermore, chemical weed control involves safety risks and may enhance environmental pollution. The continuous use of herbicides may cause weed shift and development of weed resistance to herbicides (Zhang, 2003). Dinitroanaline herbicides as pendimethalin and trifluralin are available in Pakistan and are being used by cotton growers for controlling *Trianthema portulacastrum*. This group of herbicides is ineffective against *Cyperus rotundus* while S-metolachlor (Dual Gold 960 EC) is suggested to be effective in controlling all major weeds of cotton including purple nutsedge (Cheema *et al.* 2005b; Jarwar and Baloch, 2005). Sorghum allelochemicals have been reported to have a phytotoxic effect on purple nutsedge (Mahmood and Cheema, 2003).

The organic chemical compounds having herbicidal properties may be combined with allelopathic water extracts at lower doses (Cheema *et al*., 2005a). This, on the one hand, may improve the efficacy of allelopathic extracts and on other hand may provide opportunities for reducing herbicidal doses and hence the cost of weed control could be lowered, promoting sustainable environmental safety. Therefore, it was contemplated in the present studies to evaluate the possibility of using allelopathic Sorgaab in combination with lower (one half and one third of label dose) S. metolachlor doses for getting effective control of purple nutsedge in cotton.

Materials and Methods

A two-year (2003, 2004) field investigation for controlling purple nutsedge in cotton was undertaken at the Research farm of the Department of Agronomy, University of Agriculture, Faisalabad, Punjab, Pakistan under irrigated conditions. The soil belongs to the Lyallpur soil series (Aridisol-fine-silty, mixed, hyperthermic Ustalfic, Haplargid in USDA classification and Haplic Yermosols in the FAO classification scheme). Sorgaab was prepared by following the procedure devised by Cheema and Khaliq (2000). The experiment was laid out in RCBD (Randomized Complete Block Design) with four replications in plots measuring 7 m × 3 m. Sorgaab at 12 and 15 l ha^{-1} was sprayed as pre-emergence alone or tank mixed with one half and one third lower doses of S. metolachlor as a pre-emergent spray. S. metolachlor at 2.15 kg a.i. ha^{-1} (label dose) pre-emergence was used as the standard treatment and the untreated treatment was used as control. The seedbed was prepared by giving two cultivations and planking the field once. The trial was conducted in a field where previous history showed heavy infestation of purple nutsedge. Cotton cultivar FH901 was sown by a single row hand drill in 75 cm spaced rows on moist seedbed. Fertilizer was applied at 115 kg N, 57 kg P_2O_5 ha^{-1} in the form of urea and triple super phosphate, respectively. Sorgaab and S. metolachlor (Dual Gold 960 EC) were applied in the respective plots with volume of spray at 300 l ha^{-1}, using a Knapsack hand sprayer fitted with T.Jet nozzle. All Agronomic operations except those under study were kept normal and uniform for all treatments. Data on purple nutsedge density and biomass were recorded at 15, 45 and 75 days after sowing (DAS) from two randomly selected quadrats (50×50 cm) from each experimental plot and then averaged.

Purple nutsedge dry weight was recorded after drying in an oven at 80°C for 48 h. Data on cotton leaf area index at 60, 90, 120 DAS, cotton plant height (cm), number of bolls per plant, boll weight (g), and seed cotton yield (kg ha^{-1}) were recorded for each plot. Data collected was analyzed by Fisher's analysis of variance technique. Least significance difference (LSD) test was applied at 0.05 probability level to compare treatment means (Steel and Torrie, 1984).

Results and discussions

Density and dry weight of Cyperus rotundus

The population of Cyperus rotundus was significantly inhibited by all treatments as compared to control (Table 1). Sorgaab at 12 or 15 l ha^{-1} sprayed as pre-emergence suppressed the population of purple nutsedge by 31 to 56% and 35 to 52% at 15, 45 and 75 DAS respectively. This suppression is due to the allelopathic activity of Sorgaab against purple nutsedge. Almost similar effects were recorded on dry weight of this noxious weed (Table 2). These findings are in line with work of Cheema *et al*. (2004) who suggested 67% reduction in dry weight of purple nutsedge with Sorgaab. The higher rate of Sorgaab has shown no effect which is contradictory to the earlier findings of Mahmood and Cheema (2003) who stated that a higher rate of allelochemicals was more inhibitory. Effects of Sorgaab at 12 and 15 l ha^{-1} in combination with one half and one third doses of S. metolachlor at 1075 and 717 g a.i. ha^{-1} were more pronounced and statistically equal to the label rate of S. metolachlor at 2.15 kg a.i. ha^{-1} during both years of study indicating the possible reduction in herbicide dose by 50-67%. This supports the objectives of initiating present studies and in line with work of Cheema *et al*. (2003) who found 58-71 % inhibition with Sorgaab combined with reduced rates of S. metolachlor as compared to control.

Table 1. Effect of sorgaab in combination with reduced S-metolachlor rates on purple nutsedge.

Treatments		Purple nutsedge control (%)					Purple nutsedge dry weight reduction (%)		
Extract/ Herbicide (PRE)	Rate	15 DAS		45 DAS	75 DAS		45 DAS	75 DAS	
		2003	2004	*	2003	2004	*	2003	2004
S. metolachlor	215 kg a.i. ha^{-1}	70	72	82	91	79	86	90	83
Sorgaab	12 l ha^{-1}	53	31	41	52	36	34	43	42
Sorgaab	15 l ha^{-1}	44	33	39	51	38	45	47	42
Sorgaab + S. metolachlor	12 l + 1075 g a.i ha^{-1}	69	72	77	92	72	77	88	81
Sorgaab + S. metolachlor	15 l + 717 g a.i ha^{-1}	62	67	73	85	67	75	80	76
Un-treated		0	0	0	0	0	0	0	0
LSD p=0.05		28	13	16	14	12	6	14	9

DAS= Days after sowing, PRE= Pre-emergence, * Mean of the two years, as year effect was non-significant

Table 2. Effect of sorgaab in combination with reduced S-metolachlor rates on leaf area index and leaf area duration

Treatment		Leaf area index			Leaf area duration (days)	
Extract/herbicide (PRE)	Rate	60 DAS	90 DAS	120 DAS	60-90 DAS	90-120 DAS
		*	*	*	*	*
S. metolachlor	2.15 kg a.i. ha^{-1}	2.19 a†	4.38 a	4.32 a	98.57 a	229.00 a
Sorgaab	12 l ha^{-1}	1.36 b	3.27 c	3.39 c	69.54 c	169.43 c
Sorgaab	15 l ha^{-1}	1.37 b	3.17 c	3.33c	68.14 c	165.63 c
Sorgaab + S. metolachlor	12 l + 1075 g a.i ha^{-1}	2.15 a	4.28 a	4.23 a	96.36 ab	224.00 a
Sorgaab + S. metolachlor	15 l + 717 g a.i ha^{-1}	2.18 a	4.04 b	4.00b	93.24 b	213.83 b
Un-treated	-	1.23 b	2.70 d	2.55 d	58.90 d	137.61 d
LSD p=0.05		0.185	0.202	0.191	4.98	8.064

DAS= Days after sowing, PRE= Pre-emergence, †=Means not sharing a letter in common differ significantly at p= 0.05. * Mean of the two years as year effect was non-significant

Cotton growth

Leaf area index (LAI) of cotton at 60, 90 and 120 DAS (Table 3) was significantly affected as compared to control. The LAI at 60 DAS gave similar results among the treatments with label rate of S.metolachlor, ½ and 1/3rd dose of S. metolachlor combined with Sorgaab at 12 and 15 l ha^{-1}. At 90 DAS, the maximum LAI was achieved by label rate of S. metolachlor which was statistically on par with ½ dose of herbicide combined with Sorgaab at 12 l ha^{-1} followed by 1/3rd dose of herbicide combined with Sorgaab 15 l ha^{-1}. At 120 DAS, in the year 2004, the same trend was found as at 60 DAS but in the year 2003, 1/3rd dose of herbicide combined with 15 l ha^{-1} showed less LAI than the label rate and ½ dose of herbicide combined with Sorgaab at 15 l ha^{-1}.

Table 3. Effect of sorgaab in combination with reduced S-metolachlor rates on cotton growth parameters

Treatment		Crop growth rate ($gm^{-2}d^{-1}$)		Plant height (cm)	
Extract/herbicide (PRE)	Rate	60-90 DAS	90-120 DAS		
		*	*	2003	2004
S. metolachlor	2.15 kg a.i. ha^{-1}	5.63 a†	3.11	147.85 a	143.35 a
Sorgaab	12 l ha^{-1}	3.68 b	2.71	139.45 b	131.40 b
Sorgaab	15 l ha^{-1}	4.05 b	2.66	138.70 bc	134.70 ab
Sorgaab + S. metolachlor	12 l + 1075 g a.i ha^{-1}	5.94 a	2.69	147.15 a	141.30 a
Sorgaab + S. metolachlor	15 l + 717 g a.i ha^{-1}	4.68 b	2.85	148.25 a	142.60 a
Untreated	-	2.05 c	2.54	131.70 c	125.65 b
LSD p=0.05		1.283	1.034	7.016	9.704

DAS= Days after sowing, PRE= Pre-emergence, †=Means not sharing a letter in common differ significantly at p=0.05, *Mean of the two years as year effect was non-significant.

Sorgaab alone at 12 and 15 l ha^{-1} showed statistically similar effects but were different as compared to the control. The decrease in LAI (Figure 1) after 90 DAS, during the year 2004 was due to the lower rainfall received during the cropping season (23.91 mm) as compared to the year 2003 (52.56 mm) and there was a shortage of atleast two irrigations during August and September 2004. Pettigrew (2004) also reported that moisture deficit affected cotton growth, yield and yield parameters. Increase in LAI of cotton crop was due to the better weed control that promoted the cotton plant to utilize the resources in a better way and solar radiation without any interference (Irshad and Cheema, 2004).

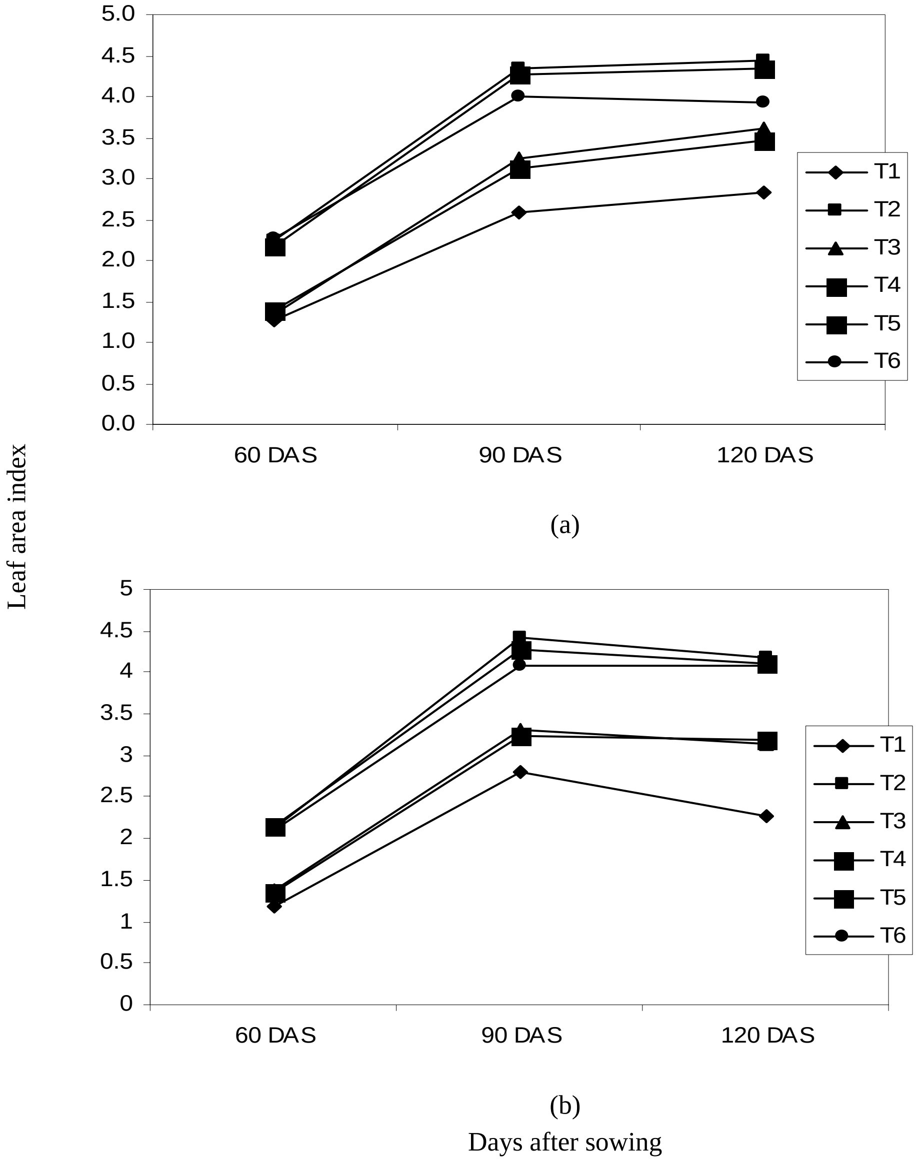

Figure 1. Leaf area indices at 60, 90 and 120 DAS (a) 2003 (b) 2004

Crop growth rate was less after 90 DAS because of shifting of resources to reproductive growth and away from vegetative growth. Data on cotton plant height at maturity revealed that all the treatments significantly enhanced the plant height as compared with control (Table 4). Three treatments, i.e. label rate of herbicide, ½ dose of herbicide combined with Sorgaab at12 l ha^{-1} and 1/3rd dose of herbicide combined with Sorgaab 15 l ha^{-1} showed statistically similar effects. Boquet et *al.* (2004) reported that increases in lint yield were associated with increases in plant height.

Table 4. Effect of sorgaab in combination with reduced S-metolachlor rates on seed cotton yield and its components

Treatment		Sympodial branches (plant^{-1})	Number of bolls (plant^{-1})	Boll weight (g)	Seed index (g)	Ginning out turn (%)	Seed oil contents (%)	Seed cotton yield (kg ha^{-1})
Extract/ Herbicide (PRE)	Rate	*	*	*	*	*	*	*
S. metolachlor	2.15 kg a.i. ha^{-1}	25.10 a†	28.68 a	3.92 a	94.33 a	40.03 a	21.04 a	2006.67 a (33.85)
Sorgaab	12 l ha^{-1}	20.98 b	18.73 b	3.49 b	85.36 b	39.05 abc	19.07 c	1719.17 b (14.67)
Sorgaab	15 l ha^{-1}	20.73 b	18.45 b	3.53 b	83.79 bc	38.81 bc	19.02 c	1698.33 b (13.28)
Sorgaab + S. metolachlor	12 l + 1075 g a.i ha^{-1}	24.08 a	27.50 a	3.98 a	91.29 a	39.96 a	20.46 b	1972.50 a (31.57)
Sorgaab + S. metolachlor	15 l + 717 g a.i ha^{-1}	23.45 a	27.33 a	3.82 a	91.86 a	39.62 ab	20.48 b	1925.00 a (28.40)
Un-treated	-	19.10 b	15.43 b	3.14 c	80.99 c	38.06 c	18.17 d	1499.17 c (0)
LSD p=0.05		2.474	3.837	0.199	3.484	1.092	0.368	118.5

PRE= Pre-emergence, † Means not sharing a letter in common differ significantly at p=0.05, Figure in parenthesis show per cent increase over control. *Mean of the two years as year effect was non-significant

Cotton yield and yield parameters

All the treatments significantly improved the seed cotton yield over control (Table 4). Maximum seed cotton yield was achieved with the application of the label rate of S. metolachlor (34%) which was statistically on par with ½ and 1/3rd dose of S. metolachlor combined with Sorgaab at 12 l ha^{-1} (32%) and 15 l ha^{-1} (28%) respectively. Sorgaab alone at 12 and 15 L ha^{-1} showed statistically similar results and yield was increased by 15% and 13 % respectively as compared to control treatment. The increase in seed cotton yield was possibly due to the better *Cyperus rotundus* control which reduced the weed crop competition. This increased the LAI, number of bolls per plant and boll weight of cotton (Table 4). Boquet *et al.* (2004) reported that increases in lint yield were associated with increases in boll weight and boll number. Cheema *et al*. (2003) also reported the increase in seed cotton yield and concluded that herbicidal dose can be reduced up to 67% when combined with concentrated Sorgaab.

Conclusions

Sorgaab (sorghum water extract) in combination with reduced rates of herbicide S. metolachlor by one half to one third was quite effective in suppressing the density and dry weight of purple nutsedge in cotton and was almost on par with the label rate of the herbicide. This would be helpful in reducing the herbicide usage and thereby promoting environmental safety.

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MANAGEMENT OF *Parthenium hysterophorus* L. IN PAKISTAN

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Abstract. Parthenium weed [*Parthenium hysterophorus* L.] has been rapidly spreading in Pakistan for the last 20 years, especially in the rain-fed districts of northern Punjab, replacing the local flora. Several studies were carried aiming at designing an integrated management program for this noxious alien weed. Seven chemical herbicides namely Chwastox, Atrazine, Ametryn, Butachlor, Glyphosate, Buctril Super and Bromoxynil+MCPA were used at recommended (R) as well as lower rates *i.e.* 0.75R, 0.50R and 0.25R. All the test herbicides were proved effective against the target weed even at the lowest rate tested. In another study, aqueous extracts of the leaves of *Azadirachta indica* (L.) A. Juss., *Ficus bengalensis* L., *Melia azedarach* L., *Mangifera indica* L. and *Syzygium cumini* (L.) Skeels., were evaluated for their herbicidal potential. Leaf extracts of *F. bengalensis* and *M. indica* were found to be highly effective in suppressing the germination and seedling growth of *P. hysterophorus*. In an attempt to search for a biological control agent for the control of *P. hysterophorus,* larvae of a mealy bug species was found to feed on the weed, resulting in complete death of the invader. Recently a Mexican beetle (*Zygogramma bicolorata* Pallister) feeding on Parthenium was observed. Parthenium was also used as green manure for maize crop, where soil amendment with 3% Parthenium green manure resulted in a maize growth equivalent to recommended NPK fertilizers.

Key words: Biological control, chemical control, integrated management, Pakistan, *Parthenium hysterophorous.*

Introduction

Parthenium hysterophorus L. is an aggressive weed of family Asteraceae. It is native to the subtropics of north and south America but has invaded Asia, Africa and Australia during the past 50 years. Since then the weed has not only naturalized itself in many countries but has spread at an alarming rate. Parthenium posses a serious health risk, particularly to the urban populations as it moves into new areas and consolidates established ones. The chemical analysis has indicated that all the plant parts including trichomes and pollens contain toxins called sesquiterpene lactones.

The major components of toxin being Parthenin (Belz *et al*. 2007) and other phenolic acids such as caffeic acid, vanillic acid, anisic acid, chlorogenic acid and parahydroxy benzoic acid are lethal to human beings and animals (Oudhia, 1998). In addition to health hazards, a lot of available data also highlights its impact on agriculture as well as natural ecosystems (Evans, 1997). There are reports of total habitat change in native Australian grasslands, open woodlands, river banks and floodplains due to Parthenium invasion (Chippendale and Panetta, 1994). Similar invasions of national wildlife parks have also been reported in southern India (Evans, 1997).

The weed has been rapidly spreading in Pakistan for the last 15-20 years. It has now become a major wasteland weed and is rapidly replacing the native flora in rain-fed areas of the province Punjab and is also spreading in north western Frontier Province and Kashmir (Javaid and Anjum, 2005). This paper presents various chemical, phytochemical (through allelopathic plant extracts), biological and cultural practices for the management of this noxious alien weed in Pakistan.

Materials and Methods

Chemical control

Parthenium seeds were sown in earthen pots containing sandy loam soil. Three parthenium seeds were sown in each pot and one week after germination, plants were thinned to one plant per pot. Recommended dosages of seven herbicides, namely Chwastox (4 ml l^{-1}), atrazine 38 SC (4 ml l^{-1}), Ametryn 40 EC (10 g l^{-1}), Butachlor 60 EC (8 ml l^{-1}), glyphosate 41 SL (16 ml l^{-1}), Buctril Super (2.5 ml l^{-1}) and bromoxynil + MCPA (4 ml l^{-1}) were selected to evaluate their herbicidal potential to control *Parthenium*. The recommended (R) as well as lower doses *i.e.* ¾R, ½R and ¼R of each of the seven chemical herbicides were sprayed on 5 and 8 weeks old parthenium plants..

Phytochemical control

Fresh leaves of five trees viz. *Azadirachta indica, Ficus bengalensis, Melia azadarach, Mangifera indica* and *Syzygium cumini* were collected from University of the Punjab, Quaid-e-Azam Campus Lahore, Pakistan. After a thorough washing with sterilized water, leaves were dried in an oven at 40°C until constant weight. To obtain a 10% (w/v) aqueous extract, 10 g crushed dry leaf material of each of the five test species was soaked in 100 ml distilled water for 36 hrs at 25°C and filtered. Further dilutions of 8, 6, 4 and 2% (w/v) were prepared by adding appropriate quantity of distilled water to the 10% stock solution. The extracts were stored at 4°C. Seeds of *P. hysterophorus* were sown on a filter paper seedbed in sterilized Petri dishes. The filter papers were moistened with 2.5 ml of aqueous root and shoot extracts of the test allelopathic grasses, and leaf extracts of allelopathic trees. Control was treated similarly with distilled water. There were three replicates of each treatment with 10 seeds per Petri dish. The Petri dishes were incubated at 25 ºC for 7 days. Germination, root and shoot length, and seedling fresh biomass was recorded at the end of the experiment after 7 days. Data were analyzed statistically using Duncan's Multiple Range Test as the mean separation procedure (Steel and Torrie, 1980).

Biological control

Field surveys were carried out in Parthenium growing areas of the Punjab province, Pakistan, from 2003 – 2005 to search for a potential bio-control agent.

Parthenium as green manure

A pot experiment was conducted to use parthenium as green manure. There were six treatments *viz*. control, recommended rates of Nitrogen, Phosphorus and Potassium fertilizers, and 1, 2, 3 and 4% (w/w) Parthenium green manure. Maize was grown as the test crop the biomass of maize was recorded 60 days after sowing, and analyzed statistically using Duncan's Multiple Range Test as the mean separation procedure (Steel and Torrie, 1980).

Results and Discussion

Chemical Control

The seven chemical herbicides used, namely Chwastox, Atrazine, Ametryn, Butachlor, Glyphosate, Buctril Super and Bromoxynil+MCPA were effective against the target weed even at the lowest rate tested, *i.e.* 25% of the recommended rate (Table 1)

Use of Aqueous Extracts

The aqueous extract of all the five test tree species exhibited herbicidal effects against the germination of *P. hysterophorus.* Extracts of *F. bengalensis* and *M. indica* were more inhibitory than the rest, where the extract of the lowest concentration tested (2% w/v) of these two test species significantly retarded the germination of *P. hysterophorus.* The most effective treatment in suppressing germination of the weed was the 10% (w/v) extract of *F. bengalensis* where germination % was 95% less than that observed in the control (Table 2).

Table 1. Days required for complete death of *Parthenium* by the application of different herbicides.

Herbicide	Dose	Days taken for complete death of *Parthenium*	
		5 weeks old plants	8 weeks old plants
Atrazine	R	7 e	12 d
	0.75R	9 cd	12 d
	0.50R	9 cd	12 d
	0.25R	9 cd	12 d
Ametryn	R	11 c	14 c
	0.75R	11 c	14 c
	0.50R	11 c	14 c
	0.25R	11 c	14 c
Bromoxynil+MCPA	R	7 e	7 f
	0.75R	7 e	7 f
	0.50R	7 e	7 f
	0.25R	7 e	7 f
Butachlor	R	13 b	18 b
	0.75R	14 b	20 a
	0.50R	14 b	20 a
	0.25R	16 a	20 a
Glyphosate	R	8 d	12 d
	0.75R	10 c	12 d
	0.50R	10 c	12 d
	0.25R	10 c	12 d
Chwastox	R	8 de	8 ef
	0.75R	8 de	8 ef
	0.50R	8 de	8 ef
	0.25R	9 cd	9 e
Buctril Super	R	4 f	4 g
	0.75R	4 f	4 g
	0.50R	4 f	4 g
	0.25R	4 f	4 g

Within a column, means followed by the same lowercase letter are not significantly different by the Duncan's Multiple Range Test (p-0.05).

The aqueous leaf extracts of *M. azaderach* used at 2-10 (w/v) concentrations were proved most effective in reducing both plumule and radicle length of *P. hysterophorus* seedlings. The toxicity of the aqueous leaf extracts tested increased with increasing concentrations (Table 2). Among the rest of the tree species, extracts of *F. bengalensis* and *S. cumini* were effective in retarding plumule and radical length, where all except 2% (w/v) aqueous leaf extracts of these tree species significantly suppressed plumule length. Similar impact of *F. bengalensis* extracts was also recorded on radical length. However, all the aqueous leaf extracts significantly declined the radical length of test weed. The effect of aqueous leaf extract of *M. azaderach, S. cumini* and *F. bengalensis* on seedling biomass of *P. hysterophorus* was similar to that of their effects on plumule and radical length (Table 2).

Table 2. Effect of aqueous root and shoot extract of five plant species on germination and early seedling growth of Parthenium.

Plant species	Treat. (% w/v)	Shoot extract				Root extract			
		Ger.* %	Shoot Length (cm)	Root Length (cm)	Fresh wt. (mg)	Ger.* %	Shoot Length (cm)	Root Length (cm)	Fresh wt. (mg)
Control	0	100a	1.85b	1.9a	8.2a	100a	1.85a	1.9ab	8.2a
Azadirachta indica	2	100a	2.11a	1.11de	7.4ab	100a	1.18bc	1.97a	7.7a
	4	100a	2.11a	0.99ef	7.6ab	100a	0.93bcd	1.64b	5.8cde
	6	89a	1.81bc	0.96f	5.98cde	70cd	0.89cde	1.33c	4.5gh
	8	72b	1.69bcd	0.95f	6.16cd	75bc	0.70def	1.10cde	4.0hi
	10	48c	1.37e	0.87f	5.0efg	55efg	0.60ef	0.80g	3.2ij
Ficus bengalensis	2	75b	1.67bcd	1.76ab	6.3cd	70cd	1.79a	1.85ab	7.58ab
	4	55c	1.41e	1.13de	5.1efg	76bc	1.54a	1.31cd	6.6bc
	6	60bc	1.43e	0.97f	4.9fg	64cdef	1.24b	0.90fg	5.7cdef
	8	25d	1.11f	0.85f	3.8hi	60defg	1.02bcd	0.97efg	5.4defg
	10	5e	0.10g	0.24h	0.94k	52fg	0.95cde	0.85fg	4.73fgh
Melia azadarach	2	91a	1.8bc	1.72b	6.6bc	85b	1.77a	1.93a	8.0a
	4	65bc	1.6cde	1.52c	4.6gh	65cde	1.74a	1.81ab	6.3cd
	6	65bc	1.35e	1.45c	3.6i	58defg	1.22b	1.65b	4.8efgh
	8	42c	0.16g	0.37h	2j	35h	0.37fg	0.80g	2.8jk
	10	38cd	0.11g	0.10i	0.0.k	27h	0.14gh	0.32hi	0.9l
Mangifera indica	2	60bc	1.77bc	1.87ab	8.1a	60defg	1.81a	1.93a	8.5a
	4	60bc	1.51de	1.51c	6.4cd	50g	1.68a	1.64a	7.8a
	6	35d	0.97f	0.63g	5.6defg	37h	1.22b	1.06def	5.8cde
	8	20d	0.0g	0.0i	0.0k	10i	0.45fg	0.1ij	2.0k
	10	25d	0.0g	0.0i	0.0k	00i	0.0h	0.0j	0.0l
Syzygium cumini	2	99a	1.38e	1.80ab	6.2cd	85b	1.8a	1.70ab	7.5ab
	4	84a	45bcd	1.61d	1.25cde	50g	1.68a	1.36c	6.4cd
	6	69b	1.06f	0.31h	2.6j	10i	0.8de	0.53h	2.3jk
	8	50c	0.0g	0.0i	0.0k	00i	0.0h	0.0j	0.0 l
	10	53c	0.0g	0.0i	0.0k	00i	0.0h	0.0j	0.0 l

*Ger – germination. In each column values followed by the same letter are not significantly different by the Duncan's Multiple Range Test (p= 0.05).

The aqueous leaf extracts of *A. indica* were the least toxic on both plumule and radical length of the weed, however, seedling biomass of Parthenium was significantly reduced by the 10% (w/v) extract of *A. indica* (Table 2). Similarly, extracts of *M. indica* failed to retard the plumule and radical length, as well as seedling biomass of the weed. Conversely the lower concentrations of 2-6% (w/v) of this test tree species promoted the seedling growth of Parthenium (Table 2). This study reveals that the aqueous leaf extracts of *M. azaderach F. bengalensis* and *S. cumini* are highly effective against germination and growth of Parthenium and can be used to control this noxious weed.

Biological control

Recently, in 2005 attack of the Mexican beetle *Zygogramma bicolorata* on Parthenium leaves was observed in different parts of Lahore (capital of province Punjab), about 100 km away in Chhanga Manga Forest. Both the adult and larvae of the beetle were found to feed on leaves of Parthenium. However, the defoliation of Parthenium by the beetle was not found possibly because of low population of this insect at present. The beetle was introduced to the neighbouring country India in 1984 (Jayanth, 1987), and probably have entered Pakistan via

India. During the period October-November 2005, heavy infestation of a mealy bug species feeding on leaves, stem and flower heads of Parthenium were observed by the authors. The infected plants first showed symptoms of dieback and ultimately dried to death. (Plate1). Among more than 260 phytophagous arthropods species reported from *Parthenium* from its native homeland, only 144 species have actually fed on the weed (McClay *et al.* 1995). However, there were no earlier reports of plant pathogen or insect pest feeding on this invasive plant.

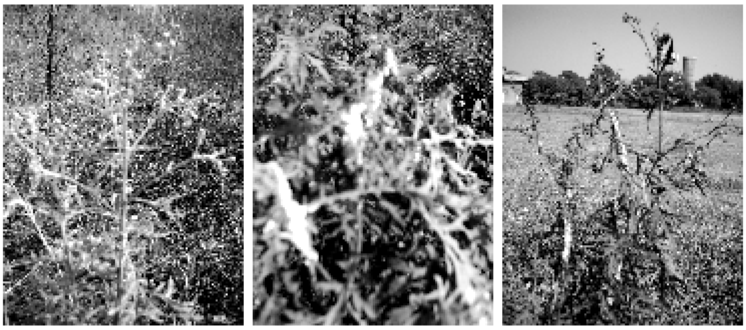

Plate 1. Different stages of damage of *Parthenium* by mealy bug (white patches show the population of mealy bug)

The mealy bug was also found feeding on four other weed species namely, *Achyranthes aspera* L., *Malvestrum tricuspidatum* A. Gray, *Sida spinosa* L. and *Xanthium strumarium* L. However, none of the field crops of grown in the season in the area *viz.* rice (*Oryza sativa* L.), maize (*Zea mays* L.) and sorghum (*Sorghum bicolor* L.) were attacked by the insect larvae. Further studies regarding the identification, ecology and biology of the mealy bug are underway.

Parthenium as a green manure

Shoot length in maize was significantly increased both by the recommended dosages NPK fertilizers and 4% (w/w) rate of the green manure of Parthenium (Figure 2A). All the green manure and the NPK treatments significantly enhanced the shoot dry biomass. A gradual increase in shoot biomass of maize was observed with the increase in Parthenium green manure quantity up to 3% (w/w) and a decline was recorded thereafter. Maize plants treated with 3% (w/w) of Parthenium green manure showed a greater shoot biomass when compared to the recommended NPK fertilizers treatment (Figure 2B). Sudhakar (1984) has also reported an enhanced growth of rice due to application of Parthenium green leaf manure. In contrast to the findings of the present study, Singh *et al.* (2005) reported adverse effects of Parthenium residues on growth of *Brassica campestris, B. oleracea* and *B. rapa.* Similar adverse impacts of Parthenium residues have also been reported by Batish *et al.* (2002a) on growth of *Cicer arietinum* and *Raphanus sativus.*

Parthenium has been reported to spread rapidly so a single control method is not adequate to stop its further spread in the country. It can only be managed effectively by developing an integrated approach involving many options in combination such as the use of effective herbicides, development of environment friendly new herbicides, introduction of biological control agents, and adapting cultural practices.

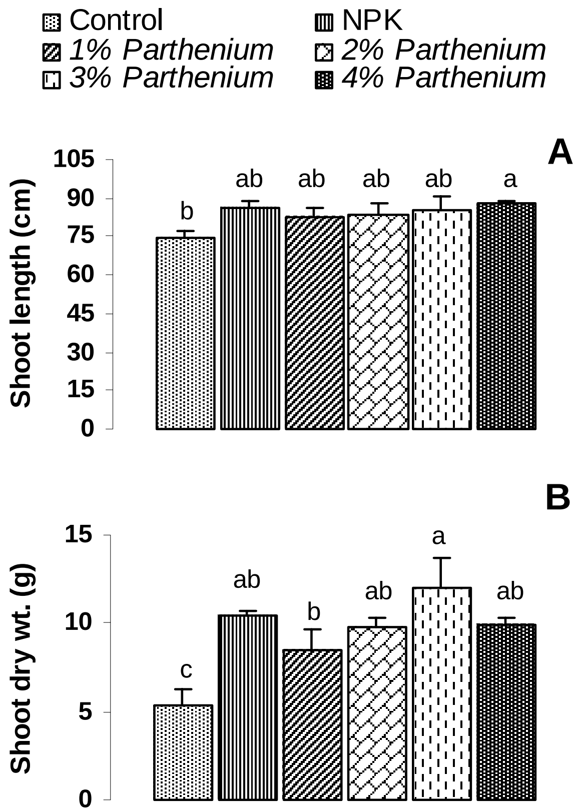

Figure 2. Effect of NPK fertilizers and Parthenium green manure on growth of maize. Vertical bars show the standard error of means of three replicates. Values with different letters are significantly different ($p < 0.05$) by the Duncan's Multiple Range Test.

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SCREENING OF DIFFERENT FIELD CROP VARIETIES FOR THE INFECTION OF *Cuscuta chinensis* Lam. UNDER FIELD CONDITIONS

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Abstract: Thirty two varieties of nineteen field crops were evaluated to study their resistance for the infection of *Cuscuta chinensis* Lam. which is a holoparasitic, invasive weed. Crop seeds were planted on soil beds at the size of 1.50 m x 0.75 m arranged in a Randomized Complete Block Design and replicated twice. The resistant check and the susceptible check used for the experiment were maize (*Zea mays* var. Badra) and red onion (*Allium cepa* var. Vedalan), respectively. Ten (10) cm long cuttings of the parasite were entwined on crop seedlings. Entwining habit, formation of haustoria, growth of the parasite on the host and symptoms of damage were used as criteria to determine disease resistant crops / varieties. Bittergourd (*Momordica charantia* var. MC 43), greengram (*Vigna radiata* var. MI 5 and Ari), soybean (*Glycine max* var. Pb 1 and Pm 13), blackgram (*Vigna mungo* var. MI 1), green chili (*Capsicum annum* var. MI 1, MI 2, MI Hot, KA 2 and Arunalu), spinach (*Spinacia oleracia* var. Yoda), tomato (*Lycopersicon esculentum* var. Donna 091), brinjal (*Solanum melongena* var. SM 164), water spinach (*Ipomoea aquatica* var. Chai tai), cabbage (*Brassica oleracea* var. White cabbage) and cucumber (*Cucumis sativus* var. LY 58), carrot (*Daucas carota* var. Kuroda) were found to be susceptible for the infection. Groundnut (*Arachis hypogoea* var. Walawa and Indi), raddish (*Raphanus sativus* var. Bigball and Beralu) and okra (*Abelmoschus esculentus* var. Haritha and Sudu bandakka) were resistant for the infection. However, the parasite grew and flowered on cowpea (*Vigna ungiculata* var. MI 35, Dawala, Waruni and Wijaya) and vegetable cowpea (*Vigna ungiculata* subspp. *sesquipedilis* var. Polon mae and Ac 00555) showing different responses for the infection, but the vine and haustoria died after few days indicating that these crops were tolerant for the infection.

Key words: Cuscuta, parasitic weeds, resistant and tolerant crops

Introduction

Cuscuta chinensis Lam. is a holoparasitic invasive weed belonging to the family Convolvulaceae and sub family Cuscutoidea (Dassanayaka and Fosberg, 1980). It grows parasitically on different crops (Dawson *et al*. 1994) and a large number of weeds (Wijesundara *et al*. 2001). It also shows a seed dormancy and exists in the environment for a long period. Therefore, control of the parasite is very difficult in an agricultural field where susceptible perennial crops are grown. Crop rotation using disease resistant crop varieties is a good controlling method for such situations (Quadrai *et al*. 1985). Different field crop varieties were screened to determine disease resistant crop / varieties for infection of *Cuscuta chinensis* Lam. under field conditions.

Methodology

Experiment was conducted at the National Plant Quarantine Service (NPQS), Katunayake, Sri Lanka in 2005/2006 *Maha* season (main cultivation season; Ocotber to February). Seeds of different varieties of spinach, tomato, brinjal, water spinach, bittergourd, greengram, soybean, blackgram, groundnut, cowpea, vegetable cowpea, green chili, cabbage, okra, radish and cucumber were planted on 1.50 m x 0.75 m beds arranged in a Randomized Complete Block Design and replicated twice. Red onion (*Allium cepa*) and maize (*Zea mays*) were used as susceptible and resistant crops, respectively. Two to three weeks old seedlings of each variety were infected by entwining 15-20 cm long stem cuttings of *Cuscuta chinensis* Lam. (2 -3 cuttings/seedling). Introduction of the parasite was repeated three times for a successful infection. The entwining habit, penetration of host's tissues by haustoria, increase of the vine length of the parasite and symptoms of host's were observed within 2 – 3 weeks. The penetration of haustoria in host's tissues was studied using cross sections of the host's stem and observing under the light microscope. Positive marks were given when the parasite grew on hosts' tissues showing the above activities and negative marks were given when the parasite did not show such growth on hosts (Table 1).

Results and Discussion

Three types of crops / varieties (susceptible, resistant and tolerant) could be identified (Table 1). If the parasite showed the entwining habit on the host's body, penetration of haustoria in host's tissue, increasing the wine length and if the host showed yellowing and stunted growth, such hosts have been identified as susceptible to the parasite. They were bitter gourd (*Momordica charantia* var. MC 43), green gram (*Vigna radiata* var. MI 5 and Ari), soybean (*Glycine max* var. Pb 1and Pm 13), black gram (*Vigna mungo* var. MI 1), green chili (*Capsicum annum* var. MI 1, MI 2, MI Hot, KA 2 and Arunalu), spinach (*Spinacia oleracia* va. Yoda), tomato *Lycopersicon esculentum* var. Donna 091), brinjal (*Solanum melongena* var. SM 164), water spinach (*Ipomoea aquatica* var. Chai tai), cabbage (*Brassica oleracea*, var. White cabbage), cucumber (*Cucumis sativus* var. LY 58) and carrot (*Daucas carota*, var. Kuroda).

If the parasite did not show the entwining habit, penetration of haustoria in host's tissue, increasing the vine length and if the host showed stunted growth such hosts have been identified as disease resistant hosts. They were groundnut (*Arachis hypogoea*, var. Walawa and Indi), raddish (*Raphanus sativus* var. Bigball and Beralu) and okra (*Abelmoschus esculentus* var. Haritha and Sudu bandakka). Although the parasite showed the entwining habit on okra varieties (Haritha and Sudu bandakka) they did not follow the next two steps (penetration of haustoria and increase of vine length). Therefore, both okra varieties were resistant for the infection.

However, cowpea (*Vigna unguiculata* var. MI 35, Dawala, Waruni and Wijaya), vegetable cowpea (*Vigna unguiculata* subsp. *sesquipedilis* var. Polon mae and Ac 00555) showed a different infection response. In this case, the parasite grew and flowered on the host but after a few days, the vine and haustoria dried and died. This means that the parasite does not spread further on the host. Therefore, they were categorized as tolerant crops. As the parasite produces flowers and seeds on tolerant crops, such a situation helps to spread the parasite and such crops cannot be taken for crop rotation. However, the host-parasite relationship for the tolerant crops should be further studied.

Table 1. Screening of different field crop varieties for the infection of *Cuscuta chinensis* Lam. under field conditions in *Maha* 2005 / 2006.

Family and Botanical name	Crop and Variety	Entwining habit	Haustorial entry	Increase of vine length	Symptoms/ reaction of the host	Overall assessment
Alliaceae						
Allium cepa	Red onion var. Vedalan (Susceptible check)	+	+	+	Yellowing & stunted; finally died	Susceptible
Apiaceae						
Daucas carota	Carrot var. Curoda	+	+	+	Yellowing & stunted	Susceptible

Table 1. continued

Family and Botanical name	Crop and Variety	Entwining habit	Haustorial entry	Increase of vine length	Symptoms/ reaction of the host	Overall assessment
Brassicaceae						
Brassica oleracea	Cabbage					
	var. White cabbage	+	+	+	Yellowing & stunted	Susceptible
Raphanus sativus	Raddish					
	var. Bigball	(-)	(-)	(-)	No change in colour & growth	Resistant
	var. Beralu	(-)	(-)	(-)	No change in colour & growth	Resistant
Chenapodiaceae						
Spinacia oleracea	Spinach					
	var. Yoda	+	+	+	Yellowing & stunted	Susceptible
Convolvulaceae						
Ipomoea aquatica	Water spinach					
	var. Chai tai	+	+	+	Yellowing & stunted	Susceptible
Cucurbitaceae						
Cucumis sativus	Cucumber					
	var. LY 58	+	+	+	Yellowing & stunted, finally died	Susceptible
Momordica charantia	Bittergourd					
	var. MC 43	+	+	+	Yellowing & stunted, finally died	Susceptible
Fabaceae						
Arachis hypogoea	Groundnut					
	var.Walava	(-)	(-)	(-)	No symptoms	Resistant
	var. Indi	(-)	(-)	(-)	No symptoms	Resistant
Glycine max	Soybean					
	var. Pb-1	+	+	+	Yellowing & stunted, finally died	Susceptible
	var. PM 13	+	+	+	Yellowing & stunted, finally died	Susceptible
Vigna radiata	Greengram					
	var.MI 5	+	+	+	Yellowing & stunted	Susceptible
	var.Ari	+	+	+	Yellowing & stunted	Susceptible
Vigna mungo	Blackgram					
	var. MI6	+	+	+	Yellowing & stunted	Susceptible
Vigna unguiculata	Cowpea					
	var. Dawala	+	+	+	Vine and haustoria died after few days	Tolerant
	var. Wijaya	+	+	+	Vine and haustoria died after few days	Tolerant
	var. MI 35	+	+	+	Vine and haustoria died after few days	Tolerant
	var. Waruni	+	+	+	Vine and haustoria died after few days	Tolerant
Vigna unguiculata subsp. *sesquipedalis*	Vegetable cowpea					
	var. Ac 00555	+	+	+	Vine and haustoria died after few days	Tolerant
	var. Polon Mae	+	+	+	Vine and haustoria died after few days	Tolerant

Table 1. continued.

Family and Botanical name	Crop and Variety	Entwining habit	Haustorial entry	Increase of vine length	Symptoms/ reaction of the host	Overall assessment
Malvaceae						
Abelmoschus	Okra					
esculentus	var. Haritha	+	(-)	(-)	No symptoms	Resistant
	var. Sudu bandakka	+	(-)	(-)	No symptoms	Resistant
Poaceae						
Zea mays	Maize					
	var. Badra (Resistant check)	(-)	(-)	(-)	No symptoms	Resistant
Solanaceae						
Capsicum annum	Green chili					
	var. MI 1	+	+	+	Yellowing & stunted growth	Susceptible
	var. MI 2	+	+	+	Yellowing & stunted growth	Susceptible
	var. MI Hot	+	+	+	Yellowing & stunted growth	Susceptible
	var. KA 2	+	+	+	Yellowing & stunted growth	Susceptible
	var. Arunalu	+	+	+	Yellowing & stunted growth	Susceptible
Lycopersicon	Tomato					
esculentus	var. Donna 091	+	+	+	Yellowing & stunted growth	Susceptible
Solanum	Brinjal					
melongena	var. SM 164	+	+	+	Yellowing & stunted growth	Susceptible

Conclusions

Groundnut (*Arachis hypogoea*, var. Walawa and Indi), raddish (*Raphanus sativus*, var. Bigball and Beralu) and okra (*Abelmoschus esculentus* var. Haritha and Sudu bandakka) were resistant for the infection. Bitter gourd (*Momordica charantia* var. MC 43), green gram (*Vigna radiata* var. MI 5 and Ari), soybean (*Glycine max* var. Pb 1and Pm 13), black gram (*Vigna mungo* variety MI 1), green chili (*Capsicum annum* var. MI 1, MI 2, MI Hot, KA 2 and Arunalu), spinach (*Spinacia oleracia* var. Yoda), tomato (*Lycopersicon esculentum* var. Donna 091), brinjal (*Solanum melongena* var. SM 164), water spinach (*Ipomoea aquatica* var. Chai tai), cabbage (*Brassica oleracea* var. White cabbage), cucumber (*Cucumis sativus* var. LY 58) and carrot (*Daucas carota* var. Kuroda) were susceptible while Cowpea (*Vigna unguiculata* var. MI 35, Dawala, Waruni and Wijaya), vegetable cowpea (*Vigna unguiculata* subsp. *sesquipedilis* var. Polon mae and Ac 00555) were tolerant for the infection *Cuscuta chinensis* Lam. under field conditions.

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BIOLOGICAL CONTROL OF WEEDS USING GOATS AND ITS' IMPACT ON WEED SEED BANK IN MAIZE

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Abstract: Field and green house experiments were done to reveal the impact of integrating goats as farming components on weed competition, crop performance and weed seed bank of maize (*Zea mays* L). Use of goats to graze on the weeds during the off-season prior to planting of maize, addition of goat manure to the field throughout the year and a combination of both grazing and addition of manure were compared with a normal crop of maize without any involvement of goat rearing as a farm component, in the same field over a period of three years. It was observed that allowing the goats for repeated grazing on the weed growth during the off season prior to planting of maize showed the highest potential to control weeds in maize and to deplete the soil seed bank appreciably after three years. The other two treatments involving manure addition + grazing and manure addition alone were next in order. However, addition of manure alone was still better than a normal crop without goat integration. The weeds that were controlled and whose seed reservoir was depleted significantly include *Cyperus rotundus* L. *Trianthema portulacastrum* L. *Echinochloa colonum* Link and *Phyllanthus niruri* L.

Key words: Biological weed control, goats, integration

Introduction

Farming system approach was observed to be a resource management strategy for achieving economic and sustainable agricultural production and to meet the diverse requirement of farm household while preserving the resource base and maintaining high environmental quality (Rangasamy, 1994). A judicious mix of any one or more activities like dairy, biogas, mushroom, silkworm rearing and apiary with crop production was reported to help in effective recycling of various by-products increasing gainful employment and income of small and marginal farmers. Though, herbicides offers selective and economic weed control they need to be integrated with other cultural and biological options considering the environmental degradation in terms of weed shift and residue hazards consequent to the exclusive use of herbicides (Kathiresan, 2007). Therefore, with recent emphasis on reduced herbicide use, alternative options need to be explored for tackling persistent weed problems. Small ruminant sector plays an important role in the natural economy where they are very useful in semiarid and arid zones where they can sustain themselves on sparse vegetation and extreme climatic conditions. Goat rearing gains importance mainly on account of the short intervals, higher rates of prolificacy and the ease with which they can be marketed. Weed control could be obtained by goat grazing since they prevent the annual replacement of seed reserves and that very few seeds ingested by goats remain viable (Tim Johnson, 1994). Goat grazing reduced the infestation of spotted knapweed and seed head production by the weed (Williams and Prather, 2003). Hence, the impact of integrating goat rearing in rainfed millet cultivation was explored with emphasis on weed control supplement.

Materials and Methods

Field experiments were taken up at Annamalai University Experimental farm, Tamil Nadu, India from 2003 for two years in the same field comparing different modes of integrating goat component in maize crop under rainfed conditions. The experimental field was predominated by perennial weeds *Cyperus rotundus L.*, *Cyanodon dactylon* Perse. Along with rare occurrence of annuals like *Trianthema portulacastrum L.* and *Echinochloa colonum L.* The fields were segmented to four main plots of dimension 15 x 24 m^2 fenced with naturally available semi-permanent fencing materials and goats were integrated in these segments for different purposes. These include grazing alone, goat manuring alone, and grazing + manuring that were compared as main treatments along with a fallow as untreated control during the off season (Jan – June) under split plot design. The goats of local breed were allowed to graze in the field at a stocking rate of 10 goats per hectare in the treatment plots that involve goat-grazing and their voidings during day and night were collected and incorporated in the treatment plots that involve goat manuring. Laboratory and field experiments were taken to trace the role of various modes of goat integration on the soil weed seed bank. In the laboratory study taken up to trace the impact of goat integration on the soil weed seed bank, soil samples were taken from the individual plots in which the various modes of goat integration were tried. The collected soil were spread on shallow trays (holding a kg of the individual soil sample) and left undisturbed and exposed to sun with optimum soil moisture. Individual weed seed germination was recorded cumulatively up to seven days. After 15 days, the germinated weed seeds were uprooted and the soil was treated with GA_3 to induce the dormant seeds for germination and was recorded up to seven days. The seed germination was recorded for every kg of soil and computed to seeds reserve per hectare.

During the crop season that followed grazing by goats, maize crop (*Zea mays* L) was sown with hybrid Bhanu during 2003 and composite CO1 during 2004 adopting spacing of 60 cm x 30 cm and 60 cm x 20 cm, respectively. Each segment accommodating goat integration was further subdivided into subplots wherein maize was raised with different weed control treatments. The sub treatments comprised of an unweeded control, hand weeding (twice), pre emergence alachlor 1.5 kg ha^{-1}, intercrop of blackgram, alachlor + hand weeding and alachlor + intercrop. An additive intercrop of mungbean, cultivar ADT 5 was raised in a row in between two rows of maize during the experimental years in treatment plots that accommodated intercropping. Pre emergence herbicide alachlor (Lasso 50 EC) was sprayed with a floodjet knapsack sprayer fitted with flood jet deflector nozzle using 500 l of spray fluid and 12 psi of pressure on the 3rd day after crop sowing in respective treatment plots. Observations recorded included weed counts and weed biomass on 60 days after treatment (DAT), weed control index and grain yield at harvest. The data recorded on weed seed bank and field experiment were statistically analysed to draw the standard error difference and ultimately the critical difference was worked out at 0.5 % probability as suggested by Panse and Sukhatme (1978). The data on weed biomass was recorded in gm^{-2} and weed control index was worked out by the formula suggested by Thakur and Sharma (1996).

Results and Discussion

Goat integration in rainfed agriculture as a farming element along with maize was observed to influence the weed infestation and crop performance in millets raised subsequently. Grazing by goats in the off season reduced the weed infestation during cropping season and weed suppression was significantly superior to all the other main treatments compared (Table 1). Grazing on the annual weeds in their vegetative stage, without allowing them to set seeds, thereby blocking the enrichment of weed seed bank contributed for suppression of annual weeds. As regards to perennial weeds, repeated grazing led to the exhaustion of food reserves in the underground propagules that reflected on the reduced infestation during cropping seasons. Though the inclusion of goats had reduced the weed population in subsequent seasons through their feeding habit, addition of fresh goat manure brought down the weed control effect slightly by favouring a higher weed count and biomass especially with annuals, when compared to integrating goats for grazing alone. This is attributed to the re-infestation by annual weed seeds through the goat manure by virtue of the process of endozoochory. However, the weed control advantage in goat manuring compared to untreated control might be due to reduced soil pH and reduced recuperation of soil weed seed bank (Johnson, 1994).

Table 1. Effect of goat integration and weed control options in maize.

Treatments	Weed control Index (%)		Maize grain yield kg ha^{-1}	
	2003	2004	2003	2004
Main Treatments				
Untreated control	81.8	80.49	33.07	19.96
Grazing	84.02	85.18	36.87	24.45
Penning	82.67	81.58	36.47	24.09
Grazing and Penning	82.32	82.50	44.29	27.17
CD (p=0.05)	1.06	0.86	1.05	1.06
Sub Treatments				
Unweeded control		-	27.29	20.98
Twice hand weeding	96.4	95.87	48.06	26.08
alachlor at 1.5 kg ai ha^{-1}	71.24	71.97	32.83	23.38
Intercrop blackgram	67.03	96.66	32.19	23.05
alachlor+handweeding	96.07	95.71	47.01	25.75
alachlor+blackgram	80.78	78.99	38.66	24.31
CD (p=0.05)	0.42	0.04	1.40	0.70

Among the weed control treatments, in cropping season significantly higher grain yields were recorded in hand weed plots registering 4806 and 2608 kg ha^{-1} during 2003 and 2004, respectively, and it was on par with alachlor application followed by hand weeding (Table 1). These results are in conformity with the reports of Thakur and Sharma (1996) and Mundra *et al.* (2003).

The weed seed bank studies (Table 2) revealed a reduction of 47.62%, 37.5%, 44.4% and 38.9% in *Cyperus rotundus*, *Cyanodon dactylon*, *Trianthema porlulacastrum*, *Echinochloa colonum* seeds, respectively, by grazing alone. This is comparatively better than the integration of goats for grazing as well as manuring wherein 28.57%, 18.75%, 35.4% and 33.3% reduction of *C. rotundus*, *C. dactylon*, *T. portulacastrum, E. colonum* seeds were recorded.

Table 2. Impact of various modes of goat integration on the weed seed bank

Treatments	*Cyperus rotundus*		*Cyanodon dactylon*		*Trianthema portulacastrum*		*Echinochloa colonum*	
	million seeds ha^{-1}	% decrease	million seeds ha^{-1}	% decrease	million seeds ha^{-1}	% decrease	million seeds ha^{-1}	% decrease
Un-weeded control	8.4	-	6.4	-	3.6	-	3.6	-
Goat grazing	4.4	47.62	4.0	37.5	2.0	44.4	2.2	38.9
Penning	7.6	9.52	6.0	6.25	2.4	7.69	3.0	22.2
Grazing and penning	6.0	28.57	5.2	18.75	2.2	35.4	2.4	33.3
CD (p=0.05)	0.44	-	0.24	-	1.64	-	1.92	-

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THE INFLUENCE OF MINIMAL WATER INPUT ON WEED DIVERSITY AND YIELD LOSS IN DIRECT-SEEDED RICE CULTIVATION

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Abstract: A study was conducted to determine rice yield loss due to weeds and status of weed composition and dominant weeds under minimal water input rice ecosystem. Two experiments were carried out in off season (March to August 2004) at two different locations namely, MARDI Bertam, Seberang Perai, Penang and Ladang Merdeka, Mulung, Kelantan (KADA). The experimental design was a split plot in Randomized Complete Block Design (RCBD) with 4 replicates. The main plots were the weeded treatments where all regular weed control methods applied with an un-weeded treatment. The subplot consisted of 5 water regimes treatments; T1 = continuous flooded condition (control), T2 = early flooding for the first month [30 DAS (days after sowing)] followed by saturated condition, T3 = early flooding up to panicle initiation stage (55 DAS) followed by saturated condition, T4 = continuous saturated condition, T5 = continuous field capacity condition. Pre-germinated rice seeds (*Oryza sativa* L) *var.* MR220 was broadcasted into each plot at a seeding rate of 150 kg/ha. Results show that rice yield loss due to weed competition were up to 90.9% in KADA and up to 53.4% in Bertam. Total grain yield of rice was also significantly affected by reduced water level treatments (saturated and field capacity conditions) in weed control plots to up to 40% and 61% in MARDI and KADA, respectively. In MARDI, grassy weeds (*i.e. Leptochloa. chinensis* and *Echinochloa* sp.) and sedges (*Fimbrystylis miliacea*) were more dominant (SDR up to 32.7%) under minimal water (saturated and field capacity) conditions. On the other hand, broadleaved weeds (*i.e. Monochoria vaginalis, Limnocharis flava,* etc.) were more dominant (SDR up to 66.5%) in flooded treatments. In KADA *Ischaemum rugosum* showed the highest percentage of SDR in all treatments with increasing trend of SDR as the water level reduced to saturated and field capacity conditions.

Key words: Minimal water input, weed diversity, weed control, rice, *Oryza sativa*

Introduction

Global climatic change and increase desertification due to deforestation and urban development lead to reduced water availability as well as induces changes in land use (Kathiresan, 2005). At the same time, the need to meet the growing demand for food will require increased crop production using less water. Regulated deficit irrigation provides a means of reducing water consumption while minimizing adverse effects on yield. Scarcity and growing competition for fresh water resources will reduce its availability for irrigation. Rice cultivation will be badly affected by this phenomenon. Weeds have been recognized as one of the major constraints on yield and quality of rice (Tsuru, 1991). The menacing effect of weeds inflicted more yield losses than any other pest. There were changes in weed flora from broad-leaved weeds and sedges in transplanted rice culture to competitive grassy weeds in direct seeded rice culture (Azmi and Mashhor, 1995). The composition of the weed flora may differ depending on location (Janiyah and Moody, 1983), water supply (Bhan, 1983), cultural practices (Benasor and De Datta, 1983; Mabbayad *et. al.* 1983), and the inherent weed flora in the area and the crop growth.

Rice yield is drastically reduced as a consequence of increased weed infestations due to limited water supply (Becker and Johnson, 1999).In a glasshouse trial, few or no weed seedlings emerged when the soil was flooded; whereas at field capacity, all weed species emerged readily (Smith and Fox, 1973). Continuous shallow ponding and shallow ponding until panicle initiation (55 days after sowing - DAS) also reduced weed species number, density and biomass as compared to saturated soil throughout (Kent and Johnson, 2001). The highest result on rice growth and yield were also obtained from flooding condition compared to continuous saturated condition (Mohankumar and Alexander, 1989). Quantitative information on shifts in weed flora brought about by changing water management practices can provide valuable indications for future weed control strategies during water crisis situation. The objective of this experiment is to evaluate the effect of different water regimes on weeds infestation and rice yield under field condition.

Materials and Methods

The trial was carried out in the off season of 2004 at MARDI Bertam, Seberang Perai and Ladang Merdeka, Mulung, Kelantan (KADA). A total of 40 individual plots (10 m x 25 m) were constructed where 20 were plots with weed control practices [water regimes treatment + all regular weeds control methods (chemical and cultural)] and another 20 were unweeded plots (water regimes treatment only). Five water regimes were used in this study namely; T1 = continuous flooded condition (10 cm water level), T2 = flooding for the first 30 DAS (Day After Sowing) followed by saturated condition, T3 = flooding up to panicle initiation stage (55 DAS) followed by saturated condition, T4 = continuous saturated condition, T5 = continuous field capacity condition. All crop management practices (land and plots preparation, fertilizer, herbicides and pesticides application, etc..) were followed the MARDI's Rice Cultivation Manual 2002. The experiment was arranged in a split plot in Randomized Complete Block Design (RCBD) with 4 replicates. All crop management practices (land and plot preparation, fertilizers, herbicides and pesticides application) followed MARDI's Rice Cultivation Manual 2002. Pre-germinated MR220 rice seeds were broadcasted into each plot at a seeding rate of 150 kg/ha. Water regime treatments were introduced into each plot at 7 DAS.

Weeds were collected at random in each plot using census count quadrate (0.5 m x 0.5 m) method at 30, 60 and 90 DAS (Kim and Moody, 1983). All plots were first marked on a map so as to minimize bias when placing a quadrate again in the plot. Four quadrates were taken in each plot. Weeds present in each quadrate were collected, washed and dried at 80^{o}C to constant weight, and weighed. The summed dominance ratio (SDR) of the weed species was computed using the following equations (Janiya and Moody, 1989):

$$\text{SDR} = \frac{\text{relative density (RD) + relative dry weight (RDW)}}{2}$$

where,

$$\text{RD} = \frac{\text{Density of a given species}}{\text{Total density}} \times 100$$

$$\text{RDW} = \frac{\text{Dry weight of a given species}}{\text{Total dry weight}} \times 10$$

Rice yields were obtained from the center of each plot with a harvested area of 5 m x 5 m at 115 DAS and the data were converted to kg/ha at 14% moisture. Weight of dry weeds and rice yield data were analyzed and subjected to Analysis of Variance using the Statistical Analysis System (SAS). Mean separation was done using Duncan New Multiple Mean Range Test (DNMRT).

Results and Discussion

A different weed composition was observed in two different locations studied (Tables 1 and 2). A total of twelve and five weed species were observed in MARDI, Seberang Prai and KADA, Kelantan respectively. In MARDI Bertam, *Monochoria vaginalis* and *Limnocharis flava* were the most dominant weeds in flooded conditions (T1 - T3), while *Fimbristylis miliacea*, *Echinochloa colona*, *E. crus-galli* and *Leptochloa chinensis* were the most dominant weeds in minimal water condition (T4 and T5). In KADA, *Ischaemum rugosum*, *Leptochloa chinensis* and *Echinochloa crus-galli* were the most dominant weeds in all treatments. Within water regime treatments, unweeded plots showed a higher number of weed species as compared to weeded plots in both locations.

Table 1. Effect of different water regimes on Summed Dominance Ratio (SDR) of weeds in MARDI Bertam field at 60 DAS during off season 2004

Weed Species	Water Regime Treatments									
	T1		T2		T3		T4		T5	
	W	UW	W	UW	W	UW	W	UW	W	UW
Limnocharis flava	66.5^{a}	15.5^{abc}	29.7^{a}	35.0^{a}	25.0^{a}	6.7^{b}	14.1^{a}	16.2^{abc}	6.4^{b}	14.0^{ab}
Monocharia vaginalis		27.7^{a}		31.6^{ab}	9.4^{b}	32.1^{a}		4.3^{bc}	2.3^{b}	20.5^{a}
Leptochloa chinensis		5.7^{bc}	20.7^{a}	8.8^{bc}	8.5^{b}	10.0^{b}	1.6^{b}	23.7^{ab}	19.7^{ab}	9.7^{ab}
Frimbristylis miliacea	11.9^{b}	23.4^{ab}	12.3^{b}	16.6^{abc}		22.3^{ab}		25.8^{a}	32.7^{a}	23.4^{ab}
Echinochloa colona	6.2^{b}	7.7^{abc}	29.3^{a}	0.7^{c}	7.1^{b}	6.5^{b}	8.4^{a}	1.0^{c}	6.2^{b}	4.7^{b}
Echinochloa crus-galli	1.4^{b}	13.6^{abc}			25.0^{a}	5.8^{b}				9.5^{ab}
Sagittaria guyanensis	14.0^{b}	1.8^{bc}	8.0^{b}	7.1^{bc}						
Ludwigia hyssopifolia		2.1^{bc}		0.3^{c}		6.7^{b}		7.0^{abc}		8.7^{ab}
Cyperus iria		2.0^{bc}				8.5^{b}				1.4^{b}
Bacopa rotundifolia							1.0^{b}		2.0^{b}	
Cyperus haspan						1.4^{b}		0.2^{c}		
Panicum repens		0.6^{c}							5.7^{b}	

Within a each column, means followed by the same letters are not significantly different by the DMRT (p = 0.05).

Table 2. Effect of different water regimes on Summed Dominance Ratio (SDR) of weeds in KADA field at 60 DAS during off season 2004

Weed Species	Water Regime Treatments									
	T1		T2		T3		T4		T5	
	W	UW	W	UW	W	UW	W	UW	W	UW
Ischaemum rugosum	62.3^{a}	55.0^{a}	68.3^{a}	44.4^{a}	76.2^{a}	49.6^{a}	75.0^{a}	31.0^{ab}	79.0^{a}	28.9^{ab}
Leptochloa chinensis	19.4^{b}	8.5^{bc}	17.4^{b}	6.3^{b}	7.3^{b}	1.2^{c}	8.0^{b}	2.2^{c}	3.7^{b}	3.4^{b}
Echinochloa crus-galli	18.3^{b}	33.5^{ab}	14.6^{b}	35.1^{a}	21.1^{b}	33.7^{b}	17.1^{b}	46.1^{a}	17.0^{b}	53.0^{a}
Ludwigia hyssopifolia				5.2^{b}		3.5^{c}		7.8^{b}		5.5^{b}
Cyperus iria		3.1^{c}		9.0^{b}		12.1^{c}		8.5^{c}		9.1^{b}

Within a column, means followed by the same letters are not significantly different by the DMRT (p = 0.05).

The percentage of summed dominant ratio of weed was greatly affected by water regimes in both locations (Tables 1 and 2). In MARDI Bertam, *L. flava* showed the highest percent SDR (66.5%) in continuous flooded (T1) weeded plots while in un-weeded plots *M. vaginalis* recorded the highest SDR (27.7 %). In T2, the highest SDR in weeded plots were *L. flava (*29.7%) and *E. colona* (29.3 %) while in un-weeded plots the highest SDR were from *L. flava* (35%) and *M. vaginalis* (31.6%). In T3, *L. flava* (25%) and *E. crus-galli* (25%) gave the highest SDR for weeded plots and *M. vaginalis* (32.1%) gave the highest SDR for un-weeded plots. In T4, *L. flava* (14.1%) and *E. colona* (8.4%) showed the highest SDR in weeded plots while, *F. miliacea* (25.8%) and *L. chinensis* (23.7%) showed the highest in un-weeded plots. In T5, *F. miliacea* obtained the highest SDR (32.7%) in weeded plots with a similar trend was also observed in un-weeded plots.

In KADA, *I. rugosum* showed the highest percentage of SDR in all treatments with increasing trend of SDR as the water level reduced to saturated and field capacity condition. In T1, SDR of *I. rugosum* were 62.3% in weeded and 55 % in un-weeded plot. SDR of *I. rugosum* in T2 were 68.3% in weeded and 44.4% in un-weeded plots. In T3, SDR of *I. rugosum* were 76.2% and 49.6% in weeded and un-weeded plot respectively. In T4, *I. rugosum* (75%) and *E. crus-galli* (46.1%) gave the highest SDR values in weeded and un-weeded respectively. In T5, the similar trend as in T4 was also observed. Results obtained from Smith and Fox (1973) in a glasshouse trial and Kent and Johnson (2001) in a field trial supported the above results. The total grain yield of rice was also significantly affected by reduced water level treatments (saturated and field capacity conditions) in weed control plots to up to 40% and 61% in MARDI and KADA respectively (Tables 3 and 4).

Table 3. Effect of different water regime treatments on rice grain yield during off season 2004 in MARDI, Bertam

Weed Control	Rice Yield (kg/ha)		
Treatment	Weeded	Un-weeded	% Yield Loss
T1	3475.0[a]	1618.7[a]	53.4
T2	2510.7[ab]	1729.3[a]	31.1
T3	2591.0[ab]	1460.0[a]	43.7
T4	2386.0[ab]	1713.3[a]	27.6
T5	2085.0[b]	1476.0[a]	29.2
Means	2609.5[A]	1599.5[B]	38.7

Within a row, means followed by the same letter are not significantly different by the DMRT (p=0.05)

Table 4. Effect of different water regime treatments on rice grain yield during off season 2004 in KADA

Weed Control	Rice Yield (kg/ha)		
Treatment	Weeded	Un-weeded	% Yield Loss
T1	3146.2 [a]	302.6 [ab]	90.4
T2	3190.2 [a]	443.9 [a]	86.1
T3	2925.1 [a]	355.8 [ab]	87.8
T4	2064.2 [b]	379.2 [ab]	81.6
T5	1217.5 [c]	111.2 [b]	90.1
Means	2508.6[A]	302.6 [B]	87.9

Within a row, means followed by the same letter are not significantly different by the DMRT (p=0.05)

The highest rice grain production was recorded at T1 (3475 kg/ha), which is significantly different to that of T5 (the lowest 2085 kg/ha), but not significantly different compared to T2, T3 and T4 treatments in MARDI. In KADA, the highest grain production was recorded at T2 (3190.2 kg/ha) which significantly different when compared to T4 (2064.2 kg/ha) and T5 (the lowest 1217.5 kg/ha), but not significantly different compared to T1 and T3 treatments. A significantly higher (p<0.05) rice yield was observed between plots with weed control and un-weeded plots in KADA (up to 90.9 % reduction) and MARDI (up to 53.4 % reduction). Similar results were also observed by Mohankumar and Alexander (1989).

Conclusion

Variability in water regimes affected the weed species and its distribution, and rice yields differently. Grass weeds and sedges were dominant under minimal water (saturated and field capacity) conditions especially in MARDI and broadleaved weeds were dominant in flooded treatments. Flooded conditions favored rice growth and yield and resulted in an effective suppression of weeds compared to the plots under field capacity. All flooding treatment, T1, T2 and T3, resulted similar degree of reduction in weed infestations, and increased rice production significantly.

Acknowledgements

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HERBICIDE RESISTANCE IN *Echinochloa spp* IN THE PHILIPPINES

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Abstract: Nineteen biotypes of two *Echinochloa* species namely, *Echinochola crus-galli* and *E. glabrescens* collected from rice fields in Nueva Ecija and Iloilo provinces were screened for butachlor+propanil resistance. One species from Cotabato served as the susceptible check. Resistance screening showed that 18 populations (89.5%) exhibited resistance to the herbicide and one population was classified as developing resistance. Of these biotypes, six were *E. crusgalli* and 11 were *E. glabrescens.* Dose response assays using butachlor+propanil on the 18 populations showed that 17 were not controlled at twice the recommended rate for field application (2 l ha^{-1}) of the herbicide. In addition, seven biotypes survived a herbicide concentration of 4 l ha^{-1}. Dose-response assays using butachlor and propanil separately indicated that these species were also resistant to these herbicides. Butachlor+propanil concentration required to reduce the number of survivors by 50% (LD_{50}) in all resistant biotypes ranged from 0.6 l ha^{-1} (0.42 kg a.i) to 2.9 l ha^{-1} (2.03 kg a.i) indicating a lower sensitivity to this herbicide by 1.9 to 9.1 times when compared to the susceptible species. The same trend could be established with LD_{50} values obtained in the dose response assays using butachlor alone and propanil alone. Development of resistance was due to the use of butachlor and then butachlor+propanil for extended time periods in these provinces. The study illustrated that farmers need to implement integrated weed management strategies with the use of non-chemical means and herbicides with different mechanisms of action to manage herbicide resistant weeds.

Key words: Barnyardgrass, butachlor, *E. crusgalli* (ECHCG), *E. glabrescens* (ECHGL), propanil, dose response assays

Introduction

In the Philippines, the first documented rice weed tolerant to 2, 4-D was *Sphenoclea zeylanica* found in San Rafael, Bulacan in Central Luzon (Sy and Mercado 1983). Migo *et al*. (1986) conducted a research using different doses of 2,4-D on *S. zeylanica* collected in different rice areas in the country and found different responses of the weed populations to 2,4-D. Some farmers in Nueva Ecija province have also reported the ineffectiveness of some herbicides such as butachlor and butachlor+propanil to control some weed species in their rice areas (Juliano 2005, unpublished data). After the work of Migo *et al*. (1986), there have been no studies on herbicide resistance in weeds associated with rice in the Philippines. It was in this context that this study was conceptualized.

The intensive monocropping in areas where direct seeding rice is practiced prompted farmers to use herbicides as the primary means of weed control. A survey conducted by the Philippine Rice Research Institute (PhilRice) and the Philippines Bureau of Agricultural Statistics (BAS) in 2001 in direct seeded areas revealed that acetanilides such as butachlor and pretilachlor dominate the herbicides used in direct seeded rice (Casimero *et al*. 2005). PhilRice and the Australian Centre for International Agricultural Research (ACIAR) also conducted a survey in Nueva Ecija and Iloilo in 2004. These provinces were chosen because Iloilo (>90%) and Nueva Ecija (60%) lead the highest number of farmers adopting direct seeding in the Philippines (Marsh *et al*. 2005). It was found that the acetanilides were still widely used by farmers with butachlor+ propanil also being more commonly used followed by butachlor and pretilachlor. The use of sulfonylureas and aryloxyphenoxypropionates (AOPPs) was found to be increasing. Thus, the development of resistance in weeds could possibly occur as most of the herbicides the farmers have been using for extended time periods belong to the same herbicide family.

Major weeds in the PhilRice-BAS survey in 2001 were dominated by *Echinochloa* spp., *L. chinensis, I. rugosum, F. miliacea, C. rotundus, and S. zeylanica* and these weed species were also identified as the dominant weeds in the PhilRice-ACIAR survey in 2004. With these results on herbicide use, there is a probability that these weed species may have developed resistance to the herbicides already used in these provinces. This study aimed to determine the existence of herbicide resistance of the *Echinochloa* species associated with direct seeded rice in Nueva Ecija and Iloilo provinces. It also determined the level of herbicide dose that the resistant biotypes can tolerate.

Materials and Methods

All experiments on resistance screening and response dose assays were conducted at the screenhouse of PhilRice Central Experiment Station in Nueva Ecija province, Philippines from June 2005 to February 2006. Mature seeds of *Echinochloa* species were collected from monocropped direct seeded rice fields in Nueva Ecija and Iloilo provinces. Seeds were identified, characterized, dried, labeled, and stored prior to use.

Screening for herbicide resistance

Of the seeds collected, eighteen populations of two *Echinochloa* species were screened for resistance to butachlor+propanil. This herbicide is the most commonly used herbicide in the two provinces based from previous surveys conducted. One population of *Echinochloa glabrescens* collected from General Santos City, South Cotabato was used as the susceptible check. The seedlings were screened using the recommended rate of the commercial formulation of butachlor+propanil at 1 l ha^{-1} or 0.7 kg a.i ha^{-1} (192 l ha^{-1} spray volume) with an application at 6-8 days after seeding (DAS). The procedure of Valverde *et al*. (2000) on whole plant assays was used in the screening. Seeds of suspected resistant weed species were pregerminated and transferred to clay pots after two days. At 6 days after transplanting, the plants were thinned to three uniform seedlings per pot before herbicide application. A total of 30 plants at three plants per pot (equivalent to 10 replications) of similar size and growth stage (at 2-leaf stage) of each population were selected. A manual hand sprayer was calibrated and used in the experiment. Each population received two passings of the herbicide.

Populations were classified as resistant, developing resistance or susceptible based on the number of plants that survived in each population (Llewellyn & Powles, 2001). All populations which survived herbicide treatment were recorded as unaffected and expressed as percentage of the controls. Populations were resistant if more than 20% of the plants survived the herbicide treatment, developing resistance if 1-20% of the plants that survived, and susceptible if all plants were killed by the herbicide treatment. The weed biotypes classified as resistant were further subjected to response dose assays using butachlor+propanil, butachlor alone, and propanil alone.

Dose response assays using butachlor+propanil.

Whole plant assay was also followed in the dose response assay of the resistant populations. The treatments were: T1 – control (no herbicide applied), T2 – recommended rate (1 l ha^{-1} or 0.7 kg a.i ha^{-1}), T3 – two times the recommended rate (2 l/ha or 1.4 kg a.i/ha), and T4 – four times the recommended rate (4 l ha^{-1} or 2.8 kg a.i ha^{-1}). The susceptible check was also subjected to response dose assay using butachlor+propanil at the following rates: T1 – control (no herbicide spray), T2 – 250 ml ha^{-1} (0.175 kg a.i ha^{-1}), T3 – 500 ml ha^{-1} (0.350 kg a.i ha^{-1}), and T4 – 750 ml ha^{-1} (0.525 kg a.i ha^{-1}).

Dose response assay using butachlor

This assay was done to determine the effect of butachlor separately from butachlor+propanil on all the resistant barnyard grass populations. The treatments included: T1 – control (no herbicide applied), T2 – recommended rate (1 l ha^{-1} or 0.6 kg a.i ha^{-1}), T3 – two times the recommended rate (2 l ha^{-1} or 1.2 kg a.i ha^{-1}), T4 – four times the recommended rate (4 l ha^{-1} or 2.4 kg a.i ha^{-1}). A sterilized Maligaya clay loam soil was used in the pots. Seeds were soaked for 24 hrs before they were sown. There were three seeds in each pot. Treatments were applied at three days after seeding.

Dose response assay using propanil

Technical grade propanil (98%) was used as there is no available commercial formulation of the herbicide propanil in Philippine markets. The treatments were: T1 – control (no herbicide spray), T2 – recommended rate (2 kg ha^{-1} or 2 kg a.i ha^{-1}), T3 – two times the recommended rate (4 kg ha^{-1} or 4 kg a.i ha^{-1}), and T4 – four times the recommended rate (8 kg ha^{-1} or 8 kg a.i ha^{-1}). Pre-germinated seedlings were transplanted into soil-filled clay pots (5 seedlings per pot). Before herbicide application (at 3-leaf stage), the plants were thinned to three plants per pot.

Data and statistical analyses

The number of survived seedlings was counted at 7, 14, and 21 days after treatment (DAT). All experiments were repeated once and data is presented as averages of two experiments. Data obtained in the resistance screening and response dose assays were analyzed statistically through the Analysis of Variance (ANOVA) and treatment means were compared using the t Tests or Least Significant Difference (LSD). The LD_{50} values required to reduce the number of survivors of the resistant and susceptible populations by 50% in relation to the untreated control were determined based on the response dose curve plotted using *Statistical* Program. All LD_{50} values obtained were compared using Tukey's test. The ratio of the LD_{50} of the resistant biotype over the LD_{50} of a reference, susceptible biotype is called resistance index (RI) or R/S ratio and was obtained through the following equation: RI = LD_{50} resistant population/LD_{50} susceptible reference.

Results and Discussion

Screening for resistance

Seventeen (89.5%) of the 19 populations of *Echinochloa* species exhibited resistance to butachlor+propanil, one population of *E. glabrescens* (5%) was rated as developing resistance to this herbicide, and one susceptible (5%) population collected from General Santos City, Cotabato was used as the check (Table 1). Most of the populations in Nueva Ecija (37%) were classified as resistant plants (40-60% survivors) while 16% of the population from Iloilo had 61-80% and 21-40% survivors. Further, 11% and 16% of the populations from Nueva Ecija and Iloilo, respectively, had above 60% survivors indicating highly resistant populations. The prolonged use of butachlor+propanil for weed control in both provinces may have attributed the increased in the selection for resistance (Juliano, L. 2005, unpublished data). The results may suggest that if farmers will not stop using butachlor+propanil to control weeds, the percentage of resistant plants may reach very high levels.

Table 1. Survival and resistance rating of *Echinochloa* species collected from farmers' fields screened for herbicide resistance using butachlor+propanil at 1 l ha^{-1} (0.7 kg a.i ha^{-1}).

Species	Place of collection	No. of plants survived	% Survival	Resistance rating
E. crusgalli	Bucot, Aliaga, Nueva Ecija	15	50	Resistant
E. glabrescens	Bcot, Aliaga, Nueva Ecija	18	60	Resistant
E. glabrescens	Aglipay, Rizal, Nueva Ecija	20	66.7	Resistant
E. crusgalli	Bertrese, Quezon, Nueva Ecija	15	50	Resistant
E. crusgalli	Lusok, Bongabon, Nueva Ecija	15	50	Resistant
E. crusgalli	Soledad, Sta. Rosa, Nueva Ecija	17	56.7	Resistant
E. glabrescens	Baluga, Talavera, Nueva Ecija	16	53.3	Resistant
E. glabrescens	Rang-ayan, Munoz, Nueva Ecija	12	40	Resistant
E. glabrescens	La Purisima, Aliaga, Nueva Ecija	15	50	Resistant
E. crusgalli	Acuit, Barotac Nuevo, Iloilo	17	57	Resistant
E. glabrescens 1	Acuit, Barotac Nuevo, Iloilo	21	70	Resistant
E. glabrescens 2	Acuit, Barotac Nuevo, Iloilo	20	66.7	Resistant
E. glabrescens 3	Acuit, Barotac Nuevo, Iloilo	20	66.7	Resistant
E. crusgalli	Monpon, Barotac Nuevo, Iloilo	12	40	Resistant
E. glabrescens	Monpon, Barotac Nuevo, Iloilo	15	30	Resistant
E. glabrescens	Dapitan, Pototan, Iloilo	10	33.3	Resistant
E. glabrescens	Hamabalud, Pototan, Iloilo	5	16.7	Developing resistance
E. glabrescens	Sinibaan, Dingle, Iloilo	10	33.3	Resistant
E. glabrescens	Gen. Santos City, Cotabato	0	0	Susceptible

Note: Species or populations followed by numbers in parenthesis were collected in different fields in the same village. Resistance rating – resistant (>20% survivors), developing resistance (1-20% survivors), susceptible (zero survivors).

Dose response assay using butachlor+propanil

All the nine populations collected in Nueva Ecija survived herbicide application at the recommended rate (1 l ha^{-1}) with survival rates ranging from 48.3% to 66.7% (Figure 1). All populations tolerated herbicide application at twice the recommended rate (2 l ha^{-1}) with percentage survival ranging from 10-62% of the total plants in each population. Three populations survived herbicide doses as high as four times the recommended rate for field application (4 l ha^{-1}) with survivors ranging from 10-37%. The same trend could be established in Iloilo where all the nine populations obtained high survival ratings (Figure 2) with only ECHGLHam population having a low survival percentage (16.7%). All the other populations survived herbicide treatments which were twice the recommended rate with survival rates ranging from 10–58%. Four of the resistant populations survived application of the herbicide up to four times the recommended rate. The results further indicated that more than 4 l ha^{-1} of butachlor+propanil is needed to control 90% of the highly resistant populations in both provinces.

Dose response assays using butachlor and propanil

All populations in both provinces except for ECHGLHam from Iloilo survived twice the recommended rate (2 l ha^{-1}) for field application of butachlor (Figures 3 and 4). Ten populations survived herbicide treatments up to four times the recommended rate. The populations resistant to butachlor+propanil were also resistant to butachlor. The selection for butachlor resistance when used alone could have resulted from the previous use of this herbicide before shifting to butachlor+propanil due to the inefficacy of butachlor to control weeds (unpublished data).

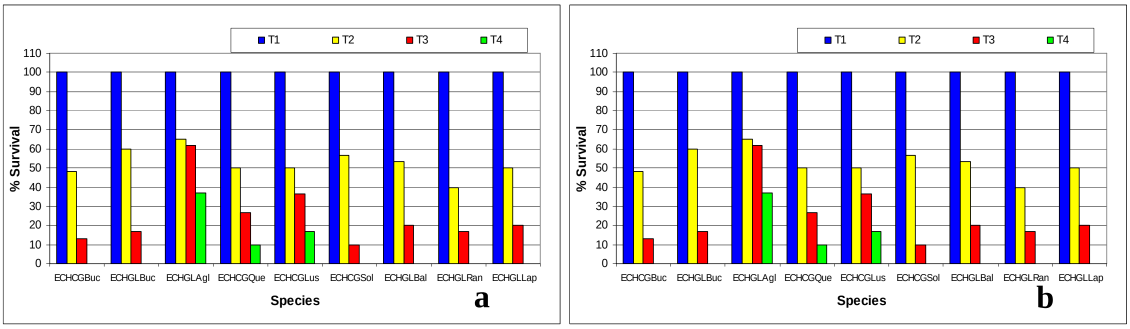

Figure 1. Per cent survival of *Echinochloa* species from Nueva Ecija (a) and Iloilo (b) provinces treated with butachlor + propanil at increasing rates. T1 – control, T2 – recommended rate (1 l ha^{-1}), T3 – 2x recommended rate (2 l ha^{-1}), T4 – 4x recommended rate (4 l ha^{-1}).

For propanil assay, seven populations were used and all populations except *E. crusgalli* (ECHCGBuc) from Nueva Ecija survived concentrations up to 4 times the recommended rate (8 kg/ha) of propanil (Figure 3). Although propanil alone is not used in rice areas in both provinces, results showed that its combination with butachlor provided selection for propanil resistance.

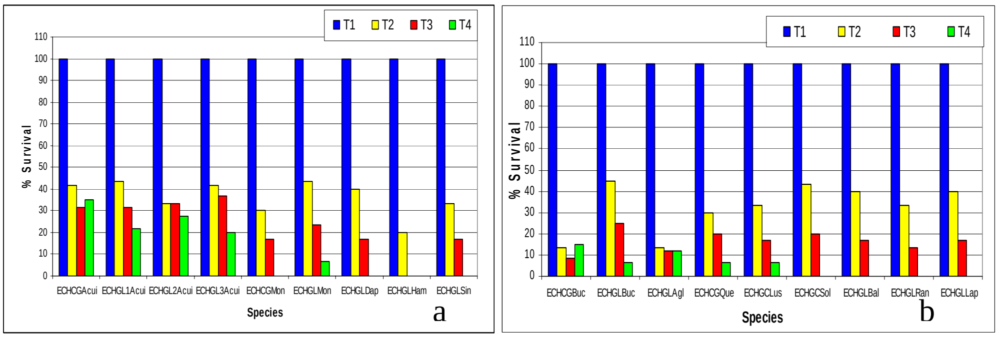

Figure 2. Per cent survival of *Echinochloa* species from Nueva Ecija (a) and Iloilo (b) provinces treated with butachlor at increasing rates. T1 – control, T2 – recommended rate (1 l/ha), T3 – 2x recommended rate (2 l ha^{-1}), T4 – 4x recommended rate (4 l ha^{-1}).

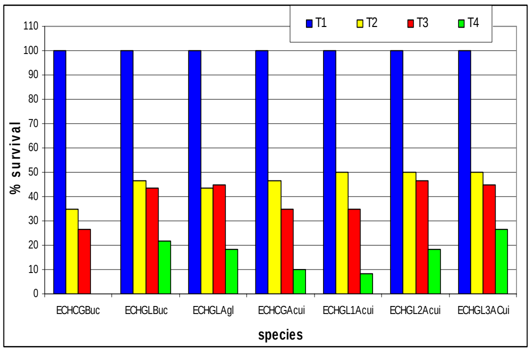

Figure 3. Per cent survival of *Echinochloa* species treated with propanil at increasing rates. T1 – control, T2 – recommended rate (2 kg ha^{-1}), T3 – 2x recommended rate (4 kg ha^{-1}), and T4 – 4x recommended rate (8 kg ha^{-1}). .

Lethal dose (LD_{50})

Of the 19 populations, four biotypes had high LD_{50} ranging from 1.8 l/ha (1.26 kg a.i) to 2.9 l ha^{-1} (2.03 kg a.i) of butachlor+propanil (Table 2). Minimum and maximum resistance ratios of the biotypes indicated that these four highly resistant biotypes were 5.6 to 9 times less sensitive to butachlor+propanil than the susceptible biotype.

A positive correlation was found between the resistance category of the *Echinochloa* species and the LD_{50} rate of butachlor + propanil. In this case, the four highly resistant biotypes; ECHGLAgl from Nueva Ecija, ECHCGAcui, ECHGL1Acui and ECHGL3Acui all from Iloilo, had high LD_{50} and high resistance rating to butachlor+propanil. Differences in the LD_{50} indicated several levels of resistance present in all resistant biotypes

Table 2. Butachlor + propanil concentration required to reduce by 50% the number of survivors of resistant *Echinochloa* biotypes.

Species	Place of collection	LD_{50} (l ha^{-1})	LD_{50} (kg a.i)	R/S ratio
E. crus-galli	Bucot, Aliaga, Nueva Ecija	1.0 b	0.70	3.1
E. glabrescens	Bcot, Aliaga, Nueva Ecija	1.3 c	0.91	4.1
E. glabrescens	Aglipay, Rizal, Nueva Ecija	2.9 d	2.03	9.1
E. crusgalli	Bertrese, Quezon, Nueva Ecija	1.0 b	0.70	3.1
E. crus-galli	Lusok, Bongabon, Nueva Ecija	1.0 b	0.70	3.1
E. crus-galli	Soledad, Sta. Rosa, Nueva Ecija	1.15 b	0.81	3.6
E. glabrescens	Baluga, Talavera, Nueva Ecija	1.10 b	0.77	3.4
E. glabrescens	Rang-ayan, Munoz, Nueva Ecija	0.80 a	0.56	2.5
E. glabrescens	La Purisima, Aliaga, Nueva Ecija	1.0 b	0.70	3.1
E. crus-galli	Acuit, Barotac Nuevo, Iloilo	1.25 c	0.88	3.9
E. glabrescens 1	Acuit, Barotac Nuevo, Iloilo	2.15 d	1.51	6.7
E. glabrescens 2	Acuit, Barotac Nuevo, Iloilo	1.05 b	0.74	3.3
E. glabrescens 3	Acuit, Barotac Nuevo, Iloilo	1.80 d	1.26	5.6
E. crus-galli	Monpon, Barotac Nuevo, Iloilo	0.80 a	0.56	2.5
E. glabrescens	Monpon, Barotac Nuevo, Iloilo	1.0 b	0.70	3.1
E. glabrescens	Dapitan, Pototan, Iloilo	0.75 a	0.53	2.3
E. glabrescens	Hamabalud, Pototan, Iloilo	0.60 a	0.42	1.9
E. glabrescens	Sinibaan, Dingle, Iloilo	0.75 a	0.53	2.3
E. glabrescens	Gen. Santos City, Cotabato	0.32 a	0.22	-

In a column values followed by the same letter are not significantly different by the Tukey's test ($p=0.05$).

The LD_{50} obtained in the dose response assays using butachlor alone are shown in Table 3. Minimum and maximum resistance ratios indicated 1.9 to 2.7 times less sensitivity of the resistant biotypes than the susceptible species to butachlor. On the other hand, LD_{50} obtained in the response dose assays using propanil alone are presented in Table 4. Propanil resistant biotypes were 2.3 to 3.1 less sensitive compared to the susceptible species.

The continuous use of a single herbicide for several years (nine and seven years in Iloilo and Nueva Ecija respectively) in monocropped areas (3 croppings/year in Iloilo and 2 croppings/year in Nueva Ecija) provided selection for resistant populations (unpublished data). This also explained why farmers in these areas have been using higher doses of herbicides and increasing the number of applications per season. What was perceived to be a result of reduced herbicide efficacy is actually herbicide resistant of the weeds to butachlor and then to butachlor+propanil.

Studies on weed management options, especially on the use of non-chemical strategies that would effectively reduce resistant weed populations should be conducted at the farm level. Extensive survey with systematic sampling in areas where rice is monocropped intensively should also determine the extent of area that herbicide resistant weeds may possibly occur. Other dominant weed species should also be screened against herbicides used in these areas for possible herbicide resistance. Herbicide resistance mechanisms need to be established for the development of effective herbicide management strategies.

Table 3. Butachlor concentration required to reduce by 50% the number of survivors of resistant *Echinochloa* biotypes.

Species	Place of collection	LD_{50} (l ha^{-1})	LD_{50} (kg a.i)	R/S ratio
E. crus-galli	Bucot, Aliaga, Nueva Ecija	0.6 a	0.36	1.9
E. glabrescens	Bcot, Aliaga, Nueva Ecija	0.9 c	0.54	2.8
E. glabrescens	Aglipay, Rizal, Nueva Ecija	0.6 a	0.36	1.9
E. crus-galli	Bertrese, Quezon, Nueva Ecija	0.7 a	0.42	2.2
E. crus-galli	Lusok, Bongabon, Nueva Ecija	0.75 a	0.45	2.3
E. crus-galli	Soledad, Sta. Rosa, Nueva Ecija	0.85 a	0.51	2.7
E. glabrescens	Baluga, Talavera, Nueva Ecija	0.8 b	0.48	2.5
E. glabrescens	Rang-ayan, Munoz, Nueva Ecija	0.7 a	0.42	2.2
*E. glab*rescens	La Purisima, Aliaga, Nueva Ecija	0.8 b	0.48	2.5
E. crus-galli	Acuit, Barotac Nuevo, Iloilo	0.8 b	0.48	2.5
E. glabrescens 1	Acuit, Barotac Nuevo, Iloilo	0.85 b	0.51	2.7
E. glabrescens 2	Acuit, Barotac Nuevo, Iloilo	0.75 a	0.42	2.3
E. glabrescens 3	Acuit, Barotac Nuevo, Iloilo	0.8 b	0.48	2.5
E. crus-galli	Monpon, Barotac Nuevo, Iloilo	0.7 a	0.42	2.2
E. glabrescens	Monpon, Barotac Nuevo, Iloilo	0.85 b	0.51	2.7
E. glabrescens	Dapitan, Pototan, Iloilo	0.8 b	0.48	2.5
E. glabrescens	Hamabalud, Pototan, Iloilo	0.6 a	0.36	1.9
E. glabrescens	Sinibaan, Dingle, Iloilo	0.75 a	0.45	2.3
E. glabrescens	Gen. Santos City, Cotabato	0.32 a	0.19	-

In a column values followed by the same letter are not significantly different by the Tukey's test (p=0.05).

Table 4. Propanil concentration required to reduce by 50% the number of survivors of resistant *Echinochloa* biotypes.

Species	Place of collection	LD_{50} (l ha^{-1})	LD_{50} (kg a.i)	R/S ratio
E. crus-galli	Bucot, Aliaga, Nueva Ecija	1.5 b	1.5	2.3
E. glabrescens	Bcot, Aliaga, Nueva Ecija	1.8 b	1.8	2.8
E. glabrescens	Aglipay, Rizal, Nueva Ecija	1.7 b	1.7	2.7
E. crus-galli	Acuit, Barotac Nuevo, Iloilo	1.8 b	1.8	2.8
E. glabrescens1	Acuit, Barotac Nuevo, Iloilo	2.0 b	2.0	3.1
E. glabrescens2	Acuit, Barotac Nuevo, Iloilo	2.0 b	2.0	3.1
E. glabrescens3	Acuit, Barotac Nuevo, Iloilo	2.0 b	2.0	3.1
E. glabrescens	Gen. Santos City, Cotabato	0.64 a	0.64	-

In a column values followed by the same letter are not significantly different at by the Tukey's test (p=0.05).

Farmer education on the development and implications of herbicide resistance in rice weeds is necessary to empower them to make sound weed management decisions. An advocacy program would entail linking research values and weed management with several stakeholders. A shared responsibility must be established and each group of people must become aware of the implications of herbicide resistance and how best to work as a team to lessen the impact of this problem.

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ALLELOPATHIC EFFECTS OF AQUEOUS EXTRACTS FROM RICE LEAVES AND DECOMPOSING RICE DEBRIS ON THE SEED GERMINATION AND GROWTH OF BARNYARD GRASS (*Echinochloa crusgalli* (L) Beauv.)

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Abstract: Laboratory and greenhouse experiments were carried out to observe the allelopathic effects of aqueous extract of fresh rice leaves and the decomposing rice straw on seed germination and growth of barnyard grass. Twelve rice varieties namely, Acheh Puteh, Anak China, Anak Ikan, Anak Ikan China, MR 14, MR 15, MR 59, Makmur, Manik, Seberang, Sekencang and Y1021 were used in both experiments. In the laboratory experiment, five concentrations of leaf extract *e.g.* 0, 50, 37.5, 25 and 12.5 g/l and in greenhouse experiment, four concentrations of rice debris *e.g*. 0, 2.5, 5.0 and 10.0 g dry weight/1000 g soil were used. The allelopathic effect of rice leaf extract significantly decreased seed germination and radicle and plumule length of barnyard grass. The highest reduction in germination was rendered by Y 1021 followed by Anak China and the lowest reduction was observed in MR 14. On an average although the radicle length was slightly increased, the plumule length was reduced. The highest reduction of plumule length was rendered by Y 1021 followed by MR 15 and the lowest reduction was under Acheh Puteh. Fresh weights of the weed were reduced, with the highest reduction under Y 1021, followed by MR 15 and the lowest reduction under MR 14. The concentrations of leaf extracts could not produce any significant difference. In greenhouse study, the varieties as well as the debris concentrations made significant differences in their allelopathic effects and seed germination, seedling length and seedling dry weight was reduced. Among the varieties, the highest reduction of seedling length and dry weight were recorded with Anak Ikan and the lowest reduction of seedling length and dry weight were noted with Sekencang.

Keywords: Aqueous leaf extract, barnyard grass, decomposing rice debris, rice allelopathy, rice

Introduction

Allelopathy is a phenomenon where direct or indirect effects of one plant to another occur by releasing compounds into the environment through root exudation, leaching by dew and rain, and volatilization or by decaying plant tissues. Allelopathy in rice crops may act as a cultural weed control in the agro ecosystems; consequently it may reduce excessive use of herbicides. Preliminary studies have shown that several accessions of rice germplasm in the field were found to decrease the growth of ducksalad (Dilday *et al*. 1994) and barnyard grass (Olofsdotter and Navarez, 1996). Chou and Lin (1976) observed that the aqueous extracts of decomposing rice residues in soil inhibited the root growth of rice and lettuce seedlings. A large volume of rice straw always is left in the field, which is incorporated in the soil during land preparation. If the rice variety posses this potentiality of releasing allelochemicals when decomposing in soil, it may serve as a means of biological weed control along with increasing the nutrient status and improvement of soil texture. Therefore, this study was conducted to investigate the effects of aqueous extracts of rice leaves and the decomposed rice straw of selected rice varieties on the germination and growth of barnyard grass, a major rice weed,.

Materials and Methods

Experiment 1: Effect of aqueous extracts of rice leaves
Aqueous extracts of 12 rice varietes/lines (Acheh Puteh, Anak China, Anak Ikan, Anak Ikan China, MR 14, MR 15, MR 59, Makmur, Manik, Seberang, Sekencang and Y1021) were

made by agitating cut fresh leaves of the varieties for 24 hr on an orbital shaker (150 rpm, Firstek Scientific Model S102; Hsin Chuang, Taiwan, ROC) after being placed in a flask with 100 ml distilled water. The extract was strained to remove solid materials. Four concentrations of the extracts (full strength (50 g/l), three-quarter strength (37.5 g/l), half strength (25.0 g/l) and one-quarter strength (12.5 g/l) were prepared by diluting the extract with distilled water immediately before use. Three concentrations of polyethylene glycol (PEG) 8000 MW of 1.25, 2.5 and 5% were also included as controls to see possible osmotic effects of the extracts. A distilled water control was included for comparing the effects of extracts or PEG concentrations. The pH of three PEG concentrations ranged from 5.50 to 5.67 and that of extracts ranged from 6.42 to 7.09.

Twenty five seeds of barnyard grass were placed in separate petridishes lined with 9 cm Whatman No.2 filter papers. Five milliliters of extract (or distilled water for controls) were added to wet each filter paper. All treatments were replicated four times in a CRD design. The covered petridishes were then incubated at 25°C with a day: night light ratio of 13 hr:11 hr in a laboratory at Universiti Kebansaan Malaysia. The germination percentage (GP), radicle length (RL), plumule length (PL) and fresh weight (FW) of the weed seedlings were recorded 5 days after seed placement. The seeds with 2 mm radicle length were considered as germinated seeds.

Experiment 2: Effects of decomposing rice debris

Air-dried culms and leaves of the varieties were cut into small pieces (4 – 6 cm) and dried in an electric oven at 60°C for 7 days. The oven-dried plant samples were then again cut into smaller pieces (0.5 – 1.0 cm) before mixing with soil (sandy loam, pH = 5.5 and contained N% = 0.030, P = 1300 mg/l, K = 12.90 mg/l and Fe = 106.02 mg/l). The soil (500 gm) and plant debris mixture was then put in black polythene bags (8 cm x 5 cm), which were placed on plastic saucer to prevent loss of water soluble toxic substances. Four concentrations of plant debris *e.g.* 0, 2.5, 5.0 and 10.0 g dry weight per 1000 g soil were used in the experiment. Fifty ml of distilled water was added to each polybag everyday to keep the soil moist for 10 days before placement of weed seed.

Twenty five non-dormant seeds of barnyard grass were sown on the soil surface of each polybag and the polybags were watered regularly (twice a day) to maintain moisture content at 80% field capacity. There were four replications for each treatment placed in a RBD design. All polybags were placed in a greenhouse having a temperature of approximately 25°C at night and 34°C during the day.

The number of seed germinated was counted at 8 days and 28 days after sowing. The seeds with 2 mm plumule above the soil surface were considered as germinated. The plant heights of ten randomly selected weed seedlings were measured at 30 days after sowing. The dry weights of selected 10 seedlings were recorded after drying for seven days in oven at 70°C. Average inhibition (%) = (GP + SL + DW)/3 was calculated. The analyses of variances of the data (of both the experiments) were done by using the statistical program MINITAB. The pooled mean values were separated on the basis of Least Significant Differences (LSD) at p=0.05.

Results and Discussion

Effect of aqueous extracts of rice leaves

Effect on seed germination: On an average, seed germination ($p<0.01$) of barnyard grass was reduced by 12% due to the treatments. The highest reduction was with Y1021 (35.6%), followed by Anak China (22.8% reduction) and the lowest reduction was with MR 14 (2.6%) (Table 1). Seed germination of barnyard grass was increased in Manik (a 8.2% increase) and

MR 14 (8.4% increase). Chung *et al.* (2000) also illustrated differences in rice varieties in their effects on seed germination of barnyard grass. The difference of effects within the concentrations used in this study was not significant (Table 2). Ebana *et al*. (2001) used leaf extract of rice cultivar PI312777 and Rexmont on seed germination of lettuce where they found no difference among the extracts. Polyethylene glycol (PEG) did not affect seed germination, radicle length, plumule length and fresh weight of the test weed (data not presented), which indicated that the reduction was not caused by osmotic potential.

Table 1. Average inhibition of aqueous leaf extracts of rice varieties on germination and growth of barnyard grass

Rice variety	Germination		Radicle length		Plumule length		Fresh weight		Average inhibition (%)
	No.	% reduction	Length (cm)	% reduction	Length (cm)	% reduction	Weight (mg)	% reduction	
Acheh Puteh	21.7	6.86	2.34	17.02	4.91	4.47	73.19	10.74	9.77
Anak China	18.0	22.75	2.27	19.50	2.92	43.19	59.75	27.13	28.14
Anak Ikan	21.7	6.86	3.10	-9.93	4.71	8.36	72.00	12.19	4.37
Anak Ikan China	22.1	5.15	3.21	-13.83	4.81	6.42	77.75	5.18	0.73
MR 14	22.7	2.58	3.94	-39.72	4.88	5.06	83.19	1.45	-8.38
MR 15	18.3	21.46	2.06	26.95	2.81	45.33	56.50	31.10	31.21
MR 59	21.7	6.86	3.79	-34.40	4.79	6.81	75.50	7.93	-3.20
Makmur	22.1	5.15	3.34	-18.44	4.05	21.21	63.37	22.72	7.66
Manik	22.3	4.28	4.06	-43.97	4.90	4.67	80.06	2.36	-8.17
Seberang	21.5	7.72	3.44	-21.98	4.65	9.58	77.44	5.56	0.27
Sekencang	19.7	15.45	2.55	9.57	3.31	35.60	67.44	17.76	19.59
Y1021	15.0	35.62	1.50	46.81	1.95	62.06	49.81	39.25	45.94
LSD (5%)	2.24	9.62	1.18	41.80	0.58	11.31	12.36	15.07	13.99
Mean	20.57	11.73	2.97	-5.20	4.06	21.06	69.67	15.28	10.66
Control (No rice)	23.3		2.82		5.14		82.00		

Sign "-"indicates no inhibition rather increase

Effect on radicle and plumule length: A significant reduction in radicle and plumule lengths was observed with the straw of varieties Y 1021 (46.8%) and MR 15 (27.0%) (Table 1). However, the radicle lengths of the weed were increased ranging from 9.9% to 44%, in comparison to control with the application of leaf extracts from 7 rice varieties. Ebana *et al*. (2001) remarked a suppressive effect of similar concentrations of leaf extracts on the root elongation of lettuce but in this study rice leaf extracts did not affect the radicle length of barnyard grass (Table 2). In case of plumule length, although significant effects of variety and concentration were found ($p<0.01$), the significance of extract concentrations disappeared when the control treatment was excluded. On an average, plumule length was reduced by 21% due to allelopathic effects of rice leaf extracts, with the highest reduction in Y 1021 (62.0%), followed by Anak China (43.0%) and the lowest reduction under Acheh Puteh (4.5%) and Manik (4.7%) (Table 1). The interaction effect of variety and concentrations was non-significant in case of radicle length but was significant in case of plumule length.

Effect on fresh weight: On an average, fresh weight of barnyard grass was reduced by 15.3% due to allelopathic effects of rice leaf extracts (Table 2). The highest reduction was noticed in Y 1021 (39.2%), followed by MR 15 (31.1%). The concentrations used in this study did could not induce differences in seed germination and growth of barnyard grass. When average inhibition percentage was calculated it was seen that almost 11% inhibition in germination and growth of weed seedlings occurred due to effects of aqueous extracts of rice leaf. The variety Y 1021 produced the highest suppressive effects (46.0% inhibition), followed by MR 15 (31.2% inhibition) and the lowest suppression occurred due to Seberang (> 1% inhibition).

Ebana *et al*. (2001) with similar concentration of rice leaf extracts found significant differences in root elongation of barnyard grass.

Table 2. Average inhibition of different concentrations of aqueous extracts of rice leaves on germination and growth of barnyard grass

Extract concentrations (g/l of water)	Germination		Radicle length		Plumule length		Fresh weight		Average inhibition (%)
	No.	% reduction	Length (cm)	% reduction	Length (cm)	% reduction	Weight (mg)	% reduction	
1.25	20.6	11.59	2.88	-2.13	4.06	21.01	69.85	14.82	11.32
2.50	20.8	10.73	3.02	-7.09	4.06	21.01	70.60	13.90	9.64
3.75	20.2	13.30	2.77	1.77	4.09	20.43	69.77	14.91	12.60
5.00	20.7	11.16	3.18	-12.76	4.03	21.60	68.44	16.54	9.13
LSD (5%)	NS	NS	NS	NS	NS	NS	NS	NS	NS
Mean	20.57	11.69	2.96	-5.05	4.06	21.01	69.67	15.04	10.67
Control (No rice)	23.3		2.82		5.14		82.00		

NS = non significant, "-" indicates no inhibition rather increase

Effects of decomposing rice debris

Effect on seed germination: On an average, seed germination was reduced by over 8% due to effects of decomposing plant debris of rice varieties (Table 3). The highest reduction was found with Anak Ikan (22.2%), followed by Acheh Puteh (15.5%) and the lowest reduction was noticed with Y 1021 (>1%). The seed germination reduced progressively with increasing debris concentrations (Table 4). Therefore, water-soluble allelochemicals might be released and affect seed germination of the weed. Similar inhibitory effects of rice straw mixture with sand on the emergence of barnyard grass were also noted by Chung *et al*. (2000) in Korea.

Table 3. Average inhibition of decomposing debris of rice varieties on the germination and growth of barnyard grass

Rice variety	Germination		Seedling height		Seedling dry weight		Average inhibition (%)
	Number	% reduction	Length (cm)	% reduction	Weight (mg)	% reduction	
Acheh Puteh	15.21	15.50	10.44	28.64	92.50	47.89	30.68
Anak China	15.29	15.05	11.17	23.65	100.83	43.19	27.30
Anak Ikan	14.00	22.22	09.93	32.12	82.50	53.52	35.95
Anak Ikan China	16.46	8.55	11.53	21.19	110.83	37.56	22.43
MR 14	17.33	3.72	12.28	16.06	97.50	45.07	21.62
MR 15	17.17	4.61	10.90	25.49	92.50	47.89	26.00
MR 59	18.54	-3.00	11.68	20.16	115.00	35.21	17.46
Makmur	15.87	11.83	12.40	15.24	112.50	36.62	21.23
Manik	17.37	3.50	11.47	21.60	100.83	43.19	22.76
Seberang	16.83	6.50	10.96	25.08	108.33	38.97	23.52
Sekencang	16.04	10.89	13.60	7.04	146.67	17.37	11.77
Y1021	17.83	0.94	12.31	15.86	125.83	29.11	15.30
LSD (5%)	4.59	26.63	2.56	17.50	45.34	25.54	18.89
Mean	16.49	8.35	11.55	21.01	89.87	39.63	23.00
Control (No rice)	18.00		14.63		177.50		

The sign "-" indicates no inhibition, but an increase

Effect on seedling height: On an average, weed seedling height was reduced by 21% due to the effects of plant debris. The highest reduction of seedling height was noticed with Anak Ikan (32.1%), followed by Acheh Puteh (28.6%) and the lowest reduction was with Sekencang (7.0%) (Table 4). The results were similar to that of Chung *et al*. (2000) who reported that some Korean varieties of rice exhibited strong inhibitory effects on seedling height of barnyard grass.

Effect on seedling dry weight: On an average, seedling dry weights of the weed were reduced by 40% due to incorporation of rice debris with the soil in comparison to the control. Among the varieties, the highest reduction was noted with Anak Ikan (53.5%), followed by Acheh Puteh (47.9%) and the lowest reduction was found with Sekencang (7.4%) (Table 4). When the average inhibition percentages were calculated, it was observed that the variety Anak Ikan was highly suppressive (35.9% inhibition) to the growth of barnyard grass, followed by Acheh Puteh (30.7% inhibition) and the lowest suppressive variety was noted with Sekencang (11.8% inhibition). Similar findings were also observed by Chung *et al*. (2001) in Korea.

Table 4. Average inhibition of different concentrations of rice debris on the germination and growth of barnyard grass

Concentration of rice debris (g/1000 g soil)	Germination		Seedling length		Seedling dry weight		Average inhibition (%)
	Number	% reduction	Length (cm)	% reduction	Weight (g)	% reduction	
2.50	17.00	5.55	12.97	11.35	92.50	47.89	21.60
5.00	16.58	7.89	11.98	18.11	100.83	43.19	23.06
10.00	15.91	11.61	09.71	33.63	82.50	53.52	32.92
LSD (5%)	1.52	NS	0.81	5.54	14.34	8.08	5.98
Mean	16.50	8.35	11.55	21.03	91.94	48.20	25.86
Control (No rice)	18.00		14.63		177.50		

NS = non significant

The present study suggests that many Malaysian rice varieties, especially Anak Ikan, Acheh Puteh, Anak China, Anak Ikan China, MR 15, MR 14, Manik, Makmur, and Seberang are allelopathic in nature rendering a 20 to 36% reduction of barnyard grass growth. Allelopathic effects of these varieties on weed growth are mainly exhibited through decomposition of rice straw in the soil. Aqueous extracts are found inhibitory to weed growth when the leaves are collected at the early growth stages *i.e.* at 6-leaf stages and with higher concentrations (Ebana *et al*. 2001).

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ALLELOPATHIC POTENTIAL OF CITRUS FOR WEED MANAGEMENT

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Abstract: The allelopathic potential of *Citrus junos* waste from food processing industry after juice extraction was investigated. Powder of *C. junos* waste inhibited the growth of roots and shoots of alfalfa (*Medicago sativa*), cress (*Lepidium sativum*), lettuce (*Lactuca sativa*), crabgrass (*Digitaria sanguinalis*), timothy (*Phleum pratense*) and ryegrass (*Lolium multiflorum*). Significant reductions in the growth of roots and shoots were observed as the powder concentration increased. In order to isolate the growth inhibitory substance in the waste; neutral, acidic and basic fractions were separated from the methanol extract of *C. junos* waste. All fractions inhibited the growth of lettuce seedlings, but by far the greatest inhibition was observed with the neutral fraction. Thus, the neutral fraction was further purified and an allelopathically active substance was isolated. The structure of the substance was determined from high-resolution MS and ^{1}H- and ^{13}C-NMR spectral data as abscisic acid-β-D-glucopyranosyl ester (ABA-GE). The ABA-GE inhibited shoot and root growth of lettuce seedlings at concentrations greater than 0.3 μM, and the concentrations for 50 % inhibition of shoot and root growth were 2.3 and 1.4 μM, respectively. The concentration of ABA-GE in *C. junos* waste was determined to be 17.9 mg kg^{-1} dry weight. Its concentration in *C. junos* waste appears to account mostly for the observed inhibition of tested plant seedlings. These results indicate that *C. junos* waste is allelopathic with potential for use in agriculture to suppress weed emergence, which should be investigate further in the field.

Key words: *Citrus junos,* abscisic acid-β-D-glucopyranosyl ester, allelopathy, waste, weed management.

Introduction

The place of origin of citrus is Southeast Asia and many kinds of citrus species are currently cultivated in the world. Some citrus fruits are processed into juice as an ingredient in beverage, sauces and salad dressings for their special flavor. After juice extraction, these fruit pulp is mostly dumped as waste at large expense. The manipulation of food processing wastes is now becoming a very serious environmental issue. It seemed, therefore, worthwhile seeking how to make use of the waste of citrus fruits.

Citrus fruits contain a variety of biologically active compounds including terpenoids and flavonoids (Davies 2000; Iwase *et al.* 2000; Ogata *et al*. 2000; Uchiyama *et al.* 2004). It was also found that an aqueous methanol extract of *Citrus junos* fruit peel inhibited growth of several plant species including weed species (Fujihara and Simizu 2003). These findings suggest that *C. junos* peel possesses potent allelopathic activity and the peel may be effective as a weed suppressive agent. In this study, the allelopathic potential of *C. junos* waste was determined under laboratory conditions, and the main allelopathic substance present in *C. junos* waste was isolated.

Materials and Methods

Material
The fruit waste (peel and pulp) of *Citrus junos* Sieb. ex Tanaka after juice extraction was obtained from a local food processing company. The fresh waste was freeze-dried and powdered in a mortar and pestle.

Bioassay

Six species, alfalfa (*Medicago sativa* L.), cress (*Lepidium sativum* L.), lettuce (*Lactuca sativa* L.), crabgrass (*Digitaria sanguinalis* L.), timothy (*Phleum pratense* L.) and ryegrass (*Lolium multiflorum* Lam.) were chosen for bioassay as test plants because of their known germination behaviors. Powder of *C. junos* waste was mixed with sterilized quartz sand (1 g) in a 3.5-cm Petri dish and moistened with 1 ml of distilled water according to the method of Shilling *et al.* (1992).

Seeds of the test species were germinated on filter paper (No. 2, Toyo, Japan) in the dark at 25°C for 1- 3 days. Ten germinated seeds of each species were then arranged on the surface of sand in each Petri dish. Seedlings were incubated in the dark at 25°C for 48 h, and shoot and root length of these seedlings was measured. The experiment was repeated five times with 10 plants for each determination.

Extraction and separation of C. junos waste

Powder of *C. junos* waste was extracted with 80% aqueous methanol for three days at 4°C. After filtration using filter paper, the filtrate was concentrated to give an aqueous extract. The extract was partitioned five times with an equal volume of dichloromethane and all five dichloromethane phases were combined. The biological activities of the dichloromethane and aqueous extract were determined by the lettuce bioassay.

The aqueous phase was adjusted to pH 8.3 with 1 M KOH and passed through an anion exchange column of Dower (1x8, Cl^-) and the column was washed with water. The effluent and washings were directly passed through a cat-ion exchange column of Dowex (50Wx8, H^+). Then, the effluent and washings were combined to form a neutral fraction. The anion exchange column was eluted with 2 M formic acid and the eluate was collected as the acidic fraction. The cat-ion exchange column was eluted with 2 M HCl and eluate was collect as the basic fraction. The biological activities of neutral, acidic and basic fractions were determined using the lettuce bioassay.

Purification of active component

The neutral fraction was loaded onto a column of Diaion HP20, and eluted with water, 20, 40, 60 and 80% (v/v) aqueous methanol, and methanol. The biological activity of the fractions was determined by using the lettuce bioassay, and the inhibitory activity was found in fractions obtained by elution with 60 and 80% aqueous methanol. After evaporation, the residue was further purified by a column of Sephadex LH-20, and eluted with water, 20, 40, 60 and 80% (v/v) aqueous methanol, and methanol. The inhibitory activity was found in fractions obtained by elution with water and 20% aqueous methanol. The fractions were combined and evaporated, and the residue was dissolved in water and loaded onto reverse-phase C_{18} Sep-Pak cartridges. The cartridge was eluted with water, 20, 40, 60 and 80% (v/v) aqueous methanol, and methanol. The activity was found in fractions obtained by elution with 20 and 40% aqueous methanol. After evaporation, the active residue was finally purified by HPLC (10 mm i.d. x 50 cm, ODS AQ-325; YMC Ltd, Kyoto, Japan) eluted at a flow rate of 2 ml min^{-1} with 30% aqueous methanol, detected at 220 nm. Inhibitory activity was only found in a peak fraction eluted between 147- 151 min, yielding an active component (4.9 mg) as a colorless resin.

Quantification of abscisic acid-β-D-glucopyranosyl ester

The powder of *C. junos* fruit waste was extracted with methanol and concentration of abscisic acid-β-D-glucopyranosyl ester in the extraction was determined using HPLC described above. The experiment was repeated three times with four assays for each determination.

Results and Discussion

Biological activity of C. junos waste powder

C. junos powder suppressed the growth of six test plants at concentrations greater than 0.1 - 0.3 mg per Petri dish for roots and shoots (Figure 1). Increasing the concentrations increased the inhibition of the root and shoot growth of all test plant species, although its effectiveness differed with species. These results suggest that *C. junos* waste possesses allelopathic activity and may work as a plant inhibiting agent.

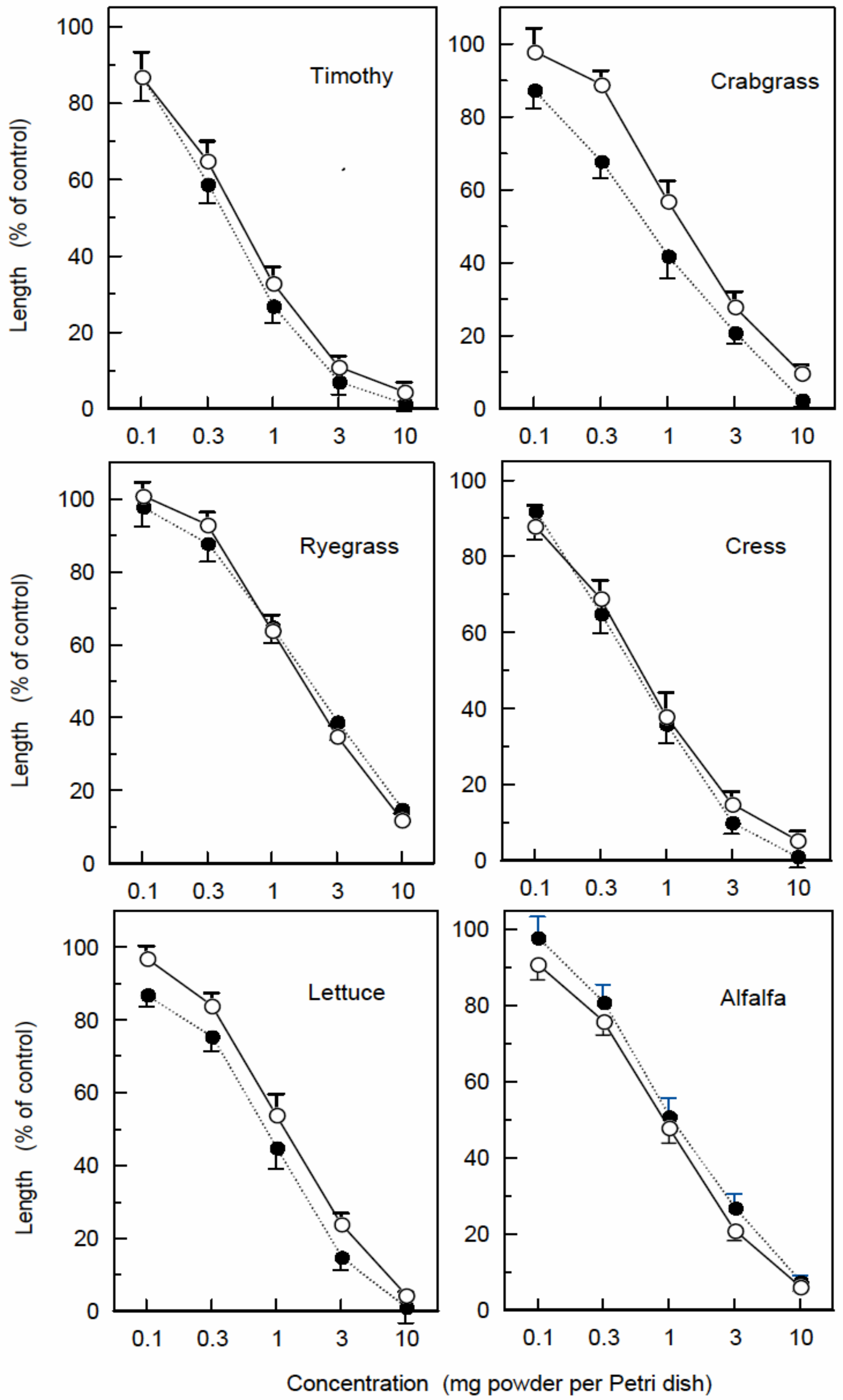

Figure 1. Effects of *C. junos* waste powder on the roots (●) and shoots (○) growth of timothy, crabgrass, ryegrass, cress, lettuce and alfalfa seedlings.

Allelopathic potential of aqueous methanol extract of C. junos waste

The powder of *C. junos* waste was found to possess potent allelopathic activity (Figure 1), the waste was extracted with aqueous methanol and the extract was partitioned with dichloromethane. Both the aqueous and dichloromethane fractions inhibited shoot and root growth of lettuce seedlings. However, the inhibitory activity in the aqueous fraction was much greater than that of the dichloromethane fraction. At a concentration of 10 mg dry weight peel equivalent ml^{-1}, the aqueous and dichloromethane fractions caused 72 and 38% inhibition of the lettuce shoots and 86 and 35% inhibition of the lettuce roots, respectively, compared to the length of control seedlings. The aqueous fraction was separated by ion exchange columns to neutral, acidic and basic fractions, and their biological activities were determined. All three fractions suppressed shoot and root growth of the lettuce seedlings, with the most marked inhibition being achieved by the neutral fraction. Thus, purification and isolation of allelopathic substances proceeded using only the neutral fraction.

Isolation and identification of allelochemical in C. junos waste

The neutral fraction was purified by columns of Diaion HP20 and Sephadex LH-20, C_{18} Sep-Pak cartridge and HPLC. An allelopathically active substance was isolated as a colorless residue. The molecular formula of the substance was determined to be $C_{21}H_{31}O_9$ (*m/z* 427.1987; calculated for 427.1968) based on its high-resolution mass spectrum. The ^{1}H NMR and ^{13}C NMR spectra of the substance are shown in Table 1. From a comparison of these data with those reported in the literature (Koshimizu *et al.* 1968; Loveys and Milborrow, 1981; Kato-Noguchi *et al*. 2002), the substance was identified as abscisic acid-β-D-glucopyranosyl ester (ABA-GE).

Biological activity

The biological activities of ABA-GE and (+)-ABA were determined with lettuce seedlings. At concentrations greater than 0.3 and 0.1 μM, respectively, ABA-GE and (+)-ABA inhibited the growth of the lettuce shoots and roots. When percentage length of test plants was plotted against logarithm of the concentrations, all concentration-response curves were linear between 20-80% inhibition. The concentrations required for 50% inhibition of the lettuce shoots in the assay (defined as I_{50}), as interpolated from the concentration-response curves, were 2.3 and 0.84 μM for ABA-GE and (+)-ABA, respectively, and those of the lettuce roots were 1.4 and 0.48 μM for ABA-GE and (+)-ABA, respectively. Comparing I_{50} values, the inhibitory activity of ABA on the shoots and roots, respectively, was 2.7 and 2.9-fold greater than that of ABA-GE. Thus, the activity of ABA-GE on the shoots and roots of the lettuce seedlings was about one third of that of (+)-ABA.

The ABA-GE was considered to be a physiologically-inactive main conjugated ABA in plants, and an end-product of ABA metabolism rather than storage or transport form (Milborrow 1970 and 1978; Neill *et al.* 1983; Zeevaart, 1983; Lehman and Vlasov, 1988). However, recently an ABA-GE-cleaving enzyme, apoplastic β-D-glucosidase was detected (Dietz *et al*. 2000). The enzyme releases free ABA from the ABA-GE, which indicates ABA-GE may have important physiological functions in plants. The effectiveness of ABA-GE on the inhibition of growth coupled to the discovery of this key enzyme (Dietz *et al.* 2000) suggest that exogenously applied ABA-GE may be absorbed by plant roots and hydrolyzed by apoplastic β-D-glucosidase, with the subsequent release of ABA, which would potentially inhibit plant growth.

Concentration of ABA-GE in C. junos waste

The concentration of ABA-GE in *C. junos* waste powder was determined to be 17.9 mg kg^{-1} dry weight (42 μmol). At concentrations greater than 0.3 μM, ABA-GE inhibited the growth of the lettuce shoots and roots. Thus, ABA-GE concentration in *C. junos* waste appears to account mostly for the observed inhibition of tested plant seedlings. In addition, other potentially inhibitory compounds might be in the *C. junos* waste powder.

Conclusions

Many investigations have attempted to exploit allelopathy of plants for weed control purposes in a variety of agricultural settings (Inderjit, 1996; Seigler, 1996; Duke *et al.* 2000). Synthetic chemical herbicides may continue to be a key component in most integrated weed management systems, but controlling weeds through allelopathy is one strategy to reduce herbicide dependency (Putnam, 1988; Einhellig, 1996; Weston, 1996; Duke *et al*. 2000). The present research suggests that *C. junos* waste powder may be potentially useful for weed management in an agricultural field setting, which should be investigate further in the field for practical application of the powder. There may be the potential benefits of turning a previously useless waste product into a valuable soil amendment and improving the economics of the food processing industry.

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STRESS-INDUCED "MOMILACTONE B" ACCUMULATION IN RICE SEEDLINGS AND RICE RHIZOSPHERE

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Abstract: Injury and heavy metal stresses increased momilactone B secretion from rice roots into the medium. The UV-irradiation also increased the concentration of momilactone B in shoots and roots of rice seedlings. The concentration in 90-min UV-irradiated shoots and roots, respectively, was 31.8- and 3.6-fold greater than that of non-irradiated shoots and roots. Accumulation of momilactone B occurred in medium in which UV-irradiated seedlings were grown. The UV-irradiation increased not only production of momilactone B in rice seedlings but secretion of momilactone B into the rice rhizosphere. As momilactone B acts as an antimicrobial and allelopathic agent, secretion of momilactome B into the rhizosphere may provide a competitive advantage for root establishment through local suppression of soil microorganism and inhibition of the growth of competing plant species.

Key words: Allelopathy, momilactone B, *Oryza sativa*, phytoalexin, rhizosphere, UV-irradiation, secretion.

Introduction

The plant rhizosphere is a densely populated area in which plant roots must compete with invading root systems of neighboring plants for space, water, and mineral nutrients, and with other soil-bore organism, including bacteria and fungi. A great number of compounds are screted into rhizosphere as root exudates, and play an active role in the regulation of symbiotic and protective interactions with microbes and neighboring plant species (McCully 1999; Hawes *et al.* 2000; Bais *et al.* 2004). For example, isoflavonoids and flavonoids present in the root exudates of a variety of leguminous plants activate the *Rhizobium* genes responsible for the nodulation process (Peters *et al.* 1986). Root exudates also act as antimicrobials against rhizospheric microflora, providing the plant with defensive advantages (Hawes *et al*. 2000; Bais *et al*. 2004).

Recent research has shown that , momilactone B is constitutively secreted from the roots of rice plants (Kato-Noguchi and Ino 2003), which suggests that momilactone B may play an important role in defense mechanism in rice rhizosphere. This paper described that injury, heavy metal and UV-irradiation stresses increased the secretion of momilactone B from rice plants into the rice rhizosphere.

Materials and Methods

Plant materials and injury stress treatment

Seeds of rice (*Oryza sativa* L. cv. Koshihikari) were surface sterilized in 70 % (v/v) aqueous ethanol for 15 min, rinsed five times with distilled water and allowed to germinate on a sheet of moist filter paper at 25 °C with a 12-h photoperiod in a growth chamber. Light was provided from above with a white fluorescent tube (120 µmol $m^{-2}s^{-1}$ at plant level; FL40SBR, National, Tokyo, Japan). All further manipulations were carried out under sterile conditions. After 3 days, uniform seedlings, in groups of 100, were transferred onto a sheet of plastic mesh which was floated on distilled water (300 ml) in a plastic container, and grown for 7 days at 25°C with a 12-h photoperiod. Rice seedlings were injured by cutting the first leaves with scissors, and the seedlings were incubated for 5 days at 25°C with a 12 hr photoperiod. For determination of momilactone B, the medium in the container was collected.

Heavy metal treatment

Rice seeds were germinated for 3 days and hydroponically grown for 10 days as described above. Rice seedlings and plastic mesh were together transferred onto 1 mM $CuCl_2$ solution (300 ml) in plastic container, and the seedlings were incubated for 5 days at 25°C with a 12 hr photoperiod. For determination of momilactone B, the medium in the container was collected.

UV irradiation

Rice seeds were germinated for 3 days and hydroponically grown for 10 days as described above. Rice seedlings were then exposed to UV-irradiation of 20, 40 or 90 min per day (emission peak 253 nm; 10 µmol $m^{-2}s^{-1}$ at plant level; GL15, Toshiba, Tokyo) for 5 days. The plastic container was covered with aluminum film so that only rice shoots were exposed. For determination of momilactone B, rice seedlings were harvested and the medium in the container was collected at day 0, 2, 5 after UV-irradiation.

Determination of momilactone B in medium and seedlings

The medium in the container was filtered and the filtrate was loaded onto a column of synthetic polystyrene adsorbent. Momilactone B was then separated and quantified as described by Kato-Noguchi *et al*. (2002).

Rice shoots or roots (*ca*. 10 g fresh weight) were homogenized with 80 % (v/v) aqueous methanol and the homogenate was filtered through filter paper. The residue was homogenized again with methanol and filtered. The two filtrates were combined and evaporated in vacuo at 35°C to give an aqueous residue. The aqueous residue was loaded onto a column of synthetic polystyrene adsorbent and purified, and momilactone B was quantified as described by Kato-Noguchi *et al.* (2002).

Results and Discussion

Effect of injury stress on the momilactone B secretion from rice

The concentration of momilactone B was 3.1-fold greater in the medium of the stressed seedlings than that of the non-stressed seedlings (Figure 1), which suggests that the injury stress may increase momilactone B secretion from rice seedlings into the medium.

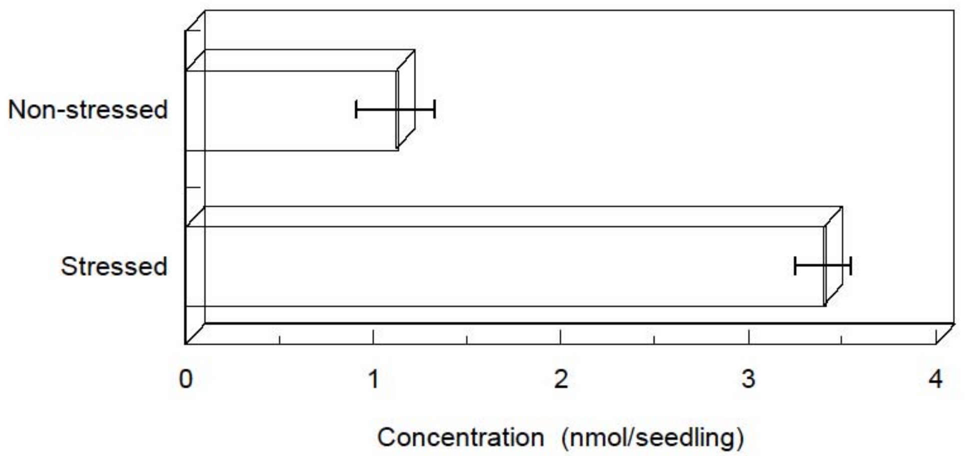

Figure 1. Effect of injury stress on momilactone B concentration in rice medium. Momilactone B was determined at 5 days after stress tretment. Means ± SE from three independent experiments with three assays for each determination (n = 9) are shown.

Effect of heavy metal on the momilactone B secretion from rice

The concentration of momilactone B was 3.7-fold greater in the medium containing $CuCl_2$ than that in control medium (Figure 2), which suggests that heavy metal stress may increase momilactone B secretion from rice seedlings into the medium.

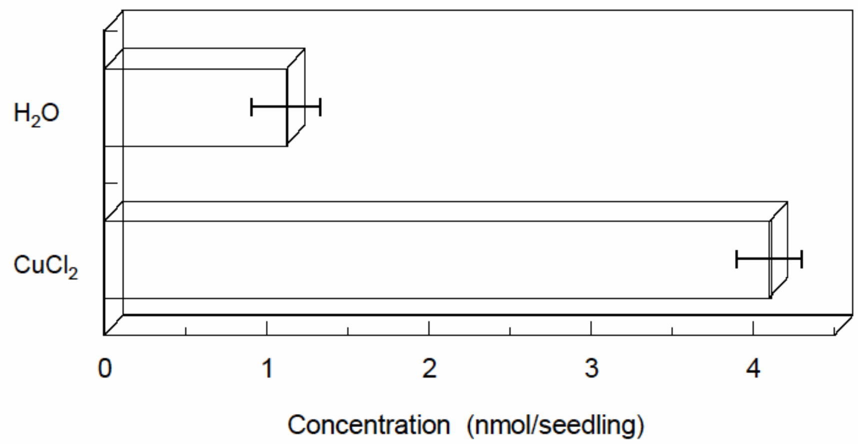

Figure 2. Effect of $CuCl_2$ on momilactone B concentration in rice medium. Momilactone B was determined after incubation in 1mM $CuCl_2$ or H_2O for 5 days. Means ± SE from three independent experiments with three assays for each determination (n = 9) are shown.

Effect of UV-irradiation on the concentration of momilactone B in rice seedlings

UV-irradiation increased the concentration of momilactone B in shoots and roots of rice seedlings and increasing the irradiation increased the concentration, with the concentration in shoots always much greater than that in roots. The concentration in 90-min UV-irradiated shoots and roots, respectively, was 31.8- and 3.6-fold greater than that of non-irradiated (0 min) shoots and roots. The UV-irradiation did not significantly affect the root and shoot length, and fresh and dray weight of the seedlings.

Momilactone B is synthesized from geranylgeranyl diphosphate. The cyclization step producing *syn*-copalyl diphosphate from geranylgeranyl diphosphate is the branch point in the biosynthesis of momilactone B and gibberellins (Wilderman *et al.* 2004 Jung *et al.* 2005). UV-irradiation induced gene *OsCyc1* encoding *syn*-copalyl diphosphate synthase (Otomo *et al.* 2004). Therefore, the production of momilactone B was probably increased by UV-irradiation due to increasing activity of *syn*-copalyl diphosphate synthase. However, momilactone B has only been known to increase in rice leaves by UV-irradiation (Kodama *et al.* 1988a; Otomo *et al.* 2004), but it was found that momilactone B also increased in the roots by UV-irradiation. Only rice shoots were exposed to UV-irradiation as described in the section of Materials and methods. Thus, momilactone B in roots was increased without exposure of the roots to UV-irradiation, which suggests that momilactone B in rice roots may be transported from rice shoots. This may be the first finding of momilactone B transportation.

Effect of UV-irradiation on the momilactone B secretion from rice

The concentration in medium of UV-irradiated seedlings at day 0 was similar to that in medium of non-UV-irradiated seedlings (Figure 3). However, accumulation of momilactone B occurred in the medium in which UV-irradiated seedlings were grown. At day 5, the concentration in the medium of UV-irradiated seedlings became 2.5-fold grater than that in the medium of UV-irradiated seedlings at day 0, and this accumulated momilactone B (3.4

nmol per seedling) in the medium was 47% of momilactone B in the seedlings (6.8 and 0.44 nmol per shoot and root, respectively) at day 5. High level of momilactone B in the medium compared with the level in the seedlings suggests that this exudation of momilactone B from rice seedlings may be active.

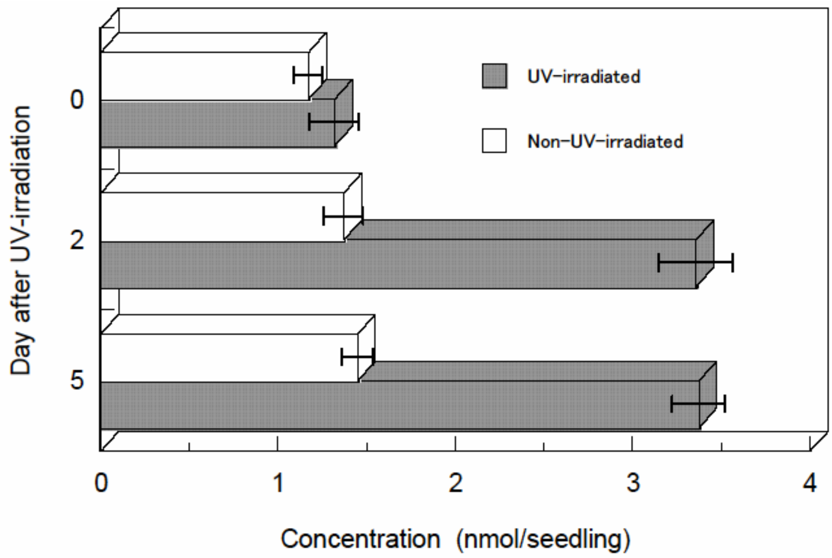

Figure 3. Effect of UV-irradiation on momilactone B concentration in rice medium. Momilactone B was determined after UV-irradiation for 5 days. Means ± SE from three independent experiments with three assays for each determination (n = 9) are shown.

In the present experiments, only rice roots were immersed in the medium as described in the section of Materials and Methods. Thus, the rice seedlings probably secrete momilactone B from their roots into the medium. Although mechanisms of the exudation are not well understood, it is suggested that plants are able to secrete a wide variety of compounds from root cells by plasmalemma-derived exudation, endoplasmic-derived exudation, and proton-pumping mechanisms (Hawes *et al.* 2000; Bais *et al.* 2004). Through the root exudation of compounds, plants are able to regulate the soil microbial community in their immediate vicinity, change the chemical and physical properties of the soil, and inhibit the growth of competing plant species (McCully, 1999; Hawes *et al*. 2000; Bais *et al*. 2004).

Conclusions

The results described in the present research suggest that UV-irradiation increased not only production of momilactone B in rice seedlings (Figure 3) but secretion of momilactone B into rice rhizosphere, and momilactone B may be produced in rice shoots and secreted into the medium through rice roots. Injury stress and heavy metal stress also increased secretion of momilactone B into rice rhizosphere (Figures 1 and 2). Momilactone B exhibits antimicrobial properties and are synthesized as a part of defensive response to the bacterial and antifungal activities (Nojiri *et al.* 1996; Takahashi *et al.* 1999; Tamogami and Kodama, 2000). Momilactone B also acts as an allelochemical, as momilactone B was first isolated from seed husks of rice as a growth and germination inhibitor (Kato *et al*. 1973; Takahashi *et al.* 1976). Therefore, secretion of momilactone B into the rhizosphere may provide a competitive advantage for root establishment through local suppression of soil microorganism and inhibition of the growth of competing plant species.

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RICE PLANTS RELEASE "MOMILACTONE B" THROUGHOUT THEIR LIFE CYCLE

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Abstract: Momilactone A and B are contained in the rice plants and released into the neighboring environment from rice throughout its life cycle. The release levels of momilactone A and B from rice increased until flowering initiation, and then decreased. Momilactone A and B inhibited the growth of *Echinochloa crus-galli* and *Echinochloa colonum* at concentrations greater than 1 μM. Comparing I_{50} values (concentrations required for 50 % inhibition of plant growth), the inhibitory activities of momilactone B on the growth of *E. crus-galli* and *E. colonum*, respectively, were 4.7- to 19.2-fold greater than those of momilactone A, which indicates that the effectiveness of momilatone B on the growth inhibition is much greater than that of momilactone A. Momilactone A and B inhibited the growth of rice seedlings at concentrations greater than 100 μM. The effectiveness of momilactone A and B on the growth inhibition of rice seedlings were 1 % of those on the growth of *E. crus-galli* and *E. colonum*, suggesting that the toxicities of momilactone A and B to rice seedlings are probably much less than those to *E. crus-galli* and *E. colonum*. No visible damage to rice seedlings by momilactone A and B was also observed. These results suggest that rice plants may be able to inhibit the growth of their neighboring plants due to the secretion of momolactone A and B into their rhizosphere without serious toxicity of momilactone A and B to rice plants themselves. Thus, momilactone A and B may play an important role in rice defense mechanism in the rhizosphere for the competition with invading root systems of neighboring plants. The involvement of momilactone B for the defense mechanism is greater than momilactone A.

Key words: Allelopathy, growth inhibitor, momilactone B, *Oryza sativa*, rice, root exudates

Introduction

Rice has been extensively studied with respect to the selection of varieties possessing great alleloapthic activity as a part of a strategy for weed control purposes (Olofsdotter, 2001; Olofsdotter *et al.* 1995; 2001; Takeuchi *et al.* 2001). A large number of rice varieties inhibited the growth of several plant species when were grown together under the field or/and laboratory conditions (Dilday *et al.* 1994; 1998; Hassan *et al*. 1998; Mattice *et al*. 1988; Kim *et al.* 1999; Azmi *et al.* 2000), suggesting that rice may produce and release growth inhibiting alleochemical(s) into neighboring environment. However, the chemical identifications involved in the rice allelopathy have not been accomplished.

A potent compound causing the growth inhibitory effect was recently isolated from rice root exudates and identified as momilactone B from its spectral data (Kato-Noguchi *et al.* 2002). Momilactone B was first isolated with momilactone A from rice husks as growth inhibitors involved in seed dormancy (Kato *et al*. 1973; Takahashi *et al*. 1976), and both momilactone A and B were later found in rice leaves and straw as phytoalexins (Cartwright *et al.* 1977; Kodama *et al*. 1988). The productions of momilactone A and B are triggered by pathogen infection, including fungi, bacteria and virus (Cartwright *et al.* 1977; Jung *et al*. 2005), UV irradiation (Kodama *et al*. 1988; Otomo *et al*. 2004). In this paper, release level of momilactone A and B throughout life cycle of rice plants and growth inhibitory activities against weed species and rice itself were determined to know the functions of momilactone A and B in rice allelopathy.

Materials and Methods

Plant materials

Seeds of rice (*Oryza sativa* L. cv. Koshihikari) were surface sterilized in 3 mM solution of Benomyl [methyl-1-(butylcarbamoyl)benzimidazole-2-ylcarbamate] for 24 h, rinsed several times with distilled water and allowed to germinate and grow as described by Kato-Noguchi *et al.* (2002). The germinating seeds, in groups of three, were transferred onto a plastic float on 3 L of one-half-strength of Kimura B nutrient solution (0.18 mM $(NH_4)_2SO_4$, 0.27 mM $MgSO_4{\cdot}7H_2O$, 0.09 mM KNO_3, 0.09 mM KH_2PO_4, 0.05 mM K_2SO_4, 0.18 mM $Ca(NO_3)_2{\cdot}4H_2O$, 0.04 mM Na EDTA-Fe_3H_2O and 0.8 mM Na_2SiO_3; pH 5.0) which was in a plastic pot (24 cm id x 30 (height) cm), and grown for 130 days in a greenhouse under natural day light conditions. The nutrient solution in the pot was renewed every two days and only roots of the plants were immersed in the solution during the incubation.

For determination of momilactone A and B, rice plants and culture solutions in the pot were collected at 0, 15, 30, 45, 60, 75, 80, 85, 90, 110, and 130 days after being transferred to hydroponics.

Determination of momilactone A and B in rice plants

Rice shoots or roots (*ca.* 10 g fresh weight) were homogenized with 100 ml of 80% (v/v) aqueous methanol and the homogenate was filtered. The residue was homogenized again with 100 mL of methanol and filtered. The two filtrates were combined and evaporated in vacuo at 35 °C to give an aqueous residue. The aqueous residue was loaded onto a column of synthetic polystyrene adsorbent, and eluted with distilled water, 20 and 80% (v/v) aqueous methanol, and methanol. After evaporation, the methanol fraction was dissolved in 50% aqueous methanol and loaded onto a reverse-phase C_{18} Sep-Pak cartridge. The cartridge was first eluted with 50% aqueous methanol to remove impurities, and then with methanol to release momilactone A and B. The fraction of momilactone A and B was chromatographed by HPLC (1.0 i.d. x 50 cm, ODS AQ-325; YMC Ltd, Kyoto, Japan; eluted at a flow rate of 2 ml min^{-1} with 70% aqueous methanol, detected at 220 nm). Momilactone A and B were quantified by measuring its peak height on the chromatogram of HPLC.

Determination of momilactone A and B in rice culture solutions

Rice culture solution in the pot was filtered and separated by column of synthetic polystyrene adsorbent and a reverse-phase C_{18} Sep-Pak cartridge, and momilactone A and B was quantified by HPLC as described above.

Bioassay of momilactone A and B

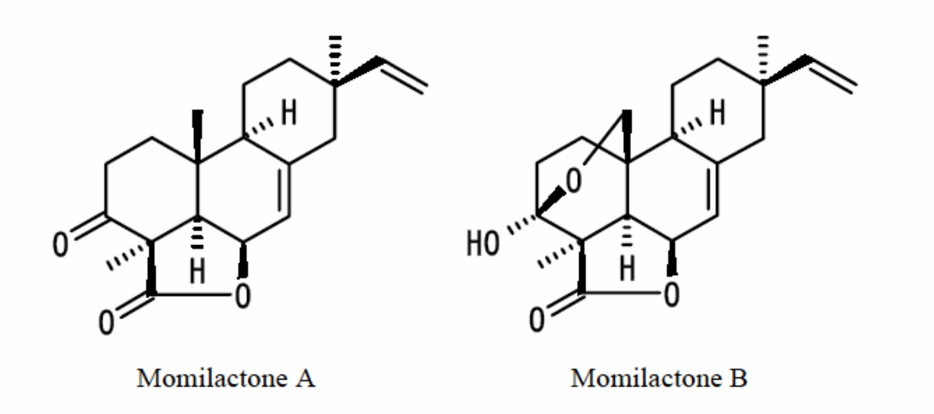
Momilatone A and B were dissolved in a small volume of methanol, added to a sheet of filter paper in a 5.5-cm Petri dish and dried. The filter paper in the Petri dishes was moistened with 2 ml of a 0.05% (v/v) aqueous solution of Tween 20. After sterilization, seeds of *Echinochloa crus-galli* (L.) Beauv, *Echinochloa colonum* (L.) Link and rice were germinated on filter paper in the darkness at 25°C for three days. Ten germinated seeds of each species were separately arranged on the filter paper in the Petri dishes, and incubated in the darkness at 25°C for two days. The length of roots and shoots of these seedlings were measured. Control seedlings were sown on the filter paper moistened with the aqueous solution of Tween 20. Bioassay was repeated six times using a randomized design with 10 plants for each determination (n = 60).

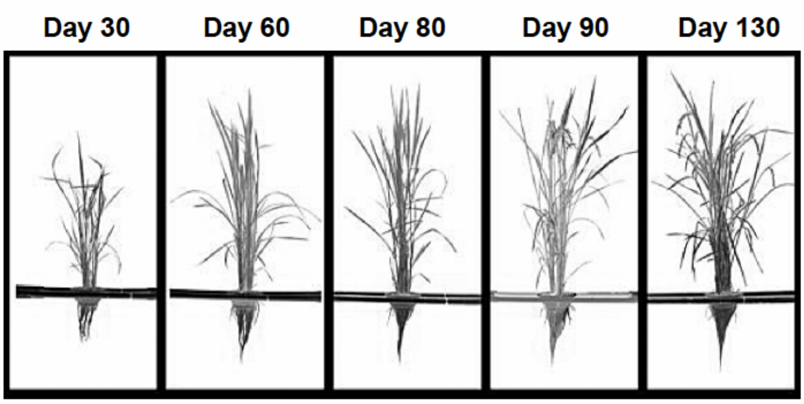
Results and Discussion

Concentrations of momilactone A and B in rice plants

The shoot length of rice plant increased rapidly until day 80 when flowering started Figure 1. At day 120, the plants started to turn yellow from green upon their senescence. Momilactone A and B were found in shoots and roots of rice plants throughout the experiments. The level of momilactone A and B in the shoots and roots increased over the vegetative growth stage until flowering initiation, and then decreased. However, the increases of momilactone A and B in the shoots was much greater than those in the roots. In addition, rice shoots and roots contained much greater momilactone A than momilactone B.

Momilactone A and B are probably synthesized from geranylgeranyl diphosphate in rice leaves (Omoto *et al*. 2004; Wilderman *et al*. 2004). Thus, momilactone A and B in the roots may be transported from the shoots.





Release level	(μg / plant / day)				
Momilactone A	0.02	0.8	1.1	0.8	0.3
Momilactone B	0.04	1.3	2.3	1.4	0.5

Figure 1. Release level of momilactone A and B from rice plants. Means from three independent experiments with three assays for each determination are shown.

Concentrations of momilactone A in culture solutions

Momilactone A and B were found in all culture solutions in which rice plants were grown hydroponically. The culture solution was renewed every two days during experiments, which indicates that rice plants may release momilactone A and B into the solutions throughout their life cycle. The concentrations of momilactone A and B in the culture solution increased rapidly from day 30 until day 80 when flowering started, and then decreased sharply (Figure

1). The release rate of momilactone A and B, respectively, at day 80 was 1.1 and 2.3 μg plant^{-1} day^{-1}, which was 55- and 58-fold greater than that at day 30.

In the present experiments, only rice roots were immersed in the culture solution as described in the section of Material and Methods. Thus, the rice plants probably release momilactone A and B from their roots into the solution. Although concentration of momilactone A in rice plants was much greater than that of momilactone B, release level of momilactone B was greater than that of momilactone A, which suggests that momilactone B was selectively released into the culture solutions by roots than momilactone A.

Inhibitory activity of momilactone A and B on two weed species

The biological activities of momilactone A and B were determined with two weed species. Momilactone A and B, respectively, inhibited the growth of roots and shoots of *E. crus-galli* at concentrations greater than 10 and 1 μM. Momilactone A and B, respectively, also inhibited the growth of roots and shoots of *E. colonum* at concentrations greater than 10 and 1 μM. Increasing the concentrations increased the inhibition of both root and shoot growth of the two weed species. When the root and shoot lengths of the weed plants were plotted against the logarithm of momilactone A and B concentrations, there were logistic concentration-response curves. Comparing I_{50} values (the concentrations required for 50% inhibition of the growth of *E. crus-galli* and *E. colonum* in the assay), the inhibitory activity of momilactone B on the growth of *E. crus-gall* was 7.4-fold greater than that of momilactone A on the growth of *E. crus-gall*, and the inhibitory activity of momilactone B on the growth of *E. colonum* were 19.2-fold greater than that of momilactone A. Thus, effectiveness of momilatone B on the growth inhibition is much greater than that of momilactone A.

Toxicities of momilactone A and B on rice seedlings

Momilactone A and B are produced in rice plants under biotic and abiotic stress conditions (Kodama *et al*. 1988; Jung *et al.* 2005; Otomo *et al.* 2004) and possess growth inhibitory activities. Therefore, toxicities of momilactone A and B to rice seedlings themselves were determined. No visible damage to rice seedlings by momilactone A and B was observed except for occurrence of growth inhibition in the seedlings. Momilactone A and B, respectively, inhibited root and shoot growth of rice seedlings at concentrations greater than 100 and 300 μM. The I_{50} values of momilactone A and B on rice root and shoot were not obtained because of their week inhibitory activities, and the inhibitory activities of momilactone A and B, respectively, on the root and shoot growth of rice seedlings were 1 - 2 % and 0.6 - 2 % of those on the root and shoot growth of *E. crus-galli* and *E. colonum*. Thus, the effectiveness of momilactone A and B on the growth inhibition of rice seedlings was much less than that on the growth of *E. crus-galli* and *E. colonum*, which suggests that the toxicities of momilactone A and B to rice seedlings may be much less than those to the two weed species.

Conclusion

Momilactone A and B inhibited the growth of *E. crus-galli* and *E. colonum* seedlings, but these inhibitory activities on the growth of rice seedlings were weak. Momilactone A and B were found to be released from rice plants over their entire life cycle. Rice usually is planted at a very high density in the paddy field and water in the field is not replaced usually during their growing season (Rao *et al.* 1989; Akita and Tanaka 1992). Thus, accumulation of momilactone A and B released from rice plants may occur in the field conditions sufficiently to inhibit germination and growth of neighboring plants. Both growth inhibitory activity and release level of momilactone B were greater than those of momilactone A.

These results suggest that rice plants may be able to inhibit the growth of their neighboring plants due to the secretion of momolactone A and B into their rhizosphere without serious toxicity of momilactone A and B to rice plants themselves. Thus, momilactone A and B may play an important role in rice defense mechanism in the rhizosphere for the competition with invading root systems of neighboring plants. However, the involvement of momilactone B for the defense mechanism is greater than momilactone A.

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WEED SCOUTING IN PAKISTAN

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Abstract: The diversity of the weed flora present in Pakistan is evident from the 75 plant families having weeds, which is a clear indicator for the occurrences of the diverse cropping systems and ecological zones. However, at the same time, fixed crop rotations have given rise to specific and intense weed problems, necessitating immediate solutions. The present study led to the documentation of major weeds in different ecosystems of Pakistan. Ten currently important weeds of Pakistan, together with colonizers and invaders, and the weeds of future importance to Pakistan were identified.

Key words: Weed flora, diversity, Pakistan

Introduction

The weed problems reflect environmental conditions and agronomic practices unique to individual fields. Keeping records of weed severity and the individual species present in each field can help farmers in better decision making. There needs to be a priority in scouting fields, for determining weed pressure and to identify the weeds present in the field. This will eventually lead to predicting weed severity and knowledgeable management for better yields. Freckleton and Watkinson (2002) argue that weed population dynamics are generally better viewed as resulting from the impacts of broad-scale types of management, as well as temporal variability in population numbers and the significance of chaotic dynamics is likely to be minimal. From as anthropocentric point of view weeds are plants ‘out of place’. It is however not easy to classify them on a narrow set of botanical criteria (*e.g.* morphological, phonological or taxonomic). In consequence, guides to weed species assemblages frequently employ a habitat/land use based classification.

Situations that prohibit species substitution in the composition of the weed flora have to be created in cropping patterns to prevent changes in genetic structure of species population within the weed community and arrest the development of weed abundance while maintaining biodiversity in the agro-ecosystem. However, not all non-crop species in a field are damaging and that within a crop there will be non-competitive biomass. Thus the biological and ecological factors leading to the long term persistence need to be recognized and identified. Enumerating population dynamics (change in abundance) of individual weed species and the weed community structure is important for predicting change. The purpose of this paper is to describe the basis status of weed diversity in Pakistan.

Materials and Methods

Life forms, seed dispersal, reproductive system, life cycle, vegetative reproduction structures, plant population densities, invasiveness, habitat, and uses of the weeds were considered in the present study. The overview presented in this paper outlines the families that have weedy members. The study is a simple representation of the families within a super-order and order that gives an overview of the complex matrix in understandable form.

Results and Discussion

The weeds in Pakistan are represented by over 400 species belonging to 75 plant families. Out of these 16 families are Monocotyledons, 57 are Dicotyledons and two belong to

Pteridophyta. Out of these 40 per cent are perennials and 5.3 per cent acted as biennials and 65 weeds are the principal species of important crops. The 10 most important weeds identified in Pakistan based on this study are presented in Table 1, where *Cyperus rotundus* topped the list.

Table 1. The ten most important weeds of Pakistan.

Scientific Name	Family	Number of habitats
Cyperus rotundus L.	Cyperaceae	18
Cynodon dactylon (L.) Pers.	Poaceae	12
Convolvulus arvensis L.	Convolvulaceae	12
Sorghum halepense (L.) Pers.	Poaceae	9
Avena fatua L.	Poaceae	9
Rumex dentatus L.	Polygonaceae	8
Fumaria indica (Haussk.) Pugsley	Fumariaceae	7
Lathyrus aphaca (L.)	Leguminosae	6
Silybum marianum (L.) Gaertn.	Compositae	6
Echinochloa colona (L.) Link	Poaceae	6

The aggressive colonizers and invaders are and interesting groups of plants including trees shrubs and the low growing mat forming species are presented in Table 2.

Table 2. Aggressive colonizers and invaders in Pakistan

Aggressive colonizers	Invaders
Silybum marianum	*Parthenium hysterophorus*
Euphorbia helioscopia	*Broussonitia papyrifera*
Imperata cylindrical	*Prosopis juliflora*
Typha spp.	*Lantana camara*
Eichhornia crassipes	*Emex australis*
Cirsium arvense	

In this study 127 weeds were identified to be found in the field borders, therefore, every precaution should be taken for prevention of entry into fields. However this is an indicator of diversity in the weed flora. The weeds of family, Compositae, in Pakistan, are represented by approximately 44 genera and more than 60 species (Khalid, 1995).The grass weeds in Pakistan belong to about 36 genera with 50 important weeds infesting crops both in Rabi and Kharif seasons (Khalid, 2004).

Based on the study, the weeds of future importance to Pakistan include *Trianthema portulacastrum* L., *Amaranthus viridis* L., *Digera muricata* (L.) Mart, *Chenopodium album* L**.,** *Conyza bonariensis* (L.) Cronq., *Eclipta alba* (L.) Hassk., *Parthenium hysterophorus* L., *Xanthium strumarium* L., *Leucas cephalotes* (Roth) Spreng., *Medicago polymorpha* L., *Salvia moorcroftiana* Wall., *Vicia sativa* L., and *Anagallis arvensis* var. coerulea (L.) Gouan.

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INTEGRATED WEED MANAGEMENT IN FENNEL (*Foeniculum vulgare* Mill.) GROWN AT FOUR SEED RATES

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Abstract: Field investigations were carried out to evaluate the comparative effectiveness of different weed control strategies in fennel (*Foeniculum vulgare* Mill) grown at four seed rates (4, 6, 8 and 10 kg ha^{-1}). Weed control treatments were pendimethalin (825 g a.i. ha^{-1}), allelopathic sorghum (*Sorghum vulgare* L. Moench) water leachate (15 l ha^{-1}+pendimethalin 412.5 g a.i. ha^{-1}), hand weeding (twice) and a weedy check was maintained as control. A split-plot design with seed rates in the main and weed control strategies in sub plots was used with three replicates at a net plot size of 5.0 m x 2.5 m. Weed flora of the experimental site comprised of canary grass (*Phalaris minor*), lambs quarter (*Chenopodium album*) and swine cress (*Cornopus didymus* L. Smith). Results revealed that increasing the seed rate from 4 to 6 kg ha^{-1} significantly ($p \leq 0.05$) decreased the total weed density and dry weight. A further increase in seed rate did not influence weed density and dry weight. Allelopathic sorghum water leachate+pendimethalin reduced total weed density and dry weight by 86 and 76 % over the control while pendimethalin alone and hand weeding gave 29-4% and 100% suppression of weed density and dry weight, respectively. The yield recorded under all the tested seed rates was similar. Pendimethalin alone, allelopathic sorghum water leachate+pendimethalin and hand weeding gave yield increases of 125, 166 and 350%, respectively over control.

Key words: Integrated weed management, fennel, allelopathy

Introduction

Commercial crops occupying large acreages have received priority of attention for optimizing their factors of production. Some other crops which though wanting study, in view of their importance in other specific spheres, have not received due consideration so far. On account of its numerous pharmaceutical uses fennel (*Foeniculum vulgare* Mill.) ranks among the most important medicinal plants. It is native to Mediterranean and Southern Europe. It can be cultivated up to an altitude of 6000 feet and on marginal lands. In Pakistan, fennel cultivation on large scale is limited by many production constraints. Widespread weed infestation in fennel is of major concern to the growers. All winter weed flora grow profoundly in fields where fennel is cultivated but farmers locally are reluctant to use herbicides because of the perception that this will damage the medicinal value of the produce. Public concerns over safety have put tremendous pressure to assess toxicological and environmental impacts of synthetic chemicals and more stringent protocols are being developed (Dayan *et al.* 1999).

Fast-developing herbicide resistant weed biotypes are also posing a serious threat to agricultural production (Heap, 2002). This necessitates looking for some new approaches for weed management that are effective, environmentally safe and economically viable. Crop allelopathy may be useful to minimize serious problems in the present agricultural production such as environmental pollution, unsafe products, human health concerns, depletion of crop diversity, soil sickness and reduction of crop productivity (Khanh *et al*. 2005). Allelopathic crop water leachates such as sorghum (*Sorghum bicolor*), brassica (*Brassica napus*) and sunflower (*Helianthus annuus* L.) are being investigated as an alternative to herbicides for weed control and have shown an ability to inhibit the density and dry weight of weeds (Cheema and Ahmad, 1992; Khaliq *et al.* 1999). Water leachates from sorghum residues inhibited germination and decreased root and shoot growth of corn (*Zea mays* L.) and wheat (Guenzi *et al*., 1967). The growing crops suppress growth of certain weed species, while residues of some crops also inhibit the seed germination of weeds by the release of phytotoxins (Narwal, 1994). The allelopathic potential of different crops such as sorghum (Cheema *et al*. 2000), sunflower (Cheema *et al.* 2005), rice (Xuan *et al*. 2003), brassica (Turk *et al*. 2003) and wheat (Narwal *et al.* 1998) has been described. It is suggested that herbicides and allelopathic products may work complementarily and the herbicidal dose may be reduced when applied in combination with allelopathic products (Cheema *et al.,* 2002a). Seed rates affect weed dynamics in crops. Schenke and Köpke (1994) and Pallutt (2000) demonstrated that the weed cover decreased when the winter cereals' seeding rate got higher.

The presented studies were conducted to compare the relative weed dynamics in fennel grown at varying seeding rates and look for the possibility of utilizing the allelopathic potential of sorghum in combination with reduced rates of herbicides for managing weeds.

Materials and Methods

The investigations were carried out at the Agronomic Research Area, University of Agriculture, Faisalabad, Pakistan, during the year 2004-05. The experiment was laid out in a split plot design and replicated thrice. The net plot size was 2.5 m x 5.0 m. Fennel was sown with four seed rates at 4, 6, 8 and 10 kg ha^{-1}. Weed management strategies were pendimethalin at 412.5 g a.i. ha^{-1} (half of label dose) tank mixed with sorghum water leachate (sorgaab) at 15 l ha^{-1}, label dose of pendimethalin (825 g a.i. ha^{-1}) and two hand weedings. A weedy check was maintained. Seed rates and weed control practices were randomized in main and sub plots, respectively. Sorghum water leachate was prepared according to the procedure described by Cheema *et al.* (2002b). Seedbed was prepared by ploughing and sowing was done in 45 cm spaced rows with the help of a single row hand drill. Nitrogen (50 kg ha^{-1}) was applied in two doses, *i.e.* half at sowing and the remainder with the first irrigation. Phosphorus (100 kg ha^{-1}) was applied at the time of sowing. Two hand weedings (first after one month of sowing and the second after the third irrigation) were done. The crop received four irrigations; first at the completion of germination, second after fifteen days from the first irrigation, third at flowering and the last irrigation was applied at seed formation. Crop was harvested manually on 2nd May 2005. Data on different traits of fennel were recorded following standard procedures. Individual and total weed density was recorded on 40 and 70 days after sowing from two quadrats of 50 cm x 50 cm. Weeds were clipped from ground surface and fresh weight recorded. These were sun-dried and then placed in an oven at 70°C for 48 h and dry weight recorded on a digital balance. Data recorded were analyzed statistically using Fisher's analysis of variance technique (Steel and Torrie, 1984). Least significant difference test ($p \leq 0.05$) was used to separate the treatments means.

Results and Discussion

Weed flora of the experimental site comprised mainly of canary grass (*Phalaris minor* L.) and lambs quarters (*Chenopodium album* L.), while swine cress (*Cornopus didymus* L. Smith) was found in lower densities. Increasing seed rates of fennel had a suppressive effect on total weed dry weight (Table 1). At 40 days after sowing (DAS), 45–49% less total weed dry weight was recorded when seed rate of fennel was increased up to 10 kg ha^{-1}. Increasing seed rate beyond 6 kg ha^{-1} did not decrease total weed dry weight significantly ($p<0.05$). At 70 DAS, seed rates of 6, 8 and 10 kg ha^{-1} recorded a reduction of 23, 3 and 80% as compared with that recorded for 4 kg ha^{-1} seed rate.

Different weed control methods significantly reduced the total weed dry weight (Table 1). At 40 DAS, pendimethalin (825 g a.i. ha^{-1}) gave only 4% reduction in total weed dry weight while combined application of sorghum water leachate at 15 l ha^{-1} with pendimethalin (412.5 g a.i. ha^{-1}) reduced total weed dry weight by 76% at this stage and by 80% at 70 DAS.

Table 1. Influence of different seeding rates and weed management practices on total weed dry weight

Treatments	Total Weed Dry Weight (g per 0.25 m^2) 40 DAS	70 DAS
Seed Rates (S)		
S_1 = 4 kg ha^{-1}	10.46 a	16.66 a
S_2 = 6 kg ha^{-1}	5.80 b	12.87 ab
S_3 = 8 kg ha^{-1}	5.90 b	16.09 ab
S_4 = 10 kg ha^{-1}	5.38 b	3.27 c
LSD (p=0.05)	2.89	5.03
Weed Control Practices (WP)		
WP_0 = Weedy check (control)	4.14 a	16.66 a
WP_1 = Pendimethalin at 825 g a.i.ha^{-1}	3.98 a	12.87 ab
WP_2 = Sorgaab at 15 l ha^{-1} + pendimethalin at 412.5 g a.i. ha^{-1}	0.00 a	0.00 c
WP_3 = Hand weeding (twice)	0.99 ab	3.27 bc
LSD (p= 0.05)	3.88	12.86
Interaction (S x WP)**		

Within a column, means followed by the same letter are not significantly different at p=0.05; DAS - days after sowing.

Hand weeding at 40 and 70 DAS gave a complete (100%) suppression in total weed dry weight as compared with the check. Interactive effect of different seeding rates of fennel and weed control methods on total weed dry weight was highly significant (Figure 1). At both the sampling dates, the increasing seed rates suppressed the total dry weight significantly. At all seeding rates, combined application of sorghum water leachate tank mixed with 50% of label dose of pendimethalin gave a good weed suppression as was recorded with label dose of pendimethalin when applied alone. Hand weeding also gave complete suppression of weeds at all seeding rates.

Seeding rates of fennel did not influence the umbels per plant, seeds per umbel, 1000-seed weight and seed yield of fennel (Table 2). However, different weed control methods influenced these plant traits significantly. Combined application of sorghum water leachate and 50% of label dose of pendimethalin gave as much number of umbels per plant, seeds per umbel, 1000-seed weight and seed yield of fennel as was recorded for label dose of pendimethalin when used alone. Hand weeding gave significantly higher values for all these characteristics than any other practice. Yield increases of 125, 166 and 350% were recorded for pendimethalin alone (825 g a.i. ha^{-1}), sorghum water leachate+pendimethalin (412.5 g a.i. ha^{-1}) and two hand weedings, respectively over the weedy check.

Possibility of suppressing weeds in fennel through manipulating seeding rate and employing allelopathic sorghum water leachate in combination with reduced dose of pendimethalin was explored in present studies. Total weed dry weight was reduced when seed rate was increased. Seeding rate and spacing both affect weed dynamics in crops. Märländer and Bräutigam (1994) showed weed density to be reduced as the number of sugarbeet plants per hectare increased, and Merkel *et al.* (2000) concluded the same from her trials in rape. Schenke and Köpke (1994) and Pallutt (2000) demonstrated that the weed cover decreased when the winter cereals' seeding rate got higher. Sorghum water leachate (15 l ha^{-1}) tank mixed with half of the label dose of pendimethalin gave as good weed suppression as was realized with label dose of pendimethalin.

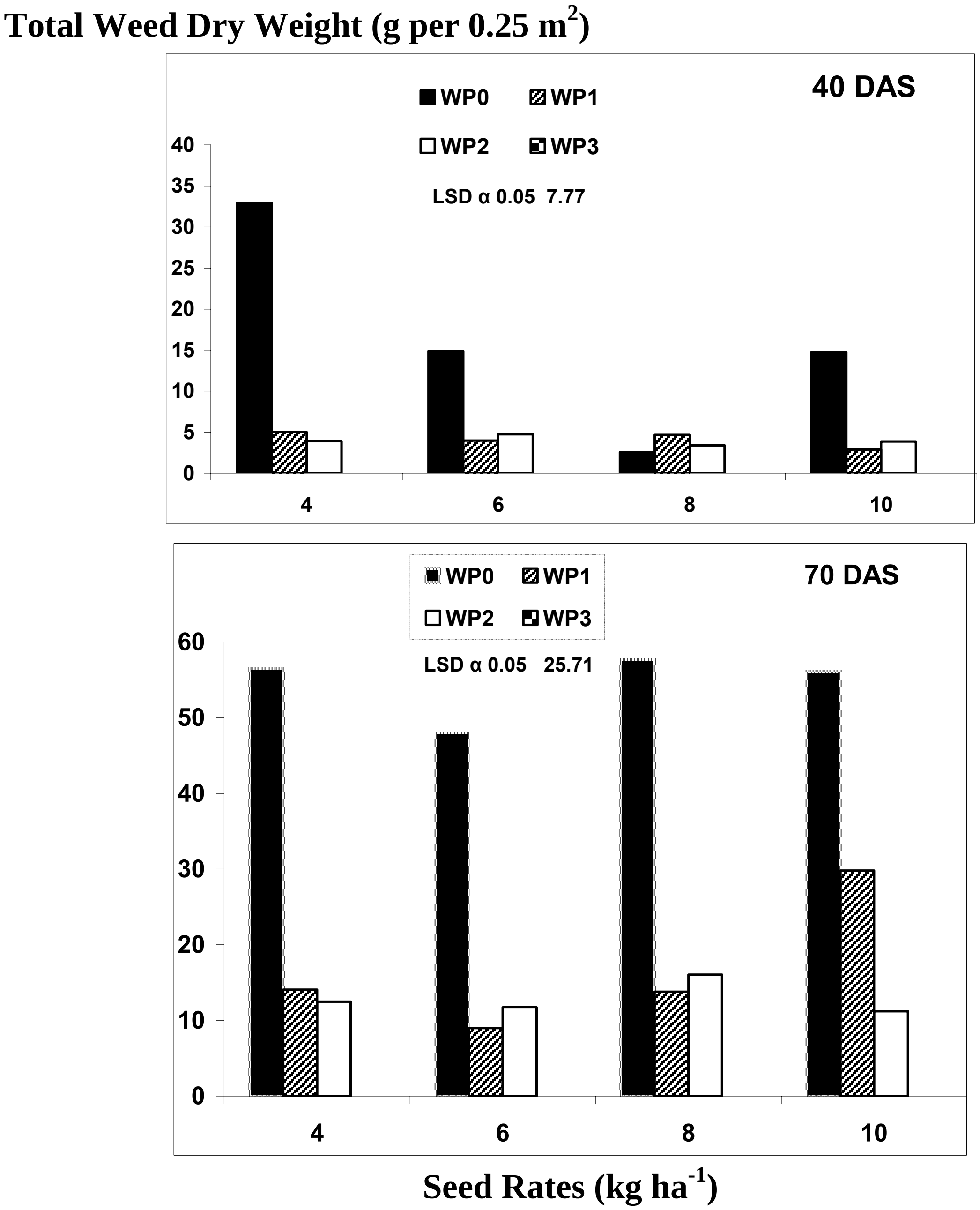

Figure 1. Interactive effect of seeding rates and weed control methods on total weed dry weight in fennel.

Putnam and DeFrank (1983) found that residues of sorghum reduce the number and biomass of common purslane and smooth crabgrass (*Digitaria ischaemum* [Schr.] Muhl.) in the fields by 70% and 98%, respectively. All parts of sorghum like roots, herbage and germinating seeds release phytoinhibitors reducing the growth of grass and broadleaf species such as green foxtail, velvetleaf, and smooth pigweed (Panasiuk *et al.* 1986; Hoffman *et al.* 1996). Cheema and Khaliq (2000) observed that spray of 'sorgaab' (water leachate of mature plants obtained after soaking in water for 24 h) reduced the weed density and biomass by 35 to 49 % and increased the wheat yield by 10 to 21 %. They concluded that sorgaab could be used as a natural weed inhibitor in irrigated wheat. The higher seed yield in the treatment containing twice hand weeding may be due to better weed control. Weed inhibition was due to suppressive effect of water leachates and compatibility with the herbicides that possibly enhanced plant food resources availability for plant growth and more translocation of photosynthates towards reproductive parts that ultimately increased grain yield (Salisbury and Ross, 1978).

Table 2. Influence of different seeding rates and weed management practices on seed yield.

Treatments	Umbels per plant	Seeds per umbel	1000-seed weight (g)	Seed Yield (Mg ha^{-1})
Seed Rates (S)				
S_1 = 4 kg ha^{-1}	17.62	110.08	11.83	0.40
S_2 = 6 kg ha^{-1}	18.72	117.57	13.53	0.41
S_3 = 8 kg ha^{-1}	18.44	105.55	12.00	0.43
S_4 = 10 kg ha^{-1}	19.57	113.17	12.68	0.43
LSD (p=0.05)	NS	NS	NS	NS
Weed Control methods (WP)				
WP_0 = Weedy Check (Control)	11.85 c	80.12 b	10.62 b	0.16 c
WP_1 = Pendimethaline at 825 g a.i.ha^{-1}	17.97 b	114.20 a	12.01 b	0.36 b
WP_2 = Sorgaab at 15 L ha^{-1} + pendimethaline at 412.5 g a.i. ha^{-1}	17.45 b	127.52 a	12.70 b	0.43 b
WP_3 = Hand weeding (twice)	18.82 a	125.14 a	13.42 a	0.72 a
LSD (p=0.05)	1.22	19.38	0.70	0.07
Interaction (S x WP)	NS	NS	NS	NS

NS – non-significant; Within a column, means followed by the same letter are not significantly different at p=0.05.

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EFFECT OF DIFFERENT ALLELOPATHIC EXTRACTS ON WEEDS AND WHEAT

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Abstract: Field studies were initiated at NWFP Agricultural University, Peshawar, Pakistan during the Rabi season of 2003-04 to study the effect of different allelopathic extracts on weeds and a wheat crop. The treatments comprised of extracts from *Ammi visnaga* and *Convolvulus arvensis*, extracted either in ethanol or $CHCl_3$, each applied either in full or half strengths. An untreated check was also included in the study. The extracts were applied after the emergence of the wheat crop as well as weeds. The data recorded were weed density per m, plant height (cm), spike length (cm), number of grains per spike, 1000 grain weight (g), grain yield per spike (g), biological yield (t ha^{-1}) and grain yield (t ha^{-1}). The major weeds were *Avena fatua, Coronopus didymus, Euphorbia helioscopia, Fumaria indica, C. arvensis, Rumex dentatus, Chenopodium album, Poa annua, Medicago denticulata,* and *Vicia sativa*. The $CHCl_3$ extract of *C. arvensis* at full strength and *A. vsnaga* half strength proved to be the best in inhibiting weeds density. The maximum grain yield of 1.153 t ha^{-1} was recorded in treatment where *C. arvensis* was extracted with $CHCl_3$ and applied at the half strength, and 1.120 t ha^{-1} in *A.visnaga* extracted in the same solvent applied at full strength. The minimum grain yield (0.79 t ha^{-1}) was recorded in the untreated plots.

Key words: Weeds, allelopathy, plant extracts, wheat

Introduction

Allelopathy refers to the beneficial or harmful effects of one plant on another plant, both crop and weed species, by the release of chemicals from plant parts by leaching, root exudation, volatilization, residue decomposition and other processes in both natural and agricultural systems. Allelochemicals produced by a plant escape into the environment and subsequently influence the seed germination, growth and development of other neighboring plants. Exploitation of allelopathy is a natural and environment-friendly technique which has the potential to be a unique tool for weed management and consequently increase crop yields. The possibility of using allelochemicals as growth regulators and natural pesticides promotes sustainable agriculture (Kruse and Strandberg, 2000; Lafleur and Mallik, 2001).

Allelopathic inhibition is complex and involves the interaction of different classes of chemicals such as phenolic compounds, flavonoids, terpenoids, alkaloids, steroids, carbohydrates, and amino acids, with mixtures of different compounds sometimes having a greater allelopathic effect than individual compounds alone. Furthermore, physiological and environmental stresses, pests and diseases, solar radiation, herbicides, and less than optimal nutrient, moisture, and temperature levels can also affect allelopathic weed suppression. Different plant parts, including flowers, leaves, leaf litter and leaf mulch, stems, bark, roots, soil and soil leachates and their derived compounds, can have allelopathic activity that varies over a growing season. Allelopathic chemicals can also persist in soil, affecting both neighboring plants as well as those planted in succession. Although derived from plants, allelochemicals may be more biodegradable than traditional herbicides but may also have undesirable effects on non-target species, necessitating ecological studies before widespread use (Lovett and Ryuntyu, 1992).

Research is continuing in all these areas and many new ideas have been developed in an attempt to understand the phenomenon of allelopathy deeply and to exploit it to enhance the production of manipulated ecosystems. Several reports address the allelopathic potential of trees species. Khan (2003) cited the advent of *Prosopis juliflora* in the local habitat. And state that it came into the region (Pakistan) in the fodder for the horses of British soldiers while the government from Australia imported Eucalyptus for afforestation purposes. Al-Humaid and Warrag (1998) reported that germination percentage of *Cynodon dactylon* seeds decreased with increasing leaf extract concentration of *Prosopis juliflora*. Chellamuthu *et al.* (1997) reported that germination of black gram (*Vigna mungo*) and sorghum (*Sorghum bicolor*) was inhibited as the rate of water extracts increased. Putnam (1984) reported that *Eucalyptus* species released volatile compounds such as benzoic, cinnamic and phenolic acids, which inhibit growth of crops and weeds growing near it. Pawar and Chawan (1999) reported that some forest trees including *Eucalyptus globolus* reduced uptake of Ca, Zn and Mg in sorghum resulting in reduced growth. Schumann *et al.* (1995) reported that *E. grandis* water extracts significantly reduced weed establishment. Cheema *et al.* (2003) have advocated the commercial utilization of sorghum water extracts for weed management in wheat. The studies on tree extracts in Pakistan. (Khan *et al*. 2004a; Khan *et al*. 2004b) exhibited the differential inhibitory effect of tree extracts on wheat and its weeds. Therefore, allelochemicals are being studied for the development of natural herbicides for sustainability of agro-ecosystems. Therefore, studies were undertaken with the objectives of identifying the response of weeds and wheat to allelochemicals extracted from different plants and to decipher the effect of different concentrations of extracts on growth of wheat and weeds

Materials and Methods

A field experiment was conducted during November 2003-04 at Malakandher Research Farm, NWFP Agricultural University Peshawar, Pakistan to investigate the effect of different allelopathic extracts on weeds and wheat crop. The experiment was laid out in a randomized complete block design, with 4 replications and the plot size was 5 x 1.50 m^2. The experiment comprised of 9 treatments including a weedy check. The allelopathic treatments were the post emergent application of *Ammi visnaga* (ethanol extracted) full strength (pure extract in ethanol), *A. visnaga* (ethanol extracted) half strength (50% pure extract + 50% water), *A. visnaga* ($CHCl_3$ extracted) full strength, *A. visnaga* ($CHCl_3$ extracted) half strength, *Convolvulus arvensis* (pure extract in ethanol) full strength, *C. arvensis* (50% pure extract + 50% water) half strength, *C. arvensis* ($CHCl_3$ extracted) full strength and *C. arvensis* ($CHCl_3$ extracted) half strength. Seeds of the wheat variety Ghaznavi-98 were sown at the rate of 120 kg ha^{-1} in the third week of November 2004. The weed density (m^{-2}), plant height (cm), spike length (cm), No. of grains $spike^{-1}$, 1000 grain weight (g), grain yield $spike^{-1}$(g), biological yield (t ha^{-1}) and grain yield (t ha^{-1}) were recorded, subjected to an ANOVA and the means were separate by the unprotected LSD test (Steel and Torrie, 1980).

Results and Discussion

Weed species infesting the experiment were wild oats (*Avena fatua*), swinecress (*Coronpus didymus*), leafy spurge (*Euphorbia helioscopia*), fumitory (*Fumria indica*), field bindweed (*C. arvensis*), curly dock (*Rumex dentatus*), common lambsquarters (*Chenopodium album*), annual blue grass (*Poa annua*), Indian sweetclover (*Medicago denticulata*) and common vetch (*Vicia sativa*). The extracts had a significant effect on weed density m^{-2}. Effective control was recorded in *Ammi visnaga* ($CHCl_3$) half strength and *C. arvensis* ($CHCl_3$) full strength application as against 101 plants m^{-2} in the weedy check (Table1). The best treatments were *C. arvensis* (ethanol) half strength and *A. visnaga* (ethanol) half strength having 37.333 and 38.667 weeds m^{-2}, respectively. The findings of Khan *et al*. (2004a; 2004b) and Khanh *et al*. (2005) have a great analogy with our inferences, suggesting that allelochemicals strongly suppress emergence of weeds.

Table 1. Effect of some plant extracts on weed density (number per m^2), plant height (cm), spike length (cm) and number of grains per spike[1].

Treatments	Weed density m^{-2}	Plant height (cm)	Spike length (cm)	No. grains per spike
Ammi visnaga (ethanol) Full Strength	53.333 c	76.800 b	8.833	60.677 a
Ammi visnaga (ethanol) Half strength	38.667 cd	78.867 ab	8.133	45.000 b
Ammi visnaga ($CHCl_3$) Full strength	39.667 cd	77.867 ab	8.633	44.667 b
Ammi visnaga ($CHCl_3$) Half strength	33.667 d	81.400 ab	9.000	45.333 b
Convolvulus arvensis (ethanol) Full strength	47.000 cd	85.000 a	8.867	44.667 b
Convolvulus arvensis (ethanol) Half strength	37.333 cd	78.400 ab	8.900	42.667 b
Convolvulus arvensis ($CHCl_3$) Full strength	33.667 d	79.333 ab	8.333	54.333 ab
Convolvulus arvensis ($CHCl_3$) Half strength	80.667 b	81.000 ab	8.100	44.667 b
Weedy Check	101.000 a	75.667 b	8.433	41.000 b
LSD (p=0.05)	19.64	7.153	NS	13.48

NS = Non-significant; Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Plant height (cm) was significantly affected by extracts of this study. The maximum plant highest height was recorded in plots treated with *C. arvensis* (ethanol) full strength followed by plots treated with *A. visnaga* ($CHCl_3$) half strength. The lowest plant height was recorded in weedy check. The spike length of wheat was not affected by the extracts. The longest spike (9 cm) was observed in *A. visnaga* ($CHCl_3$) half strength while the other treatments induced similar spike lengths. The extracts had a significant effect on number of grains per spike. The maximum numbers of grains per spike (61) were observed when *A. visnaga* (ethanol) full strength was applied, which was similar to 54 grains per spike with *C. arvensis* ($CHCl_3$ extracted), full strength. The numbers of grain in the latter treatment was similar to values of all other treatments including untreated check (Table 1). The grain weight per spike was not affected by extracts, and the highest (2.340 g) grain weight per spike was observed with the application of *A. visnaga* (ethanol) full strength followed by 2.023 g in *C. arvensis* ($CHCl_3$) full strength application. The untreated check had 1.537 g grain weight per spike (Table 2).

Table 2. Effect of some plant extracts on Grain weight per spike, 1000-grain weight, biological yield and grain yield of wheat.

Treatments	Grain weight per spike (g)	1000-grain weight (g)	Biological yield (t ha^{-1})	Grain yield (t ha^{-1})
Ammi visnaga (ethanol) Full Strength	2.340	32.750 cd	1.767	0.953 b
Ammi visnaga (ethanol) Half strength	1.870	38.467 a	1.327	0.830 bcd
Ammi visnaga ($CHCl_3$) Full strength	1.790	37.487 ab	1.960	1.120 a
Ammi visnaga ($CHCl_3$) Half strength	1.887	33.733 cd	1.600	0.944 bc
Convolvulus arvensis (ethanol) Full strength	1.860	35.703 abc	1.767	0.873 bcd
Convolvulus arvensis (ethanol) Half strength	1.810	35.197 abc	1.517	0.737 d
Convolvulus arvensis ($CHCl_3$) Full strength	2.023	33.430 cd	1.407	0.803 bcd
Convolvulus arvensis ($CHCl_3$) Half strength	1.447	34.660 bc	1.933	1.153 a
Weedy Check	1.537	30.603 d	1.960	0.790 cd
LSD (p=0.05)	N.S	3.682	NS	0.157

NS = Non-significant; Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

There was a significant difference in 1000 grain weight due the application of extracts. The highest 1000-grain weight (38.4 g) was recorded with *A. visnaga* (ethanol) half strength followed by 37.4 g observed in plots treated with *A. visnaga* ($CHCl_3$) full strength. These were similar to values obtained with *Convolvulus arvensis* (ethanol) half strength and *C. arvensis* (ethanol) full and half strength. The remaining treatments had values lower than that of the weedy check (30.6 g). The biological yield of wheat was marginally affected by the applied treatments. However, there was a random variation in the data. The highest biological yield was recorded with applications of *A. visnaga* ($CHCl_3$) full strength and the untreated check (Table 2). Grain yield (t ha^{-1}) was significantly affected by different extracts. The highest (1.153 and 1.120 t ha^{-1}) grain yield was recorded in plots treated with *C. arvensis* ($CHCl_3$) half strength and *A. visnaga* ($CHCl_3$) full strength, respectively. The treatments *A. visnaga* (ethanol) full strength, *A. visnaga* (ethanol) half Strength, *C. arvensis* (ethanol) full strength and half strength failed to out yield the weedy check (0.79 t ha^{-1}). These findings are in agreement with the earlier work of Cheema *et al*. (2003), who worked on *Sorghum bicolor* extracts.

Conclusions

The results of the study revealed that differences were observed in weed populations due to the extracts, the extracts had phytotoxic effects on weeds, but not on the wheat crop, the efficacy of the tested extracts illustrates possibilities for their selective use in wheat, and the natural products are environmental friendly, hence, need to be exploited for sustainability in the agro-ecosystems. Further studies are suggested to confirm these findings.

Acknowledgements

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BIOLOGICAL PERFORMANCE OF BISPYRIBAC-SODIUM + THIOBENCARB 915 OD IN RICE

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Abstract: Bispyribac-sodium has been widely used for direct seeded rice in the world under the trade name of "Nominee®". To expand the herbicidal spectrum and add residual efficacy, new premix product of bispyribac-sodium + thiobencarb 915 OD (15 + 900 g a.i./l oil dispersion) is being developed by Kumiai Chemical Industry Co., Ltd. Greenhouse studies were conducted at Kumiai Chemical Life Science Research Institute to evaluate weed control, phytotoxicity and other factors which may affect herbicidal efficacy. This premix product at 1.0 l/ha provided excellent weed control on grasses (*Echinochloa crus-galli*, *Ischaemum rugosum* and *Leptochloa chinensis*), sedges (*Cyperus difformis, Fimbristylis miliacea* and *Scirpus juncoides*) and broadleaved weeds (*Sagittaria trifolia, Ammannia coccinea, Aneilema keisak, Aeschynomene indica* and *Monochoria vaginalis*) at the early post-emergence stage. In addition, this premix product was not negatively affected by spray volume, rainfall and water management after application. Therefore, bispyribac-sodium + thiobencarb 915 OD is expected to be a new solution for rice farmers with a wide crop safety margin and consistent weed control.

Key words: bispyribac-sodium + thiobencarb 915 OD, bispyribac-sodium, thiobencarb

Introduction

Bispyribac-sodium (Experimental Code No. KIH-2023) is a selective post-emergence herbicide developed and being distributed by Kumiai Chemical Industry Co., Ltd. (Yokoyama *et al*. 1993; Kobayashi *et al*. 1995). Due to some excellent characteristics of bispyribac-sodium such as wide application window, broad herbicidal spectrum and high selectivity, the product has been widely used in direct seeded rice in Southeast Asian countries, Central and South American countries under the trade name of "Nominee®".

Bispyribac-sodium has an excellent foliar application efficacy against grasses, sedges and broadleaved weeds from early post-emergence through early tillering stage of *Echinochloa* spp. However, bispyribac-sodium does not provide sufficient residual efficacy at recommended dosage. In addition, herbicidal efficacy of bispyribac-sodium against *Leptochloa chinensis* which is becoming a more and more serious problem in direct seeded rice cultivation in recent years in Asian countries is not sufficient at the recommended dosage. To expand the herbicidal spectrum against grasses like *Leptochloa Chinensis* and add residual efficacy, bispyribac-sodium + thiobencarb premix product (bispyribac-sodium + thiobencarb 915 OD) has been developed by Kumiai Chemical Industry Co., Ltd.

This paper describes the results of greenhouse trials carried out at Kumiai Chemical Life Science Research Institute in Japan to evaluate weed control, phytotoxicity and other factors which may affect herbicidal efficacy of bispyribac-sodium + thiobencarb 915 OD.

Materials and Methods

Experiment 1: Herbicidal efficacy at different weed growth stages

The test chemicals used were (a) bispyribac-sodium + thiobencarb 915 OD (bispyribac-sodium + thiobencarb = 15 + 900 g a.i./l OD) at 0.5 and 1.0 l/ha and (b) Nominee® (bispyribac-sodium) 10%SC with non-ionic surfactant, Agrisol A-150K (same volume as Nominee 10% SC) at 150 ml/ha. The test chemicals were applied at different weed growth stages at 320 l/ha spray volume utilizing a hand sprayer. The target weeds were grasses namely, *Echinochloa crus-galli, Ischaemum rugosum, Leptochloa chinensis* and *Brachiaria platyphylla,* sedges namely, *Cyperus difformis, Fimbristylis miliacea* and *Scirpus juncoides,* and broadleaf weeds namely, *Sagittaria trifolia, Ammannia coccinea, Aeschynomene indica, Murdannia keisak* and *Monochoria vaginalis*.

Weed seeds were kept under wet condition until their emergence without flooding water and emerged aquatic weeds were grown under 3cm flooding condition. Just before application, water was drained out of the plastic pots. The aquatic weeds were flooded again at three days after application. The soil type was Clay-loam and the experiment was conducted in 1/1000000 ha plastic pot.

Visual observation was conducted at 7, 14, 21 and 28 days after application separately. For visual observation, 0% to 100% rating system was adopted (*i.e.* 0% (or 0) = no herbicidal efficacy to 100% (or 10) = complete control.

Experiment 2: Influence of various factors on herbicidal efficacy

Test chemical (bispyribac-sodium + thiobencarb 915 OD) was applied at the 4-leaf stage of *Echinochloa crus-galli*. The experimental conditions of test chemical, soil type and trial size and evaluation method were the same as the Trial I.

<u>Influence of spray volume on herbicidal efficacy</u>: The test chemical was applied on *Echinochloa crus-galli* at 50, 100, 200 and 400 l/ha spray volume under drained conditions. The pots were flooded at 2 days after application and water depth was kept at 3 cm during whole test period.

<u>Influence of rainfall after application on herbicidal efficacy</u>: The test chemical was applied on *Echinochloa crus-galli* at 320 l/ha spray volume under drained conditions. The sprayed plants were exposed to 60 mm rainfall at 1, 3, 6 and 24 hrs after application. The water in the pot after rainfall was drained to be 3 cm of water depth and this depth was maintained during the whole test period.

<u>Influence of the timing of flooding on herbicidal efficacy</u>: The test chemical was applied on *Echinochloa crus-galli* at 320 l/ha spray volume under drained conditions. Test pots were flooded at 6 hours, 1 day, 3 days and 5 days after application and water depth was kept at 3 cm during the whole test period.

Experiment 3: Safety to Indica Type Rice

This trial was conducted in wet and dry seeded rice. One (1) and 3 l/ha of test chemical (bispyribac-sodium + thiobencarb 915 OD) were applied at 1 and 3 leaf stage of rice (rice variety: RD-23/indica type variety) at 320 l/ha spray volume.

In wet-seeded rice condition, the test chemical was applied under drained conditions. The test pots were flooded at 3 days after application and water depth was kept at 3 cm during the whole test period. In dry-seeded rice, water was supplied from the bottom of the pots when the soil surface was dry. The pots were flooded at 14 days after application and water depth was kept at 3 cm during the whole test period.

Results and Discussion

Weed spectrum and efficacy of bispyribac-sodium + thiobencarb 915 OD were studied on key weeds (Table1). The efficacy of bispyribac-sodium + thiobencarb 1.0 l/ha was higher than that of Nominee® (bispyribac-sodium) 10% SC 150 ml/ha on *Leptochloa chinensis, Brachiaria platyphylla* and *Scirpus juncoides* at pre-emergence or the early post-emergence.

In addition, the result of pre-emergence application on broadleaved weeds indicates that bispyribac-sodium + thiobencarb has more residual activity on *Ammannia coccinea, Aeschynomene indica* and *Aneilema keisak* when compared with bispyribac-sodium alone (Table 2).

Table1. Weed spectrum of bispyribac-sodium + thiobencarb 915 OD at 1.0 L/ha

		Weed Stage					
	Weeds	Pre	2L	3L	4L	5L	6L
Grasses	ECHCG	-	10	-	10	-	8
	ISCRU	-	10	-	10	-	10
	LEFCH	10 (Pre~2L)		8	1	-	5
	BRAPP	-	8	-	1	-	0
Sedges	CYPDI	10	10	-	7	-	2
	FIMMI	10 (Pre~2L)		-	10	-	10
	CYPES	-	6 (2L~3L)		5 (4L~5L)		-
	SCPJO	3	7	-	9	-	5
Broadleaf weeds	SAGTR	10	-	10		-	-
	AMMCO	10	10 (2L~3L)		10 (4L~6L)		
	AESIN	8	10	-	10	-	10
	ANEKE	10	-	10	-	10	-
	MOOVP	6	-	10	9	-	5

0 (no efficacy) ~ 10 (complete control)

Chemical bispyribac-sodium + thiobencarb 915OD at 1.0 L/ha

Spray volume 320 L/ha

Evaluation 28DAA

Table 2. Advantage of bispyribac-sodium + thiobencarb 915 OD at 1.0 L/ha compared to Nominee® 10%SC (bispyribac-sodium alone at 150 ml/ha)

Weeds	**Chemical**	**Pre**	**2 l**	**3 l**	**4 l**	**5 l**	**6 l**
LEFCH	**bispyribac-Na + thiobencarb**	**10**		**8**	**1**	**-**	**5**
	bispyribac-Na	**5**		**5**	**1**	**-**	**5**

AMMCO	**bispyribac-Na + thiobencarb**	**10**	**10**		**10**		
AESIN		**8**	**10**	**-**	**10**	**-**	**10**
ANEKE		**10**	**-**	**10**	**-**	**10**	**-**
AMMCO	**bispyribac-Na**	**3**	**10**		**10**		
AESIN		**2**	**10**	**-**	**10**	**-**	**10**
ANEKE		**5**	**-**	**10**	**-**	**10**	**-**

> Nominee (100SC) + Agrisol A-150K 15gai/ha

Influence of spray volume on herbicidal efficacy was studied (Figure 1). 0.5 l/ha of this product was slightly affected at 50 l/ha of spray volume. however 1.0 l/ha of this product is not negatively affected within the range of 50-400 l/ha spray volume. This result suggests that bispyribac-sodium + thiobencarb can maintain stable herbicidal efficacy at various spray volumes used in many countries of the world.

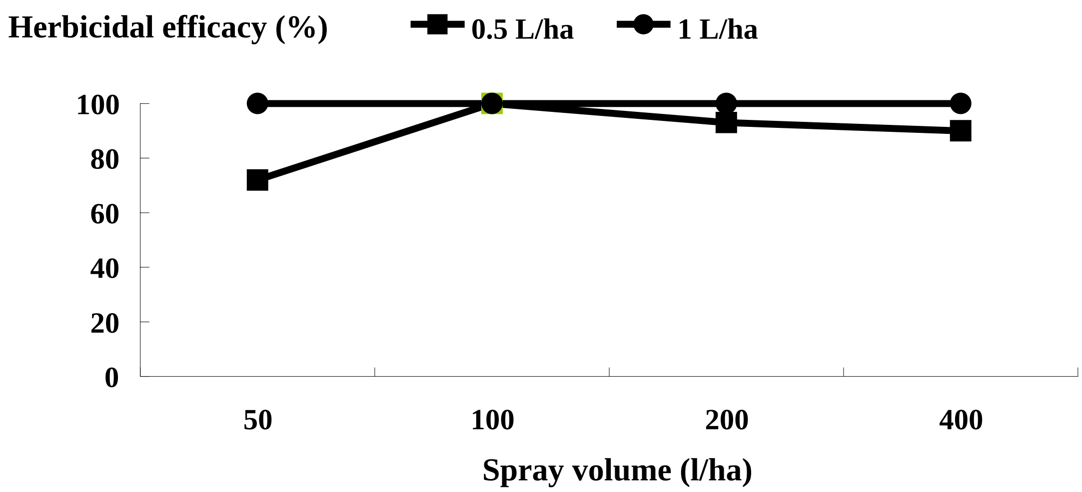

Figure1. Influence of spray volume on herbicidal efficacy of bispyribac-sodium + thiobencarb against the 4 leaf stage of *Echinochloa crus-galli*

Influence of rainfall after application on herbicidal efficacy was studied (Figure 2). When it had rainfall within 3 hrs after application of 0.5 l/ha bispyribac-sodium + thiobencarb 915 OD, herbicidal efficacy against the 4 leaf stage of *Echinochloa crus-galli* decreases significantly. However, in 6 hrs after application, even 0.5 l/ha of this product was not affected by rainfall. This result indicates that if there is rainfall within 3 hrs after application, the efficacy of this product can decrease. Therefore, rainfall within 6 hrs after application should be avoided for stable weed control.

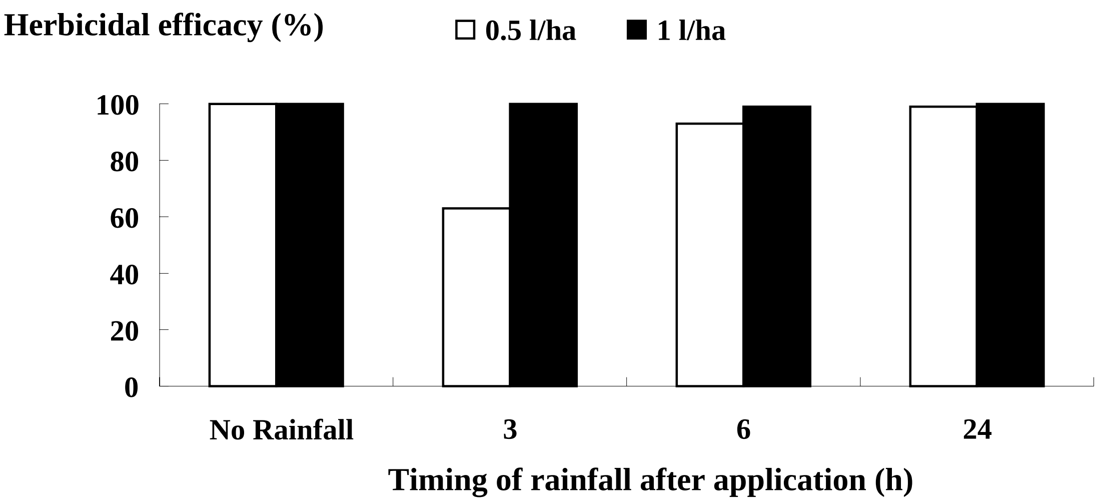

Figure 2. Influence of rainfall after application on herbicidal efficacy of bispyribac-sodium +thiobencarb against the 4 leaf stage of *Echinochloa crus-galli*

Influence of the timing of flooding after application was studied (Figure 3). The efficacy against the 4 leaf stage of *Echinochloa crus-galli* tends to decrease at 0.5 l/ha when the interval is more than 3 days between application and flood timing. Though the efficacy under no flood condition is slightly higher than 5 days interval, it suggests that flooding timing interval will affect the efficacy of this product as well as Nominee® 10% SC (bispyribac-sodium alone at 150 ml/ha). Accordingly, it is recommended that permanent flooding should be established within 3 days after application.

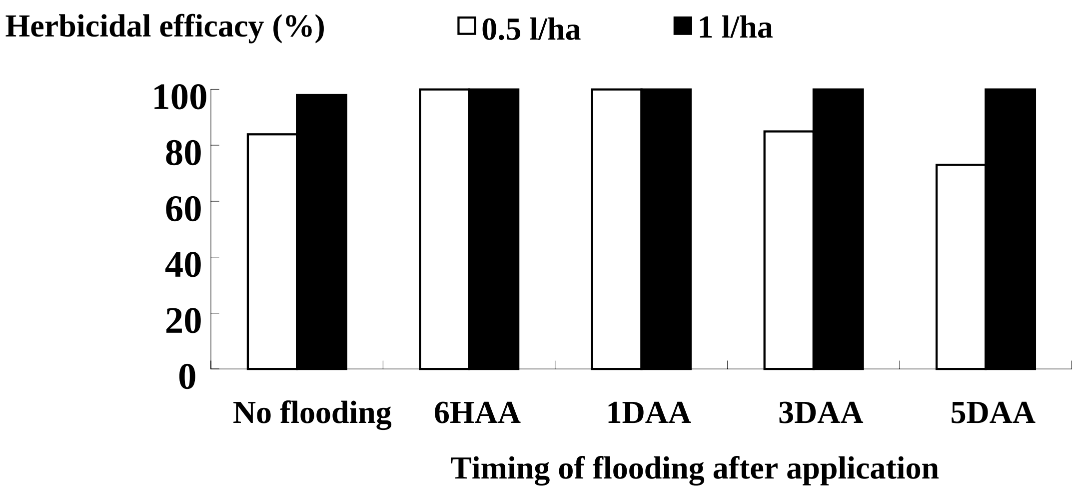

Figure 3. Influence of the timing of flooding on herbicidal efficacy of bispyribac-sodium + thiobencarb against the 4 leaf stage of *Echinochloa crus-galli*

Bispyribac-sodium + thiobencarb 915OD has high selectivity between rice and many kinds of weeds. 1.0 l/ha and 3.0 l/ha of this product were applied at 1 and 3 leaf stages of RD-23 / indica type variety. Indica type rice showed a good tolerance for 1.0 l/ha and 3.0 l/ha of this product both in wet and dry seeded rice (Figure 4).

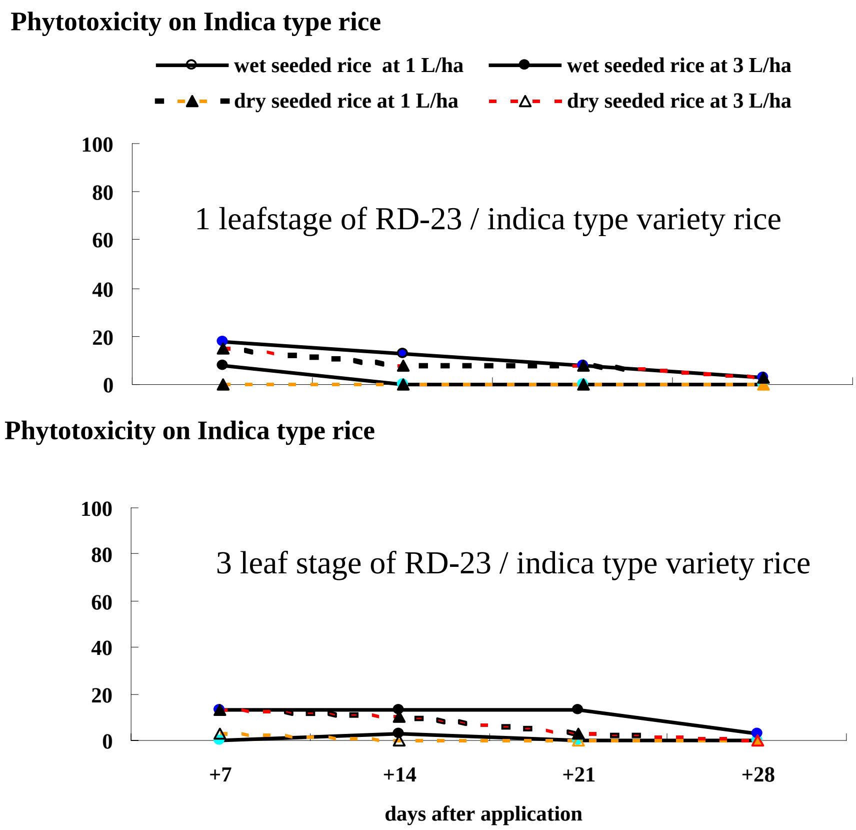

Figure 4. Phytotoxicity of bispyribac-sodium + thiobencarb on 1 and 3 leaf stages of indica type rice

These results show that a new premix product of bispyribac-sodium + thiobencarb 915 OD (15 + 900 g a.i./l oil dispersion) has high efficacy against key weeds such as *Leptochloa Chinensis* in direct seeded rice at early post-emergence stage and provide residual activity with excellent selectivity on indica type rice. Therefore, bispyribac-sodium + thiobencarb 915 OD is expected to be a new solution for rice farmers.

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INTEGRATED WEED MANAGEMENT IN SHALLOT (*Allium cepa* var. Ascalonicum)

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Abstract: A study on integrated weed management in shallot was conducted on a farmer field. The treatments consisted of three weeds management practices - hand weeding at 25 days after planting, pre-emergence herbicides applied immediately after planting, and no weeding (weedy), with sub treatments comprising of five mulching materials and bare soil. All plots were hand-weeded at 50 days after planting (DAP). The five mulching materials were rice straw, lalang leaf (*Imperata cylindrica* P. Beauv.), vetiveria leaf [*Veteveria zizanioides* (Linn.) Nash.], paragrass (*Brachiaria mutica* L) and water hyacinth (*Eichhornia crassipes* Solms.) had no significant effect (p>0.05) on weed control and yield of shallot. Integrating mulching materials with hand weeding at 25 days after planting gave the highest shallot yield, followed by the integrated pre-emergence herbicide alachlor at 1.65 kg a.i/ha with mulching materials and mulching materials with no weed control respectively. Absence of a mulch with no weed control produced the lowest shallot yield.

Key words: Shallot, integrated weed management, mulching material, herbicide, hand weeding

Introduction

Shallot (*Allium cepa* var. ascalonicum) is an annual vegetable or medicinal plants, 4-8 bulbs are formed and arise from a single parent bulb, and produced for dehydrated shallot (Tindall, 1983). Shallot is susceptible after emergence to plant stunting from weed competition. Weed present problems throughout the growing season. Hand weeding has been the traditional means of controlling weeds Zimdahl (1980). Mulching also cover the soil surface and affects the photosynthesis process of the weeds, which could reduce weed populations and promoted crop growth. Mulching with plant residue can increase soil organic matter and improve soil aeration (Brenner, 1983). However, using pre-emergence herbicides is an easy way to decrease weeds competition between shallot and weeds during early growth stage of shallot. Alachlor, metolachlor, oxyfluorfen, oxadiazon have been used as pre-emergence herbicides in shallot (Kongsaengdao *et al*.1990), but no one herbicide will control all weeds. No one method of weed control works best under all conditions. Hence, bulb onion weed management requires the use of cultivation in addition to herbicides (Roberts *et al*. 2002). In a study of shallot yield comparing various mulching materials, Kongsaengdao and Suwanrak (2002) reported that lalang leaf (*Imperata cylindrica* P. Beauv.) and vetiveria leaf [*Veteveria zizanioides* (Linn.) Nash.] had a high weed control efficacy followed by paragrass (*Brachiaria mutica* L), water hyacinth (*Eichhornia crassipes* Solms.) and rice straw (*Oryza sativa* L.) respectively. In this study is integrated weed control by combination weeds management among hand weeding, mulching and chemical control was evaluated.

Materials and Methods

The material used in this field study in Kanchanaburi province of Thailand were shallot bulbs, dry mulching materials namely lalang leaf, vetiveria leaf, paragrass, water hyacinth, rice straw and alachlor 48% EC. Shallot bulbs were planted in field plots of dimensions 1 m x 3 m at a spacing of 20 cm x 20 cm. The treatments were arranged in a 3 x 6 Factorial in a Randomized Complete Block Design (RCBD) with 3 replicates. The main plot consisted of three weeds management practices - hand weeding at 25 days after planting (DAP), pre-emergence herbicides alachor at 1.65 kg a.i/ha applied immediately after planting, no weed control. The sub plot consisted of five mulching materials: rice straw, lalang leaf, vetiveria leaf, paragrass and water hyacinth applied at a rate of 2 kg dry matter per 3 m^2 and bare soil. After 25 days, weeds in the main plot were hand weeded. The weed populations within two quadrates of dimensions 0.5 m x 0.5 m per plot were sampled to record weeds numbers and dry weights. All plots were hand weeded at 50 days after planting and investigated in the manner described above. Shallots were harvested at 77 DAP and the bulb yield was recorded.

Results and Discussion

Efficacy of mulching materials on weed control

Weeds density showed the efficacy of mulching materials on weeds control (Table 1). At 25 DAP, the weed densities (weeds number and weeds dry weight) were similar among mulching materials. Application of lalang leaf, vetiveria leaf and rice straw produced the lowest weed density followed by water hyacinth and paragrass. Plots with bare soil had the highest weed density when compared to the mulching treatments, and had 204 weeds number/m^2 and dry weight 44.9 g/m^2 . The predominant weeds were grasses weeds 63.7% such as *Leptochloa chinensis* (L.) Nees and *Acrachne racemosa* Ohwi. Broadleaf weeds were 36.3% such as *Amaranthus viridis* Linn., *Portulaca oleracea* Linn., *Eclipta alba* (L.) Hassk. Weed control efficacy at 25 days was the highest with vetiveria followed by rice straw, lalang, paragrass and water hyacinth, respectively.

Table 1. Weeds density in treatment mulching with hand weeding at 25 DAP.

Dry mulching materials at 2 kg./3 m^2	Number (No./m^2)	Dry weight (g/m^2)
Paragrass	35.5 a	8.6 ab
Lalang leaf	8.0 a	5.51 a
Vetiveria leaf	13.3 a	1.1 a
Water hyacinth	48.7 a	7.9 ab
Rice straw	38.0 a	2.2 a
Bare soil (No mulching materials)	204.0 b	44.9 b
CV (%)	84.0	171.0

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05).

Effect of integrated weeds management in shallot on weeds numbers

At 50 DAP, application of mulching materials produced significantly lower numbers of weeds than the bare soil. There was no difference among the mulching (13.6-32.9 number/m^2 while bare soil had 57 weeds. Hand weeding and herbicide application induced 27 and 11 weeds per m^2, which was significantly lower than in the un-weeded plot (51 weeds per m^2). When not weeded, lalang mulched plots had the lowest weeds numbers followed by plots with vetiveria, rice straw, water hyacinth and paragrass. Bare soil had the most weeds (Table 2).

Effect of integrated weeds management in shallot on weed dry weights

At 50 DAP, weeds in mulched plots had similar dry weights, and were significantly lower than that of the non weeded plot (Table 3). Weed dry weight of plots mulched with lalang and vetiveria were 89 and 112 g. Weed dry weights of rice straw, water hyacinth and paragrass mulched plots were similar and were between 212-242 g.

Table 2. Effect of integrated weeds management on weed number (no./m^2) at 50 DAP.

Dry mulching materials at 2 kg/3 m^2	Weed control methods			Mean
	Hand weeding	Weedy	Herbicide	
Paragrass	23.3 a	66.0 bc	9.3 a	32.9 a
Lalang leaf	14.0 a	23.3 a	3.3 a	13.6 a
Vetiveria leaf	14.7 a	33.3 ab	9.3 a	19.1 a
Water hyacinth	16.7 a	63.3 bc	8.7 a	29.6 a
Rice straw	13.3 a	38.7 ab	22.0 a	24.7 a
Bare soil	78.0 b	80.7 c	13.3 a	57.3 b
Mean	26.7	50.9	11.0	29.5

CV (%) = 66.3; Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05).

Table 3. Effect of integrated weeds management on weed dry weight (g/m^2) at 50 DAP.

Dry mulching materials at 2 kg/3 m^2	Weed control methods			Mean
	Hand weeding	Weedy	Herbicide	
Paragrass	35.9 a	571.9 b	116.9 a	241.6 ab
Lalang leaf	25.2 a	299.7 ab	11.6 a	112.1 a
Vetiveria leaf	30.4 a	206.1 a	31.7 a	89.4 a
Water hyacinth	78.2 a	560.7 b	82.7 a	240.5 ab
Rice straw	68.8 a	465.6 ab	102.6 a	212.3 ab
Bare soil	223.0 b	519.5 b	213.7 b	318.7 b
Mean	76.9	437.3	93.2	202.5

CV (%) = 84.0; Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05).

Bare soil had the highest weeds dry matter of 319 g. Weeds management practices developed significant differences in weed dry weights. Dry weights of hand weeded and herbicide applied plots were 77 and 93 g while that of the non weeded lot was 437 g. Alachlor at1.65 kg a.i/ha applied immediately after planting controlled grass weeds to a greater extent than broadleaf weeds. At 50 DAP, *Portulaca oleracea* Linn. and *Amaranthus viridis* Linn. were present in herbicide treated plots.

Per plant yields of shallot 77 DAP

The per plant yields of shallot bulbs were not different among mulching materials (Table 4). Weeds management practices such as hand weeding and herbicide produced greater per plant yields when compared to that of un-weeded plants. Hand weeding produced the highest mean shallot bulb yield of 7.58 g per plant. The application of pre-emergent herbicide reduced the shallot bulb yield to 6.94 g. per plant, and the lack of weeding produced the lowest per plant yield of 5.14 g per plant. Water hyacinth mulching with hand weeding at 25 DAP induced the highest shallot bulb yield 9.24 g per plant and bare soil with weeds gave the smallest shallot bulb yield of 1.95 g per plant.

Shallot yields

The mulching materials produced similar shallot yields (Table 5). Yield trend due to vetiveria, water hyacinth and lalang were 2,211, 2,203 and 2,166 kg/ha. Bare soil produced the lowest yield 1,771 kg/ha. Weeds management practices induced highly significant different shallot yields. Hand weeding produced the highest shallot yield follow by pre-emergence herbicide application. Lack of weed control produced the lowest shallot yield. Water hyacinth mulching with hand weeding gave the highest yield and bare soil with weeds gave the lowest yield.

Table 4. Effect of integrated weeds management on yield of shallot (g/plant) at 77 DAP.

Dry mulching materials at 2 kg/3 m^2	Weed control methods			Mean
	Hand weeding	Weedy	Herbicide	
Paragrass	7.14 ab	4.93 ab	6.96 a	6.34
Lalang leaf	6.23 b	7.24 a	7.32 a	6.93
Vetiveria leaf	8.72 ab	6.24 ab	6.26 a	7.07
Water hyacinth	9.24 a	4.67 b	7.24 a	7.05
Rice straw	6.26 b	5.83 ab	6.69 a	6.26
Bare soil	7.88 ab	1.95 c	7.17 a	5.67
Mean	7.58	5.14	6.94	6.55

CV (%) = 21.2; Within a column means followed by the same letter are not significantly different by the DMRT (p=0.05).

Table 5. Effect of integrated weeds management on total yield of shallot (kg/ha) at 77 DAP.

Dry mulching materials at 2 kg./3 m^2	Weed control methods			Mean
	Hand weeding	Weedy	Herbicide	
Paragrass	2,229 abc	1,541 ab	2,176 a	1,983
Lalang leaf	1,948 bc	2,263 a	2,288 a	2,166
Vetiveria leaf	2,726 ab	1,949 ab	1,957 a	2,211
Water hyacinth	2,889 a	1,460 b	2,261 a	2,203
Rice straw	1,900 c	1,821 ab	2,091 a	1,937
Bare soil	2,463 abc	609 c	2,239 a	1,771
Mean	2,359	1,607	2,169	2,045

CV (%) = 21.2; Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05).

Conclusions

Integrated mulching material with hand weeding at 25 DAP provided the best weed control and the high shallot yield. They were higher than when pre-emergent herbicide alachlor was applied at 1.65 kg a.i./ha, immediately after planting in combination with mulching material. The best mulching material were vetiveria and lalang followed by rice straw, water hyacinth and paragrass. The results suggest that 25 DAP is a suitable time for hand weeding because at that time shallot stand is uniform with a small canopy. If mulching is not applied, herbicide use is an alternative to control weeds in shallot

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CONTROLLING NOXIOUS INVASIVE WEED *Mikania micrantha* H.B.K. IN CHINA

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Abstract: *Mikania micrantha* H.B.K., a fast-growing vine native to tropical South and Central America, is one of the world's worst weeds. It was cultivated in Hong Kong in 1884 and now expanded widely in South China, becoming a noxious invasive weed in disturbed forests and plantation crops, causing direct economic loss in farming, wood and fruit industry and serious damage to the natural ecosystem. In order to curb the invasion and to resume the destroyed natural ecosystem, many Chinese researchers have been working on this weed and hoping to find effective ways to manage it. This paper reported managing methods of mechanical removal, herbicide spraying, biological control, using parasitic plant species and vegetation-ecological control in China. Among these methods, it was concluded that large scale mechanical removal is unsustainable, selected herbicides was effective but the possibility of causing undesired impact to environment, crops and human could not be neglected, practical use of biological control was still a long way to go, parasitic plant *Cuscuta* spp. was a new sustainable control agent, and vegetation-ecological control had a promising future to control the invader entirely. In practice, combination of multiple methods was recommended.

Key words: *Mikania micrantha,* invasive species, control

Introduction

Mikania micrantha H.B.K., a climbing perennial vine, native to Central and South America, referred to as "the weed" in this paper, is one of the first ten worst weeds in the world (Holm *et al.* 1977), although it is only of minor importance in its native ranges. The first specimen collected in China was from cultivation in Hong Kong Botanical and Zoological Garden in 1884 (Wang *et al.* 2003). This record was the earliest one in the old world. More specimens were collected in the vicinity of the Garden in the early 20th century. Currently, the weed had spread widely in mainland South China and Taiwan Island. In the mainland, it is distributed most abundantly in Hong Kong, Macao, and Pearl River delta (Zhang *et al.* 2004).

Controlling the weed was extremely difficult. The weed was photophilic and able to climb up on top of shrubs and small trees, forming a mantel suffocating the supporting plant underneath. The weed grew very fast in open and wet habitat. The total length of branching from one inter-node reached 1007 m in 12 months (Wang *et al.* 2003) in open and wet habitats. The weed produced large amount of tiny seeds with pappus. The weight was only 0.0892 g per 1000 seeds (Hu and Paul, 1994; Zhang *et al.* 2003; Yang *et al.* 2003) and spread by wind. It could root from stems and stipites and thus a broken part of stem could re-grow easily.

The weed could not tolerate deep shade. A vast area survey in China indicated that very few individuals could be found in close forest and no damaging effects were noticed (also see Huang *et al.* 2000). This finding was consistent with other reports (see Bogidarmanti, 1989; Ipor, 1991). The weed most damaged shrubs, secondary forest and forest at the early succession stage (Zan *et al.* 2003), and degrading natural ecological systems (Lan and Wang, 2001; Zhong *et al.* 2004; Zan and Wang, 2000). It also cost more clearing labor in affected agricultural lands and plantations.

Control

Physical removal
An experiment to use physical removal methods in a vast area was conducted in July 1998 and proved to be futile. A site of 2000 m2 densely grown weed was selected in Neilingding Island, an island close to Hong Kong. By using a total of 70 days manpower, the above ground portions were all cut and the roots were dug out as much as possible. The slashed weed was laid under full sunlight. Unfortunately, the weed regenerated from remnants and re-colonized 80% of the ground by 3 months and 100% by 6 months (Zan and Wang, 2000). The fast re-colonization of the weed was the result of fast sprouting from the remnant of the root system and the cut stem fragments developed adventitious roots from nodes and started to re-grow in a short time. This indicated that for effective physical removal, the measure should be repeated within several months. Although this could be done in plantations under intense care, where the weed were regularly cleared before noticeable damage appeared, over a vast area physical removal of the weed in the field was not practical.

Chemical control
The first chemical control study conducted in China was in 1994. Four herbicides, i.e., Roundup, Bentazon, Torton and Ronstar, were found to be able to suppress seed germination and seedling growth, and among which Bentazon and Torton had more inhibitory effects (Hu and But, 1994). Later, in 1999~2000, four other herbicides, *i.e.*, 24-D, Monsanto, 25% Hexazinone and 75% sulbometuron-methyl, were tested (Zan *et al.* 2001). Considering both the killing ability and the negative effect on non target species, it was found that sulbometuron-methyl being the best among them. Further tests indicated that 0.01~0.1g m-2 of sulbometuron-methyl could kill the weed totally (Wang *et al.* 2003; see also Xu, 2001).

The best time of spraying was after mass seed produced in winter germinate in spring and early summer and grow considerably. There would be no more seeds from the mass seed production which is still viable at this time. A good practice was to perform a second spraying about 15 days after the first spraying. This was necessary because it was impossible to spray all individuals in the vast area once. The second spraying was done when leaves of sprayed weed were turned brown and dried, leaving un-sprayed ones obviously green and could be spotted easily. Based on our experience, two sprays ensured no more living clump left.

From November 2001 to October 2002, sulfometuron-methyl at concentrations mentioned above were sprayed on a total area of 510 hm^2 heavily covered by the weed in Shenzhen and Neilingding Islands. Except on swampy areas, the weed was completely killed after spraying and there were only minor negative effect on other plants, vertebrates and insects. Within 5 months of monitoring after the second spray, neither sprout nor new individuals were discovered except on swampy areas. Considering the fact that devastating effects of the weed on the plant community usually appeared after several years of growth, it was reasonable to anticipate that spraying herbicide could relieve plant community from the suppression of the weed at least for several years.

The backdrop of chemical spraying was that the weed would come back several years later. Furthermore, even though selected herbicides were powerful in controlling the weed, the use of herbicide must be carried out wisely to avoid the possible negative effects to other organism and environment (Li *et al.* 2000).

Biological control
Biological control was the most attracting method to control exotic species. Many researchers had done a lot of work looking for natural enemies of the weed. In China, the weed samples with some lesions were collected in Shenzhen and Zhuhai (Han *et al.* 2001). The microbe was

then identified as *Pesudocercospora* sp. The microbe could infect the new leaves of *M. micrantha* and made them turn yellow till they perished. The fungus *Sclerotium rolfsii* which has a lethal effect on *M. micrantha* was separated for the first time (Fu e*t al*. 2003). The host range tests with 86 plant species from 42 families showed that *Actinote anteas* only completed its life cycle on *Chromolaena odorata* and *M. micrantha* (Li *et al.* 2004). However, larvae of *A. anteas* consumed *Brassica* spp. This eventually hindered the release of *A. anteas* (Han, 2007 - personal communication).

Parasitic infection

During a vast area investigation in early 2001, it was discovered that parasitic plants *Cuscuta campestris*, *C. chinensis* and *C. australis* could infect *M. micrantha* and cause death. *C. campestris* was found to be the most effective one among them (Liao *et al.* 2002; Zan *et al.* 2003). Eleven infection plots established during 2000-2001 were revisited in May 2002. Results showed that *M. micrantha* was suppressed in all plots. However, it should be noted that the growing of *Cuscuta* sp. always lagged behind that of *M. micrantha*. Investigation on the ecological safety indicated that *C. campestris* did not pose a threat on other locally available species.

Vegetation-ecological control

The weed neither existed in primary closed forest nor climbed on top of closed forest, indicating the closed forest environment is unsuitable for the weed to run wild. Hence, two pilot projects of vegetation reconstruction was established in 2001 and 2002, respectively, to create environments not favoring the weed to grow with least human intervention. Some regional tree species were selected. An idealized preference was fast growing, wide and dense canopy, high ability to reproduce, and the ability to expand easily. In addition, seedling should be available in the local nursery. 18 species were used although no single species met all preferred criteria. All caring was terminated at the end of 2002 until summer 2005 when climbers on the canopy were cut at the base. Thus the reconstruct forest almost grew by itself and it was predicted that the weed would never damage the reconstructed forest in the future. The reconstruction of the forest proved to be successful. Further, it was anticipated that the reconstructed forest would gradually expand as some trees start to fruit.

Conclusions

Physical removal could be done in small area but it is impractical for large area management. Certain herbicides could be used after careful selection. Emphasis should be placed to avoid damage to non-target plants and environments. Biological control was possible but there is a long way to go before agents could be safely and successfully released. Using certain parasitic *Cuscuta* sp. to suppress the weed was useful and found to be ecologically safe, but the growth of dodder always lagged behind the weed. Creating and environment that was unsuitable for the weed to grow would not only terminate the invasion but also reverse the invasion. Integrated management was highly recommended to combat the weed.

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EFFECT OF LOW DOSE HERBICIDES ON WEED AND YIELD OF TRANSPLANTED WINTER RICE (*Oryza sativa*)

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Abstract: A field experiment was conducted during the rainy seasons of 2004 and 2005 at Regional Research Sub-station (RRSS), Chakdaha under Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal. Pre-emergence application of a mixture of triasulfuron + pretilachlor at 0.009 + 0.5 kg a.i/ ha recorded the minimum weed density and their biomass than rest of the chemical treatments. This treatment registered a higher weed control efficiency next to the weed-free check and hand weeding twice. Hand weeding and chemical weeding treatment with triasulfuron + pretilachlor (0.009 + 0.5 kg a.i/ ha) gave significantly higher grain and straw yields. The chemical weeding treatment recorded a 39 % higher grain yield than the un-weeded control.

Key words: Low dose herbicides, transplanted winter rice, weeds.

Introduction

Transplanted rice faces the threat diverse types of weed flora, consisting of grasses, sedges and some broad-leaved weeds. Attacks of these weeds are predominant at initial 15 to 30 days after transplanting (DAT). Competition of these weeds brought about reduction in yield in transplanted winter rice by about 50 % (Anantha Kumari and Rao, 1993). Several herbicides such as butachlor, anilofos and pretilachlor have been recommended for the control of weeds in transplanted rice, which are effective against grassy weeds only. Some herbicides like 2, 4 - DEE controlled only broad-leaved weeds. Further, these herbicides are applied in large volume, which will distort our eco-system and increase the chemical load on the environment. Thus for control of mixed flora, evaluation of new herbicides with low application rates is required. The present investigation was therefore conducted to study the effect of low dose herbicides on weeds and yield of transplanted winter rice.

Materials and Methods

A field experiment was conducted during the rainy seasons of 2004 and 2005 at Regional Research Sub-station (RRSS) under Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal. The soil was sandy clay loam having a pH 7.3, organic carbon 0.58 %, total N 0.58 %, available P_2O_5 16.05 kg/ha and available K_2O 126.2 kg/ha. The experiment comprised of 11 weed control treatments, along with a weed free check, hand weeding and an un-weeded control (Table 1).

The experiment was laid out in a randomized block design (RBD) with 3 replications. Twenty-five (25) day old seedlings of rice variety IET 4094 (Khitish) were transplanted on 22nd and 24th July during 2004 and 2005 respectively. One fourth of the recommended dose of N (15 kg/ha) and full doses of P_2O_5 and K_2O (30 + 30 kg/ha) were applied before transplanting (as basal) and the remaining amount, i.e. half the amount of N (30 kg/ha), was top-dressed at active tillering and one fourth of N (15 kg/ha) at panicle initiation stage. Herbicides were applied as per the treatments after transplanting using 600 liters of water with the help of a Knapsack sprayer, fitted with flat-fan nozzle. The data on total weed population and weed biomass were taken at 30, 60 and 90 days after treatment (DAT) with the help of a random quadrat (0.5 m × 0.5 m) at two places and then converted into square meters.

Table 1. Treatments imposed in the experiment.

No.	Treatments	Concentration (%)	Dosage (kg a.i. / ha)	Time of application DAT)
T1	Butachlor	50 EC	1.50	3-5
T2	Pretilachlor	50 EC	0.50	3-5
T3	Pretilachlor	50 EC	0.75	3-5
T4	Triasulfuron	20 WSG	0.006	5-7
T5	Triasulfuron	20 WSG	0.009	5-7
T6	Triasulfuron	20 WSG	0.006	12-15
T7	Triasulfuron	20 WSG	0.009	12-15
T8	Triasulfuron + Pretilachlor	20 WSG + 50 EC	0.006 + 0.500	5-7
T0	Triasulfuron + Pretilachlor	20 WSG + 50 EC	0.009 + 0.500	5-7
T1o	Bensulfuron methyl	60 DF	0.05	20-25
T11	Bensulfuron methyl	60 DF	0.06	20-25
T12	Weed free check	-	-	15, 30, 45 and 60
T13	Two hand weedings	-	-	20 & 40
T14	Un-weeded control	-	-	-

EC – Emulsifiable Concentration, WSG – Water-soluble granules, DF – Dry formulation, DAT – Days after transplanting

The weed control efficiency (WCE) and Weed Index (WI) were calculated by the following formulae:

$$\text{Weed Control Efficiency (WCE)} = \frac{X - Y}{X} \times 100$$

where, X = weed dry weight in unweeded plot, Y = weed dry weight in treated plot

$$\text{Weed Index (WI)} = \frac{X - Y}{X} \times 100$$

where, X = grain yield of weed free plot and Y = grain yield of treated plot

After harvesting, grain and straw yields were recorded through sun-drying and percentage increase in yield over unweeded control was also calculated.

Results and Discussion

Effect on weeds

The major weed flora of the experimental site consisted of grasses *viz*. *Echinochloa crus-galli, E. colonum, Cynodon dactylon,* sedges*, viz. Cyperus rotundus, C. iria*, *Fimbristylis miliacea* and broad-leaved weeds*, viz, Ammania baccifera* and *Ludwigia parviflora*.

All treatments registered significantly lower number of weeds and weed dry matter than the un-weeded control. Mixed application of triasulfuron + pretilachlor (0.009 + 0.5 kg a.i/ ha) maintained a lower crop-weed competition from the very commencement of the crop and registered lower weed density, resulting in higher weed control efficiency. Application of triasulfuron + pretilachlor (0.009 + 0.5 kg a.i./ha) was significantly superior to all other chemical treatments in controlling weed density at 30, 60 and 90 DAT in both years of experimentation.

Among the different measures, minimum weed biomass was noted with the weed free check, which was followed by hand weeding and triasulfuron + pretilachlor (0.009 + 0.5 kg a.i/ha). All treatments significantly reduced weed biomass as compared with un-weeded control (Table 2). Triasulfuron + pretilachlor (0.009 + 0.5 kg a.i/ha) applied at 5-7 DAT was significantly superior to all other herbicidal treatments in arresting weed biomass.

Table 2. Effect of low doses of herbicides on weed density, weed biomass and weed control efficiency (Pooled data of two years)

Treatment No.	Weed density (number/m^2)			Weed biomass (g/m^2)			Weed control efficiency (%)		
	30 DAT	60 DAT	90 DAT	30 DAT	60 DAT	90 DAT	30 DAT	60 DAT	90 DAT
T_1	3.2	7.3	22.1	0.53	1.80	5.57	41.75	38.56	29.13
T_2	3.8	8.3	27.3	0.65	1.87	6.33	28.57	36.17	19.46
T_3	4.1	8.2	29.6	0.73	1.80	6.84	19.78	38.56	12.97
T_4	2.9	6.3	20.4	0.49	1.62	5.13	46.15	44.70	34.73
T_5	2.9	6.3	20.8	0.48	1.61	5.11	47.25	45.05	34.98
T_6	3.2	7.4	23.5	0.54	1.74	5.61	40.65	40.61	28.62
T_7	3.1	6.4	21.2	0.54	1.66	5.19	40.65	43.34	34.96
T_8	2.2	6.2	21.6	0.40	1.52	4.75	56.04	48.12	39.56
T_9	1.6	3.8	16.7	0.18	0.82	4.01	80.21	72.01	48.98
T_{10}	3.3	7.5	21.5	0.52	1.87	5.51	42.85	36.17	29.89
T_{11}	3.1	7.2	21.3	0.44	1.77	5.42	51.64	39.59	31.04
T_{12}	1.0	3.1	11.5	0.11	0.39	3.01	87.91	86.68	61.70
T_{13}	1.1	3.6	13.5	0.13	0.53	3.64	85.71	81.91	53.68
T_{14}	4.6	12.8	33.6	0.91	2.93	7.86	0	0	0
S.Em (±)	0.24	0.23	0.89	0.05	0.09	0.22	-	-	-
C.D. (p=0.05)	0.7	0.67	2.6	0.15	0.26	0.64	-	-	-

Refer to Table 1 for description of the treatments.

Application of triasulfuron + pretilachlor (0.009 + 0.500 kg a.i/ha) at 5-7 DAT was most effective against grasses and sedges and resulting in higher weed control efficiency at 30 (80.21%), 60 (72.01%) and 90 DAT (48.98%).

Effect on grain and straw yield of rice

Yield of rice revealed that all the measures proved superior to un-weeded control (Table 3). Among the chemical treatments, the maximum grain (4.67 t/ha) and straw yield (5.11 t/ha) were recorded with triasulfuron + pretilachlor (0.009 + 0.5 kg a.i/ha) and was on par with hand weedings and superior to rest of the treatments in both years of experimentation. Plants under this treatment produced 59% higher grain yield than un-weeded control. Higher grain yield with triasulfuron + pretilachlor was due to lower crop-weed competition, higher weed control efficiency and more nutrient uptake of crop plants. This finding is in agreement with the findings of Reddy (2005).

Thus, we may conclude that the costly and troublesome hand weeding can safely be replaced by the application of triasulfuron + pretilachlor (0.009 + 0.5 kg a.i/ha) at 5 - 7 DAT.

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Table 3. Effect of herbicides on grain and straw yield of rice (pooled data of two years).

Treatment No.	Grain yield (t/ha)	Straw yield (t/ha)	% increase over control	Weed index (%)
T_1	3.76	4.37	25.33	14.54
T_2	3.20	3.22	6.66	27.27
T_3	3.40	3.87	13.33	22.72
T_4	3.76	4.40	25.33	14.54
T_5	3.79	4.38	26.33	13.86
T_6	3.59	4.14	19.66	18.40
T_7	3.61	4.15	20.33	17.95
T_8	3.84	4.52	28.00	12.72
T_9	4.17	5.11	39.00	5.22
T_{10}	3.47	4.01	15.66	21.13
T_{11}	3.54	3.96	18.00	19.54
T_{12}	4.40	5.67	46.66	0
T_{13}	4.27	5.23	42.33	4.54
T_{14}	3.00	3.24	0	31.81
S.Em±	0.07	0.04	-	-
C.D. (p=0.05)	0.2	0.12	-	-

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WEED MANAGEMENT IN POINTED GOURD (*Trichosanthes dioica* Roxb.)

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Abstract: Weeds have been identified as the main constraint in achieving the potential yield Pointed gourd (*Trichosanthes dioica* Roxb.). A field experiment was conducted for two consecutive years at the Horticultural Research Station of Bidhan Chandra Krishi Viswavidyalaya, West Bengal, India to study the influence of different weed management treatments *viz*., wheat straw, black polythene, rice husk, sawdust (ecological weed control), paraquat dichloride at the rate of 1.0 kg ha^{-1}, 2,4-D sodium salt at 0.8 kg ha^{-1} , Butachlor at 1.25 kg ha^{-1} , Benthiocarb at 1.0 kg ha^{-1} (chemical weed control) and hand weeding (physical weed control) twice at 30 and 60 days after sowing (DAS), with a weedy check. The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replicates. The recommended cultural practices including the insect and disease management were followed. Results revealed that, out of the fourteen dominant weed species in the experimental field, eight species were monocot narrow-leaved while six were dicot broad-leaved types. Among the different treatments, paraquat dichloride at 1.0 kg ha^{-1}, 2,4-D sodium salt at 0.8 kg ha^{-1}and hand weeding exhibited better performances than the rest in reducing the population and dry matter accumulation of weeds. The vegetative growth attributes of the crop were found to have maximum values in the treatments of paraquat dichloride at 1.0 kg ha^{-1} and 2,4-D sodium salt at 0.8 kg ha^{-1}. Hand weeding also showed good performances but was costly. In relation to yield attributes, these treatments showed better performances than the rest. The early and total yield of the crop was the maximum (7.56 and 13.23 t ha^{-1}, respectively) in plots treated with paraquat dichloride at 1.0 kg ha^{-1} followed by hand weeding twice (6.69 and 11.71 t ha^{-1}, respectively).

Key words: Weed management, Pointed gourd, production improvement.

Introduction

The current agricultural production strategies involve the use of high yielding varieties of crops grown under heavy and costly inputs like fertilizers, irrigation and repeated measures of plant protection. These conditions also help in stimulating the growth of crops and weeds alike. In Pointed gourd (*Trichosanthes dioica* Roxb.), weeds are the main obstacle for its successful growing particularly during its later stages of growth as it remains in the field for longer periods. The normal practice to control weeds in this crop is through physical and cultural methods like hoeing and hand weeding and mulching, respectively. These operations, particularly hand weeding, have become expensive with the rise in labour wages. Farmers generally have to pay a major portion of the cost of cultivation for weed management. A major step in this direction could be substitution of hand weeding by use of herbicides. Hence, the present study was undertaken to evaluate different weed management in Pointed gourd (*T. dioica* Roxb.).

Materials and Methods

The experiment was conducted at the Horticultural Research Station, Bidhan Chandra Krishi Viswavidyalaya, India, during 2003-04 and 2004-05, in a sandy loam soil with a pH of 6.8. The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replicates. Treatments include different herbicides namely, paraquat dichloride at 1.0 kg ha^{-1} (Gramoxone®), 2, 4-D sodium salt at 0.8 kg ha^{-1} (Fernoxone®), Butachlor at 1.25 kg ha^{1} (Machete®), Benthiocarb at 1.0 kg ha^{-1} (Saturn®), mulching materials namely, wheat straw, black polythene, rice husk and saw dust, and hand weeding. A weedy check was used as the control. Root cuttings of Pointed gourd (*Trichosanthes dioica* Roxb.) cv. Damodar Local, were planted at a ration of 9:1 (female:male), on raised beds of 9 m x 3 m during first fortnight of November of the first year of experimentation. The spacing between rows was 3 m and hill to hill was 60 cm. The same plots were used for the second year experimentation. After first year cropping, the vines were pruned keeping a small portion of the vine above the soil surface. With the cessations of winter, sprouting was initiated from the underground portion. The fertilization was done at 90 kg N, 60 kg P_2O_5 and 40 kg K_2O per hectare. Other cultural practices were followed in time as scheduled for its cultivation. Harvesting was started from 120 days after planting (DAP) onwards and continued upto 210 DAP. Observations were taken on population and dry weight of weeds, growth and yield attributes and yield of the crop.

Results and discussion

The major weed flora present in the experimental field were *Echinochloa colona*, *Digitaria sanguinalis* and *Eleusine indica* (grasses), *Cyperus rotundus* (sedge) and *Physalis minima*, *Portulaca oleracea*, *Digera arvensis*, *Melilotus alba*, *Chicorium intybus* and *Chenopodium album* (broad leaves).

Table 1 shows that the maximum reduction in narrow leaved weed population (grasses and sedges) was observed in hand weeding treatment at 45, 90 and 135 DAP in both the years, followed by the plots treated with paraquat dichloride at 1.0 kg ha^{-1}, which rendered better performance in controlling weeds among all other herbicides and mulching materials. Lower weed populations during the second year were due to the intensive cultural practices followed during the first year. Similarly, population of broad leaved weeds increased with the age of the crop. The total dry weight of weeds increased with the increase in the age of weeds during both years. All the treatments were significantly effective in checking the dry weight of the weeds over the unweeded control and saw dust mulching. Hand weeding, 2, 4-D sodium salt at 0.8 kg ha^{-1} and paraquat dichloride showed better weed control at all the growth stages. These herbicides have been able to manage the weed density, in addition to minimize the competition offered by the newly grown weeds.

Table 1. Effect of different weed management treatments on weed population and dry weight of weeds in pointed gourd

Treatments	Weed population			Dry weight of weeds (g)		
	2003-04	2004-05	Mean	2003-04	2004-05	Mean
2, 4-D sodium salt (0.8 kg ha^{-1})	189.06	150.33	169.69	21.06	17.53	19.29
Paraquat dichloride (1.0 kg ha^{-1})	164.13	131.53	147.83	26.00	20.53	23.26
Benthiocarb (1.0 kg ha^{-1})	247.13	205.33	226.23	45.40	25.86	35.63
Butachlor (1.25 kg ha^{-1})	249.66	202.20	225.93	32.80	27.66	30.23
Hand weeding	103.60	75.13	89.37	19.20	10.26	14.73
Wheat straw	181.53	147.86	164.70	32.93	26.93	29.93
Rice husk	182.26	156.93	169.59	25.00	20.66	22.83
Black polythene	149.53	123.06	136.30	24.40	20.20	22.30
Saw dust	209.60	149.46	179.53	26.13	20.66	23.39
weedy check (control)	303.73	263.63	283.68	40.80	36.06	38.43
C.D. (p=0.05)	1.874	1.765	1.831	0.819	0.639	0.732

The result presented in Table 2 show that all the treatments exerted positive effect on vine length of pointed gourd over the control, in both years. The maximum vine length of 289 cm was recorded from the plots treated with paraquat dichloride, followed by those treated with

2, 4-D sodium salt (237 cm) and in hand weeded control. Superiority of these chemical treatments over the others was mainly due to their effectiveness in keeping the weeds under control and thus the stimulating growth of the crop.

Table 2. Effect of different weed management treatments on pointed gourd

Treatments	Vine length (cm)	Vine Number Plant^{-1}	Fruit Number Plant^{-1}	Fruit length (cm)	Fruit diameter (cm)	Fruit weight (g)
2, 4-D sodium salt (0.8 kg ha^{-1})	237.96	13.53	308.00	6.92	3.17	30.62
Paraquat dichloride (1.0 kg ha^{-1})	289.05	16.79	328.80	7.12	3.24	36.10
Benthiocarb (1.0 kg ha^{-1})	168.03	12.09	252.00	6.68	2.99	27.37
Butachlor (1.25 kg ha^{-1})	165.26	11.46	261.65	6.66	3.09	27.54
Hand weeding	226.43	13.90	285.90	7.31	3.49	36.77
Wheat straw	149.80	12.20	249.30	5.98	2.71	27.94
Rice husk	121.43	10.30	209.89	5.73	2.74	23.10
Black polythene	142.19	12.43	216.63	5.98	2.79	26.37
Saw dust	124.93	10.86	213.80	5.95	2.78	32.4
weedy check (control)	82.63	3.03	26.14	5.61	2.62	22.1
C.D. (p=0.05)	1.909	0.674	3.627	0.071	0.020	0.695

Hand weeding showed better results in controlling all categories of weeds as this treatment did not allow severe crop - weed competition in during the growth of crop. A significant effect ($p<0.05$) of treatments was observed on the number of vines per plant. In this regard, paraquat dichloride was proved to be the most effective among all other treatments, followed by 2, 4-D sodium salt and hand weeding. The increased number of vines in these treatments might be due to that these treatments effectively controlling the weed growth and thus minimizing crop-weed competition enabling the crop to grow and develop at its full capacity. Greater nutrient uptake by the plants resulting in a vigorous growth and higher total number of fruits per plot were also higher in these treatments. In relation to length, diameter and weight of fruits, a significant variation was observed during both the years. Hand weeding and paraquat dichloride showed their dominance over all other treatments. The treatments other than the above two also showed better performances than un-weeded control as they were effective in checking weed growth, thus facilitating the crop to take up nutrients and to grow upto their full vigour, which in turn helped in subsequent fruit set and development. The un-weeded control significantly ($p<0.05$) hampered the crop growth, fruit setting and development.

The early and total yield of the crop varied significantly with the different treatments in both the years of experimentation (Table 3). Paraquat dichloride applied plots produced higher yields, which was statistically significant ($p<0.05$) to all other treatments. However, other chemicals did not show such a high response and thus slightly inferior to handweeding treatment. Among the physical weed control treatments, hand weeding proved its superiority over different ecological mulching treatments.

The marginal benefit/cost ratio presented in Table 4 was positive and was the highest in the plots treated with paraquat dichloride (4.62). This was because of low added cost and high added advantage with the treatment. Hence, on economic point of view of weed control, this treatment was superior to the rest used in this study. The lowest marginal benefit/cost ratio found in plots treated with Benthiocarb, due to the higher added cost and lowest added advantage. The result corroborated the findings of Leela (1993) who opined that weed control using herbicides is more effective than hand weeding in cucurbits.

Table 3. Effect of different weed management treatments on yield of pointed gourd.

Treatments	Early yield (g ha^{-1})			Total yield (g ha^{-1})		
	2003-04	2004-05	Mean	2003-04	2004-05	Mean
2, 4-D sodium salt (0.8 kg ha^{-1})	47.95	72.80	60.37	83.92	127.40	105.66
Paraquat dichloride (1.0 kg ha^{-1})	65.35	85.90	75.62	114.36	150.33	132.34
Benthiocarb (1.0 kg ha^{-1})	39.11	47.44	43.27	68.44	83.00	75.72
Butachlor (1.25 kg ha^{-1})	42.83	51.33	47.08	74.96	87.30	81.13
Hand weeding	57.71	76.10	66.91	100.99	133.18	117.08
Wheat straw	37.71	51.19	44.45	66.00	89.59	77.79
Rice husk	23.26	39.34	31.30	40.70	68.85	59.46
Black polythene	28.40	44.69	36.54	49.70	78.22	63.96
Saw dust	29.33	37.74	33.53	44.07	46.55	45.31
weedy check (control)	13.36	16.91	3.63	25.88	26.85	26.36
C.D. (p=0.05)	1.424	2.665	2.145	2.491	4.683	3.487

The results suggest that paraquat dichloride (1.0 kg ha^{-1}) or 2, 4-D sodium salt (0.8 kg ha^{-1}) may be used in suppressing the growth of weed population as well as for increased total yield of pointed gourd (*Trichosanthes dioica* Roxb.).

Table 4. Marginal benefit/cost ratio of weed control in pointed gourd.

Treatments	Quantity required ha^{-1}	Yield g ha^{-1}	Added cost (Rs.)	Total return (Rs)	Added advantage (Rs)	Marginal benefit:cost ratio
2, 4-D sodium salt (0.8 kg ha^{-1})	1 kg	105.66	241.52	31698.00	-3426.00	-14.18
Paraquat dichloride (1.0 kg ha^{-1})	2 l	132.34	991.52	39702.00	4578.00	4.62
Benthiocarb (1.0 kg ha^{-1})	2 l	75.72	891.52	22716.00	-12408.00	-13.92
Butachlor (1.25 kg ha^{-1})	2 l	81.13	1091.52	24339.00	-10785.00	- 9.88
Wheat straw	16500 bundles	77.79	3300.00	23337.00	-11787.00	- 3.57
Rice husk	44000 kg	59.46	4400.00	17838.00	-17286.00	- 3.93
Black polythene	10000sq.m	63.96	19330.00	19188.00	-15936.00	- 0.82
Saw dust	55000 kg	45.31	2750.00	13593.00	-21531.00	- 7.82
Hand weeding	245 laboures for 7 times	117.08	5605.00	35124.00	-	-

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WEED SPECIES OF RICE OVER THE LAST DECADE IN THE MUDA GRANARY OF MALAYSIA

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Abstract: Weeds are a major constraint to rice farmers in Malaysia causing increased costs and substantial crop losses. To ascertain the principal weed species, the extent of infestations and changes over time, we conducted field surveys in the Muda rice scheme for the rice growing seasons of 2001-2005 and these were compared with data generated by the Muda Authority from 1995 through 2005. The Muda Authority focused on nine weed species that were dominant, namely *Echinochloa* species (*E. crus-galli, E. oryzicola* and *E. colona*), *Leptochloa chinensis, Oryza sativa* (weedy rice), *Ischaemum rugosum, Sphenoclea zeylanica, Marsilea crenata, Ludwigia hyssopifolia, Scirpus grossus* and *Fimbristylis miliacea*. In the off-season surveys of 2001-2005, a total of 58 weed species belonging to 26 families were recorded. The dominant species were *E. crus-galli, L. chinensis,* weedy rice, *Ludwigia hyssopifolia* and *F. miliacea.* Canonical Correspondence Analysis (CCA) indicates that some species were associated with individual years including *E. stagnina*, *Cyperus polystachyos* and *F. acuminata* with the first year of the study. While many species were broadly similar between the seasons, in part reflecting the common and prevailing practices associated with direct seeding (DS), other species did show change as in the case of weedy rice which increased eight fold between 1995 and 2001. The ingression of weedy rice is likely to be a persistent problem with the movement between farms of machinery (rotovator, combine harvester, etc.) contaminated with weedy rice seeds and while the use of uncertified rice seeds continues.

Key words: Direct-seeded rice, weed survey, weedy rice, *Echinochloa crus-galli*

Introduction

In 2005, rice was grown on an estimated 611,000 ha in Malaysia accounting for 13% of the total agricultural area. The Muda irrigation scheme is a principal rice growing area in Peninsular Malaysia and covers an area of 96,000 ha (Anon., 2004). In the late 1980s there was a rapid change in rice crop establishment practices from transplanting to direct-seeding (DS), and by the late 1990's about 80% of the Muda granary was direct sown (Morooka and Jegatheesan, 1996).

Wet-seeding is currently used on about 95% of the rice fields in the Muda area and the seed is normally scattered by hand broadcasting or by using a motorized blower. The change in crop establishment practice led to substantial changes in the weed species composition with a shift away from broadleaved weeds, which in many cases were relatively easy to control, and towards the annual grass weeds (Ho and Zuki, 1988). The increasing importance of grass weeds has increased the difficulty of effective weed control and there are needs for more information on the occurrence of individual weed species as weed control issues have become more intractable. Weed problems will reflect management practices and, in common with many other rice growing areas, there is the need for evaluation of practices with respect to the need to optimize use of irrigation water and reduce production costs. Monitoring changes in weed populations has become a priority to clearly define the emerging problems to formulate appropriate weed management strategies. This paper reports the results of studies to identify the evolving weed problems in the Muda rice area.

Materials and Methods

Weed Surveys and data analysis

A survey of weeds was conducted by the Malaysian Agricultural Research and Development Institute (MARDI) in DS rice fields in the Muda area in the off-seasons of 2001- 2005 between the rice growth stages of booting and heading**.** The surveys were conducted after farmers had weeded and applied herbicides to the rice fields and therefore these data recorded those weed species which had survived the farmers' control measures and those that are likely to pose increasing problems in subsequent crops. The survey was planned to provide a representative coverage of the whole area. Two field assistants surveyed each field. A total of 382, 428, 582, 788 and 1145 rice fields were surveyed covering the whole Muda irrigation scheme in the off season 2001, 2002, 2003, 2004 and 2005, respectively. A rating scale of 0 to 10 to denote weed cover was used (Pablico and Moody, 1985; Elazegul *et al.* 1990). Weed species and their percentage cover were recorded. The percentage cover of weed species prevailing above the rice canopy was based on the whole field after walking in and along both sides of the field, whereas the percentage cover of weeds below the crop canopy was taken from four 1 m^2 quadrates placed randomly in each rice field. Comparisons were made of the occurrence and density of individual species. Estimates of rice yields in the main- and off seasons from 2001 through 2005 were taken by MADA using random crop cuts (5 m x 5 m). A combination of quadrate and visual assessment was used to reduce the time taken to survey a field and to prevent damage to the crop.

Between 1995 and 2005, MADA conducted weed surveys in every season using similar procedures to those described above, but without quadrate assessments, and these emphasized only the nine principal species of rice weeds. Further, these surveys included only those fields where weed infestations were deemed to be damaging i.e. infesting more than 30% of the field. These infested areas were recorded together with the weed species. The rationale of selecting fields with infestation in excess of 30% coverage of weed species, as such weed coverage may inflict substantial rice yield loss. The weed species of interest to MADA were *Echinochloa species* (*E. crus-galli, E. oryzicola* and *E. colona)*, *Leptochloa chinensis*, weedy rice, *Ischaemum rugosum, Sphenoclea zeylanica, Marsilea crenata, Ludwigia hyssopifolia, Scirpus grossus* and *Fimbristylis miliacea.*

Collated data on the incidence of weed species in five seasons of 2001-2005 were analyzed based on percentage of weed infestation (above canopy) and canonical correspondence analyses undertaken (Ter Brak and Smilauer, 1998) to assess association, if any, between species prevalence, yield and average total annual rainfall in the off seasons of 2001 through 2005. Species rank order diagrams were created for the data collected by MADA at intervals from 1995 to 2005**.**

Results and Discussion

In the 2001-05 MARDI survey, a total of 58 weed species were recorded belonging to 26 families comprising of 27 broadleaved weeds, 14 grasses, 12 sedges, and 5 aquatic species. Overall, across the years (2001-2005) there was a dominance of grass weeds in terms of incidence and percentage cover. Thirty-eight weed species were found in off season 2001 and a similar number of weed species (39) were observed in 2002, 2003 and 2004, while 48 species were registered in 2005. The most widespread weed species in terms of frequency and abundance (according to percentage fields infested value) in the off-season of 2001 were *Echinochloa crus-galli* complex (*E. crus-galli* var. *crus-galli, E. crus-galli* var. *formosensis* and *E. oryzicola*) (95% of surveyed fields) > *Leptochloa chinensis* (87%) > weedy rice (82%) > *Ludwigia hyssopifolia* (64%). In the following year (off season 2002), weedy rice had become more widespread and it occurred in 93% of surveyed fields followed by *E. crus-galli* complex (92%) > *L. chinensis* (84%) > *L. hyssopifolia* (68%). Similar patterns of species rankings to those in the off-season of 2002 were observed in the off- seasons of 2003, 2004, and 2005.

While the percentage of fields with weedy rice persisted at more than 90% over the year 2002-05 (see above), the proportion of rice fields registering trace occurrence of weedy rice showed an increase from 28% in the off-season of 2001 to 51% in off-season of 2005. Such increments in area with trace occurrence or infestation of weedy rice is an indication of the improvement in land preparation techniques and changes in crop establishment method to water seeding in off season 2005 by farmers in the Muda area. A similar trend of reduced levels of weed infestation was shown for other important weeds as well. *Echinochloa crus-galli* showed an increase in infested fields with trace occurrence from 24% in the off-season of 2001 to 75% in the off-season of 2005. In addition, the number of fields registering trace occurrence of *L. chinensis,* increased from 39% in the off-season of 2001 to 55% in the off-season of 2005**,** and for *L. hyssopifolia*, the trace occurrence rose from 53% to 69%. This increase in the fields where "*trace*" was recorded, rather than higher levels of infestation, was thought to be due to improved weed management being achieved by farmers.

The ordination diagram resulting from the canonical correspondence analysis denotes the relationships between weed species, year, rainfall and yield of the Muda area (Figure 1).

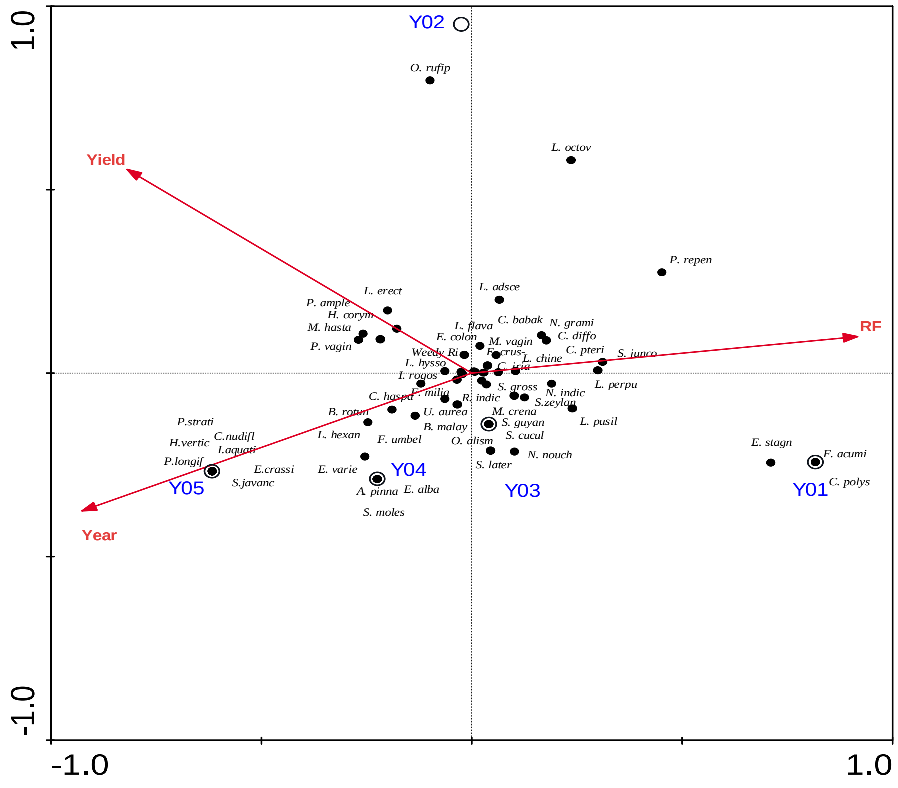

Figure 1. Canonical Correspondence Analysis ordination diagram denoting the relationships between weed species and environments {Rainfall (RF), Yield and Season} of the Muda area (off seasons, 2001 – 1005) (○; Y01 = off season 2001, Y02 = off season 2002, Y03 = off season 2003, Y04 = off season 2004 and Y05 = off season 2005). Biplot scaling of dominant weed species vectors indicating the associations of seasons (= rainfall distribution and quantity) with weed species abundance, and weed abundance and rice yield. Association of species abundance with seasons, average annual total rainfall, and yield can be ascertained by the direction of the vectors. Vector lengths indicate the relative strength of association between the respective weed species and the seasons of planting (Year), total average annual rainfall and yield. MARDI survey, 2001-05.

It is of interest that certain species are associated with particular years, as for example, *E. stagnina*, *Cyperus polystachyos* and *F. acuminata* were associated with the first year of the study (2001) while *Oryza rufipogon* was associated with 2002. *Pistia stratiotes*, *Hydrilla verticillata*, *Commelina nudiflora, Ipomea aquatica* and *Eichornia crassipes* were associated with 2005. The slightly higher number of weed species found in 2005 may have been due to an increase in water seeding which resulted in an increase of the broad-leaved aquatic weeds, though these species were of minor importance. The majority of the species recorded in the study were common to all years, and about the origin in the diagram, and these included *E. crus-galli* complex, *L. chinensis*, weedy rice, *C. iria*, *L. hyssopifolia*, *Ischaemum rugosum*, *E. colona*, *F. miliacea, Monochoria vaginalis and Scirpus grossus*.

Weed surveys undertaken by the MADA focused on the nine considered to be most damaging to the crops. Figure 2 shows the relative importance of these species in terms of area infested in four years during the off season. In 1995, 1998, and 2001, the annual grasses *Echinochloa* spp. and *L. chinensis* were ranked at the top. Over the period of the study, the rise of importance of weedy rice is noteworthy. In the off season of 1995, only about 250 ha were affected whereas by 2004 the area had risen eight fold to more than 2000 ha, or about 2% of the total rice area in the scheme.

The results of the two surveys are not directly comparable as the MADA survey included only fields that were badly infested (i.e. weeds infesting >30%) whereas in the MARDI survey fields were selected to be representative of the whole scheme. The MADA survey also only took account of those weeds that were judged to be the most serious rather than all the species. In this way the approach of only focusing on the serious weeds would fail to identify weed problems that were emerging and instead would only identify those that were major problems. Further, this survey only took into account on the taller forms of weedy rice that could be easily distinguished from the cultivated rice plants. However, it was subsequently found that there were also forms that were the same height as the crop and therefore could be overlooked. This would lead to an underestimation of the extent of field infestations.

The MARDI surveys, conducted in the off-seasons of 2001-2005, confirmed the presence of weedy rice as a significant weed problem in the area. In these surveys, weedy rice was ranked the third dominant weed in the Muda rice area in the off-season of 2001, and has become the most dominant weed in the off-season of 2002 up to off season 2005. This study revealed the rapid increase in weedy rice after the mid 1990's in all seasons compared to previous studies conducted by Azmi *et al.* (1993) and Pane (1996) in Muda area where weedy rice was not reported as a weed. Meanwhile the importance of the grasses *E. crus-galli* and *L. chinensis* as key species in the Muda area had prevailed. Azmi *et al.* (1993) and Ho and Zuki (1988) argued that the shift from transplanting to DS in the Muda fields, has resulted in the substantial increase of grassy weeds notably *E. crus-galli, L. chinensis, E. colona,* and *I. rugosum.* The results of this study suggest that weedy rice is now an important component of the weed flora and is a threat to DS rice in the Muda granary. At present, there is no selective herbicide available to control this weed, and farmers are not able to manually control this weed at early stage of the crop growing period due to its similarity to the crop and the costs of labor. Other grasses however like *E. crus-galli* and *L. chinensis* can be controlled by selective herbicides such as cyhalopfop-butyl, quinlorac, molinate, propanil, fenoxaprop-ethyl and bispyribac-sodium if applied correctly (Karim *et al*. 2004). It is suggested therefore that while chemical control measures can target *E. crus-galli* and *L. chinensis* alternative cultural control measures are required to control weedy rice. Approaches to control weedy rice that may be integrated with current practices include the use of clean certified seeds, sequential tillage for land preparation, burning of rice straw on the field, spraying of herbicides as a pre-planting procedure, manual weeding at early crop growth stage and rouging of weedy rice panicles before crop harvest. Water seeding and transplanting of rice instead of wet seeding and the rotation of the rice crop with other upland crops like maize are also possibilities to manage weedy rice. Further studies on the biology and ecology of weedy rice are however needed in order to be able to formulate effective weed control measures.

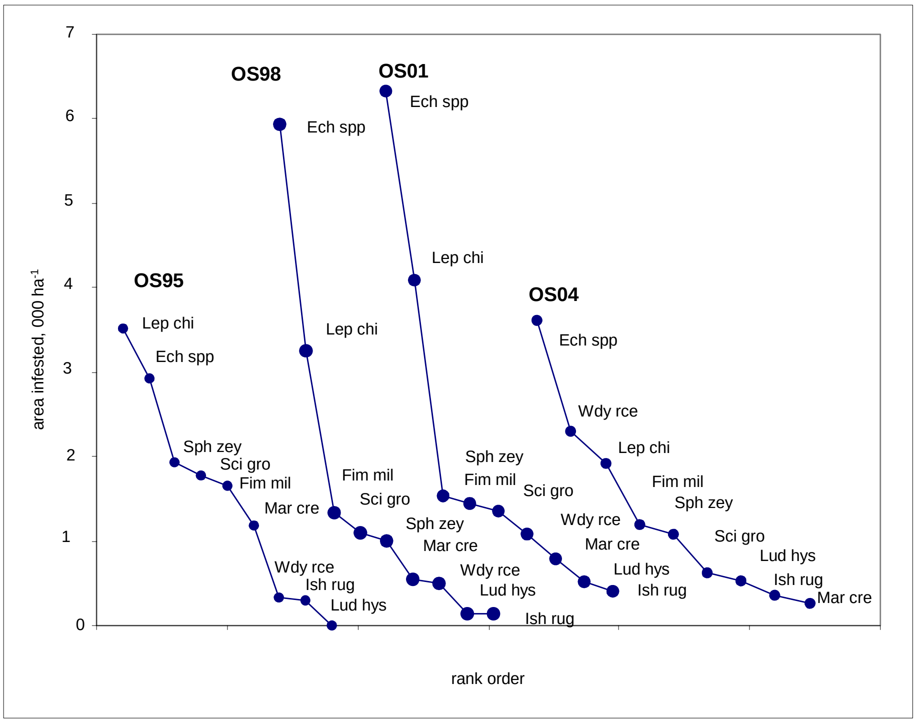

Figure 2. Rank order diagram denoting the rice areas infested by weed species in the Muda area during the off seasons at intervals 1995 – 2005, (MADA survey).

Weeds population and species diversity are dynamic in nature, and they change their abundance and dominance according to changes in rice agro-ecosystem. Rapid increases in the weed infestation and population density, especially grassy weed species such as weedy rice, *L. chinensis*, the *E. crus-galli* complex and *I. rugosum* have been recorded in the Muda granary area. The importance of these species is closely related to the changes in the establishment practices for rice in Malaysia and the shift from transplanting to DS. Weedy rice was not reported as a most noxious weed until relatively recently (Baki, 2005). The ingression of problem weeds, and weedy rice in particular, in DS fields in the Muda granary has been the result of practices that include the frequent use of shared farm machineries (rotovator, combine harvester etc.) contaminated with weedy rice seeds and the use of contaminated (uncertified) rice seeds by farmers (Azmi *et al.* 2005). The rapid establishment and success of these species as weeds is the result of repeated use of management and weed control practices that select for these species and favor their growth. Clearly, more attention needs to be given to cultural practices that help prevent the ingression of weeds and more attention to a rotation of management populations that target the build-up of problem weeds.

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NON-TARGET TOXICITY OF SOME POST-EMERGENT HERBICIDES AND INSECTICIDES AGAINST THE MEXICAN BEETLE *Zygogramma bicolorata* Pallister (CHRYSOMELIDAE : COLEOPTERA).

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Abstract: The Mexican beetle, *Zygogramma bicolorata* Pallister (Chrysomellidae : Coleoptera) is an exotic bio-control agent of parthenium, the notorious weed causing enormous crop losses and annoyance due to health hazard. After its introduction to India in 1982, the beetle has now established in many states. In nature the beetle is frequently exposed to pesticides of various kinds. Hence, laboratory studies were undertaken in the Department of Agricultural Entomology at the Bidhan Chandra Krishi Viswavidyalaya (Agriculture University), West Bengal, India to investigate the non-target toxicity of some post-emergent herbicides (at three dosages) and insecticides (at two dosages) on grubs and adults of the insect at constant temperature (25 ± 1^oC) and relative humidity ($70 \pm 5\%$). The insecticides were extremely toxic to the insect whereas, the herbicides had little direct effect. The grubs were more vulnerable to the insecticidal and herbicidal treatments than the adults. The insecticidal treatments namely, Profenofos® at 0.075 and 0.05% a.i., Spinosad® at 0.005 and 0.0038% a.i., Acetamiprid® at 0.005 and 0.004% a.i. and Imidacloprid® at 0.0045% and 0.0036% a.i. caused total mortality of the grubs within 24 to 48 hrs and of the adults within 24 to 72 hrs. Among the herbicides, glyphosate 0.08–0.2% a.i., paraquat dichloride 0.04–0.14% a.i., 2,4–D sodium salt 0.16–0.40% a.i. and metribuzin 0.04–0.11% a.i. caused 0–26.67% mortality of the grubs after 72 hrs of treatment as against 0–6.67 % in adults. The tested herbicides, though found to be safe or slightly toxic to the beetle, may affect its establishment through destruction of the weed. It is suggested that, in areas where *Z. bicolorata* has been colonized for parthenium control, protective screens should be used to prevent drifting of insecticides on the weed while carrying out spray operations in adjacent crop fields.

Key words: Pesticide toxicity, *Zygogramma bicolorata,* parthenium

Introduction

Parthenium (*Parthenium hysterophorus* L.; Asteraceae), popularly known as carrot grass, has emerged as one of the worst weeds in India due to its direct damage and harmful effects on man and animals (Khosla and Sobti, 1979). Presently it has spread over an area of 10 million ha in India (Sushilkumar, 2005). Based on well documented success of Mexican beetle, *Zygogramma bicolorata* Pallister (Chrysomelidae : Coleoptera) in controlling the weed in many countries, the beetle has been released in many areas of the country with varying results. The non-target toxicity of herbicides on *Z. bicolorata* is of immense importance as the herbicides are often utilized for the control of parthenium. In Addition, the non-target toxicity of insecticides against weed feeding herbivores is also important due to fact that they often receive insecticidal drifts particularly when the weeds grow nearer to the crop fields. Only a few attempts have been made to evaluate the effect of some herbicides on this bio-control agent and most of the herbicides have been found to be safe (Jayanth and Bali, 1993; Sushilkumar, 2005 and Patnaik *et al*. 1988). However, reports relating to toxicity of insecticides to this beetle are lacking. With this background, non-target toxicity of some post-emergent herbicides and insecticides were evaluated against the adults and grubs of the Mexican beetle, *Z. bicolorata*.

Materials and Methods

A laboratory experiment was conducted at the Department of Agricultural Entomology, Bidhan Chandra Krishi Viswavidyalaya (Agricultural University), Mohanpur, West Bengal, India with three dosages of each of the four herbicides (Table 1) and two dosages of each of the four insecticides (Table 2). Ten grubs of the third instar or five day old adults of *Zygogramma bicolorata* were placed in a 15 cm diameter Petri dish lined with filter paper and these were sprayed with 5 ml solution of the pesticide at the desired concentrations with the help of a hand atomizer. The sprayed insects were removed immediately to a separate Petri dish provided with fresh parthenium leaves as food, which were kept uncovered for half an hour to allow the spray fluid on the body of the insect, if any, to dry up. After that the Petri dishes were covered with lids and were kept $25 \pm 1^{o}C$ and $70 \pm 5\%$ relative humidity. The mortality of the insects was recorded at every 24 hrs up to 72 hrs after treatment and moribund insects were treated as dead. The observed mortality was corrected by using Abbott's formula (Abbott, 1925). Factorial CRD analysis was done for test of significance.

Results and Discussion

Effect of herbicides on the grubs and adults of Z. bicolorata

The herbicidal treatments resulted in 0-6.67% mortality of the grubs and adults after 24 hrs of treatment (Table 1). However, the average mortality obtained in case of grubs, considering all the insecticides together, was 3.33% as against 1.39% in case of adults and there was no significant difference between the two stages of the beetle in respect of mortality .When the average mortality of different herbicidal treatments were taken into account irrespective of stages of the beetle, the herbicidal treatments caused 0–5% mortality and the treatment effects were again non-significant. Similarly, the result of interaction between herbicidal treatments and stages of the beetle was non-significant which indicates that the treatments did not produce differential effect to the two stages of the insect.

After 48 hrs of treatment, the different dosages of herbicides resulted in 1.67–15% mortality of the insects irrespective of the stages of the insect. Here also the treatments failed to show any significant difference among them in respect of mortality. Only the effect of stages was statistically significant and the grubs showed higher average mortality (6.95%) than the adults (2.5%). The effect of interaction was again non-significant. Metribuzin 70% WP at 0.04% a.i. caused maximum mortality (26.67%) of grubs.

After 72 hrs, again, herbicidal treatments caused significantly higher mortality to the grubs (9.45%) than the adults (4.45%). Mortality obtained in different herbicidal treatments, irrespective of stage of the beetle, ranged from 1.67–16.67% and all the herbicides were at par among them in respect of toxicity to *Z. bicolorata* .The interaction was again statistically non-significant. Here also metribuzin at 0.04% a.i. caused the maximum mortality to the grubs (26.67%).

Table 1. Toxicity of Different Herbicides on grubs and adults of *Z. bicolorata* Pallister.

Treatments	Corrected Mortality (%)								
	24 hrs.			48 hrs.			72 hrs.		
	Grubs	Adults	Treatment (Average)	Grubs	Adults	Treatment (Average)	Grubs	Adults	Treatment (Average)
Glyphosate 41% SL 0.08%	3.33 (7.35)	0.00 (1.81)	1.67 (4.58)	3.33 (7.35)	0.00 (1.81)	1.67 (4.58)	6.67 (12.89)	6.67 (12.89)	6.67 (12.89)
Glyphosate 41% SL 0.14%	0.00 (1.81)	0.00 (1.81)	0.00 (1.81)	3.33 (7.35)	0.00 (1.81)	1.67 (4.58)	3.33 (7.35)	0.00 (1.81)	1.67 (4.58)
Glyphosate 41% SL 0.20%	3.33 (7.35)	0.00 (1.81)	1.67 (4.58)	6.67 (10.06)	0.00 (1.81)	3.34 (5.94)	10.00 (15.60)	3.33 (7.35)	6.67 (11.48)
Paraquat dichloride24% SL 0.05%	6.67 (10.06)	0.00 (1.81)	3.34 (5.94)	6.67 (10.06)	0.00 (1.81)	3.34 (5.94)	10.00 (15.60)	3.33 (7.35)	6.67 (11.48)
Paraquat dichloride 24% SL 0.09%	6.67 (10.06)	0.00 (1.81)	3.34 (5.94)	6.67 (10.06)	0.00 (1.81)	3.34 (5.94)	10.00 (15.60)	0.00 (1.81)	5.00 (8.71)
Paraquat dichloride 24% SL 0.14%	3.33 (7.35)	0.00 (1.81)	1.67 (4.58)	6.67 (12.89)	0.00 (1.81)	3.34 (7.35)	13.33 (17.87)	6.67 (12.89)	10.0 (15.38)
2,4-D sodium salt 80% WP 0.16%	0.00 (1.81)	6.67 (12.89)	3.34 (7.35)	0.00 (1.81)	6.67 (12.89)	3.34 (7.35)	0.00 (1.81)	6.67 (12.89)	3.34 (7.35)
2,4-D sodium salt 80% WP 0.28%	0.00 (1.81)	0.00 (1.81)	0.00 (1.81)	6.67 (12.89)	0.00 (1.81)	3.34 (7.35)	6.67 (12.89)	3.33 (7.35)	5.00 (10.12)
2,4-D sodium salt 80% WP 0.4%	0.00 (1.81)	3.33 (7.35)	1.67 (4.58)	6.67 (10.06)	3.33 (7.35)	5.00 (8.71)	6.67 (10.06)	3.33 (7.35)	5.00 (8.71)
Metribuzin 70% WP 0.04%	6.67 (12.89)	3.33 (7.35)	5.00 (10.12)	23.33 (28.78)	6.67 (10.06)	15.0 (19.42)	26.67 (30.99)	6.67 (10.06)	16.67 (20.53)
Metribuzin 70% WP 0.07%	6.67 (12.89)	0.00 (1.81)	3.34 (7.35)	10.00 (18.43)	6.67 (12.89)	8.34 (15.66)	13.33 (21.14)	6.67 (12.89)	10.00 (17.02)
Metribuzin 70% WP 0.10%	3.33 (7.35)	3.33 (7.35)	3.33 (7.35)	3.33 (7.35)	6.67 (12.89)	5.00 (10.12)	6.67 (10.06)	6.67 (12.89)	6.67 (11.48)
Stage (Average)	3.33 (6.88)	1.39 (4.12)	-	6.95 (11.42)	2.50 (5.73)	-	9.45 (14.32)	4.45 (8.96)	-

Factors	24 hrs		48 hrs		72 hrs	
	F test	CD	F test	CD	F test	CD
Treatments	Non-significant	-	Non-significant	-	Non-significant	-
Stages	Non-significant	-	Significant	4.18	Significant	4.98
Interaction (Treatment x stage)	Non-significant	-	Non-significant	-	Non-significant	-

Effects of insecticides on Z. bicolorata

All the insecticidal treatments resulted in total mortality of the grubs after 24 hrs except Imidacloprid® 17.8% SL at 0.0036% a.i., which caused 86.67% mortality, though the treatments were statistically at par (Table 2). Mortality of adults varied from 26.07 – 100% in different insecticidal treatments. Profenofos® 50% EC was the most toxic insecticide causing total mortality of the adults in both the dosages tested (0.05 and 0.075% a.i.), whereas Imidacloprid® was the least toxic (26.67 and 33.33% mortality, respectively, in 0.0036 and 0.0045% a.i.). Acetamiprid® 20% SP at 0.005% a.i. (86.67% adult mortality) was next to Profenofos and this treatment was statistically at par with 0.004% a.i. of the same insecticide and Spinosad® 2.5% SC at 0.005 % a.i. (both causing 80% mortality). Spinosad® 0.0038% a.i. caused 60% mortality of the adults and was significantly more toxic to the two dosages of Imidacloprid®. The insecticidal treatments caused 60–100% mortality of the beetle irrespective of its stages (*i.e.* grub or adult). The two dosages of Profenofos® caused total mortality and were the most toxic, whereas Imidacloprid® was the least toxic insecticide (60 and 63.03% mortality, respectively in two dosages). Other insecticides followed the same sequence as observed in case of adults. Among the two stages of the beetle, the grubs showed significantly higher average mortality (98.33%) than the adults (70.76%). The effect of interaction between the insecticidal treatments and stages were significant indicating that the two stages showed differential response to insecticidal treatments. As for example, the toxicity of Imidacloprid® was very low in adults (33.33 and 26.67%) than in the grubs (86.67 and 100%).

After 48 hr, all the insecticidal treatments resulted in total mortality of the grubs. The mortality of the adults was 100% in all the treatments except Imidacloprid® at 0.0045% and 0.0036% a.i. which registered 73.33 and 66.67% mortality, respectively. Taking into account all the insecticides together, the adults showed significantly lower mortality (92.5%) than the grubs (100%). The effect of treatments on the beetle, irrespective of stages, was again significant. Imidacloprid® at 0.0045 and 0.0036% a.i. caused significantly lower mortality (86.67 and 83.33% respectively) than rest of the treatments (100%). The effect of interaction between treatments and stages of the insect was significant, which was due to differences in toxicity of the two dosages of Imidacloprid® on the two stages of the beetle. After 72 hrs of treatment, the two dosages of Imidacloprid® even resulted in total kill of the adults. Thereby, the effects of stages and treatments or their interaction were non-significant.

The insecticides tested during the present investigation were extremely toxic to *Z. bicolorata,* whereas the herbicides were almost safe. Earlier, Sushilkumar (2005) observed up to 30 and 36% mortality of adults and grubs by different dosages of 2,4-D ethyl ester, whereas glyphosate and metribuzin caused maximum up to 7 and 13% mortality of the beetle by different methods of application. During the present investigation, different herbicides caused 0–26.67% and 0–6.67% mortality of the grubs and adults which was in agreement with the former author. Patnaik *et al.* (1988) also found 2,4-D to be relatively less toxic to this insect. Jayanth and Bali (1993) observed 100% mortality of the insect by topical application of paraquat, which could not be confirmed during the present investigation. The reason for higher toxicity of the lower dosage of metribuzin to the grubs, as observed during the present investigation, was not understood.

Table 2. Toxicity of Different Insecticides on grubs and adults of *Z. bicolorata* Pallister

Treatments	Corrected Mortality (%)								
	24 hrs.			48 hrs.			72 hrs.		
	Grubs	Adults	Treatment (Average)	Grubs	Adults	Treatment (Average)	Grubs	Adults	Treatment (Average)
Acetamiprid 20% SP 0.004	100.00 (89.43)	80.00 (67.88)	90.00 (78.66)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)
Acetamiprid 20% SP 0.005	100.00 (89.43)	86.67 (72.09)	93.34 (80.76)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43	100.00 (89.43)
Imidacloprid 17.8% SL 0.0035	86.67 (76.54)	33.33 (35.01)	60.00 (55.82)	100.00 (89.43)	66.67 (54.99)	83.33 (72.21)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)
Imidacloprid17.8% SL 0.0045	100.00 (89.43)	26.67 (30.79)	63.34 (60.11)	100.00 (89.43)	73.33 (64.03)	86.66 (76.73)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)
Spinosad 2.5% SC 0.0375	100.00 (89.43)	60.00 (50.77)	80.00 (70.10)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43	100.00 (89.43)
Spinosad 2.5% SC 0.05	100.00 (89.43	80.00 (67.88)	90.00 (78.66)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43	100.00 (89.43)
Profenofos 50% EC 0.05	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43	100.00 (89.43)
Profenofos 50% EC 0.075	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)	100.00 (89.43)
Stage (Average)	98.33 (87.82)	70.83 (62.35)	–	100.00 (89.43)	92.50 (81.95)	–	100.00 (89.43	100.00 (89.43	–

Factors	24 hrs		48 hrs		72 hrs	
	F test (p=0.05)	CD	F test (p=0.05)	CD	F test (0.05)	CD
Treatments	Significant	5.14	Significant	7.71	Non-Significant	-
Stages	Significant	10.29	Significant	3.85	Non-Significant	-
Interaction (Treatment x stage)	Significant	14.55	Significant	10.89	Non-Significant	-

Among the two stages of the beetle, the adults could resist the action of some of the insecticidal treatments to some extent for 24 to 48 hrs, but the grubs totally succumbed to most of them within 24 hrs. Imidacloprid® at 0.0036 and 0.0045% a.i. showed relatively lower mortality of the adults up to 48 hrs but after 72 hrs it was at par with others. Spinosad®, an insecticide of microbial origin, also caused total mortality of the grubs and adults within 24 and 48 hrs, respectively. The data obtained during the present investigation could not be compared with others as no information is available in this respect.

In conclusion it is suggested that, the herbicides should be applied with utmost care in locations where the beetle has been colonized for the control of parthenium as these may severely affect the Mexican beetle (*Z. bicolorata*), especially the grubs, by destroying the food sources. Susilkumar (2005) suggested application of herbicidal sprays by June-July, before population build-up of the beetle. It is further suggested that, in areas where *Z. bicolorata* has been colonized for parthenium control, protective screens should be used to prevent drifting of insecticides on the weed while carrying out spray operations in adjacent crop fields.

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WEED WIPER: AN INNOVATIVE METHOD FOR CONTROLLING WEEDY RICE (*Oryza sativa* f. *spontanea*) IN RICE FIELDS

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Abstract: Weedy rice (*Oryza sativa* f. *spontanea*) is a noxious weed in rice growing areas in Central Thailand. Generally, panicles of weedy rice emerge two weeks earlier than the crop-rice so that farmers could manually remove the panicles with sickles. Currently, labor has become a shortage and expensive hence demanding a cheaper but practical method to remove the weed. A simple weed wiper has been developed for herbicides, promoting seed sterility and thus reducing the seed bank of weedy rice. Two field experiments were conducted in Supanburi province to develop an application technique of herbicides as a promoter of seed sterility in weedy rice. The weed wiper was dipped in different concentrations of herbicides *i.e.* glufosinate-ammonium 15% SL, glyphosate 48% EC, paraquat 27.6% EC, quizalofop-p-ethyl 5% EC, and MSMA 72% SC, and blot-dried prior to wipe on the panicles and flag leaves of weedy rice at the flowering and three days after flowering stages. The results indicated that the weed wiper together with herbicides showed 86-98% reduction of weedy rice seeds set at both stages. However, combination of the weed wiper with paraquat and glyphosate caused crop injury. In addition, a single application with herbicides was not adequate to obtain a complete control of weedy rice as about 30% of normal panicles remained at harvest. Considering the crop safety, glufosinate-ammonium, MSMA and quizalofop-p-ethyl at the concentration of 15-30, 72 and 7.5 g ai L^{-1}, respectively, are recommended to the farmers with the weed wiper to replace the expensive labor requirement to remove weedy rice from the rice fields. The technique is cheaper and more effective than manual panicle topping-off. More importantly, this technique effectively reduced the weedy rice seed bank in the following season.

Keywords: Weed wiper, weedy rice, glufosinate-ammonium, glyphosate, paraquat, quizalofop-p-ethyl and MSMA

Introduction

Weedy rice (*Oryza sativa* f. *spontanea*) is a noxious weed occurring in rice growing areas worldwide. To date, approximately 300,000 ha of rice fields in Thailand have been infested with weedy rice. It could reduce rice yield by 10 to 100% depending on the level of infestation (Maneechote *et al.* 2004a). Herbicides for controlling of weedy rice were practiced as pre-planting, pre-emergence or early post-emergence (Azmi and Abdullah, 1998; Eleftherohorhorinos and Dhima, 2002; Ferrero, 2003; Maneechote *et al.* 2005a.). However, herbicide application for the control of weedy rice in rice cultivation is a difficult task as both belong to the same species and hence, the similarity of many morphological and physiologically characteristics. Integrated control methods are recommended in Asian countries to reduce the impact of the weed. In Malaysia, manual weeding and chemical control are recommended to be practiced with the use of clean seeds, water management and clean up farm machinery (Azmi and Abdullah, 1998). In Thailand, farmer’s participatory research indicated that integrated control methods *i.e.* use of clean seeds, eradication before sowing, hand pulling, and panicle topping-off gave a good control of weedy rice (Maneechote *et al.* 2005b).

Weedy rice plants are very competitive with the rice crop as they are generally taller and grow faster than the crop. Due to the early flowering and shattering of weedy rice, numbers of their seeds accumulated in the soil also increases. Thai farmers cut the panicles off at the flowering stage in order to deplete the seed bank in the following season and minimize the competition with rice crop. However, labor cost is an important constraint due to shortage and being more expensive. Some herbicides *i.e.* fenoxaprop and quizalofop could be sprayed on the top of weedy rice at the flowering stage to reduce filled grains (Maneechote *et al*. 2004b). However, this application technique caused crop damage when the density of weedy rice was low. Therefore, to avoid crop damage, weed wiper technique was developed to replace the expensive manual topping-off of weedy rice panicles.

Materials and Methods

Two field experiments were conducted in Supanburi province during July-September 2006. The density of weedy rice in the fields was 282 ± 66 plants m^{-2}. The panicles of weedy rice were counted in $1m^2$ of each replicate one day before herbicide application. Rice variety was Supanburi 1 sown at the rate of 125 kg ha^{-1}. At anthesis and 3 days after anthesis stages of weedy rice, a single or combinations of herbicides, glufosinate-ammonium 15% SL, glyphosate 48% EC, paraquat 27.6% EC, MSMA 72% SC and quizalofop-p-ethyl 5% EC, are carefully wiped on flag leaves and panicles of weedy rice.

The wiper equipment was simply assembled by a 2m-long bamboo stick wrapped with the 70 cm x 150 cm towel. Each herbicide was dissolved in 1 litre of water and mixed well. Each wiper was thoroughly soaked with herbicide and excessive solution was removed before use in each replicate. Plot size was 5 x 6 m^2 with 1 meter guard row. Treatments were arranged in the Randomized Complete Block Design (RCBD) with four replicates.

Twenty treated panicles of weedy rice in each treatment were collected at 14 days after wiping. Filled and unfilled grains of weedy rice/panicle were recorded. Rice yield was harvest in 2 x 2 m^2 in each replicate. Data were analyzed by ANOVA, and LSD 0.05 was used for mean comparison.

Results

Effect on seed sterility of weedy rice

All herbicides induced empty seeds of weedy rice by 86-98% when compared to the untreated panicles, when wiped at the flowering stage and three days after flowering (Tables 1 and 2). Glufosinate-ammonium at concentrations of 7.5, 15 and 30 g ai l^{-1} gave 95-97% when wiped at flowering stage. Likewise, glyphosate at 24 g ai l^{-1} applied alone or tank-mixed with paraquat at 27.6 g ai l^{-1} greatly reduced the number of filled seeds. However, they caused phytotoxicity to rice so that some panicles of rice plants underneath became abnormal (data not shown). The herbicides MSMA and quizalofop-p-ethyl were not toxic to the rice crop but the efficiency was slightly less than glufosinate-ammonium.

Effect on number of panicles of weedy rice and rice yield

At harvest, the number of weedy rice panicles was reduced from 193-252 panicles m^{-2} in untreated plot to 73-90 panicles m^{-2} after herbicide treatment (Table 3). Rice yield was increased by 1- to 4-folds of untreated control. However, single application of those herbicides reduced the number of weedy rice panicles only by 60-70% (Table 3). The remaining panicles emerging a week later could produce seeds and enrich the soil seed bank. To control the remaining panicles, a second application was recommended to farmers a week after the first application.

Table 1. Efficiency of herbicides on promoting unfilled seeds/panicle of weedy rice in Dermbang Nangbuat district, Supanburi province. Herbicides were wiped on flag leaves and panicles of weedy rice at flowering stage. Twenty panicles of weedy rice were sampled in each replicate

Treatment	Concentration (g ai l^{-1})	Weedy rice seeds/panicle			
		Total	Filled	Unfilled	% unfilled
glufosinate-NH_4	7.5	119 ab	6 b	113 ab	95 ab
glufosinate-NH_4	15	127 a	4 b	123 a	97 ab
glufosinate-NH_4	30	120 ab	3 b	117 ab	97 a
glyphosate	24	116 ab	3 b	113 ab	97 a
paraquat	27.6	114 ab	4 b	110 ab	97 ab
glyphosate+ paraquat	24+27.6	117 ab	2 b	116 ab	98 a
glyphosate+ paraquat	12+20.7	113 ab	8 b	105 b	93 b
MSMA	72	114 ab	3 b	112 ab	98 a
quizalofop-p-ethyl	7.5	109 b	4 b	105 b	97 ab
MSMA+quizalofop-p-ethyl	36+3.75	119 ab	4 b	114 ab	96 ab
panicle topping off at flowering stage	-	-	-	-	-
untreated check		127 a	110 a	17 c	14 c
CV (%)		8.34	33.9	9.35	3.57

Table 2. Effect of herbicides on promoting unfilled seeds/panicle of weedy rice in Dermbang Nangbuat district, Supanburi province. Herbicides were wiped on flag leaves and panicles of weedy rice at three days after flowering stage. Twenty panicles of weedy rice were sampled in each replicate.

Treatment	Concentration (g ai l^{-1})	Weedy rice seeds/panicle			
		Total	Filled	Unfilled	% unfilled
glufosinate-NH_4	7.5	124 bc	17 b	107 cd	86 f
glufosinate-NH_4	15	122 bc	9 cde	113 abcd	92 bcde
glufosinate-NH_4	30	119 bc	3 e	117 abc	98 a
glyphosate	24	112 c	4 de	109 bcd	97 abc
paraquat	27.6	120 bc	14 bc	106 d	89 ef
glyphosate+ paraquat	24+27.6	126 b	10 bcd	115 abcd	92 cde
glyphosate+ paraquat	12+20.7	130 ab	9 cde	121 a	93 abcde
MSMA	72	122 bc	4 de	119 ab	97 ab
quizalofop-p-ethyl	7.5	128 b	13 bc	113 abcd	90 def
MSMA+quizalofop-p-ethyl	36+3.75	119 bc	7 cde	112 abcd	95 abcd
panicle topping off at flowering stage	-	-	-	-	-
untreated check	-	142 a	127 a	14 e	10 g
CV (%)		6.82	26.13	7.15	4.21

Discussion

Removing weedy plants at early stage is quite difficult as they mimic the rice crop. Farmers could distinguish the differences at flowering stage as weedy rice flower two weeks earlier and mostly higher than crop. Cutting weedy rice panicle with sickles is a popular method in Thailand, to control weedy rice. In general, the cost of labor to remove weedy rice panicles is approximately 90 US$ ha^{-1}. To practice this weed wiper technique, the cost of single application of herbicide was about 6-12 US$ ha^{-1}. If density of weedy rice is less than 20%, a farmer using weed wiper, could finish one hectare field in a day, while on week is required for the panicle topping off using manual labor.

Table 3. Number of undamaged panicles of weedy rice and rice yield at harvest.

Treatment	Concentration (g ai l^{-1})	Panicles m^{-2}		Rice yield (t ha^{-1})	
		Exp 1	Exp 2	Exp 1	Exp2
glufosinate-NH_4	7.5	77 c	71 de	2.8 cd	4.09 ab
glufosinate-NH_4	15	73 c	69 de	3.6 ab	3.85 abc
glufosinate-NH_4	30	81 c	60 e	4.0 a	2.91 d
glyphosate	24	92 c	70 de	2.3 ef	2.25 e
paraquat	27.6	79 c	76 de	1.9 f	2.27 e
glyphosate+ paraquat	24+27.6	73 c	56 e	1.1 g	2.98 d
glyphosate+ paraquat	12+20.7	79 c	71 de	2.0 ef	3.37 cd
MSMA	72	90 c	89 cd	2.5 de	3.50 dcd
quizalofop-p-ethyl	7.5	83 c	106 c	2.2 ef	3.37 cd
MSMA+quizalofop-p-ethyl	36+3.75	90 c	91 cd	3.3 bc	3.95 abc
panicle topping off at flowering stage	-	157 b	129 b	3.2 bc	4.47 a
untreated check	-	252 a	193 a	1.1 g	1.20 f
CV (%)		13.77	17.44	12.85	13.61

Experiment 1, weedy rice panicles and flag leaves were wiped with herbicides at flowering stage.
Experiment 2, weedy rice panicles and flag leaves were wiped with herbicides at 3 days after flowering stage.

When herbicides were carefully wiped on the panicles and flag leaves of weedy rice plants, most seeds were sterilized. If the farmers are careless allowing systemic herbicides such as glyphosate touch the edge of rice leaves, weed wiper technique will produce adverse effects. Some contact herbicides such as paraquat also showed a similar disadvantage when applied as weed wiper. Unapplied spikelets developed filled grains at a rate of 4-14 seeds per panicle after paraquat application (Table 2). Glufosinate-ammonium is semi-translocated in plants resulting in a high percentage of unfilled seeds and safe to crop even if it touches some leaves of the rice crop. Similarly, MSMA and quizalofop-p-ethyl also gave a good control of weedy rice without a significant crop injury.

After wiping with herbicides, normal panicles were present at harvest, amounting to about 30-40% of the untreated check (Table 3). When the density of weedy rice is as high as those of the two experiments conducted, a second application may be required to reduce the seed rains from the remaining. Although rice yields were recovered by more than 50%, the control method during the early growth stage was necessary. Pre-sowing and after-sowing treatments with herbicides are also recommended in the fields with very high infestation of weedy rice (Maneechote *et al.* 2005a; 2006). However, eradication of weedy rice prior to sowing clean seeds and cleaning-up of the combine harvesting machines are crucial steps to prevent the rapid spread of weedy rice.

Farmers in Thailand have recently adopted using glufosinate-ammonium, MSMA and quizalofop-p-ethyl in the weed wiper technique. Use of these herbicides in the weed wiper technique was cheaper and more effective than panicle topping off manually. More importantly, this innovative technique effectively reduced the larger numbers of weedy rice seed bank in the subsequent season.

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ALTERNATIVE HERBICIDES FOR THE CONTROL OF A POPULATION OF SPRANGLETOP [*Leptochloa chinensis* (L.) Nees] RESISTANT TO ACCASE-INHIBITING HERBICIDES

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Abstract: A population of sprangletop (*Leptochloa chinenesis* L. Nees) in Supanburi province, Thailand has reported to have developed resistance to Accase-inhibiting herbicides. Laboratory study showed that this population was moderately-resistant to fenoxaprop-p-ethyl and cyhalofop-butyl while it showed a low level of resistance to quizalofop-p-tefuryl and profoxydim. The resistant population exhibited 10-, 11-, 4- and 2-fold resistance to fenoxaprop-p-ethyl, cyhalofop-butyl, quizalofop-p-tefuryl and profoxydim, respectively. A field experiment was conducted in Supanburi province to verify whether different modes of actions of herbicides could control this population. Herbicides with six modes of action *i.e*. HRAC Group A, C2, E, F1, F3 and K3 were used in the experiment. The result showed that the selected population was resistant to all ACCase-inhibiting herbicides (HRAC Group A). Density of sprangletop in untreated plots at 30 days after sowing rice was 1,097 plants m^{-2}. The results indicated that the target population was completely controlled by butachlor, clomazone, diflufenican, dimethenamid, flufenacet, oxadiargyl, oxadiazon and propanil at the rates of 1000, 270, 100, 281.3, 225, 250, 625 and 2000 g ai ha^{-1}, respectively. Rotation of different modes of action should be practiced to prevent herbicide resistance.

Key words: Alternative mode of action, herbicide resistance, sprangletop

Introduction

Sprangletop (*Leptochloa chinensis* L. Nees) is a serious grass weed in direct-seeded rice in Thailand. During the last six years, ACCase-inhibiting herbicides were commonly used to control this grass species in the rice growing areas of the central plains of the country. Fenoxaprop-p-ethyl, cyhalofop-butyl, quizalofop-p-tefuryl and profoxydim are registered in Thailand to be use as grass weed killers in rice. In 2005, the first incidence of cross resistance in sprangletop was documented (Maneechote *et al.* 2005). To date, there are many complaints from farmers that ACCase-inhibiting herbicides could not control this grass species. The objective of this study was to demonstrate that alternative herbicides with different modes of action could control sprangletop populations that are cross-resistance to ACCase-inhibitors.

Materials and Methods

Plant Materials
A population of sprangletop (*Leptochloa chinensis* L. Nees) in Sri Prachan District, Supanburi Province, Thailand, where the suspected to be resistant to fenoxaprop-p-ethyl, based on farmer's observations made in 2006, was selected as the test material (sprangletop-R). This population had been exposed to butachlor/propanil with no use of fenoxaprop-p-ethyl or any other ACCase-inhibiting herbicides. A susceptible population from Samko District, Angthong province known to be susceptible to all ACCase-inhibiting herbicides (S) was used as a control in the laboratory study.

Verification of resistance to ACCase-inhibiting herbicides

All four ACCase herbicides registered for controlling sprangletop in Thailand (fenoxaprop-p-ethy, cyhalofop-butyl, quizalofop-p-tefuryl and profoxydim) were tested on the sprangletop-R and sprangletop-S populations to access resistance level. A total of 500 seeds collected from the R and S populations were sown separately on 25 ml of 0.5% (w/v) agar-solidified water in 200 ml plastic containers (5 cm dia). Several concentrations of those herbicides had been added to the solidified water agar. At each herbicide concentration, five replicate containers were used. These sealed containers were kept under fluorescent light in the laboratory. Seven days after treatment, seedlings with green, healthy shoots extended from coleoptiles were counted as survivors. The LD_{50} (dose giving 50% mortality) values of both R and S populations were calculated from the graph data.

Verification of herbicides with different modes of action

Field experiment was conducted in Sri Prachan District during January-April 2007 to confirm resistance to all four ACCase-inhibiting herbicides and tested for alternative modes of action of herbicides for controlling this population. Treatments consisted of fenoxaporp-p-ethyl, cyhalofop-butyl, quizalofop-p-tefuryl, profoxydim, oxadiargyl, oxadiazon, flufenacet, diflufenican, dimethenamid, pretilachlor, butachlor, clomazone and propanil. Rate and time of application of each herbicide was shown in Table 1.

Table 1. Formulations, rates, and time of application of herbicides with different modes of action for controlling a population of sprangletop resistant to ACCase-inhibiting herbicides in Supanburi province.

Treatment	Rate (g ai/ha)	Time of application*	HRAC group	Modes of action
Fenoxaprop 6.9% EC	150	7 DAS	A	ACCase inhibitors
Cyhalofop-butyl 10%EC	240	7 DAS	A	ACCase inhibitors
Profoxydim 7.5% EC	218.75	7 DAS	A	ACCase inhibitors
Quizalofop-p-tefuryl 4% EC	50	7 DAS	A	ACCase inhibitors
Oxadiargyl 40% SC	125	4 DAS	E	Inhibition of protopor-phyrinogen (PPO)
Oxadiargyl 40% SC	250	4 DBS	E	Inhibition of protopor-phyrinogen (PPO)
Oxadiazon 25% EC	625	4 DAS	E	Inhibition of protopor-phyrinogen (PPO)
Flufenazet 60% WP	112.5	4 DBS	K3	Inhibition of cell division
Flufenazet 60% WP	225	4 DBS	K3	Inhibition of cell division
Diflufenican 50% WP	100	4 DAS	F1	Inhibition of carotenoid synthesis at phytoene desaturase step (PDS)
Dimethenamid 90% EC	281.25	4 DBS	K3	Inhibition of cell division
Pretilachlor 30% EC	750	after sowing	K3	Inhibition of cell division
Butachlor 60% EC	1,000	4 DAS	K3	Inhibition of cell division
Clomazone 48% EC	270	4 DAS	F3	Inhibition of carotenoid synthesis (unknown target)
Propanil 36% EC	2,000	7 DAS	C2	Inhibition of photosynthesis at photosystem II

* DAS – days after sowing; DBS – days before sowing

Treatments were arranged in a Randomized Complete Block Design (RCBD) with four replicates. Plot size was 4 x 5 m² with 0.5 m guard rows. Two weeks after treatment, visual estimates of crop injury were recorded using a scale of 0 to 10 (0 = no injury, 1 to 3 = slightly toxic, 4 to 6 = moderately toxic, 7 to 9 = severely toxic, 10 = completely killed). The number of rice and weeds in an area of 1 m^2 of each replicate was also counted. Data were analyzed by ANOVA, and LSD (p = 0.05) was used for mean comparison.

Results

Verification of resistance to ACCase-inhibiting herbicides

The laboratory experiment indicated that R population exhibited a moderate level of resistance to fenoxaprop-p-ethyl and cyhalofop-butyl and a low level of resistance to quizalofop-p-tefuryl and profoxydim. The LD_{50} values of R population were 10, 11, 4 and 2 times higher than S population, in the presence of fenoxaprop-p-ethyl, cyhalofop-butyl, quizalofop-p-tefuryl and profoxydim, respectively (Table 2). This R population has developed cross-resistance to all four ACCase-inhibiting herbicides.

Table 2. LD_{50} values of resistant (R) and susceptible (S) sprangletop grown in agar containing ACCase-inhibiting herbicides. Ratios are the value for resistant (R) divided by the value of susceptible (S).

Herbicide	LD_{50} (mg a.i. l^{-1})		R/S ratio
	R	S	
Aryloxyphenoxypropionates			
Fenoxaprop-p-ethyl	1.24 ± 0.22	0.13 ± 0.02	10
Cyhalofop-butyl	1.64 ± 0.31	0.14 ± 0.02	11
Quizalofop-p-tefuryl	0.26 ± 0.04	0.07 ± 0.01	4
Cyclohexanediones			
Profoxydim	0.22 ± 0.02	0.13 ± 0.02	2

[a]Abbreviations: LD_{50}, dose giving 50% mortality, ACCase, acetyl coenzyme A carboxylase

Verification of herbicides with different modes of action

The ACCase-inhibiting herbicides use at twice the recommended rate, were slight to moderately toxic to rice plants (Table 3).

Table 3. Efficacy of different modes of action of herbicides on a resistant population of sprangletop and rice yield at harvest

Treatment	Rate (g ai ha^{-1})	Herbicide Efficiency	Phytotoxicity to rice	Rice yield (t/ha)
Fenoxaprop-p-ethyl	150	0.0	5.5	0.4 fg
Cyhalofop-butyl	240	0.0	1.0	1.6 d
Profoxydim	218.8	0.0	2.0	1.0 e
Quizalofop-p-tefuryl	50	0.0	8.5	0.1 g
Oxadiargyl	125	9.0	1.0	2.6 bc
Oxadiargyl	250	9.6	1.5	2.9 ab
Oxadiazon	625	9.8	1.0	2.8 ab
Flufenazet	112.5	8.5	1.0	2.1 cd
Flufenazet	225	9.5	1.0	2.8 ab
Diflufenican	100	9.0	1.5	2.7 ab
Dimethenamid	281.3	9.5	2.0.	2.9 ab
Pretilachlor	750	9.5	0.5	3.0 ab
Butachlor	1000	9.8	0.5	2.2 c
Clomazone	270	9.8	2.5	2.5 bc
Propanil	2000	9.8	1.0	3.2 ab
UTC	-	0.0	0.0	0.7 ef
CV (%)				17.4

In contrast, the resistant sprangletop plants were not affected. All other herbicides were slightly toxic to rice plants at 14 days after treatment however the rice plants showed no injury symptoms at 30 days after application. The density of R population was 1,097 plants per m².

The sprangletop-R plants were mostly killed by herbicides with different modes of action to that of HRAC group A herbicides (Table 4). Flufenacet killed the sprangletop-R population and allowed gooseweed (*Sphenocloea zeylanica*, SPHZE) to emerge. In addition, clomazone gave an excellent control of srangletop but poor control of SPHZE and smallflower umbrella sedge *Cyperus difformis* (CYPDI). Complete control of the sprangletop-R population was obtained when treated with alternative herbicides such as oxadiargyl, oxadiazon, flufenacet, dimethenamid, pretilachlor and butachlor at the rates of 250, 625, 225, 281.3, 750 and 1,000 g ai ha^{-1}, respectively. Propanil gave a good control of this population with about 37 sprangletop plants m^{-2} surviving in the treated area.

Table 4**.** Number of plants/m^2 and dry weight of rice and weeds at 30 days after sowing rice.

Treatment	Rate	Rice	LEPCH		SPHZE		CYPDI	
	g ai ha^{-1}	No./m^2	No./m^2	DW (g)	No./m^2	DW (g)	No./m^2	DW (g)
Fenoxaprop	150	142 e	776 b	173 a	18 b	0.6 bc	44 c	3.0 b
Cyhalofop-butyl	240	256 a	236 c	62 b	7 cd	3.3 a	29 cd	1.7 bc
Profoxydim	218.8	97 f	296 c	77 b	38 a	0.2 bc	87 b	7.4 a
Quizalofop-p-tefuryl	50	11 g	1007 ab	210 a	15 bc	0.9 b	32 c	2.0 bc
Oxadiargyl	125	213 bcd	1 c	1 c	0 d	0 c	0 e	0.0 c
Oxadiargyl	250	206 cd	0 c	0 c	0 d	0 c	0 e	0.0 c
Oxadiazon	625	215 bcd	0 c	0 c	0 d	0 bc	0 e	0.0 c
Flufenazet	112.5	239 abc	10 c	2 c	14 bc	0.3 c	0 e	0.0 c
Flufenazet	225	263 a	0 c	0 c	0 d	0 c	0 e	0.0 c
Diflufenican	100	246 abc	54 c	13 c	0 d	0 c	0 e	0.0 c
Dimethenamid	281.3	208 abcd	0 c	0 c	0 d	0 c	0 e	0.0 c
Pretilachlor	750	224 abcd	0 c	0 c	0 d	0 c	0 e	0.0 c
Butachlor	1000	226 abcd	4 c	1 c	2 d	0 c	0 e	0.0 c
Clomazone	270	247 ab	0 c	0 c	13 bc	0.2 bc	122 a	7.4 a
Propanil	2000	240 abc	37 c	8 c	0 d	0 c	2 de	0.1 c
UTC		179 d	1097 a	202 a	0 d	0 c	0 e	0.0 c
CV (%)		13.8	102.1	63.3	102.1	165.9	95.7	113.9

Discussion

In Thailand, ACCase-inhibiting herbicides are popular among farmers as they are more convenient than other groups with different modes of action that are applied as pre-emergent or early post-emergent herbicides. During the past six years, the first report of a sprangletop BLC 1 population resistant to ACCase-inhibitng herbicides was documented in Bangkok (Maneechote *et al*. 2005). To date, many populations of sprangletop in rice growing areas in the Central Plain were suspected to have developed resistance to Group A herbicides.

To control this resistant population, herbicides with alternative modes of action are required. It was confirmed that herbicides with different modes of action to that of ACCase inhibitors, gave an excellent control of the tested sprangletop-R population. This study was a demonstration for farmers to learn that herbicide resistance problem could be easily solved by changing into herbicides with different modes of action. However, multiple resistance has rapidly developed in grass weeds such as annual ryegrass, in Australia (Owen *et al.* 2007). Hence, not all resistant sprangletop populations found in the Central Plains will be effectively controlled by those herbicides. Rotation of herbicides with different modes of action is an effective method for farmers to prevent herbicide resistance.

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INTEGRATED WEED MANAGEMENT IN WHEAT AT HIGHER ELEVATIONS

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Abstract: A field trail was conducted at the Agriculture Research Station, Chitral, NWFP-Pakistan during the wheat season of 2004-05, using a split plot design. The seed rates (Factor A) assigned to main plots were 100, 125 and 150 kg ha^{-1} and the herbicides (Factor B) assigned to subplots were Buctril super® 60 EC (Bromoxynil+MCPA) at 0.45, Topik® 15 WP (clodinafop-propargyl) at 0.04, Puma super® 75 EW (fenoxaprop-p-ethyl) at 0.75 and Isoproturon® 50 WP (isoproturon) at 1.0 kg a.i. ha^{-1}, keeping a weedy check for comparison. The effect of all these treatments was studied on weed control efficiency (%), fresh weed biomass (kg ha^{-1}), number of tillers m^{-2}, 1000-grain weight (g), biological yield (kg ha^{-1}) and grain yield (kg ha^{-1}). Statistically maximum weed control efficiency (79.0%) and minimum weed biomass (1086 kg ha^{-1}) was observed in Buctril super® 60 EC followed by Isoproturon® 50 WP (77.1 %) and (1157 kg ha^{-1}) as compared to the biomass in the weedy control (3770 kg ha^{-1}). Similarly, number of tillers (289 m^{-2}), 1000-grain weight (41.0 g), biological yield (9333 kg ha^{-1}) and grain yield (2504 kg ha^{-1}) were the maximum in Buctril super® 60 EC followed by Isoproturon® 50 WP when compared to the weedy control. Therefore, the herbicide Buctril super® 60 EC at 0.45 kg and Isoproturon® 50 WP at 1 kg a.i. ha^{-1} were the best for weed control in wheat. While among the seed rates, 125 kg ha^{-1} gave the best performance.

Key words: Wheat, *Triticum aestivum,* seed rates, densities, chemicals, herbicides, weeds

Introduction

Wheat ranks first among the cereal crops in Pakistan and occupies about 66% of the annual food cropped area (Anonymous, 1996). It has always been a subject of extensive research. The green revolution of 1960s brought a major breakthrough in wheat production of the country as production almost doubled with the extensive cultivation of short-stature, fertilizer responsive and high yielding varieties. Increased cropping intensities have made the fertilizer use indispensable. Wheat is the major source of food and is called "the king of cereals". During 2004-05, the area at the national level under wheat cultivation was 8.3580 million ha, with a production of 21.612 million tons. The area consisted of about 7.2206 million ha irrigated and 1.1374 million ha of un-irrigated land. At provincial level, in NWFP, the area under wheat cultivation was about 0.7486 million ha having 0.3133 m ha area as irrigated and 0.4353 mill. ha as rain-fed, giving a total production of 1.0911 m tons at 1458 kg ha^{-1} (MINFAL, 2005). Although more importance is given to the research-work in plain areas due to the large acreage of land available for cultivation, yet the importance of limited available land in the mountainous areas can not be ignored. Farmers in high altitude areas most often grow wheat, although they do not get yields as high as in the plains. The low yield per hectare beside many other factors could be attributed to serious weed infestation in the crop.

Weed losses are up to 30% in wheat production (Khan and Noor, 1995). Many studies have shown significant differences in competitive ability of wheat varieties against a range of weed species (Appleby *et al.* 1976; Balyan *et al.* 1991; Verschwele and Niemann, 1993; Blackshaw, 1994; Christensen, 1994). Of particular concern is the fact that modern semi-dwarf wheat cultivars are less competitive than the older standard wheats (Lemerle *et al.* 1996), making them more dependent on herbicides than the older types. The extent and reasons for the inter-varietal variation in competitive ability of wheat were recently reviewed by Lemerle *et al.* (2001).

Weeds reduce crop yields, deteriorate quality of farm produce and hence the decrease market value of wheat. Chemical control method is being emphasized in modern agriculture

(Taj *et al.* 1986). The manufacturers' recommended field doses of herbicides are selected to give reliable weed control without crop damage. Under optimum weather and soil conditions, effective weed control may be obtained with doses of herbicide below that recommended by the manufacturer (Caseley, 1990). For the crop:weed:herbicide combinations investigated there was little increase in crop yield for herbicide dose rates above 20% of recommended field rates. There is also increasing pressure on the farmer to reduce herbicide use for both economic and environmental reasons (Brain *et al.*, 1999). Annual losses in wheat amount to more than 28 billions at national level and Rs. 2 billions in NWFP (Hassan and Marwat, 2001). In Chitral, wheat crop is severely infested by *Galium aparine*, *Convolvulus arvensis*, *Fumaria indica*, *Rumex* sp., *Stellaria media*, *Medicago* sp., *Mellilotus* sp., *Coronopus didymus*, *Chenopodium* sp., *Euphorbia* sp., *Lolium* sp., *Avena fatua*, and *Anagallis arvensis* as reported by the farming community in the survey of the project area during 2004-05. The experiment was conducted with the objectives to select the best herbicide for weed control and to study the effect of different seed rates as well, thus to develop an integrated weed management strategy in wheat for the area concerned.

Materials and Methods

A field trial entitled "Integrated weed management in wheat at higher elevations" was conducted at Agricultural Research Station, Chitral during November-June 2004-05. The experiment was laid out in Randomized Complete Block Design with a split plot arrangement, having three replications. Three seed rates (Factor A) including 100, 125 and 150 kg ha^{-1} were assigned to the main plots and four post-emergence herbicides (Factor B) including Buctril super® 60 EC (bromoxynil+MCPA), Topik® 15 WP (clodinafop-propargyl), Puma super® 75 EW (fenoxaprop-p-ethyl), and Isoproturon® 50 WP (isoproturon) and a weedy check were assigned to the subplots. The dosages and the description of treatments imposed are given in Table 1. The size of each treatment was kept at 5 m x 1.8 m consisting of 6 rows each 30 cm apart with a row length of 5 m. The data on weed control efficiency (%), fresh weed biomass (kg ha^{-1}), number of tillers m^{-2}, 1000-grain weight (g), biological yield (kg ha^{-1}), and grain yield (kg ha^{-1}) were recorded.

Table 1. Treatments used in the wheat experiment of Chitral (2004-05)

Treatment No.	Seed rate (kg ha^{-1})	Herbicides (Trade names)	Active ingredients	Dose (kg a.i. ha^{-1})
1.	100	Topik® 15 WP	clodinafop-propargyl	0.04
2.	125			
3.	150			
4.	100	Isoproturon® 50 WP	isoproturon	1.00
5.	125			
6.	150			
7.	100	Puma super® 75EW	fenoxaprop-p-ethyl	0.75
8.	125			
9.	150			
10.	100	Buctril super® 60 EC	bromoxynil + MCPA	0.45
11.	125			
12.	150			
13.	100	Weedy check	---	---
14.	125			
15.	150			

The weed control efficiency was determined from weed density data before and after the herbicide application by the following formula;

$$\text{Weed control efficiency (\%)} = \frac{(\text{weed density m}^{-2}\text{ before - weed density m}^{-2}\text{ after}) \times 100}{\text{weed density m}^{-2}\text{ before herbicide application}}$$

The data recorded for each parameter were individually subjected to the ANOVA technique by using MSTATC and means were separated by using the LSD test (Steel and Torrie, 1980).

Results and Discussion

Weed control efficiency (%)

The weeds that were dominant in the experiment were *Galium aparine*, *Convolvulus arvensis, Fumaria indica, Rumex* sp., *Stellaria media, Medicago* sp., *Mellilotus* sp., *Coronopus didymus, Chenopodium* sp., *Euphorbia* sp., *Lolium* sp., *Avena fatua*, and *Anagallis arvensis*. Statistical analysis of the data showed that differences among herbicides and seed rates were significant for weed control efficiency (Table 2). It is evident from the data in Table 1 that maximum weed control efficiency (79.0 %) was recorded in Buctril super® 60 EC treatments as broadleaved weeds were more prevalent. Among the seed rates the highest weed control efficiency (54.4 %) was recorded at a seed rate of 125 kg ha^{-1}. In the seed rate 100 kg ha^{-1}, the larger spacing may have provided room for the weeds to flourish after the herbicide application. In the seed rate 150 kg ha^{-1}, there may be intra-specific competition among the wheat plants due to which weeds flourished even after the herbicide application. For the interaction of seed rate with the herbicides, maximum weed control efficiency (82.8 %) was recorded at the seed rate 125 kg ha^{-1} treated with Buctril super® 60 EC. Analogous results were reported by Khan *et al*. (2002) that herbicides application effectively controls weeds. These findings are also in conformity with those of Shahid (1994), who reported that herbicides significantly reduce weed density.

Table 2. Weed control efficiency (%) as affected by wheat seed rate and herbicides

Herbicides	Wheat seed rate			Herbicide (mean)
	100 kg ha^{-1}	125 kg ha^{-1}	150 kg ha^{-1}	
Buctril super® 60 EC	73.8	82.8	80.3	79.0 a
Topik® 15 WP	57.9	62.5	60.5	60.3 b
Puma super® 75EW	44.7	51.0	48.3	48.0 c
Isoproturon® 50 WP	71.5	81.4	78.4	77.1 a
Weedy check	0.0	0.0	0.0	0.0
Seed rate (mean)	49.5 b	54.4 a	53.5 a	

LSD (p=0.05) for herbicides = 15.1; *Means followed by the same letter in the respective categories are not differ significantly by the Least Significant Difference Test (p=0.05).

Fresh weed biomass (kg ha^{-1})

Data indicated that differences among the seed rates and herbicides were significant for fresh weed biomass. It is clear from the data in Table 3 that minimum weed biomass (1086 kg ha^{-1}) was recorded in Buctril super® 60 EC which is statistically similar to Isoproturon® 50 WP having a weed biomass of 1157 kg ha^{-1} due to the fact that there was a pronounced problem of broad leaved weeds which were effectively controlled resulting in decrease of the biomass. Maximum biomass (3770 kg ha^{-1}) was recorded in the weedy check. Among the seed rates, the highest weed biomass (2121 kg ha^{-1}) was recorded in the seed rate 100 kg ha^{-1} because the larger spacing may have provided room for the weeds to flourish even after herbicide application. For the interaction of seed rates with the herbicides, minimum weed biomass (941 kg ha^{-1}) was recorded in the seed rate 125 kg ha^{-1} treated with Buctril super 60EC and maximum weed biomass (4006 kg ha^{-1}) was recorded in the 100 kg ha^{-1} seed rate in weedy check. Similar results were reported by Khan *et al*. (2002) that herbicide application decreases fresh weed biomass. Shahid (1994) reported that herbicides significantly reduce weed biomass m^{-2}.

Table 3.Weed biomass (kg ha^{-1}) as affected by wheat seed rate and herbicides

Herbicides	Wheat seed rate			Herbicide (mean)
	100 kg ha^{-1}	125 kg ha^{-1}	150 kg ha^{-1}	
Buctril super® 60 EC	1225	941	1091	1086 c
Topik® 15 WP	1918	1833	1900	1884 bc
Puma super® 75EW	2200	2061	2091	2118 b
Isoproturon® 50 WP	1258	1036	1175	1157 c
Weedy check	4006	3776	3526	3770 a
Seed rate (mean)	2121 c	1930 a	1957 a	

LSD (p=0.05) for herbicides = 624; * Means followed by the same letter in the respective categories are not differ significantly by the Least Significant Difference Test (p=0.05).

Number of tillers m^{-2}

Analysis of the data exhibited that the number of tillers m^{-2} was significantly affected by different herbicidal treatments while the seed rates and interaction of herbicides with the seed rates were non-significant statistically. The data in Table 3 revealed that maximum (289) tillers m^{-2} were recorded in Buctril super® 60 EC followed by Isoproturon® 50 WP which produced 279 tillers m^{-2} as more weed control triggered the number of tillers to increase. The minimum (173) tillers m^{-2} was recorded in the weedy check because of dominance of the weeds. Among the seed rates, the highest number of tillers (248 m^{-2}) was recorded in the seed rate 125 kg ha^{-1}.

The number of tillers in seed rate 150 kg ha^{-1} was less because of intra-specific competition. For the interaction of seed rates with the herbicides, the differences although were non-significant statistically, yet the maximum tillers (300) were recorded in seed rate 125 kg ha^{-1} treated with Buctril super® 60 EC herbicide as there was good weed control along with optimum seed rate. The minimum number of tillers m^{-2} (165) was recorded in 100 kg ha^{-1} seed rate in the weedy check (Table 3). Tunio *et al.* (2004) reported similar results. They reported that application of herbicides significantly influenced the number of tillers m^{-2}.

Table 3. Number of tillers m^{-2} as affected by wheat seed rate and herbicides

Herbicides	Wheat seed rate			Herbicide (mean)
	100 kg ha^{-1}	125 kg ha^{-1}	150 kg ha^{-1}	
Buctril super® 60 EC	279	300	288	289 a
Topik® 15 WP	244	255	248	249 b
Puma super® 75EW	215	221	218	218 b
Isoproturon® 50 WP	262	293	282	279 a
Weedy check	165	172	184	173 c
Seed rate (mean)	233	248	244	

LSD (p=0.05) for herbicides = 46; * Means followed by the same letter in the respective categories are not differ significantly by the Least Significant Difference Test (p=0.05).

1000-grain weight (g)

ANOVA of the data revealed that 1000-grain weight was significantly affected by different herbicidal treatments while seed rates and interaction of seed rates with herbicides were non-significant statistically. The data in Table 4 revealed that maximum (41.0 g) 1000 grain weight was recorded in Buctril super® 60 EC and minimum (37.2 g) was recorded in weedy check. Although statistically non-significant, the highest numerical value of 1000 grain weight (40 g) was recorded in the seed rate 125 kg ha^{-1}. The interaction of herbicides with seed rates was non-significant statistically. The maximum 1000 grain weight (44.2 g) was recorded in 125 kg ha^{-1} seed rate treated with Buctril super® 60 EC herbicide. Best weed control channeled the flow of nutrients to the wheat grains increasing their size and weight. The minimum (37.2 g) was recorded in the seed rate 125 kg ha^{-1} under weedy control (Table 4). The seed rate 125 kg ha^{-1} seems to be optimum because in seed rate 100 kg ha^{-1} there is enough space for weeds to invade even after herbicide application while in seed rate 150 kg ha^{-1} there is intra-specific competition among wheat plants. As 1000 grain weight is a very important yield component in every crop, increase in 1000 grain weight will automatically increase the total grain yield of the crop. Hassan *et al*. (2003) reported that herbicides application increased 1000 grain weight significantly as compared to weedy check.

Table 4. 1000-grain weight (g) as affected by wheat seed rate and herbicides.

Herbicides	Wheat seed rate			Herbicide (mean)
	100 kg ha^{-1}	125 kg ha^{-1}	150 kg ha^{-1}	
Buctril super® 60 EC	38.6	44.2	40.2	41.0 a
Topik® 15 WP	38.2	38.4	38.3	38.3 b
Puma super® 75EW	37.9	38.1	38.0	38.0 b
Isoproturon® 50 WP	37.7	42.3	39.5	39.8 a
Weedy check	37.1	37.2	37.3	37.2 b
Seed rate (mean)	37.9	40.0	38.6	

LSD (p=0.05) for herbicides = 1.7; * Means followed by the same letter in the respective categories are not differ significantly by the Least Significant Difference Test (p=0.05).

Biological yield (kg ha^{-1})

Differences among the herbicides were significant while among the seed rates and interaction of herbicides with seed rates were statistically non-significant. It is evident from Table 5 that maximum (9333 kg ha^{-1}) biological yield was recorded in Buctril super® 60 EC treatment as more weed control improved the biological yield. Minimum (5811 kg ha^{-1}) biological yield was observed in weedy check. Among the seed rates the highest biological yield 8146 was recorded in 125 kg ha^{-1} (Table 5). The biological yield in seed rate 150 kg ha^{-1} was lower due to intra-specific competition among wheat plants. The interaction of herbicides with seed rates though statistically non-significant, revealed the maximum biological yield (10333) in seed rate 125 kg ha^{-1} treated with Buctril super® 60 EC herbicide. The minimum biological yield (5333) was recorded in 100 kg ha^{-1} seed rate under weedy check. The seed rate 125 kg ha^{-1} seems to be optimum because in seed rate 100 kg ha^{-1} there is enough room for weeds to establish even after herbicide application while in seed rate 150 kg ha^{-1} there seem to be intra-specific competition among the crop plants. The highest biological yield is attributable to the best weed control. Hassan *et al.* (2003) reported analogous results.

Grain yield (kg ha^{-1})

Grain yield was significantly affected by different herbicides and seed rates, while the interaction of seed rates with herbicides were non-significant statistically. The data regarding grain yield of wheat in Table 6 showed that maximum (2504 kg ha^{-1}) grain yield was recorded in Buctril super® 60 EC treatments as more weed control triggered the grain yield to go up .

Minimum (1406 kg ha^{-1}) grain yield was recorded in the weedy check. Among the seed rates, the maximum grain yield (2141 kg ha^{-1}) was recorded in the seed rate 125 kg ha^{-1} (Table 6).

Table 5. Biological yield (kg ha^{-1}) as affected by wheat seed rate and herbicides.

Herbicides	Wheat seed rates			Herbicide means
	100 kg ha^{-1}	125 kg ha^{-1}	150 kg ha^{-1}	
Buctril super® 60 EC	8333	10333	9333	9333 a
Topik® 15 WP	7200	7666	7333	7399 ab
Puma super® 75EW	6633	7100	7000	6911 b
Isoproturon® 50 WP	8000	9633	8833	8822 a
Weedy check	5333	6000	6100	5811 bc
Seed rate means	7100	8146	7720	

LSD (p=0.05) for herbicides = 811; * Means followed by the same letter in the respective categories are not differ significantly by the Least Significant Difference Test (p=0.05).

Table 6. Grain yield (kg ha^{-1}) as affected by wheat seed rate and herbicides.

Herbicides	Wheat seed rate			Herbicide means
	100 kg ha^{-1}	125 kg ha^{-1}	150 kg ha^{-1}	
Buctril super® 60 EC	2206	2846	2460	2504 a
Topik®15 WP	1926	2066	2006	1999 b
Puma super® 75EW	1833	1900	1886	1873 b
Isoproturon® 50 WP	2100	2473	2300	2291 a
Weedy check	1340	1420	1460	1406 c
Seed rate means	1881 b	2141 a	2022 a	

LSD (p=0.05) for herbicides = 321; * Means followed by the same letter in the respective categories are not differ significantly by the Least Significant Difference Test (p=0.05).

The grain yield in seed rate 150 kg ha^{-1} was lower due to intra-specific competition among wheat plants. The interaction of herbicides with seed rates was non-significant statistically. The highest grain yield (2846) was recorded in 125 kg ha^{-1} seed rate treated with Buctril super® 60 EC herbicide. The lowest grain yield (1340) was recorded in 100 kg ha^{-1} seed rate in the weedy check. Increase in grain yield in the herbicide treated plots was probably due to the efficient weed control and thus the crop efficiently utilized all the available resources. These results are in conformity with those reported by Hassan *et al.* (2003). They reported that herbicidal treatments significantly increased grain yield in wheat. These findings are also similar to Tunio *et al.* (2004) who communicated likewise.

Conclusion

Buctril super® 60 EC (bromoxynil+MCPA) at 0.45 kg a.i. ha^{-1}, a broadleaved killer, proved to the best among the tested treatments for weed control, as there was a pronounced problem of broadleaved weeds in the experimental plots, followed by Isoproturon® 50 WP (isoproturon) at 1.0 kg a.i. ha^{-1}. Among the seed rates alone, the grain yield was the highest in the seed rate 125 kg ha^{-1}. The interaction of Buctril super® 60 EC at 0.45 kg ha^{-1} with the seed rate at 125 kg ha^{-1} produced excellent results in all the weed and crop parameters.

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AN EVALUATION OF WEED MANAGEMENT OPTIONS FOR COTTON-BASED FARMING SYSTEMS IN MUZARABANI

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Abstract: On-farm trials were carried out during the 2000-2001 and 2001-2002 seasons in order to test the efficacy of different weed management options. The treatments were grouped into two, (a) mechanical options for farmers who own implements and (b) non-mechanical options for those who do not own implements. Some of the treatment combinations included pre-emergence herbicides, alachlor (Lasso®) *and* cyanazine (Bladex®) tank mixed, applied overally or in a 30 cm band application along crop row in combination with ploughs, cultivators and hoes for inter-row weeding. The band-applied herbicides were found effective in reducing the weed density during the first six-eight weeks of crop growth thereby reducing early crop weed competition. The combination of implements and pre-emergent herbicides, alachlor and cyanazine tank mixed proved to be the best in reducing weed densities. These combinations proved to be important as the use of cultivators and ploughs resulted in furrows that formed ridges and furrows that allowed water retention. This is quite important in Muzarabani, which can receive drought spells during the season. As for non-mechanical options full application of herbicides proved to be the best in reducing weed numbers and labour.

Key words: Cotton, weed management, weeding options

Introduction

Excessive weed growth is a critical factor limiting smallholder crop production in Zimbabwe (Chivinge, 1984). Focus group discussions with farmers in Muzarabani district, Zambezi Valley, where 95% of households produce cotton, identified weed control as a major problem limiting yield, the total area cultivated and therefore productivity of the farming system (Chatizwa *et al.* 2000). The common and problematic weeds found in Muzarabani were *Boerria scabra* Schumach and Thom K Schum (button weed), *Ocimum canum* Sims (Wild basil), *Trichodemsa zeylanicum* (Burm.f.) R.Br., (late weed) *Eragrostis aspera* (Jacq) Nees (Rough love grass), *Corchorus olitorius* L., (Jute), *Vernonia poeskana* Vatke & Hilder., *Beorhivia erecta* L, *Sphaeranthus fleoxuosus* L., *Panicum maximum* Jacq (Guinea grass), *Ceratotheca sesamoides* Endl., *Celosia trigyna* L.,(Silver spinach) *and Urochloa paniocoides* Beauv. (Garden Urochloa) (Mavudzi *et al*. 2001a).

Many farmers are unable to control these weeds efficiently during the critical period, despite weeding 3-4 times per season. On-farm studies in the area have demonstrated significant yields gaps in both cotton and maize crops attributable to weed competition under farmer's current weed management practices (Mavudzi *et al*. 2001a). The findings from farmer group discussions; the yield loss study and other household characterisation work were used to design a series of researcher-managed and farmer-managed field trials focusing on alternative weed management options. The trials were undertaken with the aim of reducing labour bottlenecks at peak weeding time and reducing cotton yield losses due to weeds. The objective of the study is therefore to evaluate different weed management options that integrate the use of pre-emergent herbicide mixtures (alachlor + cyanazine) with hoe and mechanical weeding.

Materials and Methods

On farm experiments were conducted during 2000-2001 and 2001-2002 seasons in Mufudzi, Muringazuva and Gutsa villages in Muzarabani district, north East of Zimbabwe. Treatments indicated in Table 1 were allocated to farmers according to their resources.

Table 1. Treatments imposed.

Treatment Number and description		Treatment Code
	Non mechanical methods	
1	Overall hand hoeing at 3, 6, and 9 weeks after crop emergence (WACE).	HH
2	Pre-emergence application of cyanazine (550 g a.i. ha^{-1}) plus alachlor (960 g a.i. ha^{-1}) in 30 cm wide bands along the crop row at cotton planting, followed by inter-row weeding with a hand hoe at 3, 6 and 9 WACE.	Hca, HH
3	Hand hoe weeding of crop rows at 2 WACE followed by post-emergence application of cyanazine (550 g a.i. ha^{-1}) in the crop inter-rows at 3 WACE.	HH, Hc
4	Pre-emergence application of cyanazine (550 g a.i. ha^{-1}) plus alachlor (960 g a.i. ha^{-1}) over the whole plot area followed by hand weeding at 3, 6 and 9 WACE.	Hca
	Mechanical methods	
5	Hand hoe weeding at 2 WACE along the row followed by inter-row cultivation with an ox-drawn cultivator at 4 WACE.	HH,OC, HH
6	Inter-row cultivation using an ox-cultivator at 3 and 6 WACE accompanied by hoe weeding in the cotton row	OC,HH
7	Inter-row cultivation using an ox-plough with mouldboard for at 3, 6 WACE accompanied by hand hoe weeding in the cotton row.	OP+D,HH
8	Inter-row cultivation using an ox-plough without the mouldboard for at 3, 6 WACE accompanied by hand hoe weeding in the cotton row.	OP-D,HH
9	Pre-emergence application of cyanazine (550 g a.i. ha^{-1}) plus alachlor (960 g a.i. ha^{-1}) in 30 cm wide bands along the crop row at cotton planting and inter-row cultivation using an ox-plough with mouldboard at 3 and 6 WACE accompanied by hoe weeding in the cotton row	Hca, OP+D, HH
10	Pre-emergence application of cyanazine (550 g a.i. ha^{-1}) plus alachlor (960 g a.i. ha^{-1}) in 30 cm wide bands along the crop row at cotton planting and inter-row cultivation using an ox-plough without the mouldboard at 3 and 6 WACE accompanied by hoe weeding in the cotton row.	Hca, OP-D, HH
11	Pre-emergence application of cyanazine (550 g a.i. ha^{-1}) plus alachlor (960 g a.i. ha^{-1}) in 30 cm wide bands along the crop row at cotton planting and inter-row cultivation using an ox cultivator at 3 and 6 WACE accompanied by hoe weeding in the cotton row.	Hca, OC, HH

Participating farmers were identified by the community according to wealth categories (Muzenda and Ellis-Jones, 2000). The farmers with the supervision of the researchers planted the daughter trials and participated in weeding, harvesting and evaluation. The trials consisted of single strips of treatments with farms as replicates. The gross plot was 300m^2. Cotton cultivar Albar SZ 8714 was planted at all sites at a plant population of 33,333 plants $ha^{-1.}$ Compound L (5% N, 18% P, 10% K) fertiliser was applied at a rate of 150 kg ha^{-1} and ammonium nitrate (34.5% N) was applied at 6 WACE at a rate of 100 kg ha^{-1}. Weeds were counted at 3, 6 and 9 wace before any weeding operation at three random positions in each of the gross plots. Crop height was measured at first boll split. Cotton bolls were also counted at first boll split on each of the sampled plants. Analysis of treatment effects on cotton yield was done by Analysis of variance (ANOVA), using farms nested in villages as the blocking factor. Partial budgets were calculated for each weeding option based on average yields. Input and output values for both seasons have been based on market prices for 2001-2002 season.

Results

In 2000-2001, treatment had no significant effect ($p<0.05$) on weed numbers for the first 6 weeks after cotton emergence (Table 2) on non-mechanical weeding options. In the following year herbicide use, either pre-emergence or early post-emergence application resulted in significantly less weed density than hand weeding.

Table 2. Mean effect of weed control methods on total weed density (number m^{-2}) at nine on-farm sites in Muzarabani

Treatment code	2000-2001 season			2001-2002 Season		
	Weed density at 3 WACE	Weed density at 6 WACE	Weed density at 9 WACE	Weed density at 3 WACE	Weed density at 6 WACE	Weed density at 9 WACE
HH	6.56	7.16	10.53	8.41	8.54	7.48
Hcab + HH	25.91	6.04	9.90	3.90	6.62	6.86
HH + Hca	-[1]	-	-	11.62	5.48	9.64
Hca-full cover	5.05	6.41	8.00	5.30	5.67	6.01
Significance	Ns	NS	*	***	*	***
S.E.D (df.96)	0.657	0.666	1.558	1.806	1.178	1.049
HH +OC	10.38	3.91	9.78	10.71	9.80	10.64
OC+HH	13.78	3.36	9.95	10.85	9.24	10.29
P+D+HH	11.42	2.70	9.35	12.98	9.46	10.94
OP-D+HH	12.03	2.69	10.24	10.70	6.76	9.43
Hcab+OP+D+HH	8.77	2.06	7.56	2.48	8.26	11.01
Hcab+OP-D+HH	6.20	3.14	6.75	3.74	7.79	11.23
Hcab +OC+HH	6.51	3.75	6.48	3.26	9.31	9.52
Significance	***	Ns	*	***	Ns	Ns
S.E.D.	1.869	0.85	1.365	1.436	1.363	1.232

[1]- Treatment not applied. Figures in brackets are the original figures. ns not significant *** = $p<0.001$ * = $p<0.05$. WACE= weeks after crop emergence.

The results in 2000-2001 and 2001-2002 seasons indicated that there was a significant ($p<0.001$) effect of treatment on the weed density at 3 and 6 WACE, respectively. Combinations of pre-emergence herbicide applied in a band over the crop row with use of mechanical equipment (T9, T10 and T11 – see Table 1) had consistently lower weed densities than combination of the same equipment with hoe weeding (T5, T6, T7 and T8 – see Table 1). A similar trend was also observed in the 2001-2002 season (Table 2).

There was no significant ($p<0.05$) effect on weed control practices observed on yield in 2000-2001 and 2001-2002 seasons (Table 3). In 2000-2001 season a higher number of bolls per plant were recorded in the plot that received a pre-emergence herbicide banded plus hand hoe weeding at 3 WACE. A significant ($p<0.05$) effect was observed when herbicide was applied overally over the whole plot, with plants being taller in this treatment than in other treatments (Table 2*)*. In mechanical options there was a significant ($p<0.05$) effect of treatment of the number of bolls per plant and plant height in 2000-2001 and 2001-2002. The treatments where pre-emergent herbicides were applied and combined using ox-plough with a mouldboard or without and ox-cultivator had more number of bolls per plant and taller plants than the rest of the treatments. In 2001-2002 season the treatments involved a combination of herbicide and ox plough with mouldboard had higher number of bolls (Table 3**)**. There was no effect ($p<0.05$) of treatments on seed cotton yield in both seasons.

Table 2. Mean effect of mechanical weed control methods on cotton yields and yield components at nine on-farm sites in Muzarabani.

Treatments	2000-2001 season			2001-2002 Season		
	Bolls Plant^{-1}	Plant height (cm)	Yield (kg ha^{-1})	Bolls Plant^{-1}	Plant height (cm)	Yield (kg ha^{-1})
HH	16.70ac	129.72	1461	9.49	79.84a	917
Hcab+HH	19.18b	129.83	1712	9.02	78.50a	788
HH+Hc	-[1]	-	-	10.77	80.32a	794
Hca-full cover	16.16a	129.82	1625	10.93	85.33b	873
Significance	*	Ns	Ns	ns	*	Ns
S.E.D	1.259	2.814	165	1.128	2.395	132.9
HH +OC	14.21a	108.77a	1812	12.12ac	83.54	981
OC+HH	16.30a	118.78b	1910	12.19ac	86.01	1077
OP+D+HH	18.03b	126.91bc	2001	13.31cd	83.02	938
OP-D+HH	16.77ab	122.85b	1806	11.94ac	81.02	902
Hcab+OP+D+HH	17.12ab	130.31c	1945	14.62d	87.28	1020
Hcab+OP-D+HH	22.09c	132.52c	2100	11.26a	82.46	863
Hcab+OC+HH	20.49bc	129.57c	1940	12.60acd	85.74	997
Significance	***	***	Ns	*	ns	Ns
S.E.D.(df.54 6[1],69[2], 615[3])	1.290	2.846	0.1495	1.012 2.024	2.426	0.1137

[1]- Treatment not applied. [1]S.E.D for bolls per plant, [2] S.E.D for plant height, [3]S.E.D for yield

Table 3. Partial budget analysis for, 2000-2001 season.

Treatment	Benefits		Weeding costs					Benefits less costs			Returns to labour
	Yields	Value	Labour	DAP	Labour for herbicide	Herbicide	Knapsack	Total	Margin	Margin over farmer method	%	Z$ hr^{-1}
HH-	1557	93420	13400	-	-	-	-	13400	80020	-		299
Hcab + HH	1557	93420	11750	-	150	2175	800	14875	78545	-1475	-2	334
Hc, HH	1557	93420	8900	-	150	1262	800	11112	82308	2288	3	462
Hca-full cover	1557	93420	3050	--	300	2740	800	6890	86530	6510	8	1419
HH, OC. HH	1931	115834	6750	1750	-	-	-	8500	107334	650	1	795
OC. HH	1931	115834	7400	1750	-	-	-	9150	106684	0		721
OP+D, HH	1931	115834	6600	3500	-	-	-	10100	105734	-950	-1	801
OP-D, HH	1931	115834	6950	3500	-	-	-	10450	105384	-1300	-1	758
Hcab + OP+D +HH	1931	115834	4150	3500	150	2175	800	10775	105059	-1625	-2	1266
Hcab + OP-D+ HH	1931	115834	4150	3500	300	2175	800	10925	104909	-1775	-2	1264
Hcab + OC+ HH	1931	115834	5400	1750	150	2175	800	10275	105559	-1125	-1	977

Partial budgets

On the on-farm daughter trials in the first season, there were no significant differences ($p<0.05$) between yields. Overall herbicide application and banded herbicide along crop row in combination with an Ox-cultivator for weeding interrow were the lowest cost weeding options giving the highest productivity for non-mechanical and mechanical weeding options respectively. Highest returns to labour were derived from overall herbicide and ox-plough with or without a mouldboard (Table 4).

Table 4. Partial budget analysis for 2000-2001 season

Treatment	Benefits		Weeding costs						Benefits less costs			
	Yields	Value	Labour	DAP	Labour for herbicide	Herbicide	Knapsack	Total	Margin	Margin over farmer method	%	Z$ hour^{-1}
HH-	843	50580	13400	-				13400	37180	-		139
Hcab, HH	843	50580	11750	-	150	2175	800	14875	35705	-1475	-4	152
Hc,HH	843	50580	8900	-	150	1262	800	11112	39468	2288	6	222
Hca-full cover	843	50580	3050	-	300	2740	800	6890	43690	6510	18	716
HH, OC. HH	968	58097	6750	1750	-	-	-	8500	49597	650	1	367
OC. HH	968	58097	7400	1750	-	-	-	9150	48947	-		331
OP+D, HH	968	58097	6600	3500	-	-	-	10100	47997	-950	-2	34
OP-D, HH	968	58097	6950	3500	-	-	-	10450	47647	-1300	-3	343
Hcab+OP+D+ HH	968	58097	4150	3500	150	2175	800	10775	47322	-1625	-3	570
Hcab+OP-D+ HH	968	58097	4150	3500	150	2175	800	10775	47322	-1625	-3	570
Hcab+ OC+ HH	968	58097	5400	1750	150	2175	800	10275	47822	-1125	-2	443

The second season gave similar results. Again there were no significant ($p<0.05$) between yields. As in the first season overall herbicide and herbicide combined with ox cultivator were the lowest cost weeding options with highest productivity being achieved with overall herbicide and hand hoe combined with ox-cultivator. Highest returns to labour were derived from overall herbicide and ox-plough with or without a mouldboard (Table 4).

Sensitivity analysis indicates that the price and hence the availability of labour is key. When this is easily available (or not valued) traditional farmer systems (hand hoe and ox-cultivator accompanied with hand hoes) are the most productive. As the labour price increases, due to unavailability or opportunity elsewhere, herbicide systems become more productive**.**

Discussion

In general therefore, cotton crops were protected from weed competition and yield loss by all of the practices or combinations of the practices tested. Weed counts assessments indicated a significant advantage for treatments including a pre-emergence herbicide e.g. in the on-farm trials on plots for which the herbicide was followed up by inter-row weeding with a cultivator or plough from which the mouldboard had been removed, or when herbicide application was compared to hand weeding. This suggests that the use of the herbicide at planting will reduce subsequent labour requirements for weeding. The better weed suppression during the first 3 weeks of the season may also increase the time "window" before it is necessary to begin a second weeding with animal draught, although this would need to be confirmed in the field and may depend on soils moisture conditions.

Previous work in Zimbabwe (Riches *et al*. 1997) has emphasized that weeding with a plough with the mouldboard in place is useful for enhancing soil moisture conservation as well as providing a labour efficient weeding option. In the current trials there was no indication of higher yields where the plough was used for weeding compared to other methods.

Though there were significant differences ($p<0.05$) in weed numbers before any weeding operations, all the methods of weeding were effective in reducing the weed numbers shown by insignificant differences in yield and individual weed species analysis. However, there was a difference in the costs and time of weeding the different weeding methods that is where a farmer needs to choose the most appropriate method according to the resources**.** Resource group one who are well resourced have got a wide range of choice they can choose any option depending on the DAP availability, labour availability and the money for purchasing herbicides. Resource group 2 with limited labour but with DAP available can choose hand hoe along the crop rows and ox-cultivator at 3 WACE this is because less labour is required and it was observed to be the second cheapest option. Farmers with limited DAP in RG2 ad RG3 can consider the option of applying a pre-emergence herbicide in combination with Ox ploughing at 3 WACE this was second list option which required less labour hours. Resource group 4 with limited labour and DAP can consider the use of herbicide either overall herbicide spray which is the cheapest option and requires less time or hand hoe along the crop row at 2 WACE and apply cyanazine between the crop row at 3 WACE, which requires less labour also, or they can plant an area that will be equivalent to the labour available so that they do not end up abandoning their fields. These trials have demonstrated that weeds can be adequately suppressed in cotton by timely use of a number of herbicide and non-herbicide weed control methods. The choice will be influenced by the resources available to the farmer.

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MANAGING BACKYARD AND CREEKLINE INFESTATIONS OF ALLIGATOR WEED [*Alternanthera philoxeroides* (Mart.) Griseb] IN THE HAWKESBURY-NEPEAN CATCHMENT, NEW SOUTH WALES, AUSTRALIA

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Abstract: Alligator weed, a major aquatic weed in Australia, poses a significant threat to waterways and properties. It has the potential to cause losses of millions of dollars from agricultural, tourism and extractive industries in a major river system in Sydney, the Hawkesbury-Nepean River catchment. The Sri Lankan community had mistakenly grown Alligator Weed as a vegetable in their backyards. Campaigns by NSW Agriculture to inform the community and discontinue this practice were launched during 1996-1999. We surveyed the backyard infestations of Alligator weed in suburban residences of the Sri Lankan community living within the catchment, to determine the extent of the problem. Overwhelming evidence was found that the Sri Lankan community is no longer a major factor in spreading Alligator weed in Sydney. The community had ceased growing the weed as a vegetable. The few persistent backyard infestations found were eradicated during 2004. Forty-four infestations, 30 of which originated from the old database were collated for future monitoring. A second strategic project surveyed the extents and spread of Alligator weed along two creek lines (McKenzie's Creek and Marsden Park Creek) within the river catchment, and assessed the level of success of recent control efforts. The creek lines were mapped with 3400 m^2 of Alligator weed found over 5 km, infesting 50 properties. The study identified deficiencies in the treatment regime that had been used by the Local Control Authority. Guidelines and protocols were developed for improved 'site-specific' control and for inspections and recording of infestations. Given that the minor infestations recorded in 1996 in the creeks had expanded to cover larger areas, a more systematic local control plan is needed to stop the spread. Elements of such a plan include educating the public, obtaining local community support, as well as training of contractors and weed control officers.

Key words**:** Alligator weed, management

Introduction

Alligator weed [*Alternanthera philoxeroides* (Mart.) Griseb.] is one of twenty Australian Weeds of National Significance (WONS), and is a declared as a noxious weed in all the council areas of the Hawkesbury-Nepean (H-N) catchment in New South Wales (Figure 1). This invader poses a significant threat to waterways and properties, with potential losses of many millions of dollars to agricultural, tourism and extractive industries in the catchment. Given the seriousness of the threat posed by Alligator weed and other aquatic weeds in this major river catchment, NSW Agriculture established the 'Hawkesbury Nepean Aquatic Weeds Task Force', which functioned from 1999 to 2004. Addressing a key issue identified by the Hawkesbury-Nepean Catchment Blueprint on Biodiversity- 'Coordinated Strategies and Action to control Aquatic and Terrestrial Weeds', a key objective of the Task Force was to identify strategic gaps in Alligator weed management, and act to contain the spread of the weed in the catchment. An outcome of the Task Force's actions was the *Hawkesbury-Nepean Alligator Weed Strategic Plan* (NSW Agriculture, 2001). Goal 3.5 of the Strategic Plan is to '*Ensure that the Sri Lankan Australian community is aware of the prohibition on the cultivation and sale of Alligator weed and is aware of alternative species*'.

This paper summarises the key findings of a collaborative pilot project initiated by the Task Force, targeting strategic gaps in Alligator weed management, and identifies key issues for future attempts to arrest the spread of Alligator weed in the H-N catchment. The Local Government Advisory Group (LGAG)- part of NSW Department of Natural Resources (DNR), funded the project, which commenced in November 2003.

Methodology

Objective 1- Suburban backyard Alligator weed infestations

It had been known since 1995 that Sri Lankan communities were growing Alligator weed as a food source in their backyards (McKenzie, 1996; Gunasekera and Adair, 1999). The weed was mistaken for a similar species – "Mukunuwenna" (*Alternanthera sessilis* (L.) R. Br. ex DC.), a popular vegetable in Sri Lanka. Some older Sri Lankans had mistakenly grown Alligator weed for up to 25 years in Australia. Investigations revealed that Sri Lankans living in major cities, such as Sydney, Melbourne, Brisbane and Canberra, and in various regional centres were all doing the same. Previous work by NSW Agriculture raised awareness of the issue in the community which led to the compilation of a database of infestations in Sydney.

The primary objective of this project was to determine the current occurrences of Alligator weed in Sri Lankan backyards across several suburbs of the Hawkesbury-Nepean catchment, and assist in eradicating these weed populations. This required (a) conducting a media campaign to raise awareness of the issue; (b) identifying the extent of backyard infestations across the catchment; (c) carrying out control free of charge; (d) Substituting Alligator weed with the real 'Mukunuwenna' or an equivalent; and (e) Updating information for each local council to monitor and control future Alligator weed infestations.

The Government Census (2001) indicated that the Sri Lankan community had an estimated 650 households in the H-N catchment, located broadly in the north-west sector of Sydney (Figure 1). The main suburbs were Kellyville, Castle Hill, Quakers Hill, Baulkham Hills, Plumpton, St. Marys and St Clair. A new list of addresses was collated from the phonebook, and 610 letters were sent with the leaflet ("Don't Mistake Alligator Weed for Mukunuwenna"). Households were also contacted by phone. Seventy letters were sent with the leaflet to properties on the old database as well. Other methods used to reach the community were: (a) Posters and leaflets placed in six grocery stores and two Buddhist temples; (b) newspaper articles in five local papers, one Sri Lankan paper and in two Newsletters of Sri Lankan Community Organisations; (c) webpage banner advertisement on main Australian Internet site for Sri Lankans – *www.ozlanka. com.au*, with photos; the advertisement ran from three months, 15 Jan-15 April 2004), and (d) SBS radio interview (for Sri Lankan audiences).

The main message in the letters, posters and leaflets was that Alligator Weed is not 'Mukunuwenna'. Other information comprised: (a) tips on identifying the two species apart from one another; (b) negative effects of Alligator weed on human health and the environment; (c) its 'Noxious Weed' status under legislation (Noxious Weeds Act 1993); (d) dispersal mechanisms; (e) offer of free home visit for control and free substitute vegetable. Feedback from the community indicated that information on human health effects had the biggest impact on them in changing their behaviour.

Providing a replacement vegetable for Alligator weed, either 'Mukunuwenna' itself (*A. sessilis*) or a large-leaf form- *Alternanthera denticulata* R. Brown, was important as part of the strong message given to the Sri Lankan community. *Alternanthera denticulata*, a native Australian species, had been successfully used in both NSW and Victoria, after testing for nutrition and community acceptance (Gunasekera and Bonila, 1999). Property inspections were carried out with landholders, and infestations treated with Wipeout 360 ™ (glyphosate), Brush-off ™ (metsulfuron methyl) or Kamba M ™ (MCPA/dicamba), if the infestation was in a grass lawn.

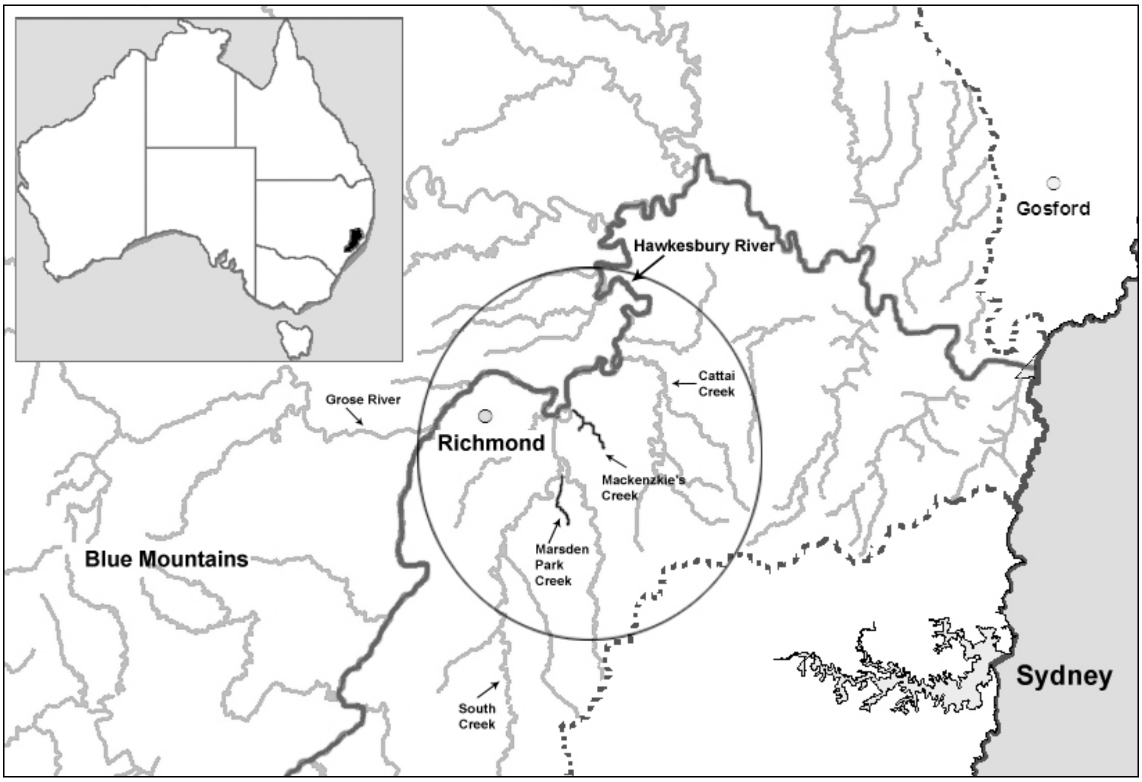

Figure 1. Part of the Hawkesbury-Nepean River catchment in NSW. Australia. Note the H-N River and locations of two small tributaries- Mckenzies Creek and Marsden Creek

Objective 2- Creekline Alligator weed infestations

Alligator weed spreads predominantly along creek lines. The weed arrives in the creek line mainly through stormwater flows and flooding when stem fragments break off from an infestation and are transported by water to new areas. Dispersal also occurs through human activities (earth moving machinery and movement of sand, soil or mulch contaminated with stem fragments) or through movement of animals. Spread is only via vegetative means, as no viable seeds are set in Australia. Often, infestations occupy the creek bed or shoreline, and spread on to the bank, riparian zones and eventually the floodplains, drainage channels and billabongs. Given the above, the second aspect of the project was to: (a) assess the spread of Alligator weed along McKenzie's Creek and Marsden Park Creek, and (b) assess the level of success of recent control efforts.

Results and Outcomes

Ten new backyard infestations were discovered through the media campaign. Four new infestations were found by mail-outs and follow up phone calls made to homes. From the 66 properties inspected from the old database of 68 properties, 30 small infestations were found again (45%). Four neighbouring properties were discovered to also have infestations. The database was updated to show 48 new property infestations for future monitoring by relevant Local Control Authorities (Table 1). About 50% of the infestations were less than 1 m^2, but a few (16%) were large enough to warrant aggressive control action, which was taken (Table 2). Herbicide treatments were carried out in 36 properties while the householders treated the remainder (Table 1).

Table 1. Updated database of infestations, 2004.

Source	Total	Control actions taken by Project	Control action by Householder
Old database	30	22	8
Neighbouring properties (Old database)	4	3	1
Located by Media campaign	10	8	2
Located by phone calls to mail out list	4	3	1
Total	48	36	12

Table 2. Size of Alligator weed infestations found in Sri Lankan backyard properties

Source	<1 m^2	1-3 m^2	4-6 m^2	10-25 m^2	Total
Old database	17	6	4	3	30
Neighbouring properties (Old database)	1	2	1	-	4
This project	7	7	-	-	14
Updated database	25	15	5	3	48

Metsulfuron methyl, the selective herbicide was used in 80% of infestations (Plate 1). All three chemicals were effective in killing the plant above the ground, but may require some follow-up treatments. A very high proportion (94%) of Sri Lankans contacted does not have Alligator weed in their properties; none were found to still cultivate the weed.

(a) 2 April 2004 (b) 29 May 2004

Plate 1. Control of a backyard infestation, among grass, by metsulfuron-methyl

The targeted creeklines- Mckenzies Creek and Marsden Park Creek (Figure 1) were surveyed over several days and the infestations mapped. Approximately 3400 m^2 of Alligator weed was found over 5 km of the two creeks and in 50 properties associated with the creeks. Being relatively new infestations, the patches were not dense. Evidence of significant activity of the biological control agent - Alligator weed flea beetle *Agasicles hygrophila* was recorded from both creeks, surveyed in February-March 2004. In some patches, grazing damage caused by the beetle had reduced visible biomass by as much as 50%; yet, the weed survived and recovered.

Minor amounts of Alligator weed had first been noticed in Mckenzies Creek around 1995, mostly in the upstream areas of the H-N catchment. From these upstream areas, infestations had quickly spread down to infest the rest of the creek, covering some 45 properties (generally 2 ha lots). A few heavily infested properties had received treatments by the control authority, but these once or twice yearly herbicide applications were found to be ineffective. In Marsden Park Creek, the infrequent treatments carried out in recent years by the control authority were found to be ineffective. Although the infestations were not very dense, the mapping indicated that Alligator weed had spread more than 1 km from its original infestation location in this creek, over a few years.

Conclusions

It appears that Alligator weed is not widespread in backyard properties of the Sri Lankan community anymore. However, as evidenced by the small number of new infestations found, the community can still be a ‘vector’. The campaigns since 1996 have been effective, and our project estimated that a very high proportion of Sri Lankans are aware of the problem, and do not contribute to spreading of the weed. The fact that a small % of infestations from this source is still in the catchment means that occasional property inspections are required to eradicate this source of infestation.

The creekline infestations are much more serious, and managing these are the legal responsibility of the local control authority- Hawkesbury River County Council (HRCC), which has weed spray operators, inspectors and supervisors, to conduct management activities. Given that minor infestations found in Mckenzies Creek and Marsden Park Creek in 1996 have grown so large, more effective management action by the control authority is essential.

A key lesson learnt from the project is that it is necessary to prioritise Alligator weed funding for control works at the head of catchments and sub-catchments. Mapping of infestations, creating a database of infestations, contacting landholders and providing advice on management, are all essential for an integrated management effort. Experience has shown that follow-up inspection and treatment is also essential. Guidelines for Alligator weed management in rural and semi-urban properties have been developed, and these need to be widely published for use by landholders. In all cases, it is necessary to detect early and investigate the sources of a new infestation (such as suppliers of mulch, sand, soil and heavy machinery), so as to prevent further spread.

It is also necessary for strategic Alligator weed management to place the onus of control on the landholders. The Local Councils are required to conduct diligent education and awareness campaigns, and regular monitoring of landholder eradication efforts. Letters of non-compliance may follow these.

Whilst private landholders may be asked to play an increased role in managing Alligator weed in their properties, other landholders, such as the local councils and government agencies also need to be held accountable for more effective management of infested road culverts, creeklines and drainage lands. Stakeholder consultations during the project confirmed the view that local communities need to be more closely involved in Alligator weed management, and this can be achieved through the formation of local Landcare groups.

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EFFECT OF HERBICIDE APPLICATION ON SOIL AND WATER CONSERVATION IN SLOPY GARDENS

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Abstract: A weeding trial was conducted to study the degree of water runoff and soil erosion as affected by herbicide application in peach and tea gardens, located in two different in the hilly area of Sichuan Basin, China. The treatments were single applications of glyphosate, paraquat, and 3 times paraquat +1 time glyphosate, tillage, and slashing of weeds (CK) — were carried out four times a year. Water runoff, soil erosion, weed biomass and degree of cover in the trial plots were monitored for three years. Soil erosion and nutrient loss by water run off in weeding trial plot of five treatments were as follows: tillage > glyphosate > paraquat > 3 times paraquat +1 time glyphosate > weed slashing (CK). The reduction in soil erosion in the trial plots with glyphosate, paraquat, 3 times paraquat +1 time glyphosate and weed slashing (CK) when compared with tillage, were 71.2%, 84.3%, 83.0%, and 89.3% respectively for the tea gardens with 18° slope, and 16.2%, 52.3%, 79.5%, and 82.8%, respectively, in tea gardens with a slope of 8° in tea. The reduction of soil erosion in peah gardens with a slope of 25° under the same treatments were 69.2%, 83.1%, 84.5%, and 91.9%, respectively, while in peach gardens with 10° slope, it was 66.1%, 78.5%, 75.8%, and 85.2%, respectively. Loss of OM, N, P K in the experimental plots with glyphosate application, when compared to paraquat application, was increased by 94.7%, 80.4%, 83.0%, and 93.5%, respectively, in tea gardens with 18° slope, and 75.9%, 77.8%, 77.8%, and 71.5%, respectively, in tea gardens with a slope 8°. The loss of these nutrients in the same comparison were 46.7%, 54.9%, 66.1%, 95.8% and 72.1%, 62.7%, 54.9%, 58.7%, respectively, in peach gardens with slopes of 25° and 10°. The biomass and total coverage degree of weed in the plots was the least in the glyphosate-treated plots and the maximum was in the plots where weeds were slashed manually (CK) in the peach gardens. Paraquat residue was observed in the top 10-cm of plots treated with the herbicide (Gramoxone®) but was not detected in the water runoff.

Key words: Sloping lands, nutrient loss, soil erosion, weed control

Introduction

There is a conflict between the increasing population and farmlands in the hilly area of the Sichuan Basin of China. This problem has been intensified due to the shifting slope land from annual cropping into perennial cropping or forests for soil and water conservation. Many slopes have been planted with fruit and non-wood tree crops and species such as tea and peach and have become the main income for farmers. Most farmers till the soil of these plantations on sloping lands for weed control, which causes surface water runoff, soil erosion and nutrient loss. Therefore a research program was conducted to determine optimal methods of weed control on these lands while minimizing damage in terms of soil and water loss land degeneration.

Materials and Methods

Field experiment

Five trial plots, each with dimensions of 20 m x 5 m were established on selected on 2 year old tea gardens with slopes of 18° and 8°, and on 15 year old peach gardens with slopes of 25° and 10°, respectively (Table 1). The trial plots of each slope l were treated with glyphosate; paraquat (Gramoxone®), 3 times paraquat +1 time glyphosate (applied at the last time in a year), tillage or Weeds slashed. (CK) The treatments were imposed in March, June, August and November.

of each year. The Syngenta (China) Investment Company Ltd. supplied Gramoxone®, and glyphosate was bought from the open market. The dosage of Gramoxone® (20% paraquat solution) and glyphosate (10% glyphosate solution) used were 3000 ml ha^{-2} and 15000 ml ha^{-2}, respectively. Tillage plots were hoed and in plots, which were slashed, the weeds were cut manually and placed on the soil surface as mulch. Prior to the imposition of treatments, the total cover of weed was determines using 5 quadrats of $1 \times 1m^2$ area in each plot. The soil was sampled to a depth of 20 cm in all trial plots using a 5 cm x 5 cm core sampler for determination of nutrients. For analysis of residual Paraquat, the samples of water runoff were collected in Aug. of the final year of experimentation in plots treated with Gramoxone, Weed slashing (CK), 3 times paraquat +1 time glyphosate. Soil samples were also collected in same trial plots at depth of 0~10 cm and10~20 cm, respectively, in December of the last year of the experiment.

Table 1. The situation and characteristics of trial plots

Trial plot	Lat. and Longi.	Alt.	Slope	Land form	Rainfall: Average annual rainfall and characteristic	Rainfall: The most rainfall in recent decades	Weeding
Tea	N30°30' 32" E103°13' 01"	670m	18°,8°	Gentle hill	1593.8mm, Abundant, concentrated and storm frequent in Summer, overcast and rainy continuously in Autumn	240	Tilling
Peach	N30°01' 56" E104°17' 56"	820m	25°	Low mountain	884.7mm, Dry in Winter, drought in Spring, storm frequent and a little waterlogged in Summer	180	Tilling
	N30°01' 27" E104°17' 50"	740m	10°				

Chemical analysis

The soil sampled for chemical analyses in trial plots on tea and peach garden was air-dried, ground and sieved using nylon screens of 2.0 mm and 0.25 mm. The samples were stored for analyzing pH, organic matter, total and available N, P, and K. Sub samples of the soil soon after sampling were analyzed for bulk density and Maximum moisture holding capacity (Table 2). The samples of eroded soil were air-dried, mixed thoroughly, passed through nylon screens of 2.0mm and 0.25mm, and stored for the analysis of organic matter, total and available N, P, and K. The water samples collected from trial plots were filtered and the soil on the filter paper weighed. The water and soil were sent to Safety Evaluation Center of Shenyang Chemical Academy and was tested by HLPC (Aglient 1100) for residual Paraquat. The methods of soil analysis used were those described by Wanru *et al.* (2000).

Results and Discussion

Water runoff

The water runoff on an average from steep and gentle slopes of plots treated with glyphosate, paraquat (Gramoxone®), 3 time paraquat + 1 time glyphosate, tillage and weed slashing(CK) was 1486.9 m^3 ha^{-1} a^{-1}, 1087 m^3 ha^{-1} a^{-1}, 642.9 m^3 ha^{-1} a^{-1}, 1197.3 m^3 ha^{-1} a^{-1} , 731.2 m^3 ha^{-1} a^{-1} in tea gardens and 145.2 m^3 ha^{-1} a^{-1}, 224 m^3 ha^{-1} a^{-1}, 178.3 m^3 ha^{-1} a^{-1}, 427.3 m^3 ha^{-1} a^{-1}, 175.2 m^3 ha^{-1} a^{-1} in peach gardens, respectively (Table 3).

Table 2. Physical and chemical properties of soil in tea and peach gardens

Trial plot	Type of soil	pH value	Bulk specific gravity	Max. moisture holding capacity	OM	Total N	Total P	Total K	Available N	Available P	Available K
			$g \cdot cm^{-3}$			$g \cdot kg^{-1}$				$mg \cdot kg^{-1}$	
Tea	Yellow soil	5.0	1.15	456.3	16.2	1.0	0.15	10.7	74.5	3.3	36.8
Peach	Purple soil	8.2	1.54	283.8	10.1	0.65	0.29	22.5	27.3	19.1	117.1

Table 3. Water runoff and soil eroded in the experimental plots#

Trial plot	Treatment	Steep slope (18° for tea, 25° for peach) Water runoff	Steep slope Soil erosion	Gentle slope (8°for tea, 10°for peach) Water runoff	Gentle slope Soil erosion
		$l/100m^2.a$	$g/100m^2.a$	$l/100m^2.a$	$g/100m^2.a$
Tea	Glyphosate	14941.7	3676.4	14795.5	3283.3
	Paraquat	6644.8	2073.3	15095.1	1911
	Tillage	8777.5	10692.8	15168	4352.9
	Weed cutoff(CK)	6725.9	1197.2	7898.2	736.9
	3 time paraquat +1 time glyphosate	6260.1	1980.9	6597.4	914.4
Peach	Glyphosate	1762.2	23970.6	1141.6	11179.4
	Paraquat	3477.4	13199.5	1003.5	7087.5
	Tillage	7153.6	77733.1	1392.8	32946.9
	Weed slashing(CK)	2580.1	6304.8	924.5	4857.3
	3 time paraquat +1 time glyphosate	2647.6	12058.6	916.8	7978.4

#The data in the table is averaged for three years from 2003 to 2005.

Water runoff was generally more from plots treated with tillage and less in those where weeds were slashed (CK). This is attributed to the placement of weeds on the soil surface after slashing, which also enhanced water infiltration. In addition, physical properties and depth of soil are also important factors affecting water runoff (Donghai *et al*. 2001).

Soil erosion

Splash erosion due to raindrops and surface erosion by water was highest in the tilled plots. This is due to the exposure of soils by tillage and the lack of roots to bind the soil (Mingshu *et al*. 1991, Guiquan *et al.* 1991). Gramoxone application reduced soil erosion to a greater extent than glyphosate as paraquat killed only the above ground part of weeds and not the roots, which held soil particles together (Fuyou, 1991). Soil erosion in slashed plots (CK) was the lowest due to the protection offered by the mulch and the presence of roots. The magnitude of soil erosion in trial plots of five treatments was Tillage > glyphosate > paraquat, 3 times paraquat +1 time glyphosate > weed slashing (CK) (Table 3).

The soil erosion in trial plots treated with glyphosate, paraquat , 3 times paraquat + 1 time glyphosate and weed slashing (CK)were reduced by 71.2%, 84.3%, 83.0%, and 89.3% , respectively in the tea gardeb with 18° slope, and 16.2%, 52.3%, 79.5%, and 82.8%, respectively in the tea garden with 8° slope. Soil erosion was reduced by the same treatments by

69.2%, 83.1%, 84.5%, and 91.9%, respectively, in the peach garden with 25° slope, and by 66.1%, 78.5%, 75.8%, and 85.2%, respectively, in peach garden with 10° slope, when compared to the tilled plots. The soil erosion in trial plots with tillage, glyphosate, paraquat, 3 times paraquat +1 time glyphosate and weed slashing (CK) was 106.9 t km^{-2} a^{-1}, 36.8 t .km^{-2} a^{-1}, 20.7 t km^{-2} a^{-1}, 19.8 t km^{-2} a^{-1}, 12.0 t km^{-2} a^{-1} and 43.5 t .km^{-2} .a^{-1}, 32.8 t .km^{-2} .a^{-1},19.1 t .km^{-2} .a^{-1}, 9.1 t .km^{-2} .a^{-1}, 7.4 t .km^{-2} .a^{-1} on slopes of 18° and 8° in tea gardens, and 777.3 t .km^{-2} .a^{-1}, 239.7 t .km^{-2} .a^{-1}, 132.0 t .km^{-2} .a^{-1}, 120.6 t .km^{-2} .a^{-1}, 63.0 t .km^{-2} .$a^{-1}$and 329.5 t .km^{-2} .a^{-1}, 111.8 t .km^{-2} .a^{-1}, 70.9 t .km^{-2} .a^{-1}, 79.8 t .km^{-2} .a^{-1}, 48.6 t .km^{-2} .a^{-1} on slopes of 25° and 10° in peach gardens, respectively. Thus, herbicide application was more effective in soil conservation of steep slopes.

Nutrient element loss of soil erosion

Generally surface erosions removed fertile topsoil, thus nutrients are lost, leading to the degradation of lands. In this study, the nutrient element loss due to soil erosion in the five treatments was similar to that of soil erosion, *i.e.* that was tillage > glyphosate > paraquat, 3 times paraquat +1 time glyphosate > weed slashing (CK) (Table 4). The loss of OM, N, P, K in the trial plot treated with glyphosate, when compared to paraquat increased by 94.7%, 80.4%, 83.0%, and 93.5%, respectively, and 75.9%, 77.8%, 77.8%, and 71.5%, respectively in tea gardens with slopes of 18° and 8°. The soil erosion was reduced by 46.7%, 54.9%, 66.1%, and 95.8%, respectively, and 72.1%, 62.7%, 54.9%, and 58.7%, respectively, in peach gardens with slopes of 25°and 10°.

Table 4. Nutrient loss in trial plots on tea and peach garden[#]

Trial plots	Treatment	Steep slope 18°for tea, 25°for peach				Gentle slope 8°for tea, 10°for peach			
		OM	Total N	Total P	Total K	Organic matter	Total N	Total P	Total K
		g/100m²							
Tea	Glyphosate	213.47	11.53	2.91	50.66	142.19	7.31	2.08	41.72
	Paraquat	109.64	6.39	1.59	26.18	80.84	4.11	1.17	24.23
	Tillage	457.06	28.47	6.68	126.14	185.32	9.36	2.38	56.75
	CK**	67.22	3.66	1.01	14.39	34.01	1.68	0.52	9.15
	3 Paraquat. + 1 glyphosate	117.85	6.24	1.55	23.66	46.88	2.46	0.64	11.17
Peach	Glyphosate	624.65	45.91	22.96	645.9	225.40	15.03	11.31	269.24
	Paraquat	425.70	29.63	13.82	329.88	130.98	9.24	7.30	169.62
	Tillage	1789.3	128.01	76.58	1899.8	655.07	45.46	37.37	743.58
	CK	178.69	12.93	6.45	152.14	72.03	5.75	5.06	107.66
	3 paraquat. + 1 glyphosate.	270.92	20.39	9.55	280.82	129.00	9.55	8.41	175.34

[#]The data in the table is averaged for three years from 2003 to 2005. [**] Slashing.

Loss of K and organic matter was clearly evident in all in trial plots. The loss of K and organic matter in tilled plots on slopes of 25° in peach gardens reached 19 t km^{-2} a^{-1} and 17.9 t .km^{-2} a^{-1}, respectively. Potassium loss was equivalent to K_2SO_4 of 40 t. Nitrogen loss in the same trial plot reached 1.3 t .km^{-2} a^{-1}, which was equivalent to 2.8 t of Urea.

Biomass and total coverage of weeds

The weed biomass (dry weights) and total coverage of weeds were minimum in plots treated with glyphosate (28.3 g.m^{-2}~65.0 g.m^{-2}, 24.2%~44.8% in tea gardens and 43.9 g.m^{-2}~58.7 g.m^{-2},41.8%~53.2% in peach gardens, respectively (Table 5).This indicated that glyphosate controlled weeds very effectively. . The impact of the weed control methods on biomass and total weed cover was in the order glyphosate > paraquat > 3 times paraquat + 1 time glyphosate > weed slashing (CK) > tillage in tea gardens.

Table 5. The biomass and total cover degree in trial plots on tea and peach garden[#]

Trial plots	Treatment	Steep slope 18° for tea, 25° for peach		Gentle slope 8° for tea, 10° for peach	
		Biomass	Total cover	Biomass	Total cover
		g.m^{-2}	%	g.m^{-2}	%
Tea	Glyposate	65.0	44.8	28.3	24.2
	Paraquat	148.2	75.6	131.8	56.8
	Tillage	256.4	90.6	243.6	78.4
	Weed slashing(CK)	218.9	88.5	206.1	78.4
	3 times paraquat+1 time glyphosate	127.5	60.0	133.8	47.0
Peach	Glyphosate	43.9	41.8	58.7	53.2
	Paraquat	118.2	60.9	111.0	67.2
	Tillage	88.0	80.8	113.4	75.2
	Weed slashing(CK)	207.2	94.8	236.6	94.2
	3 times paraquat+1 time glyphosate	118.4	60.0	86.8	65.6

[#]The data in the table is averaged for two years from 2004 to 2005.

In the peach gardens, the effect was in the order of glyphosate > paraquat > 3 times paraquat + 1 time glyphosate > tillage > weed slashing (CK) except for the biomass of the trial plot treated with tillage on slope of 25°. Although the biomass and the total coverage degree of weed was higher in the tilled plots, soil erosion was also the highest in these plots due to soil exposure and destruction of the structure by tillage, causing splash erosion and surface run off when compared to other treatments (Mingshu *et al.* 1991). A comparison of the two herbicides illustrated that paraquat was better in reducing soil erosion than glyphosate, due to the contact nature of its activity.

Residual paraquat

Paraquat residue in soil was lower in the peach garden at a soil depth of 0~10cm. There was no difference between the residual values of this herbicide at depths of 10 – 20 cm in both gardens (Table 6). Th residue was also lower at soil depth of 10 – 20 cm. Residual paraquat in soil depth of 0~10cm was the maximum in the plots treated with paraquat (Gramoxone®), moderate in the plot with 3 time paraquat + 1 time glyphosate and the minimum in the plot where weed were slashed (CK) in both tea and peach gardens. There were paraquat residues at the soil depths of 0~10cm in the plots where weeds were slashed even without Gramoxone® application

Conclusions

Soil and water conservation on both tea and peach gardens was the best in plots where weeds were slashed, moderate with herbicide application and the worst with tillage. The impact was greater on steep slopes. Herbicide application for weed control was more effective, convenient and time saving than slashing or tillage. The results obtained in the experiment showed that the conservation of soil and N, P, K of nutrient elements was better in plots treated with Gramoxone® (paraquat) than with glyphosate on both tea and peach gardens. Weed control in plots treated with Glyphosate was also better than in other plots. Paraquat residue in Gramoxone® treated plots was evident in the top 10 cm of soil, but was not detected in runoff water.

Table 6. The paraquat residue in the soil and water runoff in trial plots

Trial plots	Treatments	Soil 0~10cm	Soil 10~20cm	Water runoff
		mg kg^{-1}		
Tea	Gramoxone	3.6	0.11	NDP[§]
	Weed slashing (CK)	0.34	#LOQ	NDP
	3 times Gramoxone +1 time Glyphosate	1.0	#LOQ	NDP
Peach	Gramoxone	1.6	0.074	NDP
	Weed slashing (CK)	0.090	#LOQ	NDP
	3 times Gramoxone +1 time Glyphosate	1.1	0.10	NDP

[#]Limit of qualification, LOQ=0.05 mg.kg^{-1} in the analyses. [§] No detectable peak - Paraquat was not detected in the analyses.

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INTEGRATED WEED MANAGEMENT IN CANOLA (*Brassica napus* L.)

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Abstract: A field experiment was conducted to study the effects of mechanical and chemical methods of weed control and their combinations on the grain and oil yield of canola *cv*. Hayoola. The treatments consisted of eleven combinations of mechanical cultivations (using a cultivator) and chemical control of weeds with trifluralin and sethoxydim herbicides (pre-sowing and post-emergent herbicides, respectively) namely T1: trifluralin (pre–plant herbicides), T2: trifluralin + one cultivation, T3: trifloralin + two cultivations, T4: trifluralin + sethoxydim, T5: trifluralin + sethoxydim + one cultivation, T6: trifluralin + sethoxydim + two cultivations, T7: sethoxydim + two cultivations, T8: sethoxydim + one cultivation, T9: sethoxydim, T10: one cultivation, T11: two cultivations, T12: Total weed control, and T13: No weed control. The highest weeds control, and grain and oil yields were obtained in treatment with two cultivations plus trifluralin and sethoxydim herbicides. The highest grain yield was due to higher number of pods per plant, grains per pod and lateral branches.

Key words: Integrated weed management, mechanical cultivation, trifluralin, sethoxydim

Introduction

Canola (*Brassica napus* L) is an important oil crop and is being promoted for cultivation (Ahmadi, 2000). Weeds are a limiting factor of Canola production and mechanical, chemical, agricultural and biological methods are used for their control (Swanton and Weise 1991). Herbicides are to be used due to their effectiveness in controlling a wide spectrum of weeds, nevertheless the studies have shown that, the herbicides applications are not always successful due to different ecotypes, differential growth periods, tolerance against herbicides as well as environmental contamination (Swanton and Murphy, 1996). The repeated use of mechanical methods, sometimes results in plant damage (Buhler *et al.* 1995). Thus, the application of integrated management methods becomes necessary, where different techniques for weed management, *i.e.* agricultural, mechanical and chemical methods are used collectively (Shaw 1982). Thus, integrated weed management is considered more effective in controlling weeds, and their effectiveness was tested in the management of weeds in canola.

Materials and Methods

The experiment was carried out at the agricultural research and demonstration farm of Ramin, of the Shahid Chamran University of Ahwaz, Iran**.** The treatments were: T1: trifluralin (pre–plant herbicides), T2: trifluralin + one cultivation, T3: trifluralin + two cultivations, T4: trifluralin + sethoxydim, T5: trifluralin + sethoxydim + one cultivation, T6: trifluralin + sethoxydim + two cultivations, T7: sethoxydim + two cultivations, T8: sethoxydim + one cultivation, T9: sethoxydim, T10: one cultivation, T11: two cultivations, T12: total weed control, and T13: No weeding. Pre–plant herbicide treatments that were used one week before planting Canola were incorporated to the soil by a light disc. Trifluralin was applied at a rate of 2.5 l per ha prior to planting, while sethoxydim was used at rate of 2 l per ha before the stem elongation phase.

The herbicides were applied using a sprayer equipped with a flat fan nozzle, with a spray volume of 300 l per ha. The sprayer pressure used for trifluralin and sethoxydim herbicide applications was 250 and 200 Kpa, respectively. The cultivator was used before the stem elongation phase and before flowering (10 and 25 days after application of sethoxydim, respectively). A manually weeded control was maintained for comparison of data.

Sampling of weed species was done once in 8 days using a quadrat at a size of 0.1 m^2. The weed samples were separated to different species after being taken to the laboratory. The number of weeds was calculated, and then the weeds were dried in an oven and the dry weights were measured. The yield and yield components were estimated after the crop reached its physiological maturity, by harvesting the interior rows, leaving out 0.5 m strip in the border as the guard rows. The harvest area was 3 m^2. The oil percentage of the seeds were determined by the Soxleht method. Oil yield was calculated as per equation proposed by Bestawesy *et al.* (1991) as Oil yield per unit area = Grain yield per area***** Grain oil per cent. Statistical analysis was done using the SPSS statistical program.

Results and Discussion

Atriplex patulum L, was the most common weed in all treatments while *Malva rotundifolia* L, was abundant in the non weeded plots (T13) and when sethoxydim herbicide (T9) was applied (Table1). Control of *Malva rotundifolia*, was better with Trifloralin pre–planting herbicide, than with sethoxydim herbicide (Table1), as shown by Hejazi (2001).In addition, managing *M. rotundifolia* L., was more effective with in pre–planting herbicidal treatments with one and two – cultivations. The results also showed that, using trifluralin herbicide had more efficiency than sethoxydim herbicide in order to decrease the weed numbers of *Atriplex patulum* L. This phenomenon could be attributed to these broadleaved plants having large growing organs and straight root systems. Cultivation has also significantly decreased its numbers. The impact of the experimental treatments on weed numbers of *Convolvulus arvensis* L. was statistically significant (Table1).

The difference of total numbers of weeds was significant in the experimental treatments. The lowest total number of weeds was in the treatment trifluralin + sethoxydim + two cultivations , which was also similar to the use of trifluralin + two cultivations. The weed numbers of these were lower to the perfectly management treatment (T12). The results also agreed with those of Singh *et. al* (1999) who demonstrated illustrated that 75.9% of weeds could be managed by integrating herbicides with two cultivations.

The treatments T12, T6 and T3, developed the most numbers of pods in Canola plants and those of T9 and T13 had the lowest numbers (Table2). In general, the two – cultivations increased the numbers of pods on Canola plants. The use of two- cultivations with trifluralin pre–plant herbicide increased numbers of pods significantly (Table2) as shown by Hatami (2000) and Bestawesy *et al* (1991).

Grains per pod and seed yield was the highest in the plots, which were weed free (T12) followed by pre and post – plant applications of herbicides with two cultivations (T6) (Table 2). Evaluating the correlation coefficient matrices of seed yields and its elements showed that there was a negative and significant correlation (r = -0.75**) between weed numbers and grain yield and between grain number in pod (r=0.60*) and pod number in bushes and a positive significant relationship (r=0.65*) between grain number and seed yields. Bestawesy *et al.* (1991) reported that, weed a cumulative decrease in weed numbers reduces, the competitive effects thus, enhancing radiation distribution within the canopy and thus improve the microclimate, which in turn promoted the development of pod numbers in bushes, seed in pods and grain yields .

Harvest index was the lowest in the un-weeded plots due to severe weed competition (Table2). The difference in grain oil content between plots with different treatments was not significant. The results showed that the oil yield increased due to greater the grain yields There was a significant positive correlation (r=0.98**) between grain yield and oil yield.

Table1. Means of weed number and control percentage in different weed control treatments.

Treatments		Total		*Sinapis arvensis, Lolium temulentum and Trifolium spp*		*Convolvulus arvensis*		*Atriplex patulum*		*Malva rotundifolia*	
		Weed No.	Control (%)	Weed No.	Control (%)	Weed No.	Control (%)	Weed No.	Control (%)	Weed No.	Control (%)
T_1	Trifluralin	182 c	49	41b	26	26 b	18	50 a	6	64 b	37.0
T_2	Trifluralin + one cultivation	88 d	75	14 de	74	9 c	71	34 b	36	30 c	71.0
T_3	Trifluralin + two cultivations	52 e	85	10 e	81	7 cd	78	19 d	65	16 de	84.0
T_4	Trifluralin + Sethoxydim	174 c	51	33 c	39	27 b	16	49 a	8	65 b	36.0
T_5	Trifluralin + Sethoxydim + one cultivation	79 d	78	10 e	81	9 c	72	28 bc	47	32 c	69.0
T_6	Trifluralin + Sethoxydim + two cultivations	45 e	87	7 e	86	5 d	85	16 d	69	16 de	84.0
T_7	Sethoxydim + two cultivations	58 de	84	10 e	81	7 cd	78	19 d	64	21d	79.0
T_8	Sethoxydim + one cultivation	92 d	74	14 de	74	12 c	63	33 b	38	33 c	67.0
T_9	Sethoxydim	223 ab	37	41b	25	39 ab	11	51 a	4	102 a	0.1
T_{10}	One cultivation	101 d	71	18 d	68	12 c	63	36 b	32	35 c	66.0
T_{11}	Two cultivations	62de	83	14 de	73	7 cd	78	19 d	63	20 d	81.0
T_{12}	Control all of weeds	0 f	100	0 f	100	0 e	100	0 e	100	0 f	100.0
T_{13}	Without weed control	243 a	0	55 a	0	32 a	0	53 a	0	102 a	0.0
Significance level		**		**		**		**		**	

Within a column, values followed by the same letter are not significant different by the LSD test (p=0.05).

Table2. Means of oil yield, harvest index, grain yield and yield components in different weed control treatments.

Treatments		No. of Pods Plant^{-1}	No. of Grains Pod^{-1}	Total Grain Weight (g)	Grain Yield (g.m^{-2})	Oil Yield (g.m^{-2})	Harvest Index (%)
T_1	Trifluralin	113.2 ^{de}	18.92 ^{b}	3.79 ^{a}	151.30 ^{bc}	70.06 ^{dc}	44.03 ^{c}
T_2	Trifluralin + one cultivation	148.1^{d}	21.09 ^{ab}	3.88 ^{a}	211.36 ^{b}	98.66 ^{c}	50.01 ^{ab}
T_3	Trifluralin + two cultivations	221.3 ^{b}	22.29 ^{a}	4.15 ^{a}	285.20 ^{ab}	131.81 ^{ab}	56.27 ^{ab}
T_4	Trifluralin + Sethoxydim	114.6 ^{de}	18.97 ^{b}	3.87 ^{a}	157.80 ^{bc}	73.85 ^{dc}	45.37 ^{c}
T_5	Triflyralin + Sethoxydim + one cultivation	142.6^{d}	21.07 ^{ab}	3.77 ^{a}	225.60 ^{b}	105.44 ^{bc}	50.80 ^{ab}
T_6	Trifluralin + Sethoxydim + two cultivations	223.2 ^{b}	22.85 ^{a}	4.17 ^{a}	301.56 ^{ab}	141.31 ^{ab}	57.71 ^{ab}
T_7	Sethoxydim + two cultivations	185.7^{c}	21.16 ^{ab}	4.11^{a}	283.88 ^{ab}	124.79 ^{b}	55.62 ^{ab}
T_8	Sethoxydim + one cultivation	137.9 ^{d}	20.87 ^{ab}	3.70 ^{a}	205.18 ^{b}	95.49 ^{c}	52.24 ^{ab}
T_9	Sethoxydim	95.6 ^{f}	16.01 ^{c}	3.80 ^{a}	99.50 ^{d}	46.11 ^{e}	42.27 ^{c}
T_{10}	One cultivation	125.0 ^{d}	20.72 ^{ab}	3.90 ^{a}	200.46 ^{b}	93.17 ^{c}	52.05 ^{ab}
T_{11}	Two cultivations	183.0 ^{c}	21.2 ^{ab}	4.09 ^{a}	280.00 ^{ab}	132.71 ^{ab}	55.06 ^{ab}
T_{12}	Control all of weeds	333^{a}	22.88 ^{a}	4.2 ^{a}	355.55 ^{a}	168.45 ^{a}	61.25 ^{a}
T_{13}	Without weed control	95.1^{f}	15.85 ^{c}	3.75 ^{a}	94.6 ^{d}	43.56 ^{e}	41.37 ^{c}
Significance level		**	**	Ns	**	**	**

Within a column, values followed by the same letter are not significant different by the LSD test (p=0.05).

Conclusions

The results showed that the most effective weed control was achieved in the treatments using two–cultivations with trifluralin and sethoxydim herbicides, as well as two-cultivations with trifluralin. The absence of weeding (T13) or with sethoxydim herbicide produced the lowest yield s and oil contents due to the competition by weeds. Generally, the most effective control was with on two – cultivations, however due to the absence of significant differences between this treatment and when trifluralin and sethoxydim was applied, the use of two–cultivations and trifluralin seems to be more desirable for weed management in canola.

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BIO-EFFICACY OF BENSULFURON METHYL 0.6% + PRETILACHLOR 6.0% GR ON WEED MANAGEMENT IN TRANSPLANTED RICE

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Abstract: The problem of weeds has become more acute due to the increased use of inputs such seeds of high yielding varieties (HYV's), fertilizer and irrigation in rice. A field experiment was carried out during winter seasons of 2005-06 at Bidhan Chandra Krishi Viswavidyalaya, Nadia (22.57°, 88.20°; 7.8 m latitude), India to find out an efficient and economic weed management practice in transplanted rice. The treatment comprised un-weeded control, hand weeding, bensulfuron methyl 0.6% + pretilachlor 6% GR (52.5 + 525, 60 + 600 (X), 75 + 750, 120 + 1200 (2X) g a.i./ha), pretilachlor 50% EC at 600 g a.i./ha, and bensulfuron methyl (Londax®60% DF) at 60 g a.i./ha. The predominant weed flora in the experimental fields were *Echinochloa colona, E. crus-galli* and *Cynodon dactylon* as grasses; *Cyperus iria, C. difformis, C. rotundus,* and *Frimbristylis spp.* among sedges; and *Enhydra fluctuans, Ludwigia perviflora, Marselia quadrifolia* and *Sphenoclea zeylanica* among broad leaf weeds. The density and dry weight of weeds decreased significantly at 30-60 days after treatment (DAT) in the plots treated with bensulfuron methyl 0.6% + pretilachlor 6.0% GR, irrespective of the dose used. This herbicide mixture yielded 1.47 t more per hectare than the untreated control when used at 52.5 + 525 g a.i./ha. A higher cost-benefit ratio was observed in this treatment followed by plots treated only with bensulfuron methyl (Londax®). No phytotoxicity has been observed on the test variety IET 4786 (Shatabdi) at all doses of bensulfuron methyl 0.6% + pretilachlor 6.0% GR.

Key words: Bio-efficacy, herbicide mixture and rice

Introduction

Growth requirement of crops and weeds are identical. It is well known that light, water, space and mineral nutrient are the four important factors for which weed compete with the crop. In addition to that some weeds act as alternate host of insect pests and diseases and exude toxic materials that are detrimental for crop growth, resulting in heavy reduction in crop yield. Weed problems have become more acute due to increase in the use of inputs such as seeds of high yielding varieties (HYV's), fertilizer and irrigation. Estimated loss of yield due to weeds is as high as 15-30% in transplanted rice. Although transplanting gives rice a head start over direct seeded condition, as weeds have wide adaptability to compete with crops, it is necessary to invent potential devices for enabling the crop to fight against the awesome challenge of obnoxious harmful weeds. Over the few decades chemical herbicides have dominated weed management options available in developing countries including India and the share of herbicides to total pesticide market is increasing (Yaduaraju *et al.* 2005). Hence, a study was conducted to find out a suitable and economically viable weed management measure in transplanted rice fields.

Materials and Methods

The field experiment was carried out during winter seasons of 2005-06 at the Instructional Farm of Bidhan Chandra Krishi Viswavidyalaya, Jaguli (22.57°, 88.20°; 7.8m latitude), Nadia, W.B., India. The experimental soil was clay loam with pH 6.9. The soil nutrient status of the experimental is given in Table 1.

Table 1. Soil nutrient status of the experimental site

Parameter	Unit
Organic Carbon (%)	0.58
Total Nitrogen (kg/ha)	1282
Phosphorus (kg/ha)	15.52
Potassium (kg/ha)	130.63

The rice variety used in the experiment was IET 4786 (Satabdi) and fertilized with 120:60:60 kg N: P_2O_5:K_2O per ha, respectively. The experiment was laid out in Randomized Complete Block Design with nine treatments and three replicates. The treatment comprised an un-weeded control, hand weeding, bensulfuron methyl 0.6% + pretilachlor 6% GR (52.5 + 525, 60 + 600 (X), 75 + 750, 120 + 1200 (2X) g a.i./ha), pretilachlor 50% EC at 600 g a.i./ha, bensulfuron methyl (Londax 60% DF) at 60 g a.i./ha. The required quantity of commercial formulation was mixed with adequate quantity of sand and broadcasted uniformly for treatments T_1, T_2, T_3, T_6 and T_7 whereas for other treatments (T_4 and T_5) the required quantity of commercial formulation was applied by using a knapsack sprayer fitted with a WFN 040 nozzle with a spray volume of 500 l/ha. All other recommended package of practices was followed uniformly to raise the crop. Weed samples were collected from a quadrate size of 0.5 m x 0.5 m at 30 and 60 days after transplanting (DAT) for assessing weed density and weed dry weight. All the data under observation were subjected to statistical analysis.

Results and Discussion

Predominant weed flora

The predominant weed flora in the experimental fields were *Echinochloa colona, E. crus-galli, Cynodon dactylon* among grasses; *Cyperus iria, C. difformis, C. rotundus, Frimbristylis spp.* among sedges; *Enhydra fluctuans, Ludwigia perviflora, Marselia quadrifolia* and *Sphenoclea zeylanica* among broad leaf weeds.

Effect on density and dry weight of weed

Weed density and total dry weight of weeds varied significantly due to the weed control treatments (Table 2). All the mixed herbicide, bensulfuron methyl 0.6% + pretilachlor 6.0% GR treatments recorded significantly lower total weed population than the standard herbicide treatments, *i.e.* pretilachlor 50% EC, bensulfuron methyl (Londax® 60% DF), butachlor 5% G and untreated control. The dose of mixed herbicide, bensulfuron methyl 0.6% + pretilachlor 6.0% GR at 52.5 + 525 g a.i./ha, 60 + 600 g a.i./ha and 75 + 750 g a.i./ha, were equally effective against all the species *viz. E. colona, C. dactylon, C. iria, C. rotundus, Frimbristylis spp., L. perviflora, M. quadrifolia* and *S. Zeylenica,* etc. and statistically, they were in par with each other.

Furthermore, bensulfuron methyl 0.6% + pretilachlor 6.0% GR @ 52.5+525 g a.i./ha, 60 + 600 g a.i./ha and 75 + 750 g a.i./ha were superior ($p<0.05$) over the tested standards *viz.* pretilachlor 50% EC, bensulfuron methyl (Londax®60% DF), butachlor 5% GR and untreated control. The density of weeds decreased significantly at 30 and 60 DAT in the plots treated with bensulfuron methyl 0.6% + pretilachlor 6.0% GR irrespective of the dose used. The highest density of weeds per m^2 area was observed in the un-treated plot, followed by butachlor 5% GR, bensulfuron methyl (Londax® 60% DF), pretilachlor 50% EC and bensulfuron methyl 0.6% + pretilachlor 6.0% GR at 30 and 60 DAT. Similar results were also reported by Rao and Singh (1997).

Table 2. Bioefficacy of Bensulfuron methyl 0.6% + Pretilachlor on weed density and dry weight in rice during Rabi 2005-06

Treatments	Dose (g. a.i./ha)	Total weed density (no /m^2)						Total weed dry weight (g /m^2)	
		Broad leaf		Sedges		Grass			
		30 DAT	60 DAT	30 DAT	60 DAT	30 DAT	60 DAT	30 DAT	60 DAT
Bensulfuron methyl 0.6%+ Pretilachlor 6% G	52.5 + 525	6.00	3.70	6.30	3.00	6.30	3.70	7.0	7.66
Bensulfuron methyl 0.6%+ Pretilachlor 6% G	60 + 600	4.70	2.30	5.00	2.30	5.70	3.40	6.1	5.38
Bensulfuron methyl 0.6%+ Pretilachlor 6% G	75 + 750	3.70	1.00	4.70	0.97	4.30	0.30	4.9	4.31
Pretilachlor 50% EC	600	37.60	49.50	27.30	50.75	9.60	14.70	19.5	30.6
Bensulfuron methyl (Londax®60% DF)	60	12.00	13.00	10.60	11.30	41.60	43.40	10.3	15.83
Butachlor 5% G	200	36.90	52.95	33.00	55.97	15.70	51.00	24.1	31.66
Hand weeding	-	7.03	5.70	5.30	3.00	4.60	4.30	6.8	9.70
Untreated control	-	58.00	63.97	42.00	66.00	56.00	78.00	43.7	76.75
CD at p=0.05		2.50	3.30	2.25	1.91	2.63	2.86	2.34	3.90

Among the weed control treatments, application of bensulfuron methyl 0.6% + pretilachlor 6.0% GR at 52.5+525 g a.i./ha gave significantly lower total dry weight at 30 and 60 DAT as compared to pretilachlor 50% EC, bensulfuron methyl (Londax® 60% DF), butachlor 5% GR and the untreated control. The reduction in weed dry weight in plots treated by bensulfuron methyl 0.6% + pretilachlor 6.0% GR at 52.5 + 525 g a.i./ha was non significant even at the higher doses.

Effect on crop phytotoxicity

None of the treatments of bensulfuron methyl 0.6% + pretilachlor 6.0% GR at 52.5 + 525, 60 + 600 (X), 75 + 750 and 120 + 1200 (2X) g a.i./ha showed any of the phytotoxicity symptoms on rice crop.

Effect on crop yield and yield attributes

The number of effective tillers/m^2, number of filled grains/panicle, and thousand grain weight were higher in the plots treated with the herbicide mixture the rest of the treatments, including untreated plot. The yield data presented in the Table 3 clearly envisages a significant impact of bensulfuron methyl 0.6% + pretilachlor 6.0% GR at 52.5 + 525 g a.i./ha over pretilachlor 50% EC, bensulfuron methyl (Londax® 60% DF), butachlor 5% GR and untreated control. The treatment bensulfuron methyl 0.6% + pretilachlor 6.0% GR produced 1.47 t more yield per hectare than the untreated control when used at 52.5 + 525 g a.i./ha. Increasing the dose in the mixture to 75 + 750 g a.i./ha did not show any significant increase in yield. Banerjee *et al.* (2005) also reported that pre-emergence application of butachlor + butachlor mixture proved beneficial for successful cultivation of paddy.

Effect on monetary returns

A higher cost-benefit ratio was observed in plots treated with bensulfuron methyl 0.6% + pretilachlor 6.0% GR treatments followed by bensulfuron methyl (Londax® 60% DF), pretilachlor 50% EC, hand weeding and butachlor 5% GR (Table 3).

Table 3. Bio efficacy of bensulfuron methyl 0.6% + pretilachlor on yield component, yield and cost: benefit ratio in rice during Rabi 2005-06

Treatments	Dose (g. a.i./ha)	No. of effective tiller/m^2	No. of filled grins/ panicle	Test weight (g)	Grain yield (t/ha)	Straw yield (t/ha)	Additional yield over control	Cost: benefit ratio
Bensulfuron methyl 0.6% + Pretilachlor 6% G	52.5 + 525	352.8	86.3	22.4	4.51	5.19	8820	1:7.8
Bensulfuron methyl 0.6% + Pretilachlor 6% G	60 + 600	364.3	86.9	22.6	4.68	5.22	9840	1:7.9
Bensulfuron methyl 0.6% + Pretilachlor 6% G	75 + 750	390.3	90.2	22.8	5.00	5.24	11760	1:7.96
Pretilachlor 50% EC	600	325.8	83.1	22.0	3.61	4.81	3420	1:3.3
Londax 60% DF (Bensulfuron methyl)	60	335.6	86.2	21.8	3.80	5.18	4560	1:5.7
Butachlor 5% G	200	314.7	85.2	21.6	3.40	4.74	2160	1:2.2
Hand weeding	-	341.3	85.5	22.3	4.31	5.06	7620	1:2.4
Untreated control	-	306.5	81.3	21.0	3.04	3.85	-	-
CD at p=0.05		8.6	0.43	NS	0.56	0.05	-	-

The results conclude that the mixture herbicides, bensulfuron methyl 0.6% + pretilachlor 6.0% GR was the most effective in controlling all the weed species found in the experimental plots, and significantly increased the yield at the dosage of 52.5 + 525 g a.i./ha. No phytotoxicity symptoms were observed on the test variety IET 4786 (Shatabdi) at all the doses of this herbicide mixture. Hence, bensulfuron methyl 0.6% + pretilachlor 6.0% GR at the rate of 52.5 + 525 g a.i./ha, used as a pre-emergent application can be recommended for the management of weeds in transplanted rice.

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ESTIMATION OF SEED PRODUCTION BY PLANT SIZE IN UMBRELLA SEDGE (*Cyperus iria* L.) AND GOOSE WEED (S*phenoclea zeylanica* Gaertn.)

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Abstract: Estimations of seed production depending on plant size were investigated in Umbrella sedge (*Cyperus iria* L.) and Goose weed (*Sphenoclea zeylanica* Gaertn.), representative annual weeds in paddy fields in the tropics. *Cyperus iria* was collected at Batalagoda, Sri Lanka on July 1985 and *S. zeylanica* was collected from Kumamoto, Japan on August 1990. As for umbrella sedge, number of spikelet including ten seeds per inflorescence (Y) was estimated by total length of rachis (X cm) as Y=12.8X+107.8 (r^2=0.683) for matured inflorescence and Y=10.4X+52.5 (r^2=0.718) for immature inflorescence. As for Goose weed, 1 cm of inflorescence produced approximately eleven capsules, including 120-230 seeds and no inflorescence was observed in plants below 20 cm in height. Relationship between estimated length of total inflorescence (Y cm) and plant height (X cm) was given by Log (Y) = 0.0349X-1.321 (r^2=0.803). These equations provide a rough estimate of seed production in both weeds though further investigations such as seasonal changes and intraspecific variation should be necessary.

Key words: Goose weed (*Sphenoclea zeylanica* Gaertn.), seed production, umbrella sedge (*Cyperus iria* L.)

Introduction

Number of seeds produced by a plant is the basic information required in studying the biology of weeds as it greatly affecting the size of seed bank in the soil. The amount of seed fluctuates depending on the plant size of weed. This is known as plasticity which is an essential characteristic of weed. In order to establish the simple and practical methods to estimate the amount of seed production, it is necessary to accumulate data on the relationship between plant size and seed production. For this purpose, estimations of seed production depending on plant size were investigated in Umbrella sedge (*Cyperus iria* L.) and Goose weed (*Sphenoclea zeylanica* Gaertn.), which are noxious annual lowland weeds in the tropics.

Materials and Methods

Umbrella sedge (Cyperus iria L.)
Eighty nine inflorescences of umbrella sedge (*C. iria*) were collected randomly from paddy fields and nursery beds in the Rice Research and Development Institute at Batalagoda, Sri Lanka on July 1985. Samples were divided into two groups, 41 matured inflorescences with complete flowering, and 48 immature inflorescences before flowering. Number and length of rachis branches, and total number of spikelets including those occurring directly from the base of inflorescence were measured as shown in Plate 1.

Goose weed (Sphenoclea zeylanica Gaertn.)
Samples of Goose weed (*S. zeylanica*) were collected from paddy fields infested severely by this naturalized weed in Kashima Town, Kumamoto Prefecture in the middle of August 1990. The number and length of inflorescence, number of capsule, floret and flower buds (Figure 1) were measured with the samples using a quadrate of 60 cm x 60 cm. Number of seeds in a capsule was determined with a zoom stereo microscope.

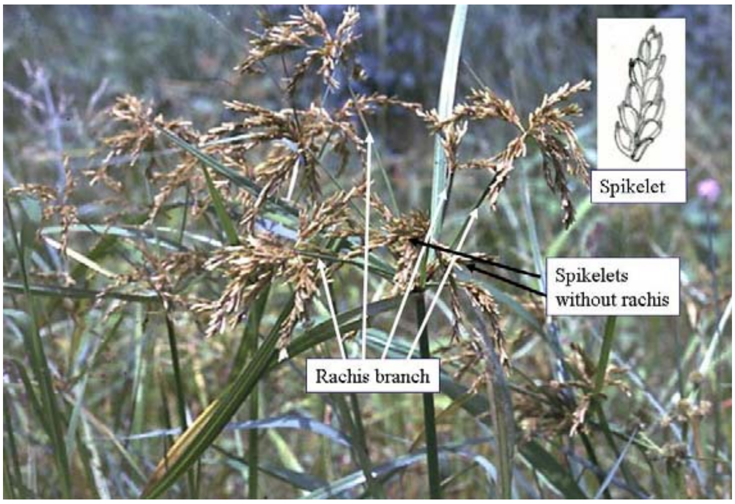

Plate 1. Composition of inflorescence of *C. iria* for measurement

Figure 1. Composition of inflorescence of *S. zeylanica* for measurement

Results and Discussion

Umbrella sedge

The correlation coefficients of the simple linear regression (Y=aX+b) were obtained between the total number of spikelets (Y) and number of rachis branches, length of the longest rachis branch, number of spikelet on the longest rachis branch, and total length of rachis branch (X), respectively, for both groups of inflorescence as shown in Table 1.

The highest coefficient of correlation for total number of spikelet was obtained with number of spikelet on the longest rachis branch for matured and immature inflorescence. In the inflorescence with few rachis branches, spikelets on the longest rachis branch occupied the greater part of total number of spikelet. Total length of rachis branch showed second significance. Total length of rachis branch can be used to estimate the number of spikelet which includes ten seeds approximately, considering the simple and practical measurement. It was reported that one plant of umbrella sedge produced 1930 seeds (Kasahara, 1968). It could be supposed that the sample had 6.6 cm of total rachis branch length belonging to small size inflorescence group because the biggest inflorescence had 80.9 cm of total rachis branch with 1141 spikelets in this investigation.

Table 1. Relationships between total number of spikelet and several parts of inflorescence in *C. iria*

Inflorescence	Part of inflorescence	A	B	r^2
Mature	Number of rachis branches	158.5	-460.7	0.635
	Longest rachis branch	61.6	-8.0	0.611
	Number of spikelet on longest rachis branch	5.5	-26.7	0.938
	Total length of rachis branch	12.8	107.8	0.683
Immature	Number of rachis branches	148.2	-410.5	0.607
	Longest rachis branch	95.2	-85.9	0.688
	Number of spikelet on longest rachis branch	5.13	24.1	0.956
	Total length of rachis branch	10.4	52.4	0.718

Goose weed

Density of Goose weed in severely infested paddy field was 323 plants m^{-2} (ranging from 186 to 528 plants m^{-2}), which is the average value for three quadrates. The investigations were carried out at the early ripening stage of rice plant. Since Goose weed came into bloom at this season, it was considered that this invasive weed could produce seeds required to naturalize in the central Kyushu. The number of inflorescences on a plant was measured with 242 plants in the quadrate Nos. 1 and 2. In the plants exceeding approximately 20 cm in height, single inflorescence was observed and multiple inflorescences were observed in those exceeding 50 cm in height (Figure 2).

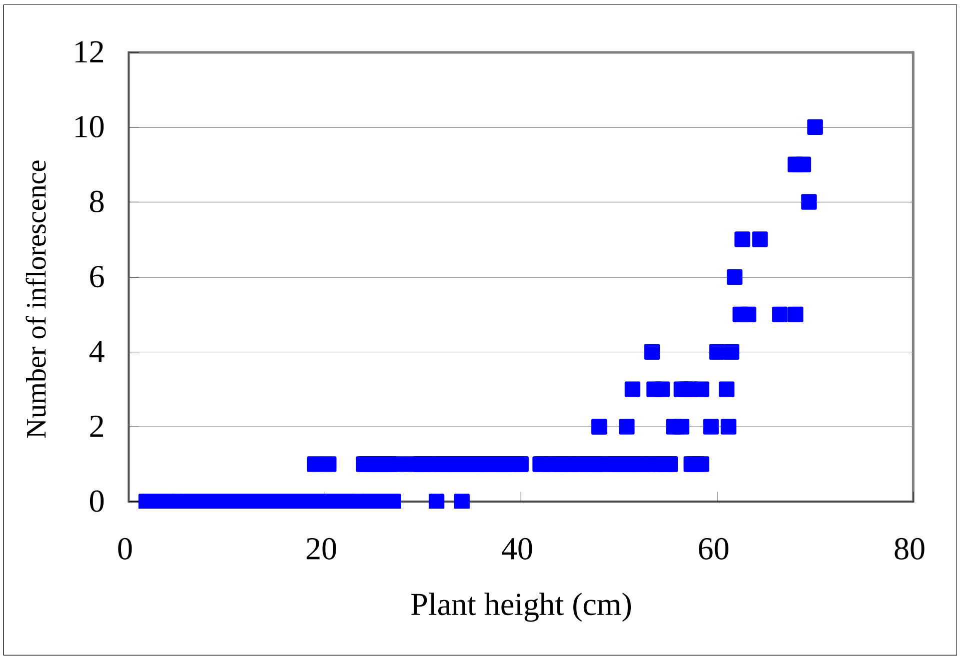

Figure 2. Relationship between plant height and number of inflorescence in *S. zeylanica* at the middle of August in Kumamoto Prefecture

Inflorescence of Goose weed is a dense spike of botrys type, producing florets in its tip as its elongation. The capsule, flower, flower bud (which could be determined with the naked eye) and developing flower bud were found in the inflorescence at the time of investigation as illustrated in Figure 2. A significant linear regression (Y=12.46X-1.34; r²) was obtained for total number of capsule, flower and flower bud (Y) with the length of inflorescence (X) as shown in Figure 3. It was calculated that one centimeter of inflorescence could produce approximately eleven capsules.

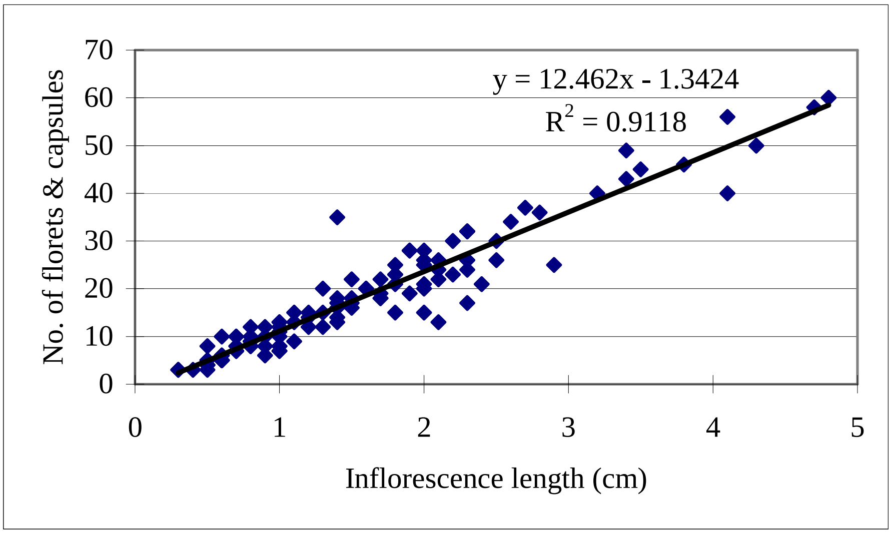

Figure 3. Relationship between length of inflorescence and total number of capsule, flower and flower bud in *S. zeylanica* at the middle of August in Kumamoto Prefecture

The number of seeds was measured with several capsules from the lower part of inflorescence as shown in Plate 2. The number of seed in a capsule ranged from 120 to 230.

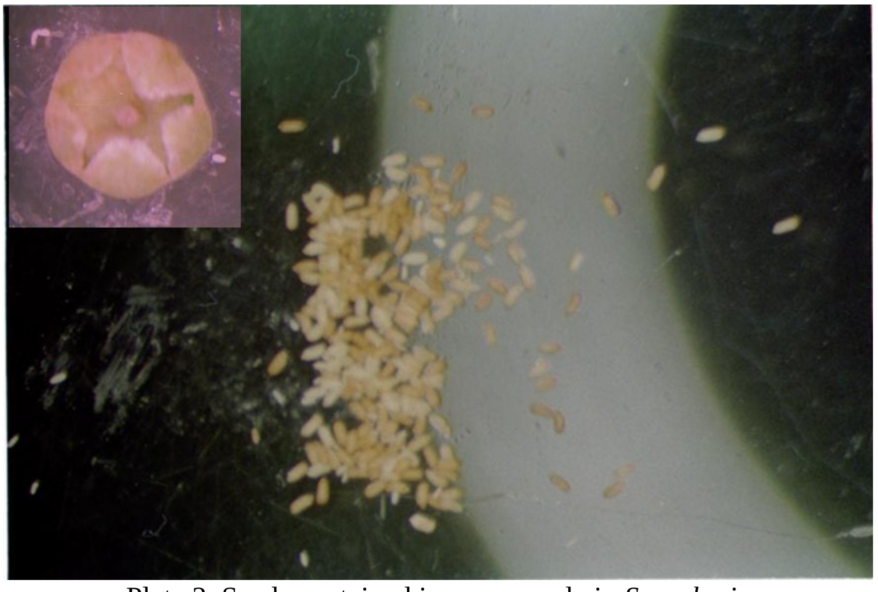

Plate 2. Seeds contained in one capsule in *S. zeylanica*

According to above measurements, the maximum number of Goose weed seeds produced in the transplanted paddy fields severely infested by the weed in the Kumamoto Prefecture of Japan was estimated as four million m^{-2} assuming that each of the 323 plants produced one inflorescence of 4.7 cm in length. Furthermore, the relationship between estimated length of total inflorescence (Y cm: length of longest inflorescence *x* number of inflorescence in a plant) and plant height (X cm) was given by an equation, Log(Y) =0.0349X-1.321 (r^2=0.803). This equation would provide the possible size of inflorescence for various plant sizes of the weed growing among rice plants in paddy fields. Goose weed produces the smallest seed at a seed weight of 0.0089 mg, among 103 major weed species found in Thailand (Noda *et al*. 1984). However, estimations made considering the plant size crucial for understanding the seed production dynamics, especially in a weed species producing numerous minute seeds such as Goose weed.

In conclusion, these mathematical expressions stated in this paper would provide rough estimates of seed production in both weeds. Though further investigations such as seasonal changes and intraspecific variation should be needed for better understanding of the seed production dynamics of a species, the results of the present study highlights the importance in considering the growth pattern.

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CURRENT STATUS OF THE HERBICIDE CONTAMINATION OF NEW ZEALAND AQUIFERS

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Abstract: Herbicides remain an important input into agricultural production in New Zealand. Although the total herbicide use now is considerably less compared to that in the 50's, their use rose by 25% between 1994 and 2004, and in 2004 herbicides sales accounted for 65% of the $210 million value of total pesticide sales. Surveys of use patterns have repeatedly identified the pastoral farming sector as the largest consumer of herbicides, with synthetic auxins and glyphosate being the major contributors. The substantial economic benefits arising from the use of herbicides in New Zealand have been questioned in recent years as a result of non-target spray drift damage and contamination of water resources. Contamination of groundwater has now become a global concern. The status of New Zealand aquifers, with respect to herbicide contamination, has been systematically monitored since 1990. In this paper all available publications and reports from Local Bodies and research results from the science community are summarised and critically reviewed. Some herbicides have been detected in drinking water – the concentrations however have been very low. On the other hand, groundwater contamination by herbicides has been detected in most regions of New Zealand, particularly at high risk locations (e.g. high groundwater, proximity to open water bodies, free draining soils). Thus far some 26 herbicide active ingredients have been found in New Zealand aquifers. Among these, triazines cause the most concern, with simazine showing the highest occurrence. Alachlor, bromacil and some phenoxy hormones also had a high frequency of detection. Despite the number of detections, however, they exceeded the MAV (Maximum Acceptable Values) in less than 1% of sampled wells.

Key words: Aquifer, groundwater, herbicide contamination

Introduction

Herbicide use is of considerable financial benefit to farmers and no other method of weed management has been as cost- and time-effective. The benefits arising from the use of herbicides are sometimes overshadowed by concerns over negative environmental and health effects (Ministry for the Environment, 2002); the off-site transport and subsequent contamination of water resources being one of them. Groundwater contamination can be attributed to either point or non-point sources (Léon *et al.* 2001). The discharge of a discrete identifiable source, such as a waste pipe, is a point-source contamination, while transport processes through and over the soil surface such as soil surface runoff, interflow, preferential flow and leaching, and spray drift lead to non-point-source pollution. There are several reasons why any pesticide contamination of aquifers should be avoided. Firstly, groundwater is an important drinking water source in New Zealand with about 50% of community water supplies relying partially or completely on groundwater (Davies, 2001). Secondly, uncontaminated groundwater proves agricultural management systems to be environmentally sustainable, as required by our Resource Management Act, 1991. This has implications with respect to trade and non-tariff barriers. Last but not least, the potential environmental impacts of herbicides in water resources are manifold. Residues can adversely affect reproductive cycles in many aquatic organisms, cause greater susceptibility to diseases, and lead to behavioural and growth changes in aquatic plants and animal life. Ultimately, they can accumulate in the food chain and affect humans (Krieger *et al*., 2001).

In June 2006, New Zealand's food exports totalled more than NZ$ 12,100 million and logs and wood exports were valued at NZ$ 1,960 million (Statistics New Zealand, 2007). The New Zealand economy, with a merchandise export value of NZ$ 32,430 million (Statistics

New Zealand, 2007), depends on the use of agrichemicals to optimise its agricultural production and generate high quality products to meet international standards. The frequency of herbicide applications and the total amount applied are the most important factors for determining the risk of water contamination (Müller *et al*. 2002). In the year 2002/03 total pesticide sales were dominated by herbicides with 2,540 t out of a total amount of 3,892 t (Manktelow *et al.*, 2005). According to the FAO classification of pesticides, the three herbicide categories phosphonyls (1,048 t), phenoxy hormones (765 t) and triazines (209 t) accounted for 80% of the total herbicide use (Manktelow *et al.* 2005). Surveys of use patterns have identified the pastoral farming sector consuming the largest volume of herbicides. Given the importance of agriculture to the New Zealand economy and the prevalent use of herbicides in arable and pasture production systems, contamination of groundwater resources is likely. However, only a limited amount of published information is available on the state of New Zealand groundwater contamination by pesticides (Taylor *et al*. 1997).

This paper summarises past and recent survey results of herbicide contamination of water resources in New Zealand. The overall scope is to outline the current level, spatial distribution and trends of water contamination, and to discuss the implications of the findings.

Materials and Methods

The status of New Zealand aquifers with respect to herbicide contamination has been monitored since 1990. Here we summarise and critically review results published in scientific journals and documents available from regional councils. We surveyed all regional councils with regard to their monitoring practices under the Resource Management Act. The compiled information includes location of the wells, extent and objective of the monitoring programme, ratio of the positive results and detection limits. To understand the background of the contaminations, we compared detection frequencies with the intrinsic herbicide properties responsible for the environmental fate of herbicides, and summarised the results in the index 'Groundwater Ubiquity Score' (GUS). The GUS index is a screening approach developed to evaluate the leaching potential of pesticides (Gustafson, 1989). It takes into account their partition coefficient (K_{oc}) and the degradation half-lives in soil (DT_{50}): GUS = $\log(DT_{50})(4 - \log(K_{oc}))$. As information on herbicide degradation and sorption in New Zealand soils is limited, these properties were taken from overseas data for calculating the GUS index. However, interactions of herbicides with New Zealand soils are expected to be different from those in overseas soils due to New Zealand soils' distinctive physico-chemically parameters (Rahman and James, 1995). Thus, local information was used, where available.

Results and Discussion

Four national assessments of pesticides in groundwater have been conducted since 1990 at intervals of four years (Close, 1993; Close, 1996; Close and Rosen, 2001; Close and Flintoft, 2004). Some more intensive monitoring on a regional basis has been undertaken by a number of regional and district councils following the previous national measurement campaigns (Table 1). These studies had different objectives and methods. The first regional study on pesticide monitoring was undertaken in 1975/76 in the Piako and Waihou rivers. Among the surveys carried out by regional bodies, the annual surveys by Environment Canterbury were the most extensive. Environment Waikato focussed on sites considered to be at high risk of contamination through either shallow groundwater and/or high pesticide usage and sampled about 20 selected wells at a relatively high temporal pattern over a period of three years (Hadfield and Smith, 1999).

Table 1. Overview of national and regional groundwater monitoring programmes

Regional Council	Level	Start	Number of sites	Sampling frequency	Target areas
Northland	national	1994	6	4-yearly	high risk
Auckland	national	1990	9 – 14	4-yearly	high risk
Waikato	national	1998	8	4-yearly	high risk
	regional	1995-98	20	quarterly	high risk
		1998	24	4 sites quarterly & 20 annually	most vulnerable
Manawatu	national	1994	8	4-yearly	high risk
	regional	1997	40	3-yearly	
Taranaki	national	1994	2	4-yearly	coverage#
	regional	1995	35	one-off	
		2002	6	one-off	
Wellington	national	1994	5	4-yearly	high risk
	regional	1996	14	one-off	
Hawkes Bay	national	1990	9 - >20	4-yearly	coverage#, risk
Bay of Plenty	national	1990	8 – 13	4-yearly	
Canterbury	national	1994	5	4-yearly	coverage# high risk
	regional	1989	50-150	annually	
		recently	< 5	annually	
Otago	national	1994	5 – 10	4-yearly	high risk
Southland	national	1994	3	4-yearly	
	regional	1999	10	twice in 2 years	
West Coast	national	1998	3	4-yearly	high risk

Selection of sites representing the entire region.

A synthesis of the results for herbicides detected, grouped according to their chemical categories (FAO, 1996), amount used and a mobility index is given in Table 2. The potential risks to humans implied by these findings can be only partially addressed by comparison to the standards established for drinking water by the Ministry of Health (2000). Concentrations of individual pesticides were generally low compared to these standards. Herbicides exceeded the Maximum Acceptable Value (MAV) only in less than 1% of sampled wells.
Five herbicides (atrazine, bromacil, cyanazine, MCPA, mecocrop) have been detected in concentrations higher than their MAV. In total 26 herbicides have been found. Among these compounds, triazines cause the most concern due to their high leaching potential. However, as the analysis for triazine herbicides is straightforward, most of the analysis protocols included them. Among single compounds, simazine showed the highest frequency of occurrence. High frequencies of detection for single compounds have also been noted for alachlor (12) and bromacil (14). Other categories of concern are phenoxy hormones, amides and uracils.

The GUS index demonstrates that the majority of herbicides detected in groundwater resources fall into the category ‘leaching’ (17 leachers). Almost 30% of the herbicides detected in groundwater are classed as ‘transitional’ or ‘improbable leachers’. This indicates that processes other than matrix flow are important for the contamination. Preferential flow and point sources need to be considered here. When locally available information on herbicide properties is taken into account, the index calculations changed the risk assessment for two compounds: 2, 4-D (L instead of T) and metribuzin (T instead of L).

Given the amount and type of herbicides used, land topography, rainfall pattern, and the variations in analytical methods, the wide variability seen in the survey results between the regions is not surprising. In general, groundwater contamination is more likely in shallow, unconfined aquifers with permeable soils and high groundwater recharge. This is supported by the results from Canterbury, where a high percentage of wells (87%) were shallow (< 30 m depth), and wells where herbicides had been found were shallower than 18 m. But herbicides have also been detected in deep wells, e.g. in the Pukekohe area frequent detection of the mobile and very persistent herbicide picloram (up to 0.9 μg l^{-1}) has been reported for a well that is 64.5 m deep and cased to 39.5 m (Hadfield and Smith, 1999).

Table 2. Synthesis of information on herbicide detections in groundwater

Chemical category (FAO)	Use[#] (t)	Herbicide	GUS*	Detections	Concentration range (μg l^{-1})	Mean (μg l^{-1})
Phenoxy hormones	746	2,4-D	2.70 (T)	4	0.05-0.9	0.27
		2,4,5-T	4.70 (L)	1	0.1	0.1
		MCPA	3.77 (L)	1	61	61
		MCPB	3.09 (L)	1	2.1	2.1
		mecoprop	3.57 (L)	5	0.51-420	85.36
Triazines	245	atrazine	3.56 (L)	45	0.01-37	1.97
		cyanazine	1.97 (T)	1	0.7	0.7
		hexazinone	4.43 (L)	33	0.04-0.24	0.14
		metribuzin	3.56 (L)	10	0.02-1.2	0.33
		propazine	3.86 (L)	42	0.01-2.5	0.52
		simazine	3.35 (L)	71	0.01-1.6	0.18
		terbuthylazine	2.95 (L)	71	0.01-2.0	0.40
Amides	67	alachlor	2.08 (T)	12	0.02-3.67	0.62
		metolachlor	3.32 (L)	5	0.036-4.5	0.98
Dinitroanilines	2	oryzalin	1.59 (IL)	1	0.19	0.19
		pendimethalin	0.59 (IL)	2	0.03-0.046	0.038
		trifluralin	0.17 (IL)	2	0.02-0.3	0.16
Urea derivates	15	diuron	2.58 (T)	10	0.03-9.5	3.44
Sulfonylurea		primisulfuron	3.40 (L)	1	2.7	2.7
Uracils	5	bromacil	4.44 (L)	14	0.02-6.37	1.41
		terbacil	4.70 (L)	4	0.08-6.05	3.38
Other hormone types	125	picloram	5.46 (L)	4	0.02-2.8	1.04
		triclopyr	4.49 (L)	4	0.02-200	67.61
Other herbicides	70	bentazone	3.21 (L)	3	0.015-0.18	0.12

[#] Holland and Rahman (1999); * GUS (Groundwater ubiquity score) >2.8: leacher (L); 1.8 to 2.8: transition (T); <1.8: improbabale leacher (IL).

The first National Survey (Close, 1993) included re-sampling of some of the wells where pesticides were detected. The results confirmed a high temporal variability of aquifer contamination. Environment Waikato (Hadfield and Smith, 1999) also assessed temporal variability in its intensive monitoring programme (Table 1). The highest overall concentrations were detected during summer months. This is consistent with findings from overseas, where herbicide levels in groundwater showed pronounced seasonal variability in agricultural areas, with maximum values often following spring applications (Felding, 1995).

In a few cases a relationship between herbicide detections and usage was noticeable. In Southland, the occurrences of several of the herbicides detected were linked to specific land uses. Propazine, terbuthylazine and hexazinone are or have been used in the Edendale Forest Nursery, and contamination of metribuzin was attributed to horticultural operations (potatoes) in the area (Hughes, 2000). Detections of bromacil were clearly linked in different regions to its non-agricultural use, *e.g.* along railway lines for long-term weed control (Hughes, 1997). High pesticide detections could often be traced back to poor management practices, particularly procedures for storage and mixing of chemicals prior to application and disposal and clean-up after application.

In general, reductions of pesticide concentrations in surface and groundwater require management strategies that focus on reducing chemical use and subsequent transport in the hydrologic system. Some of the mitigation strategies include (a) dissemination of the 'Best Management Practices' and improve training for pesticide users, (b) exploring alternatives to the use of pesticides (Integrated Pest Management), (c) reducing the frequency of spraying (re-visit spray schedules by the industry), (d) making more efforts toward organic production and associated research, (e) establishing management practices that reduce non-point source contamination, (f) establishing continuous pesticide container recycling programmes and collection systems for unwanted agricultural chemicals to prevent inappropriate storage, (g) improving packaging of pesticides to minimise waste production, (h) strengthening legal controls on handling waste, storage and transport of pesticides, (i) promoting better wash down/clean up facilities for growers and relocation of wells away from these areas, (j) encouraging regular checks for spraying equipment through Contractors' organisations, (k) ensuring better certification/licensing of pesticide sale, and (l) revision of the pesticide approval system in relation to high risk compounds.

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EFFECT OF HERBICIDES ON WEED POPULATION, BIOMASS, CHLOROPHYLL CONTENT AND SOIL MICROFLORA IN TRANSPLANTED SUMMER RICE (*Oryza sativa* L)

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Abstract: A field experiment was conducted during the period January to May 2002 and 2003 to study the effects of some herbicides on weed population, biomass, chlorophyll content and soil microflora in transplanted summer rice. The rice variety was IET 4786. Besides the non weeded control, hand weeding at 20 and 40 DAT, 4 applications of pyrazosulfuran-ethyl (PSE) 10 WP (20, 25, 50 and 100 g ha^{-1}) and 3 applications of acetachlor (100, 150 and 200 ml ha^{-1}) were used. All the herbicides were applied as pre-emergence herbicides. The predominant weed species were barnyard grass (*Echinochloa crus-galli),* ricefield flatsedge *(Cyperus iria),* water fern *(Marselia quadrifolia),* Enhydra *(Enhydra fluctuans) and* hoorah grass *(Fimbristylis littoralis)*. PSE proved better than acetachlor in controlling weeds. The highest grain yield of rice and lowest total weed population, biomass and chlorophyll content of weeds were recorded where PSE 10 WP at 100g/ha was applied. PSE has stimulatory and acetachlor has detrimental effects on soil beneficial microorganisms (N-fixing bacteria and P - solubilizer).

Key words: Summer rice, PSE 10 WP, acetachlor, weed management, soil micro-flora

Introduction

Rice (*Oryza sativa* L.), one of the most important staple food crops is subjected to heavy yield loss (41.8%) due to weed infestation. In general, the yield loss due to uncontrolled weed growth under Indian conditions ranged between 18-20% in transplanted rice. (Balasubramanian and Duraiswamy, 1996). The age old traditional cultural methods such as hand weeding or hoeing are slow and laborious and uneconomic. In recent years, farmers have developed inclination to use chemical methods of weed control (Mukhopadhyay, 1992). Though several herbicides for controlling weeds in transplanted rice have been recommended, the use of herbicides is quite limited due to lack of technology regarding rates, times and methods of application.

The indiscriminate use of synthetic herbicides for controlling weeds may offer potential hazards to users, consumers, live stock, wild life, soil micro flora and to other soil environments. Modern agricultural fields are generally treated with high rates of synthetic nitrogenous fertilizers, pesticides, and herbicides, which develop adverse effects on the soil chemical and biological environments. Hence, the present investigation was conducted to identify the effect of low rates of herbicides on weed population, biomass, chlorophyll content and soil microflora in transplanted summer rice.

Materials and Methods

A field experiment was conducted during the summer season of 2002 and 2003 at ‘C’ Block Farm (23°N latitude, 89°E longitude and 9.75 m above mean sea level), Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal. The soil of the experimental field was sandy clay loam in texture, pH 6.9, organic carbon 0.65%, total nitrogen 0.06%, available P_2O_5 20 kg ha^{-1} and available K_2O was 120 kg ha^{-1}. The experiment was laid out in a randomized block design with 3 replications. There were 9 treatments comprising of 4 rates of pyrazosulfuran-ethyl (PSE) 10 WP *i.e.* 20, 25, 50 and 100 g ha^{-1}, 3 rates of acetachlor *i.e.* 100, 150 and 200 g ha^{-1}, which were compared with hand weeding (at 20 and 40 days after transplanting - DAT) and an non weeded control. Both PSE and acetachlor were applied as pre-emergence herbicides at 6 and 7 days after transplanting, respectively. Yield was measured at harvest and the effects of the above treatments on soil microflora and chlorophyll content of weeds was studied.

Healthy seedlings of rice (CV IET-4786) were transplanted at a spacing of 20 cm x 10 cm in the 3rd week of January during 2002 and 2003 after applying 120 kg N, 60 kg P_2O_5 and 60 kg K_2O ha^{-1}. Half N, full of each P_2O_5 and K_2O were applied at planting and the balance N was top dressed at 4 weeks after transplanting. The recommended cultural practices and plant protection measures were adopted. Weed samples were collected from a quadrat of size 0.5 m x 0.5 m at 15, 30 and 60 DAT for assessing weed density and weed dry weight. Weed control efficiency was calculated with the formula.

$$\text{W.C.E. (\%)} = \frac{(X-Y)\, x\, 100}{X}$$

Where, X = Weed dry weight in control plot *i.e.* unweeded plot,
Y = Weed dry weight in treated plots

The enumeration of the microbial population was done on agar plates containing appropriate media following serial dilution technique and pour plate method (Pramer and Schmidt, 1964). The count was taken out 3rd and 5th day of incubation. Jensen's agar medium was used for containing aerobic non-symbiotic nitrogen fixing bacteria. Total number of phosphate solubilizing microorganisms was estimated in Pikovskaia's agar medium. For estimation of total chlorophyll content in weeds, optical density of weed samples were recorded at 645 and 663 nm with the help of spectrophotometer using 80% acetone and the amounts of chlorophyll were calculated with the equations-

$$\text{Total chlorophyll (mg/g)} = \frac{20.2\,(A645) + 8.02\,(A663)}{1000\, x\, W}\; x\, V$$

Where, V = final volume of chlorophyll extract in 80% acetone (ml)
W = fresh weight of the samples (g.)
A = absorbance at specific wavelengths

Results and Discussion

The predominant weed species infesting the experimental plots were barnyard grass (*Echinochloa crus-galli),* ricefield flatsedge *(Cyperus iria),* water fern *(Marselia quadrifolia),* Enhydra *(Enhydra fluctuans) and* hoorah grass *(Fimbristylis littoralis).* Weed density and total dry weight of weeds varied significantly due to the weed control treatments. All the weed control treatments recorded significantly lower total weed population than in the weedy check. Application of PSE at 100 g ha^{-1} developed significantly lower weed populations (1.31 to 13.28 m^{-2}) over the crop growth period. Weed populations in plots sprayed with Acetachlor at all three rates and PSE at lower rates were similar (Table 1).

Application of PSE at 100 g ha^{-1} produced significantly lower total dry weights (Table 1) when compared to the other treatments at 15, 30 and 60 DAT (0.078, 0.18 and 0.5 g m^{-2}, respectively). In general all plots that received herbicidal treatments produced a lower weed biomass than the non weeded control plots. Moorthy (1997) also observed that the application of PSE lowered the weed biomass and enhanced the grain yield.

Table 1. Effect of different weed control treatments on density and dry weight of weeds and weed control efficiency in summer rice (Mean data of 2 years)

Treatment	Weed density ($No.m^{-2}$)			Weed dry weight ($g\ m^{-2}$)			Weed control efficiency (%)		
	15 DAT	30 DAT	60 DAT	15 DAT	30 DAT	60 DAT	15 DAT	30 DAT	60 DAT
PSE 10 WP at 20g ha^{-1}	9.00	18.67	43.30	0.30	1.22	2.53	46.90	21.99	15.75
PSE 10 WP at 25g ha^{-1}	7.33	14.30	38.31	0.22	1.07	2.28	58.45	32.51	23.85
PSE 10 WP at 50g ha^{-1}	6.29	9.00	25.00	0.18	0.42	0.87	66.58	70.55	70.17
PSE 10 WP at 100g ha^{-1}	1.31	4.31	13.28	0.08	0.18	0.50	85.32	87.66	81.76
Acetachlor at 100 ml ha^{-1}	7.00	11.35	35.36	0.26	0.77	2.15	52.46	50.32	29.44
Acetachlor at 150 ml ha^{-1}	6.00	9.29	29.66	0.18	0.69	1.93	65.36	56.73	37.24
Acetachlor at 200 ml ha^{-1}	5.67	6.67	23.34	0.16	0.46	0.88	71.53	69.60	68.43
Hand Weeding (20 & 40 DAT)	14.61	6.00	33.00	0.52	0.30	1.23	5.40	82.54	57.56
Control (Weedy check)	22.71	31.35	52.31	0.55	1.54	2.97	0.00	0.00	0.00
CD (p=0.05)	1.52	1.19	2.36	0.09	0.59	1.02	15.08	9.84	12.14

PSE, Pyrazosulfuran-ethyl; DAT= Days after transplanting.

The highest weed control efficiency at all observations (85.32, 87.6 and 81.76%, respectively) were in the plots treated with PSE at 100 g ha^{-1}, followed by hand weeding at 20 and 40 DAT and PSE at 50g ha^{-1}. Application of PSE at all rates effectively reduced weed biomass at 15 DAT, and at 30 and 60 DAT, it was effective only when applied at 50g ha^{-1} or more. Similarly, acetachlor was effective only when applied at 150g ha^{-1} in all crop growth periods. PSE controlled the weeds more efficiently than acetachlor by reducing chlorophyll content of leaves and the effect is progressively more conspicuous with crop maturity. The lowest chlorophyll content was recorded in treatment where PSE was applied at 100 g ha^{-1} (Figures 1, 2, 3 and 4).

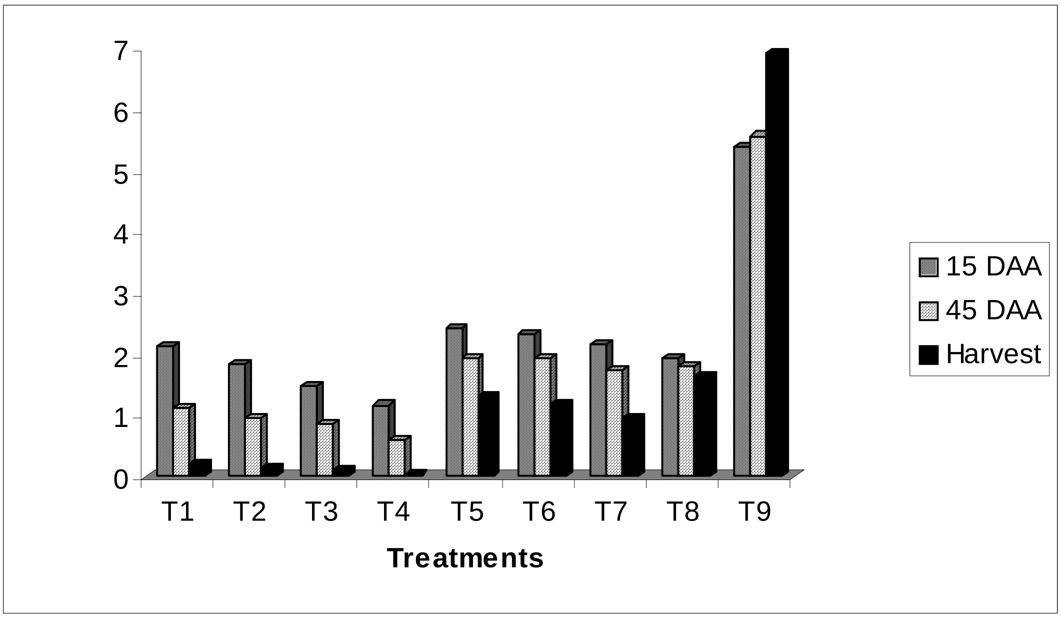

Figure 1. Bioefficacy of herbicides on chlorophyll content (mg g^{-1} of fresh weight) of *Echinochloa crus-galli* at different days after application.

Application of pyrazosulfuran-ethyl at 100g ha^{-1} produced the maximum grain yield (7.19 t ha^{-1}) but maximum straw yield (11 t ha^{-1}) was recorded in treatment where hand weeding was done at 20 and 40 DAT. Minimum grain and straw yield (3.37 and 6.93 t ha^{-1}, respectively) were found in un-weeded control plot. All the treatments where herbicides were applied and also hand weeding recorded significantly higher grain yield when compared to the non - weeded control. This was in agreement with the earlier findings of Mukhopadhyay *et al* (1990).

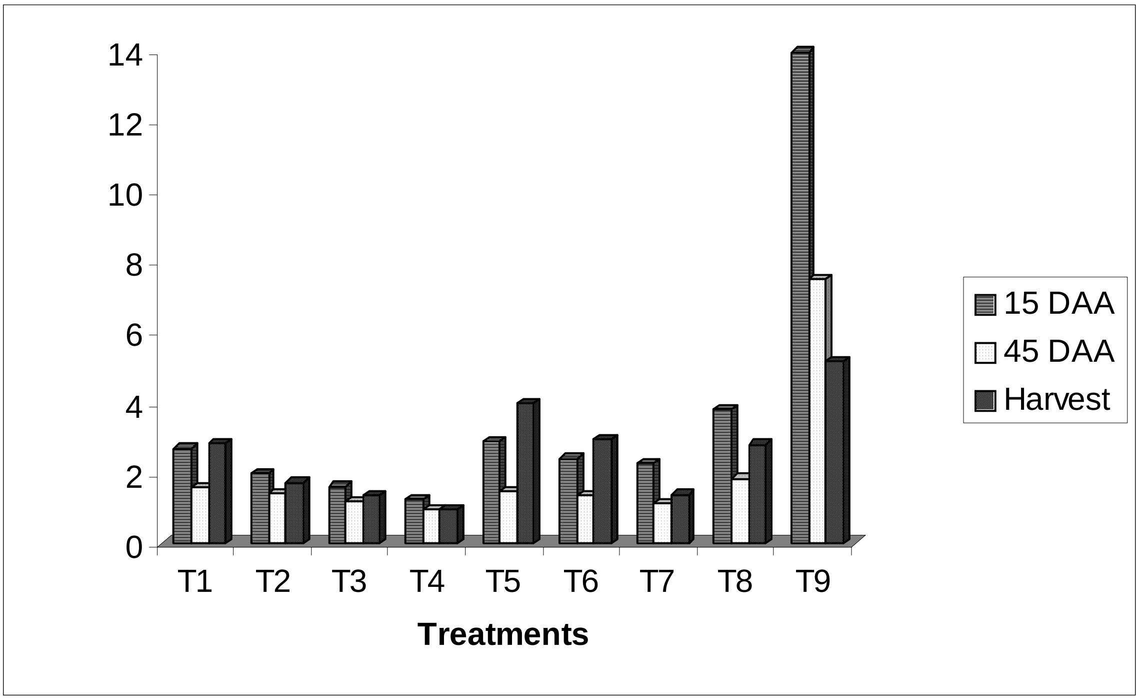

Figure 2. Bioefficacy of herbicides on chlorophyll content (mg g^{-1} of fresh weight) of *Enhydra fluctuans* at different days after application

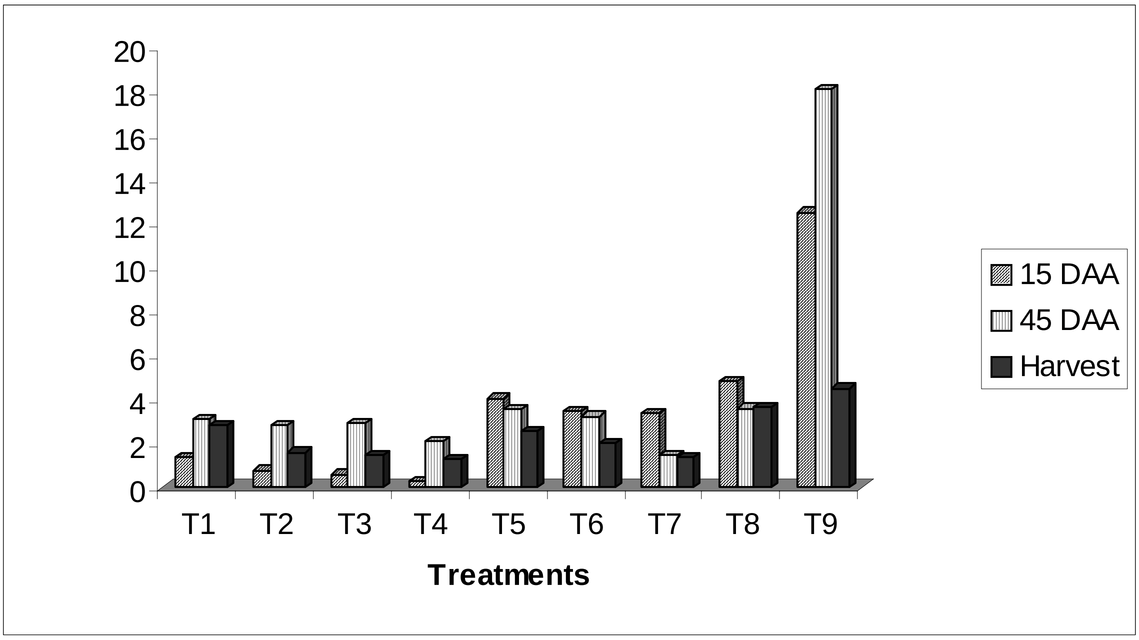

Figure 3. Bioefficacy of herbicides on chlorophyll content (mg g^{-1} of fresh weight) of *Cyperus iria* at different days after application

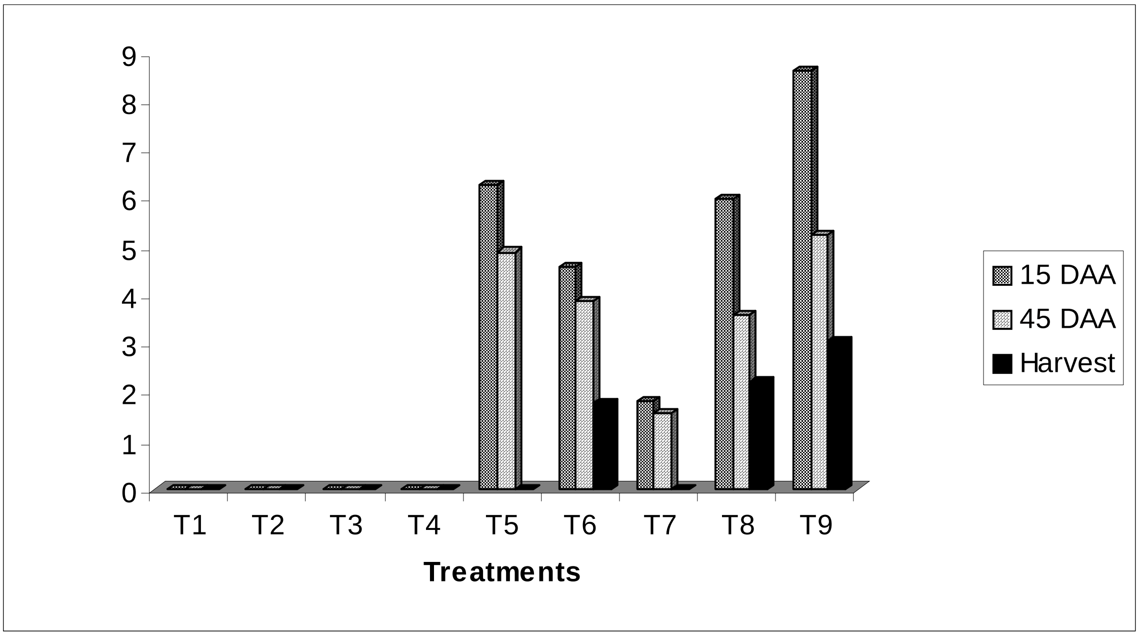

Figure 4. Bioefficacy of herbicides on chlorophyll content (mg g^{-1} of fresh weight) of *Marselia quadrifolia* at different days after application

High levels of pyrazosulfuran-ethyl caused a significant enhancement in the proliferation of both non-symbiotic nitrogen fixers and phosphorus solubilizing microbes in the soil rhizosphere over that of the soils in non weeded plots of summer rice at each stage of sampling (Table 2).

Table 2. Effect of different weed control treatments on rice yields and soil micro flora CFU X 10^4 g^{-1} (Mean data of 2 years)

Treatment	Grain yield (t ha^{-1})	Straw yield (t ha^{-1})	Non symbiotic N fixing bacteria			P - solubilizing bacteria		
			30 DAT	45 DAT	60 DAT	30 DAT	45 DAT	60 DAT
PSE 10 WP at 20g ha^{-1}	5.19	7.15	180.5	193.5	215.0	19.0	35.0	43.5
PSE 10 WP at 25g ha^{-1}	5.23	8.05	212.0	222.0	232.0	21.5	59.0	64.5
PSE 10 WP at 50g ha^{-1}	6.20	8.41	226.0	233.0	248.5	25.5	59.6	75.0
PSE 10 WP at 100g ha^{-1}	7.19	9.11	237.0	249.5	266.0	31.5	63.5	82.5
Acetachlor at 100 ml ha^{-1}	5.70	8.20	173.5	262.5	151.0	16.0	13.5	11.0
Acetachlor at 150 ml ha^{-1}	6.02	8.20	157.5	149.0	140.0	19.0	10.0	8.0
Acetachlor at 200 ml ha^{-1}	6.30	9.07	136.0	127.5	116.0	9.0	9.0	5.0
Hand Weeding (20 & 40 DAT)	6.11	11.00	157.0	163.0	173.5	13.5	23.5	36.0
Control (Weedy check)	3.73	6.93	178.0	180.5	212.5	18.5	30.5	39.0
CD (p=0.05)	1.26	2.11	10.0	15.84	11.80	5.84	5.25	5.63

There was however a progressive increase in the population of non-symbiotic N-fixing bacteria and P solubilizer in the soil rhizosphere of summer rice by increasing the application of PSE from 20 g ha^{-1} to 100 g ha^{-1}. The highest microorganism population was recorded when PSE was applied at 100 g ha^{-1}. On the other hand, acetachlor exerted harmful effects on the population of non-symbiotic N fixing bacteria and phosphorus solubilizing bacteria in the soil rhizosphere.

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THE CONTRIBUTION OF RICE SEED TRAITS TO EARLY GROWTH AS AN INDICATOR OF COMPETITIVENESS

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Abstract: Previous studies suggest that rice seedling vigor, as characterized by early growth, is a good indicator of a rice cultivar's ability to compete against weeds. To identify those characteristics that contribute to seedling vigor, seeds of 20 diverse rice cultivars were studied and grown in greenhouse experiments. Seed weight (SW), embryo length, and endosperm length differed ($p<0.001$) between cultivars. Embryo length and endosperm length were positively related to SW, as was leaf area at 6 DAS (LA6). Larger embryos were associated with the shorter intervals to the period at which total seedling dry matter regained the initial seed weight (RSW) after germination. Differences in the interval to RSW, which commenced shortly after 6 DAS for many cultivars, were closely related to LA6.

Key words: embryo size, endosperm length, seedling vigor, seed weight

Introduction

Weeds are a major constraint in direct-seeded rice where competition for resources between rice and weeds is common since rice establishment is almost always accompanied by weed recruitment. With direct seeding (DS), good crop establishment and achieving an appropriate plant population density is important to attain good yields. The attribute of good seedling vigor could contribute to this. Further, it is essential that rice cultivars grown for DS are competitive against weeds in temperate (Redoña *et al.* 1996) or in tropical regions (Widawsky and O'Toole, 1996).

Studies (Ni *et al*. 2000; Perez de Vida *et al.* 2006) indicate that early vigorous growth enhances the ability of rice to compete with weeds and that substantial differences exist between genotypes (Redoña *et al.* 1996). Others have suggested that seedling growth could be used as an indicator in screening for competitiveness (Zhao *et al.* 2006). Some studies have focused on rice seedling growth traits but not seed size and component traits such as endosperm and embryo sizes or seed changes that immediately follow germination and prior to photosynthetic stage. Pandey *et al.* (1994) reported that embryo weight and length in indica rice cultivars were correlated with seed and seedling vigor. Further, he suggested that rice embryo length can be used to screen for embryo size since the two are strongly correlated. In barley, the size of seed has been shown to be positively related to early growth (Kaufmann and Guitard, 1967).

Understanding the relations between rice seed traits and early seedling growth will be of benefit to plant breeders by enabling them to select for rice cultivars that have greater seedling vigor and that are, in turn, likely to be more competitive with weeds. The objective of this study is to determine the relations between seed and seedling growth traits that are associated with seedling vigor with a view to using this as an indicator of competitiveness with weeds under field conditions.

Materials and Methods

Greenhouse experiments were conducted at the International Rice Research Institute in Los Baños, Laguna, Philippines on June 12-24, 2005 and April 23 to May 5, 2006.

Cultivars studied

Twenty rice (*Oryza sativa* L.) cultivars with a range of early vigor were identified from previous field experiments (Johnson and Cope, 2004, unpublished; Zhao, 2006). The cultivars comprised of modern lines, landraces, *aus, indica,* and *japonica* types and *O. sativa* x *O. glaberrima* interspecific hybrids to provide a wide range of genotypes. These cultivars were AUS196, Azucena, CT13377-4-2-M, CT6510-24-1-2, IR36, IR60080-46A, IR65907-116, IR66424-1-2-1-5, IR68702-072-1-4-B, IR77384-12-35-3-25-1-B, IR77384-12-35-3-25-3-B, IR77384-12-35-3-6-7-B, IR77384-12-35-B-22-4-B, Maravilha, NSG 19, PSBRc09, Sabita, SRN1, Vandana, and WAB56-125. Seeds were harvested from the previous season and had been stored in identical conditions.

Seedling establishment and measurements

Thirty three uniform and healthy seeds were surface-sown in rows 25 mm apart in plastic trays (336 mm x 260 mm x 115 mm) filled with sterilized soil. Seeds were then covered with a thin layer of sterilized soil. The trays, with eight completely randomized replicates, were stood in shallow water (ca.10 mm) to ensure an adequate supply of water. At 2, 4, 6, 8, 10, 12 days after sowing (DAS), seedlings were pulled out, washed carefully, placed in plastic bags, and then placed into an insulated box to prevent moisture loss and deterioration of the samples. Shoot length and root depth were measured with a calibrated steel rule. Leaf area was measured using LI-3100C area meter (LICOR, 4421 Superior St., Lincoln, NE, USA). Roots, culms, leaf blades, and seeds were detached and oven-dried separately at 70°C for five days. The number of days taken for the seedlings to reach the biomass equal to initial seed weight (RSW) were estimated from the changes in seedling dry mass over 12 days. We used total leaf area at 6 DAS (LA6) as an indicator of seedling vigor. Prior to sowing, eight replications of 100-seed lots were dried at 45 °C for 3 d. All dried seeds and plant samples were weighed in a semi-analytical balance AX205 (Mettler-Toledo, GmbH, CH-8730 Uznach, Switzerland).

For embryo and endosperm length measurements, twenty randomly selected mature seeds of each of the 20 cultivars from the same seed lot used for earlier experiments were dried at 45 °C for 3 d, de-hulled, and then glued onto glass slides in such a way that the portion of the seed where the embryo is located was most exposed. Embryo images were taken with an Olympus DP70 digital camera attached to a SZX7 microscope (Olympus Corporation, Tokyo, Japan) and their lengths measured using Image ProPlus 5.1 software (Media Cybernetics, Singapore).

Data analysis

A combined ANOVA was conducted on all parameters using Genstat version 7 (2004), the days to RSW were determined by regression of the total dry matter using SAS release 9 (SAS Institute Inc., 2005).

Results and Discussion

Results for five cultivars which are representatives of a range of vigor scores based on ground cover are shown in Table 1. The cultivars were scored for seedling vigor (1= least vigorous, and 5=most vigorous) in previous field observations, which showed that Maravilha (Zhao, 1996) was the least vigorous while Sabita and NSG19 (Johnson and Cope, 2004, unpublished) were the most vigorous. The rest had intermediate ground cover. The cultivars differed significantly ($p<0.001$) in their SW, embryo length, RSW, LA6. Sabita (30.8 mg) and NSG 19 (27.2 mg) had the heaviest seeds among the cultivars and incidentally both had high vigor scores. IR36 (21.5 mg) and Maravilha (22. 9 mg) had the lightest seeds. The embryo length of mature seeds ranged from 1.5 mm (AUS 196) to 1.8 mm (NSG 19, Sabita), while the endosperm length ranged from 5.8 mm (AUS 196) to 8 mm (Sabita). Pandey (1994) found moderate variations in embryo length among and within indica and japonica rice cultivars but mean embryo length was higher in the latter.

Table 1. Cultivar differences in seed traits (mean and $LSD_{0.05}$ values are for the 20 cultivars)

Cultivars	Seedling vigor[a]	Seed weight (mg)	Embryo length (mm)	Endosperm length (mm)	RSW[b] (day)	LA6[c] (cm^2)
AUS196	4.3	26.1	1.5	5.8	6.5	1.65
IR36	4.5	21.5	1.6	6.8	6.6	1.27
Maravilha	2.5	22.9	1.7	7.1	7.3	0.82
NSG 19	4.8	27.2	1.8	7.2	6.5	2.20
Sabita	5	30.8	1.8	8.0	6.3	2.54
Mean	-	23.9	1.6	6.9	6.7	1.42
± $LSD_{.05}$	-	0.866	0.063	0.161	0.404	0.191

[a] Seedling vigor: 1= least vigorous, 5 = most vigorous, [b] RSW = Regain seed weight = number of days taken for the seedling to attain biomass equal to initial seed weight. [c] LA6= leaf area at 6 DAS.

Most of the cultivars reached RSW shortly after 6 DAS and while they had started to photosynthesize they continued to utilize seed reserves. It was not however determined whether reserves available after RSW were used for growth or for respiration. Our data indicated that most of the cultivars had not completely exhausted their seed reserves until 8 DAS or a few days after RSW commenced suggesting that the remaining seed reserve may be in excess (McWilliam et al., 1970) of the seedling requirement. RSW was longest for Maravilha (7.3 d) and shortest for Sabita (6.3 d).

Table 2 shows the correlations among seed traits in five selected rice cultivars. Seed weight was associated with both embryo length and endosperm length (r = 0.461* and 0.426*, respectively) and was strongly correlated with LA6 (r=0.785**). Krishnasamy and Seshu (1989) reported that the positive effect of seed weight on total dry matter at 14 DAS was due to a contribution of a larger amount of food reserves available in bigger seed. The positive correlation (r = 0.506*) between embryo and endosperm lengths in this study is similar to the results of Pandey (1994) in indica rice. A more pronounced embryo size – grain weight relationship was reported by Richards and Lukacs (2002) in wheat. Bremner (1963) suggested that a compounded linear effect of embryo and endosperm sizes was responsible for large plants in wheat.

Table 2. Correlations among seed and seedling growth traits up to 12 DAS

	Embryo length	Endosperm length	RSW	LA6
Seed weight	0.461*	0.426*	-0.177 ns	0.785**
Embryo length		0.506*	-0.489*	0.433 ns
Endosperm length			-0.048 ns	0.241 ns
RSW				-0.557*

*, ** significant at p=0.05 and 0.01, respectively; ns= not-significant.

Our results suggest that embryo length was important for increased growth as it was associated with RSW (r = -0.489*). Similar findings in barley were reported by Lopez-Castañeda *et al.* (1996). In wheat, Bremner *et al.* (1963) showed that the size of the embryo was crucial to seedling growth up to six days after imbibition but thereafter, the endosperm had a larger role than the embryo. Rice endosperm length was not associated with any of the very early seedling traits and it was not determined whether the effect of endosperm on growth could have been masked by early RSW (6.3 to 7.3 d) or that endosperm conversion to seedling was delayed. The absence of a correlation between endosperm length with early seedling growth traits does not necessarily mean that endosperm is not important for early growth in rice but that it may be just a weak indicator of the amount of food reserves in rice since the bulk of SW is due to the endosperm. It was observed that Sabita produced the highest LA6 and it had the largest SW (30.8 mg) and longest endosperm (8.0 mm). Lopez-Castañeda *et al.* (1996) reported that earlier utilization of the seed reserve was more important than its rate of utilization. It is possible that the germinating seeds may use up seed reserves at a faster rate but the conversion to seedling dry mass is similar for different seed weights. Early RSW was associated with early leaf expansion as shown by its highly significant correlations with LA6 (r = -0.590**) and therefore, with dry matter accumulation (Namuco *et al.* 2005). In the case of more vigorous NSG 19 and Sabita, the negative association of leaf area with RSW is an indication of initial high seedling growth rate.

Conclusions

In our studies, the early onset of autotrophic growth is associated with early vigorous growth and dry matter production. The RSW is an indicator of the onset of autotrophic growth and as such may have a role in the prediction of initial growth rates. Greater initial seed weights were related to enhanced leaf area at 6 DAS and large embryos were associated with rapid early growth. Sabita and NSG 19 displayed the characteristics of rapid early fast growers and may be considered potential parents in breeding for weed suppressive cultivars.

Seedling vigor is a complex trait and may be influenced by various edaphic and climatic factors. It warrants further attention due to its suggested importance in successful crop establishment and its role in rice competitiveness against weeds. Further studies on the characteristics of early growth traits of seedlings may provide additional insights to their role in seedling vigor. However, a necessary step is the validation of observations under field conditions before the findings could be of use as a screening tool.

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EFFECT OF MULCHING MATERIALS FOR WEED CONTROL IN TOMATO (*Lycopersicon esculentum* Mill.) IN THAILAND

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Abstract: The experiments were conducted at Nong Khai Horticultural Experiment Center, Nong Khai province, Thailand from 2001 to 2002. Different mulching materials including straw, cogon grass, grey and black plastic were compared while using standard chemical control: oxadiazon and metribuzin. Additional treatments included hoe weeding were. The predominant weeds were *Melochia corcholifolia* L. for the first year and *Commelina diffusa* Burm.f. in the second year. Best results in terms of tomato yields and net returns were obtained with mulching materials straw and cogon grass.

Key words: Mulching materials, tomato, weed control, benefit

Introduction

Weed competition limits quantity and quality of tomato planting. Many weed species become plant hosts for insects and disease (Anomynous, 2002). In Thailand, weeds in tomatos were recorded as follows: grasses - *Digitaria adscendens* Henry, *Eleusine indica* Gaertn., *Echinochloa colona* (L.) Link., *Leptochloa chinensis* (L.) Nees. and *Dactyloctenium aegyptium* (L.) Willd., broadleaved weeds - *Trianthema portulacastrum* L., *Portulaca oleracea* L., *Amaranthus viridis* L., *Ageratum conyzoides* L. and *Euphorbia hirta* L., and sedges: *Cyperus rotundus* L.(Anonymous, 2002). The difficult to control broadleaved weeds were *Abutilon theophrasti* Medik. and *Solanum ptycanthum* Dun.(Mcgiffen and Masiunas,1991). In the Philippines and Thailand, broadleaved weeds were found to be dominant. Examples are *Ageratum conyzoides* L. *Trianthema portulacastrum* L. *Portulaca oleracea* L., etc. (Pancho, 1986; Kongsaengdao and Suwannarak, 2000). Cultural practices are used to complement weed control. These include deep plowing at 30-40 cm, sun drying for 2-3 weeks and harrowing to decrease weed population which includes disease and insect or pest of tomato. Manual weeding has also been reported to promote high yield during 2-10 weeks after transplanting (Kongsaengdao and Suwannarak, 2000).

Weed management by using natural mulching materials has been evaluated from time to time. Mulching materials can shade sunlight to protect weed seed germination, inhibit weed growth, decrease soil erosion (Agboola and Udom, 1967; Rajput and Singh, 1970) increase nutrient, C.E.C, organic matter and microorganism activities (Lai, 1975). As a result of weed control with mulching materials such as cogon grass, rice straw, banana leaf, and rice hull the tomato yield increased. Earlier studies in Thailand (Yingwiwattanapong *et al*. 2000) suggest that several mulching materials such as synthetic fiber cloth, controlled weeds 7-8 months, followed by cogon grass for 3-4 months, soybean hull and rice hull for 2-3 months, straw and newspaper sheet for 1-2 months in ornamental plant of *Curcuma alismatifolia* Gagnep control weeds and increase flower yield.

Weed control in younger three years of coffee crop was successful by using napier grass, guinea grass, guatemala grass, banana leaf and stem, bark of coffee fruit, stem of maize and sorghum as mulching materials (Thammakhet, 1999). The objective of this experiment was to compare mulching materials with herbicides for weed management in tomato.

Materials and Methods

The experiments were conducted at Nong Khai Horticultural Experiment Center, Thailand by using Randomized Complete Block Design with 4 replications. Experiments comprised 7 treatments as follows: (a) mulching with straw at 1 kg/m², (b) mulching with cogon grass at 1 kg/m², (c) mulching with grey and black plastic, (d) oxadiazon at 1 kg ai/ha, (e) metribuzin at 0.44 kg ai/ha, (f) Hoe weeding at 15 and 45 days after transplanting, and (g) untreated check. The tomato variety peto 94 was used in the first year and SK 4 in the second year of the experiments. Thirty day old tomato seedlings were transplanted at a spacing of 50 cm x 100 cm. Each plot comprised 4 rows, therefore one "rai" (0.16 ha) consisted of 3200 plants. Basal fertilizer application was done with 15-15-15 (N-P_2O_5-K_2O) at 25 kg/rai and composting was done at the rate of 0.5 kg/plant. After 15, 35 and 55 days transplanting, a fertilizer mixture at 12-24-12 (N-P_2O_5-K_2O) at 30 kg/rai were applied.

Weed density and weed weight were recorded at 60 days after transplanting (DAT) by using two quadrates (50 cm x 50 cm) per plot and identified by species. Plant height was measured using 10 plants/plot, shading diameter of the plant was measured from north to south and from east to west then measures included fruits/plant, and size of fruit by length, width and thickness of fruit from 10 fruits/plot. Yields were harvested from 2 m x 5 m area for 5 times and cost analysis was obtained for return or investment.

Results and Discussion

First year experiment

Weed population: At 60 DAT, there were 2 types of weeds: narrow-leaved weeds and broadleaved weeds. The dominant weeds in untreated check plots were broadleaved weeds (Table 1)

Table 1. Weed species and density in untreated check plots as affected by mulching materials on weed control in tomato variety peto 94 (2001).

Weed species	Type	No./0.25 m^2	%
Melochia corchorifolia L.	Broadleaved weed	6.2	49.02
Physalis minima L.	Broadleaved weed	2.4	18.63
Mimosa invisa Mart.	Broadleaved weed	1.9	14.71
Sesbania rostrata Brem.	Broadleaved weed	1.1	8.82
Digitaria adscendens Henry	Narrow-leaved weed	0.9	6.86
Echinochloa colona (L.) Link	Narrow-leaved weed	0.2	1.96
Total		12.7	100.00

The dry weight of broadleaved weed weight in the plots mulched with different treatments is given in Table 2. The total of weed weight was significantly reduced with mulching. Hoe weeding and mulching with straw gave less weed weight, good weed control. The plastic treatment had highly weed density than any other treatments. Generally, black color was found to inhibit seedling germination of weeds. Weed density of untreated check was significantly higher than the rest of the treatments (Table 3). Therefore, any successful weed management practices should consider weed species, density and climatic conditions. The experiment in USA, metribuzin had a good efficacy to control broadleaved weeds such as *Abutilon theophrasti* Medik. (Mcgiffen and Masiunas, 1991).

Tomato growth and yield: At 60 DAT, mulching with straw and cogon grass gave a plant height (Table 3) significantly higher than other treatments (73 cm and 72 cm, respectively).

The natural mulching material can shade to protect weed germination and conserve soil moisture (Agboola and Udom, 1967; Rajput and Singh, 1970) including decay to the organic matter (Limthong and Luengwutiroj, 1998). The chemical control with oxadiazon gave the shortest plant at 59 cm. and similar plant height with metribuzin manual weeding and untreated check.

Table 2. Weed weight as affected by mulching materials on weed control in tomato variety peto 94 (2001).

Treatments	Weed weight (g/0.25 m^2)		
	Broad-leaved weeds	Narrow-leaved weeds	Total
Mulching with straw	3.70 ab	1.33	5.03 a
Mulching with cogon grass	11.93 abc	3.40	15.33 ab
Mulching with plastic	21.15 c	16.63	37.78 b
Oxadiazon 160 g a.i./rai	9.00 abc	0	9.00 ab
Metribuzin 70 g a.i./rai	12.25 abc	0.65	12.90 ab
Hoe weeding at 15 and 45 day	1.40 a	1.65	3.05 a
Untreated check	19.05 bc	3.07	22.12 ab
CV (%)	86.5	28.9	126.0

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05).

Table 3. Weed number, plant height and yield as affected by mulching materials on weed control in tomato variety peto 94 (2001).

Treatments	Weed number (no/0.25 m^2)	Plant height (cm) [1/]	Yield (g/plant)
Mulching with straw	1.75 a	73.7 a	4,825 a
Mulching with cogon grass	5.00 a	72.2 a	3,800 a
Mulching with plastic	2.13 a	60.8 b	1,000 b
Oxadiazon 160 g ai/rai	2.13 a	59.3 b	2,750 ab
Metribuzin 70 g ai/rai	0.50 a	62.5 b	2,425 ab
Hoe weeding at 15 and 45 day	1.25 a	62.2 b	2,350 ab
Untreated check	12.00 b	65.3 b	3,150 ab
CV (%)	85.0	6.6	55.2

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05). [1/]Random from 10 plants/plot

Tomato yields were recorded to be higher when mulching with straw, cogon grass, plastic, oxadiazon, metribuzin, hand hoe and untreated check at 4825, 3800, 1000, 2750, 2425, 2350 and 3150 g/plant respectively (Table 3). The highest yields were obtained from mulching with straw followed by cogon grass. This is because natural mulching materials encourage tomato root to penetrate deep into the soil which helps greater uptake of nutrients and moisture (Thammakhet, 1999).

Second year experiment:

Weed population: Grasses, broadleaved weeds and sedges present insecond year experiment are presented in Table 4. The weed weight of grasses was not significantly different among the treatments. The treatment of using herbicide metribuzin gave the lowest weed weight 0.8 g/ quadrate. Broadleaved weed weights were significantly higher in untreated check than other cultural and mechanical control treatments. The weights of sedges were lower than the others and were not significant among treatments (Table 5).

Table 4. Weed species and density in untreated check plots as affected by mulching materials on weed control in tomato variety SK. 4 (2002).

Weed species	Type	No./0.25 m^2	%
Commelina diffusa Burm.f.	Broadleaved weed	50.0	46.73
Digitaria adscendens Henry	Narrow-leaved weed	17.5	16.37
Cyperus iria L.	Sedge	11.0	10.28
Melochia corchorifolia L.	Broadleaved weed	5.5	5.14
Ludwigia hyssopifolia (G.Don)Exell	Broadleaved weed	5.5	5.14
Lindernia ciliata Pennell	Broadleaved weed	5.5	5.14
Mollugo pentaphylla L.	Broadleaved weed	2.5	2.34
Euphorbia hirta L.	Broadleaved weed	2.0	1.87
Borreria laevis (Lamk.) Griseb.	Broadleaved weed	2.0	1.87
Tridax procumbens L.	Broadleaved weed	1.0	0.93
Cynodon dactylon Pers.	Narrow-leaved weed	1.0	0.93
Mimosa invisa Mart.	Broadleaved weed	1.0	0.93
Ageratum conyzoides L.	Broadleaved weed	1.0	0.93
miscellaneous[1]		1.5	1.40
Total		107.0	100.00

[1]*Fimbristylis miliacea* (L.)Vahl, *Amaranthus viridis* L. and *Brassica juncea* Czern. & Coss.

Table 5. Weed weight as affected by mulching materials on weed control in tomato variety SK. 4 (2002).

Treatments	Weed weight (g/0.25 m^2)			
	Narrow-leaved weeds	Broadleaved weeds	Sedges	Total
Mulching with straw	25.75	29.32 a	0	55.07 a
Mulching with cogon grass	21.22	24.56 a	0	45.78 a
Mulching with plastic	21.09	8.02 a	0.31	29.42 a
Oxadiazon 160 g ai/rai	7.27	13.43 a	0.05	20.75 a
Metribuzin 70 g ai/rai	0.81	39.39 ab	0	40.20 a
Hoe weeding at 15 and 45 day	21.37	6.16 a	0.69	28.22 a
Untreated check	16.82	84.75 b	0.65	102.22 b
Total	114.33	205.63	1.70	321.66
CV (%)	115.8	111.4	147.6	60.5

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05).

Mulching significantly reduced the weed dry weights when comared to untreated check (Table 5). In untreated check plots, *Commelina diffusa* Burm.f was the dominant weed as it had long stems and many branches, followed by *Digitaria adscendens* Henry. The treatment using the herbicide metribuzin gave the lowest weed weight while untreated check plots gave the highest weed weight.

<u>Tomato growth and yield:</u> Mulching with straw, cogon grass and plastic, did not result in a significant change in plant height of tomato (Table 6). These three treatments were different from the rest of the four treatments. Mulching with straw gave the highest plant diameter (93 cm) followed by mulching with cogon grass, plastic, hand hoe, metribuzin and untreated

check. Oxadiazon gave the lowest diameter at 73 cm (Table 6). Similar results were reported by Kittipong (1997) where straw could control weeds in vegetables and conserve soil moisture to promote vegetable growth and prevent weed germination.

Table 6. Plant height, shading diameter and number of fruits per plant as affected by mulching materials on weed control in tomato variety SK. 4 (2002).

Treatments	Plant height (cm) [1]	shading diameter(cm) [1]	number of fruits (fruit/plant) [1]
Mulching with straw	63.9 a	93.7 a	230.5 b
Mulching with cogon grass	60.4 a	85.7 b	237.6 b
Mulching with plastic	64.5 a	83.1 bc	263.9 a
Oxadiazon 160 g ai/rai	52.6 b	73.6 d	157.5 c
Metribuzin 70 g ai/rai	51.4 b	77.5 bcd	165.8 c
Hoe weeding at 15 and 45 day	53.1 b	78.7 bcd	152.1 c
Untreated check	53.6 b	75.5 cd	167.1 c
CV (%)	7.5	6.9	8.1

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05). [1]Random from 10 plants/plot

Mulching with plastic gave the highest fruits/plant (263) followed by mulching with cogon grass and straw. The rest of the treatments gave higher yields when compared to the untreated check. The results of the present study are similar that reported by Kiani and Faravani (2003) where weeds affected the morphology and physiology of tomato. There were no significant difference in width, length and thickness of fruits (Table 7).

Table 7. Width, length and thickness of tomato fruit as affected by mulching materials on weed control in tomato variety SK. 4 (2002).

Treatments	Width (cm)	Length (cm)	Thickness (cm)
Mulching with straw	3.24	4.20	0.423
Mulching with cogon grass	3.27	4.15	0.453
Mulching with plastic	3.22	3.74	0.423
Oxadiazon 160 g ai/rai	3.34	4.03	0.465
Metribuzin 70 g ai/rai	3.34	4.00	0.443
Hoe weeding at 15 and 45 day	3.20	3.92	0.430
Untreated check	3.25	3.95	0.448
C.V.(%)	8.4	7.2	10.7

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05).

The tomatoes were harvested five times, with an interval of 5-7 days. The results showed that mulching with cogon grass gave the highest yield at 3858 g/plant followed by mulching with straw (Table 8), which were significantly higher when compared to the rest of the treatments.

Cost and benefit analysis

The fixed cost of planting tomato seeds is 700 baht, land preparation 600 baht, labor for planting 1200 baht, pesticide (except herbicide) 500 baht, chemical for fruit process 500 baht, fertilizer 1600 baht, decomposed fertilizer 300 baht, fuel 750 baht, agricultural materials (bamboo stick, straw, plastic case) 4000 baht, harvesting cost 350 baht, sale cost 350 baht, herbicide application 100 baht, fertilizer application 50 baht, land rent 1000 baht, and the total of cost production is 12000 baht/rai, excluding weeding cost (Table 9).

Table 8. Yield of tomato for five times as affected by mulching materials on weed control in tomato SK. 4 variety (2002).

Treatments	Yield 1st harvest (g/plant)	Yield 2nd harvest (g/plant)	Yield 3rd harvest (g/plant)	Yield 4th harvest (g/plant)	Yield 5th harvest (g/plant)	total (g/plant)
Mulching with straw	136.64 ab	426.39 ab	1241.32 ab	1005.81	786.20 a	3596.36 a
Mulching with cogon grass	158.00 a	545.83 a	1388.89 a	966.18	799.33 a	3858.23 a
Mulching with plastic	110.85 ab	469.44 ab	1019.45 bc	753.28	236.81 c	2589.83 b
Oxadiazon 160 g ai/rai	95.00 b	328.43 b	814.24 c	887.71	328.06 bc	2453.44 b
Metribuzin 70 g ai/rai	90.96 b	387.51 ab	922.22 bc	969.68	338.91 bc	2709.29 b
Hoe weeding at 15,45 day	120.52 ab	482.84 ab	999.18 bc	748.81	467.71 bc	2819.07 b
Untreated check	89.53 b	446.57 ab	883.34 c	785.88	265.18 b	2470.49 b
CV (%)	25.9	26.5	20.9	30.3	32.3	17.2

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05). [1]Yield was harvested from 20 plants and then average per plant.

Table 9. Investment of weeding and benefit per rai as affecting by mulching materials on weed control in tomato variety SK 4 (2002).

Treatments	Weeding Cost (Baht/rai)	Yield (Kg./rai)[1]	Tomato sale price (Baht/rai)[2]	Fixed cost (Baht/rai)	Benefit (Baht/rai)
Mulching with straw	3,300	11,508.35	34,525.05	12,000	19,255.05
Mulching with cogon grass	4,900	12,346.34	37,039.38	12,000	20,139.02
Mulching with plastic	3,300	8,287.46	24,863.38	12,000	9,563.38
Oxadiazon 160 g ai/rai	450	7,851.01	23,553.03	12,000	11,103.03
Metribuzin 70 g ai/rai	200	8,669.73	26,009.19	12,000	13,809.19
Hoe weeding at 15 and 45 day	960	9,021.02	27,063.06	12,000	14,103.06
Untreated check	-	7,905.57	23,716.71	12,000	11,716.71

[1] 1 rai comprises with tomato at a population density of 3,200 plants, 1 rai = 1600 m²

[2] Calculation with tomato price

The calculation of weeding cost depended on the market price and prevalent labor cost in agriculture. The tomato yield was the highest in plots mulched with cogon grass. The price of tomato price is 3 baht/kg. Mulching with cogon grass gave the highest net return of 20139 baht followed by mulching with straw, hoe weeding, metribuzin, untreated check, oxadiazon and plastic sheet, respectively (Table 9).

Conclusions

Mulching with cogon grass and straw gave excellent weed control and promoted plant height more than any other weed control methods. Mulching with straw gave a better shading diameter than any other treatments, while cogon grass and straw mulches gave high yield and net returns benefit.

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FATE OF PARAQUAT IN SRI LANKAN SOIL TYPES

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Abstract: The herbicide paraquat (1,1-dimehtyl 1-4,4'- bipyridinium dichloride) has been widely used in agriculture for more than hundred countries as a contact herbicide for many crops. It is necessary to monitor the fate of pesticides in the environment, especially soil and water, due to the heavy use of pesticides in the world today. A major part of a paraquat application that reaches the soil within the typical good agricultural practice (GAP) is strongly adsorbed to soil of wide variety of texture. This is in equilibrium with an extremely low concentration in soil solution. However, paraquat in soil solution is biodegradable and completely mineralized by soil microorganisms. A quantitative laboratory scale experiments were carried out to obtain preliminary data on fate of paraquat using two different soil types of Sri Lanka, such as the Distribution co-efficient and Adsorption isotherms. Furthermore, this study has been extended to simulated rice field environment grown in a poly tunnel. An electroanalytical detection scheme previously established by our research group for analysis of paraquat was utilized for the quantification of paraquat in the environment.

Key words: Paraquat, eletcroanalytical chemistry, soil

Introduction

Paraquat is a bipyridinium type, contact herbicide and it is highly toxic to humans. Despite its high mammalian toxicity, paraquat is one of the most frequently used pesticides in over 130 countries world wide. There is a high potential to contaminate water resources with paraquat due to its higher water solubility (about 620 g l^{-1} in pH 7.2 at 20°C) (European Commission Health & Consumer Protection Directorate-General, 2003; Taylor, 2001)

Paraquat has been analyzed using a wide range of different analytical techniques such as spectrophotometry (Jain, 1993), liquid chromatography, mass spectrometry (Jain, 1993), polorography, electroanalytical methods, etc. Electroanalytical techniques offer a promising solution for their direct quantitative determination. Many electroanalytical methods have been established for the determination of paraquat in varies matrices including chathodic stripping voltammetry, capillary zone electrophoresis, square wave voltametry, cyclic voltametry and chemically modified electrodes (Bagchi, 1993; Navaratne, 2000; Souza, 2005; Walcarius, 1996; Zen, 1996).

Due to the frequent use of paraquat and its high-toxicity, it is necessary to detect the levels of this herbicide in the environment such as water resources, paddy field's run-offs and in soil. Many other countries have extensively reported the fate of paraquat in their respective soils (Tsai, 2004).This study was mainly focused on the determination of paraquat in a simulated rice field environment to monitor the fate of paraqut in Sri Lankan soil types and water.

Materials and Methods

Materials
Paraquat used in the experiments was the commercial formulation purchased from the Sri Lankan market. The percentage of the activie ingredient in the commercial formulation was calculated using analytical grade paraquat standard (Methlviologendichloride hydrate, 98%), which was purchase for Sigma-Aldrich (Germany). Common salts prepared using NaCl, KCl and NaOH, were analytical grade chemicals obtained from BDH (England). Analytical grade

stearic acid (BDH, England) was used as the electrode modifier. Sodium dithionite (98 %) was purchased form Hopkin and Willams (England). All the solvents used for the experiments were double distilled.

Methods
Amperometric experiments were carried out using potentiostat (CV-1B) and X-Y recorder from Bioanalytical systems (USA). UV-Visible experiment ware conducted in UV-Visible spectrophotometer (Shimadzu 160 UV-Visible). Amperometric determination of paraquat was done using stearic acid-coated glassy carbon electrode (GCE), following the method reported by Navaratne (2000). Two drops of 1% stearic acid in distilled CH_2Cl_2 was introduced on a cleaned surface of GCE. All electrochemical experiments were conducted with respect to a saturated calomel reference electrode (SCE). Amperometric detection of Paraquat at coated electrodes was performed at potential - 0.65 V vs. SCE.

The UV-Visible detection method was used as the alternative method for the detection of paraquat by electrochemical method. The UV-Visible spectrophotometric detection of paraquat was done at 260 nm (direct UV detection) and at 394 nm (paraquat sodium dithionite complex). In the paraquat complexation, a sample was reacted with 2 cm^3 of 2 % sodium dithionite solution in 0.1 mol dm^{-3} NaOH and the colour intensity was measured in the wave length at 394 nm. A 10 mm quarts cell was used for the measurements of both standard and unknown samples. Calibration standard were constructed in both methods to accomplish the unknown determinations.

Determination of Apparent partition coefficient (K_D)
Soil samples, collected from paddy fields of Rice Research & Development Institute in Batalagoda and Agriculture Research Station at Gannoruwa, Sri Lanka were initially dried for 24 h at a constant temperature (100°C – 105°C). Two soil types from Batalagoda and one from Gannoruwa were used for the experiments. Standard series (1800 ppm – 3000 ppm) of commercial samples (concentration of paraquat was predetermined) of paraquat were prepared. An aqueous solution (100 cm^3) of paraquat (known concentration) was then mixed with sieved (100 mesh) soil (50 g), stirred for 10 min, and allowed to reach equilibrium for 20 minutes. A 25 cm^3 (one from each concentration) sample from the supernatant solution was withdrawn and analyzed for the paraquat content, in triplicate, using the amperometric method. The Apparent Partition Coefficient (K_D) value was then determined for each concentration. Adsorption isotherms were constructed to understand the adsorption behavior of Paraquat in to soil. The experiment was repeated with Bentonyte clay and K_D value was determined.

Introduction of Paraquat in a simulated rice filed environment.
For this purpose, four rice beds (1 m $\times$1 m) were prepared in a polytunnel, and they were filled up to 0.25 m with paddy soil obtained from the Rice Research Development Institute (RRDI), Batalagoda, Sri Lanka. The same type of soil was used in all the four beds for replicate measurements. Weeds were introduced to these soil beds and allowed to grow under saturation condition. Paraquat was introduced to the rice beds. In this experiment, a paraquat solution of 720 mg dm^{-3} was introduced to the field before cultivation of rice. Levels of paraquat were measured using the amperometric method, which was described above. Sampling was started after two days of application of paraquat. The sample was continued until no paraquat was detected within the sensitivity of amperometric method. Two water samples from the top water and one leach out (25.0 cm^3) from each bed were collected and analyzed, in triplicate, using the amperometric calibration curves. After one week of paraquat application, rice plants were cultivated in the beds.

Results and Discussion

As reported earlier, non-electroactive stearic acid modified G.C. was used as an amperometric sensor to detected paraquat at -0.65 V vs. SCE. This experiment resulted in a linear calibration curve and hence determination of unknown paraquat samples from rice bed was possible. Furthermore, the analytical characteristics with respect to sensor calibration are: a minimum detection limit of 6.0 mg dm^{-3} based on the signal to noise ratio of 3, a linear dynamic range of 20.0 mg dm^{-3} and 150.0 mg dm^{-3}, and a sensitivity of 0.042 μA mg^{-1} dm^{3}.

The variation of K_D of paraquat between soil and water phases with concentration, as determined with the aid of amperometric measurements, is shown in Figure1.

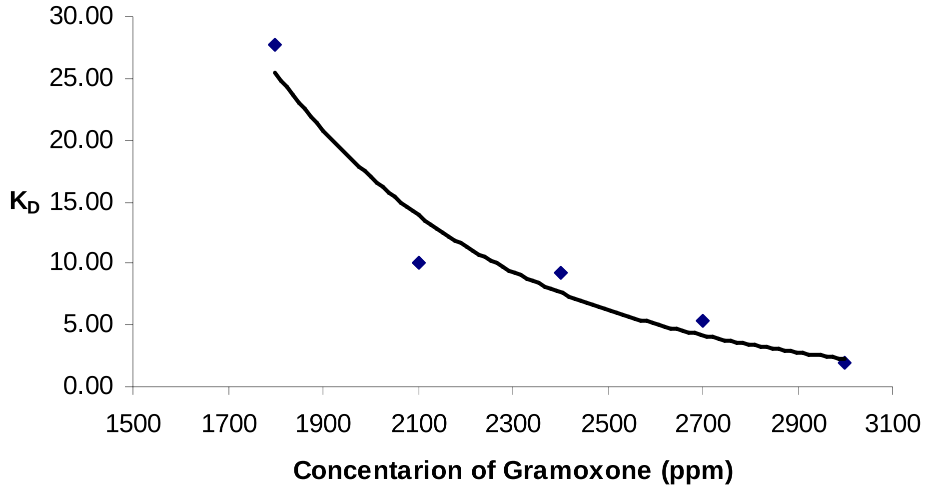

Figure 1. Variation of average K_D (triplicate) with the initial concentration of paraquat

Steady state amperometric detection of paraquat per cent in water samples, which were collected from the model rice bed showed the variation in paraquat concentration with time as given in Figure 2. Paraquat is strongly adsorbed to clay and organic particles (Roberts, 2002). However, K_D values of paraquat between soil and water obtained in this research are lower than those reported for commercial clay (Figure 1). This was further verified by the experiment conducted with bentonyte clay with respect to apparent partition coefficient (the calculated K_D is 92). Bentonyte clay is rich in clay partials they by adsorption of paraquat is high. In contrast K_D values found with soil types used to study were far below. It may be due to the sandy nature of soil used in our experiments. The mechanism of paraquat adsorption into soil occurs via the attachment of positively charged paraquat ions with negatively charged clay and organic matter. Due to strong ionic interaction, adsorption of paraquat is irreversible, and hence desorption occurs at a very slow rate. Consequently, paraquat, when present at low levels, results in even lower concentrations in the supernatant. Such low levels would be far below the minimum detection limit of electro analytical techniques.

Levels of paraquat in the fields were constant during the initial 12 hrs and then it was decreased, and finally equilibrium was established (Figure 2). The levels of paraquat were less than the detection limit of the electro analytical sensor after 96 hrs. Langmuir adsorption isotherm, which is a plot of reciprocal of amount, adsorbed *vs* the reciprocal bulk concentration resulted in a linear relationship with a regression coefficient of Y = 1.0736X - 0.7665. Furthermore, the Fruendlich adsorption isotherm, a plot of ln (amount adsorbed) *vs* ln (bulk concentration) shows linearity up to 7.78 mol dm^{-3}, beyond which the graph is leveled off (Figure 3). The linearity in the Langmuir isotherm favors the initial monolayer adsorption of paraquat to the soil particles. Adsorption of paraquat would continue even after completion of the monolayer until the saturation limit is reached according the Fruendlich isotherm.

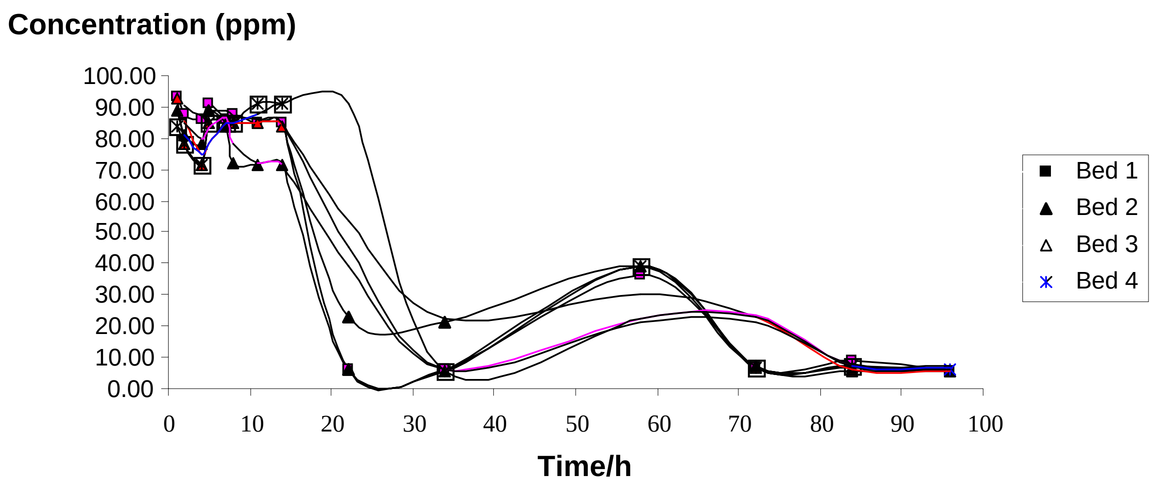

Figure 2. Variation of the levels of paraquat with time of sampling (in supernatant water). Each point is an average of three trials.

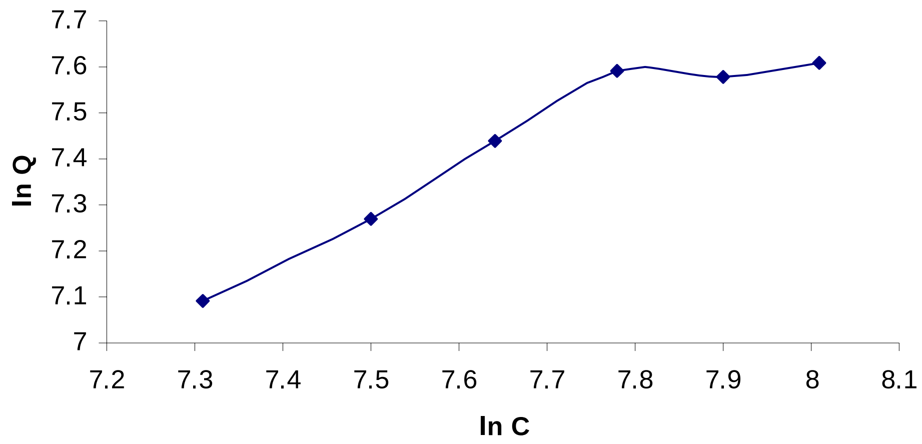

Figure 3. Fruendlich isotherms using electrochemical method.

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SOLITO® A NEW HERBICIDE FOR BROAD SPECTRUM WEED CONTROL IN ASIAN DIRECT-SEEDED WET-SOWN RICE

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Abstract: Weeds and particularly grasses are highly competitive in tropical direct seeded wet-sown rice and cause severe yield losses when they are not controlled timely. Further, in many direct seeded rice areas heavy infestations of *Leptochloa chinensis* have built up following drier water management practice and repeated use of post-emergence herbicides lacking control of this particular species. Solito®, a combination of pretilachlor/pyribenzoxim (EC 320, 300+20 g active ingredient per litre), applied at 6-10 days after seeding at 1-1.5 litre of product per hectare, has been evaluated in several South East Asian countries and Sri Lanka for weed control in direct seeded wet-sown rice. The product was safe to the rice crop and controlled all major weed species present in the trials such as *Leptochloa chinensis*, *Echinochloa crus-galli* , *Ischaemum rugosum* as well as annual broadleaves and sedges. Early control of all weed species combined with a high safety to rice ensured maximum rice yield. The product will be a useful weed management tool to Asian farmers who want to control weeds early and reliably in direct seeded wet-sown rice.

Key words: Direct-seeded rice; herbicide; pretilachlor; pyribenzoxim

Introduction

Weeds and particularly grasses such as *Echinochloa* spp. are highly competitive in tropical direct seeded wet-sown rice and can reduce yield by 45-75 % when not controlled (Johnson *et al.* 2003; Moody 1990). Saturated soil conditions required for the optimal growth of rice seedlings at and during the first two weeks after sowing are ideal conditions for the germination of *Leptochloa chinensis* and *Ischaemum rugosum* (Chin, 2001*)*. *Leptochloa* spp. is not well controlled by several frequently used rice herbicides such as quinclorac, molinate as well as post-emergence acetolactate synthase inhibitors such as bispyribac-sodium, pyribenzoxim and penoxsulam. Repeated use of herbicides based on those active ingredients has caused an increase of *Leptochloa* infestations in major direct-seeded wet-sown rice cultivation areas such as Sri Lanka, Vietnam, Thailand, Malaysia and the Philippines.

Sofit®, a combination of pretilachlor and the safener fenclorim applied at 0-4 days after seeding provides excellent control of *Leptochloa* and *Echinochloa* as well as most annual broadleaves and sedges. However, farmers may miss the application window when heavy rain occurs just after seeding. Solito® , a combination of pretilachlor/pyribenzoxim (EC320, 300+20 g active ingredient per litre and a built-in wetter), applied at 6-10 days after seeding at 1-1.5 litre of product per hectare provides a flexible application window to farmers as well as season long control of all important weed species in tropical wet-sown rice with a single application.

Materials and Methods

The experiments were carried out at multiple locations during several rice cropping seasons between 2004 and 2006 in farmers' fields situated in major direct-seeded wet-sown rice production areas in Malaysia, Sri Lanka, Thailand and Vietnam. Soil preparation, seeding of rice, water management, fertilization and pest management were carried out according to recommended practices in the respective areas. In addition, a yield trial was carried out at Syngenta R&D station in Cikampek, West Jawa.

Experiments were laid out following a completely randomized block design (2-4 replicates) with 15-50 m^2 plots. Herbicides were applied with a knapsack sprayer equipped with a single flood jet nozzle at 400-450 l/ha spray volume. Fields were completely drained at the time of herbicide application. Water was reintroduced 1-3 days after application in Vietnam, Malaysia and Sri Lanka and between 2 and 7 days in Thailand. Crop tolerance and control of each individual weed species were assessed visually at 15, 30 and 50 days after seeding following a 100% scale (0% = no control, no crop damage; 100% full control, complete crop destruction).

Results and Discussion

No damage to rice was reported in any trial after application of the combination pyribenzoxim/ pretilachlor at all tested application rates and timings. The combination pyribenzoxim/pretilachlor provided excellent control of both *Echinochloa* and *Leptochla* sp. when applied at 6-10 DAS at 1.0-1.5 l/ha in Vietnam, Malaysia and Sri Lanka (Tables 1, 2 and 4). Performance of the product was slightly more variable in Thailand, mainly due to delayed water reintroduction in some trials (Table 3). Other weed species including *Ischaemum rugosum* as well as annual broadleaves and sedges were completely controlled by the combination pyribenzoxim/pretilachlor (data not shown).

Table 1. Control of grass weeds in direct-seeded wet-sown rice in Sri Lanka, average across 5 trials carried out in 2005 and 2006.

Herbicide Treatment	Application timing	% Control of *Echinochloa* spp. 40-45 DAA	% Control of *Leptochloa* spp. 40-45 DAA
Pretilachlor/pyribenzoxim EC 320, 1 l/ha	6 DAS	100	99
	8 DAS	100	99
Pretilachlor/pyribenzoxim EC 320, 1.25 l/ha	8 DAS	100	99
	10 DAS	100	100
Bispyribac-sodium SC 100, 30 g a.i/ha	15 DAS	98	38

DAS= Days after Seeding; DAA= Days after Application

Control of *Echinochloa* was equivalent or superior to standard bispyribac-sodium applied 10-15 DAS at 20-30 g ai/ha in Malaysia, Vietnam and Thailand. Bispyribac-sodium failed to control *Echinochloa* at several locations in Malaysia whereas the mixture pyribenzoxim/ pretilachlor performed well. This result may indicate that the combination is a possible option for the management of ALS-resistant *Echinochloa*. Control of *Leptochloa* with the combination was clearly superior to straight bispyribac-sodium confirming the additional activity of pretilachlor in the combination against both grass species.

Table 2. Control of grass weeds in direct-seeded wet-sown rice in Vietnam, average across 14 trials carried out in 2005 and 2006.

Herbicide Treatment	Application timing	% Control of *Echinochloa* sp. 40-45 DAA	% Control of *Leptochloa* sp. 40-45 DAA
Pretilachlor/pyribenzoxim EC 320, 1 l/ha	6 DAS	99	98
	8 DAS	97	95
Pretilachlor/pyribenzoxim EC 320, 1.25 l/ha	8 DAS	96	91
	10 DAS	98	97
Bispyribac-sodium SC 100, 20 g ai/ha	15 DAS	90	68

Table 3. Control of grass weeds in direct-seeded wet-sown rice in Thailand, average across 9 trials carried out in 2004 and 2005

Herbicide Treatment	Application timing	% Control of *Echinochloa* sp. 40-45 DAA	% Control of *Leptochloa* sp. 40-45 DAA
Pretilachlor/pyribenzoxim EC 320, 1.5 l/ha	8 DAS	98	94
	10 DAS	93	88
Bispyribac-sodium SC 100, 30 g ai/ha	10 DAS	91	86

Table 4. Control of grass weeds in direct-seeded wet-sown rice in Malaysia, average across 16 trials carried out in 2005 and 2006

Herbicide Treatment	Application timing	% Control of *Echinochloa* sp. 40-45 DAA	% Control of *Leptochloa* sp. 40-45 DAA
Pretilachlor/pyribenzoxim EC 320, 1.5 l/ha	8 DAS	100	100
	10 DAS	100	96
Bispyribac-sodium SC 100, 20 g ai/ha	12 DAS	59	25

A trial carried-out under heavy *Echinochloa* and *Leptochloa* infestation showed that pretilachlor/pyribenzoxim applied at 8 DAS at 1.5 l/ha provided the highest rice grain yield in comparison with various standards (Table 5).

Table 5. Effect on yield of different herbicide treatments in direct seeded wet-sown rice in Cikampek, West Jawa, 1 trial wet-season 2005-6

Herbicide Treatment	Application timing	% Maximum crop damage	% Control of *Echinochloa* sp. 40-45 DAA	% Control of *Leptochloa* sp. 40-45 DAA	Yield in t/ha grain
Untreated			0	0	0.96 D
Propanil/butachlor, EC 550 2 l/ha	8 DAS	11	77	71	3.58 B
Pretilachlor/pyribenzoxim EC 320, 1.5 l/ha	8 DAS	6	97	91	5.06 A
Bispyribac-sodium SC 100, 20 g ai/ha	15 DAS	12	92	22	2.52 C
Fenoxaprop-p-ethyl/ethoxysulfuron SC 89, 0.5 l/ha	15 DAS	16	64*	55*	3.18 C

Fimbristylis, *Cyperus* and *Monochoria* were controlled at 90% or more by all herbicide treatments; 4 replicates, 20 m^2 harvested per plot; letters indicate differences in yield at 5% LSD Duncan test. * Lack of control of new weed germinations.

Results presented in Table 5 was based on the excellent crop tolerance of the mixture combined with an early application timing and season-long control of all weed species present including both *Echinochloa* and *Leptochloa* spp.

In conclusion, the results of extensive field tests carried-out with the combination pyribenzoxim/ pretilachlor in tropical direct-seeded wet-sown rice have shown that the combination provides a constant high level of control of all major weed species with a single herbicide application. Application of the product 6-10 days after seeding just before water introduction extends the application window for early season herbicides and gives more flexibility to farmers to meet the optimal of timing application and thereby provide cost effective and robust weed control. Excellent tolerance of the product to rice and early control of all major weed species ensure maximum yield. The low application rate of the product and the optimized built-in wetter provide further convenience to farmers. Extensive registration studies have also confirmed that the product is safe to farmers and the environment.

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BIOCHEMICAL ADAPTATION OF PURPLE NUTSEDGE (*Cyperus rotundus* L.) TO FLOODING: ALCOHOL DEHYDROGENASE ACTIVITY IN DRYLAND AND WETLAND ECOTYPES

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Abstract. Increasing populations of purple nutsedge (*Cyperus rotundus* L.) in rice grown in rotation with vegetables is apparently due to increasing adaptation of this weed to flooded soil, thus it is now an emerging weed problem in flooded rice. Laboratory studies were conducted to determine the basis for its adaptation to flooded soil by comparing activities of enzymes of alcoholic fermentation in dryland and wetland ecotypes. Alcohol dehydrogenase (ADH) activity was detected in the roots of seedlings of both ecotypes. Induction of enzyme activity was observed in both ecotypes after 24 hrs hypoxia amounting to as much as 46-fold in the dryland ecotype and 14-fold in wetland ecotype. When hypoxia was prolonged to 48 hrs, the dryland purple nutsedge showed a continuous rise in ADH activity while down-regulation was exhibited by the wetland ecotype. This was coupled with a pronounced decrease in soluble sugars in the wetland ecotype upon germination and during hypoxia of up to 48 h, suggesting that catabolism of soluble sugar is fairly active in the wetland tuber when oxygen is limited, indicating a pronounced Pasteur effect. A slow decrease observed after 48 hrs hypoxia suggests that utilization of soluble sugar by the wetland ecotype is highly regulated during hypoxia, and a high steady-state sugar level in the tuber is sustained. In contrast, there was an increase in starch and soluble sugars in the dryland tuber upon germination up to 48 hrs hypoxia. Our results suggest that tolerance of wetland purple nutsedge to flooding is due to its ability to slow down its fermentation rate during prolonged hypoxia. This was obtained by down-regulation of its ADH activity and by reducing its utilization of carbohydrate reserves, resulting in sustainability of its substrate supply to avoid starvation, injury and death in the event of prolonged flooding.

Key words: Weed adaptation, hypoxia, anoxia, flooding tolerance, Pasteur effect

Introduction

Over the years, the continuous rotation pattern of rice during the wet season and vegetables during the dry season in rainfed rice areas in the Philippines is naturally selecting purple nutsedge that can grow in both wetland and dryland fields, and causing increased populations of this weed in flooded rice. From being a minor weed in flooded rice in the 1970s, purple nutsedge was the second most dominant weed in flooded rice in central Luzon, Philippines in the late 1990s (Baltazar *et al*. 1999). This suggests that mechanisms of adaptation to flooding to survive in an oxygen-deficient environment may have evolved in the wetland ecotype.

When exposed to anaerobic environment, plants normally shift energy production from aerobic respiration to alcoholic fermentation, which is an important adaptation for survival under anaerobic conditions. Increase in enzymes of glycolysis and ethanolic fermentation, pyruvate decarboxylase (PDC) and alcohol dehydrogenase (ADH) under anaerobic conditions has been reported in rice (Ellis and Setter, 1999). Alcohol dehydrogenase (ADH, alcohol:NAD^+ oxidoreductase, E.C. 1.1.1) is the most studied enzyme relating to anaerobiosis. ADH activity was reported to increase in most plants in response to anaerobic. The maintenance of a fairly high level of fermentable sugar during conditions of oxygen deficiency is also important so that the plant will not run out of fuel for the sustenance of its growth and metabolism.

Determining the mechanism of adaptation of purple nutsedge to flooding will help understand weed plasticity and adaptability to a wide range of environmental conditions and assist in predicting weed response to the environment. Data from this research can be used to provide basic information that will help enhance flood tolerance in crops as well as in developing ecological weed control approaches such as flooding and other kinds of habitat-manipulation management strategies. This study was conducted to determine the importance of alcoholic fermentation and carbohydrate availability to flooding tolerance of purple nutsedge by comparing its alcohol dehydrogenase activities and sugar and starch content in dryland and wetland ecotypes.

Materials and Methods

Plant material
Tubers of purple nutsedge were collected from flooded rice fields and from vegetable growing areas to represent wetland (also called lowland) and dryland (also called upland) ecotypes, respectively. The tubers were grown in their respective habitats (dryland or wetland) in the screen house of the International Rice Research Institute.

Enzyme extraction
Enzyme extraction was done following the method of Valdez (1995). At 1 week after tuber sprouting, roots samples were ground with a cold extraction buffer (1 g tissue per 6 ml buffer) using ice-cold mortars and pestles. The extraction buffer was adjusted to pH 7.5 with 1 N potassium hydroxide (KOH). From the crude extract, 200 ul was transferred to an Eppendorf tube containing 3% bovine serum albumin (BSA). With BSA as the standard, the remaining extract was centrifuged and analyzed for protein content using the Bradford method (Bradford, 1976). The aliquot for ADH assay was centrifuged at 13,000 rpm for 3 min at 4°C. The supernatant was then transferred to a clean Eppendorf tube and kept in ice until assay.

Alcohol dehydrogenase assay
Alcohol dehydrogenase was assayed in the acetaldehyde to ethanol direction using the method optimized by Valdez (1995). The assay mixture consisted of 830 ul of 62.4 mM TES at pH 7.0, 20 µl crude extract, 50 µl of 3.4 mM nicotinamide adenine dinucleotide, reduced form (NADH), and 100 µl of 200.32 mM acetaldehyde. The enzyme activity was measured by following the disappearance of NADH at 340 nm at 30°C for 7 min. One unit of the enzyme was defined as the amount of enzyme catalyzing the oxidation of 1 µmole of NADH per minute at such temperature**.**

Hypoxic treatment
Hypoxic treatment was based on the method of Ellis and Setter (1999) with slight modifications. The experiment was conducted in airtight Erlenmeyer flask with gas inlet and outlet. Young intact seedlings of dryland and wetland purple nutsedge were immersed in water through which nitrogen gas was flushed vigorously until the concentration of oxygen dropped to 0.05 mol m^{-3}. After hypoxia for 24 to 48 hrs, the roots were excised from the seedlings and assayed for enzyme activity.

Soluble sugar, starch and total carbohydrate content
Tubers were peeled and cut into small pieces, oven-dried at 70°C for 24 hrs, then ground into fine powder. Portions of the samples were weighed (200 mg), placed in a test tube and analyzed for total soluble sugar content based on the anthrone reaction following the procedure of Fales (1951). The residue obtained after extraction of soluble sugar was used for starch analysis following the method of McCready *et al*. (1950). Diluted perchloric acid was

used to effect hydrolysis of starch and the released sugar was assayed based on anthrone reaction. The amount of total carbohydrates in the tubers of dryland and wetland purple nutsedge at various hypoxic treatments was determined by summing up the values obtained for the amount of soluble sugar and starch of each replicate of every treatment.

Results and Discussion

Alcohol dehydrogenase activity

Upon germination, ADH activity in the roots of 7-day old seedlings was detected in both ecotypes. When the seedlings were subjected to hypoxic treatment for 24 h, a sharp increase in ADH activity was manifested (Figure 1). When the hypoxic treatment was extended to 48 hrs, the dryland ecotype manifested a continuous sharp increase in ADH activity while down-regulation was exhibited by the wetland ecotype. The dryland and wetland purple nutsedge differed in their strategies to survive long-term hypoxia or anoxia. It is possible that the dryland ecotype follows Mode 1 of anoxia tolerance as proposed by Gibbs and Greenway (2003) where fermentation is greatly enhanced to compensate for the low energy production, thus, meeting the energy requirement to support plant growth. The uninterrupted boost in the induction of the enzyme activity indicates that the plant strongly exhibits the Pasteur Effect in order to sustain its growth in the oxygen-deficient environment. However, this mode of adaptation to anoxia is not highly sustainable due to the fact that it entails high substrate requirement. More often than not, this mechanism leads to substrate depletion.

In contrast, the mechanism of adaptation by the wetland ecotype is probably intermediate between Modes 1 and 2 as suggested by Gibbs and Greenway (2003). At early periods of hypoxia, there was a pronounced increase in the activity of the enzyme, implying that wetland ecotype also exhibits the Pasteur Effect as a means of generating energy. However, during prolonged hypoxia, slowing down of fermentative metabolism was manifested by the wetland ecotype. The mechanism of down-regulation of metabolism by the wetland ecotype during long-term hypoxia ensures sustainability of the fermentation process. This is to avoid early depletion of substrate, which eventually leads to starvation in the long run. Thus, at the biochemical level, an important characteristic that distinguishes the wetland purple nutsedge from the dryland ecotype is its frugality in utilizing its energy reserve to sustain metabolic processes during prolonged hypoxia. The wetland ecotype observes prudence in its metabolism, a quality that turns out to be beneficial for long-term survival in the oxygen-deficient environment in flooded rice fields. Higher plants have an absolute requirement for oxygen to sustain metabolism and growth. Oxygen functions as terminal electron acceptor, and is thus important in respiration. As a consequence, most plant tissues suffer from irreversible damage during anoxia. Some agricultural practices such as natural flooding and irrigation can bring about oxygen deficiency around the rooting zone of the plant. This is due to the fact that gases diffuse 10,000 times slower in water than in air (Mohanty *et al*. 2000). Differences in the regulation of ethanolic fermentation can occur between plant species resulting in varying responses to certain environmental stresses such as hypoxia or anoxia brought about by flooding. Comparison of the fermentative metabolism of dryland and wetland purple nutsedge is important as it could shed light on the observed tolerance of wetland ecotype and intolerance of dryland ecotype to low oxygen condition.

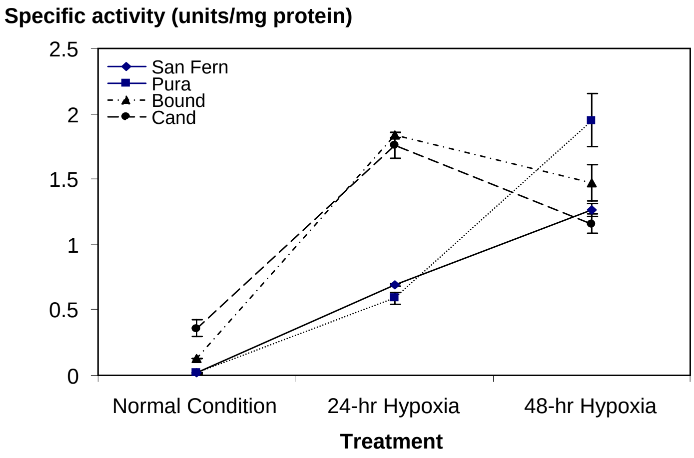

Figure 1. Changes in alcohol dehydrogenase activity in roots of 7-day old seedlings of dryland (San Fernando and Pura) and wetland (Boundary and Candaba) purple nutsedge at germination and after 24 h to 48 h hypoxia.

Carbohydrate content in tubers

The tubers of purple nutsedge serve not only as major vegetative propagules, but also as sources of carbohydrate to fuel its metabolic processes. The tubers of the wetland ecotypes are bigger, more plump and more robust, while the tubers of the dryland ecotypes are smaller and leaner (Figure 2). The biomass of the wetland tubers is also much heavier and significantly higher than the biomass of the tubers of the dryland ecotype (Figure 3).

A B

Figure 2. Tuber morphology of wetland (A) and dryland (B) purple nutsedge

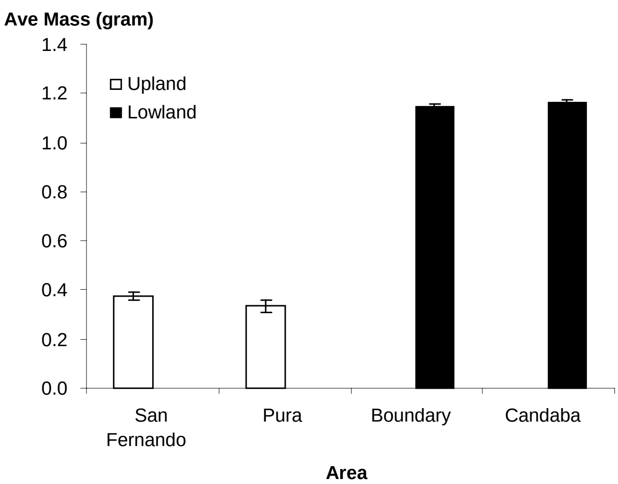

Figure 3. Tuber biomass of dryland (San Fernando and Pura) and wetland (Boundary and Candaba) purple nutsedge

Although the amount of total carbohydrate in the tuber of the wetland purple nutsedge was lower on per cent basis (Figure 4A), its tendency to develop bigger tubers with greater biomass correlated well with its ability to accumulate higher amounts of carbohydrate. Thus, on a per tuber basis, the amount of total carbohydrate in the wetland tuber is significantly higher than that of the dryland tuber (Figure 4B). The bulk of carbohydrate reserve in the wetland tuber was in the form of soluble sugar, at levels much higher than that in the dryland tuber (data not shown). The dryland tubers stored its carbohydrate in the form of starch (data not shown).

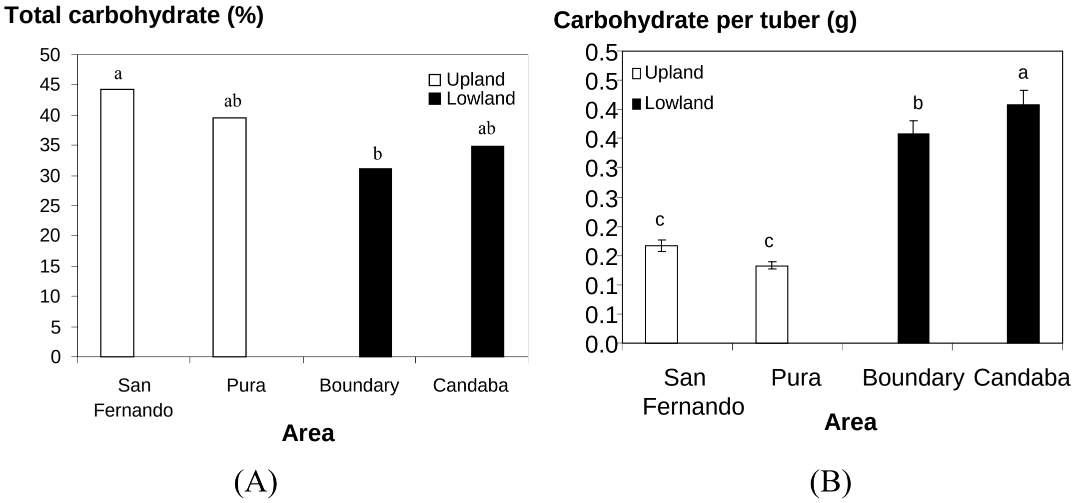

Figure 4. Total carbohydrate content of dryland (San Fernando and Pura) and wetland (Boundary and Candaba) purple nutsedge expressed as percentage by mass (A) and per tuber basis (B)

While the wetland tuber accumulated high amounts of soluble sugar before germination, a pronounced decrease in soluble sugars was observed upon germination and during hypoxia of up to 48 hours (data not shown). This suggests that catabolism of soluble sugar is fairly active in the wetland tuber when oxygen is limited, indicating a pronounced Pasteur effect. Slow decrease after 48 hr hypoxia suggests that utilization of soluble sugar by the wetland ecotype is highly regulated during hypoxia, such that a high steady-state sugar level in the tuber is sustained. Modulation in the consumption of carbohydrate reserve by the wetland ecotype will prevent depletion of substrate during prolonged periods of hypoxia. An increase in soluble sugar content in the dryland tubers during hypoxia agrees with the observed increase in alcohol dehydrogenase activity in the roots of dryland purple nutsedge.

Results of our studies indicate that the tolerance of wetland purple nutsedge to hypoxia is due to its capacity to accumulate large amounts of soluble sugar, coupled with its ability to slow down its fermentation rate during prolonged hypoxia. This is done by down-regulation of its alcohol dehydrogenase activity and by slow release of its stored soluble sugars, ensuring sustainability of substrate supply in the long run to avoid starvation that might lead to injury and plant death in the case of prolonged flooding.

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Hydrocotyle umbrellata L.: A NEW INVASIVE AQUATIC PLANT IN THAILAND

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Abstract: Water hyacinth (*Eichhornia crassipes* Solms) is one of the most well-known invasive aquatic plants in the world In Thailand, apart from this species, there are plants species, which are gradually and steadily increasing their area of invasion, such as narrowleaf cattail (*Typha angustifolia*), Florida type water lettuce (*Pistia stratiotes* L.) and water milfoil (*Myriophyllum brasilliensis* Cambess.). The Dollarweed (*Hydrocotyle umbellata* L.) is also an invasive species and was intentionally introduced as an ornamental plant in aquaria about 10 years ago. The invasiveness of this plant was studied for its growth, competitiveness and reproduction ability, which is limited to the vegetative part. Even if the plant produces seeds, seedlings have never been found in the experimental field. The plant growth in water started increased 1 to 140 leaves within 3 months. The plant can compete against Asiatic pennywort (*Centella asiatica* L.) in experimental plots. At present, *Hydrocotyle umbellata* L has been found increasing its areas of invasion gradually in both aquatic and terrestrial habitats.

Key words: Ornamental plant, vegetative reproduction, *Hydrocotyle umbellata*

Introduction

Many aquatic plants were introduced for aquarium decoration or as general ornamental plants. Water hyacinth (*Eichhornia crassipes* Solm.) is a classic example of introduction of such an invasive aquatic plant through human intervention, which has made it distributed through out the world. Thailand is now facing invasion of some aquatic invasive plants, which were intentionally introduced to the country. Water lettuce (*Pistia stratiotes* L.), the only species of genus *Pistia* L., especially the Asian type, could be found in closed water body such as swamps, ditches in orchards and ponds. However, the Florida type was introduced as an ornamental aquatic plant and was sold in Sunday markets. At present the plant can be found in many water bodies especially along roadsides or near communities.

Water milfoil (*Myriophyllum brasilliensis* Cambess.) was introduced for ornamental purpose about 20 years ago. Today the plant is still being sold as aquarium and aquatic ornamental plant in the market. The plant grows very well in the absence of natural enemies in canals, streams and swamps all over northern Thailand. The narrowleaf cattail (*Typha angustifolia* L.) is the most invasive plant in Thailand at present. It was introduced to Thailand for flower arrangements and as an ornamental plant about 30 years ago. Presently, the plant grows luxuriantly in swamps, shallow ponds, vacant paddy fields or waste land where soil is with high moisture, including roof-gardens in high building in downtown Bangkok. The plant can form pure stands after occupying the area within few months. Numerous tiny seeds with hair are blown by the wind to far away places make the plant distribute very fast.

Hydrocotyle umbellata L., is an aquatic, or water-loving, perennial plant with many common names such as Dollarweed, marsh or water pennywort or navelwort, native to America (NRCS, USDA, 2007). The plant was introduced to Thailand more than 10 years ago and sold in market as an ornamental plant. With shiny round leaves and easy growing, the plant has become popular in aquariums plant few years later. The plant can produce roots at every node and new shoots from lateral buds at leaf axils. Thus, it can propagate through vegetative parts, *i.e.* nodes with a leaf. Generally, this plant can grow vigorously as natural enemies are not found in aquaria (Plate 1)

Plate 1. Dollarweed (*Hydrocotyle umbrellata* L.)

The plant is still popular as an aquarium plant and through misidentification with Asiatic pennywort, it is consumed as a vegetable. Moreover, the entry route of ornamental plants is an important pathway of spreading these species far and fast, by human beings. Thus, the purpose of this study was to evaluate weed potential of Dollarweed in Thailand, aiming at preventing the spreading of this plant in nature.

Materials and Methods

Propagation ability of vegetative parst.

As Dollarweed (*Hydrocotyle umbrellata* L.) is a creeping plant and new shoots emerge from the lateral buds at every leaf axis, the shoot apex with four leaves of Dollarweed was selected and was cut at 0.5 - 1 cm away from both sides of the node. Each node with a leaf was placed in a beaker containing three different media namely, *i.e.* 100 ml water, 5 g soil + 100 ml water, and 10g soil + 100 ml of water, with 5 replicates. All beakers were placed at room temperature under natural conditions. Water levels in beakers were maintained at the same level throughout the experiment. The plant growth parameters such as root growth, leaf number and number of newly emerging shoots were recorded every 3 days for a period of one month.

Growth of Dollarweed

Cuttings containing one node with a mature leaf were grown in 34 cm x 47 cm x 12 cm pots containing soil at 8 cm deep, totaling to 38 pots. The water level was kept up to the top of the pot throughout the experiment, t simulate the flooded conditions. The number of leaves in each pot was recorded every 7 days up to 168 days.

Competition with other plant

Asiatic pennywort [*Centella asiatica* (L.) Urban], which is an indigenous vegetable and considered as a medicinal plant in Thailand, was used for competitive studies with Dollarweed. Both plants were grown in a 100 cm x 100 cm pots with a fixed population density, *i.e.* 5 plants per pot, while using the replacement series method to have the ratio of the number of Dollarweed:Asiatic pennywort in each pot as 5:0, 4:1, 3:2, 2:3, 1:4 and 0:5, with 3 replicates. Pots were watered everyday to field capacity. The leaf number of each plant was recorded every 7 days up to 154 days, and the first record was made at 14 days after planting. At the end of the experiment, all the plants were removed, cleaned to remove soil and weighed for fresh weight. Thereafter, all leaves were removed for measurement of the leaf area using leaf area meter (Hayashi Denkoh, ACC-400). All plants were dried in an oven at 70^{o}C for 3 days and the dry weight was recorded for individual plant

.

Allelopathic potential

Allelopathy is considered as an important character of plants if they are to become strong weeds or an invasive plant. Roots, leaf stalks and leaves were prepared fresh and dried with 0 (control), 0.01, 0.05 and 0.1 g of materials placed separately between 10-10 agar (0.3%) in ∅ 29 mm x 130 mm test tube. Then 5 seeds of lettuce were sown in each tube. The experiment was carried out with 3 replicates. The tubes were sealed with a transparent film and kept in 25^{o}C under dark conditions. The root length and shoot height of each plant were recorded.

Field observations.

The area of infestation of Dollarweed in the wild was surveyed in the north, northeast and central region of Thailand. The habitat of the infestation was also recorded.

Results and discussion

Propagation ability of vegetative part

The planting material with the first node and the youngest leaf of the shoot apex, without roots, was green for a week and after that the plant died under all conditions. Although some plants had new short roots, they died later. A similar trend was observed with the first node in water + 5 or 10g of soil, but the plant green for a shorter period (Table 1). When the panting material with the second node was used, only the plant in water medium was green until the end of the experiment, but those in water + soil died. However, those that survived in water did not produce new leaves. The panting materials with the third node survived under all conditions but only 2 out of 5 plants in water produced new but small leaves. All the planting materials with the fourth node survived and grew well, producing new leaves in all growing conditions tested.

Table 1. Number of Leaves of Dollarweed at 1 month after growing in water

Condition of growing	Order of node from shoot apex			
	1st node	2nd node	3rd node	4th node
Water	B-0	1	1-2	1-2
Water + 5g soil	0	x	1	2
Water + 10g soil	0	X	1	2

*Growth of Dollarweed***.**

In the first 7 days of the experiment, only few plants started producing new shoots, which later became the main shoot. The number of leaves of the plants increased gradually with time. After the lateral buds elongated with leaves on the branch, the number of leaves increased around 50 days after planting (Figure 2). The number of leaves increases after second and third branches were fully developed.

Competition with other plants

The number of leaves. The Average number of leaves at the onset of experiment for Dollarweed was 1, and for Asiatic pennywort 5. The number of leaves of both plants increased gradually with time (Figure 3). The increase in the number of leaves of both plants was followed a similar pattern, with the number leaves per plant in Dollarweed was marginally higher than that of the Asiatic pennywort. The number of leaves of the Asiatic pennywort was markedly higher than that of Dollarweed in the pots where this species was grown 3 and 4 plants of Asiatic pennywort. However, at the end of the experiment, the total number of leaves per plant of the Asiatic pennywort was higher than that of the Dollarweed except in pots where the number of Asiatic pennywort was less. At the end of the experimentation, the average leaf number per plant for Dollarweed + Asiatic pennywort was 360+0, 527+455, 708+302, 386+577, 591+694 and 0+594 in pots where Dollarweed were grown alone, and in pots where Dollarweed plants : Asiatic pennywort was grown at a ratio of 4 + 1 Asiatic pennywort, 3 + 2, 1 + 4, and Asiatic pennywort alone, respectively.

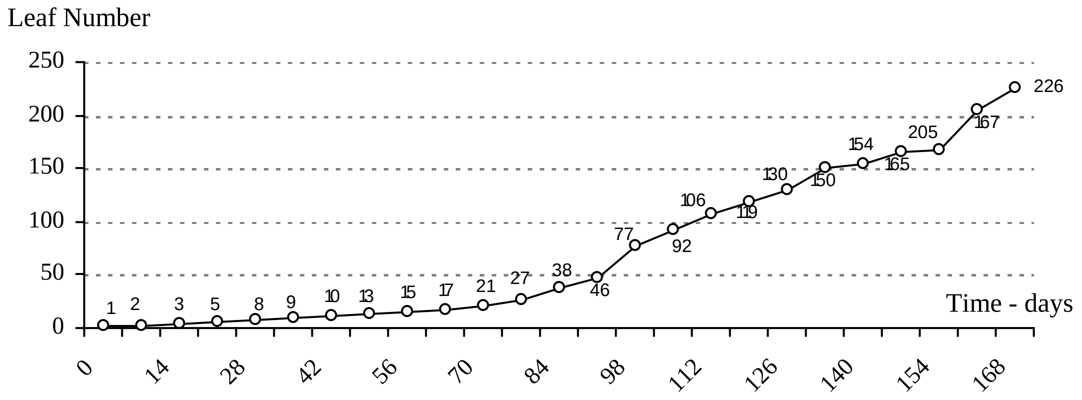

Figure 2. Growth of Dollarweed in pot (number of leaves)

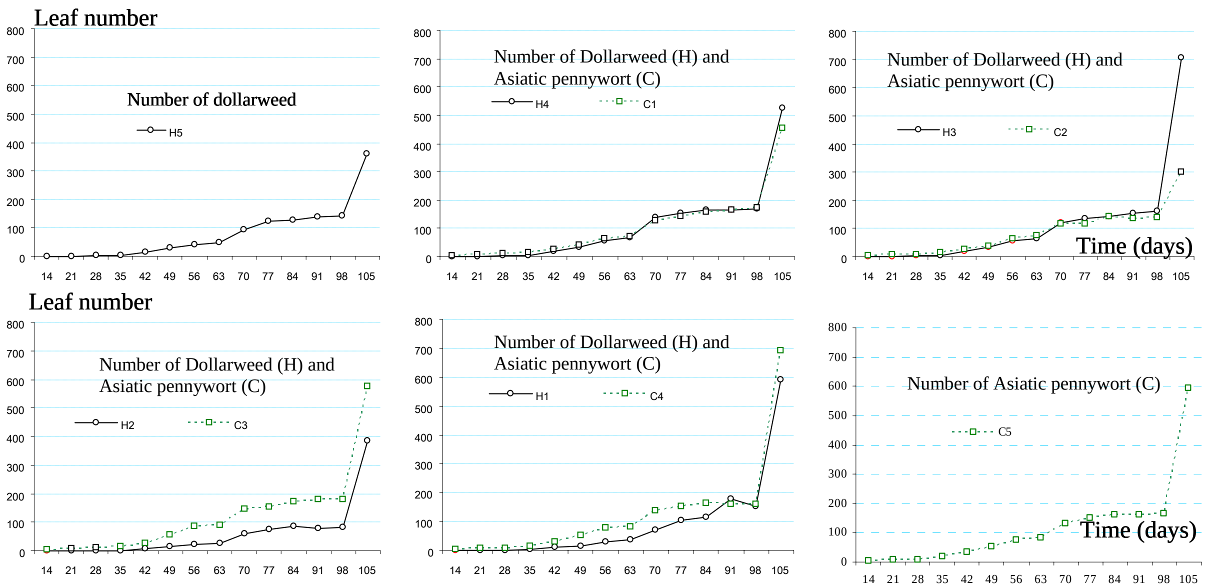

Figure 3. Individual average number of leaves of Dollarweed (H - solid line) and Asiatic pennywort (C – dotted line) growing in 1 m x 1 m pot

Leaf area: The average leaf area of each treatment is shown in Figure 4. The average leaf area of both plants follow the same trend of change, and less competition was observed in the same species with the higher leaf area. Leaf area for individual plants of Dollarweed was 692, 1,110, 1,5159, 1,429 and 3,259 dm^2 when grown at 5, 4, 3, 2 and 1 plant per pot in combination with 0, 1, 2, 3, and 4 plants of Asiatic pennywort. The leaf area of Asiatic pennywort was 1,810, 924, 1502, 1055 and 902, respectively (Figure 4).

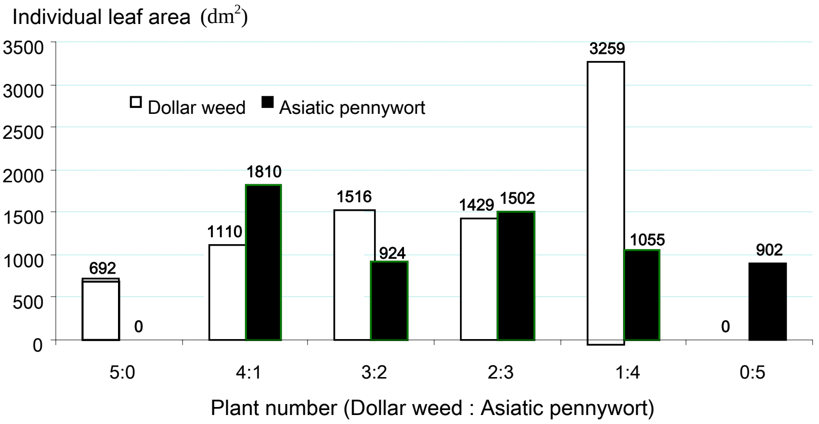

Figure 4. Average leaf area of individual plant at the end of the experiment.

Fresh weight and dry weight: The average fresh weight of Dollarweed was higher than the Asiatic pennywort in all the treatments that included both species. The change of dry weight showed similar pattern to that of fresh weight, except that when the Dollarweed alone pots showed a lower dry weight per pot when compared that with Asiatic pennywort alone.

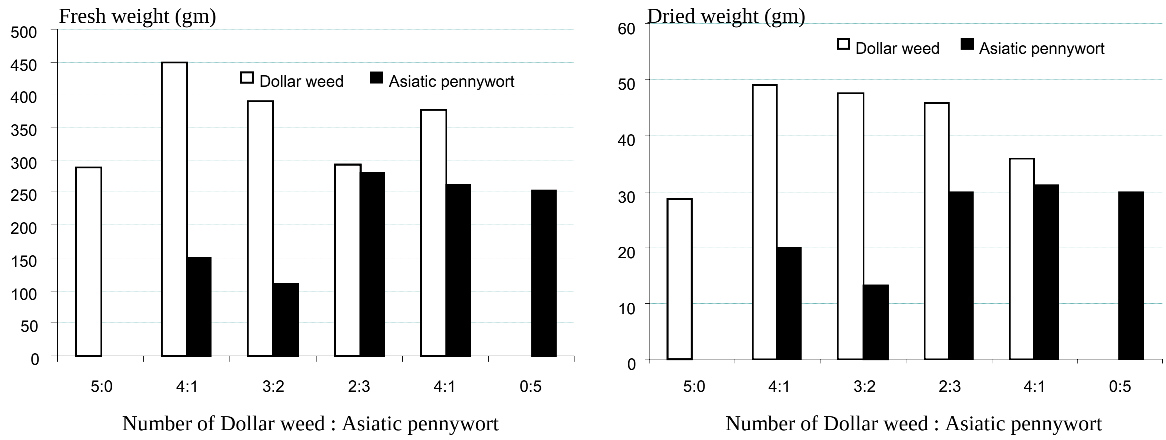

Figure 5. Average fresh and dry weight of individual plant of Dollar weed and Asiatic pennywort

Allelopathic potential: Lettuce grown in 0.3% agar with fresh dried leaf (DL), dried shoot (DS), dried root (DR), fresh leaf (FL), fresh shoot (FS) or fresh root (FR) at 0.01, 0.05 or 0.1g are shown in Figure 6. The root growth of lettuce decreased at higher rates of the Dollarweed. Both root and shoot growth of lettuce grown with dried Dollarweed was less than that under fresh Dollarweed. The root growth of lettuce did not show any progress when treated with all the rates of dried root of Dollarweed. The growth of lettuce (fresh weight) was 6.3, 1.2 and 0% of control when treated with 0.01, 0.05 and 0.1 g of the roots of Dollarweed, and shoot growth was 0 in all rates of dried root.

Field observations

Infestation of Dollarweed was found in a mango orchard where the plant was grown as an ornamental plant. It formed a thick mat on which a man could walk. The plant had to be cut to move out from the body of water, but small pieces of plant were spread out along the water way in the orchard, thus the plant could compete against torpedograss (*Panicum repens* L.). In a village in northern Thailand, the plant grew throughout the yard in front of a house even under dry soil conditions. The plant infestation was found both in the water along roadside as well as on uplands with moist soil or dried areas. In some swamps the plant occupied both the water and more than 1 meter up in the bank, forming a thicket, which was previously occupied by a global invasive plant swamp, morning glory (*Ipomoea aquatica* Forsk). In all the locations it was noted that the plant had no damages from any natural enemies.

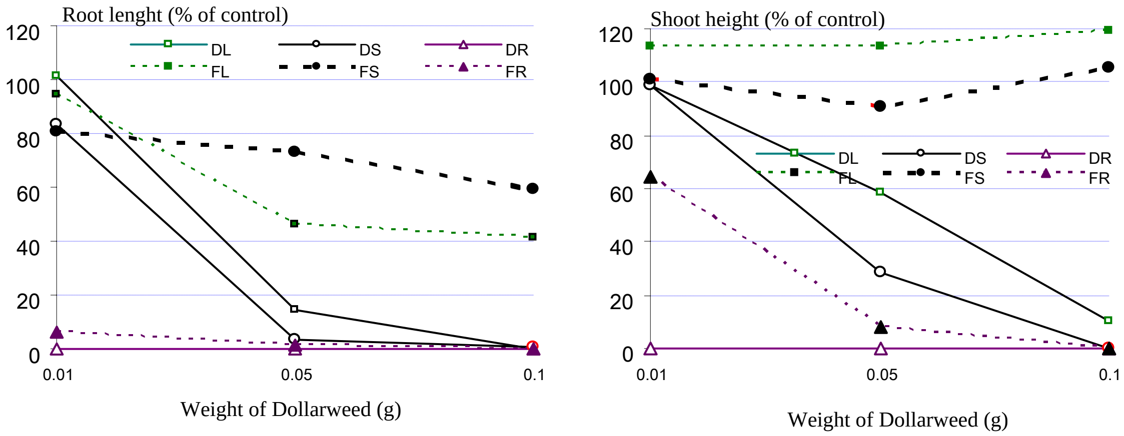

Figure 6. Effect of various parts of Dollarweed on growth of lettuce (root length and shoot height - % of control)

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Since the plant has shiny green leaves it is attractive as an ornamental plant. Thus, the spread can be faster through the human intervention in the ornamental plant market. The plant roots at every node, new shoot emerge from the bud at leaf axil, and young shoots (2nd -3rd leaf from shoot tip) can be used for propagation. The plant can grow rapidly (to reach 226 leaves within 168 days or around 6 months), and could form a pure colony and compete against other plants. In the case of grown together with the Asiatic pennywort, the leaf number and leaf area of Dollarweed is affected, however, its mass (dried weight) could be much higher than Asiatic the pennywort. Moreover, the plant exhibits allelopathic effects on other plants. These characters demonstrate the strong weedy nature of the species (Muenscher, 1980). Moreover, the plant is non-native to the Asian continent, thus without natural enemies in the introduced environments, including Thailand.

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INTEGRATED WEED MANAGEMENT IN WHEAT (*Triticum aestivum* L.) IN GANGETIC ALLUVIUM SOILS OF EASTERN INDIA

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Abstract: Field experiments revealed that all weed management methods improved the growth and yield parameters of wheat. The treatment having hoeing + isoguard plus at 1250 g ha^{-1} induced the best growth and yield, which was comparable to manual weeding on two occasions. Among non-chemical treatments hoeing with a Dutch hoe was comparable to a single manual weeding. Different categories of weeds viz. grasses, sedges and broad leaves reduced the grain yield by 34 per cent. Hoeing + isoguard plus was the most effective treatment for lowering the population and dry weights of all weeds. Isoguard plus was the most economical treatment which gave highest net return and benefit over other methods of weed management; while hoeing with Dutch hoe was much more cost effective among non-chemical treatments.

Key words: Hoeing, isoguard-plus, wheat.

Introduction

To mitigate the food production of increased rate of population in India in the future, the area under wheat has to be increased in non-traditional states such as West Bengal, Assam and other parts of eastern India by adopting sound agro-techniques such as, introduction of short-heat tolerant high yielding cultivars, management of inputs and control of abiotic and biotic stresses. Among the biotic stresses, weeds play a significant role in deciding the productivity of wheat crop. Tiwari and Parihar (1993) stated that 34.5 per cent loss of wheat grain could be reduced through integrated approach of weed management. This system also minimizes ground water contamination and health hazards, ensures food safety, and protection of endangered species and the non-development of herbicide resistance weeds. Keeping these views in mind an integrated approach of weed management was carried out during winter seasons of 2004-05 and 2005-06 using the high yielding dwarf variety of wheat (PBW 343) to evaluate the effectiveness of different weed management practices to study the effect of different methods of weed management in new alluvial zone on growth and yield of wheat crop, determine the effect of different categorized weed pressure on growth and yield of high yielding variety of wheat, and calculate the economics of weed management practices.

Materials and Methods

A field experiment was carried out in the Research Farm of Bidhan Chandra Krishi Viswavidyalaya, Kalyani, Nadia, West Bengal, India during winter seasons of 2004-05 and 2005-06 with eight treatments comprising of – hoeing with Dutch hoe at 21 days after sowing (DAS); isoguard plus (isoproturon 750 g a.i + 2, 4-D sodium salt 500 g a.i) at 1250 g ha^{-1} at 28 DAS; hoeing with Dutch hoe at 21 DAS + isoguard plus 1250 g ha^{-1} at 28 DAS; metribuzin 175 g a.i ha^{-1} at 28 DAS; hoeing with Dutch hoe at 21 DAS + metribuzin 175 g a.i. ha^{-1} at 28 DAS; manual weeding once at 21 DAS; manual weedings twice at 21 and 42 DAS; and weedy check on wheat (*Triticum aestivum* L., cv. PBW 343). The experimental was laid out in a randomized complete block design with three replicates. The size of the gross plot was 19.32-sq. m. and that of net plot was 11.5 m² with 23-cm row spacing. The experimental soil was sandy loam with varying pH levels between 7.2 to 7.3, organic carbon 0.47 to 0.52%, available nitrogen 203.42 to 205.45 kg ha^{-1}, available phosphate 7.16 to 15.90 kg ha^{-1} and exchangeable K_2O 181.30 to 215.84 kg ha^{-1}. The maximum temperature varied from 23.1 to 37.6°C during 2004-05 and from 25.4 to 38.0°C during 2005-06 and the minimum temperature varied between 11.1 to 22.5°C during 2004-05 and between 8.3 to 21.4°C during 2005-06. The total precipitation during the period of investigation was 146.5 mm and 1.2 mm during 2004-05 and 2005-06, respectively. The relative humidity varied from 32.9 to 99.7% during first year and from 31.0 to 99.0% during second year of experimentation.

The crop was sown on 25.11.2004 and 30.11.2005 and harvested on 26.03.2005 and 20.03.2006, respectively. The crop was grown with recommended package of practices. Plant protection measures were taken as and when necessary during both the years. Necessary observations on crop and weeds were recorded at the appropriate times.

Results and Discussion

Effect on growth parameters of the crop

Plant height, tiller density and dry matter production of wheat (Table 1) was significantly increased by the adoption of all methods of weed management when compared to the weedy check plot. Improvement in plant height is supported by the findings of Sardana *et al.* (2001) and Pandey and Verma (2002), and increase in tiller density is confirmed by the findings of Dixit and Bhan (1997).

Table 1. Growth parameters of wheat as influenced by weed management practices.

Treatments	Plant height at harvest (cm)		Tiller density (m^{-2})		Dry matter production (g ha^{-1})	
	2005	2006	2005	2006	2005	2006
Hoeing with Dutch hoe at 21 DAS	87.3	74.8	256	263	76.9	85.3
Isoguard plus 1250 g ha^{-1} at 28 DAS	91.2	77.5	284	281	82.6	87.2
Hoeing at 21 DAS + Isoguard plus 1250 g ha^{-1} at 28 DAS	90.7	78.5	282	261	84.2	88.9
Metribuzin 175 g ha^{-1} at 28 DAS	91.0	76.7	267	260	73.8	82.9
Hoeing at 21 DAS + Metribuzin 175 g ha^{-1} at 28 DAS	91.0	76.7	282	284	77.5	84.2
One manual weeding at 21 DAS	87.1	73.3	265	256	74.7	82.2
Two manual weedings at 21 and 42 DAS	93.2	78.1	295	309	83.0	89.5
Weedy check	85.2	66.2	241	234	53.3	66.8
CD (p=0.05)	1.95	4.55	30.3	35.8	7.3	9.1

DAS = days after sowing.

Effect on yield components of the crop

Spike density, spike length, spike weight, filled grain density and test weight of grain (Table 2) were influenced due to adoption of weed management practices. Maximum values of the said components were obtained from the two manual weedings closely followed by the treatment obtaining hoeing + isoguard plus. This finding collaborates with the findings of Marwat *et al.* (2003).

Effect on grain and Bhusa yields of wheat

Grain yield of wheat increased significantly in the treatments which used hoeing + isoguard plus, hoeing + metribuzin and two manual weedings (Table 3). Other weed management practices such as the application of isoguard plus alone, metribuzin alone, one manual weeding and hoeing alone also improved the grain yield of wheat. This finding is supported by the findings of Sardana *et al.* (2001), Singh *et al.* (2002) and Marwat *et al.* (2003).

Table 2. Yield components of wheat as influenced by weed management practices

Treatments	Spike density (m^{-2})		Spike length (cm)		Filled grains per spike		1000 grain weight (g)	
	2005	2006	2005	2006	2005	2006	2005	2006
Hoeing with Dutch hoe at 21 DAS	247	253	9.03	10.83	36.2	38.8	36.33	36.53
Isoguard plus 1250 g ha^{-1} at 28 DAS	274	275	9.37	10.73	37.0	40.3	39.17	38.70
Hoeing at 21 DAS + Isoguard plus 1250 g ha^{-1} at 28 DAS	271	252	9.20	11.03	45.3	48.0	40.83	38.58
Metribuzin 175 g ha^{-1} at 28 DAS	262	248	10.07	9.13	32.7	35.0	37.50	38.60
Hoeing at 21 DAS - Metribuzin 175 g ha^{-1} at 28 DAS	261	271	9.83	11.27	36.3	44.3	38.17	37.87
One manual weeding at 21 DAS	262	243	9.97	11.00	46.3	40.7	37.67	37.57
Two manual weedings at 21 and 42 DAS	268	278	10.30	11.67	47.7	51.7	40.67	43.00
Weedy check	225	206	9.63	9.10	32.3	34.0	32.33	32.48
CD (p=0.05)	15.5	10.6	NS	0.41	3.8	4.3	3.48	3.36

DAS = days after sowing, NS = Not significant

Table 3. Grain and Bhusa yields of wheat and production economics under weed management practices (Mean values of two years).

Treatments	Grain yield (g ha^{-1})	Bhusa yield (g ha^{-1})	Total cost (Rs ha^{-1})	Gross return (Rs ha^{-1})	Net return (Rs ha^{-1})
Hoeing with Dutch hoe at 21 DAS	30.54	50.31	17473	35033	17560
Isoguard plus 1250 g ha^{-1} at 28 DAS	32.69	52.21	17247	37254	20006
Hoeing at 21 DAS + Isoguard plus 1250 g ha^{-1} at 28 DAS	34.00	52.55	18775	38433	19658
Metribuzin 175 g ha^{-1} at 28 DAS	31.62	46.17	17359	35384	18025
Hoeing at 21 DAS - Metribuzin 175 g ha^{-1} at 28 DAS	33.28	46.53	18887	36932	18045
One manual weeding at 21 DAS	30.68	47.80	26294	34782	8488
Two manual weedings at 21 and 42 DAS	32.72	51.80	31409	39018	7609
Weedy check	21.36	37.10	15945	24789	8844
CD (p=0.05)	3.06	7.72			

DAS = Days after sowing; Rate of grain and bhusa were taken as per local market rate.

Affect on weed populations and weed dry weight

The numbers of different weeds such as grasses, sedges and broad leaves and their dry weights were significantly reduced due to weed management methods (Table 4). Weed numbers and their dry weights were significantly reduced with the treatments of two manual weedings followed by the combined effect of hoeing and isoguard plus or metribuzin or from the use of any one chemical alone. This indicated that different categorized weeds could easily be controlled by the suitable combination of mechanical and chemical methods.

Table 4. Weed population and dry weight as affected by weed management practices in wheat.

Treatment	Weed population (m^{-2})				Weed dry weight (g m^{-2})			
	40 DAS		60 DAS		40 DAS		60 DAS	
	2005	2006	2005	2006	2005	2006	2005	2006
Hoeing with Dutch hoe at 21 DAS	12.5 (157.0)	13.6 (184.5)	14.0 (195.5)	13.7 (187.2)	15.5 (239.8)	12.8 (163.3)	17.0 (288.5)	16.2 (261.9)
Isoguard plus 1250 g ha^{-1} at 28 DAS	6.9 (47.1)	6.1 (36.7)	10.5 (110.3)	6.2 (38.0)	9.4 (87.9)	7.3 (53.3)	14.2 (201.1)	7.6 (57.2)
Hoeing at 21 DAS + Isoguard plus 1250 g ha^{-1} at 28 DAS	13.0 (168.5)	7.6 (57.3)	4.7 (21.6)	5.5 (29.8)	7.0 (48.5)	6.9 (47.6)	6.0 (35.5)	7.6 (57.2)
Metribuzin 175 g ha^{-1} at 28 DAS	5.8 (33.5)	5.0 (24.5)	11.4 (129.5)	3.6 (12.5)	8.7 (75.6)	5.1 (25.5)	14.7 (215.6)	6.0 (35.5)
Hoeing at 21 DAS - Metribuzin 175 g ha^{-1} at 28 DAS	5.7 (32.0)	4.8 (22.5)	6.7 (44.3)	4.4 (18.6)	6.7 (44.8)	4.9 (23.5)	6.0 (35.5)	6.2 (38.0)
One manual weeding at 21 DAS	12.1 (146.0)	7.9 (61.9)	15.9 (252.3)	6.4 (40.5)	13.2 (173.8)	7.7 (58.8)	19.8 (391.5)	8.1 (65.1)
Two manual weedings at 21 and 42 DAS	5.1 (25.5)	3.6 (12.5)	2.6 (6.3)	5.6 (30.4)	6.0 (35.5)	4.6 (20.7)	3.1 (9.1)	4.0 (15.5)
Weedy check	26.8 (717.2)	17.6 (309.3)	16.0 (255.5)	17.7 (312.8)	17.3 (298.8)	17.8 (316.3)	16.7 (278.3)	20.8 (432.1)
CD (p=0.05)	3.4	1.5	1.9	1.9	3.6	3.9	7.5	3.3

DAS = days after sowing, Transformation values ($\sqrt{x+0.5}$) were statistically analyzed, Figures within the parenthesis are actual values, Production economics and additional income from weed management practices

Economics of weed control

The additional incomes and benefits from weed management practices (mean values of two years) are presented in Table 5.

Table 5. Additional income from different methods of weed management practices.

Treatments	Cost of weeding (Rs ha^{-1})	Additional yields (g ha^{-1})		Additional income (Rs. ha^{-1})	Benefit from method of weed management (Rs. ha^{-1})
		Grain	Bhusa		
Hoeing with Dutch hoe at 21 DAS	1364	9.18	13.21	10244	8880
Isoguard plus 1250 g ha^{-1} at 28 DAS	1162	11.33	15.11	12464	11302
Hoeing at 21 DAS + Isoguard plus 1250 g ha^{-1} at 28 DAS	2526	12.64	15.45	13692	11166
Metribuzin 175 g ha^{-1} at 28 DAS	1262	10.26	9.07	10595	9333
Hoeing at 21 DAS - Metribuzin 175 g ha^{-1} at 28 DAS	2626	11.92	9.43	12143	9517
One manual weeding at 21 DAS	9240	9.32	10.70	9993	753
Two manual weedings at 21 and 42 DAS	13860	13.36	14.70	14229	369
CD (p=0.05)	Nil	-	-	-	-

DAS = Days after sowing

The cost of cultivation, gross return and net return (mean values of two years) are presented in Table 5. Manual weeding either once or twice was more expensive than mechanical (hoeing), chemical (herbicide) or combination of both. As a result, net returns were obtained from the latter treatments. The highest net return (Rs. 20,006/-) was obtained from the Isoguard Plus alone, closely followed by the combination of hoeing and Isoguard Plus (Rs. 19,658/-). Though the maximum additional income (Rs. 14,229/- ha^{-1}) was received from two manual weedings, the lowest benefit amounting Rs. 369/- ha^{-1} was obtained from the same treatment due to the very high costs of weeding. Maximum benefit (Rs. 11,302/- ha^{-1}) was obtained from the plot receiving isoguard plus only followed by the treatment having hoeing + isoguard plus (Rs. 11,166/- ha^{-1}). The benefits obtained from the other treatments were also high when compared to manual weeding. Among non-chemical treatments hoeing was more cost-effective treatment than manual weeding

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INVESTIGATION ON THE WEED SEEDBANK UNDER DIFFERENT WEED MANAGEMENT METHODS IN YOUNG TEA [*Camellia sinensis* (L. Kuntze)]

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Abstract: A field experiment was conducted at the low country station of the Tea Research Institute, Sri Lanka during the period of 1994-'95 to investigate the density of weed seed bank in soil in relation to different weed management methods during the first year of planting tea. Manual weeding (hand and slash weeding) at 2-18 week intervals and several herbicides at varied intervals were imposed on plots with or without mulching. Weed seed banks were determined using both seed germination and "Malone's seed extraction" methods before and 12 months after imposition of treatment (MAT). There was no significant difference in seed density between plots prior to imposition of treatments. However, weed seed density at both 0-5 and 5-15 cm depths was significantly affected by the treatments at 12 MAT. AT the 0-5 c soil depth, the highest weed seed density of 4168 m^{-2} was observed from mulched plots hand-weeded every 6 weeks. Comparable densities were found in manually weeded plots except those weeded every two weeks and those treated with 2, 4-D + paraquat and glufosinate ammonium. The greatest seed density of 5520 m^{-2} at 5-15 cm depth was from unmulched plots, slash weeded every 6 weeks and this was comparable to that of hand weeding every 18 weeks. The least weed seed density at each depth was recorded from the plots treated with oxyfluorfen + paraquat. The seed density at 0-5 cm depth in all herbicide treated plots and that was manually weeded at every 2 weeks was comparable, whereas at the soil depth of 5-15 the seed density was not significantly different among treatments except for slash weeding every 6 weeks. A majority of seeds were found in plots weeded every 6 weeks was of desirable herbs. Hand weeding at 6-8 week intervals or application of oxyfluorfen + paraquat or glyphosate was found to be effective in the mitigation of weed seed bank in tea soil.

Key words: Weed seedbank, manual weeding, chemical weeding, mulching, weed management in tea.

Introduction

Weed management in tea plantations is a critically important operation, particularly during early establishment tea (Somaratne, 1988), as weeds interfere with tea and interrupt the major field operations such as plucking, manuring and pruning. Weed menace becomes more adverse with delayed weeding for more than three months (Prematilake *et al.* 1999). Among the possible causes of heavy occurrence of weeds in tea fields, presence of a 'weed seed stock' which is termed "weed seed bank" in soil a major factor as it releases viable seeds for germination followed by growth of weeds as a continued process. Weed seed bank has been referred to as the reserve of seeds present in the soil and on its surface (Roberts, 1981). Seed banks in cultivated soils are derived from seeds produced *in situ* and those that have been introduced from elsewhere (Froud-Williams *et al.* 1983). Quantitative studies on weed seeds in soil under different agronomic conditions are particularly important in providing a basis for advice on control measures (Wilson and Cussans, 1975). Furthermore, investigations on species composition provide a basis for estimating the future weed infestation (Ball and Miller, 1989) thereby helps in the planning of control strategies (Roberts, 1981).

Understanding of the size of the weed seed bank in soil under various weed management techniques in young tea is necessary and will provide an insight as to how and when such techniques are imposed for more sound and cost-effective weed management. The objectives of the present study is therefore to investigate the density of weed seed bank in soil and its composition in relation to various manual and chemical weed management methods in newly planted tea at low altitude.

Materials and Methods

A field experiment was conducted during June 1994 to December 1995, at the low country station of the Tea Research Institute, Sri Lanka, where the site (at Ratnapura) is located at an elevation of 30 m amsl (the latitude: 6^0 41'.N & longitude 80^0 24' E). The soil type is an Ultisol sandy loam, annual rainfall is 2500-3000 mm and the mean ambient temperature is 28°C.

Tea (*Camellia sinensis* L. Kuntze), Clone-K 16/3, was planted in holes of 45 cm deep at a spacing of 1.2 m x 0.6 m in June, 1994. The test field was previously rehabilitated with Mana grass (*Cymbopogon confertiflorus*) for a period of 24 months and well cleaned. Forty plots each consisting of 30 tea plants, were demarcated leaving two rows of tea as guard rows in each plot. Shade was provided by planting of Gliricidia (*Gliricidia sepium*) at a spacing of 2.4 m x 3.0 m.

Treatment combinations

Five manual weeding (hand and slash weeding) treatments (T1-T4) and five herbicide combinations (T6-T9) were applied to mulched and un-mulched plots (Table 1). Hand weeding was carried out in mulched plots at intervals of 2 (T1), 6 (T2), 12 (T3) and 18 (T4) weeks and slash weeding of un-mulched plots was done at 6-week intervals (T5). Herbicide treatments applied to mulched plots included Round up® (glyphosate, 36%) at 0.99 kg a.i. together with kaolin at 3.42 kg ha^{-1} (T6), Fernoxone® (2, 4-D, 73%) at 0.73 kg a. i. + Gramoxone® (paraquat 20%) at 0.15-0.22 k a.i.ha^{-1} (T7) and Basta® (glufosinate ammonium, 15%) at 0.2 kg a.i.ha^{-1} (T8), Goal-2E® (oxyfluorfen, 24%) at 0.29 kg a.i.ha^{-1} + paraquat at 0.15-0.22 kg a.i. ha^{-1} were given to mulched (T9) and un-mulched plots (T10). The experimental design was RCBD with four replicates. All plots were hand weeded prior to allocation of treatments. In the mulching treatments, tea inter rows were mulched with *Cymbopogon* grass at a rate of 37 tonnes ha^{-1} soon after planting tea and repeated twice. In un-mulched plots (T5), the weeds in tea inter-rows were retained as a live ground cover, slashed and removed every six weeks. Oxyfluorfen (T9 and T10) was first applied to the bare soil soon after planting tea prior to mulching. Another, three applications of oxyfluorfen + paraquat and the other herbicides alone or as mixtures were applied between tea rows when weeds were 10-15 cm tall at number of rounds. A knapsack sprayer fixed with a Poli jet nozzle (Orifice size 042) was used to spray all herbicides at 550 l ha^{-1}. In all herbicide-treated plots, unattended weeds by herbicides were hand-weeded prior to application of fertilizer mixture (T-200 at 1500 kg ha^{-1} yr^{-1}), at two-month intervals.

Weed seed bank determination

The weed seed density was assessed following the germination method as described by Brenchley and Warrington (1930) and Roberts (1970) and was supported by additional seed extraction (Malone, 1967). A total of 15 soil samples were extracted randomly to the depth of 0-5 and 5-15 cm soon after plot marking (from an area of 202 cm^2) in June 1994, within and between of tea rows in each plot, using a metal pipe (4.3 cm diameter) and stored in bags at a mean temperature of 30°C until processing. Samples from the same depth in each plot were bulked to make a composite sample weighing 800 g and each was placed on a tray. Five trays filled with sterilized sand were used as controls. All trays were then placed in the screen house and were protected from contamination by foreign air-borne weed seeds. Soil was kept moist throughout facilitating weed seed germination and frequently re-arranged to avoid differential light effects. A similar sampling process was followed 12 months after imposition of treatments (MAT). Then, seedling emergence, counting, identification and removal of each weed were continued until no further emergence occurred by disturbing soil occasionally to facilitate germination of buried seeds. Subsequently, the balance of seeds in soil was isolated using Malone's seed extraction method in order to determine the actual number of viable seeds present.

Statistical analysis

Count data (Seed No.) were log transformed and they were subjected to ANOVA using the SAS package. Mean separation was done using Least Significant Difference (LSD) at p=0.05. The original values were used for interpretation of results.

Results and Discussions

Weed seed density prior to imposition of treatments

There were large number of weed seeds with a wide variation, however, no significant differences in seed density was observed between plots before planting at both depths of 0-5 and 5-15 cm (Table 1). However, the highest (2903 m^{-2}) and the least (815 m^{-2}) weed seed densities at 0-5 cm depth occurred in plots to be hand weeded every 12 and 18 weeks, respectively.

Table 1. Mean density of soil weed seed bank prior to and 12 months after planting tea.

Treatment No.	Mean* seed density (No. m^{-2})							
	Prior to planting				12 months after planting			
	0-5 cm depth		5-15 cm depth		0-5 cm depth		5-15 cm depth	
	Log	Actual	Log	Actual	Log	Actual	Log	Actual
1	3.05	1346	3.20	1976	3.11	1601	3.048	1481
2	3.17	2026	3.21	1791	3.57	4168	3.290	2163
3	3.43	2903	3.25	2199	3.51	3293	3.233	2033
4	2.46	852	3.13	2371	3.55	3723	3.368	2653
5	3.06	2223	3.43	2891	3.39	2709	3.715	5519
6	3.27	1890	3.21	2297	3.12	1430	3.130	1506
7	3.10	1667	3.20	1618	3.29	2514	3.100	1314
8	2.61	2322	3.32	2557	3.29	2514	3.273	2276
9	3.01	1235	3.10	1816	3.00	1241	2.998	1026
10	2.36	1223	2.94	1235	3.00	1241	3.180	1621
LSD (p=0.05)		NS		NS		0.434		0.39

Mulching and - manual weeding: T1-every two weeks; T2-every six weeks; T3-every 12 weeks; T4-every 18 weeks; & chemical weeding: T6-glyphosate (0.99 kg a.i ha^{-1}) + kaolin (3.42 kg ha^{-1}); T7-2, 4–D (0.73 kg a.i ha^{-1})/ paraquat (0.15-0.22 kg a.i ha^{-1}); T8- glufosinate ammonium (0.2 kg a.i ha^{-1}); T9-oxyfluorfen (0.29 kg a.i ha^{-1}) / paraquat (0.15-0.22 kg a.i ha^{-1}); Un-mulched and T5-slash weeding-every six weeks, and T10-oxyfluorfen (0.29 kg a.i ha^{-1})/paraquat (0.15-0.22 kg a.i ha^{-1}).
* Mean value among 4 replicates, NS = not significant at 0.05 level

At the depth of 5-15 cm, the highest density (2890 m^{-2}) was observed in plots that were slash-weeded every 6 weeks. However, the seed densities observed in the present study (up to 7900 m^{-2}) at the depth of 0-15 cm is relatively lower when compared with the 13000-27000 seeds m^{-2} observed in old tea fields as reported by Eden (1949) and this may be attributed to previous ground cover with establishment of Mana (*C. confertiflorus*) followed by ground cleaning before planting of tea. The variation in seed density is attributed to the *in situ* weed density, proximity to source, soil conditions, burial depth, cropping regime, fertilizer and organic manures *etc.* (Boli and Watkinson, 1993).

Impact of various manual and herbicidal treatments on weed seedbank, 12 MAP

Though not significant, the weed seed density at the depth of 0-15 cm was varied with time *i.e*. before and 12 MAP, having relatively many seeds in plots weeded manually at intervals exceeding two weeks (Figure 1). However, there was a significant difference in seed density between treatments in 1995, *i. e*. 12 MAT, to which the differences in seed number at both depths (0-5 and 5-15 cm) largely contributed.

Impact of manual weeding on the seedbank

The least seed density obtained from hand weeding at two-week intervals (weed free treatment) (Fig. 1) was significantly less ($p<0.05$) than that of other hand weeding treatments but comparable to that of all herbicide treatments in 1995. This was also relatively lower than that reported prior to imposition of treatments. Reduced seed density may be attributed to the lower seed influx from *in situ* production. Weed removal during the onset of vegetative growth prevented reproductive output. However, greatest species diversity observed in this treatment may probably be due to less intense intra-specific competition. In contrast, though not significant, a relatively greater weed seed count in all other manual weeding treatments compared to those in 1994, may mostly be attributed to the subsequent replenishment of the seedbank with *in situ* seed production, which further depended upon the frequency of weeding and the type of weeds present. A higher seed density recorded in mulched plots hand weeded every 6 weeks was largely due to (66%) the greatest density recorded (4168 m^{-2}) at 0-5 cm depth (Table 1). Such higher density was mainly attributed to the production of seeds by short season species such as *Mollugo pentaphylla*, *Oxalis corymbosa*, *Stemodia verticillta*, *Peperomia pellucida* and *Lindernia cordifolia,* which are considered to be favourable for tea.

Total weed seed density (log No./m²)

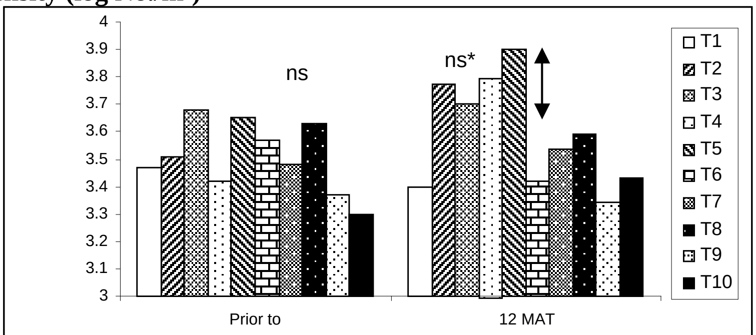

Fig.1: Mean total weed seed density (Log values) in the soil seedbank at the depth of 0-15 cm *prior to* and *12 months after imposition of treatments [MAT]*

LSD (0.05)

For treatments, refer to Table 1.

Higher densities to which almost 60% of seeds were contributed from 0-5 cm depth, in plots manually weeded at 12 and 18 week intervals were the result of high *in situ* seed rain from species such as *Borreria latifolia*, *B. leavis*, *Cleome viscosa*, *O. barellieri,* and *Digitaria sanguinalis.* The greatest seed density recorded from unmulched plots, slash weeded every 6 weeks resulted from an initial high density (5115 m^{-2}) as well as the *in situ* production of seeds from weeds left as a live ground cover, where almost 67% of seeds were located within (5520 m^{-2}) 5-15 cm depth (Table 1). Cracks formed by excessive drying during a dry spell followed by washing out of seeds with rains into the cracks and coarse textured soil might explain such an increased density in the deep layer. These are in agreement with the findings of Harper (1977) and Hopkins and Graham (1983).

Weed seed density in chemically treated plots

Weed seed density in herbicide-treated plots did not vary before and 12 MAP indicating the maintenance of the *status quo* from weed control. The treatments of oxyfluorfen + paraquat with and without mulching, which had the least seed density and glyphosate had resulted in successful control of an array of weeds preventing further seed replenishment (Table 1). Many authors have also concluded that effective weed control with herbicides has led to a decreased seedbank (Hurle, 1974; Fogelfors, 1991). The higher seed density in un-mulched *vs* mulched plots treated with oxyfluorfen + paraquat may be attributed to the immigration of seeds from elsewhere and deposition on the ground. Slight increase in seed number in 2, 4-D/paraquat treated plots was ascribed to the seed influx from some uncontrolled and tolerant weed species present until hand pulling. It could be concluded that the weed seedbank in soils of young tea fields could be kept under control with rational use of herbicides such as oxyfluorfen alone or with paraquat or glyphosate at correct time, or manual weeding at 6-8 week intervals.

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RESIDUAL EFFECTS OF PROPANIL IN SIMULATED RICE FIELD ENVIRONMENT

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Abstract: 3,4-dichloroaniline (3,4-DCA), the major degraded product of propanil, is detected at stearic acid-modified electrodes, using steady-state amperometry. This reliable, convenient analytical method provides a reasonable minimum detection limit of 2.0×10^{-8} mol dm^{-3}. Although 3,4-DCA is electroactive at bare glassy carbon (GC) electrodes, an uncontrollable noise prevents precise measurements at such electrodes. Consequently, stearic acid-modified GC electrodes reduce noise, allowing the detection 3,4-DCA at lower levels. This methodology is successfully extended for the detection of 3,4-DCA at ppm levels in water of a model rice bed after application of the recommended dose of propanil.

Key words: 3,4-DCA, electrochemistry, pesticidal fate, propanil.

Introduction

Propanil [N-(3,4-dichlorophenylpropionamide)] is a very effective and widely used post emergence selective weedicide in rice fields (Vidotto, 2007). It is degraded in the environment by several means: chemical or enzyme catalysed hydrolysis, or microbial or photochemical decomposition (Fava, 2005). One or more of these modes are always possible in the real environment, and hence the rate of the formation of the principal degraded product, 3,4-DCA, has many contributions. Consequently, it would be a complex task to predict the rate of degradation of propanil in the real environment with respect to each individual mode. Nevertheless, the fate of propanil in the environment can be indirectly determined by monitoring the levels of 3,4-DCA. Quantitative determination of pesticides and/or their residues in the Sri Lankan agricultural environment is a crucial issue because the effect of a pesticide in the local environment, where soil constituents and atmospheric conditions are different from those of the country of manufacture, would probably not be the same as reported.

Gas chromatographic analysis followed by quantification with a nitrogen-phosphorus detector (NPD) is recommended by the US Environmental Protection Agency for the detection of propanil at ppb levels (USEPA, 2006). Further, many attempts have recently been reported to include a solid phase extraction (SPE) step, which integrates sampling, extraction, concentration and sample introduction in one step, in conjunction with chromatographic analysis (Namera, 1999; Krasnova, 2000). Similar techniques can also be employed for the detection of 3,4-DCA. Although these advances in analytical chemistry improves the minimum detection limit and the sensitivity of detection, such techniques are associated with some inherent drawbacks such as economic factors, complexity of analysis, environmental aspects, etc. Electrochemical methods, in particular, amperometry, is a viable alternative in this regard despite the fact that it may not be sensitive to ppb level unless a preconcentration step similar to that in SPE is incorporated.

In this research, levels of 3,4-DCA is regularly monitored in surface water as well as in the leachate in a simulated rice field environment after introduction of propanil. Use of steady-state amperometric methods at stearic acid-modified GC electrodes is successfully demonstrated for the determination of 3,4-DCA with adequate accuracy and precision.

Materials and Methods

Three rice beds of the same dimensions (1.0 m × 1.0 m) were prepared in a polytunnel, each of which was filled up to 0.25 m with paddy soil obtained from the Rice Research and Development Institute (RRDI), Batalagoda. Weeds were introduced to the soil beds and allowed to grow under saturation. After weeds were grown, the field was filled up to 3 cm with water, and surface water and leachates were analyzed for any residual 3,4-DCA that may have been present from previous rice cultivation cycles. Water was then completely drained, and the recommended dose of Gramoxone was introduced. After 2 days of drying, water was reintroduced and normal agricultural practices were conducted, and rice plants were cultivated. The recommended dose of propanil was introduced 10 days after cultivation, and representative water samples were collected to determine the levels of 3,4-DCA using amperometric methods reported elsewhere (Navartne, 2001).

Propanil applied to the rice field was purchased from the local market, whose strength was detrmined using a technical grade (96 %) sample of propanil. A standard sample of 3,4-DCA (98 %) was purchased from Fluka. Acetonitrile, used for the preparation of 3,4-DCA standards, was distilled prior to use, and the other chemicals used were of analytical grade.

Cyclic voltammetric analysis and amperometric determination were performed in a three-electrode cell, consisting of a saturated calomel reference electrode (SCE), a platinum counter electrode and a stearic acid coated GC working electrode. All potentials were reported with respect to the SCE. The electrolyte was 0.1 mol dm^{-3} NaCl dissolved in the mixed H_2O/CH_3CN (1:3) medium. CV-1B Cyclic Volatammograph (Bioanalytical Systems, USA) provided suitable potential programs for all electrochemical experiments, and the responses were recorded on an X-Y Recorder (Bioanalytical Systems, USA).

Results and Discussion

Cyclic voltammetric studies of 3,4-DCA at bare GC electrodes in 0.1 mol dm^{-3} NaCl show an intense oxidation peak at +0.70 V, which is probably associated with the oxidation of the amine moiety (Figure 1). Consequently, many potentials in the vicinity of +0.70 V were attempted for amperometric determination of 3,4-DCA, and the amperograms obtained at +0.70 V produced the best quality. Hence, this potential was selected as the optimal potential for amperometric detection of 3,4-DCA. However, amperometric responses obtained at the bare GC electrode were noisy, probably due to electrode fouling. This situation was overcome by surface modification of the GC electrode with stearic acid, a nonelectroactive surfactant. Although cyclic voltammetric peak currents are decreased after coating, the performance of the stearic acid-modified GC electrode is better in many respects. For instance, minimum detection limit (MDL) and the sensitivity of the modified electrode for 3,4-DCA were determined to be 2.0×10^{-8} mol dm^{-3} (= 3 ppb) and 1.23 A mol^{-1} dm^3, respectively. The MDL of the amperometric method developed is comparable to sophisticated analytical methods, which is a great achievement in electroanalytical chemistry. A sample amperometric response obtained with sequential additions is shown in Figure 2. The calibration curve constructed using the current measurements of Figure 2 can be used for quantification of 3,4-DCA in water samples.

The levels of 3,4-DCA, detected using the amperometric method developed, after introduction of propanil show an initial increasing trend up to 4 days followed by a decrease up to the 8th day (Figure 3). After a period of 8 days, the concentration of 3,4-DCA levels off at an average value of 8.9×10^{-7} mol dm^{-3}. Although propanil is at undetectable levels after 7 days, residual levels of 3,4-DCA, which is stable and toxic than propanil, can be detected in water even after 50 days of propanil application. Initial increment of the levels of 3,4-DCA in the fields is due to the degradation of propanil applied to the fields. This degradation process is completed within a period of 4 days, according to the results obtained. Although the presence of propanil was not detected in this research, the results could be used for investigation of the fate of propanil in the rice field environment.

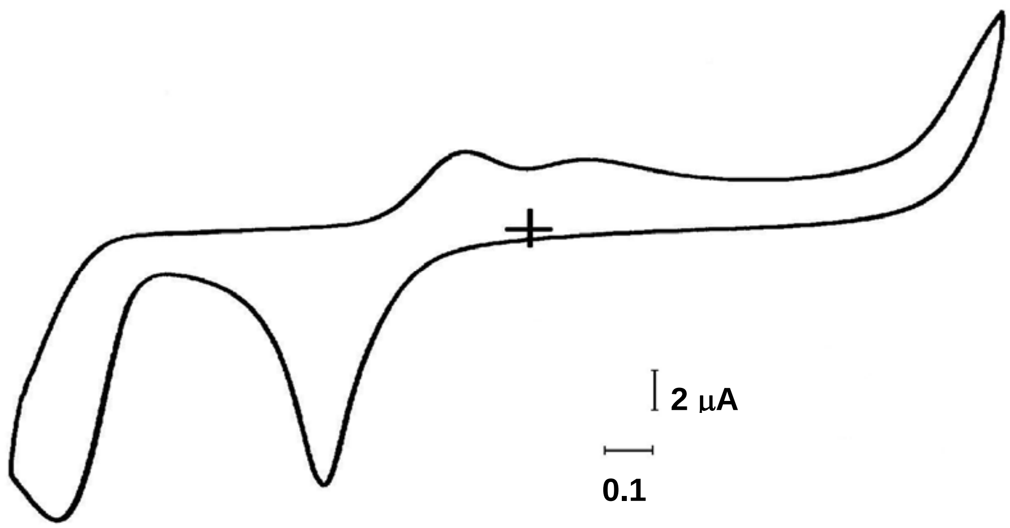

Figure 1. Cyclic voltammogram of 3,4-DCA (9.88×10^{-4} mol dm^{-3}) in 0.1 mol dm^{-3} NaCl (H_2O/CH_3CN 3:1) at bare GC electrode. Scan rate 100 mV s^{-1}, N_2 saturated, potential range +1.0 V to -1.0 V vs. SCE.

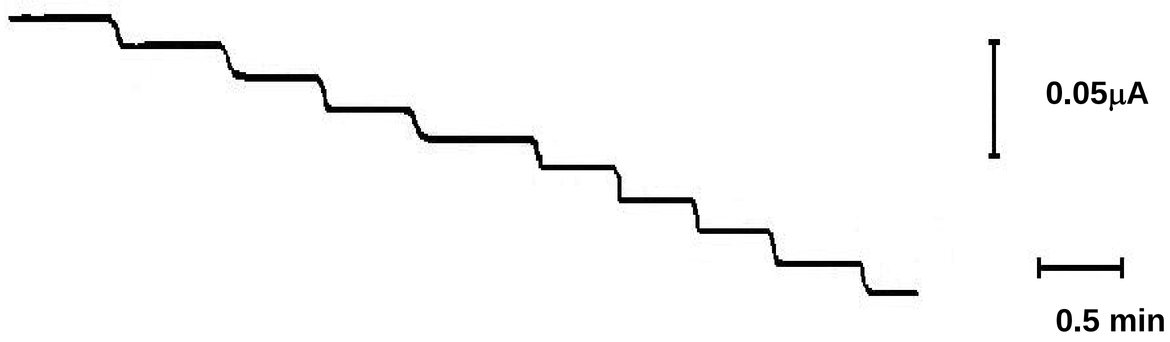

Figure 2: Steady-state amperometric responses of sequential additions of 50 µl of 1.0×10^{-3} mol dm^{-3} 3,4-DCA into 25.00 cm^3 of 0.1 mol dm^{-3} NaCl (H_2O/CH_3CN 1:3) at a potential of +0.7 V vs. SCE.

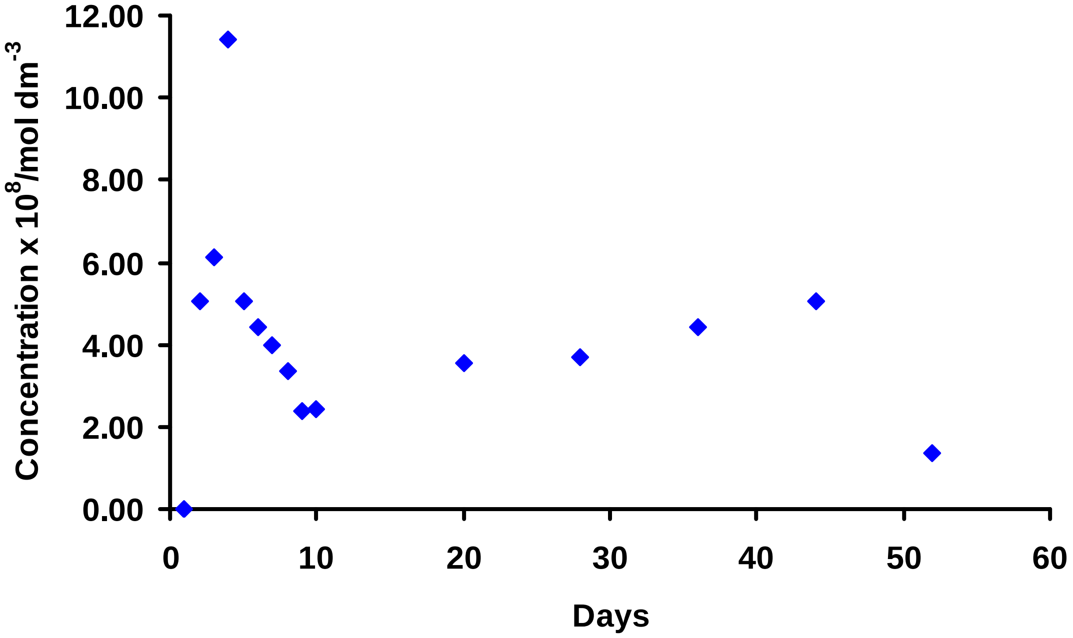

Figure 3. The average concentration of 3,4-DCA of water collected from three rice beds.

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LEAF DISCOLORATION OF SEASHORE PASPALUM (*Paspalum vaginatum*) cv. "SALAM" CAUSED BY CHLORIMURON-ETHYL

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Abstract: The study was conducted on the leaf discoloration of seashore paspalum (*Paspalum vaginatum*) cv. Salam caused by chlorimuron-ethyl in 6 golf courses in Hainan and Guangzhou during 2005 and 2006 respectively. The leaf discoloration of seashore paspalum occurred at ≥ 0.15 kg ha^{-1} of chlorimuron-ethyl regardless of the turf quality and temperature. In warm season turf zones of China, the application of chlorimuron-ethyl at 0.1 kg ha^{-1} is recommended for weed control when temperature increases above 15°C regardless of turf quality. The rate of 0.15 kg ha^{-1} of chlorimuron-ethyl is recommended in G or F turf quality of seashore paspalum.

Key words: Turfgrass, yellowing, quality, Seashore paspalum, chlorimuron-ethyl
Abbreviations: G, - good turf quality; F - fair turf quality; B - bad turf quality; DAT - days after treatment

Introduction

Seashore paspalum (*Paspalum vaginatum*) has been popularly adopted in the golf courses of Southern China. Chlorimuron -ethyl can be applied in Burmudagrass, *Zoysia* and Seashore paspalum to control weeds. However, although Chlorimuron-ethyl was safe to Burmudagrass, and *Zoysia* , sometimes, leaf discoloration of Seashore paspalum (*cv.* 'SALAM') has been observed after the application in lower temperature or in bad quality turf (Xue, 2001; Pan *et al.* 2006). The objective of this study was to determine the tolerance rate of 'Salam' seashore paspalum with chlorimuron in turfs of different quality during periods of different temperatures and the length of time needed for the turf to recover fully.

Materials and Methods

The study were carried out in 6 golf courses in southern China in 2005~2006 dividing into two experiments: high temperature (28°C~38 °C in Hainan, from Jun. to Oct., 2005): with chlorimuron at the rate of 0, 0.1, 0.15 and 0.2 kg ha^{-2} in Meishi golf (B = bad turf quality) and the West coast golf (G = good turf quality), Red valley golf (F = fair turf quality). Low temperature (15°C ~25°C in Guangzhou, from Nov. 2005 to Mar. 2006) with chlorimuron at the rate of 0, 0.1, 0.15 and 0.2 kg ha^{-2} in South Olympic golf (B) and the South Ttaoyuan golf (G), Hill view golf (F).

The turf quality in the courses used for the experiment were at three levels: Good (abbreviated as G), Fair (F) and Bad (B).Golf turf quality evaluation includes homogeneity, coverage, density, intensity, color, greenness, green lasting period, flatness, smoothness, flexibility and recovery ability (Huang and Liu, 2000). Good turf quality means well-balanced height and leaf color, no nakedness on the ground, uniform growth form of stem and leaves on the ground, flat surface, coverage between 97.5% to 100%, with >3 branches/cm^2, greenish-blue, high density of greenness, long green period, strong ability of recovery and no obvious appearance of color change after mowing; Fair turf quality means balanced turf, coverage between 90% to 97.5%, with 2~3 branches /cm^2, pale green, medium density of greenness, proper green period and recovery ability, obvious color change after mowing; Bad turf quality means uneven height and leaf color, exposed ground, non-uniform growth form of stem and leaves on the ground, uneven surface, coverage under 90%, with <2 branches /cm^2, gray green, low density of greenness, short green period, bad ability of recovery, obvious color change after mowing.

Treatments were applied as a broadcast spray in 150 l ha^{-1} of water with a SWOS-08 hand pump sprayer. Mowing was done 2 days before herbicide application and proper irrigation was done after herbicide application. The experimental design was a randomized complete block with three replications. Plot size was 1 m x 2 m. The degree of turf injury was based on scale from 0 to 5 where 0 = no injury; 1 = ignored minor leaf discoloration; 2 = minor leaf discoloration; 3 = obviously leaf discoloration with some plant necrosis; 4 = severe leaf discoloration and necrosis (this would not be acceptable); 5 = leaf discoloration leading to death. Injury ratings were done at 4, 7, 15 and 21 DAT. The time periods required for fully recovery were also recorded.

Results and Discussion

Leaf discoloration of Seashore paspalum (cv. 'SALAM') at different temperatures

The results of this experiment showed that the leaf discoloration of Seashore paspalum caused by chlorimuron varied according to the temperature. The leaf discoloration changed faster at high temperature (28°C ~38 °C) than at low temperature (15°C ~25°C). Pale-green leaves occurred 4 DAT. At high temperature, the time needed for full recovery was 12 DAT for turfs of good quality and 21 DAT for turfs of bad quality, at 0.2 kg ha^{-1} of chlorimuron. At low temperatures, the recovery times were 18 and 25 DAT for turfs of good and bad quality respectively (Tables 1 and 2).

Leaf discoloration of Seashore paspalum (cv. 'SALAM') at different rates of chlorimuron

The experiments indicated that the degree of leaf discoloration of Seashore paspalum caused by chlorimuron increased as the rate increased at both high and low temperatures. However, it was much different in G and B turfs. There was no damage in G turfs at both sites at the rate of 0.1 kg ha^{-1}. At the rates of 0.15 kg ha^{-1} and 0.2 kg ha^{-1}, injury occurred at both sites regardless of the temperature. However, the recovery was easier at 0.15 kg ha^{-1} than at 0.2 kg $ha^{-1}$of chlorimuron (Tables 1 and 2).

Table 1. The leaf discoloration of Seashore paspalum in different quality turf caused by chlorimuron-ethyl at high temperature (Hainan, China 2005).

Site (turf quality)	Rate (kg ha^{-1})	4 DAT	7 DAT	15 DAT	Full recovery (Days)
West Coast golf (A)	0	0	0	0	No injury
	0.1	0	0	0	No injury
	0.15	1.2	1.3	0	8
	0.2	1.5	2.0	1.0	10
Red Valley golf (B)	0	0	0	0	No injury
	0.1	1.0	1.1	0	8
	0.15	1.5	2.0	1.3	16
	0.2	2.2	3.0	2.0	20
Meishi golf (C)	0	0	0	0	No injury
	0.1	1.1	1.2	0	9
	0.15	2.1	2.2	1.1	16
	0.2	3.2	3.3	2.1	21

DAT=days after treatment Application time: Aug. 23rd~24th, 2005, Temperature: 30°C-38°C

Leaf discoloration of Seashore paspalum (cv. 'SALAM') in different turf quality

At both high and low temperatures, the degree of leaf discoloration of Seashore paspalum caused by chlorimuron was related to turf quality. For example, at high temperature, the degree of leaf discoloration of Seashore paspalum was 2.0 and 1.0 in G turf, but 3.3 and 2.1 in B turf at 7 and 15 DAT at 0.2 kg ha^{-1}of chlorimuron. The corresponding values at low temperature were 1.8 and 1.5 in G turf and 3.0 and 2.0 in B turf at 7 DAT and 15 DAT at 0.2 kg ha^{-1}of chlorimuron. The period required for full recovery were 18 and 12 DAT in G turf at high temperature, and 25 and 21 DAT in B turf at low temperature at 0.2 kg ha^{-1}of chlorimuron (Tables 1 and 2).

Table 2. The leaf discoloration of Seashore Paspalum in different quality turf caused by chlorimuron-ethyl at low temperature (Guangdong, China 2005~2006).

Site (turf quality)	Rate (kg ha^{-1})	4 DAT	7 DAT	15 DAT	Full recovery (Days)	Remarks
South Taoyuan golf (A)	0	0	0	0	No injury	Temperature goes up after application
	0.1	0	0	0	No injury	
	0.15	0	1.0	0	8	
	0.2	0	1.8	1.5	18	
Hill View golf (B)	0	0	0	0	No injury	Temperature goes down rapidly after application
	0.1	0	0	0		
	0.15	0	1.2	1.0	14	
	0.2	0	2.0	1.8	20	
South Olympic golf (C)	0	0	0	0	No injury	Temperature goes down slowly lasting for more than 110 days
	0.1	0	1.0	0	8	
	0.15	0	2.1	1.5	18	
	0.2	0	3.0	2.0	25	

DAT=days after treatment Application time: Feb 8, 2006, 15 -21°C in South taoyuan; Oct 13, 2005, South Olympic, 20-25 °C; Nov. 21, 2005, Hill view golf, 18°C -22°C.

Discussion

In golf turf in Southern China, it is suggested that the best time for application of chlorimuron in quality G seashore paspalum is after March and before October at 0.1~0.15 kg ha^{-1}. If the weed infestation is high, the rate should be increased to 0.17 kg ha^{-1} from May to August. In quality F turf, application of chlorimuron should be done after April and before September at 0.09~0.14 kg ha^{-1}. If the weed infestation is very high, the rate should be increased to 0.15 kg ha^{-1} from May to August. In quality B turf, application of chlorimuron should be done at 0.09~0.1 kg ha^{-1} when temperature increases up to 15°C. If the weed infestation is very high, frequent application with 0.09~0.12 kg ha^{-1} is recommended. In warm season turf zones of China, the application of chlorimuron-ethyl at 0.1 kg ha^{-1} is recommended for weed control when the temperatures are above 15^{o}C. The rate of application at 0.15 kg ha^{-1} of chlorimuron-ethyl is recommended in G or F turf quality of seashore paspalum.

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AVAILABLE TECHNOLOGIES AND FUTURE RESEARCH CHALLENGES FOR MANAGING WEEDS IN DRY-SEEDED RICE IN INDIA.

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Abstract: In India, dry-seeding (DSR) is extensively practiced in rainfed lowlands, uplands, flood-prone areas and in irrigated 'aerobic rice'. Weed infestation continues to be a serious problem in DSR. This review aims at (a) enlisting major weeds reported in DSR in India**,** (b) assess the yield losses due to weeds, (c) document and critically evaluate the available weed management options for DSR in India, and (d) discuss future research needs and strategies to continue to manage weeds effectively and economically in DSR in India. Abour 136 species belonging to 82 genera are associated with DSR in India. *Cyperus rotundus* L., *Cynodon dactylon* (L.) Pers., *Echinochloa colona* (L.) Link, and *E. crus-galli* (L.) P. Beauv. were reported as major problematic weeds in all DSR growing states of India. Aerobic soil conditions and dry-tillage practices, besides alternate wetting and drying conditions, are conducive for germination and growth of highly competitive weeds, which cause grain yield losses of 50–91%. Hand pulling, simple-tool-aided weeding, or hand- or animal-drawn-implement weeding is still common on small farms growing upland DSR in India. Several herbicides were found effective by researchers in managing weeds in DSR. However, an integrated weed management strategy involving both direct and indirect weed management technologies is essential for effective weed management in DSR in order to ensure higher rice productivity. Keeping in view of dynamic nature of weeds in DSR, future research on weed management in DSR in India should focus more on on-farm research. The weed management technologies generated through on-farm research needs to be integrated in the form of decision making frame works to aid farmers make better decision making and improve DSR productivity through effective and economical weed management.

Key words: Dry-Seeded Rice, India, Herbicides, Integrated Weed Management.

Introduction

Dry-seeded rice (DSR) is a traditional practice developed by farmers to suit the agro ecological conditions. Hand broadcasting or dibbling seeds into furrows or drill-seeding in rows by machine is used for seeding at shallow depths into moist, aerobic soil (Rao *et al.* 2007). Subsequently, rice is raised as a dry-land crop or the field is kept flooded during much of the season depending on the soil and climatic conditions. In India, dry-seeding is extensively practiced in rainfed lowlands, uplands and flood-prone areas (Misra *et al*. 2005). Dry-seeding is also practiced in irrigated 'aerobic rice' in Indo- Gangetic Plains (Singh *et al*. 2006). In rainfed uplands, rice is mostly dry-seeded. Upland rice occupies about 7.1 million hectares and accounts for 13% of total Indian rice area (Frolking *et al*. 2006). Approximately 6.2 million hectares of upland rice area is present in eastern zone alone, comprising of the states of West Bengal, Orissa, Assam, Bihar, eastern Madhya Pradesh, eastern Uttar Pradesh and the north-eastern hill region. The DSR is also grown in Tamilnadu, Karnataka, Andhra Pradesh and Kerala in the southern part of India. Productivity of DSR is often reported to be low, mainly due to the inadequate use of nutrient inputs, inefficient water management, and problems associated with weed management.

The Indian rice demand projection for sustaining the present level of calorie supply has been estimated to exceed 158 million tones by 2010, which amounts to an annual productivity growth of 2.4 percent (Siddiq, 2000). The target is no doubt challenging, but it is not unachievable given the potential opportunities and avenues yet to be exploited and rapid advances being made in crop research. To attain the target, it is essential to increase the prevailing low productivity of rainfed lowlands, uplands and flood-prone areas where DSR is grown in India. Yields of DSR have remained low. Weeds of DSR in India are quite adapted to the micro-ecological conditions and are one of the major reasons for low productivity (less than one ton ha^{-1}) of DSR (Rao *et al*. 2007). Thus one of the ways to meet the demands of increasing populations is to increase DSR productivity by identifying available weed management technologies for their effective management and prevent the losses caused by weeds. This review aims to (a) list major weeds reported in DSR in India**, (**b) assess the yield losses due to weeds, (c) document and critically evaluate the available weed management options for DSR in India, d) discuss future research needs and strategies to continue to manage weeds effectively and economically in DSR in India,

Importance of Weeds in DSR in India

Many farmers resort to dry-seeding of rice in rainfed areas to take advantage of early rains. In DSR the weeds emerge with the crop and offer competition for growth factors and thus affect the grain yield. In some instances, weed competition in DSR is so intense that failing to control weeds results in no rice yield (Moody and Mukhopadhyay, 1982). Thus, although labor and its associated costs may be reduced for crop establishment, other technologies such as effective weed management are essential to overcome constraints imposed by direct dry-seeding.

Weed flora in dry-seeded rice

The extent of weed competition depends on the type of weed species, its density, and cultural practices. Over 1800 plant species have been reported as weeds of rice in South and Southeast Asia. Among these, 70 species belonging to 56 genera are reported to occur in DSR in India (Moody, 1989). The present review of literature revealed that 136 species belonging to 82 genera are associated with DSR. The most commonly reported weeds in DSR in different states of India are presented in Table 1

Cyperus rotundus and *Cynodon dactylon* are major problems in upland condition particularly in poorly managed fields (Singh, 2005). *Ischaemum rugosum* and *Fimbristylis miliacea* were the most common weeds in DSR using no-tillage system, but were almost absent in transplanted areas (Singh *et al*. 2001). Changes in rankings of dominant weeds were observed by Singh *et al*. (2005) in response to dry-seeding of rice in the Indo-Gangetic plains. After four seasons of rice cropping, at 56 days after planting, *Echinochloa colona* and *Commelina diffusa* were dominant in DSR in comparison with *Cyperus iria* and *E. colona* in transplanted rice. Manual weeding 30 days after sowing led to increased importance of *I. rugosum* in DSR.

In rainfed dry-seeded lowland rice, the initial stage field situation is similar to that of uplands which hastens the germination and growth of weeds such as grasses, some sedges and broadleaf weeds. With the accumulation of water in the fields during later stages of the monsoon, certain specific water tolerant weeds and aquatic weeds besides wild rice pose problems (Moorthy and Saha, 2001). The dominant weeds before submergence were: *Echinochloa colona*, *Digitaria sanguinalis*, *Cyperus iria*, *C. rotundus* and *Commelina benghalensis*. The weeds that came after submergence were: *Fimbristylis miliacea*, *Scirpus articulatus* and aquatic weeds like *Ipomoea aquatica*, *Marsilea quadrifolia*, *Limnophila indica*, *Ottelia alismoides*, *Erriocaulon sieboldianum* and *Ceratoptus thalictroides* (Moorthy and Saha, 1997). Singh and Singh (2001) observed *Ipomoea aquatic*a, *Scirpus* spp., *Typha* spp., *Eichhornia crassipes, Nitella* spp., *Nymphea stillata* and *Oryza* spp. (volunteer./weedy rice) during post flood periods.

Table 1. Weed species most commonly reported in dry-seeded rice in India.

Weed species [a]	Family	State [b]
Aeschynomene indica L.	Fabaceae	HP,ORS,
Ageratum conyzoides L.	Asteraceae	ASM, CSG, HP, KTK, MP, ORS, UP, UTR
Alternanthera sessilis (L.) R. Br. ex DC.	Amaranthaceae	CSG, KTK, MP, ORS, UTL
Amaranthus viridis L.	Amaranthaceae	CSG, HAR, UP, UTR, MP
Ammannia baccifera L.	Lythraceae	TN, UP, UTR
Caesulia axillaris Roxb.	Asteraceae	CSG, MP, UTR, UP
Celosia argentea L.	Amaranthaceae	ORS, UP, UTR
Commelina benghalensis L.	Commelinaceae	ASM, BHR, CSG, HP, MP, ORS, RAJ, UP, UTR, KTK, DEL
Commelina communis L.	Commelinaceae	CSG, MP
Commelina diffusa Burm. f.	Commelinaceae	CSG, UTR
Cynodon dactylon (L.) Pers.	Poaceae	All DSR growing states in India
Cyperus difformis L.	Cyperaceae	ASM, BHR, HP, KTK, MP, ORS, TN, UP, UTR, WBL
Cyperus iria L.	Cyperaceae	ASM, CSG, DEL, HAR, HP, KTK, MP, ORS, TN, UP, UTR, WBL
Cyperus rotundus L.	Cyperaceae	All DSR growing states in India
Dactyloctenium aegyptium (L.) Willd.	Poaceae	ORS, JRK
Digitaria ciliaris (Retz.) Koel.	Poaceae	ASM, ORS, UP
Digitaria sanguinalis (L.) Scop.	Poaceae	CSG, MP, ORS, RAJ, UP, UTR, WBL
Echinochloa colona (L.) Link	Poaceae	All DSR growing states in India
Echinochloa crus-galli (L.) P. Beauv.	Poaceae	All DSR growing states in India
Echinochloa spp.	Poaceae	All DSR growing states in India
Eclipta prostrata (L.) L.	Asteraceae	CSG, KTK, MP, ORS, RAJ, TN, UP, UTR
Eleusine indica (L.) Gaertn.	Poaceae	ASM, CSG, DEL, KTK, MP, ORS, UP
Fimbristylis miliacea (L.) Vahl	Cyperaceae	ASM, CSG, MP, ORS, TN, UP, UTR, WBL
Ischaemum rugosum Salisb.	Poaceae	CSG, HP, MP, ORS, UTR
Leptochloa chinensis (L.) Nees	Poaceae	ORS, UP, UTR
Lindernia ciliata (Colsm.) Pennell	Scrophulariaceae	ORS, UP
Ludwigia perennis L.	Onagraceae	ORS, UTR, WBL
Marsilea quadrifolia L.	Marsileaceae	ORS, TN
Monochoria vaginalis (Burm. f.) C. Presl	Pontederiaceae	HP, UP
Oryza sativa L. (weedy rice, red rice)	Poaceae	KTK, ORS
Panicum repens L.	Poaceae	ORS
Paspalum distichum L.	Poaceae	BHR, UP, UTR
Paspalum scrobiculatum L.	Poaceae	ORS, UTR
Phyllanthus fraternus G.L. Webster	Euphorbiaceae	ASM, BHR, CSG, HAR, HP, MP, ORS, UP, UTR
Portulaca oleracea L.	Portulacaceae	KTK , ORS, UP
Setaria glauca (L.) P. Beauv	Poaceae	HP, JRK, ORS, UTR
Sorghum halepense (L.) Pers.	Poaceae	UP, UTR
Sphenoclea zeylanica Gaertn.	Campanulaceae	ORS, UTR
Trianthema portulacastrum L.	Aizoaceae	HAR, MP, TN, UP, UTR
Xanthium strumarium L.	Asteraceae	RAJ

[a] Based on the large number of references cited in this review. [b] ASM=Assam., BHR=Bihar., CSG=Chattisgarh., DEL=Delhi., HAR=Haryana., H.P.= Himachal Pradesh; JRK=Jarkhand., KTK=Karnataka., MP=Madhya Pradesh., ORS=Orissa., RAJ=Rajasthan., TN=Tamil Nadu., UP=Uttar Pradesh., UTR=Uttaranchal., WBL=West Bengal.

Irrigated dry-seeded 'aerobic rice' is a new water saving system being developed for lowland areas with water shortage and for favorable upland areas with access to supplementary irrigation (Belder *et al*. 2005). The major weeds associated with DSR rice in the aerobic rice system were *E. crus-galli* (L.) P. Beauv., *E. colona* (L.) Link, *Dactyloctenium aegyptium* (L.) Willd., *Leptochloa panicea* (Retz.) Ohwl, *Caesulia axillaris* Roxb., *Euphorbia hirta* L., *Lindernia* sp., *C. benghalensis* L., *Eclipta prostrata* (L.) L., *Trianthema portulacastrum* L., and *Portulaca oleracea* L. (Singh *et al*. 2006; 2007)

Weed competition in dry-seeded rice

Weed infestation continues to be a serious problem in DSR. Aerobic soil conditions and dry-tillage practices, besides alternate wetting and drying conditions, are conducive for germination and growth of highly competitive weeds, which cause grain yield losses of 50–91% (Choubey *et al*. 2001; Moorthy and Saha, 2002). The critical period of crop weed competition was identified to be 4 weeks (Tiwari and Singh, 1991) to 45 days (Naidu and Bhan, 1980) after sowing in DSR. If weeds are not removed in earlier stages, they compete severely with upland rice and will reduce grain and hence removal during early stages (30 DAS) is imperative. Pandey *et al*. (2003) reported that in order to obtain high yields of spring rice, the crop should be kept weed-free during 40-120 DAS.

Weed problems are also critical in rainfed dry-seeded lowland rice. Moorthy and Saha (2001) concluded that maintaining the weed-free condition through out the rainfed dry-seeded lowland rice crop growth (both pre-submergence and post-submergence stages) helps in increasing grain yield substantially. However, first 45 days (Singh *et al*., 1999), 30-60 days (Moorthy and Saha, 2005).after sowing period is considered the critical period for crop weed competition in this rice culture.

Available technologies

Research on weed management in DSR in India aimed at two broad approaches to weed control: indirect (preventative or anticipatory) techniques which are aimed at minimising weed development, and direct (active control) techniques aimed at controlling weeds which are present in the crop. It seems certain, however, that neither indirect nor direct weed control practices alone can solve weed problems in DSR. An integrated approach with better information on which to base weed control decisions will be needed to solve problems in DSR.

<u>Indirect weed control technologies</u>: In indirect means, most of the research in India is aimed at the possibility to manipulate the weed infestations by means of important agronomical practices such as tillage including stale seed bed, variety choice and intercropping.

Tillage: Land preparation plays an important role in DSR and could be an effective way of controlling weeds, thereby minimizing cost and maximizing profit (Ram, 2000). Increasing the number of tillage operations before sowing improved the crop stand, reduced weed infestation and, thereby, increased the yield significantly when compared with that achieved by conventional tillage. Summer ploughing rather than conventional tillage decreased weed dry weight at harvest by 15.8-53.2 % and increased grain yield by 47.4-56.3 % (Sharma, 1997). Ram (2000) showed that cost of tillage, labor used for weeding, total human labor used for cultivation, weed dry weight, weeding cost and total variable cost of cultivation were found to be lower with the use of a rotavator as compared to other conjunctive use of tillage implements. Singh *et al*. (2005) have shown that differing tillage practices in wheat selectively alter the relative abundance of grass and sedge weeds in the succeeding rice crop.

The stale seedbed technology generally recommended for raising small grains under weed-free upland conditions can be used in direct sown lowland rice as a low-cost, ecofriendly, and energy efficient weed control method. (John and Mathew, 2001). Renu *et al*. (2000) observed that the use of paraquat in a stale seedbed was more effective than mechanical weeding in DSR. Sharma *et al*., (2004) recorded higher net returns with the stale seedbed.

'Beushening', a time tested traditional rice cultivation system, is common throughout rainfed rice areas of eastern India. In this process weeds get buried in soil and rot along with some rice plants (Saha *et al*., 1999). After 10-12 days of beushening operation, the rice plants rise up and become erect. The remaining weeds are removed by manual operation. Beushening method of weed control is considered as good as hand weeding (Saha and Srivastava, 1993). Moorthy and Rao (1994) reported 77% control of weeds at 70 DAS through this practice alone. However, in the process of beushening, rice plants are killed and it becomes difficult to maintain the required plant population (Saha *et al*., 1999).

Cultivar: Adoption of weed-competitive genotypes is regarded as an effective tool for integrated weed management in DSR (Rao *et al*. 2007). Significantly lower weed populations and weed dry weights were recorded with SBR 34-69-1, Kalaguni (Hussain and Gogoi, 1996); Jagannath (Singh and Reddy, 1984); Cultivar Vaidehi and cv. Barh Avarodhi (Singh and Singh, 2001a); RR 51-1. RR 20-158 and Vandana (Mishra, 1997). The competitive ability of these varieties was attributed to lower light penetration in the canopy and early growth and quicker initial canopy coverage (Hussain and Gogoi, 1996), tall and fast growing nature (Gogoi and Kalita, 1989); rapid growth rate and more canopy coverage (Singh and Singh, 2001a); early emergence of plumule, initial higher leaf area index, tiller number, number of roots per plant, and root depth (Mishra, 1997).

*Intercropping***:** The use of green manure intercrops having bioherbicidal characteristics or weed smothering capability would have the additional benefit of adding biomass to soil (Singh *et al*., 2007). Intercropping with cowpea (fodder) and greengram (seed) lowered populations and dry matter accumulation of weeds (Hussain and Gogoi, 1996). Deferred seeding of black gram in DSR fields (30cm) with one weeding may be recommended for better yield, weed suppression, and better economics. Singh *et al*. (2007) concluded that application of wheat residues mulch at 4t ha^{-1} and Sesbania intercropping for 30 days were equally effective in controlling weeds associated with DSR and increasing net returns

Direct weed control technologies: Direct physical control methods include removal of weeds by hand, with weeding tools (hoe, scythe and spade), or with mechanical implements. Hand pulling, simple-tool-aided weeding, or hand- or animal-drawn-implement weeding is still common on small farms growing upland DSR in India. These methods are safe for the environment but are labor-intensive.

Manual weeding: The labor input per ha is 86 to 152 person-days for two weeding operations in DSR (Singh, 2005). The delay in first weeding increased the person days required for weeding. Quite often, weeding is delayed or cancelled due to poor weather conditions, the lack of labor or the expensive labour costs. Other problems with manual weeding include damage to the rice crop when weeders move through the field, and mistaken removal of rice instead of weeds because of the difficulty in distinguishing grassy weeds from rice. Manual weeding can be implemented only when weeds have reached a sufficient size to be pulled and it has an inherent opportunity cost. Manual weeding is therefore often practiced late as evidenced by yield loss comparisons of the effects of manual weeding at 21–30 DAS with those from the use of early post-emergence herbicides (Singh *et al.*, 2005a). Hand weeding twice (15 and 30 DAS) or thrice (15 30 and 45 DAS), is essential to obtain satisfactory weed control and rice grain yield. (Dutta and Gogoi, 1994).

Mechanical weeders: Mechanical weed control with the help of simple weeders is the only practical means for small and poor farmers to increase the productivity of rainfed DSR. Weeding with dry weeder (20 DAS) and mulching + dry land weeder (20 DAS) proved effective in reducing weed growth (Hussain and Gogoi, 1996). Mechanical weeding (20 DAS) followed by hand weeding (35 DAS) resulted in significantly lower weed dry weight (Nair *et al*. 2002). Sarma and Gogoi (1996) reported that in rainfed upland rice in India a manually operated peg-type dryland weeder and a twin wheel hoe were effective in weed control when used twice at 20 and 30 days after emergence. Another dryland weeder (with a straight-line peg arrangement) has also shown excellent performance across a range of soil types with varying soil moisture levels and weed intensity, providing a labor saving of approximately 57% compared with hand weeding (127 person-days ha^{-1}) (Subudhi, 2004).

Herbicides: Herbicides are considered to be an alternative/supplement to hand weeding method. However, in DSR, herbicide activity is unstable because of various environmental conditions. Most of the research on weed management in DSR is on evaluation of herbicides for managing weeds. Herbicides reported to be effective in DSR in India are summarized in Table 2. Behera *et al*. (1997) reported that resource rich, medium and large farmers prefer premergence application of butachlor followed by hand weeding, while the resource poor, small and marginal farmers preferred a finger hoe supplemented with hand weeding. Pendimethalin or pretilachlor as pre-emergence application followed by hand weeding were effective in controlling weeds in DSR rice in India (Singh, 2005). Singh *et al*. (2006) reported that, in the FIRBS, (i) ethoxysulfuron at 18 g a.i. ha^{-1} applied at 21 DAS was effective for controlling broadleaf weeds and (ii) fenoxaprop-p-ethyl + ethoxysulfuron at 50 + 18 g a.i. ha^{-1}, applied at 21 DAS, and pendimethalin followed by chlorimuron + metsulfuron at 1000 fb 4 g a.i. ha^{-1} applied at 3 DAS followed by 21 DAS gave broad-spectrum weed control.

Herbicide combinations control more species of weeds than single herbicide treatment (Behera and Jena, 1998). Triclopyr at 500 g a.i. ha^{-1}, bensulfuron at 60 g a.i. ha^{-1}, ethoxysulfuron at 18 g a.i. ha^{-1}, and 2,4-D (ester) at 500 g a.i. ha^{-1}, all applied at 21 days after seeding (DAS), were equally effective in enhancing rice grain yields by controlling broadleaf weeds (Singh *et al*. 2006). However, effective and economical management of mixed population of both grass and broadleaf weeds was observed with herbicide combinations such as fenoxaprop-p-ethyl+ethoxysulfuron at 50+18 g a.i $ha^{-}1$, applied at 21 DAS, and pendimethalin followed by (*fb*) chlorimuron+metsulfuron at 1000 *fb* 4 g a.i. $ha^{-}1$ applied at 3 *fb* 21 DAS (Singh *et al*. 2006).

Registered herbicides, when used within label specifications, are cost-effective and safe for both the rice crop and non-target species. Reports of herbicide-resistant biotypes (314 herbicide resistant biotypes of 183 species (110 dicots and 73 monocots) have been found in 58 countries (www.weedscience.org/in.asp)) and have increased rapidly in recent years due to widespread adoption of herbicides. In India, to date, no herbicide resistance is reported in the weeds of DSR. However, herbicides must be used judiciously, and when possible, should be supplemented by other weed control techniques.

Table 2. Herbicides and their combinations reported to be effective in controlling weeds in direct dry-seeded rice in India.

Herbicide	Mode of action, chemical group [a]	Class of weeds controlled [b]	Application time[c]	State[d]
Cyhalofop-butyl	Inhibition of acetyl CoA carboxylase (ACCase), aryloxyphenoxy-propionates (FOPs)	G,B,CS	POE	CSG, JRK,UP, UTR,
Fenoxaprop-p-ethyl	As above	G, CB, CS	POE	UP
Bensulfuron-methyl	Inhibition of acetolactate synthase (ALS) (acetohydroxy-acid synthase, AHAS), sulfonylureas	B, S	POE	UP
Chlorimuron+ metsulfuron	As above	G, B	POE	UP,
Ethoxysulfuron	As above	B, S	POE	UP
Metsulfuron	As above	G, B	POE	UP
Pyrazosulfuron-ethyl	As above	G,S,B	PRE, POE	ORS
Propanil	Inhibition of photosynthesis at photosystem II - amides	G,B,S	POE	ASM, DEL,HR, KTK, MP, ORS, PUN, TN, UP, UTR,
Paraquat	Photosystem-I- electron diversion - bipyridiliums	G,B, S	BSR	KER
Oxyfluorfen	Protoporphyrinogen oxidase (PPO) inhibitors, diphenylethers chlorophyll synthesis inhibitor)	B, G	PRE	HP, MP, RAJ, UTRL
Oxadiazon	Inhibition of protoporphyrinogen oxidase (PPO) - oxadiazoles	G, B	PRE	JRK, KTK, MS, ORS, RAJ,
Dinitramine	Microtubule assembly inhibition, dinitroaniline (cell division inhibitor)	G, B, S	PRE	KTK,
Pendimethalin	As above	G, CB	PRE, EPOE	All states in India
Dithiopyr	Microtubule assembly inhibition, pyridines (cell division inhibitor)	G, B	PRE	UTR,
Butachlor	Inhibition of cell division (inhibition of very long fatty acids), chloroacetamides	G, CB	PRE	All states in India
Pretilachlor+safener	As above	G, S, CB	PRE, POE	TN,UTR
Anilofos	Inhibition of cell division (inhibition of very long chain fatty acids), others	G, CB	PRE, EPOE	All states in India
Molinate	Inhibition of lipid synthesis, –not ACCase inhibition - thiocarbamates	G, B	PPI, POE	ORS
Thiobencarb	As above	G, CB	PRE	All states in India
2,4-D	Synthetic auxins (action like indole acetic acid), phenoxy-carboxylic acids	B, S	POE,	All states in India
Triclopyr	Synthetic auxins (action like indole acetic acid), pyridine carboxylic acids	B, S	POE	UP
Quinclorac	Synthetic auxins (action like indole acetic acid), quinoline carboxylic acids	G, CB	PRE, EPOE, POE	ORS, UTR,

[a]Herbicide classification by mode of action, 2003 Weed Science.org and Tomlin (1997); [b]G=grasses; CG=certain grasses, B=broadleaf weeds; CB=certain broadleaf weeds; S=sedges; CS=certain sedges.[c] BSR=Before seeding rice; PPI=Pre plant incorporation; PRE=pre-emergence (0–3 DAS); EPOE= early post-emergence (4–20 DAS);

POE=post-emergence (20 DAS and later). [d] ASM=Assam; BHR, Bihar; CSG=Chattisgarh; DEL=Delhi; HAR=Haryana; H.P.=Himachal Pradesh; JRK=Jarkhand; KER=Kerala; KTK, Karnataka; MP=Madhya Pradesh; ORS=Orissa; RAJ=Rajasthan; TN=Tamil Nadu; UP=Uttar Pradesh; UTR=Uttaranchal.,

Integrated weed management

A limited number of studies were conducted in India on IWM in DSR. The direct ad indirect weed management methods discussed so far has their own advantages and disadvantages. Any single method used in isolation can not provide effective and season long weed control due to various factors operating in the system. Hence, integration of several weed control methods has proven to be effective. An integrated weed management strategy involving summer plowing, thiobencarb, application and inter-crop cultivation is essential for effective weed control in direct-dry-seeded, flood prone, lowland rice, in order to ensure higher N-use efficiency and crop productivity.

Sharma *et al*. (2004) reported that pendimethalin+one hand weeding+cris-cross sowing and pendimethalin+one hand weeding gave similar yields as well as net returns and were significantly superior to two hand weedings. Higher seed rate (75 and 100 kg/ha) integrated with butachlor (Angiras and Sharma, 1998) or hand weeding (Moorthy, 1997) practices would provide season long weed control and higher yield of rainfed dry-seeded upland rice. Integration of Oxadiazon (Jena *et al*. 2002), butachlor (Singh and Singh, 2001), anilophos (Kolhe and Tripathy, 1998), thiobencarb (Kolhe and Tripathy, 1998) with hand weeding and butachlor with a twin wheel hoe (30 DAE) (Dutta and Gogoi, 1994) resulted in optimal weed control and grain yield.

Future research needs

As cultural practices and weed management methods change, the weed flora also changes (Rao *et al*. 2007). Weeds are dynamic and they change as the situation changes. It is our task to be able to predict these changes or at least be aware of them, to learn how to control the important weeds, and to prevent build-up of difficult-to-control species or to lessen their impact. Future research in India on weed management in DSR needs to answer the following questions enlisted under the following research priority areas

Need for more on-farm research

To answer relevant questions more on–farm research is needed in different agro-ecological regions of DSR growing states in India. They are: (a) What are the major problematic weeds in farmers' fields?, (b) What is the extent of on-farm yield losses caused by weeds? c) Are the available weed management technologies reaching the farmer?, (d) How effective are the weed management practices presently used by farmers in different agro-ecological zones?, (e) Are there any innovative weed control measures used by farmers that needs research attention?, (f) Why do farmers adopt the technologies that are used at present?, (g) What are the areas of research that farmers want in different agro-ecological regions?, (h) How are weed control decisions made by the farmers?, and (h)Can we improve the decision making process of the farmer for reducing the losses caused by weeds?

Breeding rice cultivars competitive against weeds

Certain questions need to be answered prior to initiation of breeding for weed competitiveness. They are: (a) Is the genetic variation in weed competitiveness among parents large enough for breeding?, (b) Are weed competitiveness and its related traits heritable?, (c) Can we combine high yield potential with strong weed competitiveness in DSR?, (d) What traits can be used as indicators of weed competitiveness among rice cultivars?, (e) What role does allelopathy play in competitiveness of rice cultivars?, (f) How could one differentiate competitive ability of rice against weeds and its allelopathy?

Evolving effective and economical integrated weed management technology for DSR
Which herbicides are suitable for weed control in DSR under a varying range of environmental conditions? What are the need based and location specific technological options (direct and indirect) for different agro-ecological regions to enable farmers to use them as components of IWM to effectively and economically manage weeds in DSR in India.

Understanding the ecology and physiology of major weeds
What are the survival mechanisms of the weeds in DSR?, What are the factors that are detrimental to the growth of predominant weeds?, Which stage of the life history of the weed is more easy to manage?, What factors are to be focused to shift the crop-weed balance in the favor of crop? Which measures are needed to prevent the spread and management of problematic weedy rice?

Monitoring the changes in weed community across time
What are the biological and ecological factors responsible to the long term persistence of species in weed communities, and for their temporal and spatial variation at the field level? , What factors influence the soil weed seed bank?, How does current weed management practices effect the weed community?, Is there any build up of difficult-to-control weed communities? Is there any herbicide resistance developed among weeds? Can we shift the weed community from "difficult-to-control weed community" to "easily manageable weed community"?

Herbicide tolerant transgenic/non-transgenic rice cultivars
Could herbicide tolerant rice cultivars be developed using biotechnological tools ? How could one avoid the possible gene flow? Can we prevent "super weeds" unintentionally emerging due to future scientific research activities?

Assessing long term effect of herbicide use on environment
How could we prevent any adverse effect that herbicides use may have on the soil, water and air of the DSR agro-ecosystem? What impact with the herbicides has on the flora and fauna in rice ecosystems and on human health?

Understanding the climate change impact on weed/DSR balance
How would climate change influence the growth and competitiveness of weeds and crop in DSR? What technological options are needed for managing weeds effectively and economically under changed climate scenario?

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USING THE WEED SEED WIZARD TO UNDERSTAND AND MANAGE THE WEED SEEDBANK

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Abstract: Weeds are an important factor in every agronomic system. The "Weed Seed Wizard" is a simulation model being developed to help co-ordinate management of weed seed within the seedbank. The model uses field management and weather records, and simulation of important aspects of seed biology, to track and predict the number, ages, soil depth, dormancy levels, viability and germination of seeds in the soil, in order to predict the amount of weeds appearing each year. Two versions of the model are being created: a relatively simple and user-friendly version for providing decision and management support via the web; and a 'research' version, incorporating more detailed representations of soil, seeds and biological processes, for integrating current understanding of the underlying seedbank dynamics and exploring these dynamics in more detail. In this paper, we explain the structure of the model and show how it can simulate the effect of decisions regarding choice of crop, sowing date, seeding rate, tillage, grazing management and herbicide application on weed germination, weed density, crop yield and the long-term sustainability of the system. We also show how the seed dormancy, germination requirements, competitiveness, and herbicide resistance of different weed species or populations can strongly affect the dynamics of the system, and thus the choice of an appropriate management strategy.

Key words: individual-based, seed ecology, simulation model, seedbank dynamics, weed control

Introduction

If you think weeds are a problem, think again! The real problem is the weed seedbank hidden beneath your feet! Weeds are the symptom. They compete with crops and pastures to reduce yield, contaminate grain and may be poison stock. But the seedbank is the disease. If you kill every last weed, your crop will look clean but the weeds will be back next year all the same. We need to take long-term strategies for managing the seedbank: starving it of new recruits; exposing seeds to their natural enemies; burying them where they will eventually rot and die; and exploiting their dormancy mechanisms to trigger germination in conditions where weeds can be easily killed or will have little effect. Thus we can slowly but surely strangle the seedbank to death.

To help move towards this long-term view of weed management, the "Weed Seed Wizard" project aims to construct computer simulations of weed seedbank dynamics for a variety of species. As a national Australian project, with participants in the states of Western Australia, South Australia, New South Wales and Queensland, it will target major in-crop annual weeds from each cropping region. For the southern states, target weeds include annual ryegrass (*Lolium rigidum* Gaud.), barley grass (*Hordeum leporinum* Link.), wild radish (*Raphanus raphanistrum* L.), wild oat (*Avena fatua* L.), brome grass (*Bromus* spp.) and silver grass (*Vulpia* spp.). Northern weeds include sweet summer grass (*Brachiaria eruciformis* Sm. Griseb), liverseed grass (*Urochloa panicoides* P. Beauv.), barnyard grass (*Echinchloa colona* L. Link), native jute (*Corchorus trilocularis* L.), bladder ketmia (*Hibiscus trionum* L.), fleabane (*Conyza* spp.) and sowthistle (*Sonchus oleraceus* L.). Different weed species persist in the soil (or seedbank) for different periods. Some species have little or no dormancy and germinate with the opening rains, completely depleting the seed bank. Other species with dormancy require specific environmental (light, moisture, temperature, accumulated degree days, etc) cues for dormancy to be broken and germination to commence. These species can persist for several years. The Wizard will be based on the vast collection of existing documented knowledge about each species, and will then be validated against further data collected from a number of trials in each of the participating states.

As seedbank dynamics and weed seed persistence are complicated and often hard to predict, one objective is to construct a practical decision-aid tool, incorporating current scientific knowledge, that can help farmers and consultants manage weed populations in real agricultural contexts. Eventually this tool will be self-calibrating (have the capacity to adjust its parameters in response to ongoing observation records to better predict weed populations in a particular field). Another objective is to build a modelling framework that can be used for more theoretical investigation of the complex mix of factors affecting the weed seedbank, and thus contribute to our understanding of seedbank dynamics. As part of the Sustainable Cropping Systems program within the CRC for Australian Weed Management, the "Weed Seed Wizard" project will contribute to developing crop and pasture systems for sustained productivity and profitability through these objectives.

The Model

The Weed Seed Wizard will take into account conservation tillage systems (including No-Till and Zero-Till) combined with strategic use of a range of other techniques, such as soil inversion, autumn tickle, crop competition, selective and nonselective herbicides, crop topping, swathing, seed catching, and burning or grazing for stubble management. It will also incorporate recent advances in understanding of the factors that affect germination and death of weed seeds, in order to predict the amount of weeds appearing each year. We envisage that the Wizard will operate as an adjunct to field record-keeping software, using farmer records concerning field management decisions, the site, and other observations. Such records might include crop sown, sowing date, seeding rate, tillage and grazing management, herbicide application, crop yield, weed density, rainfall etc. Eventually the Wizard might be part of a farm management system that integrates farmer record keeping with simulations of biological and physical processes including weed populations, soil water and nutrition, insect pests and diseases.

A prototype of "The Wizard" has been designed and implemented in the Java programming language. The model includes representations of the soil, the daily weather, the plant population, and the individual seeds within the soil. It simulates the moisture and temperature within the soil; the dormancy, after-ripening and germination of the seeds; the effects of competition between different plant species on seed set; and the effects of management actions on plants and seeds. A graphical user interface (GUI) has been designed for the tool. The GUI includes windows where the user can adjust the initial conditions of the simulated field and adjust model parameters such as the period of simulation. It also includes scenario windows where the user can add, delete or edit scheduled management actions and view the simulated output regarding the states of the soil seedbank, plant populations, crop yield etc. These can be used to compare a number of different management scenarios. Standard data formats have been designed for required data on herbicides, plant species, types of cultivation and management scheduling and routines have been implemented to read this data into the model.

Model Structure and Capabilities

The "Weed Seed Wizard" is being built in an 'object-oriented' web-friendly programming language (Java) that enables different objects within the natural system to be described clearly as separate 'objects' or modules within the program. This facilitates adaptation of the model for different purposes and users. The primary objects in the model (Figure 1) are an *input* *interface* allowing the user to configure the model, set up the initial state of the system and schedule management events; an *event queue* including all events that affect the system; *data lists* including all required model parameters; a representation of the changing *state* of the system; and an *output interface* for presenting the output of the model.

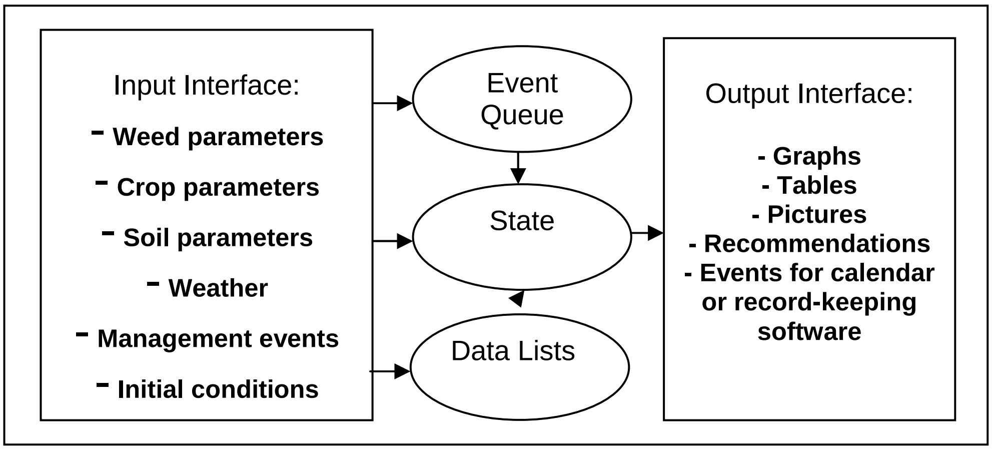

Figure 1. The overall modular structure of the Weed Seed Wizard

Input interface

The input interface consists of a main GUI containing drop-down menus and a number of tabbed panes or windows (Figure 2), which in turn contain buttons, editable text fields, sliders and drop-down lists. One of these tabbed panes gives a short introductory tutorial for the model. Others allow the user to configure the model, set up the initial state of the system and specify what output is produced by the model. Another kind of tabbed pane, called scenario panes, allows the user to set up a schedule of past and planed management events. The example scenario pane showing in Figure 2 is the default list of management events. Note the list of scheduled management events appearing in the middle of the pane and the list of predicted annual crop losses due to weeds displayed towards the bottom of the pane.

In configuring the model, the user will have options such as changing the time step used by the model, or deciding whether future weather is simulated using historical averages or replicated from actual historical. (The historical weather data can be downloaded from weather databases or entered directly from the user's records). The user will also be able to decide whether to simply use existing data files specifying model parameters regarding weed and crop species, herbicides, soil types and cultivation events, or to modify these data files to match their own conditions and observations. Setting up the initial state of the system will involve making estimates of the number of weed seeds of different species in the seedbank at some initial time. The estimation of current seedbank numbers can be made more accurate by simulating several historical years using recorded weather data and management schedules.

In Figure 2, note that two scenario panes are available in addition to the default scenario. The user can actually set up any number of scenario panes, and use them to compare the predicted effectiveness of alternative management schedules. Management schedules can be set up from scratch or existing schedules can be duplicated and modified. Events can be added individually or as compound events, such as all events over a year for a wheat season or all events over three years for a wheat-wheat-lupin rotation. Users can copy, delete, and edit existing events in schedule lists by clicking on them. Management schedules can be saved and later reloaded, and interesting sets of scenarios can be saved and reloaded as a group. It is envisaged that future versions of the Wizard will allow users to download a record of historical and planned management actions from the field record-keeping software they may already be using, and be able to suggest 'optimal' strategies for controlling weeds for a given initial state of the weed seedbank.

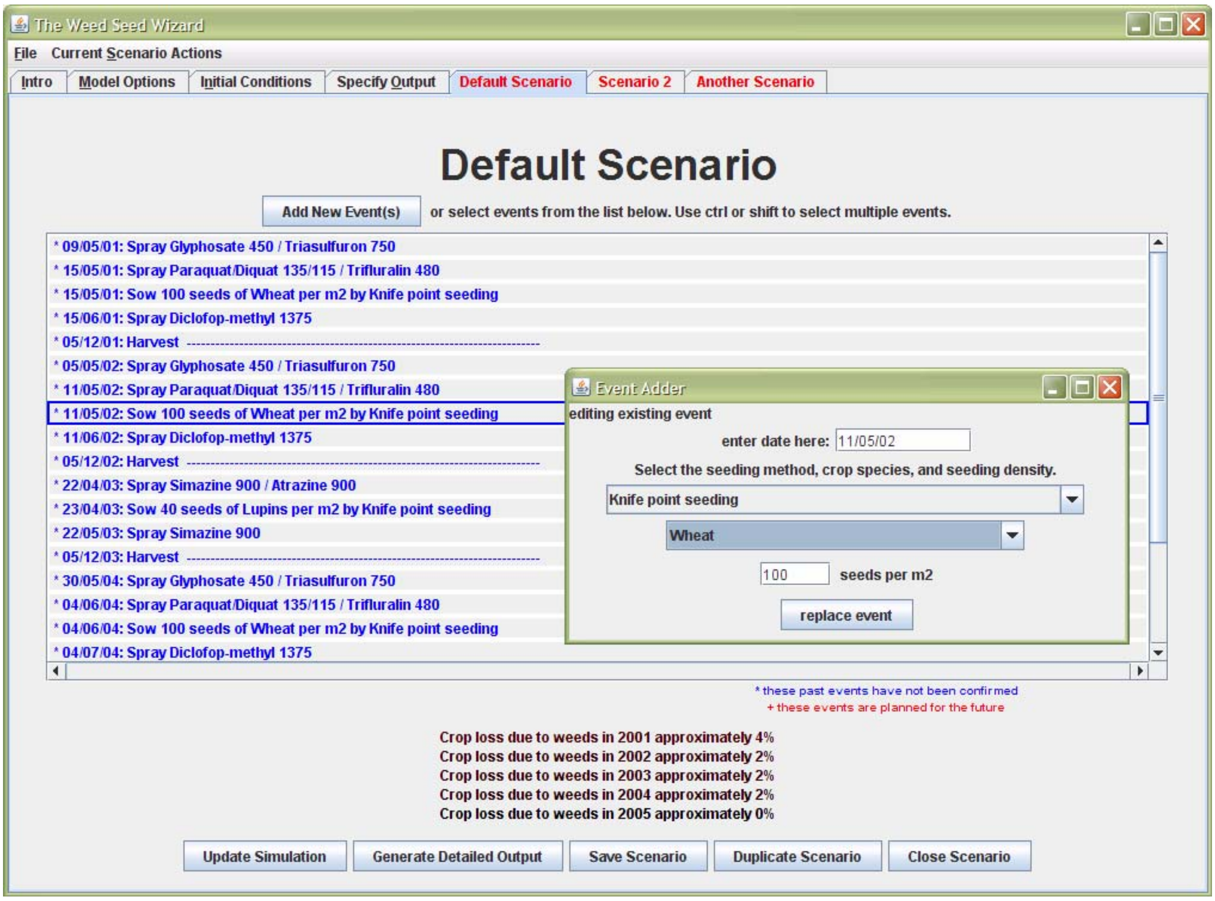

Figure 2. The main Weed Seed GUI containing drop-down menus and a number of tabbed panes or windows (background) and a smaller pop-up window for editing events (foreground).

Event queue

The simulation runs by using an event queue. This is a list of events ordered according to their scheduled date and time. This structure allows new events to be easily inserted into the queue as required, by the user or automatically by the program itself, in response to the results of previous events and the evolving state of the system. The queue includes 'occasional events' entered by the user, such as herbicide application, cultivation etc; simulated occasional events, such as rainfall events, plant germination and death, and seed rain; as well as regular state updates and record keeping.

Data lists

The simulation depends on a large number of parameters. These parameter values are read from the standardised *xml files* by *parsers* in the initiation phase of running the program to create equivalent data list structures that can be used by the program. These include a herbicide list, a plant species list, a cultivation and seeding options list, and a soil layer list. There is also a default list of management events and a list of compound events. The user can easily modify characteristics of an item in a data list, or add new items (such as new weed species, or new herbicide options). The standardised structure of the xml files facilitates communication with other software and databases. The parameter values in these files are being drawn from literature, expert opinion, laboratory experiments and ongoing field trials across the country.

State

The state object is the heart of the Wizard (Figure 3). Once the initial state, the event queue and the data lists have been set up (by the user or with the provided defaults), the events in the event queue are applied to the state one after the other using the parameters specified in the data lists to simulate the dynamics of the system. The state object includes sub-objects representing the *environment* (conditions such as rainfall, potential evaporation and temperature), the *soil* (consisting of a number of *layers*), the *seedbank* (consisting of individual *seed cohorts*), the *plant population* (consisting of individual *plant cohorts*), and a *record* of important aspects of the evolving state of the system. Each object within the system 'knows' certain data (a seed cohort knows its species, age, dormancy, germination status, moisture content, accumulated degree days, and position within the soil) and communicates at least some of that data with other objects (seed germination depends on temperature and moisture level in the relevant soil layer).

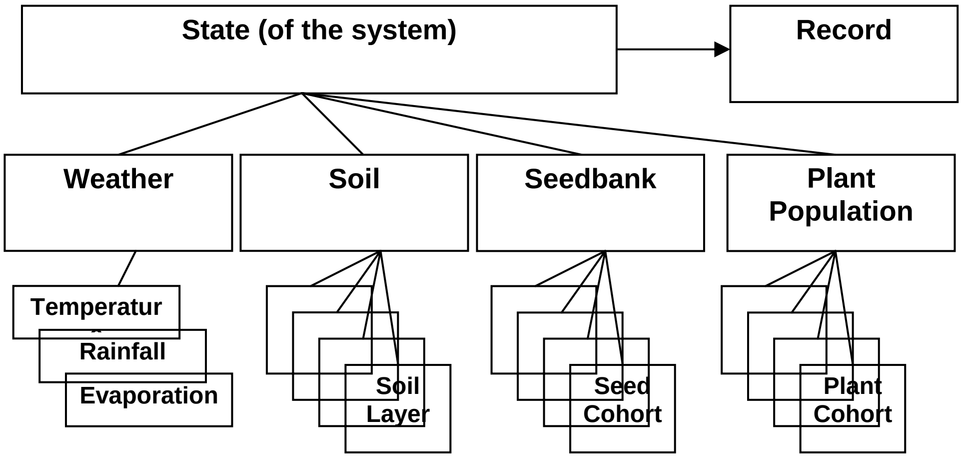

Figure 3. The Weed Seed Wizard's modular representation of the dynamic state of the agronomic system. The record is used to produce the output for the user.

Output interface

The basic output given by the model for a given scenario is the list of predicted crop losses shown in Figure 2. The user also has the option of producing a series of graphs displaying the information stored in the record, including the numbers of seeds in different layers in the seedbank and weed populations over time. Another option is to output results in a detailed tabular form, specifying weed numbers before and after management events. In future, we will add the capacity to give pictures indicating the predicted weed densities, or a list of recommended actions for the user.

Results and Discussion

The current version of the Wizard is able to produce reasonable simulations of the population dynamics of ryegrass and wild radish for a range of standard integrated management strategies for these species. For example, it can give reasonable predictions of the effects of standard knockdown, pre-emergent and post-emergent herbicide applications, 'double-knock' herbicide strategies, variable sowing densities, seed-catching and strategic mouldboard (inversion) and 'tickle' ploughing events (see Figure 4 for an example).

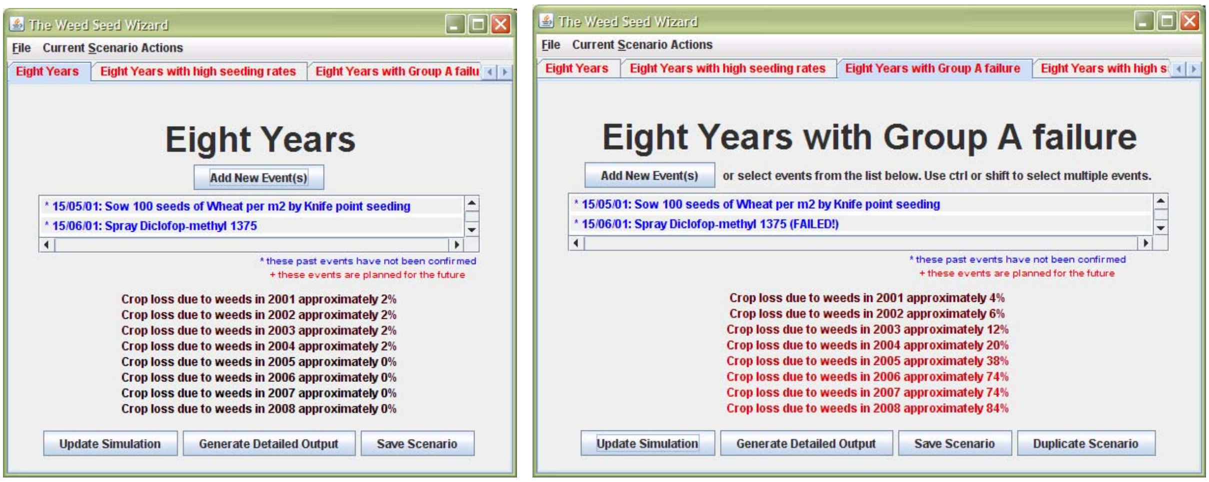

Figure 4. Comparing two similar scenarios running over eight years. The only difference between the two scenarios is that in the scenario on the right, it is assumed that the weed has evolved resistance to the selective in-crop herbicide and so the kill rate of this herbicide against the weed is reduced from 90% to 20%. Note the difference in the predicted crop losses.

The Weed Seed Wizard will integrate current knowledge of weed biology and the effects of management techniques to simulate and predict annual weed populations. It will allow us to explore how the dormancy, germination requirements, competitiveness, and herbicide resistance of different weed species or populations can affect the dynamics of the system, and thus the choice of an appropriate management strategy. Through incorporating the effects of herbicides and a range of other strategic options, the Wizard will help us design a truly integrated weed management system. By giving deeper insight into how different factors interact to determine weed seedbank levels and helping farmers manage weeds within their fields, the Wizard will contribute to building the sustainability of Australian crop and pasture systems. Because of its modular structure and dependence on standardised data files, the model can be easily adapted to cropping systems and weed species throughout the world.

Acknowledgements

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EFFECT OF SEEDLING EMERGENCE TIME ON THE PERFORMANCE OF MAYWEED (*Anthemis cotula* L.): AN ALIEN INVASIVE SPECIES IN KASHMIR HIMALAYA

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Abstract: Biological invasions, caused by non-native invasive species are a major factor contributing to ecosystem perturbations and hence are being actively pursued worldwide. Mayweed (*Anthemis cotula* L.), a native of southern Europe-West Siberia is an aggressive invasive species in Kashmir Himalaya, India. Among the myriad of attributes, seedling emergence time is critical to its successful colonization of habitats with varying levels of soil disturbance. Field studies revealed that the species recruits individuals over an extended period of time from September to May. This recruitment period is interspersed by harsh snowy winter (December-February) and hence the established plants of the species are constituted of pre-winter and post-winter populations, with former contributing to the fitness component of its life history and latter to the survival component. Except for number of achenes per capitulum and achene weight, all other investigated parameters such as stem height, number of primary branches per plant and number of capitula per plant were significantly higher in individuals belonging to pre-winter population in than the individuals constituting the post-winter plants. Fecundity of pre-winter individuals was further enhanced if decapitated by a specific herbivore that results in over-compensatory growth. Thus, seedling emergence time is of pivotal importance to the establishment, colonization and spread of this obnoxious invasive weed in the Kashmir Himalaya.

Key words: *A. cotula*, emergence time, invasiveness

Introduction

Biological invasion by alien species are among the most important driving forces of evolution on our human-dominated planet and distinctive features of this evolution now include homogenization of biota, proliferation of opportunistic species and decline of biodiversity (Eppstein and Molofsky, 2007). The growing awareness of the biological, economic and social impacts of invasive plant species has propelled the current staggering interest in identification of life-history and ecological traits that contribute to invasiveness of some species in their non-native range. This interest is further fuelled by the central importance of such studies in providing a scientific basis for the development of strategies aimed at prevention and management of invasive species (Richardson and Pyšek, 2006).

While species invasiveness has been associated with a variety of individual traits such as high seed production, dispersal, vegetative reproduction, broad tolerance or phenotypic plasticity (Baker, 1965; Sutherland, 2004; Pyšek and Richardson, 2007), yet seedling emergence time, known to significantly influence survival, growth, competitive ability and ultimately fitness of established adult plants (Geber and Griffen, 2003), has been rarely considered as a key trait contributing to invasiveness of the species with protracted seedling recruitment pattern. The present study thus, examined the influence of emergence date of *Anthemis cotula* seedlings on the vegetative, reproductive and fitness attributes of adult plants, and the relationship between phenotypic variation in germination time to environmental factors, and invasiveness of the species in the Kashmir Himalaya.

Materials and Methods

Study species

Mayweed (*Anthemis cotula* L.), also known as Stinking Chamomile, is an annual bushy, branched ill-scented herb of the Asteraceae. It is native to southern Europe-west Siberia but at present has cosmopolitan distribution (Erneberg, 1999). This invasive species can follow a winter or summer life cycle resulting in the formation of two distinct (pre and post-winter) populations. After emergence in the fall, *A. cotula* forms a basal rosette for winter survival. In a winter annual life cycle, the rosette bolts in the spring, growing to a height of about 25-125 cm. Capitula are 15-30 mm in diameter and contain white neuter ray florets and yellow hermaphroditic disc florets. The plant is strongly self- incompatible and mostly pollinated by insects.

Study area

The study was carried out in the Mirza Bagh campus of the University of Kashmir, Srinagar, J&K, India, lying within the geographical co-ordinates of 34° 5' to 34° 6' N latitude and 74° 8' to 74° 9' E longitude at an altitude of 1,586 meters above mean sea level. This three hectare area is an abandoned wasteland previously occupied by grassland that now supports vegetation dominated by *A. cotula*. The temperature at the study site ranges from an average daily maximum of 22°C and minimum of 3°C. It receives annual precipitation of about 70 cm, mostly in the form of snow during winter months.

Sampling

In June 2003, three study sites within the Mirza Bagh campus designated as S1, S2 and S3 were selected for studies of seedling establishment, survival and reproductive differences among the individuals of pre-winter and post-winter populations. Average distance between the three study sites ranged from 75 to 100 m. Seedlings emergence pattern in *A. cotula* was monitored in three randomly laid permanent quadrats ($1m^{-2}$) at each study site, which were surveyed after an interval of 15 days during the study period from the stage of seedling recruitment to maturation *i.e.* from September 2003 to July 2004, except during the period when the entire study area was covered with snow. The seedlings, after attaining distinctive appearance, were marked using color paint following Khushwaha *et al*. (1981). The marked seedlings were again counted on the subsequent census to record mortality and survival of the individuals. The seedlings without color marking on each sampling date constituted the new recruits which after counting were marked with the color paint in the permanent quadrats to distinguish between previous and new recruits on successive sampling dates. Mortality data of pre-winter and post winter individuals was used to construct population survivorship curves. At maturity, just prior to dispersal of achenes, data on attributes, namely stem height, number of primary branches per plant, number of capitula per plant, number of achenes per capitulum and average weight of 1000 achenes per plant were recorded. Data were statistically analyzed using SPSS version 10.0.

Results

Seedling recruitment pattern

Seedling emergence in *A. cotula,* a winter annual, starts in early autumn (September) and lasts till spring (May). During this entire period, recruitment occurs in two main pulses. The recruitment pattern shows a relationship with temperature and precipitation (Figure 1). A major pulse of recruitment occurs in autumn prior to onset of winter (December-February) and a minor pulse occurs during spring (April).

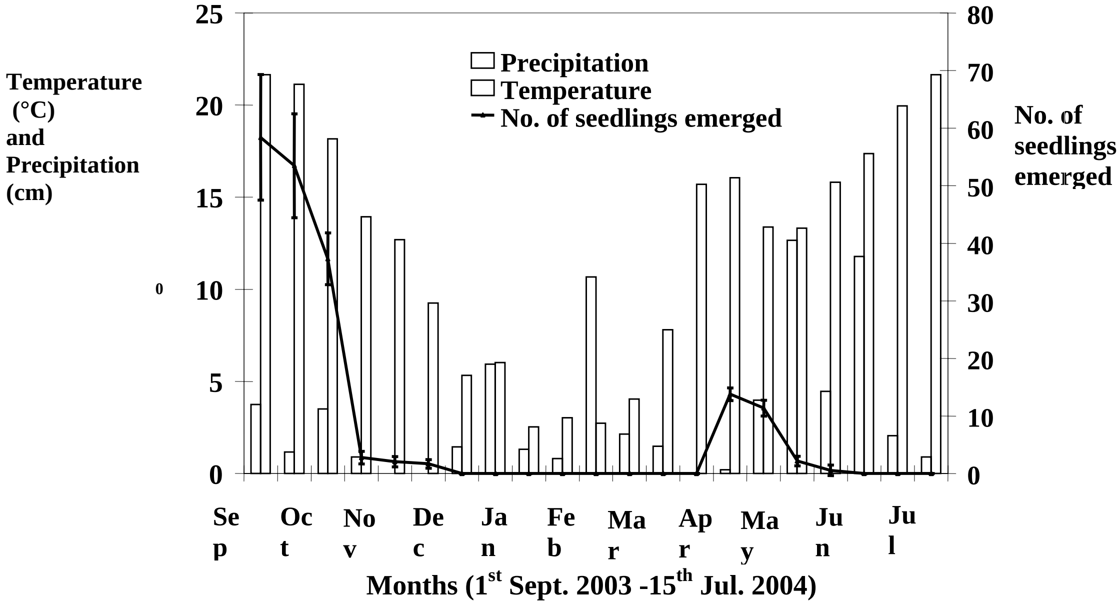

Figure 1. Temporal Variation in seedling emergence (Mean ± SD) of *A. cotula* (continuous line) in relation to temperature and precipitation (bars).

There was an irregular mortality pattern in the populations investigated. The population size was higher during autumn and thereafter declined considerably. Since the established populations of *A. cotula* in the Kashmir Himalaya consist of individuals of both the pre- as well as the post-winter seedlings, the numbers surviving during the study period at each site are presented as survivorship curves (Figure 2). Pre-winter population exhibits a Type II curve while as post-winter population showed Type I survivorship curve.

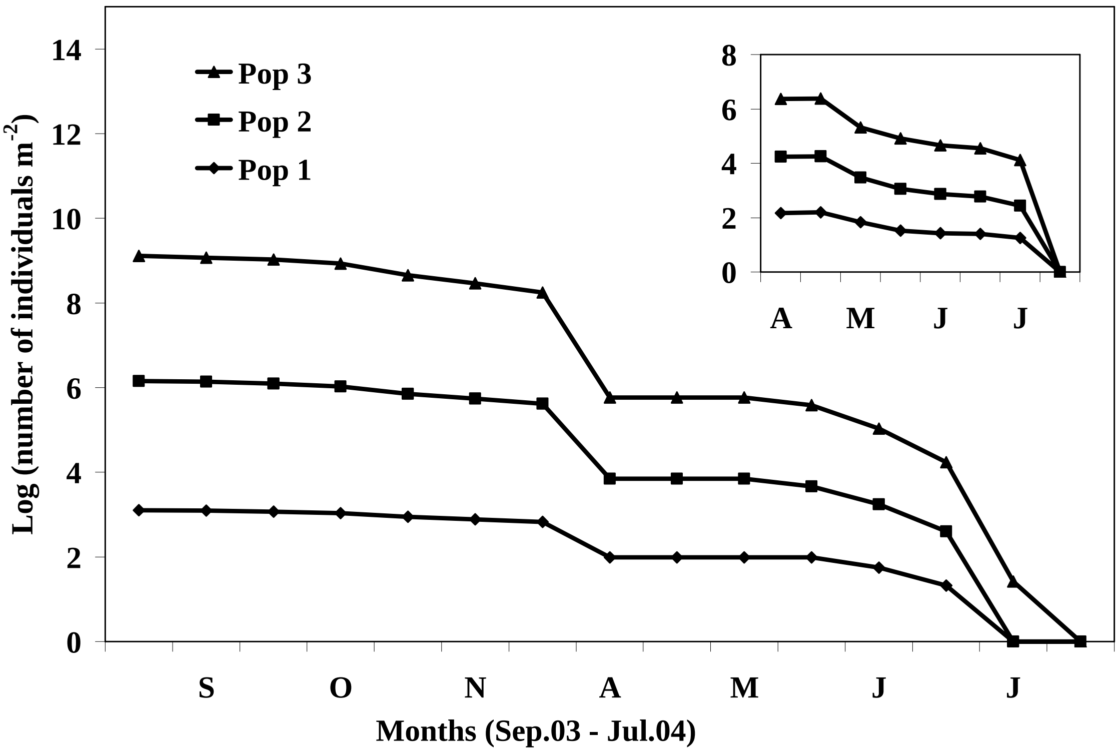

Figure 2. Survivorship curves for pre (main) and post-winter (inset) populations in *A. cotula* in Kashmir Himalaya.

Emergence time and fitness attributes

The individuals of the two populations differed significantly in respect of vegetative parameters, such as stem height ($p<0.0001$), number of primary branches per plant ($p<0.0001$), total number of branches per plant ($p<0.0001$) and reproductive attributes such as number of capitula per plant ($p<0.0001$) and number of achenes per plant ($p<0.0001$). However, statistically insignificant differences between the two populations were recorded in the number of secondary branches per primary branch ($p<0.047$) and average achene weight ($p<0.033$). Particularly important was the significant increase in the numbers of primary branches per plant in individuals of pre-winter population because it influenced the number of capitula and hence the number of reproductive propagules and subsequent invasiveness of the species in the Kashmir Himalaya (Table 1).

Table 1. Comparison of vegetative and reproductive attributes of pre- and post-winter populations of *A. cotula* across different study sites. (Mean ± SD based on 3 replicates).

Attribute	Population	Pop 1	Pop 2	Pop 3
Stem height (cm)	Pre-winter	91.80 ± 6.92	87.87 ± 5.94	87.47 ± 5.07
	Post-winter	42.00 ± 5.06	36.67 ± 5.62	37.56 ± 2.70
No. of primary branches/ plant	Pre-winter	14.13 ± 1.86	14.67 ± 1.74	13.87 ± 1.32
	Post-winter	1.89 ± 0.72	2.22 ± 0.89	1.89 ± 0.72
No. of secondary branches/primary branch	Pre-winter	9.87 ± 0.58	9.87 ± 0.75	9.67 ± 0.66
	Post-winter	8.67 ± 0.72	9.44 ± 0.58	9.33 ± 0.58
Total no. of branches/ plant	Pre-winter	132.47 ± 18.35	135.27 ± 19.39	122.33 ± 14.82
	Post-winter	14.00 ± 3.61	17.44 ± 8.46	16.00 ± 6.35
No. of capitula/plant	Pre-winter	1095.20 ± 137.23	1146.27 ± 253.61	1034.67 ± 213.37
	Post-winter	99.11 ± 27.28	152.44 ± 76.38	120.56 ± 58.53
No. of achenes/capitulum	Pre-winter	156.07 ± 2.77	154.27 ± 3.20	157.73 ±4.16
	Post-winter	125.56 ± 3.31	131.33 ± 2.91	125.44 ± 6.66
No. of achenes/plant	Pre-winter	173432.60 ± 22659.44	180632.80 ± 38974.10	166912.27 ± 31910.43
	Post-winter	13005.00 ± 3428.71	20509.44 ± 10515.13	15590.33 ± 7551.83
Wt. of 1000 achenes (g)	Pre-winter	0.489 ± 0.013	0.496 ± 0.030	0.510 ± 0.028
	Post-winter	0.448 ± 0.004	0.473 ± 0.037	0.477 ± 0.034

Discussion

The present study clearly reveals temporal variability in germination time and seedling recruitment of *A. cotula* in the Kashmir Himalaya. Time of propagule germination, is determined by the interplay between abiotic and biotic factors and the mode of natural selection on germination time Venable (1984). Such biotic and abiotic factors seem to be operative in *A. cotula* also since initiation of germination in autumn coincides with the favorable temperature and moisture conditions. Experimental studies on achene germination (Rashid, *et al.* 2007) also agree with such conclusions. Besides, senescence of the existing vegetation during autumn not only creates favorable 'regeneration niche' in terms of germination promoting light environments but also allows the seedlings to avoid inter-specific competition by niche pre-emption (Allaie, *et al*. 2005). Notwithstanding the benefits that accrue to the species upon seedling recruitment in autumn, considerable seedling mortality occurs mainly due to intra-specific competition and frost-heaving of soil. However, the surviving seedlings over-winter as vegetative rosettes which according to Regehr and Bazzaz (1979) are capable of substantial photosynthesis and energy storage during winter. These rosettes grow rapidly with the onset of favorable environmental conditions during spring and produce a large number of branches which subsequently bear a large number of flower heads.

In contrast to pre-winter population, the individuals of post-winter seedling population experience less mortality but the adult plants are smaller in size and produce a limited number of primary lateral branches. These attributes significantly constrain the production of propagules necessary for occupation of virgin areas by this species. Thus, the existence of an ecological trade-off in the life history of A. *cotula* is easily discernible with individuals of pre-winter population contributing to fecundity and those of post-winter population ensuring survival and continuance of the species in the invaded habitats. Importance of the timing of emergence in determining fitness of individuals has also been reported by Teasdale *et al.* (2004). The present study, therefore, conclusively demonstrates the importance of early seedling emergence in the successful colonization and spread of this invasive species in the Kashmir Himalaya.

Acknowledgements

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EFFECTS OF WOOD VINEGAR AND SULFONYLUREA-BASED HERBICIDES ON THE GROWTH AND PROTEIN FORMATION OF BARNYARDGRASS (*Echinochloa crus-galli* var. *crus-galli*)

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Abstract: The effects of wood vinegar and reduced dosages of four sulfonylurea-based herbicides on the growth and nitrogen content of barnyardgrass (*Echinochloa crus-galli* var. *crus-galli*) at 1- and 2-leaf stages were investigated. The herbicides used were pyrazosulfuron-ethyl+esprocarb (PSE), pyrazosulfuron-ethyl+molinate (PSM), pyrazosulfuron-ethyl+mefenacet (PSMF), and halosulfuron-methyl+pyriminobac-methyl+pretilachlor (HMPMP). Two wood vinegar concentrations were tested: 500 WV (1 ml/500 ml water) and 1000 WV (1 ml/1000 ml water). For each of these dilutions, 50% and 30% of the recommended application rate was applied and pots for 100%, 50% and 30% alone and no herbicide (0%) treatments were used as control. Results revealed that 100% and 50 + 1000 WV treatments of HMPMP showed similar effects on the growth of barnyard grass at both leaf stages. The 50 + 1000 WV treatments of the PSE and PSMF obtained higher efficacy than 50% but lower than 100% at 1-leaf stage. The 50 + 1000 WV HMPMP and PSMF showed a higher efficacy in controlling growth of barnyard grass than PSE and PSM. The decrease in dry weight of 2-leaf stage barnyard grass treated with 50% HMPMP plus 1000 WV was accompanied by a severe decrease in nitrogen and branched-chain amino acid contents, and high inhibition of AHAS activity. Results also showed that the efficacies of the herbicides in 500 WV are lower than those in 1000 WV treatments.

Key words: Wood vinegar, sulfonylurea-based herbicides, barnyardgrass

Introduction

Wood vinegar is gaining popularity for use in agricultural crop productivity (Yoshimoto, 1994) and probably in herbicidal applications (Kim *et al.* 2000; Mu *et al.* 2003). It has a superb fertilizing effect that may enhance herbicide absorption and translocation in the plant system of young seedlings that can increase herbicidal efficacy (Gigliotti *et al.* 2005). It contains more than 12 kinds of organic acids, 13 kinds of phenol, 19 kinds of carbonyl, and 9 kinds of alcohol (Kim *et al.* 2001) which can all act as growth regulating herbicides on the germinating seeds. Studies have shown that use of material rich in dissolved organic carbon can affect herbicidal degradation, sorption, mobility and efficacy (Gigliotti *et al.* 2005; Cox *et al.* 2006). Dissolved organic matter (DOM) may reduce herbicide sorption through stable DOM-herbicide interactions (Businelli, 1997) or by competing with herbicide molecules for sorption sites on the soil surface (Lee *et al.* 1990). It also enhances herbicide sorption by adsorbing on the soil particles and providing additional sites for pesticide sorption (Gigliotti *et al.* 2005). Wood vinegar may possibly increase the absorption and translocation of sulfonylurea-based herbicides in the plants. Thus, the objective of this study was to investigate the effects of wood vinegar and reduced dosages of sulfonylurea-based herbicides on barnyard grass (*Echinochloa crus-galli* var. *crus-galli*).

Materials and Methods

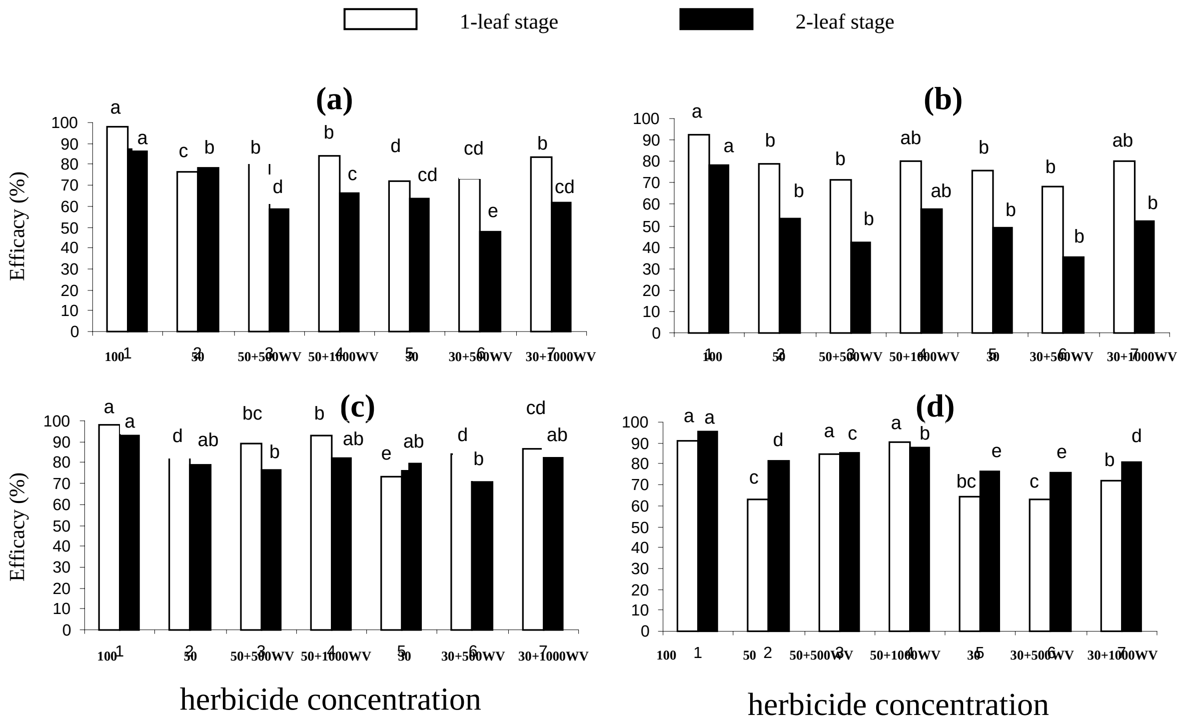
Four granular sulfonylurea-based herbicides were tested namely: pyrazosulfuron-ethyl+esprocarb, pyrazosulfuron-ethyl+molinate, pyrazosulfuron-ethyl+mefenacet and halosulfuron-methyl+pyriminobac-methyl+pretilachlor. Two wood vinegar concentrations were tested: 500 (1 mLl/500 ml water) and 1000 (1 ml/1000 ml water) where 50% and 30% of the recommended application rate was applied. Pots for 100%, 50%, 30% and 0% of the recommended herbicide application rate were used as control. To prepare the plant materials, barnyard grass (2 g, around 80 seeds) was placed in a Petri dish with 15 ml water and incubated in growth chamber (30°C, 24 hrs white light) for 36 hours. The seeds were then sowed in a 0.01m^3 (0.41 m x 0.24 m x 0.11 m) pot containing complete fertilizer (3 g) and 1:1 ratio of clay:sand mixture filling the pot to the half. Water level was kept at 3.0~3.5-cm depth during the whole duration of the experiment. The plants were treated at 1- and 2-leaf growth stages using a total volume of 3 L wood vinegar solution. The harvest was collected 10 and 12 days after treatment (DAT) for 1- and 2-leaf stage, respectively. The samples were oven-dried and dry weights were recorded. For the total nitrogen content analysis, 1 g of dried ground tissue was digested in 12 ml concentrated sulfuric acid for 2 hours at 470°C. Total nitrogen content was measured by the conventional Kjeldahl method (Kjeltec Auto1035/38 Sampler System, Tecator AB, Sweden). For the amino acid analysis, extraction was done by digesting dried samples (0.1 g) in 4 ml of 6N HCl at 110°C for 24 hrs. The mixture was filtered using Whatman filter paper no. 1, added with 5 ml of 6N HCl by washing the filter paper, and dried completely using a rotatory evaporator. After drying, the sample was added with 3 mL 0.2N citric acid and filtered with 0.2 μm membrane filter paper. Analysis was done using amino acid analyzer (Pharmacia Biotech Co., Model Biochrom 20, Swiss). Acetohydroxy acid synthase (AHAS; EC 4.1.3.18) activity was assayed at 3, 6, 9 and 12 DAT according to the procedure of Singh *et al.* (1988).

Results and Discussion

Study (Kim *et al.* 2000) showed that foliar application of wood vinegar exhibited herbicidal activity against *Echinochloa crus-galli* at different growth stages. However, no inhibitory effect was observed when wood vinegar was soil-applied. Herbicidal activity of acetic acid has also been tested and has been shown to have broad-spectrum activity (Chinery, 2001).

Effect of wood vinegar concentration on the efficacy of the herbicides
The trend on the efficacy of the herbicides (Figure 1) shows that the 500 WV treatments generally have lower efficacy as compared to 1000 WV treatments. The exact behavior of the herbicides when mixed with WV is not yet known. However, the observed trend could possibly be due to the lower pH (~3.94) in 500 WV medium since the herbicides undergo degradation in lower pH. Sulfonylurea (SU) herbicides undergo faster degradation by chemical hydrolysis at lower pH (Sarmah and Sabadie, 2002). There is also an increased microbial activity in soil treated with higher wood vinegar concentration (Zhu, 2004) that possibly contributed to the microbial degradation of both SU and the mixed herbicides.

Effect of wood vinegar and herbicides on the growth of barnyard grass
Table 2 shows that in 1-leaf stage, all treatments significantly reduced the dry weights compared to the control. Figure 1 shows that 50% plus WV treatments of HMPMP obtained similar efficacy with that of 100% while 50 + 1000 WV treatments of PSE and PSMF obtained significantly higher efficacy than 50% alone. It also shows that 50% plus WV treatments of PSM obtained higher but not significantly different efficacy than 50% alone. For the 2-leaf stage (Figure 1), only 50% plus WV treatments of HMPMP obtained a significantly higher efficacy compared to 50% alone which is also similar with that of 100% showing the possibility of increasing its efficacy from 50% alone to 50% plus wood vinegar treatments. For the 30% treatments, all herbicides showed increasing efficacies from 30% alone to 30 + 1000 WV treatments in 1-leaf stage and PSMF of 2-leaf stage. Results showed that WV can increase the efficacies of 50% PSE, PSMF and HMPMP in controlling growth of 1-leaf stage and only 50% HMPMP for the 2-leaf stage.



Figrue 1. Effect of wood vinegar and sulfonylurea herbicides on the growth of 1- and 2-leaf stage barnyard grass (*Echinochloa crus-galli var. crus-galli*): (a) pyrazosulfuron-ethyl+ esprocarb; (b) pyrazosulfuron-ethyl+molinate; (c) pyrazosulfuron-ethyl+ mefenacet; (d) halosulfuron-methyl+pyriminobac-methyl+pretilachlor.

Table 1. Effect of wood vinegar on the efficacy of sulfunylurea herbicides on the growth of barnyard grass (*Echinochloa crus-galli var. crus-galli*).

Herbicide application rate (%)	Wood vinegar (volume)	Pyrazosulfuron-ethyl+esprocarb		Pyrazosulfuron-ethyl+molinate		Pyrazosulfuron-ethyl+mefenacet		Halosulfuron-methyl+ pyriminobac-methyl+ pretilachlor	
		dry weight (g)							
		1 LS	2 LS	1 LS	2 LS	1 LS	2 LS	1 LS	2 LS
100	-	0.07d	0.74d	0.24c	1.25c	0.07d	0.42c	0.29c	0.27d
	-	0.78bc	1.24d	0.70bc	2.65b	0.52bc	1.19bc	1.22b	1.06bc
50	500	0.58bc	2.35c	0.95b	3.29b	0.35cd	1.33bc	0.50c	0.85bcd
	1000	0.53c	1.94c	0.66bc	2.39bc	0.24cd	1.01bc	0.32c	0.70cd
	-	0.92b	2.06c	0.79b	2.91b	0.87b	1.15bc	1.18b	1.35b
30	500	0.87bc	2.97b	1.04b	3.66b	0.53bc	1.68b	1.22b	1.36b
	1000	0.55c	2.18c	0.65bc	2.72b	0.44cd	0.92bc	0.92b	1.08bc
control	-	3.28a	5.68a	3.28a	5.68a	3.28a	5.68a	3.28a	5.68a

100 = recommended herbicide application rate, 50 = 50% of the recommended application rate, 30 = 30% of the recommended application rate, 500 = 1 ml wood vinegar/500 ml water, 1000 = 1 ml wood vinegar/1000 ml water, DW = dry weight, LS = leaf stage.

Data showed a more evident increase in efficacy from 50% alone to 50% plus WV treatments at 1-leaf stage than at 2-leaf stage. This could be due to the growth stage of the plants since the younger the plants are, the more susceptible they are to the attack of herbicides. Also, wood vinegar contains organic acids that can help bypass the natural barrier and defense mechanisms of the plants that would facilitate co-migration of the herbicide. The vigorous growth of plants promoted by wood vinegar can also be a major contributing factor for high efficacy even at lower herbicide dosage because of increased uptake and translocation of herbicides.

Effect of wood vinegar and herbicides on the nitrogen content

Results (Figure 2) revealed that WV treatments of all herbicides obtained a decreasing trend in the nitrogen content compared to the reduced herbicide application rate alone. Only HMPMP gave a clear significant difference in nitrogen content between 50 + 1000 WV and 50 and 100% while the rest of the herbicides obtained similar nitrogen contents in these treatments. The effect of 1000 WV plus reduced application rate of HMPMP was even more evident at the 2-leaf stage. Only WV treatments of 30% PSMF and HMPMP obtained significantly lower values compared with 30% alone. Data also showed that though there were no similar values in the 50 + 1000 WV and 100% HMPMP in controlling growth of 2-leaf stage barnyard grass, there was a severe decrease and nitrogen contents in 1000 WV treatments of both 30 and 50% HMPMP that obtained significantly similar values with that of 100%. The results showed that 50% herbicides applied with WV can reduce nitrogen contents in plants treated with more than 50% alone.

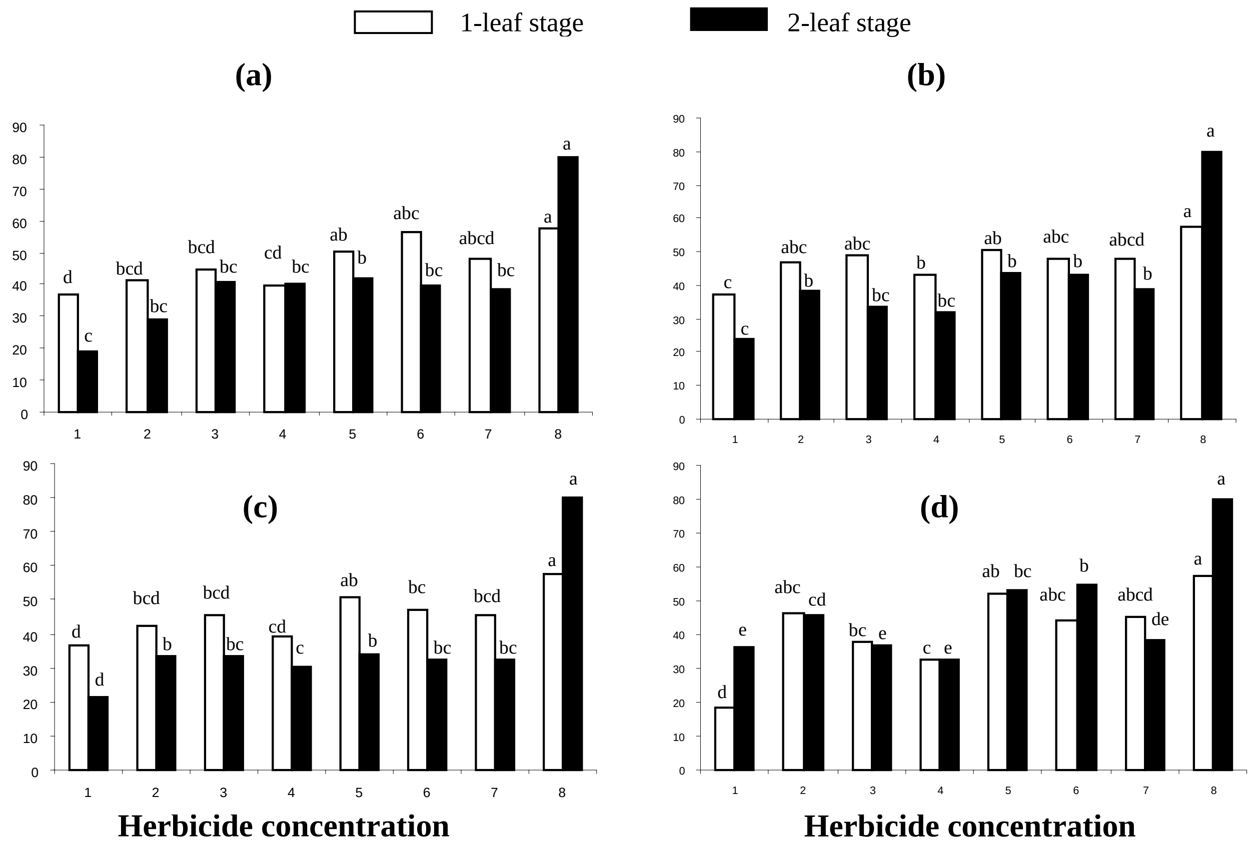


Figure 2. Effect of wood vinegar and sulfonylurea herbicides on the nitrogen content of barnyard grass (*Echinochloa crus-galli* var *crusgalli)* at 1- and 2-leaf stage (a) pyrazosulfuron-ethyl+ esprocarb; (b) pyrazosulfuron-ethyl+molinate; (c) pyrazosulfuron-ethyl+mefenacet; (d) halosulfuron-methyl+pyriminobac-methyl+pretilachlor.

Effect of wood vinegar and HMPMP on the branched-chain amino acid content and the acetohydroxyacid synthase (AHAS) activity

The herbicide that showed the best controlling effect on the growth and nitrogen content of barnyard grass was considered for the analysis of branched-chain amino acid content and assay of AHAS activity. Results (Figure 3) showed that the branched chain amino acids valine and isoleucine in 50 + 1000 WV HMPMP was significantly lower compared to 50% HMPMP alone but significantly higher than that of 100% treatment.

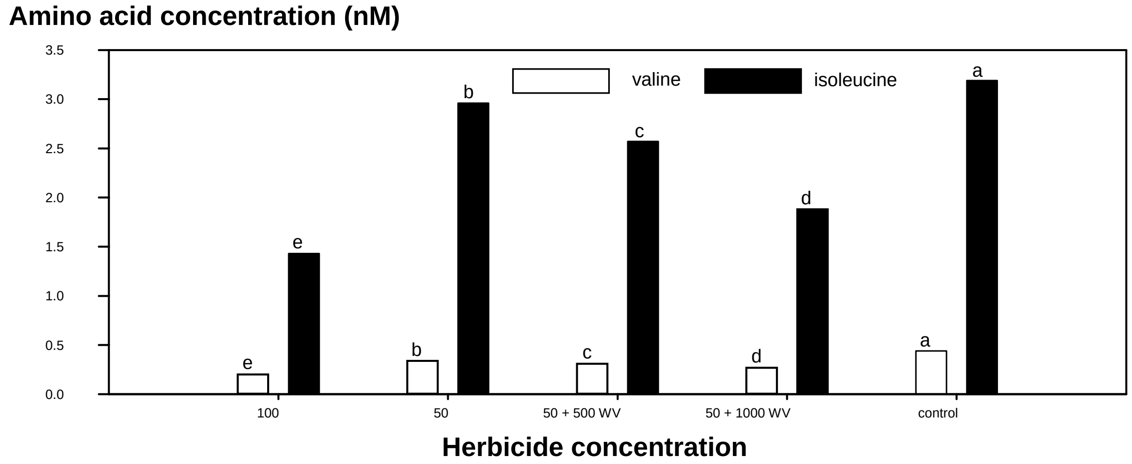


Figure 3. Effect of wood vinegar and halosulfuron-methyl+pyriminobac-methyl+pretilachlor on the branched-chain amino acid content of 2-leaf stage barnyard grass (*Echinochloa crus-galli var. crus-galli*).

The effect of WV and HMPMP on the AHAS activity (Figure 4) showed that 50 + 1000 WV HMPMP caused a significantly higher inhibition of AHAS activity compared to 50% alone and that enzyme activity was already greatly inhibited from the start. The high inhibition of AHAS activity and decrease in branched-chain amino acid in HMPMP plus WV may be attributed to the high concentration of sulfonylurea and pyrimidinyl carboxy type of herbicides in HMPPM which are both affecting the AHAS activity and eventually the branched-chain amino acid synthesis.

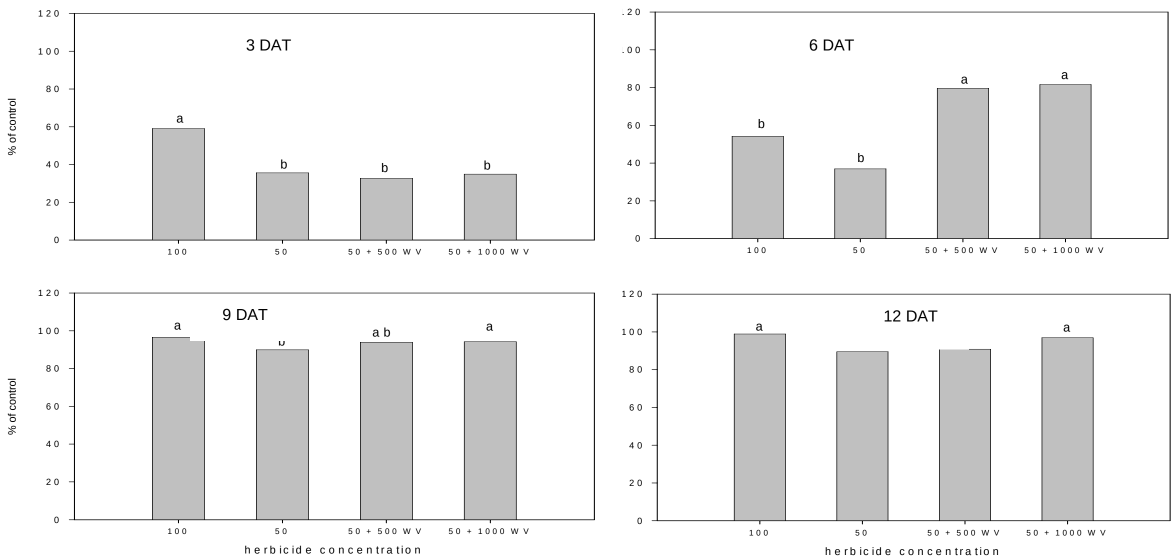


Figure 4. Effect of wood vinegar and halosulfuron-methyl+pyriminobac-methyl+pretilachlor on the acethohydroxyacetic acid synthase of 2-leaf stage barnyard grass (*Echinochloa crus-galli var. crus-galli*).

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CHEMICAL WEED MANAGEMENT IN WHEAT (*Triticum aestivum* L.) IN RAINFED AREA OF PAKISTAN

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Abstract: Dry areas have much more problems of weeds when compared to irrigated areas due to aggressive behavior of weeds. Thus weeds in dry regions competing with crops and lower the yields. A self-conducted survey in Kohat-Pakistan showed that the major weeds were *Chatham's oxyacantha, Avena fatua, Cirsium arvense, Phalaris minor, Convolvulus arvensis, Medicago denticulata, Malcolmia sp., Silybum marianum, Setaria sp., Rumex crispus, Lolium sp., Fumaria indica, Polygonum sp., Vicia sativa and Lathyrus* sp. The yield reduction due to these major weeds ranged from 20-50 % in wheat. An experiment was conducted on the effect of various herbicides for weed control in wheat during the rabi season 2005-2006. The experiment comprised of eight treatments, viz; seven herbicides and a weedy check. The herbicides were; Topik® 15 WP (clodinafop-propargyl) at 0.05 kg, 2, 4-D® 70 SL (2,4-D) at 0.7 kg, Buctril M® 40 EC (bromoxynil + MCPA) at 0.49 kg, Isoproturon® 50 WP (isoproturon) at 1.0 kg, Aim® 40 DF (Chlorfluazuron) at 0.02 kg, Logran extra 64 WG (terbutryn + triasulfuron) at 0.16 kg, Puma Super® 75 EW (fenoxaprop-P-ethyl) at 0.93 kg a.i ha^{-1} and a weedy check. The data recorded on weed kill efficiency (%), fresh weed biomass (kg ha^{-1}), 1000-grain weight (g), biological yield (kg ha^{-1}), and grain yield (kg ha^{-1}) were significantly affected by the different herbicidal treatments. The use of Buctril M® 40EC induced the best performance, with maximum weed kill efficiency (47.2%) and minimum fresh weed biomass (400 kg ha^{-1}) as compared to weed biomass (1102 kg ha^{-1}) in the weedy check. Similarly, number of spikes (506 per m^2), number of grains per spike (57.3), 1000- grain weight (46.6 g), biological yield (16750 kg ha^{-1}) and grain yield (1970 kg ha^{-1}) were the highest in Buctril M® 40 EC treatments as compared to weedy control plot.

Key words: *Triticum aestivum,* weed control, herbicides

Introduction

In irrigated and non-irrigated areas winter weeds drastically reduces yield of cereals, especially wheat. Hand weeding and mixture of herbicides puma super 75 EW and Buctril-M 40 EC has effectively controlled winter weeds (Khan *et al.* 2003). In the NWFP, the area under wheat cultivation is about 0.732 million ha. One third of this area is irrigated, while two third is rainfed, giving a total production of 1.064 million tons at the rate of 1454 kg ha^{-1} (Anonymous, 2005). Weed interference is one of the important but less noticed constraints, contributing towards low yield of wheat.

It has been estimated that crop losses due to weed competition throughout the world as a whole, are greater than those resulting from the combined effect of insect pests and diseases. Weeds may encourage the development of fungal diseases, provide shelter for pests and act as host plants for parasitic nematodes. There are several reasons for entirely eliminating weeds from the crop environment. With the rising costs of labor and power, the use of herbicides will be the only acceptable option of weed control in the future. The infested situations need the development of package of weed management technology, helpful to minimize the weed competition losses. The control of weeds is basic requirement and major component of management in the production system (Young *et al.* 1996).

Conventional methods of weed control are weather dependent, laborious and costly. Thus, chemical weed control has been proved to be relatively efficient, and economical in controlling the weeds (Majid and Hussain, 1983). Weed control has resulted in higher yield in wheat by increasing number of tillers. Among the weed control methods, the chemical control is of recent origin, which is being emphasized, in modern agriculture (Taj *et al*. 1986). The low yield per acre beside many other factors could be attributed to serious weed infestation in the crop. Weed losses are upto 30% in wheat production (Khan and Noor, 1995).

The objectives of the experiment were to find out the most effective herbicide for control of weeds in wheat crop in the rainfed conditions of Kohat and to study the effect of different herbicides on the yield of wheat.

Materials and Methods

The experiment was conducted at Barani Agriculture Research Station, Kohat on variety Kt-2000. The experiment consisted of eight treatments replicated four times, using a Randomized Complete Block Design. The plot size was 5 m x 1.5 m, consisting of six rows each 25 cm apart and 5 m long. The treatments included seven herbicides and a weedy check. All the herbicides were applied post-emergence. All the herbicides mentioned in Table 1 were applied with the help of a knap sack sprayer. While spraying the herbicides, all the precautionary measures were kept in mind to avoid any danger due to the misuse of the herbicides. The crop was harvested at maturity when the grains were fully mature and the crop had senesced. In order to determine the effects of the aforesaid treatments, data were recorded on the weed kill efficiency (%), fresh weed biomass (kg ha^{-1}), number of spikes per m^2, number of grains per spike, thousand grains weight (g), biological yield (kg ha^{-1}), grain yield (kg ha^{-1}) and Cost-benefit ratio. The data recorded for each trait was subjected to ANOVA technique by using MSTATC computer software and means were separated by using Fisher's LSD test (Steel and Torrie, 1980).

Table 1. Treatments used in kohat experiment 2005-06.

Trade name	Common name	Rate (kg a.i. ha^{-1})
Puma super® 75 EW	fenoxaprop-P-ethy	0.93
Topik® 15 WP	Clodinafop-propargyl	0.05
Isoproturon® 50 WP	Isoproturon	1.00
Logran extra® 64 WG	terbutryn + triasulfuron	0.16
Aim® 40 DF	Chlorfluazuron	0.02
2, 4-D® 70 SL	2,4-D	0.70
Buctril M® 40 EC	bromoxynil + MCPA	0.49
Weedy check	---	---

Results and Discussion

Weed control efficacy (%)

The weed species infesting the experiment were *Carthamus oxyacantha, Avena fatua, Cirsium arvense, Phalaris minor, Convolvulus arvensis, Medicago denticulata, Malcomia* sp., *Silybum marianum, Setaria* sp., *Rumex crispus, Lolium* sp., *Fumaria indica, Polygonum* sp., *Vicia sativa* and *Lathyrus* sp. There was a significant ($p<0.05$) effect of different herbicides on weed kill efficiency (Table 2). The maximum weed-kill efficiency (47.2%) was recorded in Buctril-M® 40 EC treatment. Thus, Buctril-M® 40 EC has effectively controlled all weeds, which ultimately increased the final yield. Analogous results were reported by Khan *et al*. (2003) who reported that herbicides application effectively controlled weeds.

Table 2. Effect of different herbicides on weed density and different parameters of wheat.

Treatments	Weed - kill efficacy (%)	Fresh weed Biomass (kg ha^{-1})	No. of spikes per m^{-2}	No. of Grains per Spike	1000-grain weight (g)	Biological yield (kg ha^{-1})	Grain yield (kg ha^{-1})	Cost-benefit ratio (CBR)
Weedy check	5.26 c	1102 a*	400	50.2	41.46 c	10850 b	1553 b	----
Puma super® 75 EW	25.8 b	785 b	460	51.8	41.86 bc	12000 b	1630 b	2.10
Topik® 15 WP	29.1 b	791 b	459	52.4	42.18 bc	11000 b	1695 b	1.69
Isoproturon® 50 WP	32.2 b	435 c	499	51.4	41.86 bc	11200 b	1780 ab	3.97
Logran extra® 64 WG	36.2 ab	466 c	440	55.9	43.89 abc	11900 b	1703 b	2.47
Aim® 40 DF	36.7 ab	778 b	476	54.1	41.50 c	12500 b	1723 b	2.87
2, 4-D® 70 SL	38.4 ab	771 b	431	56.5	44.46 ab	12400 b	1930 a	9.79
Buctril M® 40 EC	47.2 a*	400 c	506	57.3	46.69 a*	16750 a*	1970 a*	10.71
LSD $_{0.05}$	13.3	238	NS	NS	2.948	3438	193.2	

*In each column, means followed by the same letter are not significantly different at p=0.05 by the LSD test.

Fresh weed biomass (kg ha^{-1})
There was significant effect of different herbicides on fresh weed biomass (Table 2). The maximum fresh weed biomass (1102 kg ha^{-1}) was recorded in the weedy check while minimum fresh weed biomass (400 kg ha^{-1}) was recorded in the Buctril-M® 40 EC plots. The difference in the weed biomass in different treatments can be due to the phytotoxic effects of herbicides. Analogous results were reported by Khan *et al*. (2003). They reported that herbicides application decreased the fresh weed biomass as compared to the weedy check. These findings are also in conformity with those of Shahid (1994), who reported that herbicides significantly reduced fresh weed biomass m^{-2}.

Number of spikes per m^2
Different herbicides had similar effects on the number of spikes m^{-2}. Comparison of the treatment means reflected that maximum number of spikes m^{-2} (506) were recorded in Buctril M® 40 EC herbicide while minimum number of spikes m^{-2} (400) were counted in the weedy check. The possible reason for increase in number of spikes m^{-2} by Buctril M® 40 EC may be the effective weed control, which increased nutrients availability to the crop. Khan *et al.* (2003) reported that application of herbicides significantly influenced the number of tillers per m^2.

Number of grains per spike
Number of grains spike^{-1} was non-significant statistically. The highest (57.3) number of grains spike^{-1} recorded in Buctril-M® 40 EC treatment. The lowest (50.2) number of grains spike^{-1} was counted in the weedy check. The lowest number of grains spike^{-1} obtained in weedy check was probably due to the weed competition against the wheat plants which might have greatly reduced the flow of nutrients towards the grains in spikes.

Thousand grain weight (g)
The herbicides had significant effects on thousand grain weight. The highest thousand grain weight (46.6 g) recorded in Buctril M® 40 EC plots (Table 2). The lowest thousand grain

weight (41.46 g) was recorded in the weedy check. The increased thousand grain weight due to Buctril M® 40 EC is attributed to the availability of resources to the wheat crop. As thousand grain weight is a very important yield component in every crop therefore increase in grain weight will automatically increase the total grain yield of the crop. Hassan *et al*. (2003) reported that herbicides application increased 1000 grain weight significantly when compared with the weedy check.

Biological yield (kg ha^{-1})

Herbicides had significant effect on the biological yield (Table 2). The maximum biological yield of (16750 kg ha^{-1}) was recorded in Buctril-M® 40 EC plots, while the minimum biological yield (10850 kg ha^{-1}) was obtained in the weedy check. The highest biological yield is attributable to the better weed management. Hassan *et al.* (2003) reported analogous results.

Grain yield (kg ha^{-1})

Analysis of variance of the data exhibited that herbicides had significant effect on the grain yield of wheat (Table 2). The data indicated that maximum grain yield of (1970 kg ha^{-1}) was produced by those plots to which Buctril M® 40 EC was applied. It was however, statistically at par with plots receiving 2,4-D® 70 SL producing grain yield of 1930 (kg ha^{-1}) and Isoproturon® 50 WP (1780 kg ha^{-1}). The minimum grain yield of (1553 kg ha^{-1}) was obtained in weedy check plots, which was statistically comparable with Puma Super® 75 EW (1630 kg ha^{-1}), Topik® 15 WP (1695 kg ha^{-1}) and all remaining herbicide applied plots. The highest grain yield obtained from Buctril M® 40 EC and other top scoring treatments was perhaps due to their phototoxic effect on weeds, while the lowest grain yield obtained from weedy check plots was probably due to the weed competition with crop. As there was a preponderance of broad leaf weeds hence the grass killers failed to surpass the weedy check. The failure of Logron extra® 64 WG and Isoproturon® 50 WP is attributed to their weaker control of broad leaf weeds. Hence, it can be concluded that Buctril M® 40 EC and 2,4-D® 70 SL are the best choice for broad leaf weed control in wheat. Analogous results were reported by Punia *et al*. (1996) and Khan *et al*. (1999) who obtained higher yield with same herbicides as compared to weedy check.

Cost-Benefit Ratio (CBR)

The cost-benefit ratios were found to be significant for different treatments (Table 2). Maximum approximate cost-benefit ratio was recorded with Buctril M® 40 EC (1: 11) followed by 2, 4-D® 70 SL treatments (1: 10). The lowest cost-benefit ratio was recorded for the Topik® 15 WP treatment (1: 2). These values indicated that all the herbicidal treatments gave acceptable cost-benefit ratio as compared to the yield in the weedy check. The possible reason for the highest return of Buctril M® 40 EC and 2,4-D® 70 SL herbicides might be their lower cost and timely weed control as compared to other herbicides. Similar work has been reported by Pattanaik *et al*. (1996).

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STUDIES ON THE BIO-EFFICACY OF TREFLAN® 48 EC (TRIFLURALIN) AGAINST THE WEEDS OF POTATO

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Abstract: The type and density of weed flora are responsible for reducing tuber yield of potato as well as quality of produced tuber. The crop grows under competition for nutrients, water and weather elements due to the competitive ability of infested weeds. The extent of yield loss of potato ranged from 16 per cent to 76 per cent. A field experiment was conducted during winter season of 2002-03 and 2003-04 at the farmers' field of Surakalna, Burdwan district of West Bengal, India in Randomized block design with eight treatments replicated thrice to evaluate the bio-efficacy of Treflan® 48 EC (Trifluralin) on weeds associated with potato (*Solanum tuberosum* L) cv. Kufri Jyoti. Predominant weed flora in the experimental field were *Amaranthus spp., Portulace oleraceae, Euphorbia hirta, Phylanthus niruri, Chinopodium album* and *Cyperus rotundus*. The highest weed control (74.4%) was observed with Treflan 48® EC (Trifluralin) at 960 g a.i/ha closely followed by Pedimethalin® 30 EC at 1125 g a.i/ha and hand weeding treatments. It was further clear from the experiment that the trend of decrease in density and dry matter of weed ultimately resulted in increases in tuber yield. The increase in tuber yield due to different weed management treatments ranged from 17.3 to 49.68 per cent. Application of Treflan® 48 EC (Trifluralin) at 960 g a.i/ha resulted in the lowest weed infestation in potato which ultimately reflected on the maximum improvement in tuber yield. A succeeding crop of rice was taken in both the years after potato and no residual effect was observed in rice. All yield attributes and grain yield of rice were not influenced by weed management treatments in potato.

Key words: Treflan® 48 EC, weed control, potato

Introduction

India is the leader in Sub-tropical potato production. National scenario showed the picture of increasing annual compound growth rate (ACGR) of 2.4%, in contrast to all the major feed crops, which had either very negligible increase or a decrease in ACGR. So, it is an important crop where research efforts should be made to minimize the biotic and abiotic factors which negatively affect the production. Being a heavy feeder crop, potato needs heavy dressing of fertilizer with frequent irrigation which ultimately encourages weed growth also. The type and density of weed flora have their individual accountability for reducing tuber yield of potato as well as quality of produced tuber. The crop will grow under stress of nutrients, water and weather elements due to the competitive ability of infested weeds. The extent of yield loss of potato crop ranged from 16 per cent to 76 per cent. Prasad and Singh (1995) indicated clearly that a potato crop should be kept under a weed management programme at least up to 40 days of planting. Crop weed competition becomes acute during the early growth stage. Once the crop canopy of potato covered the soil, mostly the annuals are effectively suppressed. Fortunately, the cultural requirement of potato crop namely earthing up is done around this period and manual weeding is complementary to the cultural operation. However, cultural weed control is not feasible and economically not viable. Hand weeding in potato sometimes injures potato roots (Pal Singh *et al.* 2007). Chemical weed control greatly reduces yield losses and is regarded as a useful supplement to mechanical weed control. The present programme, therefore, was undertaken to find out an effective herbicide schedule for controlling the weeds in potato.

Materials and Methods

The field experiment was conducted during the winter season of 2002-03 and 2003-04 at the farmer's field of Surakalna, Burdwan district of West Bengal, India having neutral (pH = 6.8) sandy clay loam soil of medium fertility to find out an effective and economically sound method of weed management where Kufri Jyoti variety of potato was used.

Table 1. Soil characteristics of the experimental site.

Parameter	Unit
Organic Carbon (%)	0.61
Total Nitrogen (%)	0.062
Phosphorus (kg/ha)	16.25
Potassium (kg/ha)	190

The experiment was laid out in a randomised block design with eight treatments replicated thrice with a plot size of 7 x 5 m^2. The treatments were T_1: Treflan® 48 EC at 480 g a.i/ha, T_2: Treflan 48® EC at 600 g a.i/ha, T_3: Treflan® 48 EC at 720 g a.i/ha, T_4: Treflan® 48 EC at 960 g a.i/ha, T_5: pedimethalin® 30 EC at 1125 g a.i/ha, T_6: Fluchloralin® 45 EC at 1125 g a.i/ha, T_7: hand weeding twice at 20 and 40 days after planting (DAP) and T_8: untreated control. All herbicides were sprayed and incorporated in the soil at three days before planting potato seed tubers. Cut tubers were planted 45 cm apart in furrows with an inter-plant spacing of 20 cm. The recommended doses of fertilizer were applied as per standard schedule. Biometrical observations such as identification of predominant weed species, weed population and weed dry weight and yield of potato were recorded as per schedule and standards. A succeeding crop of rice was also taken in both the years to evaluate the residual effect of applied herbicides. The data so obtained as described earlier were subjected to statistical analysis by the analysis of variance method (Panse and Sukhatme, 1978).

Results and Discussion

Associated weed species

The predominant weed species in the field were namely *Amaranthus spp., Portulace oleraceae, Euphorbia hirta, Phylanthus niruri, Chinopodium album* and *Cyperus rotundus.* The observation clearly indicated that cultivation of potato in winter season was mostly hampered by broad leaved weeds.

Total weed population and dry weight

Total weed population differed significantly with different methods of weed management starting from first (30 DAP) to last date (60 DAP) of observations (Table 2). The maximum weed population was recorded in the un-weeded control treatment at all dates of observation. Both the chemical and cultural (hand weeding twice) methods of weed management resulted in considerable reduction of total weed biomass over un-weeded control treatments at all dates of observation. The experimental results revealed that the populations as well as dry weights of weeds were significantly influenced due to the application of herbicides. The lowest weed population and weed dry weight at the initial stage (30 DAP) was observed when the crop received cultural means of weed management. Application of Treflan® at 960 g a..i/ha, pedimethalin® at 1125 g a.i/ha or fluchloralin® at 1125 g a.i/ha significantly reduced total number as well as dry weight of weeds in potato at 30 and 60 DAP over lower doses of Treflan®.

Table 2. Effect of treatments on population and dry weight of weeds and tuber yield of potato.

Treatment	Dose (g a.i/ ha)	Total number of weeds/m^2		Weed dry weight (g/m^2)		Weed control over untreated (%)	Tuber yield (t/ha)	Yield increase over untreated (%)
		30DAP*	60 DAP	30DAP	60 DAP			
Treflan® 48 EC	480	24.52	31.33	16.60	34.50	37.2	22.12	17.3
Treflan® 48 EC	600	22.55	30.30	15.33	32.86	39.3	22.80	21.1
Treflan® 48 EC	720	18.20	22.25	12.40	24.69	55.6	23.61	25.26
Treflan® 48 EC	960	10.10	12.48	6.21	10.73	74.4	28.20	49.68
Pedimethalin® 30 EC	1125	11.23	13.10	6.35	10.95	73.7	28.00	48.61
Fluchloralin 45® EC	1125	11.75	15.76	7.01	14.80	68.4	27.70	47.02
Hand weeding twice		6.33	12.90	2.98	5.38	74.0	27.82	47.56
Untreated control		37.80	49.90	30.18	70.76		18.84	
SE_m (±)		1.73	1.62	1.70	2.18		0.69	
$CD_{0.05}$		5.16	4.79	5.02	6.48		2.05	

Data pooled over two years. DAP* – days after planting

Pre-planting application of Treflan® at 960 g a.i/ha efficiently controlled the weeds starting from the initial stage of crop growth up to 60 DAP. This might be due to the fact that such herbicides had the optimum lethal effect for all dominant weeds and persist in the soil up to the critical growth period of the potato crop.

The percentage of weed control due to Treflan® 48 EC, pendimaethalin® 30 EC and fluchloralin® 45 EC over the untreated control had ranged from 37.2 to 74.4 per cent at 60 DAP. Overall performance regarding control of weeds in potato, Treflan® at 960 g a.i/ha was found to be the best among the other herbicides, which was closely followed by the treatment receiving pendimethalin® at 1125 g a.i/ha. However, it was earlier reported by Bhattacharya *et al.* (2005) that pendimethalin® was an effective chemical for controlling weeds in potato.

Tuber yield of potato

The impact of different weed management treatments on tuber yield of potato is shown in Table 2. Applied weed control treatments improved the tuber yield significantly over the untreated control. The increase in tuber yield ranged from 17.3 to 49.68 per cent. The experimental results showed that Treflan® at 960 g a.i/ha gave the highest tuber yield (28.2 t/ha), which however, did not differ significantly with the treatments receiving pendimethalin® at 1125 g a.i/ha, fluchloralin® at 1125 g a.i/ha and hand weeding (28.0, 27.7 and 27.82 t/ha, respectively). The lowest tuber yield of potato (18.84 t/ha) was recorded under untreated control treatment. Different chemical treatments tried in the experiment did not show any phototoxic symptoms to the crop potato.

Effect on succeeding crop

A succeeding crop of rice was taken in both the years after potato and no residual effects were observed in rice (Table 3). All yield attributes and grain yield of rice were not influenced by weed management treatments.

Treflan® 48 EC at 960 g a.i/ha had significantly controlled the weeds of potato and as efficiently as pendimethalin® 30 EC at 1125 g a.i/ha, fluchloralin® 45 EC at 1125 g a.i/ha and hand weeding at 20 and 40 days after planting. Application of Treflan® 48 EC was not harmful to the succeeding crop of rice. Therefore, Treflan® 48 EC (Trifluralin) at 960 g a.i/ha can profitably be used as an alternative measure to combat the weed infestation especially in situations of labour scarcity at the peak period to overcome the costly and time consuming hand weeding.

Table 3. Residual effects of Treflan® and other herbicides on the succeeding rice crop in the same plot (pooled over two years).

Treatment	Dose (g a.i/ha)	Number of effective tillers/m^2	Straw yield (t/ha)	Grain yield (t/ha)
Treflan 48® EC	480	180.0	3.21	2.25
Treflan 48® EC	600	178.0	3.15	2.21
Treflan 48® EC	720	178.0	3.19	2.20
Treflan 48® EC	960	182.3	3.21	2.28
Pedimethalin® 30 EC	1125	180.3	3.24	2.26
Fluchloralin® 45 EC	1125	181.7	3.08	2.25
Hand weeding twice		182.0	3.12	2.20
Untreated control		180.0	3.05	2.24
$SE_m\pm$		1.89	0.28	0.26
$CD_{0.05}$		NS	NS	NS

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EFFECT OF HERBICIDES ON WEED POPULATION, WEED DRY WEIGHT AND YIELD OF ONION AND SUBSEQUENT RESIDUAL EFFECTS ON CABBAGE, OKRA AND BLACKGRAM

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Abstract: A field experiment with haloxyfop®10 EC applied at varying rates and times of application along with pendimethalin® 30 EC (100 g a.i/ha PPI), fluchloralin® 45 EC (900 g a.i/ha PPI) and oxyfluorfen® 23.5 EC (250 g a.i/ha), along with an untreated check was conducted during 2000-2001 and 2001-2002 to study the effect of herbicides on grassy weeds in onion .Subsequent residual effects of haloxyfop was also studied on cabbage, okra and blackgram. Haloxyfop® 10 EC was applied at 14 or 21 DAT. Pendimethalin® 30 EC and fluchloralin® 45 EC was incorporated prior to planting whereas oxyfluorfen was applied 3 days after transplanting of onion. The dominant grassy weeds were *Echinochloa crus-galli, Echinochloa colona, Cynodon dactylon, Digiteria arvensis* and *Eleusine indica*. Application of haloxyfop® 10 EC 100 g a.i/ha either at14 DAT or at 21 DAT significantly reduced the number as well as dry weight of grassy weeds in both the years and significantly increased the yield of onion crop. Highest onion yield was recorded with oxyfluorfen® 23.5 EC followed by pendimethalin® 30 EC , fluchloralin® 45 EC and haloxyfop® 10 EC 100 g a.i/ha. Haloxyfop® at 100 g a.i/ha was comparable with pendimethalin® at 1000 g a.i/ha preplant application, fluchloralin® at 900 g a.i/ha and oxyfluorfen® at 250 g a.i/ha. Lower rates of application of haloxyfop were less effective in controlling grassy weeds. Haloxyfop has no significant residual effect on subsequent cabbage, okra and blackgram crop.

Key words: Haloxyfop, oxyfluorfen, pendimethalin, onion

Introduction

Onion (*Allium cepa*) is an important vegetable all over the world. It is grown in India and consumed by common people all year round. Onion is consumed fresh in salads or added into cooking dishes as a spice. This crop has a very shallow root system , short stature , small leaves and slow growth rates. Therefore, it is a poor competitor with weeds. In transplanted onion the critical stages is from the time of transplantation to nine weeks and yield reduction estimates ranged between 4.45 and 70.5% (Garcia *et al.* 1994). Application of pendimethalin at 1.32 kg a.i/ha significantly reduced weed population and enhanced yield of onion in Pakistan (Marwat *et al.* 2005) Oxyfluorfen at 100ga.i/ha with hand weeding significantly reduced weed populations and increased the yield of onion (Maity *et al*. 2005).The objective of the study was to evaluate the efficacy and effective dose of herbicides in controlling weeds in onion and to determine the residual effect on succeeding crop of cabbage, okra and blackgram.

Materials and Methods

The experiment on onion was conducted on farmers fields in Barsat Block in 24 parganas, North district of West Bengal, India during winter seasons of 2000-2001 and 2001-2002.The treatments consisted of (a) Haloxyfop® 10 EC 40 g a.i/ha at 14 days after transplanting (DAT), (b) Haloxyfop® 10 EC at 60 g a.i/ha at 14 DAT, (c) Haloxyfop® 10 EC at 80 g a.i/ha at 14 DAT, (d) Haloxyfop® at 100 g a.i/ha at 14 DAT, (e) Haloxyfop® 40 g a.i/ha at 21 DAT, (f) Haloxyfop® at 60 g a.i/ha at 21 DAT, (f) Haloxyfop® 80 g a.i/ha at 21 DAT, (g) Haloxyfop® 100 g a.i/ha at 21 DAT, (h) Pendimethlin® 30 EC 1000 g a.i/ha pre-plant application, (i) Fluchloralin® 45 EC 900 g a.i/ha pre-plant application, (j) Oxyfluorfen® 23.5 EC 250 g a,i/ha at 3 DAT and (k) an untreated check. The experiment was conducted in a Randomized Complete Block Design with three replicates. The plot size was 10 m x 5 m. The onion variety used was Sukhsagar.

Seedlings of onion was transplanted during first week of November and harvested in first week of March in both years. The crop received 80 kg N, 40 kg P_2O_5 and 20 kg K_2O/ha. Half nitrogen and full P and K rates were applied at transplanting and the balance N was applied at 30 days after transplanting. Weed counts and dry weights were taken at 30 and 60 days after transplanting and at harvest. The herbicides were applied as per treatments.

Results and Discussion

The predominant grassy weeds found in onion field were *Echinochloa crus-galli, E. colona, Cynodon dactylon, Nigeria arvensis* and *Elusine indica.* Number of grassy weeds were significantly lower at 30 and 60 DAT with application of pendimethalin® 30 EC at 1000 g a.i/ha pre-plant application, fluchloralin® 45 EC at 900 g a.i/ha pre-plant application, oxyfluorfen® 23.5 EC at 250 g a.i/ha at 3 DAT and haloxyfop10 EC at 100 g a.i/ha at 14 DAT or 21 DAT over lower doses of haloxyfop® well as untreated check in both years (Table1). Oxyfluorfen® 23.5 EC at 250 g a.i/ha was the most effective in reducing numbers of grassy weeds. Marwat *et al*. (2005) reported that application of pendimethalin® at 1.32 kg a.i/ha significantly reduced weed population of onion in Pakistan.

Table 1. Effect of herbicides on number of grasses and yield of onion (t/ha) in2000-2001 and 2001-2002.

Treatment	Dose g a.i. /ha	Time of application	Number of grasses per m²						Bulb yield (t/ha)	
			30 DAT		60 DAT		Harvest			
			00-01	01-02	00-01	01-02	00-01	01-02	2000-2001	2001-2002
Haloxyfop® 10 EC	40	14 DAT	20.1	19.8	27.3	25.8	32.1	30.3	7.74	6.78
Haloxyfop® 10 EC	60	14 DAT	16.5	14.3	22.5	19.2	31.3	25.6	7.75	6.83
Haloxyfop® 10 EC	80	14 DAT	10.3	9.7	11.8	12.5	16.3	17.5	8.15	7.58
Haloxyfop® 10 EC	100	14 DAT	4.3	5.1	5.1	6.8	10.1	8.9	8.82	8.53
Haloxyfop® 10 EC	40	21 DAT	20.4	22.3	27.8	29.3	36.3	34.8	7.58	6.59
Haloxyfop® 10 EC	60	21 DAT	18.7	19.8	24.2	25.5	34.5	26.2	7.62	6.71
Haloxyfop® 10 EC	80	21 DAT	12.5	14.3	14.5	16.1	18.7	17.5	7.82	7,08
Haloxyfop® 10 EC	100	21 DAT	6.6	6.2	7.3	8.2	12.8	10.7	8.35	8.02
Pendimethalin®30 EC	1000	PPI	3.9	4.1	4.2	5.0	8.2	5.4	11.08	10.18
Fluchloralin® 45 EC	900	PPI	3.6	3.3	5.1	4.8	7.8	5.2	10.92	10.05
Oxyfluorfen® 23.5 EC	250	3 DAT	2.3	2.5	3.7	2.9	5.5	3.1	11.26	10.41
Untreated check			21.3	24.8	32.7	36.1	38.1	42.3	7.45	6.23
CD (p=0.05)			1.56	1.78	1.81	2.16	2.14	2.48	5.17	0.476

DAT=Days after transplanting, PPI= Pre plant incorporation

The bulb yield of onion was highest with application of oxyfluorfen® 23.5 EC at 250 g a.i/ha at 3 DAT followed by pendimethalin® 30 EC at 1000 g a.i/ha as a pre-plant application, fluchlorain® 45 EC at 900 g a.i/ha pre-plant application and haloxyfop® 10 EC at 100ga.i/ha at 14 or 21 DAT over other treatments including untreated check (Table 1). The weeds were effectively controlled by the application of above treatments resulting in better growth of onion crop and thereby enhancing yield. Application of oxyfluorfen® 100ga.i/ha with hand weeding increased yield of onion in India (Maity *et al*, 2005). Pendimethalin® was also reported to be an effective herbicide in controlling weeds in onion crop (Singh *et al.* 1992; Marwat *et al.* 2005; Maity *et al*. 2005)

Dry weights of grassy weeds and weed control efficiency (WCE) as influenced by different herbicide application is presented in Table 2. Dry weights of grassy weeds was lowest with application of oxyfluorfen® 23.5 EC at 250 g a.i/ha at 3 DAT followed by pendimethalin® pre-plant application, fluchloralin® 45 EC at 900 g a.i/ha pre-plant application and haloxyfop10 EC at 14 DAT or 21 DAT over lower doses of haloxyfop® and untreated check at 30 and 60 DAT in both years. Weed control efficiency was highest with oxyfluorfen® 23.5 EC at 250 g a.i/ha at 3 DAT followed by pendimethalin®30 EC at 1000 g a.i/ha pre-plant application, fluchloralin® 45 EC at 900 g a.i/ha pre-plant application and haloxyfop®10 EC at 100 g a.i/ha at14 or 21 DAT over lower doses of haloxyfop® and the untreated control. The results agree with those of Maity *et al.* (2005).

Table 2. Effect of herbicides on dry weight of grasses (g/m^2) and WCE in onion

Treatments	Dose g a.i /ha	Time of Appli.*	Dry weight of grasses g/m^2						Weed control efficiency					
			30 DAT		60 DAT		Harvest		30 DAT		60 DAT		Harvest	
			00-01	01-02	00-01	01-02	00-01	01-02	00-01	01-02	00-01	01-02	00-01	01-02
Haloxyfop® 10 EC	40	14 DAT	16.5	14.7	44.3	40.4	76.4	70.1	12.2	27.2	21.3	34.0	13.7	27.2
Haloxyfop® 10 EC	60	14 DAT	13.2	12.1	39.1	33.7	62.1	56.3	29.8	40.1	40.1	47.4	29.8	41.5
Haloxyfop® 10 EC	80	14 DAT	7.8	6.5	28.3	22.4	46.7	40.5	58.5	67.8	49.7	65.1	47.2	57.9
Haloxyfop® 10 EC	100	14 DAT	3.9	3.2	8.9	7.5	14.4	12.3	79.3	84.2	84.2	88.3	83.7	87.2
Haloxyfop® 10 EC	40	21 DAT	18.3	16.1	52.1	46.1	81.5	78.2	2.65	20.2	7.46	28.1	7.9	18.8
Haloxyfop® 10 EC	60	21 DAT	14.1	13.8	46.9	40.5	68.2	61.5	25.0	31.7	16.7	36.8	22.9	36.1
Haloxyfop® 10 EC	80	21 DAT	9.5	6.8	32.3	28.7	51.8	46.7	49.5	66.4	42.6	55.2	41.5	51.5
Haloxyfop 10 EC	100	21 DAT	5.3	4.5	11.8	9.2	19.6	16.5	71.8	77.7	79.0	85.6	77.8	82.7
Pendimethalin® 30EC	1000	PPI	1.2	1.1	5.3	4.5	10.8	10.1	93.6	94.6	90.6	92.8	87.8	89.5
Fluchloralin® 45 EC	900	PPI	1.3	1.4	6.1	5.6	10.3	9.7	93.1	93.1	89.2	91.3	88.4	90.0
Oxyfluorfen® 23.5 EC	250	3 DAT	0.8	0.7	4.9	4.2	8.1	9.1	95.7	96.5	91.3	93.4	90.8	90.6
Untreated check			18.8	20.2	56.3	64.1	88.5	96.3						
CD (p=0.05)			0.76	0.69	4.28	5.16	3.44	6.08						

* Time of application, DAT=Days after transplanting, PPI= Pre plant incorporation

The residual effect of haloxyfop® 10 EC at different doses was studied on succeeding crops of cabbage, okra and blackgram and the data is presented in Table 3. There was no significant residual effect of haloxyfop®10EC at different rates on the yields of succeeding crops of cabbage, okra and blackgram.

Table 3. Residual effect of Haloxyfop10EC on succeeding crops of cabbage, okra and blackgram

Treatments	Dose g a.i/ ha	Time of application	Yield (t/ha)					
			Cabbage		Okra		Blackgram	
			2000-01	2001-02	2000-01	2001-02	2000-01	2001-02
Haloxyfop® 10 EC	40	14 DAT	21.13	18.12	7.23	6.57	1.15	1.27
Haloxyfop® 10 EC	60	14 DAT	20.23	17.75	7.18	6.83	1.18	1.20
Haloxyfop® 10 EC	80	14 DAT	21.51	17.90	7.15	6.67	1.21	1.18
Haloxyfop® 10 EC	100	14 DAT	20.85	18.37	7.15	7.00	1.17	1.20
Haloxyfop® 10 EC	40	21 DAT	21.33	18.14	7.23	6.91	1.19	1.23
Haloxyfop® 10 EC	60	21 DAT	21.48	17.81	7.02	7.15	1.20	1.21
Haloxyfop® 10 EC	80	21 DAT	21.27	18.08	6.78	7.09	1.23	1.25
Haloxyfop® 10 EC	100	21 DAT	20.91	18.40	7.05	6.73	1.21	1.20
Untreated control			21.02	18,05	6.85	6.97	1.19	1.28
CD (p=0.05)			NS	NS	NS	NS	NS	NS

NS= Not Significant

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SURFACTANTS ENHANCE PRIMISULFURON ACTIVITY IN COMMON LAMBSQUARTERS (*Chenopodium album* L.)

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Abstract: Common lambsquarters (*Chenopodium album* L.) is one of the most widely distributed weed species in the world and is competitive with 40 crops. Greenhouse and laboratory studies were conducted to determine the effect of non-ionic (Induce®) and organosilicone (Silwet L-77®) surfactants on primisulfuron activity and droplet contact angle on common lambsquarters and to examine the pattern of primisulfuron droplet spread on leaf surface using scanning electron microscopy. The activity of primisulfuron at 0, 20, 40, 60 and 80 g ai ha^{-1} was assessed 3 weeks after treatment in terms of percent injury and fresh weight. The contact angles of both sides of the 1 µl droplets on the leaf surface were measured using a goniometer. Primisulfuron activity on common lambsquarters in terms of percent injury and fresh weight was significantly enhanced by surfactant type and primisulfuron rates. There was no difference between the two surfactants in enhancing primisulfuron activity at all rates of primisulfuron. Both surfactants significantly reduced contact angles of primisulfuron droplets on common lambsquarters leaf surfaces. Silwet L-77® when mixed with primisulfuron had the lowest contact angle on the leaf surface due to very low surface tension of the solution. Scanning electron micrographs showed that with Silwet L-77® the droplet spread was more and it gradually blended with the leaf cuticular surface. The increased activity of primisulfuron with the surfactants was related to the lower contact angle and greater spread of spray droplets on leaf surface of common lambsquarters.

Key words: *Chenopodium album,* common lambsquarters, contact angle, primisulfuron, scanning electron microscopy, surfactant.

Introduction

Common lambsquarters (*Chenopodium album* L.) is one of the most important weeds in corn and soybean. In field corn and sugar beets (*Beta vulgaris* L.), yield reductions of 11 and 48%, respectively, have been associated with interference by this weed (Beckett *et al.* 1988; Schweizer, 1983). In vegetable crops, season-long interference by common lambsquarters has resulted in 36% yield reductions in tomato with 64 plants per meter of row (Bhowmik and Reddy, 1988) and 65% yield reductions in lettuce (*Lactuca sativa* L.) with 1.33 plants per meter of row (Santos *et al.* 2004).

Primisulfuron is a selective postemergence herbicide and its activity is often affected by surfactant type and the target weed species. Nandula et al. (1995) reported that primisulfuron provided greater wirestem muhly [*Muhlenbergia frondosa* (Poir.) Fern.] control with methylated vegetable oil concentrate as compared to nonionic surfactant, whereas, in field experiments, primisulfuron (39 g ai ha^{-1}) provided greater control of itchgrass (*Rottboellia cochinchinensis*) when applied with nonionic surfactant than with organosilicone or methylated seed oil blend (Strahan *et al.* 2000). It is apparent from these works that activity of primisulfuron can be increased by proper selection of adjuvant for a particular weed species. The objectives of this study were to (a) measure the contact angle of primisulfuron droplets with and without surfactants on leaf surface, (b) examine the pattern of primisulfuron droplet spread on leaf surface, and (c) determine primisulfuron activity on common lambsquarters with and without surfactants.

Materials and Methods

Common lambsquarters plants were grown in 10 cm^2 plastic pots containing Hadley fine sandy loam soil with 3.5% organic matter and a pH of 6.5 in greenhouse with an average temperature of 20 ± 2^{o}C with natural sunlight. Plants were thinned to 5 per pot and were watered as needed. Plants were at five- to six-leaf stage for all experiments.

Contact angle of 1-µl droplets of primisulfuron (Beacon®; water-dispersible granule) alone, with Induce® (non-ionic surfactant, blend of alkyl aryl polyoxylkane ethers, free fatty acids, and dimethyl polysiloxane) at 0.25% (v/v) and with Silwet L-77® (organosilicone surfactant, polyalkyleneoxide-modified heptamethyltrisiloxane 7.5 EO, 100%) at 0.1% (v/v) were measured on the adaxial surface of the fourth to sixth fully expanded leaves from the tip of the plants. The contact angles of both sides of the 1 µl droplets on the leaf surfaces were measured using a Contact Angle Goniometer (Rame-hart, Inc., 43 Bloomfield Avenue, Mountain Lakes, NJ 07046). Each value was the mean of contact angles of two sides of the droplet. The experimental design was completely randomized. Contact angle measurements were replicated four times, and the experiment was repeated. Data were subjected to ANOVA using the General Linear Model procedure (SAS 1992), and means were separated using Fisher's Protected LSD test at p = 0.05.

Plant specimens were prepared for scanning electron microscopy by mounting leaf segments on aluminum stubs using double sticky carbon tape. 1-µl droplets of primisulfuron alone, with Induce® at 0.25% (v/v) and with Silwet L-77® at 0.1% (v/v) were applied on the mounted leaf segments. Silver paint was used to attach the margins of leaves to the aluminum stubs. These leaf specimens were air dried for 30 minutes and examined under scanning electron microscope (JEOL-JSM 840, USA, 11 Dearborn Road, Peabody, MA 10960).

Effect of surfactants on primisulfuron activity was determined using 0, 20, 40, 60, and 80 g ai ha^{-1} rates, where 40 g ai ha^{-1} represented the manufacturer suggested use rate. All rates of primisulfuron were applied alone, with Induce® at 0.25% (v/v) and with Silwet L-77® at 0.1% (v/v). An untreated control was included for the comparison. Spray solutions were applied using a CO_2-backpack sprayer with TeeJet XR 11004VS nozzles at 152 kPa using a spray volume of 190 l ha^{-1}. Primisulfuron activity was assessed 3 weeks after treatment in terms of percent injury and fresh weight. Percent injury was estimated visually on a scale of 0 (no injury) to 100% (plant death). Plant shoots were clipped at the soil surface and the fresh weights were recorded. The experimental design was completely randomized. Treatments were replicated three times and the experiments were repeated. Data were subjected to ANOVA using the General Linear Model procedure (SAS 1992), and means were separated using Fisher's protected LSD test at p= 0.05.

Results and Discussion

Contact angles of 1 µl droplet of primisulfuron on the adaxial leaf surface of common lambsquarters were higher when applied without surfactant (Figure 1). High wax content per unit of leaf area in common lambsquarters (Sanyal *et al.* 2006) could be a probable reason for high contact angle of primisulfuron droplets on the leaf surface. In general, leaf wax content and spread area of the spray droplet is inversely related (Chachalis *et al*. 2001). However, composition, physical structure, and orientation of leaf wax also play important roles in this regard (Juniper, 1960; Whitehouse *et al.* 1982). Both surfactants significantly (with a *P* value of <0.0001 in ANOVA) reduced contact angles of primisulfuron droplets on common lambsquarters leaf surfaces (Figure 1). Silwet L-77® when mixed with primisulfuron had the lowest contact angle on the leaf surface due to very low surface tension of the solution. Surfactant reduces the surface tension and contact angle of herbicide droplets, thereby improving the coverage and increasing the chance for a herbicide to absorb into the plant tissue (Kocher and Kocur, 1993). Addition of surfactant to spray solution enhances the solution's ability to wet the leaf surface. However, there are variations of wetting properties among surfactants (Wells, 1989; Knight and Kirkwood, 1991). Our data show that Silwet L-77® reduced the contact angle of primisulfuron droplets more than Induce®, similar to that reported by Wells (1989) and Knight and Kirkwood (1991).

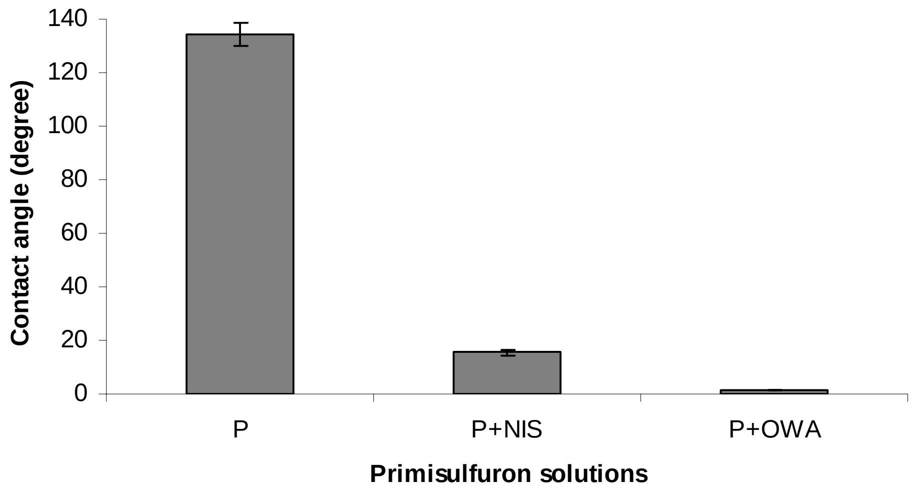

Figure 1. Contact angle of 1-µl droplets of primisulfuron without surfactant (P), with Induce® (non-ionic surfactant) (P+NIS), and with Silwet L-77® (organosilicone surfactant) (P+OWA) on the leaf surface of common lambsquarters. Primisulfuron was used at 40 g ai ha^{-1}, Induce® at 0.25% (v/v), and Silwet L-77® at 0.1% (v/v).

Scanning electron micrographs showed the spreading pattern of primisulfuron droplets with Induce®and Silwet L-77® (micrographs not shown). Both surfactants spread primisulfuron droplets to a greater extent as compared to that without a surfactant. With Silwet L-77® the droplet spread was more and it gradually blended with the leaf cuticular surface in such a way that the boundary of the droplet was not visible.

The primisulfuron rates and surfactants had significant effects on per cent injury and fresh weight of common lambsquarters as determined by the probability values for each of the variables are given in Table 1.

Table 1. Probability values from ANOVA for effect of surfactant types and primisulfuron rates on injury and fresh weight of common lambsquarters.

	Source[1]	Injury	Fresh weight
Percent injury	ST	<0.0001	<0.0001
	PR	<0.0001	<0.0001
	ST*PR	<0.0001	<0.0001
Fresh weight	ST	<0.0001	<0.0001
	PR	<0.0001	<0.0001
	ST*PR	<0.0001	<0.0001

[1]Surfactant types (without surfactant, with nonionic surfactant Induce®, and with organosilocone surfactant Silwet L-77®), primisulfuron rates (0, 20, 40, 60 and 80 g ai ha^{-1}), and their interactions were denoted by ST, PR, and ST*PR, respectively.

At 40 g ai ha^{-1} primisulfuron injured common lambsquarters only 26% when applied without a surfactant, whereas, 55 and 57% injury occurred when applied with Induce® and Silwet L-

77®, respectively (Figure 2). There was no difference between the two surfactant treatments in terms of percent injury of common lambsquarters at all rates of primisulfuron.

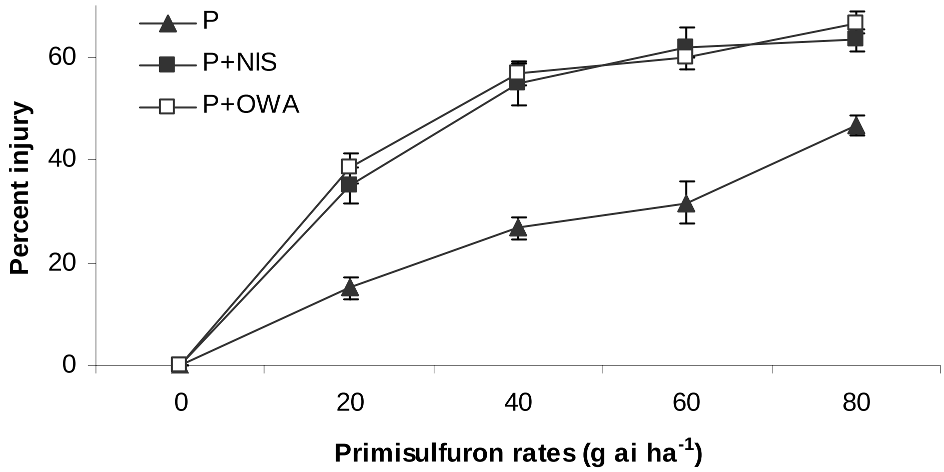

Figure 2. Effect of various rates of primisulfuron on common lambsquarters injury. All rates of primisulfuron were applied alone (P), with Induce® (non-ionic surfactant) at 0.25% (v/v) (P+NIS), and with Silwet L-77® (organosilicone surfactant) at 0.1% (v/v) (P+OWA). LSD values for primisulfuron rates and surfactant types were 4.18 and 3.24, respectively.

Both surfactants reduced the fresh weight of common lambsquarters as compared to the treatment without any surfactant (Figure 3). The fresh weight was sharply reduced from 0 to 40 g ai ha^{-1} rate when applied with a surfactant, whereas, from 40 to 80 g ai ha^{-1} fresh weight reduction was not significant. No differences in fresh weights were observed between two surfactant treatments at 40 g ai ha^{-1} or higher rates of primisulfuron.

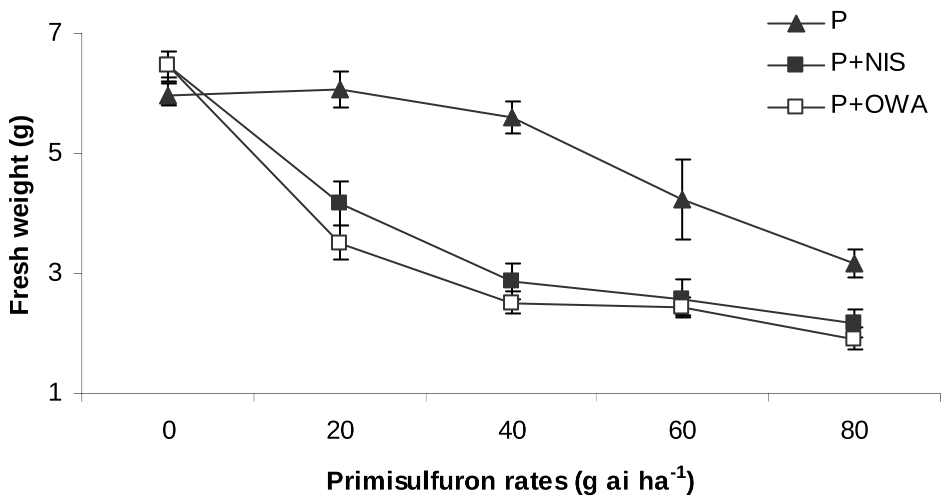

Figure 3. Effect of various rates of primisulfuron on common lambsquarters fresh weight. All rates of primisulfuron were applied alone (P), with Induce® (non-ionic surfactant) at 0.25% (v/v) (P+NIS), and with Silwet L-77® (organosilicone surfactant) at 0.1% (v/v) (P+OWA). LSD values for primisulfuron rates and surfactant types were 0.49 and 0.38, respectively.

The greater spread with a lower contact angle with the surfactants may cause greater absorption of the herbicide into the plant tissue and as a result caused higher activity of primisulfuron. In quackgrass, the shoot regrowth was reduced by primisulfuron application to a greater extent with the linear alcohol ethoxylate additive 1412-70 as compared to Scoil, Sylgard 309, Agridex, or X-77 (Gillespie and Vitolo, 1993). In contrast, Morton and Harvey (1994) reported that giant foxtail control was not affected by primisulfuron rates or adjuvants. Control of johnsongrass and quackgrass were greatest with primisulfuron plus the ethoxylated alcohols 1412-60 and 1412-70, which had chain lengths of 14 and 12 carbon atoms and 60 or 70% ethoxylation (Dunne *et al.* 1994). Our results showed that primisulfuron activity on common lambsquarters was greatly enhanced by the organosilicone surfactant Silwet L-77® and the non-ionic surfactant Induce®.

Acknowledgement

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ALLELOPATHIC POTENTIAL OF INVADED WEEDS ON A DIVERSE WEED SPECTRUM OF POTATO (*Solanum tuberosum* L.)

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Abstract: Parthenium (*Parthenium hysterophorus*), Chromolaena (*Chromolaena odorata*) and Lantana (*Lantana camara*) are major invaded weeds in India. These weeds are reported to have pronounced allelopathic effects and higher nutrient contents in their biomass. The allelopathic potential and manurial value of invaded weeds were investigated through a series of laboratory and field experiments for exploitation as components of a weed management programme for potato (*Solanum tuberosum* L.). The effect of utilizing invaded weeds as a nutrient source for maximizing the production potential, monetary returns and energy input-output of potato was studied.

Key words: Allelopathic potential, chromolaena, lantana, nutrient source, parthenium, potato

Introduction

Parthenium (*Parthenium hysterophorus*), chromolaena (*Chromolaena odorata*) and lantana (*Lantana camara*) are major invaded weeds in India. These weeds are reported to have higher nutrient contents and pronounced allelopathic effects in their biomass. The term allelopathy refers to any direct or indirect harmful effect by one plant on another through the production of chemical compounds released into the environment (Rice, 1984). Allelopathy is one of the major non-chemical and non-hazardous means of weed management in sustainable crop production. Potato (*Solanum tuberosum* L.) is an important crop in the Indian sub continent. It has got a unique capacity to produce high energy and protein ha^{-1} day^{-1}. It is heavily fertilized to get higher biomass and yield per unit area. Potato receives larger amounts of the nutrients through inorganic (synthetic fertilizers) and organic sources (FYM and green manures). Recently, the availability of organic manures like FYM and green manures has been continuously declining in Indian agriculture. These invaded weeds can be used as a substitute for FYM. Thus, there is a feasibility of sustaining the potato yield by supplying major nutrients through combined application from inorganic and organic sources. So, the allelopathic potential and manurial value of the invaded weeds were investigated through a series of laboratory and field experiments, for exploitation as components of weed and nutrient management programmes in Potato (*Solanum tuberosum* L.).

Materials and Methods

The study comprised of three laboratory studies and one field experiment. Testing the allelopathic potential of invaded weeds (Parthenium, Chromolaena and Lantana) was conducted as three different laboratory experiments using air-dried leaf, stem, flowers and their combinations soaked in distilled water (1:10 w/v) for 1, 3 and 7 days. Aqueous extracts were collected and filtered in Whatman No.1 filter paper. Viable seeds of finger millet (*Eleusine coracana*) were soaked overnight, surface sterilized and later spread on Petri plates at 25 per plate over a moist filter paper. It was maintained with a slow and continuous exposure to 10 ml of aqueous extracts of three allelopathic plant products and 10 ml of distilled water (for comparison as control), observed for germination, dry matter and vigour index (Abdul-baki and Anderson, 1973) on the 8th day. Testing the allelopathic and manurial potential of invaded weeds under field conditions was conducted during *rabi* seasons of 2002-

2004 at the University of Agricultural Sciences, Bangalore, Karnataka, India. The soil of the experimental field was sandy loam in texture. The experiment was laid out in a randomized block design with 15 treatments, *viz*. (T_1) 75% Nitrogen (N) as inorganic + 25% N as *Parthenium hysterophorus* incorporation, (T_2) 75% N as inorganic + 25% N as *P. hysterophorus* compost, (T_3) 75% N as inorganic + 25% N as *Chromolaena odorata* incorporation, (T_4) 75% N as inorganic + 25% N as *C. odorata* compost, (T_5) 75 % N as inorganic + 25% N as *Lantana camara* incorporation, (T_6) 75% N as inorganic + 25% N as *L. camara* compost, (T_7) 50% N as inorganic + 50% N as *P. hysterophorus* incorporation, (T_8) 50% N as inorganic + 50% N as *P. hysterophorus* compost, (T_9) 50% N as inorganic + 50% N as *C. odorata* incorporation, (T_{10}) 50% N as inorganic + 50% N as *C. odorata* compost, (T_{11}) 50% N as inorganic + 50% N as *L. camara* incorporation, (T_{12}) 50% N as inorganic + 50% N as *L. camara* compost, (T_{13}) 100% N as farmyard manure (FYM) (m) 100% NPK alone (125:100:125 kg ha^{-1}), and (T_{14}) 100% NPK+ FYM at 10 t ha $^{-1}$. The treatments from T_1-T_6 were uniformly applied with 75% N as inorganic and remaining 25% N was supplied through weed biomass incorporation and compost. The treatments from T_7-T_{12} were applied with 50% N as inorganic and remaining 50% N was supplied through weed biomass incorporation and compost respectively. The different sources like parthenium, chromolaena, lantana and FYM were analyzed for nitrogen content and the results are given in Table 1.

Table. 1. Nutrient composition of invaded weeds and FYM used in the experiment.

Organic manures	Nutrient composition on oven dry weight basis (%)			Carbon (%)	C : N ratio
	N	P	K		
FYM	0.61	0.43	0.72	24.40	40.00
Parthenium biomass	2.63	0.23	1.40	48.50	18.44
Parthenium compost	1.44	0.17	0.92	21.64	15.02
Chromolaena biomass	2.51	0.47	2.15	45.90	18.29
Chromolaena compost	1.37	0.22	0.69	22.37	16.26
Lantana biomass	2.40	0.31	1.73	53.40	22.25
Lantana compost	1.09	0.14	0.45	22.42	20.56

The potato variety used in the experiment was Kufri Jyothi with the spacing of 45 cm x 20 cm. The crop was harvested 84 days after planting. Economics of the treatment were computed based upon existing market prices. Energy equivalents for direct and indirect sources were calculated (Mittal *et al*. 1985) for all inputs.

Results and Discussion

Testing the plant products for allelopathic potential under laboratory conditions showed a clear difference in the potential among them in eliciting allelopathic inhibition (Table 2). Plant extracts of parthenium significantly inhibited the germination and reduced the dry matter production of finger millet. Leaf extract of parthenium significantly reduced the germination percentage of finger millet (51.1%), compared to stem, flower and their combinations. This reduced germination might be due to the presence of sesquiterpene lactones mainly parthenin, traces of coronopillin and phenolics (Kanchan and Jeyachandra, 1979). Moreover, the presence of parthenin in leaf extract might have caused mutation and inhibition of seed germination as well as growth (Patil and Hegde, 1988).The other plant extracts like chromolaena and lantana extracts did not influence the germination percentage and dry matter production but influenced the vigour index.

Table 2. Effect of allelopathic plant extracts on germination and growth of finger millet.

Treatments	Parthenium extract			Chromolaena extract			Lantana extract		
	Germ. %	DM (mg)	VI	Germ. %	DM (mg)	VI	Germ. %	DM (mg)	VI
Extracts									
Leaf (L)	51.1	0.5	27.6	97.8	1.6	160.9	95.1	1.3	121.4
Stem (S)	97.8	1.3	131.9	98.7	2.0	198.5	98.7	1.7	165.4
Flower (F)	94.7	1.2	115.1	98.2	1.7	170.4	95.1	1.5	145.5
L+S+F	95.1	1.2	114.2	97.8	1.9	181.5	96.4	1.6	158.4
Control	100.0	1.8	158.9	99.6	2.1	209.1	100.0	1.7	165.0
S.Em. ±	1.6	0.1	7.7	0.7	0.1	6.1	1.4	0.1	10.9
CD (p=0.05)	4.7	0.2	17.2	NS	NS	17.6	NS	NS	31.4
Soaking periods									
1 day	79.7	1.0	93.4	97.1	1.8	177.5	95.7	1.4	135.6
3 day	90.1	1.2	114.3	98.1	1.9	181.4	97.1	1.6	156.0
7 day	93.3	1.4	131.6	100.0	1.9	193.3	98.4	1.6	161.6
S.Em. ±	1.3	0.1	6.0	0.6	0.1	4.7	1.1	0.1	8.4
CD (p=0.05)	3.7	0.2	17.2	NS	NS	13.6	NS	NS	NS
S.Em. ±	2.8	0.1	13.3	1.2	0.1	10.5	2.4	0.2	18.8
CD (p=0.05)	8.2	NS	NS	NS	NS	NS	NS	NS	NS

Germ. %= Germination Percentage DM= Dry matter VI= Vigour Index

Among soaking periods, a shorter soaking period of 1 day duration had higher inhibition in germination and reduction in dry matter production and vigour index compared to longer soaking periods (3 and 7 day durations). The longer soaking periods resulted in lesser inhibition of germination and improved growth of finger millet, which might be due to the microbial degradation of allelochemicals from plant products over the period of time.

The broad spectrum of weeds observed in the experimental field were *Eleusine indica*, *Digitaria marginata*, and *Dactyloctenium aegyptium*, *Cynodon dactylon* (grasses), *Parthenium hysterophorus*, *Borreria hispida*, *Amaranthus viridis* and *Commelina benghalensis* (broad leaved weeds) and *Cyperus rotundus* as the lone sedge. Performance of the allelopathic invaded weeds on the broad spectrum of weeds under field conditions revealed that the density of grasses, sedges and total weeds m^{-2} did not show any significant difference due to weed utilization treatments (Table 3). But, the highest total weed count in potato was recorded with application of 100% N as FYM (100.3 m^{-2}). This might be due to cattle manure serving as the source of monocot weeds (Jane and Kenneth, 1994), which was in conformity with the present investigation. However, a higher broad leaved population was observed in the plots where substitution of nitrogen by parthenium green biomass (35.3 and 24.7 m^{-2}, respectively) had occurred. This was due to emergence of parthenium plants in those plots. In the present study, the utilization of invaded weeds for allelopathic potential did not inhibit the germination and emergence of broad spectrum of weeds under field conditions. It might be due to allelochemicals released from the weeds which would have degraded by soil microbes and other complex factors during longer periods of decomposition in the soil. This result was clearly supported by the results of the laboratory experiments conducted by the authors (Table 2).

In utility point of view, integration of organic weed sources and inorganic nutrient sources produced a higher tuber yield of potato rather than application of fertilizer or FYM alone (Table 3). The highest total tuber yield was recorded with 100% NPK + 10 t FYM (T_{15}: 21.05 t ha^{-1}) which was on par with 25% N supplied as chromolaena incorporation (T_3: 20.11 t ha^{-1}). It was closely followed by the application of 25% N either as parthenium compost (18.27 t ha^{-1}) or lantana incorporation (18.20 t ha^{-1}). Application of 100% NPK + 10 t FYM, substitution of 25% N through chromolaena or lantana weed incorporation or parthenium compost could be attributed to better availability of nutrients, which helped in greater synthesis of carbohydrates and their translocation, better improvement in vegetative growth and increased yield attributes and yield (Sasani *et al*. 2003). Substitution of 100% N as FYM (12.68 t ha^{-1}) and 50% N as lantana compost (13.02 t ha^{-1}) recorded significantly lowest tuber yields. This might be due to temporary immobilization of nitrogen during earlier stages of potato, which in turn, reduced the yield attributes and yield.

Table 3. Effect of different treatments on weed density, yield attributes and yield of potato

Treatments	Grasses (m^2)	BLW (m^2)	Sedges (m^2)	Total weeds (m^2)	Tuber weight (g)	Tubers plant^{-1}	Tuber yield (t/ha)
T_1	7.7 (61.3)	4.9 (24.7)	2.1 (4.6)	9.4 (90.6)	209.65	4.78	16.14
T_2	7.7 (59.3)	3.0 (9.7)	2.9 (10.6)	8.9 (79.6)	219.10	4.98	18.27
T_3	7.6 (59.3)	0.7 (0.0)	2.9 (10.7)	8.3 (70.0)	234.29	5.35	20.11
T_4	7.2 (53.3)	3.0 (10.0)	1.9 (4.0)	8.0 (67.3)	208.87	4.78	16.52
T_5	8.4 (71.6)	1.8 (3.3)	2.5 (8.7)	9.1 (83.6)	212.26	5.22	18.20
T_6	7.3 (56.4)	3.1 (9.3)	3.2 (11.3)	8.6 (77.0)	188.83	4.73	15.63
T_7	6.8 (50.3)	5.9 (35.3)	1.9 (4.0)	9.4 (89.6)	191.58	4.43	14.15
T_8	8.4 (72.0)	2.0 (4.7)	2.5 (6.6)	9.1 (83.3)	184.90	4.67	14.22
T_9	6.4 (42.7)	2.6 (7.3)	2.3 (8.0)	7.5 (58.0)	190.84	4.37	14.24
T_{10}	8.8 (77.3)	1.5 (2.0)	1.4 (2.5)	9.0 (81.8)	184.58	4.43	14.50
T_{11}	7.6 (60.3)	3.0 (8.7)	1.7 (4.5)	8.5 (73.5)	182.63	4.37	13.78
T_{12}	5.9 (38.5)	1.7 (2.7)	4.5 (21.3)	7.8 (62.5)	187.23	4.27	13.02
T_{13}	9.7 (96.3)	1.0 (0.7)	1.7 (3.3)	9.9 (100.3)	175.16	4.05	12.68
T_{14}	8.2 (67.3)	2.8 (8.7)	1.9 (4.0)	8.9 (80.0)	209.10	4.78	17.26
T_{15}	7.3 (54.3)	1.4 (2.0)	2.9 (12.0)	8.2 (68.3)	239.18	5.63	21.05
S.Em$\pm$	0.88	0.48	0.72	0.73	6.40	0.16	0.38
CD (p=0.05)	NS	1.39	NS	NS	18.53	0.46	1.10

Figures in parantheses indicates the original values BLW = Broad leaved weeds

A higher benefit:cost ratio and energy input-output was obtained with application of 25% N as chromolaena to potato (3.2 and 2.6, respectively). The higher benefit:cost ratio was due to lower cost of cultivation and higher yield (Figure 1).

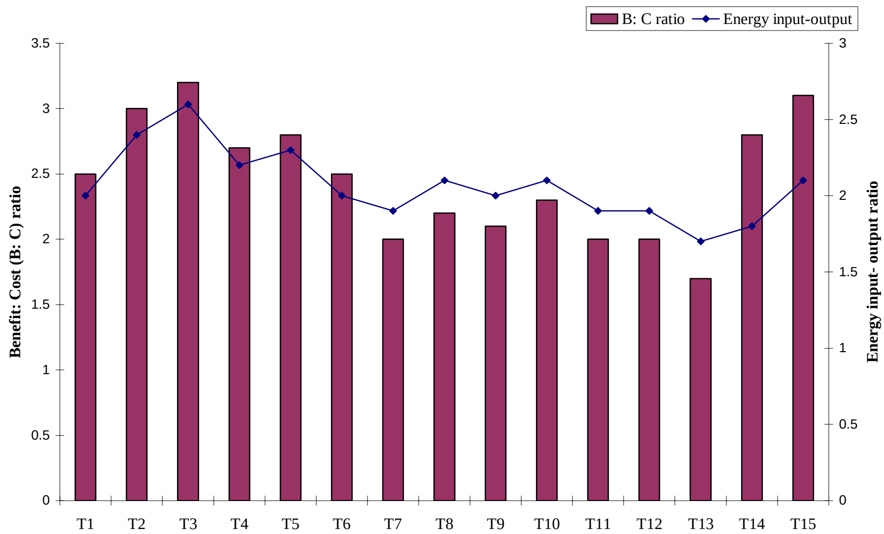

Figure 1. B: C ratio and energy input-output ratio as influenced by weed utilization treatments

The higher energy input-output because of lesser use of synthetic fertilizers as compared to the application of 100% NPK + 10 t FYM ha^{-1} to potato (3.1 and 2.1, respectively). However, the lesser monetary returns and energy input-output were obtained under 100% N as FYM due to significantly lesser tuber yield (1.7 and 1.7, respectively).

Conclusion

The results revealed that allelopathic potential of invaded weeds did not influence the diverse weed spectrum but may be utilized as a nutrient source to maximize the production potential of potato.

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INTEGRATED EFFECTS OF ALLELOPATHY AND SYNTHETIC HERBICIDES ON WHEAT AND ITS' WEEDS IN PESHAWAR

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Abstract: Studies were undertaken during 2005–06 in Agricultural University Peshawar, Pakistan to screen accessions of natural herbicides – aqueous allelopathic extracts for their impact alone and possible synergistic/antagonistic effects in combination with synthetic herbicides on growth of wheat and its weeds. Fresh parts of different plants namely sorghum (*Sorghum bicolor*), sunflower (*Helianthus annuus*), johnson grass (*Sorghum helepense*), neem (*Azadirachta indica*), eucalyptus (*Eucalyptus camaldulensis*) and acacia (*Acacia nilotica*) were dried, chopped, soaked in water in ratio of 2:10 (w/v) and filtered to obtain the suspected allelopathic extracts. Synthetic herbicides included: Buctril M® 40 EC (bromoxynil + MCPA), Puma super® 75 EW (fenoxaprop–p-ethyl) and Affinity® 50 WDG (carfentrazone– ethyl ester). All six plant extracts were solely applied twice as foliar sprays at 30 and 50 days after sowing (DAS) of wheat (*Triticum aestivum*), while the three synthetic herbicides were solely sprayed as per recommended rates once at 30 DAS. In all possible combinations, doses of synthetic herbicides were reduced to half and applied mixed with full dose of plant extracts at 30 DAS. Common weeds of the area included *Avena fatua*, *Convolvulus arvensis*, *Rumex dentatus*, *Phalaris minor*, *Ammi vesnaga*, *Coronopus didymus*, *Poa annua*, and *Fumeria indica*. Weeds were most inhibited in plots treated with Affinity® (6 weeds m^{-2} with 3.07 g dry biomass) and combination of sunflower extract with Affinity® (6.33 weeds m^{-2} with 6.0 g m^{-2} dry biomass). Sunflower extract alone reduced (70 cm) wheat plant height. Plots treated with the combination of sunflower + Affinity® produced the highest (5200 kg ha^{-1}) wheat grain yield. Control plots produced the lowest (4100.33 kg/ha) grain yield. It was concluded that a combination of half dose of Affinity® with sunflower extract was as effective as the full dose of Affinity® in controlling weed growth and enhancing wheat grain yield.

Key words: Allelopathic extracts, herbicides, integration, weed control

Introduction

Presently about two third by volume of the pesticides used in agricultural production are herbicides. The growing environmental and health concerns from the use of these pesticides have stimulated interest in new technologies. The ability of some natural plant compounds to inhibit growth of other plants/organisms has opened new horizons for future research. Improvement in crop productivity and environmental protection through eco-friendly control of weeds, pests, crop diseases, conservation of nitrogen in croplands and synthesis of novel agrochemicals based on these natural products have been the center of attention of scientists engaged in allelopathic research (Chon *et al.* 2003).

Wheat is the basic component of human diet in Pakistan but its average yield has never crossed 30 – 35 % of its optimum yield potential (Sarwar and Nawaz, 1985) and weed infestation has been one of the major causes (Khan *et al.* 2002). The annual losses in wheat due to weeds in Pakistan amount to Rs. 28 billions (Hassan and Marwat, 2001). Weeds are controlled through indiscriminate use of synthetic herbicides (Majid *et al.* 1983; Salarzai *et al.* 1999) which has resulted in serious ecological and environmental problems. Therefore, there is a need for environmentally safe herbicides that are equally or more effective and selective than currently available synthetic herbicides. There is a strong feeling that allelopathic research can be applied to so many current weed problems (Putnam *et al.* 1983; Norwal, 1999). Putnam (1984) reported that eucalyptus species released volatile compounds such as benzoic, cinnamic and phenolic acids, which inhibited growth of crops and weeds growing near it. Thakur and Bhardwaj (1992) reported that leachates from *E. globulus* leaves significantly reduced maize germination but were ineffective on wheat germination. Qasem (1993) assessed allolopathic effects of 54 weed species on wheat under laboratory and glass house conditions. Weeds substantially varied in their effects. *Ranunculus asiaticus* completely prevented wheat seed germination. Roots in general appeared more sensitive to allelopathic effects than shoots. Duhan and Lakshminarayana, (1995) reported that *A. nilotica* tree bark extracts inhibited seed germination and seedling growth of *C. tetragonoloba* and *P. glaucum*. Schumann *et al.* (1995) reported that water extracts of *E. grandis* significantly reduced weed establishment. Cheema *et al.* (1997) reported reduction in weed biomass by 33 – 53 % and increase in wheat yield (7 – 14 %) by application of sorghum (*Sorghum bicolor*) and sunflower (*Helianthus annuus*) water extracts. Anjum and Bajwa (2005) reported that sunflower allelochemicals have potential as possible alternatives for achieving sustainable weed management.

In view of the recognized importance of allelochemicals in biological control of weeds, an experiment was conducted under field conditions with the objectives to screen different natural aqueous allelopathic extracts for their herbicidal potential alone and in combination with synthetic herbicides against weeds of wheat.

Materials and Methods

Field based studies were undertaken during 2005 – 06 at the Research Farm of NWFP Agricultural University in Peshawar, Pakistan to evaluate aqueous natural extracts of different plants namely sorghum (*Sorghum bicolor*), sunflower (*Helianthus annuus)*, johnson grass (*Sorghum helepense*), neem (*Azadirachta indica)*, eucalyptus (*Eucalyptus camaldulensis)* and acacia (*Acacia nilotica)* for their herbicidal potential alone and in combination with synthetic herbicides. For preparing plant extracts fresh plant parts of these species were dried in shade, chopped, soaked in tap water in ratio of 2:10 (w/v) and filtered. Synthetic herbicides included: Buctril® M® 40 EC (bromoxynil + MCPA), Puma super® 75 EW (fenoxaprop–p-ethyl) and Affinity® 50 WDG (carfentrazone–ethyl ester). Wheat (*Triticum aestivum* cv. Saleem 2000) was sown on 6 November, 2005 in Randomized Complete Block Design (RCBD) with three replicates. All six plant extracts were solely applied twice as foliar spray at 30 and 50 days after sowing while all three synthetic herbicides were solely sprayed as per recommended rates once at 30 d after sowing. In all possible combinations, doses of synthetic herbicides were reduced to half and applied mixed with full dose of natural herbicides 30 d after sowing. Common weeds of the area included: *Avena fatua, Convolvulus arvensis, Rumex dentatus, Phalaris minor, Ammi vesnaga, Coronopus didymus, Poa annua* and *Fumeria indica.* Data on weed density (m^{-2}), dry weed biomass (gm m^{-2}), wheat plant height (cm) and wheat grain yield (kg ha^{-1}) were recorded sixty days after sowing. The entire data were individually subjected to analysis of variance and the means were compared by least significant difference (LSD) test using MSTAT software.

Results and Discussion

The impacts of different plant extracts and synthetic herbicides as well as their combinations on the growth of wheat (*Triticum aestivum)* and its weeds are shown in Table 1. Analysis of data revealed that weed density m^{-2}, dry weed biomass (g m^{-2}), wheat plant height (cm) and wheat grain yield (kg ha^{-1}) were significantly ($p<0.01$) affected by all treatments used.

Weed density m^{-2}

Combination of eucalyptus (*Eucalyptus camaldulensis)* extract and Puma super® (fenoxaprop–p-ethyl) enhanced weed germination which resulted in maximum (45.33) weed density m^{-2} indicating a stimulatory effect at low concentration (2:10 w/v) of the extract (Table 1). Lovett (1989) also reported that biological activities of receiver plants to allelochemicals are known to be concentration-dependent with a response threshold. Responses are, characteristically, stimulation at low concentrations of allelochemicals and inhibition as the concentration increases. Identical results were also reported by Anjum and Bajwa (2005) and Nasim *et al.* (2005).

Table 1. Effect of different natural and synthetic herbicides and their combinations on growth parameters of wheat and its weeds

Treatments	Growth parameters of wheat and its weeds			
	Weeds density m^{-2}	Dry weed biomass (g m^{-2})	Wheat plant height (cm)	Wheat grain yield (kg ha^{-1})
Sorghum	27.33 H	10.00 H	85.67 AB	4600 G
Sunflower	30.33 FG	11.20 F	70.00 O	4500 I
John grass	32.33 EF	13.03 D	79.00 HIJ	4400 K
Neem	35.33 CD	16.00 C	73.67 LMN	4450 J
Eucalyptus	35.00 CD	18.03 B	75.33 KLM	4430 J
Acacia	37.00 C	18.07 B	84.67 A-D	4540 H
Buctril® M	14.33 LM	06.93 K	75.33 KLM	4750 E
Puma super	16.00 JKL	08.07 J	73.00 MN	4650 F
Affinity	06.00 P	03.07 M	85.33 ABC	4900 D
Sorghum+buctril M	15.33 KLM	08.04 J	76.00 KL	4750 E
Sorghum+puma super	18.00 J	11.00 FG	86.00 A	4895 D
Sorghum+affinity	07.00 OP	09.03 I	73.00 MN	4960 C
Sunflower+buctril M	16.00 JKL	07.03 K	79.00 HIJ	5000 B
Sunflower+puma super	17.00 JK	08.00 J	82.00 D-G	4900 D
Sunflower+affinity	06.33 P	06.00 L	83.00 B-E	5200 A
Johnson grass+buctril M	16.33 JKL	08.00 J	79.33 G-J	4250 MN
Johnson grass+puma super	18.00 J	12.00 E	80.00 F-I	4190 O
Johnson grass+affinity	08.00 NOP	07.00 K	82.67 C-F	4900 D
Neem+buctril M	23.33 I	11.17 F	77.00 JK	4260 M
Neem+puma super	33.33 DE	13.20 D	77.67 IJK	4150 P
Neem+affinity	10.00 N	09.10 I	83.67 A-E	4320 L
Eucalyptus+buctril M	28.00 H	13.17 D	81.67 E-H	4200 O
Eucalyptus+puma super	45.33 A	16.10 C	71.67 NO	4320 L
Eucalyptus+affinity	13.33 M	10.37 GH	77.00 JK	4480 I
Acacia+buctril M	22.33 I	10.13 H	76.00 KL	4230 N
Acacia+puma super	29.00 GH	11.07 FG	83.33 A-E	4730 E
Acacia+affinity	09.00 NO	09.00 I	80.00 F-I	5180 A
Contol	40.00 B	26.00 A	76.00 KL	4100.33 Q

Within a column, means followed by the same letter are not significantly different by the Least Significant Difference test (p=0.05).

Affinity® (carfentrazone–ethyl ester) was the most toxic to weeds and reduced their density (6 weeds m^{-2}) due to its broad-spectrum nature. Combination of sunflower (*Helianthus annuus)* extract and affinity also reduced weed density to 6.33 m^{-2} which was statistically similar to weed density as obtained with the full dose of affinity indicating a synergistic effect of both natural and synthetic herbicides in controlling weeds. Weed density control ranking of top five herbicides and their combinations was Affinity® = sunflower+Affinity® > sorghum

(*Sorghum bicolor*)+Affinity® > johnson grass (*Sorghum helepense*)+Affinity® > acacia (*Acacia nilotica*)+Affinity® (Figure 1). An *et al.* (1996) and Alsaadawi (1992) reported inhibition of seed germination by sunflower extract. Roots of weeds which succeeded to germinate became brown, stunted, void of root hair and ultimately dried. Qasem (1993) noted a rapid inhibition of seedlings' roots tips respiration when exposed to allelochemicals.

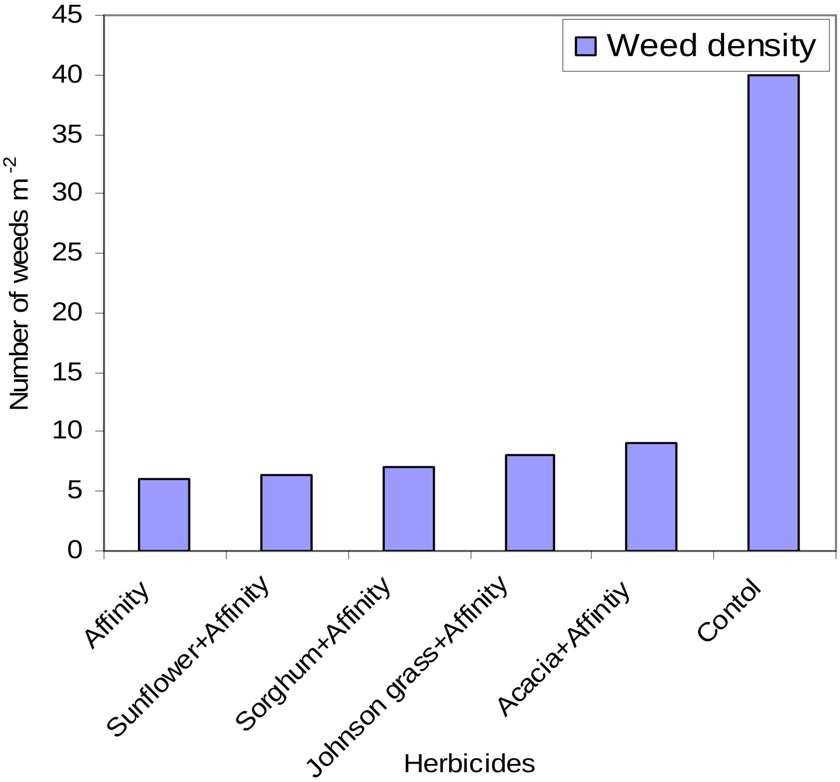

Figure 1. Weeds density control ranking of top five treatments.

Dry weed biomass
Table 1 revealed that dry weed biomass was the most inhibited by Affinity (3.07 g m^{-2}) closely followed by the combination of sunflower extract and affinity which resulted in 6 g m^{-2} dry weed biomass. The maximum (26 g m^{-2}) dry weed biomass was recorded for control plots. Sole affinity retarded above ground growth while combining Affinity® with sunflower extract did more than that. Reduced radicle and coleoptile extension, swelling or necrosis of root tips, curling of root axis and lack of root hairs ultimately resulted in reduced uptake of nutrients and hence dry weight accumulation. These gross morphological effects may be secondary manifestations of primary events caused by a variety of more specific effects acting at the cellular or molecular level in the receiver plants. Similar results were reported by An *et al.* (1996) and Alsaadawi (1992).

Wheat plant height (cm)
Table 1 indicates that maximum (86 cm) wheat plant height was observed in plots treated with the combination of sorghum (*Sorghum bicolor*) extract and puma super which again revealed a synergestic effect of both plant extracts and synthetic herbicides. The minimum plant height of 70 cm was recorded in plots treated the sole sunflower extract thereby indicating a toxic effect of natural herbicide on plant elongation. Roots of some plants treated with sorghum extract became brown, stunted and void of root hair which ultimately restricted nutrient flow to the meristematic apical bud and hence reduced plant height. Qasem (1993)

noted a rapid inhibition of seedlings' roots tip respiration when exposed to allelochemicals and thereby yielded similar effects.

Wheat grain yield

Data in Table 1 revealed that a maximum of 5200 kg ha^{-1} and 5180 kg ha^{-1} wheat grain was recorded for sunflower + Affinity® and acacia (*Acacia nilotica)* + Affinity® combinations respectively which revealed a synergestic effect of both natural and synthetic herbicides. The minimum (4100 kg ha^{-1}) wheat grain yield was recorded in control plots. Wheat was comparatively more tolerant to the natural herbicides than weeds and hence its yield was enhanced. Ranking of the top five herbicides and their combinations based on wheat grain yield increase was sunflower+Affinity® = acacia+Affinity® > sunflower+Buctril M® > sorghum+Affinity® > Affinity® (Figure 2). These results are in line with those of Shahid *et al.* (2006) and Khan *et al.* (2004).

On the basis of these results, it was concluded that a combination of sunflower with half of the recommended dose of affinity exhibited almost similar weeds control and far more wheat grain yield increase as those obtained with the full recommended dose of affinity and hence is recommended for better economic returns and environmental protection.

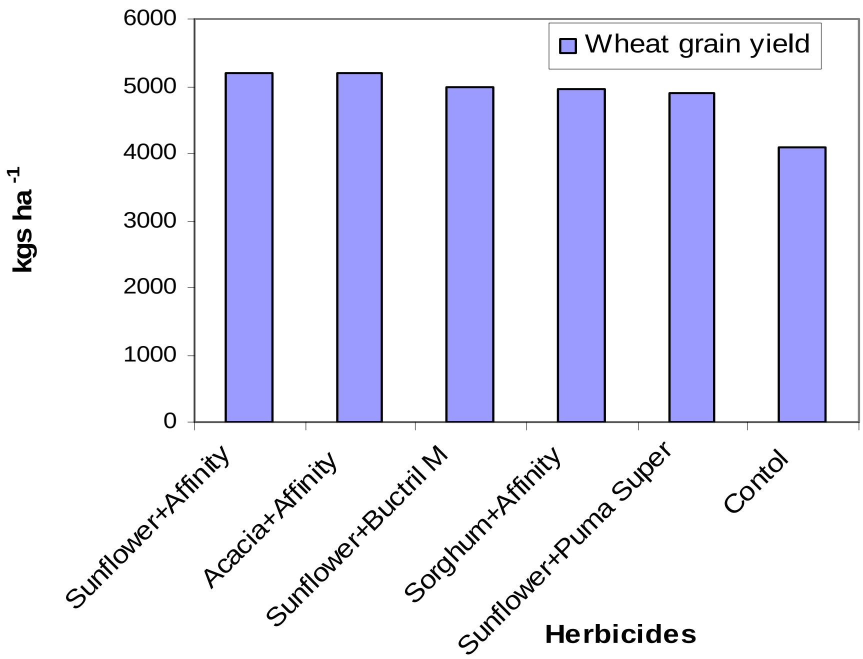

Figure 2. Ranking of top five treatments based on grain yield increase of wheat

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ALLELOPATHIC EFFECT OF ANTHERS OF A NEWLY-BRED RICE VARIETY 'K21' ON GROWTH OF BARNYARD GRASS AND LETTUCE

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Abstract: This study was conducted to investigate allelopathic potential of anthers of 3 rice cultivars Dongjinbyeo (female parent), Kouketsumochi (male parent), and K21 (hybrid), using their inhibitory effects on barnyardgrass and lettuce, and to determine their phenolic compounds by HPLC. Based on the results of bioassay and HPLC analysis in three rice varieties, the variety which had higher amount of phenolic compounds showed higher inhibitory effects on test plants. The variety Kouketsumochi showed the highest inhibitory effects and also contained the highest amounts of phenolic compounds, followed by K21 and Dongjinbyeo.

Key words: Allelopathy potential; phenolic compound; K21.

Introduction

The allelopathic potential of rice has received a great attention in the recent past since Dilday *et al.* (1991) identified rice cultivars exhibiting allelopathic potential against ducksalad [*Heteranthera limosa* (Sw.) Willd]. In addition, allelopathic potential has also been reported from numerous crops such as barley, cucumber, oats, rice, sorghum, sunflower, tobacco and wheat. The incorporation of allelopathic traits into cultivating rice varieties, which could reduce the rate of applying herbicides for weed control is worth exploring (Khush, 1996).

Ma (2002) developed an allelopathic rice cultivar K21, with high yield and quality by crossing between Donginbyeo (a non-allelopathic, but a high yielding variety with good agronomic traits) and Kouketsumochi (a potent allelopathic cultivar, but tall and susceptible to lodging). This study was conducted to investigate existence of allelochemicals in the anthers of K21 and what kinds of the allelochemicals are in the anthers, and finally allelopathic potential of the anthers of rice.

Materials and Methods

Effect of methanol extract of rice anthers
The anthers of rice varieties K21 (hybrid), Donginbyeo (female parent) and Kouketsumochi (male parent) were collected from the flowered rice plants. Methanol extraction was done for 24 hrs at 26±2°C. The extracts were filtered through Whatman No. 1 paper and diluted to the concentrations of 1.0, 2.5, and 5.0% for bioassay using lettuce and barnyardgrass. The shoot and root lengths of the test plants were measured 7 days after treatment.

Identification of phenolic compounds in K21
Two grams of anthers of the three rice cultivars were separately homogenized using a mortar and pestle with liquid nitrogen. Extraction was made with 40 ml of 100% HPLC grade methanol with shaking for 24 hrs. Hexane and ethyl acetate fraction were made and then the collected ethyl acetate fraction was dried by evaporation at 40°C under vacuum condition. The dried ethyl acetate phase was dissolved with 1 ml of methanol. Ten µl of the extracted sample was injected into a Phenomenex LUNA C18 column (4.6 mm x 250 mm) fitted with a guard column. The UV-absorbing compounds eluted from the column were monitored at 280 nm.

Results and Discussion

Kouketsumochi, a potent allelopathic rice showed the greatest inhibitory effect on test plants, followed by K21 and Dongjinbyeo. The inhibitory effect was increased as the concentration of extracts increased. The per cent inhibition observed in the root growth of barnyardgrass treated with 2.5% extracts from Kouketsumochi, K21, and Dongjinbyeo were 89.5, 83.4, and 47.4%, respectively (Table 1).

Table 1. Inhibitory effect of rice pollen (including anther) extracts on the growth of lettuce and barnyardgrass.

Test plant	Cultivar	Shoot length[1)]			Root length		
		1.0%	2.5%	5.0%	1.0%	2.5%	5.0%
		% inhibition[2]					
Letuce	Kouketsumochi	13.8 a[3)]	42.5 a	63.2 a	18.0 a	83.2 a	93.9 a
	K21	10.1 b	31.8 a	60.1 a	14.2 a	65.5 a	88.1 a
	Dongjinbyeo	4.1 b	14.8 a	55.0 a	8.6 a	29.6 b	86.9 a
Barnyardgrass	Kouketsumochi	15.7 b	66.7 a	85.6 a	24.3 a	89.5 a	100 a
	K21	36.3 a	52.1 b	79.4 a	22.3 b	83.4 a	100 a
	Dongjinbyeo	2.9 b	33.7 b	80.3 a	14.3 b	47.4 b	100 a

[1)]in the case of lettuce, hypocotyl length, [2]Average of 3 replicates. [3]Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05).

The HPLC analysis of phenolic compounds showed that all three varieties had similar peak patterns, indicating the presence similar types of phenolic compounds. The hybrid K21, showed a diverse kinds of phenolic compounds. However, Kouketsumochi contained the highest amount of phenolic compounds. The total amounts of phenolic compounds in Kouketsumochi, K21, and Dongjinbyeo were 1.7276, 0.9637, and 0.4184 mg/g F.W., respectively (Table 2).

Table 2. Phenolic compounds in three varieties of rice pollen (including anther) identified by HPLC analysis.

Phenolic compounds	RT* (min)	Cultivar: Dongjinbyeo	K21	Kouketsumochi
		mg/g FW		
Tannic acid	11.67	0.0513	0.2218	0.0841
ρ-Aminobenzoic acid	16.24	ND**	0.0042	ND
Catechol	18.87	ND	0.0273	ND
ρ-Hydroxybenzoic acid	19.27	0.1584	0.2172	1.0251
ρ-Hydroxyphenyl	19.88	0.0171	0.0294	0.4133
Caffeic acid	21.16	0.0073	0.0098	0.0161
ρ-Coumaric acid	26.80	0.0185	0.0436	0.0355
Ferulic acid	28.96	0.0084	0.0450	0.0099
trans-Cinnamic acid	42.49	0.0066	0.0107	0.0084
Benzophenone	54.91	0.1508	0.3547	0.1351
Total phenol compounds		0.4184	0.9637	1.7276

*RT - Retention time, **ND - not detected

The results of the bioassay and HPLC analysis in three rice varieties indicated that the variety that has the higher amount of phenolic compounds may have a higher inhibitory effect on the test plants. The variety Kouketsumochi showed the highest inhibitory effects and contained the highest amounts of phenolic compounds, followed by K21 and Dongjinbyeo.

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CROP PERFORMANCE AND INCIDENCE OF WEED SPECIES WITH THE INTRODUCTION OF DIRECT SEEDED RICE IN THE RICE-WHEAT CROPPING SYSTEM OF NORTHERN INDIA

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Abstract: Sustainability of the rice-wheat cropping system of the Indo-Gangetic Plains is essential to regional food security. Changing rice establishment from transplanting to direct seeding offers the potential to reduce costs through labour and water savings in addition to improving productivity. Studies over five cropping cycles have confirmed that despite seasonal variation rice yields under both wet seeding and dry can be equivalent to transplanted rice, with intensive weed control. In contrast, conservation (zero) tillage in drill seeded rice production, gave lower yields. The inclusion of appropriate early post emergence chemical weed control followed by manual weeding for rice improved yields over conventional hand weeding. In wheat, conservation tillage gave lower (9%) yields compared to conventional tillage practices. Continuous direct seeding resulted in succession in the weed flora. In the absence of control incidence of *Cyperus rotundus* was low under wet seeding but high in all dry seeding methods. Conversely, wet seeding selected strongly for *Ischaemum rugosum* and *Leptochloa chinensis* whereas *Echinochloa colona* was common under all direct seeding practices. These rapidly emerging shifts in the weed flora emphasize the need for on-farm systems of weed monitoring and response in terms of weed control method.

Key words: Weeds, rice, direct seeding, crop establishment

Introduction

Rice and wheat are the staple food crops of India, contributing to nearly 80% of the total food-grain production (Timsina and Connor, 2001). A total of 13.5 million ha of land is under the rice wheat rotation in south Asia, of which 10 million ha is in India and largely in the Indo-Gangetic Plains (IGP) in the north of the country (Ladha *et al*., 2000). Demand for food grains in India is expected to grow and over the next three decades an estimated 30-50% production increase is required to meet domestic demands. There has, however, been stagnation in rice productivity in recent years, a declining rice yield trend has been observed (Padre and Ladha, 2004), and reports of labour and water shortage for agriculture in areas with high production potential. Pandey and Velasco (2002) argued that low wage rates and adequate water supply favor transplanting but direct seeding is likely to increase in circumstances of labor scarcity and increasing wage rates. Singh *et al*. (2001a; 2003; 2005) have reported the potential for direct seeding of rice as a replacement for transplanting and described early findings of a long term system trial designed to examine the impact of changes in rice and wheat crop establishment methods on crop yield and on the weed flora in rice. Responses in rice weed community structure to changes in the rice-wheat rotation have been recorded (Singh *et al*. 2001b) and reduced tillage agriculture is well known to have an impact on weed community composition (Derksen *et al*. 1993). This paper assesses the implications of these changes to sustainable direct seeding.

Materials and Methods

In 2000, a long term replicated field experiment was established at the field station of G.B. Pant University of Agriculture and Technology, Uttaranchal, India. Five rice establishment methods were included in factorial combination with two wheat establishment methods (Table 1) and three weeding levels. Further experimental details are given in Singh *et al*. (2005). The experiment was continued for five cropping seasons and selected results are reported for 2004 and 2005 rice seasons.

Table 1. Experimental details (DAS - days after seeding; DAT - days after transplanting) (From Singh *et al.* 2005).

Crop	Establishment method	Land preparation and sowing	Herbicide used in intensive weeding
Rice	Conventionally transplanted – TP	Ploughed, puddled and levelled; hand transplanted 21-day-old plants at 20 cm × 20 cm spacing.	Butachlor 1.5 kg a.i. ha^{-1}, 2 DAT.
	Wet seeded (pre-germinated) - WS	Ploughed 12-15 cm, puddled and levelled; sown with drum seeder at 35 kg ha^{-1}, 20 cm row spacing.	Anilophos 0.4 kg a.i. ha^{-1}, 7 DAS, 2-3 leaf stage of rice.
	Dry drill seeded – DS	Ploughed, harrowed; drill seeded, 50 kg ha^{-1}, 20 cm row spacing.	Pendimethalin 1.0 kg a.i. ha^{-1}, 1 DAS.
	Dry drill seeded with stale seed bed treatment – DSf	As DS, with a flush irrigation and subsequent 7 d glyphosate application prior to seeding; 50 kg ha^{-1}, 20 cm row spacing.	Pendimethalin 1.0 kg a.i. ha^{-1}, 1 DAS.
	Dry drill seeded with zero tillage – ZT	Flush irrigation and subsequent 7 d glyphosate application prior to drilling; 50 kg ha^{-1}, 20 cm row spacing.	Pendimethalin 1.0 kg a.i. ha^{-1}, 1 DAS.
Wheat	Conventional tillage	Ploughed, harrowed; drill seeded, 100 kg ha^{-1}, 20 cm row spacing.	Isoproturon 1 kg a.i. ha^{-1} + metsulfuron methyl 4 g a.i. ha^{-1} , 35 DAS.
	Zero tillage	Drill seeded into rice stubbles after paraquat, 0.5 kg a.i. ha^{-1}, 100 kg ha^{-1}, 20 cm row spacing.	Isoproturon 1 kg a.i. ha^{-1} + metsulfuron methyl 4 g a.i. ha^{-1} , 35 DAS.

Rice for transplanting was sown in a nursery at the same time as plots were direct seeded, and transplanted 28 days (d) later. Rice plots were sub-divided and either - intensively weeded (herbicide followed by manual weeding) - "*clean weed*", - manually weeded at 28 d (and 56 d if required) after crop establishment - "*hand weed*", or un-weeded - "*control*". In the wheat phase of the rotation, weeds were managed in all plots with post-emergence herbicide and hand weeding. In each rice plot, weed abundance (weed density and biomass, by species) was measured in two 0.25 m x 1 m quadrates covering 5 crop rows at 28 d, 56 d after crop establishment, and prior to manual weeding, and at harvest. In transplanted rice, after puddling, a water depth of 100 mm was maintained for one month following planting. In wet seeded plots, after puddling the soil was kept saturated for 21 DAS to ensure rice establishment. Dry seeding was into aerobic soil. In all plots supplemental irrigation was given to all plots as required after 28 DAS/DAT and plots were drained where excessive monsoon rain caused water depths in excess of 100 mm. Rice and wheat yields were taken from 5 m^2.

Results and Discussion

Yields

Mean grain yield of rice in 2004 was marginally higher than in 2005 (4.78 v. 4.42 t ha^{-1}; SED = 0.09). Wet seeding gave the highest yield in weed free plots in 2004 (7.1 t ha^{-1}), whilst dry seeded rice (6.46 t ha^{-1}) gave the highest in 2005. The lowest yields in weed free plots were from zero-tillage (5.46 t ha^{-1}). Transplanted rice gave an average yield of 6.6 t ha^{-1} across the two years (Figure 1).

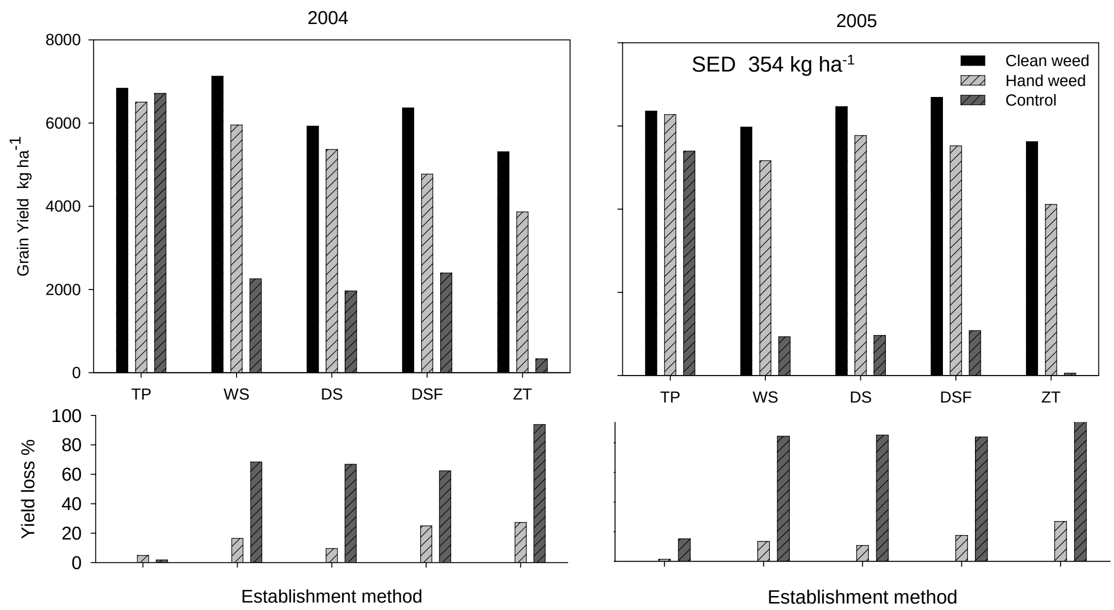

Figure 1. Rice grain yield under different methods of establishment and weed management, and % yield losses in the hand weed and control plots as compared to clean weeded plots, 2004 and 2005.

Losses of rice grain yield due to weeds were slightly higher in 2005 than the previous year though there were similar patterns in both years. Across 2004 and 2005, compared to the clean weeded plots, competition from weeds in un-weeded plots was 8% under transplanting plots, 76% in WS, 75% in DS/DSF and 96% under ZT. Hand weeding significantly reduced yield losses which were least in TP (3%) but under the direct seeding options ranged from 10% with DS to 27% with ZT. There were no significant effects of the different wheat establishment practices on the grain yield of the subsequent rice crop.

Effects of tillage practices for the rice on the subsequent wheat crop were not consistent across the two years. Across the two years however, wheat yields were significantly ($p = 0.034$) lower after zero-tillage for wheat as compared to conventional tillage (3555 vs. 3933 kg ha^{-1}). This finding is not consistent with those of Tripathi *et al.* (1999) who recorded comparable yields of wheat from zero- and conventional tillage irrespective of different rice seeding/transplanting methods.

Weed species incidence

At the start of the trial in 2000 *Echinochloa colona, Caesulia axillaris*, and *Fimbrystylis miliacea* were the most abundant weeds in both the transplanted and dry-seeded rice plots, when un-weeded plots (Singh *et al*. 2001a). Fourteen species were commonly recorded over the five cropping seasons: *Caesulia axillaris, Commelina diffusa, Cynotis* spp, *Cyperus difformis, C. iria, C. rotundus, E. colona, E. crus-galli, Eragrostis japonica, F. miliacea, Ischaemum rugosum, Leptochloa chinensis* and *Paspalum distichum* (Singh *et al*. 2005). After four cropping cycles in 2004, *I. rugosum*, *E. crus-galli*, *L. chinensis*, *E. colona* and *C. rotundus* were recorded in all direct seeded plots although the establishment methods had selectively influenced the relative likelihood of their presence. Probabilities of occurrence of these weed species are given in Figure 2 in control plots at 28 and 56 DAS and at 56 DAS after hand weeding. *E. colona* was a species common to all establishment methods although it was most prevalent in ZT plots. Hand weeding at 28 DAS reduced its likelihood of occurrence in all plots at 56 DAS except DS, indicating a prolonged emergence period in soil which in the surface layers is partially aerobic. Reduced presence in plots given an early flush irrigation suggests that seed dormancy is easily broken and there is a transient seed bank in the surface layers of the soil. ZT favoured the growth of *I. rugosum* and *E. crus-galli* and, particularly in the case of *E. crus-galli,* the incidence of these species tended to increase between 28 to 56 days. A prolonged emergence period for *E. crus-galli* is reflected in its relatively high incidence in the WS and ZT plots despite hand weeding at 28 DAS. Only in WS plots was there was a high probability of *L. chinenis* occurring, with incidence increasing from 28-56 DAS, while in the control plots of DS and DSF by 56 DAS it was absent. The abundance of *C. rotundus* was promoted under DS, DSF and ZT, and there was relatively low incidence in puddle plots (WS). Incidence of this species declined between 28-56 DAS in the control plots. A decline in the incidence of this species after 28 DAS may have been due to competition from other species, changes in the moisture regime, completion of life cycles or a combination of these.

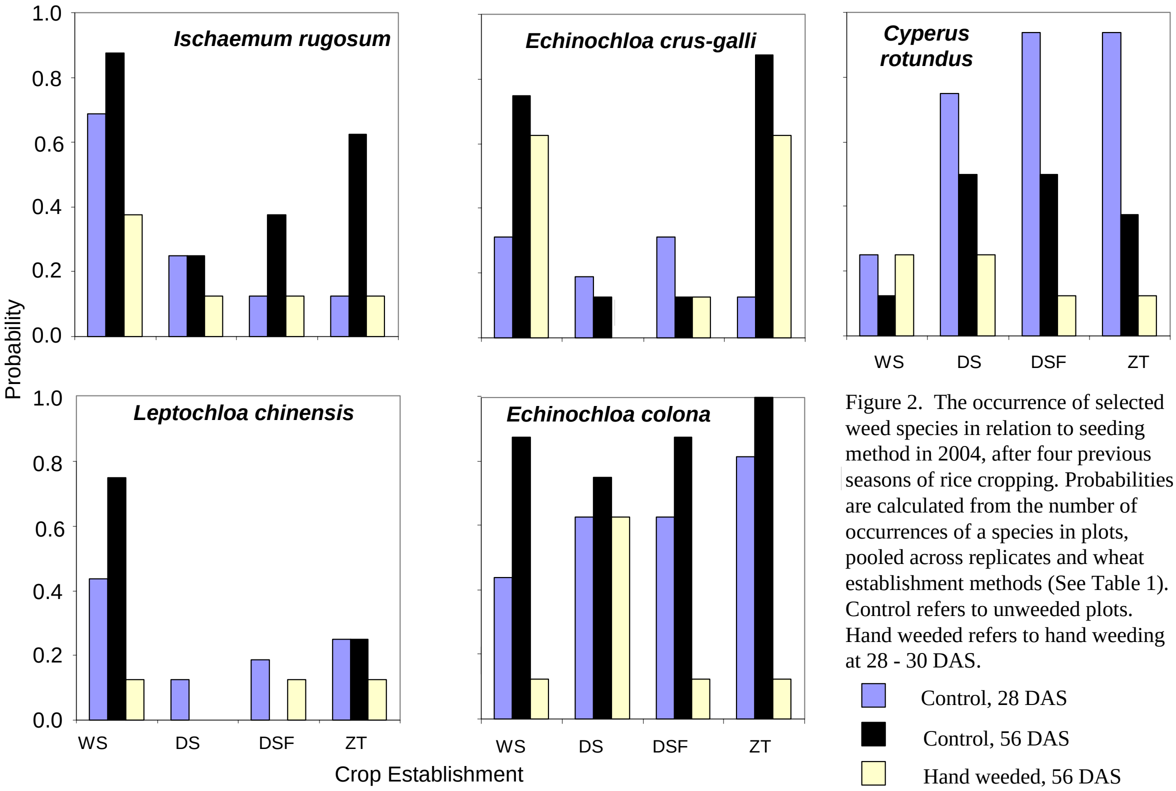

Figure 2. The occurrence of selected weed species in relation to seeding method in 2004, after four previous seasons of rice cropping. Probabilities are calculated from the number of occurrences of a species in plots, pooled across replicates and wheat establishment methods (See Table 1). Control refers to unweeded plots. Hand weeded refers to hand weeding at 28 - 30 DAS.

The selective effects of land preparation and crop establishment on recruitment of rice weeds and by implication soil seed banks are well known (Mortimer and Hill, 1999) and these results highlight the responsiveness of major rice weeds. As a single observation in time, they suggest that underlying weed succession may result from cultural practices. Further analysis (not shown) of population dynamics support the hypothesis of succession for *C. rotundus* since increases in density were seen over years at comparable census points although spatial heterogeneity over plots precluded statistical significance. Whilst substantially reducing weed biomass and protecting yield, hand weeding alone did little to control the principal weed species in most instances. Populations present after hand weeding in rice would contribute to the seed rain of that season and in some instances may result in substantial crop losses due to competition.

Understanding the underlying dynamics of weed communities underpins decision support systems that identify important target weeds and potential shifts in populations, and determines appropriate control measures. Successful yield protection from weed competition in direct seeded rice relies on effective early chemical control followed by manual weeding, the latter to remove aggressive late emerging species not adequately suppressed by the developing rice canopy.

These studies indicate that *E. colona* is likely to be a threat to rice yields in the direct seeding systems and, unless suppressed by flooding after crop establishment the weed is likely to require either post-emergent herbicide application or repeated hand weeding in order to control it. A change to dry, direct seeding with either conventional or zero-tillage for improving productivity has the potential to reduce time and costs but may increase the risk of ingress of *C. rotundus*. The limited knowledge of factors influencing the germination dynamics, seed longevity and dormancy for many of the key weed species is a limitation to prediction of the likely impacts of crop management on weed populations.

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EFFECT OF PHYSICOCHEMICAL PROPERTIES OF TWO SURFACTANT SERIES ON THE DISTRIBUTION OF ^{14}C GLYPHOSATE

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Abstract: A study was conducted to examine the effects of surfactants from tallow amine ethoxylate and nonylphenols ethoxylate series with average 2 to 15 moles of ethylene oxide (EO) on the distribution of ^{14}C-glyphosate and phytotoxicity of glyphosate on two weed species – sicklepod (*Cassia obtusifolia* L.), which contains 93% polar waxes and teaweed (*Sida spinosa* L.) containing 14% polar waxes. Presence of surfactant may improve physicochemical properties of glyphosate. The surface tension (ST) and contact angle (CA) values of surfactant with and without glyphosate showed a significant increase as the EO number increased in case of both tallow amine ethoxylate and nonylphenols ethoxylate surfactant series. Test weed plants were treated with ^{14}C-glyphosate alone and formulated with different EO in tallow amine ethoxylate and nonylphenols ethoxylate surfactant series to determine the pattern of ^{14}C glyphosate distribution. Surface tension, contact angle and ^{14}C-distribution were significantly affected by both the presence of different waxes on plants and addition of adjuvants in glyphosate. Higher absorption was recorded in *S. spinosa* than *C. obtusifolia* when glyphosate was formulated with tallow amine ethoxylate, ^{14}C absorption increased with the increase in EO number which is in contrary to the values of ST and CA. However, in case of nonylphenols ethoxylate surfactant series, the increase in absorption was recorded with the decrease in ST and CA values. Study indicated that there was a linear relationship between the physical properties, absorption and efficacy of glyphosate with the addition of nonylphenols ethoxylate surfactant series. The per cent control was higher with higher EO in tallow amine ethoxylate series and with lower EO in nonylphenols ethoxylate surfactant series.

Keywords: Surfactants, tallow amine ethoxylate and nonylphenols ethoxylate series, physicochemical properties

Introduction

The epicuticular waxy layer on plant surfaces is the main barrier for foliar-applied herbicides to reach the target site (Kirkwood, 1991; Sharma and Singh, 1999). Plants grown under dry conditions have smaller leaves, thicker cuticles with more trichomes, and more compact leaf structure that reduces spray interception, retention, and absorption, affect the performance of surfactant on herbicide (Van Valkenburg, 1982, Ryerse *et al*. 2004). Additional factors like environment, application and chemical/ formulation have been shown to influence the role of adjuvants in herbicide efficacy (Elmore *et al.* 1998). Surfactant improved the bioefficacy and rainfastness of glyphosate (De Ruiter *et al.* 1996). Nalewaja and Matysiak (1995) reported that a surfactant having HLB value of about 15 was optimum for enhancing glyphosate activity applied to wheat. Efficacy of applied herbicide has been correlated with HLB of added surfactant (Holloway and Stock, 1990), which is a function of EO content. Efficacy is generally maximized with surfactants having HLB of 12 to 15 (Matsui *et al*. 1992). Surfactants improve spray solution characteristics (*e.g.* surface tension, solubility) and add to enhanced wetting of the plant surface, better retention, and higher rates of absorption. However, the beneficial effect of surfactants on spray performance greatly depends on surfactant chemistry (Silcox and Holloway, 1989).

Elmore *et al.* (1998) reported that there are various factors influencing the efficacy, retention and penetration of herbicide to the plant foliage *e.g.* adjuvant type, plant species, age of the plants, epicuticular wax and environmental conditions etc. Addition of different type of adjuvants affects the efficacy, absorption and translocation of glyphosate, which is a complex scenario (Liu, 2004). The improved wettability and absorption mainly results from complex interactions among the pesticide, surfactant, carrier water, and the plant surface (Hull *et al.* 1982; Sharma *et al.* 1996). Tank mixing of glyphosate with surfactants, reduced the values of ST and CA to a great extent (Sharma *et al.* 1996; Sun and Singh, 1998). It is important to use surfactant around the cuticle micelle concentration (CMC) with herbicides in order to obtain the maximum effectiveness of herbicidal sprays (Sharma *et al.* 1989). The beneficial effect of surfactant concentrations beyond the CMC has been reported by McWhorter and Barrentine (1988). In the study the main object was to understand the use of different type of surfactant - tallow amine ethoxylate and nonionic alkoxylate series, varying in their physicochemical characteristics with average POE of 2 to 15, with glyphosate. The effect of these surfactants on the absorption of ^{14}C glyphosate and phytotoxicity of glyphosate on two weed species – sicklepod which contains 93% polar waxes and prickly sida 14% polar waxes (Harr *et al.* 1991) was determined.

Materials and Methods

Seeds of prickly sida and sicklepod plants were sown in plastic pots of 15 cm x 10 cm size containing commercial potting medium (Metro-Mix 500, Scotts-Sierra Horticultural Products Co., Marysville, OH) in a greenhouse maintained at 25/16ºC (± 0.5 ºC) day/night temperatures, 70% (± 5%) relative humidity. For ^{14}C distribution study, the plants were grown in 6.5x5.5cm plastic pots. The plants were watered and fertilized with Tracite foliar fertilizer (Helena Chemical Co., Collierville, TN) containing 20-20-20 (N-P-K) once 3 weeks after planting to promote optimum growth. Weed seedlings were thinned to four seedlings for efficacy and one seedling for ^{14}C absorption studies, respectively. Plants were at four leaf stage at application.

^{14}C Glyphosate absorption studies.
Glyphosate was applied at 0.56 kg/ha ± surfactant for ^{14}C glyphosate absorption studies. Plants were transferred to a growth chamber a week before receiving the treatments, which maintained an average photosynthetic photon flux density (PPFD) of 200 $\mu E/m^2/S$ at the plant level; day/night temperatures of 25/16ºC (± 0.5ºC) and the relative humidity of 55/70% (± 5%), respectively. Each plastic pot containing single seedling represented one replication and there were three replications for each treatment. The experiment was repeated once. The third fully expanded leaf at 4- leaf stage of each plant was carefully covered by aluminum foil, then the plants were sprayed with glyphosate at 0.56 kg/ha ± surfactant using a pressurized air chamber track sprayer (Allan Machine Works, Midland, MI) delivering 189 l/ha at 138 *k*Pa. After removing the aluminum foil, five x 2 µl droplets of ^{14}C glyphosate treatment were applied to the adaxial leaf surface in the median part of the third leaf. The quantity of ^{14}C glyphosate applied on to the leaf was determined by dispensing a same number of droplets directly into 7 ml vials containing scintillation liquid (ScintiVerse, R. J. Harvey Inc., Hillsdale, NJ). At 72 hrs after treatment, treated plants were dissected into two sections (treated leaf and rest of the plant tissue). To recover all non-absorbed ^{14}C-glyphosate, the treated leaf was washed 2 x 4 ml of water:ethanol (1:1 v/v) and then rinsed 2x3 ml of ethanol. A 200 µl sub-sample from each washing was dispensed into a polyvinyl vial containing 7 ml of scintillation liquid and then quantified using a liquid scintillation counter (LSC) (Packard Instrument Inc., Meriden, CT). Plant samples were oven dried at 50ºC for 48 hrs and were combusted utilizing a biological oxidizer (R. J. Harvey Inc., Hillsdale, NJ). The $^{14}CO_2$ in oxidized samples was quantified by LSC to determine absorption of ^{14}C glyphosate. Foliar absorption was defined as ^{14}C in plant tissues after washing the treated leaf, and translocation

as the amounts of total ^{14}C, which traveled out of treated leaf into rest of the plant tissues. Both values were expressed as percent relative to applied dose. The recoveries were calculated by adding radioactivity in all the assayed fractions which were 95 to 97% excluding background activity. The experiment was conducted as a complete randomized design with a factorial arrangement of treatments with two factors – weed species and herbicide treatments.

Table 1: Physicochemical properties of surfactant used.

Surfactant	POE	HLB	Solubility
Series A*			
Toximul (TA 2)	2	5	methanol, kerosene, xylene
Toximul (TA 5)	5	9	methanol, kerosene, xylene, water
Toximul (TA 15)	15	14	xylene, methanol, water
Series B*			
Makon (M8)	8	12	xylene, methanol, water
Makon (M10)	10	13	xylene, methanol, water
Makon (M12)	12	14	xylene, methanol, water

*Series A = Tallow Amine Ethoxylate surfactant series (TA) and Series B = Nonionic alkoxylate surfactant series (M), and associated number with TA and M is ethlyane oxide number.

Efficacy studies

Glyphosate at a rate of 0.56 and 1.12 kg a.i/ha was used as Rodeo formulation (isopropylamine salt) (Dow AgroSciences, Indianpolis, IN), which do not contain any surfactant. Fresh aqueous solutions of tallow amine ethoxylate and nonionic alkoxylate surfactant series (Stepan Co., Northfield, IL) at 0.25% v/v (Table 1) were added with glyphosate at 0.56 kg/ha. Aqueous solutions of glyphosate (± surfactant) were prepared and applied to plant species using air pressure chamber track sprayer as described earlier. Plant injury symptoms were visually rated until 4 wks after treatment (WAT) and ratings of 4 WAT were used in the statistical analysis. A scale of 0 to 100 was used; 0 – indicating no damage and 100 – indicating complete plant death as described by Frans *et al.* (1986). The experiment was designed as complete randomized block with four replications and was repeated once. Untreated control plants were used as reference for visual evaluations and were not included in the analysis of data.

In these studies percentage data were arcsine transformed before analysis to stabilize variances. The data of two repeat studies were combined after performing a test of homogeneity of variance. Since transformation did not alter data interpretation, non-transformed data were used in the analysis. Data were analyzed using ANOVA, and means were separated by Fisher's protected LSD procedure (p = 0.05).

Results and Discussion

Efficacy studies

When glyphosate 0.56 kg/ha alone was applied, it had less phytotoxic symptoms on both weed species. Incorporation of tallow amine ethoxylate surfactant, achieved significantly higher control (72%) of sicklepod with TA15 (higher EO number) than others (Table 2). But in case of prickly sida significantly higher control (88%) was obtained with TA5. Tank mixing of nonionic alkoxylate surfactant series with glyphosate achieved reduced phytotoxicity with the increase in EO number. With the incorporation of M8 surfactant, a maximum of 61% control in sicklepod and 76% in prickly sida was obtained. Although the values of percent control with glyphosate + both surfactants were significantly higher over

0.56 kg/ha glyphosate with no surfactant but these values were either equal or lower in sicklepod and equal or higher in prickly sida than those obtained with 1.12 kg/ha glyphosate with no surfactant.

Table 2. Effect of surfactant type on the bioefficacy of glyphosate when applied to two broadleaf weed species.

Treatments	Rates (kg a.i./ha)	% Control	
		Sicklepod	Prickly sida
Glyphosate	0.56	14 p	39 m
Glyphosate	1.12	75 def	78 abcd
Glyphosate + TA2	0.56 + 0.25%	21 o	83 abc
Glyphosate + TA5	0.56 + 0.25%	45 l	88 a
Glyphosate + TA15	0.56 + 0.25%	72 efg	85 ab
Glyphosate + M 8	0.56 + 0.25%	61 hi	76 de
Glyphosate + M 10	0.56 + 0.25%	58 hij	63 h
Glyphosate + M 12	0.56 + 0.25%	52 k	24 n
LSD (p=0.05)		------ 05 ------	

TA and M - refer to Table 1. Means followed by the same letter are not significantly different at p=0.05.

Absorption studies

Although environmental conditions will cause different levels of cuticle thickness and composition (Hull *et al.* 1982) but wax contents of these weed species affect the absorption of 14C glyphosate (Harr *et al.* 1991). It is known from an earlier report that both the quantity and quality of waxes present on the leaf surface and the presence of surfactants in treatments play a major role in the absorption of the a.i. through the cuticle (Elmore *et al.* 1998). There was a significant interaction effect between the weed species and herbicide treatments applied in ^{14}Cglyphosate distribution. Significantly higher ^{14}C absorption was recorded by *S. spinosa* than by *S. obtusifolia*, with the application of ^{14}C glyphosate only (Figure 1). However, with the addition of either series of surfactant, significantly higher absorption of ^{14}C glyphosate was recorded by both weed species. Sherrick *et al.* (1986a) reported 2- to 3-folds increase in absorption of glyphosate with polyethoxylate tallow amine adjuvant. Results showed a decrease in absorption when the amount of polar waxes increased from 14% in *S. spinosa* to 93% in *S. obtusifolia* (Harr *et al.* 1991). These data confirmed that the lipoidal nature of the surface minimizes the loss of water from the plant and may act as a barrier to the absorption of agrochemicals (Holloway, 1993). Glyphosate formulated with tallow amine ethoxylate showed increased ^{14}C glyphosate absorption with an increase in POE number. These results were similar to the results reported by Stock and Holloway (1993) and Sharma *et al.* (1996). When ^{14}Cglyphosate was applied with nonionic surfactant, absorption was 6, 18, 30, and 52% in pitted morningglory (*Ipomoea lacunose* L.), prickly sida (*Sida spinosa* L.), barnyardgrass (*Echinochloa crus-galli* L.) and hemp sesbania (*Sesbania exaltata* L.), respectively (Norsworthy *et al.* 2001). Surfactant aided in leaf wetting and herbicide absorption of leaf cuticle up to a point where critical micelle concentration (CMC) is reached (Kirkwood, 1991). Critical micelle concentration is a point beyond which there is no further reduction in surface tension and contact angle of the solution. In this study, addition of surfactant affected translocation of ^{14}C glyphosate (Sharma *et al.* 1996, Feng *et al.* 2003, Liu, 2004). Sherrick *et al.* (1986a, b) noted that high concentrations of surfactants reduced glyphosate movements from treated to rest of the plant tissue due to localized necrosis.

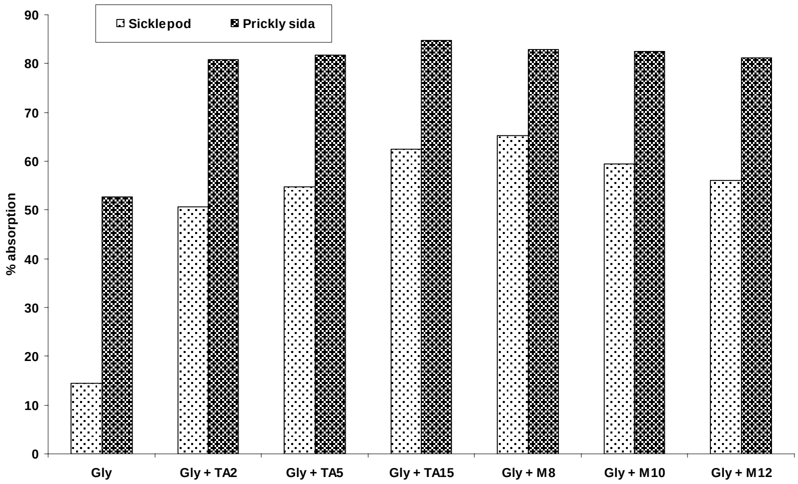

Figure1. Effect of surfactant type on the amount of ^{14}Cglyphosate absorption (% of applied) Gly = glyphosate (TA and M- refer to Table 1).

The translocation of ^{14}C glyphosate as percent of applied showed similar trends as for absorption with both the series of surfactant (Figure 2) (Feng *et al.* 2003; Liu, 2004). Data indicated the amount of ^{14}C glyphosate retained by the treated leaf was significantly higher in the case of prickly sida than sicklepod. The amount of ^{14}C glyphosate in the treated leaf decreased with an increase in POE number in tallow amine ethoxylate, but the amount increased with an increase in POE in the nonionic alkoxylate surfactant series (Figure 2). This presence of higher amounts of ^{14}C glyphosate in treated leaf of indicated the possible wax solubilization action of surfactant present in the herbicide solution.

It has been suggested further that substantially enhanced absorption and translocation may happen because of stomatal infiltration of a.i. (Stevens *et al.* 1991). The mechanism of surfactant-induced transfer of pesticide active ingredient through the cuticle is not well established (Buckovac, 1976). It could involve one or more of the following processes, as described by Stock and Holloway (1993): (i) change in solubility relationships and partitioning processes that are favorable to transfer, (ii) decrease in the resistance of the cuticle to diffusion, and/or (iii) activation of specific polar or non-polar routes through the cuticle.

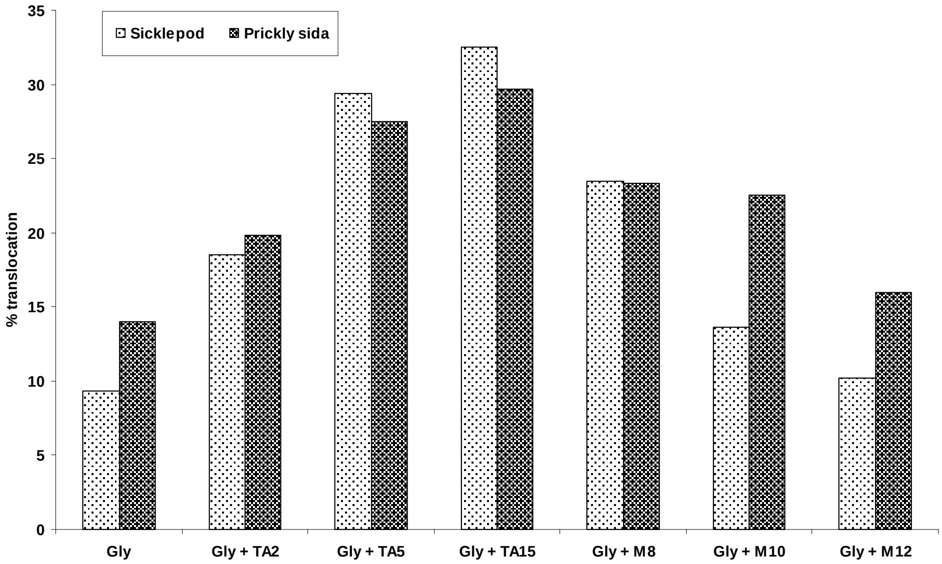

Figure 2: Effect of surfactant type on the amount of ^{14}Cglyphosate translocation (% of applied).

Results of this study provide further evidence as to the importance of adjuvant on the absorption, translocation, and efficacy of glyphosate with the addition of nonionic alkoxylate surfactant series. Although percent weed control was significantly higher with the addition of either surfactant to glyphosate but the value of control was higher with higher POE surfactant in the tallow amine ethoxylate series and with the lower POE surfactant in the nonionic alkoxylate surfactant series (Table 3). Similarly, Gaskin (1995) reported that many surfactants do not simply exert their effects on plant surfaces, but penetrate rapidly through the waxy cuticle and into the underlying tissues. The foliar absorption of a surfactant can in turn affect the absorption and translocation of a systemic herbicide, which means that the surfactant providing greatest absorption may not provide greatest efficacy.

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STICKY FLORESTINA (*Florestina tripteris* DC. Prod.) HERBICIDE SCREENING FOR A RECENT INVASIVE SPECIES IN CENTRAL WESTERN QUEENSLAND

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Abstract: *Florestina tripteris* DC. Prod. (Sticky Florestina) is an annual weed that is established in the Tambo and Barcaldine Shires of Central Western Queensland, Australia. It was accidentally introduced from Southern Texas, U.S.A in a consignment of Buffel grass seed in 1964. It is a member of the Asteraceae family and grows up to 1 m in height. *Florestina tripteris* has grey-green leaves and small white flowers. The pecies produces hundreds of seeds that are sticky and are spread easily by stock and machinery, including vehicular traffic. *Florestina tripteris* has the ability to survive dry conditions, germinating only after rain and often completing its life cycle within a month. A herbicide screening trial commenced in April 2002. The mortality of overall foliar sprayed plants was high but could have been improved by targeting plants following germination in early spring. The best three foliar overall treatments were bromoxynil 200 g l^{-1} at 4 g a.i. l^{-1} (controlling 85% of weeds sprayed), 2,4-D acid 300 g l^{-1} at 17 g a.i. l^{-1} (controlling 75% of weeds sprayed) and 2,4-D dimethylamine salt 500 g l^{-1} at 16 g a.i. l^{-1} (controlling 70% of weeds sprayed). The best three treatments from a residual seedling control trial at Barcaldine site were clopyralid 300 g l^{-1} at 6 g a.i. l^{-1} (providing 0.6% of seedling recruitment of untreated areas), and metsulfuron methyl 600 g kg^{-1} at 0.24 g a.i. l^{-1} (giving recruitment counts that were 30% of the untreated areas) and triclopyr ester 300 g l^{-1} at 4.2 g a.i. l^{-1} + picloram 100 g l^{-1} at 1.4 g a.i. l^{-1} (giving 80% of seedling recruitment of the untreated areas). An Australian permitted registration allows for the use of 200 g 2,4-D amine + 200 g alcohol alkoxylate non-ionic surfactant 100 l^{-1} water to be sprayed alone, 2-3 weeks after germinating rains, but before bud and flower stage. Once budding and flowering have begun, a tank mix of 100 g 2, 4-D amine + 20 g metsulfuron methyl + 200 g alcohol alkoxylate non-ionic surfactant 100 l^{-1} in water will kill treated plants and reduce the number of germinants emerging following the next rain.

Key words: Control, herbicide, pasture, soil-moisture, toxic, weed.

Introduction

Florestina tripteris DC. Prod. (Sticky Florestina) was first investigated as a weed threat in the Tambo and Barcaldine Shires of Central Western Queensland in 2000. It was accidentally introduced from Southern Texas, U.S.A in a consignment of Buffel grass (*Cenchrus ciliaris* L.) seed in 1964 (Bucknell per. comm., 2000). *Florestina tripteris* is a member of the Asteraceae family, with lower leaves that are grey-green, simple and opposite and upper leaves trifoliate and alternate. The entire plant is covered in very short sticky white hairs. Plants attain 10 cm to 1 m in height at maturity and bear numerous flower heads with small white flowers (Turner, 1963).

Florestina ripteris produces hundreds of seeds that are sticky and are spread easily by stock, machinery and people, particularly on areas where heavy traffic is common. It is an annual plant that has the ability to survive dry conditions and drought, germinating only after rain and sometimes completing its life cycle within a month while soil moisture is available. *F. tripteris* is a highly invasive weed that will prove hard to control because of its growth habit when invading pastures. It will grow as a monoculture along road verges and on bare disturbed areas, but tends to occur as isolated plants in amongst pasture species such as Buffel or Mitchell grass (*Astrebla pectinata* (Lindl.) F. Muell. and *A. elymoides* F. Muell. ex F.M. Bailey). Scattered plants with counts up to 5000 ha^{-1} have been recorded in pasture.

The preferred areas of *F. tripteris* include roadsides, stock routes and disturbed or overgrazed pastures (Turner 1963). Newly graded roads and fence lines have been the sites of the densest infestations. The first Queensland Herbarium record for this species from the

Tambo Shire was in April 1989 and the first record from the Barcaldine Shire was in October 1993.

Materials and Methods

Herbicide screening trials were conducted in Queensland, 25 km south of Barcaldine (145°19”06’E, 23°47”40’S) and 50 km south-west of Tambo (145°56”12’E, 250°12”58’S) in March 2002. Both sites were treated with the same herbicides (Table 1).

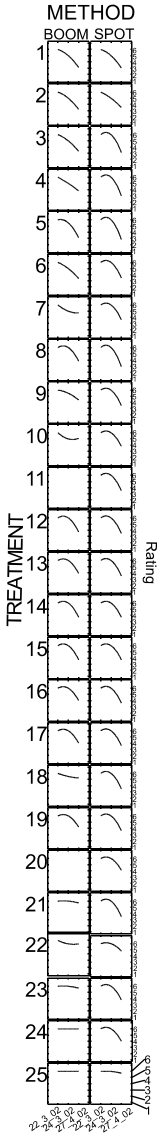
Table 1. Herbicides trialled on F. *tripteris* , listed in order of effectiveness

Treatment	Active constituent	Dilution g a.i per 10 litres
1	200 g l^{-1} bromoxynil as noe	40
2	300 g l^{-1} 2,4-D acid	168
3	500 g l^{-1} 2,4-D dimthylamine salt	160
4	300 g l^{-1} triclopyr ester	42
	+100 g l^{-1} picloram	14
5	200 g l^{-1} bromoxynil as noe	80
6	300 g l^{-1} triclopyr ester	60
	+ 100 g l^{-1} picloram	20
7	300 g l^{-1} clopyralid as tipa	30
8	360 g l^{-1} glyphosate isopropyl ammine salt	144
9	300 g l^{-1} clopyralid as tipa	60
10	600 g kg^{-1} metsulfuron-methyl	2.4
11	200 g l^{-1} fluroxypyr ester	20
12	360 g l^{-1} glyphosate isopropyl ammine salt	72
13	200 g l^{-1} fluroxypyr ester	40
14	500 g l^{-1} 2,4-D dimthylamine salt	80
15	300 g l^{-1} 2,4-D acid	84
16	200g l^{-1} fluroxypyr ester	80
17	600 g kg^{-1} metsulfuron-methyl	4
18	500 g l^{-1} dicamba as dma	80
19	360 g l^{-1} glyphosate isopropyl ammine salt	36
20	500 g l^{-1} dicamba as dma	40
	600 g kg^{-1} metsulfuron-methyl	2.4
21	500 g l^{-1} dicamba as dma	40
22	500 g l^{-1} atrazine	80
23	600 g kg^{-1} metsulfuron-methyl	1.2
24	500 g l^{-1} atrazine	160
25	control	

noe = n-octanoyl ester, tipa = triisopropanolamine salt dma = dimethylamine salt

Twenty-four treatments involving 12 products were sprayed on *F. tripteris* by the spot foliar overall spray methodology or 1 m hand-held boom application technique. Plant condition was reflected as a visual rating index (Australian Weeds Committee, 1979). Ratings ranged from 1 indicating plant death to 6 indicating normal growth (Figure 1).

The Barcaldine experiment was set out in randomly selected plots (size 10 m^2) along Queensland stock route M217. Dominant plant species within this trial site were *Acacia cambagei* R.T.Baker and *Ptilotus exaltatus* Nees var. exaltatus. Scattered throughout the site were *Acacia farnesiana* (L.) Willd., *Boerhavia schomburgkiana* Oliv., *Ocimum* *tenuiflorum*.L. and *C. ciliaris,* in conjunction with native grasses *A. elymoides* and *Astrebla squarrosa* C.E. Hubb (Milson, 1995).



The days were fine with light breezes up to 2 m per second, having no effect on herbicide application. The temperature ranged between 32°and 39°C. A model 320 hand-pump Swiss-mex® compression sprayer with 9 litre capacity with Rega® 1 mm adjustable nozzles was used. The boom hand pump sprayer was a Swiss-mex® compression sprayer with 15 litre capacity fitted with four Hardi 41212 fan nozzles spaced 34 cm apart. Twenty-three plots were subjected to a 1 m boom foliar spray at 525 l ha^{-1}. The spot spray technique used ~ 200 L ha^{-1} depending on *F. tripteris* density. Data were collected on 22nd and 23rd March 2002 (initial application), with the first herbicide effect assessment on 24th March 2002, and the second assessment made on 27th April 2002. Further assessments were made the following year (Figure 2) to determine the residual effects of herbicides upon seedling emergence. Treatment plots were replicated three times. The assessments were on 10 plants within a 0.25 m^2 quadrat. The quadrat was randomly assigned along a randomly selected entry point transect line.

The experiment near Tambo was set out (randomly selected 10 m^2 plots) amongst existing vegetation. Grazing is the primary use of this land so the predominant introduced pasture species was *C. ciliaris* in conjunction with native pasture species, including *A. elymoides*. Other plant species within the trial areas were *A. cambagei*, *Brachychiton rupestris* (T. Mitch. ex Lindl.) K. Schum, *Josephinia eugeniae* F. Muell. and *Aristida leptopoda* Benth. (Henderson, 1997). The weather parameters at the time of application were similar to the Bacauldine temperature range and wind speed. Again Swiss-mex® compression sprayers were used with spot spray and hand boom attachments.

Figure 1. Ratings determining herbicide damage to *Florestina tripteris*. Rating 1= Dead, 2 = Total stem death, 3 = Top stem death, 4= Tip necrosis, 5 = Leaves yellowing, 6 = Normal. Treatments are as in Table 1.

Three assessments of herbicide effects were made in 2002 (Figure 1). A further four assessments were made after rain in 2003 to determine the residual effects of the herbicides on seedling emergence (Figure 2). These counts were obtained in 0.25 m^2 quadrats as before. Treatments 10 and 13 were not considered when assessing the residual effects of herbicides.

Plants were healthier at the Tambo site due to higher soil moisture, while plants at the Barcaldine site were moisture-stressed and sparse in distribution. The Barcaldine boom sprayed plants were up to 1 m in height and more mature, with most flowering and some exhibiting seed set. Although the overall density was less than at the Tambo site, plant numbers in many plots were high and these plants averaged 0.75 m in height.

Analysis

The results were analysed with Systat 9 (Wilkinson, 1999a), using repeat measure multivariate analysis of variance (Wilkinson and Coward, 1999) and as outlined by (Sparkes and Panetta, 1997). A graphical presentation was used to differentiate between treatment and application method effects (Wilkinson, 1999b) (Figures 1 and 2). Readings from the 30 assessed plants from within the datum areas for each cell were averaged for each assessment period and the smoothing applied utilized a distance-weighted least squares method (Wilkinson, 1999). The 30 assessed plants per assessable cell provided adequate numbers to represent the condition of treated *F. tripteris* in the two sites and meet the conditions for sampling normally distributed data. The data were combined for analysis (Figure 1). A multivariate repeat measure analysis was carried out with each of the treatment responses and site influences as the within subject variables, *i.e.* ratings at each of the assessments x treatment (Wilkinson and Coward, 1994). The metric, due to the uneven spread of weeks after application between assessments, was 1, 2 and 5 for herbicide efficacy and 1, 2, 4, and 12, for herbicide residual effects and was used to adjust *F* values for the error caused by non-systematic assessment. Post-hoc between-subject comparisons on the final results were moderated using Tukey adjustment (Wilkinson and Coward, 1994).

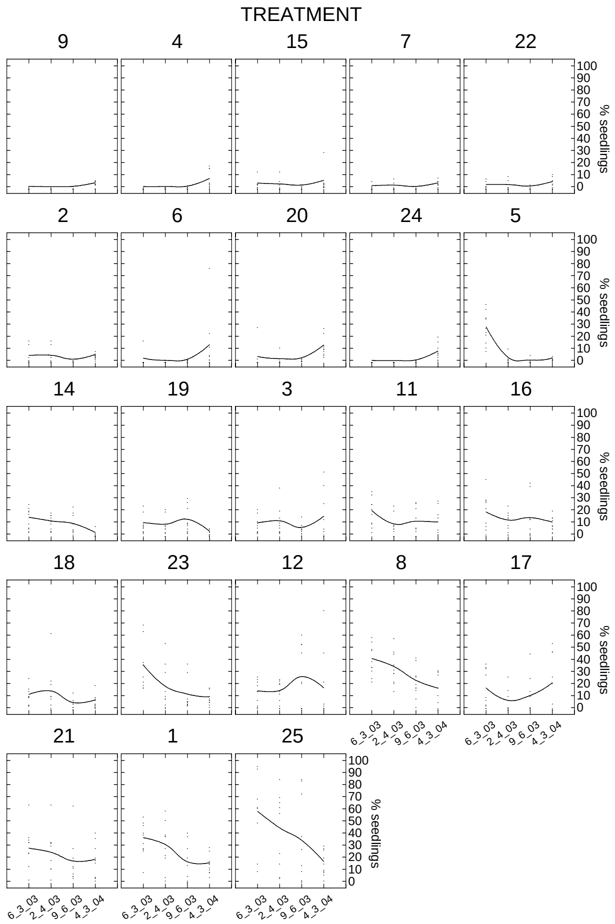


Figure 1. *Florestina tripteris* seedling emergence following herbicide application. Ratings are % ground cover per 0.25 m^2 quadrat. Treatments are as in Table 1.

Results

Herbicide efficacy

There was a significant difference between the performance of the herbicides (MS = 5.44 df = 22, 446 F = 31.37 P <0.01). There was a steady quadratic movement from healthy pre-sprayed plants to plants that moved towards necrosis at the last inspection (Table 2).

Table 2 Pair-wise comparisons between assessment times

Assessments over time	Mean difference	Std. Error of difference	p-value*	95% Confidence interval*	
1 / 2	0.234	0.013	0.000	0.203	0.265
1 / 3	2.031	0.037	0.000	1.942	2.119
2 / 3	1.797	0.042	0.000	1.695	1.898

*Bonferroni correction for multiple comparisons was used.

The Tambo (Site 1) plots treated by the spot spray method produced more effective control compared to the Barcaldine plots (Site 2) treated using a hand boom (MS = 5.93 df = 1, 446 F = 34.22 p <0.01). However, the most effective first 12 treatments had similar effectiveness at both sites, whether applied by spot spraying or hand boom. The mortality of treated plants was higher for spot spraying possibly because plants were sprayed to run-off point.

The best three foliar overall treatments were bromoxynil 200 g l^{-1} at 4 g a.i. l^{-1} (controlling 85% of weeds sprayed), 2,4-D acid 300 g l^{-1} at 17 g a.i. l^{-1} (controlling 75 % of weeds sprayed) and 2,4-D dimethylamine salt 500 g l^{-1} at 16 g a.i. l^{-1} (controlling 70% of weeds sprayed).

Residual seedling control

A significant difference existed between the residual activity of the herbicides expressed as seedling emergence post application (Wilks' Lambda (counts over time x treatment) = 0.3 df = 66, 607 F = 4.6 p<0.01). All treatments produced residual effects that were different from the control except for treatment 8, the high rate of glyphosate (MS = 950.6, 80.4 df = 22,207, F = 11.8, p = 0.2)

The best three treatments from the residual seedling control pilot trial at the Barcaldine site were clopyralid 300 g l^{-1} at 6 g a.i. l^{-1} (yielding 0.6% of seedling recruitment of untreated areas), and metsulfuron methyl 600 g kg^{-1} at 0.24 g a.i. l^{-1} (giving recruitment counts that were 30% of the untreated areas) and triclopyr ester 300 g l^{-1} at 4.2 g a.i. l^{-1} + picloram 100 g l^{-1} at 1.4 g a.i. l^{-1} (giving 80% of seedling recruitment of the untreated areas).

Discussion

The information from this trial led to a permit being issued by the Australian Pesticides and Veterinary Medicines Authority (APVMA) that was renewed in December 2006. The permit allows for the use of 400 ml 2,4-D amine (Amicide 500®) + 200 ml BS 1000® 100 l^{-1} water to be sprayed alone, 2-3 weeks after germinating rains, and before bud and flower stage. Once budding and flowering have begun, a tank mix of 200 ml 2,4-D amine (Amicide 500®) + 20 g metsulfuron methyl (Brush off®) + 200 ml BS 1000® 100 l^{-1} water will kill treated plants and reduce the number of germinants following the next rain. This tank mix can be sprayed only once for each cohort of weed emergence. This gives local authorities, landholders and conservation groups a tool to incorporate into control options and should reduce the spread of this plant.

Prevention of *F. tripteris* establishment in pasture is a key factor in strategic control planning. Variables such as rainfall patterns, soil types, associated plant communities, grazing pressure and temperature are determinants in invasiveness of opportunistic weed species (Pressland, 1984). Sustained periods of drought and movement of stock and feed are factors in the establishment *F. tripteris* in new areas. *F. tripteris* will grow along road verges on unsealed roads, increasing the likelihood of establishment in new areas. Adaptive management techniques involving herbicide application should be instituted immediately after *F. tripteris* appears following rain events, as the rosette stage is more susceptible to herbicide treatment. Treatment at the rosette stage will prevent seed from being set and enable establishment of competitive pasture species. The rosette stage may last less than two weeks depending on how much follow up rainfall occurs.

Florestina tripteris develops stands of higher density where soil disturbance has occurred, such as along the verges of roads and in consistently used stock routes. It was also noted that areas of heavily grazed pastures were more susceptible to invasion by *F. tripteris*. The road verges and stock route areas need to be surveyed and *F. tripteris* encountered needs to be treated with herbicide. Improved pasture management may assist in reducing the incidence of *F. tripteris* in grazing lands. Pot experimentation utilising controlled seed inputs will be necessary to confirm the herbicide residual effects observed in this field research.

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MOLECULAR CHARACTERIZATION OF WEEDY RICE IN SRI LANKA

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Abstract: Weedy rice is an emerging problem in Sri Lanka affecting the lowland rice (*Oryza sativa*) cultivation due to its competitive ability and enriching soil seed bank due to seed shattering. The control of weedy rice has been difficult due to its morphological similarity (mimicry) to cultivated rice. A study was undertaken to use morphological as well as molecular techniques to identify weedy rice collected from cultivated paddy fields, in an attempt to elucidate the relationship of these weedy forms with the cultivated and wild rice varieties. The two weedy rice accessions (Weedy Rice-1; collected from Puttlam district, north western province) and Weedy Rice-2 (collected from Galle district, southern province) were subjected to Random Amplified Polymorphic DNA (RAPD) analysis together with a popular new improved rice variety Bg-300, a traditional variety (*Kaluheneti*), and a annual wild type (*O. nivara*). There morphological developments were also studied in plant house conditions. Among 11 primers used in the DNA fingerprinting, 6 primers (OPF-4, OPF-16, OPF-18, OPM-10, OPM-18, OPY-11) showed specific bands that helped to differentiate the samples tested. The dendograms obtained from both molecular and morphological analysis clearly separated the cultivated varieties, wild rice and weedy rice into different clusters. The shortest genetic distance was observed between Bg-300 and *Kaluheeneti* while the highest genetic distance as observed between *O. nivara* and the weedy rice accessions used in the study. The morphological analysis indicated that weedy rice show intermediate characteristics to those of cultivated and wild (annual) rice. In both analyses, the positioning of two weedy rice accessions between the cultivated and annual wild type (*O. nivara*) indicated the potential of the occurrence of a hybrid between wild and cultivated rice.

Key words: Weedy rice, *Oryza sativa*, Bg-300, *Kaluheeneti*, *O. nivara*, DNA fingerprinting, morphological characters

Introduction

Weeds are the most important biological constraint in rice cultivation, which reduce the yield qualitatively and quantitatively, and are adapted to frequently disturbed habitats. Weedy rice has become a threat for productivity of rice and they grow in directly sown paddy fields and abounded rice fields. Weedy rice could occur due to several factors like, gene flow between wild and cultivated *japonica* types, old rice varieties becoming feral as well as crosses between cultivated rice and wild rice such as *Oryza nivara* and *O. rufipogon* (Marambe, 2005). Most of weedy rice contains AA genome, because they have evolved from the cultivated rice, and have intermediate characteristics between cultivated and wild rice. The most widely documented weedy rice is *O. sativa* in all rice-growing regions, especially, in Asian countries. Apart from “AA genome *Oryza* species”, only three other *Oryza* species have been reported as weeds in rice fields: *O. latifolia* in Central America, *O. officinalis* in the Philippines, and *O. punctata* in Africa (Vaughan *et al*. 2005). The weedy rice occurs mostly in direct-seeded rice lands, due to shifting from transplanting to dry sowing and non-use certified seeds. As they are morphologically similar to cultivated rice during the vegetative stage, with seed shattering, varying degree of secondary dormancy, higher competitive ability and showing similar responses to the herbicides as in the case of cultivated rice, have made their control more difficult. Most of the characters which weedy rice contains are determined genetically. Hence, the identification of weedy rice is important in order to reduce their infestation.

In mid 1990's, weedy rice was first identified as a threat from Vavunia, Ampara, and Batticaloa, districts (Marambe and Amarasinghe, 2000). The yield losses have been estimated to vary from 10-100% due to their competitive ability. On the basis of the awn length four different types of weedy rices have been reported in Sri Lanka. A large proportion of their seeds are unfilled and filled grains had a low ability to germinate 4 weeks after shattering (Marambe, 2005). The most appropriate methods for identification of weedy rice are DNA-based markers and there are several different DNA fingerprinting techniques such as Random Amplified Polymorphic DNA (RAPD), Amplified Fragment Length Polymorphism (AFLP), and Microsatellites. Among the DNA fingerprinting techniques, RAPD is a more powerful technique, which can identify relationship among the crop wild relatives and RAPD have been used to determine DNA diversity in a range of crops including rice (Mackill, 1995). The present study thus, attempted to identify the relationship of weedy forms of rice to cultivated and wild rices available in Sri Lanka using RAPD technique, and the morphological differences of weedy rice when compared to wild and domesticated rice.

Materials and Methods

Seed collection and planting

Seeds from five rice accessions (*Oryza sativa*) including one improved rice variety (Bg 300), one traditional rice variety (*Kaluheeneti*), one annual wild rice accession (*O. nivara*), and two weedy rice accessions were collected as described in Table 1. The cultivated varieties and wild accession were obtained from Plant Genetic Resources Centre (PGRC), Gannoruwa, and weedy accessions were collected from cultivated paddy lands at Puttlam and Galle districts.

Table 1. Selected rice varieties and their types.

Code	Variety	Type	Site of Collection
1	Bg-300	New improved	PGRC*
2	*Kaluheeneti*	Traditional	PGRC
3	*Oryza nivara*	Wild Type (annual)	PGRC
4	Weede Rice-1	Suspected weedy type	Puttlam, north western province
5	Weedy Rice-2	Suspected weedy type	Galle, southern province

*PGRC – Plant Genetic Resources Centre, Gannoruwa, Sri Lanka.

Seeds from the different accessions were soaked in water for 24 hrs and then wrapped in banana leaves for 48 hrs for germination. Thereafter, the pre-germinated rice seeds were grown in separate plastic trays for the molecular analysis and in clay pots filled with soil for the morphological analysis, under plant house conditions at the Agricultural Biotechnology Centre (AgBC), Faculty of Agriculture, University of Peradeniya. Five plants were grown in each pot with five pots per each treatment. Out of five pots, one pot from each treatment was randomly selected to measure the shoot and root dry weight. The dry weight was measured at the late vegetative stage. The fertilizer application was done according to the recommendations of the Department of Agriculture. Each pot was kept at field capacity through out the research period.

Molecular analysis

DNA extraction and RAPD analysis: The DNA was extracted based on modified CTAB (cetyltrimethylammonium bromide) method (Culling, 1992). About 300mg of fresh tender rice leaves obtained from 21 days old rice seedlings of the respective rice varieties were separately frozen at -80°C and grounded to form a fine powder. Added 1.5 ml of pre-heated 2% CTAB extraction buffer (2% CTAB, 100mM Tris-HCl pH 8.0, 20 mM EDTA, 1.4 M

NaCl) and were incubated for 30 minutes at 60°C in a shaking water bath. An equal volume of chloroform : isoamyl alcohol (24:1) was added to the tube, inverted for 10 min, and centrifuged at 8000 rpm at room temperature for 5 minutes. The upper aqueous phase was pipetted into another fresh set of Eppendorf tubes and approximately 0.6% volume of ice-cold isopropanol was added into the Eppendorf tube containing aqueous phase and were centrifuged at 5000 rpm for 5 minutes.

The aqueous phase was discarded, and 1ml of 70% ethanol solution was added and centrifuged at 12000 rpm, for 10 minutes. The washing solution was drained to dry the DNA pallet by inverting the tubes on a paper towel for an hour. DNA was dissolved in 50 µl of sterile distilled water and kept in water bath at 38°C for 30 minutes. The DNA samples were stored at 4°C until use, and the DNA quantification was carried out using a UV spectrophotometer (UV 1600-UV Visible, SHIMADZU; absorbance at 260 nm and 280 nm). Working solutions of rice DNA were prepared at 30 ng/µl DNA concentration using double distilled sterilized water.

The RAPD amplification was carried out using Techne Thermocycler (FFG05TUD, Flexigene, USA), each 30µl of reaction mixture containing 10x buffer (ABgene, UK), 25 mM of $MgCl_2$, 0.2 mM dNTP (Sigma Chemical Co., St, Louis), 5pM deca-mer primer (Operon Technologies, Inc., Alameda, CA), and 0.5 U Taq polymerase and 30ng/µl sample DNA. The amplification conditions were programmed 1 cycle of initial denaturation 94°C for 3 minutes followed by 40 cycles of 93°C for 1 minute, 35°C for 3 minutes, 72°C for 2 minutes for respectively denaturation, annealing and extension. The final extension at 72°C for 10 minutes and final hold at 4°C until electrophoresis. The amplified DNA samples were electrophoresed on 1.5% of agarose gels in 1X TBE buffer (40mM Tris base, 20mM Boric acid, 2mM Na_2-EDTA-$2H_2O$) and stained with ethidium bromide, scanned under UV light and photo documented using UV-photo-print gel system.

Eleven primers (OPE-1, OPE-6, OPF-4, OPF-6, OPF-16, OPF-18, OPG-4, OPM-4, OPM10, OPM18, and OPY-11) were initially screened with five rice accessions to identify the most promising for detecting polymorphisms. Among the tested six Operon primers (OPF-4, OPF-16, OPF-18, OPM10, OPM18, OPY-11) were selected to screen the samples. The final set of experiments were conducted a minimum of three times in independent runs to get final result.

Data analysis

Polymorphic bands were scored visually 1 for presence or 2 for absence with each primer. Genetic distances between each accession were calculated based on modified Roger's distance (Wright, 1978, with TFPGA, version 1.3). Distances were used to construct a cluster diagram by the Unweighted Pair Group Method with Arithmetic mean (UPGMA) method. Data were bootstrapped over 1000 permutation pains in order to increase the accuracy.

Morphological characteristics

The number of tillers per plant, tiller length, leaf blade length and width, and leaf area of the wild, weedy and cultivated rice were recorded at the late vegetative stage and average panicles per plant and average seeds per plant were recorded at the heading stage.

Experimental design and data analysis

The experiment was conducted in a Complete Randomized Design (CRD) with 4 replicates. The data were analyzed using the SAS computer package.

Results and Discussion

Molecular analysis

The eleven primers used in the study showed 4-12 bands per primer. Among them six primers showed specific bands, which could help differentiate at least one test sample from the others. Majority of the amplified fragments were polymorphic. The OPF-4 primer showed two clear monomorphic bands and one polymorphic band at the position of A (Plate 1a).

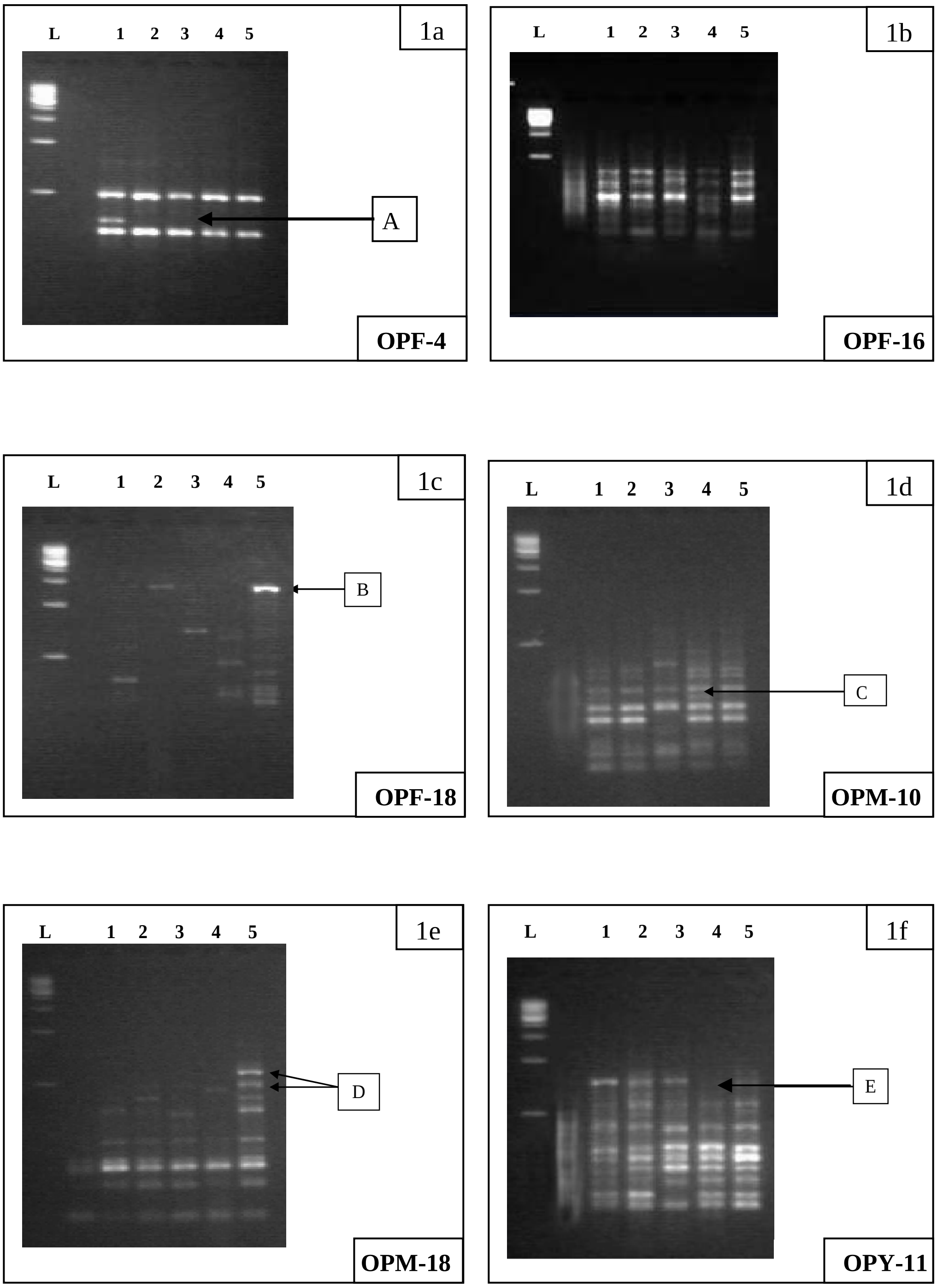

Plate 1. Gel electrophoresis of amplified DNA fragments from five different rice accessions using 6 different primers

This polymorphic band could be used as potential information for identification of BG-300 from the other rice varieties used in the present study. The OPF-16 primer showed four monomorphic bands that are common to all the rice accessions and six polymorphic bands that are specific to different rice accessions (Plate 1b). The OPF-18 primer showed nine polymorphic bands, but no monomorphic bands were observed. Each polymorphic band provides potential information to identify each accession (Plate 1c). A clear specific band was only present in Weedy Rice-2 collected from Galle district, between 2 kb and 3 kb at the B position. This information can be used to identify Weedy Rice-2 in future studies. The OPM-10 primer showed two monomophoric and nine polymorphic bands (Plate 1d). In this case, *Kaluheenati* and Weedy Rice-2 showed similar banding patterns and indicating the close relationship between these two accessions. Weedy Rice-1, which was collected from Puttlam district, showed clear polymorphic band at position ‘C’, and it could be used as potential information for identification of Weedy Rice-1 from other rice accessions used in present study. The OPM-18 primer showed four clear monomorphic bands and seven polymorphic bands (Plate 1e). It showed two clear polymorphic bands at the position of D that specify to the Weedy Rice-2.

The OPY-11 primer showed a similar banding pattern in both Weedy Rice-2 and *Kaluheenati* (Plate 1f), followed by OPM-10 (Plate 1d). It indicates that gene introgression between cultivated and weedy rice. This primer showed another specific band at the E position in all the accessions except Weedy Rice-1. This information will be useful to identify the Weedy Rice-1 from the other tested accessions.

Six primers, which were used for the analysis of the rice accessions, have produced 38 reproducible polymorphic bands. These data were used to obtain the genetic distances between the five rice accessions as described in Table 1. The smallest genetic distance was observed between Bg-300 and *Kaluheeneti*, which are the closely related cultivated rice varieties. The highest genetic distance was observed between *O. nivara* and Weedy Rice-2 followed by *O. nivara* and Weedy Rice-1 thus suggesting that these are distantly related rice accessions. Valverde (2005) reported that weedy rice could represent partially de-domesticated forms arising from back mutation with indica characteristics, probably derived from old cultivars.

Table 1. Genetic distances between five different rice accessions based on RAPD data.

Population	1	2	3	4	5
1	*****	0.1964	0.3571	0.3214	0.2857
2		*****	0.2679	0.3393	0.3036
3			*****	0.3571	0.4286
4				*****	0.3214
5					*****

Genetic distances obtained from RAPD data were used to create a cluster diagram that has shown in Figure 1. The UPGMA cluster diagram showed four major clusters as A, B C and D corresponding to the different rice accessions. The Weedy Rice-2 was clustered together with two cultivated rice varieties suggesting a close relation among these three accessions. Bg-300, *Kaluheenati* and Weedy Rice-2 separate into same cluster ‘A’ exhibiting exchange of genes between them. Cluster ‘A’ has divided into two sub clusters and two cultivated varieties were grouped together, which indicated close relationship between them.

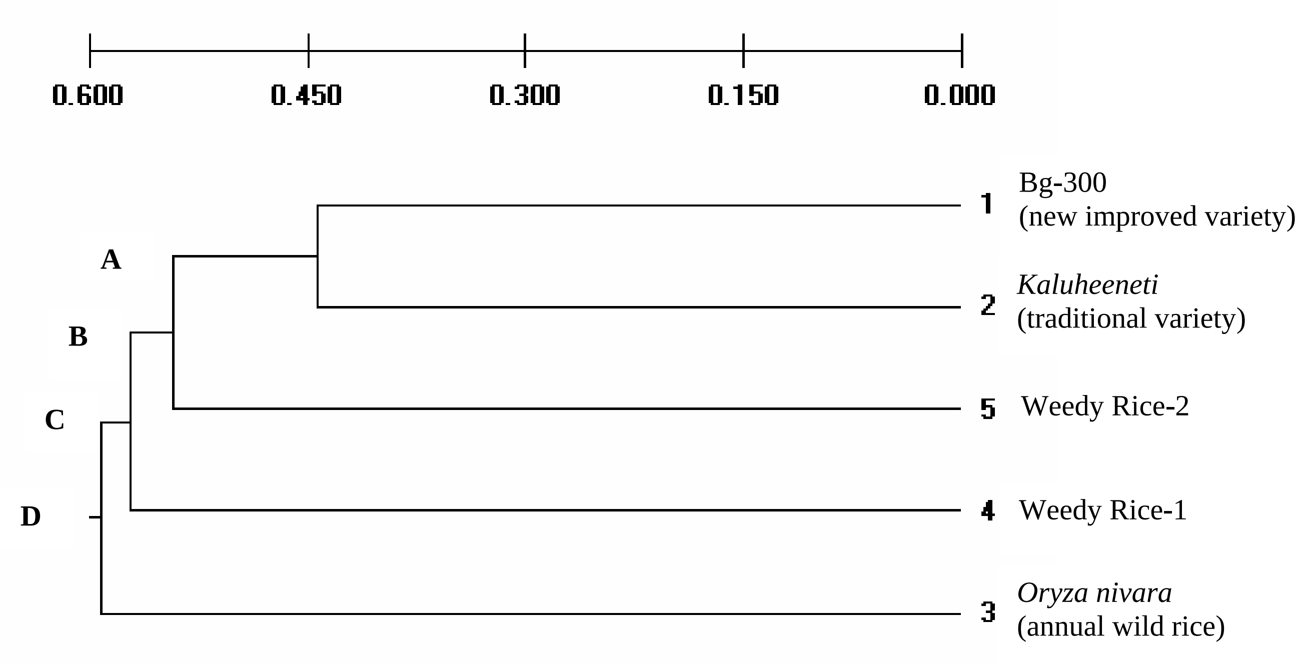

Figure 1. Dendrogram generated by OPF4, OPF16, OPF18, OPM10, OPM18 and OPY11 primers for five accessions tested.

The separation of two Weedy Rice accessions into two clusters indicated the geographical isolation between them. The position of two Weedy Rice accessions between the cultivated and wild type *Oryza nivara*, indicated the potential of the occurrence of a hybrid between wild and cultivated rice. Hybrid derivatives from the association between cultivated and wild rice are adapted to a wide spectrum of habitats from the wild to cultivated fields and gene flow from cultivated rice into wild rice frequently occurs. Weedy rice seems to rapidly evolve and adapt to particular location or condition (Vaughan *et al*. 2005).

Morphological analysis

Morphological characteristics were studied only in the case of cultivated and wild rice, and Weedy Rice-1 (Table 2). The plants of Weed Rice-2 were not included in the analysis as they were mechanically damaged due to unavoidable circumstances. The tiller length of different rice accessions at the time of the heading varied within the range of 70-100 cm and the height of the variety *Kaluheeneti* was significantly higher (p< 0.05), when compared to other three accessions (Table 2).

Table 2. Morphological characters of rice accessions

Type	Tiller length (cm)	No. of tillers/ plant	No. of panicles/ plant	No. of seeds/plant	Leaf blade length (cm)	Leaf blade width (cm)	Leaf area/plant (cm²)
Kaluheeneti	96.8o a	3 c	3 c	252 b	41.0 a	0.83 c	338 a
Bg-300	71.33 c	3 c	3 c	268 ab	30.9 b	1.15 a	300 ab
Oryza nivara	72.76 c	8 a	5 a	195 c	32.0 b	0.73 c	230 c
Weedy Rice – 1	87.25 b	5 b	4 ab	322 a	43.8 a	1.00 b	246 bc
Weedy Rice – 2	NA	NA	NA	NA	NA	NA	NA

Within a column, means followed by te same letter are not significantly different by the LSD (p=0.05).

Kaluheeneti is a traditional rice variety, which is taller than improved varieties, and it was susceptible to lodging. The weedy rice accessions collected were of intermediate height between *Kaluheeneti* and *O. nivara*. The latter shows a prostrate growth habit.

The average number of tillers per plant was significantly higher ($p<0.05$) in *O. nivara*, but most of tillers were unproductive. The lowest number of tillers was observed in Bg-300, which is an improved variety, and the number of productive tillers was higher. Weedy Rice accessions showed intermediate characters between Bg-300 and *O. nivara.* The average number of panicles per plant was significantly higher in Weedy Rice, but there were higher percentage of unfilled seeds. Awns were observed in the Weedy Rice accessions collected as well as in *O. nivara*, and shattering of seeds were observed also in these two groups. The color of the leaf sheath in *O. nivara* and weedy rice was purple, while it was pale in Bg-300 and *Kaluheenati*.

The dendrogram was obtained from analyzing descriptors (Figure 2). According to the eigen values and its proportions of the covariance matrix, three main morphological characters (length of the tillers, average tillers per plant, and average panicles per plant) have given higher contribution to form the dendrogram.

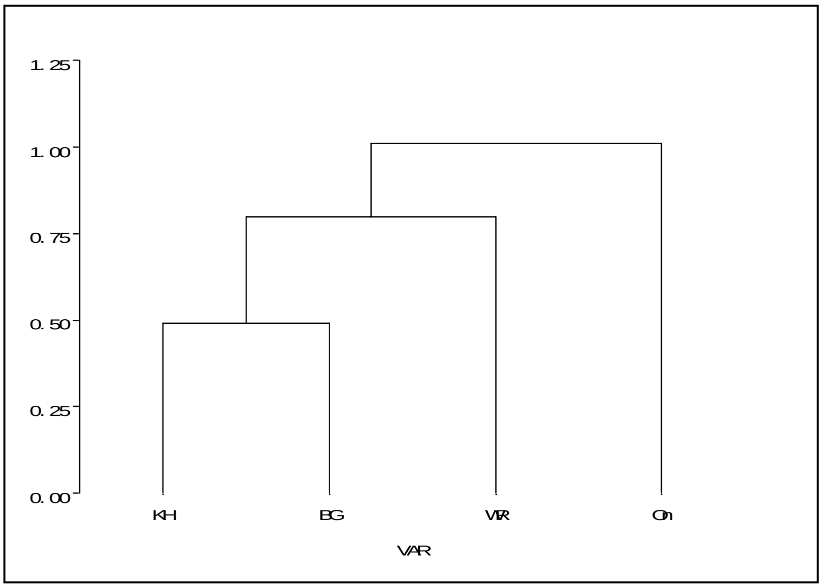

Figure 1. Dendrogram generated by the analysis of morphological parameters (KH – *Kaluhheneti*, BG – Bg 300, WR – Weedy Rice, On – *Oryza nivara*)

The dendogram shows a clear separation of cultivated (*Kaluheeneti* and Bg-300), wild (*O. nivara*) and Weedy Rice (WR) into three different clusters. The *Kaluheeneti* and Bg-300 has cluster together at similarity level 0.49 indicating the close relation between two cultivated rice accessions. The wild rice *O. nivara* is grouped in another cluster at the level of 1.01. The weedy rice accession is grouped between cultivated and wild rice accessions and it indicate the intermediate characters of the wild and cultivated rice.

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ALLELOPATHIC POTENTIAL OF WILD ONION (*Asphodelus tenuifolius* Cav.) EXTRACTS ON GERMINATION AND SEEDLING GROWTH OF CHICKPEA (*Cicer arietinum* L.)

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Abstract: Water extracts obtained from roots, shoots and fruits of wild onion (*Asphodelus tenuifolius* Cav.) harvested at maturity and soil taken from the (*A. tenuifolius*) field were used to determine their allelopathic effects on mean germination time, time taken for 50% germination, germination index, germination/emergence percentage and seedling growth of chickpea (*Cicer arietinum* L.) in the laboratory at 15^oC. Roots, shoots and fruits of wild onion were soaked individually after complete drying, in water in 1:20 (w/v) for 24 hrs. Distilled water was used as a control. Fifty seeds of chickpea were placed in each Petri dish, replicated four times. The seeds germinated were taken out from the Petri dishes and counted daily during 12 days. Twenty seeds of chickpea were sown in sand in each Petri dish to record the length and weight of root and shoot 18 days after sowing. Twenty seeds of chickpea were sown in each of controlled (normal soil) and soil taken from an *Asphodelus tenuifolius* infested field, replicated three times. The maximum time taken to mean germination was recorded in stem- and fruit extracts. Fruit extract caused the maximum reduction in germination index and germination percentage of chickpea. Different wild onion extracts significantly reduced the root, shoot length and biomass of chickpea seedlings when compared with distilled water. Reduction in root length was 26 to 38% and in shoot length were 22 to 31%. Reduction in biomass of chickpea seedlings ranged from 33 to 37 %. The maximum reduction in root, shoot length and biomass of chickpea seedlings was caused by fruit extract of wild onion. Soil beneath *A. tenuifolius* plants significantly reduced the emergence, root, shoot length, shoot dry weight and plant biomass of chickpea seedlings.

Key words: Allelopathy, *Asphodels tenuifolius*, *Cicer arietinum*, germination, emergence, seedling growth, soil bioassay

Introduction

Chickpea (*Circer arietinum* L.) is a major pulse crop of rain fed areas which constitute 88% of the total chickpea area in Pakistan. It is planted as a sole crop with zero tillage and without application of fertilizers in sandy soils in a chickpea-chickpea cropping system. Its average yield is 696 kg ha^{-1} (Anonymous, 2005) which is much lower than its potential yield. Heavy weed infestation is among several factors, which contribute for its poor yield. In Pakistan, 24-63% yield losses due to weeds in chickpea have been reported (Masood, 1993; Tanveer *et al*. 1998).

Weeds affect the crops by competing with them for different environmental recourses as well as by release of allelochemicals. When these effects occur concomitantly, the harm caused by weeds becomes even greater. Allelopathy was implicated by Suseelamma and Raju (1994), Kiemnec *et al*. (2002), Acciaresi and Asenjo (2003) and Kadioglue *et al*. (2005) when they showed water extracts of different weeds inhibited germination, root and shoot length and dry weight of different crops. Weeds may have inhibitory or promotive effects on germination and growth of crop plants (Qasem, 1993a; Alam and Islam, 2002; Kadioglue *et al*. 2005).

Different weed species vary in their allelopathic effects on germination and growth of field crops (Qasem, 1993; Hamayun *et al*. 2005). All plant parts including leaves, stems, roots, seeds and fruits (Horsley, 1977; Friedman *et al*. 1982; Kumari and Kohli, 1987; Suseelamma and Raju, 1992; 1994; Rajangram *et al*. 1992) have allelopathic potential. Different parts of the same weed also vary in their allelopathic effect on germination and growth of crops. Some plant parts are more inhibitory than others (Rehman *et al*. 1992; Burhan and Shaukat, 1999). Soil taken from *Dicanthium annulatum* (Dirvi and Hussain, 1979), *Eragrostis poaeoides* (Hussain *et al*. 1984), *Oxalis corniculata* (Chughtai *et al*. 1985) and *Imperata cylindrica* (Hussain *et al*. 1992) has also been reported to retard the germination, radicle and plumule growth of different field crops.

Wild onion (*Asphodelus tenuifolius*) is an obnoxious weed in rain fed areas of Pakistan. It is known to have allelopathic effects on germination and seedling growth of different crops (Mishra *et al*. 2001). The present study was designed to evaluate the allelopathic effect of root-, shoot-, leaf- and fruit extracts and the soil underneath *A. tenuifolius* on germination and seedling growth of chickpea.

Materials and Methods

Plants of field grown *Asphodelus tenuifolius* were uprooted at maturity and dried at room temperature. Plants were separated into roots, stems and fruits which were cut into small pieces with a scissor. The dried plant parts were immersed in distilled water for 24 hrs at room temperature in the ratio of 1:20 w/v. The aqueous extract of different plant parts was obtained. Three independent experiments were conducted with root-, stem- and fruit extracts of *A. tenuifolius* in laboratory whereas distilled water was used as control. Treatments were arranged in a completely randomized design with four replications.

Experiment No. 1

In this experiment fifty seeds of chickpea were soaked separately in extracts of different plant parts of *A. tenuifolius* for 24 h. Chickpea seeds were also soaked in distilled water as a control. After 24 hrs of soaking, seeds of chickpea were placed evenly on filter paper in Petri dishes. The Petri dishes were kept in a germinator at 15^{o}C. Subsequently distilled water was added to all treatments.

Experiment No. 2

In this experiment, seeds of chickpea were placed evenly on filter paper in Petri dishes. Three ml of extract/distilled water according to the treatment was added on the first day. Subsequently, extract and distilled water was added when needed. The Petri dishes were placed in a germinator at 15^{o}C. Germination of chickpea was observed daily according to the method of the Association of Official Seed Analysis (1990). The time to obtain 50% germination (T_{50}) was calculated according to the formula of Coolbear *et al.* (1983) as modified by Farooq *et al.* (2004). The germination index and mean germination time were calculated as described by the Association of Official Seed Analysis (1990) and equation of Ellis and Roberts (1981), respectively.

Experiment No. 3

In this experiment Petri dishes were filled with sand. Twenty seeds of chickpea were placed in each Petri dish. Respective extract was added to each Petri dish. Petri dishes were placed in a germinator at 15^{o}C for a period of 18 days. During this period, extract was added to respective Petri dishes according to requirement. Root and shoot length of seedlings was measured after 18 days. Seedlings were separated into roots and shoots and oven dried at 65^{o}C till a constant weight was obtained.

Experiment No. 4

Soil was collected to a depth of 10 cm from a plot where *A. tenuifolius* had been grown for the last two years (test) and a plot that did not have plants grown on it (control). Soil was dried at room temperature and sieved through a 2 mm mesh. Ten grams each of test and control soil was uniformly spread in 9 cm diameter Petri dishes, separately. Ten seeds of chickpea were placed uniformly on soil. Seeds were covered with the same soil as spread underneath. Soil was adequately moistened with distilled water. The dishes were incubated at 15°C. Each treatment was replicated four times. The emergence and growth of seedlings were recorded 20 days after sowing.

The data recorded were analyzed statistically by using Fisher's analysis of variance technique and LSD (p=0.05) was used to compare treatment means (Steel *et al.* 1997).

Results

Results presented in Table 1 reveal that water extracts of root, stem and fruit of *A. tenuifolius* significantly affected the time taken for 50% germination, mean germination time, germination index and germination percentage of chickpea.

Table 1. Germination of chickpea seed soaked in extract of *Asphodelus tenuifolius*

Treatment	Time taken for 50% germination (days)	Mean germination time(days)	Germination index	Germination percentage
Distilled water (control)	2.803 c	3.515 d	12.04 a	81.00 b
Root extract	3.955 b	5.097 a	9.802 b	85.00 a
Stem extract	4.220 ab	4.655 c	8.262 d	72.00 c
Fruit extract	4.318 a	4.997 b	8.673 c	72.75 c
LSD (p=0.05)	0.3479	0.04872	0.2179	7.588

Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Chickpea seeds took significantly more time for 50% germination and complete germination when soaked in root, stem and fruit extract of *A. tenuifolius* than when soaked in distilled water. Seeds soaked in root extract took less time for 50% germination but more time for mean germination than those soaked in stem and fruit extracts of *A. tenuifolius*. Root, stem and fruit extracts significantly decreased the germination index of chickpea compared with distilled water, maximum reduction being in stem extract. Stem and fruit extract of *A. tenuifolius* reduced germination percentage of chickpea seed but root extract increased the germination percentage compared with distilled water.

Table 2 shows that continuous use of extracts of different parts of *A. tenuifolius* increased the time for 50% germination as compared with distilled water but a significant increase was recorded with stem extract.

Table 2. Effect of continuous application of *Asphodelus tenuifolius* extract on germination of chickpea

Treatment	Time taken for 50% germination (days)	Mean germination time (days)	Germination index	Germination percentage
Distilled water (control)	4.128 b	4.887 b	9.810 a	90.50 bc
Root extract	4.250 b	4.773 c	9.670 a	93.50 a
Stem extract	4.810 a	5.122 a	9.408 b	91.00 ab
Fruit extract	4.210 b	5.057 a	9.255 b	88.00 c
LSD (p=0.05)	0.3043	0.1089	0.1688	2.742

Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Stem and fruit extract of *A. tenuifolius* increased but root extract decreased the mean germination time of chickpea compared to control. Germination index of chickpea was significantly decreased with stem and fruit extract of *A. tenuifolius* but was not significantly different in root extract and distilled water. Fruit extract of *A. tenuifolius* caused maximum reduction in germination percentage whereas the root extracts enhanced germination of chickpea when compared to distilled water.

Root-, stem- and fruit extracts of A. *tenuifolius* caused significant reduction in root, shoot length and dry weight of their seedlings compared with seedlings raised with distilled water (Table 3). Different extracts caused 26 to 38% reduction in root length, 22 to 31% in shoot length, 40 to 42% in root dry weight and 22 to 30% reduction in shoot dry weight over distilled water.

Table 3. Allelopathic effect of *Asphodelus tenuifolius* on seedling growth of gram

Treatment	Root length (cm)	Shoot length (cm)	Root dry weight (mg)	Shoot dry weight (mg)	Plant biomass(mg)
Distilled water (control)	23.31 a	11.03 a	44.00 a	34.50 a	78.50 a
Root extract	17.34 b (25.61)	7.565 d (31.41)	26.50 b (39.77)	25.00 b (27.53)	51.50 b (34.39)
Stem extract	14.51 c (37.75)	8.585 b (22.16)	25.50 b (42.04)	26.50 b (23.18)	52.50 b (33.12)
Fruit extract	14.36 c (38.39)	8.130 c (26.29)	25.50 b (42.04)	24.00 b (30.43)	49.50 b (36.94)
LSD (p=0.05)	2.100	0.3898	3.413	3.268	7.600

Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05). Values in parentheses show % decreases over control.

The results of the soil residual toxicity reveal the toxic nature of the soil due to leaching of toxins from *A. tenuifolius* (Table 4). Soil from the *A. tenuifolius* grown field significantly inhibited the root and shoot length, shoot dry weight, total plant biomass and emergence percentage of chickpea.

Table 4. Effect of residual toxicity of *Asphodelus tenuifolius* soil on emergence and growth of chickpea

Treatment	Emergence percentage	Root length (cm)	Shoot length (cm)	Root dry weight (mg)	Shoot dry weight (mg)	Plant biomass (mg)
Control soil	81.75 a	13.76 a	16.83 a	35.95 b	53.03 a	88.99 a
Test soil	72.50 b	9.53 b	15.99 b	38.64 a	41.40 b	79.93 b
LSD (p=0.05)	2.619	0.347	0.245	0.973	1.175	2.047

Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Discussion

Results from the experiment indicate that soaking of chickpea seed in aqueous extracts of root, stem and fruit of *A. tenuifolius* resulted in delayed germination and less germination index in comparison with distilled water. More delayed germination and less germination index with fruit extract of *A. tenuifolius* could be due to a greater inhibitory effect of allelochemicals present in fruit. These results are supported by the findings of Angiras *et al*.

(1988) who reported delayed germination of maize seed with *C. rotundus* and *E. crus-galli*. Maximum reduction in germination percentage of chickpea seed when soaked in fruit extract of *A. tenuifolius* indicates the presence of water soluble allelochemicals in maximum concentration. Noor and Khan (1994) also reported the highest reduction in *Zea mays* seed germination by *A. samans* seed extract.

Continuous application of extracts of *A. tenuifolius* appeared to have more inhibitory effects on time taken for 50% germination and mean germination time of chickpea than soaking (the maximum with stem and fruit extract). Decrease in germination index and percentage was less with continuous application than one (soaking) application. Chickpea seed either soaked in root extract or continuously supplied with root extract increased significantly the number of germinated seeds. These results are in line with those of Kadioglue *et al*. (2005). They stated that *Glycyrrhyza glabra* L., *Sorghum halapense* L. and *Reseda lutea* L. stimulated germination of chickpea at rates of 95%, 94% and 93%, respectively.

The significant inhibition of root and shoot length, and dry weight of chickpea seedling by extracts of different plant parts of *A. tenuifolius* may reflect the allelopathic potential of *A. tenuifolius* due to water soluble inhibitors (Kill and Yan, 1992). These results are supported by the findings of Suseelamma and Raju (1992, 1994) who reported inhibitory effect of extracts of stems, leaves, inflorescence and roots of *Digera muricata* on length and dry weight of root and shoot of horse gram (*Macrotyloma uniflorum*) amd root and shoot length in ground nuts (*Arachis hypogaea*). More inhibition of chickpea roots compared with shoots may be due to their more intimate contact with extract treated filter paper. Fruit and root extract caused maximum reduction in root and shoot length, respectively, which indicate that inhibition of *A. tenuifolius* differed with the plant parts. These results are in line with those of Rajangram *et al*. (1992) who observed that leaf extract of *Croton sparisflorus* was more inhibitory than stem and root extract on paddy.

It is often complicated to measure root and shoot length of a crop due to curling. Therefore, seedling dry weight may be the better criterion to study allelopathic potential of weeds on crops. Extracts of different plant parts of *A. tenuifolius* produced similar results in reducing seedling dry weight of chickpea. Decrease in emergence, root, shoot length, shoot dry weight and plant biomass of chickpea seedlings in test soil indicate that soil from the *A. tenuifolius* grown field was phytotoxic compared with controlled soil. These results are supported by Dirvi and Hussain (1979), and Hussain *et al.* (1992). They found an inhibitory effect of soil taken from *Dicanthium annulatum* and *Imperata cylindrica* fields on germination and radicle and plumule growth of different crops. Increase in root dry weight of chickpea seedlings in test soil compared with controlled soil was due to increase in thickness of roots for reasons unknown so far.

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HERBICIDE TECHNOLOGY TRANSFER IN RICE CULTIVATION

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Abstract: Transfer of new farm technology created a significant potential for increasing rice production all over the world. Agricultural productivity depends on the extent to which farmers adopt new agricultural innovations. The efforts made through various schemes increased the area and production but a variation in productivity at farm level still exists. There are number of factors that are responsible for increasing rice productivity. Several studies indicated that the use of herbicides and weed management have contributed significantly to rice yields at farm level. Most farmers stated that some of the rice technologies developed at experimental stations do not produce high yields under actual farm conditions. This is mainly due to lack of communication regarding values of the improved practices, individual factor contribution to the total yield and net return to the farmer. In view of these, an attempt was made to show the contribution of selected individual and combined production factors to the total rice yield and net return to the farmer. The study was carried using integrated demonstrations with surveys in five villages of Cuddalore District during the Samba season under irrigated conditions. Field demonstration plots were laid out variety and 300 farmers constituted the sample of the survey. Herbicide application was one of the combined production factors that contributed to the higher yields. Further, herbicide application factor gave higher yields (5059 kg/ha) in all locations and other production factors and also induced higher additional net returns (Rs. 5158/ha) to the farmer. The findings of the survey provided inputs for policy makers and administrators of the state to popularize herbicide technology and to augment productivity of rice in all the regions in the country.

Key words: Herbicide application, net return-production factors, technology transfer, yield

Introduction

Rice is the staple food of people in nearly all Asian countries and a major source of livelihood in rural economics. Transfer of New Farm Technology created a significant potential for increasing rice production all over the world. Agricultural productivity depends on the extent to which farmers adopt new agricultural innovations. Rice is the most important cereal crop is grown and consumed in most parts of India. The efforts made though various schemes increased the area and production but a variation in productivity at farm level still exists. There is a wide gap between technology generated by the scientists and the technology adopted by the farmers.

There are many factors responsible for increasing rice productivity. Weeds are one of the principle causes of low rice production, reducing the yield by 60 to 70% in India (Ghosh *et al*. 1994). Several studies indicated that use of herbicides and weed management programmes have contributed significantly to rice yields at the farm level (Clampett, 2001). Most of the farmers stated that some of the rice technologies developed at experimental stations do not produce high yields under actual farm conditions. This is mainly due to the lack of communication regarding the importance of the improved practices, individual factors contribution to the total yield and net return to the farmers. In view of these, an attempt was made in this study to determine the share of selected individual and combined production factors that contribute the total yield and to illustrate the contribution of selected production factors to the additional net return to the farmer.

Materials and Methods

The study was conducted in five villages of Cuddalore district during the Samba season (August to December) using a demonstration cum survey approach under irrigated conditions. Based on the expert opinion, six production factors such as selection of variety, purity of seed, age of seedling, and number of seedlings per square meter. *Azospirillum* application and herbicide application responsible for maximizing rice yields under farmers' field condition were selected for conducting field demonstrations to show the worthiness of these factors. Field demonstrations were laid out in the five villages in a factorial randomized block design with eight plots. The rice variety Aduthurai 38 (ADT 38) was planted in the demonstrations plots of dimensions 10 m x 8 m in each farm. The mean actual farm yield was used for calculating the contribution of selected production factors to the net return. During harvest, the grain and straw yields were recorded systematically and the data were collected with the help of a well structured, pre-tested interview schedule and suitable statistical tools were used to analyze the data.

Results and Discussion

Contribution of selected production factors to the yield

Table 1 indicates the grain yield/ha recorded at various locations as well as the mean grain yield for all the villages. The table also reveals the difference between the yield obtained by adopting different production factors and the control. Critical differences were calculated to determine the significance at 5% level. The combination of productions factors gave higher yield than both the control and the individual factors in all the five locations (Table 1)
.

Table 1. Contribution of selected production factors to the yield of ADT.38 rice cultivar in five selected villages of Cuddalore district

Demonstration Number	Grain yield (kg/ha)						Difference between the mean value of treatments and control
	Adoor	Mugaiyur	Nandi mangalam	Pinnalur	Nanjalur	Mean	
D_1	4063	4586	4263	3839	4125	4175.20	209.8
D_2	4125	4825	4475	4188	4400	4402.60	437.2
D_3	4563	5094	4813	4688	4500	4731.60	766.2
D_4	4763	5013	5025	4925	4675	4880.80	914.8
D_5	4963	4663	4800	4400	4538	4672.80	707.4
D_6	5032	5150	5125	5013	4975	5059.00	1094.6
D_7	5130	5438	5350	5238	5038	5242.80	1277.4
D_8	4013	4325	4063	3738	3688	3965.40	-

$CD_{0.05}$ = 614.13

D_1 – Selection of variety (ADT. 38 rice)
D_2 – Purity of seed (Certified seed)
D_3 – Age of seedlings (30 days old seedlings)
D_4 – Number of hills/sq.m (66 nos/m²)
D_5 – *Azospirillum* application (10 pkts/ha)
D_6 – Herbicide application (Butachlor 2.5 l/ha)
D_7 – Combined factor
D_8 – Control

An individual production factor, namely, 'Herbicide application' (D_6) produced higher yields at all the locations compared to other individual production factors. The application of weedicides after transplanting resulting in weed free conditions might have led to higher yields in all the locations. This observation derives support from Singh and Bhan (1989)

Arulchezhian and Kathiresan (1990) who reported similar findings on performance of different rice varieties influenced by different weed control methods.

An ideal plant population (D_4) was found to be the second most important production factor at Nandimangalam (5025 kg/ha), Pinnalur (4925 kg/ha) and Najalur (4675 kg/ha) villages. It is obvious that maintenance of adequate plant population during the crop period in the above locations led to higher yield. This finding is in agreement with the findings of Palaniappan and Balasubramanian (1991) who reported that maintenance of adequate plant population is essential for obtaining higher yield in rice.

The third production factor, namely, age of seedling (D_3) was responsible for a yield of 5094 kg/ha at Mugaiyur village. *Azospirillum* application (D_5) gave a yield of 4963 kg/ha at Adoor location. Younger seedlings establish better and biofertilisers compliment inorganic and organic sources. This observation derives support from Thyagarajan (1996) who reported that transplanting with younger seedlings and soil inoculation with *Azospirillum* in rice fields is essential for obtaining higher yields in rice.

From the above results, it may be inferred that adoption of improved production factors namely, selection of variety (D_1), purity of seeds (D_2), age of seedling (D_3), number of hills per square metre (D_4), *Azospirillum* application (D_5) and herbicide application (D_6) as a package helped in increasing the yield as the combined factors (D_7) recorded the maximum grain yield (5242.8 kg/ha) over the control (3965.4 kg/ha). This may be due to fact that all the individual production factors complement each other leading to maximum yield than the control.

When individual factors are considered, the 'Herbicide application' (D_6) recorded the maximum yield (5059 kg /ha) resulting in a difference of 1094.6 kg/ha over the control which is significant at 5% probability. It could be observed that this factor is relatively more important than other individual factors, because the rice crop in this area is affected by weed infestation. The competition of weed resulted in severe reduction of yield of rice. These findings are in conformity with the findings of Purushothaman and Hosamani (1990).
The next important individual factor is 'the number of hills per square meter' (D_4). The mean grain yield obtained by this demonstration is 4880.2 kg/ha which is higher than the control (3965.40 kg.ha). The difference of 914.85 kg/ha is significant at 5% probability. The optimum number of panicles per square meter is obtained by having recommended number of hills per square meter. This favors uniformity of seeds which helps to achieve the maximum yield compared to control (Pathak *et al*. 1999).

The third important individual factor is the 'age of seedling' (D_3). The mean grain yield obtained by this demonstration is 4731.6 kg/ha which is higher than the control (3965.4 kg/ha). The difference of 766.2 kg/ha is significant ($p<0.05$). The maximum yield per unit area is obtained by planting younger seedling. This favors good establishment of crop during the entire growth period and it leads to better uptake of nutrients which helps to achieve more yield compared to control.

The fourth important individual factor is '*Azospirillum* application' (D_5). The mean grain yield obtained by this demonstration is 4672.8 kg/ha which is higher than the control (3965.40kg/ha) resulting in a difference of 707.4 kg/ha, which is significant ($p<0.05$). It could be observed that *Azospirillum* application also proved relatively more important than the control, because the applied microorganisms would induce the root nodules of the rice crop that leads to higher yields. This derives support from Kandasamy *et al*. (1991) who also reported that the different rice cultivars planted in the *Azospirillum* applied fields gave maximum grain yield than the control.

The other two factors namely 'purity of seed' (4402.6 kg/ha) and 'selection of variety' (4175.2 kg/ha) recorded higher yield but were not significant, resulting in differences of 437.2 kg and 209.8 kg, respectively, over the control. Thus, the demonstration conducted in all the locations either with individual production factor or combined factors gave higher yield than the control.

Contribution of selected production factors to the net return

Table 2 presents the additional net return per hectare obtained by the respondents at different demonstration plots with selected production factors.

Table 2. Contribution of selected production factors to the additional net return obtained by the respondents

(in Rupees)

Name of the demonstration	Additional cost involved /ha	Additional gross return/ha	Additional net return/ha
Selection of variety	-	1049.00	1049.00
Purity of seed	450.00	2186.00	1736.00
Age of seedling	750.00	3831.00	3081.00
Number of hills per sq. Met	900.00	4574.25	3674.25
Azospirillum application	60.00	3537.00	3477.00
Herbicide applications	330.00	5468.00	5138.00
Combined factor	990.00	6387.00	5397.00

The mean of the actual farm yield (*i.e.* 3965.40 kg/ha) was used for the purpose of calculating the contribution of selected production factors towards additional net return. The combination of production factors gave the highest additional net return of Rs. 5397.00 when compared to individual production factors. All the factors in combination gave the highest yield due to their complementary thereby providing additional net return. Among the individual factors, herbicide application, age of seedling, number of hills per square meter and Azospirillum application gave higher additional net return per hectare. The application of herbicide after transplanting resulting in a weed free environment and the number of hills per square meter determines the plant population and the age of seedling determines the vigor of the plant. These obviously contributed to higher yield. The finding derives support from the findings of Purushothaman and Hosamani (1990).

Conclusion

The study revealed that herbicide application is one of the combined production factors that contribute to the higher yield in all the locations. Hence, that more number of demonstrations with the above production factors organized at village level by the extension workers to educate the farmers about the worthiness of selected production factors is recommended and thereby to increase their adoption level As the demonstration is working on the principle “Seeing is Believing” and ‘Learning by Doing’ it would serve as a powerful tool in understanding these factors in a complete form. Further, herbicide application factor gave higher yield when compared to the control and other production factors and also gave higher additional net return to the farmer. Further it is suggested, that concerned departments need to formulate appropriate information and communication strategies to popularize the herbicide application technology to all the regions in the country and to increase the rice productivity in a sustainable manner.

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INFLUENCE OF FARM LITTER ON WEED CONTROL AND YIELD OF CHICKPEA

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Abstract: This study was conducted with the objective of determining the effects of different farm litter/covering materials as mulches on weed control and seed yield in rain fed chickpea. Neem (*Azadiracta indica* A. Juss) tree litter applied at 5000 kg/ha suppressed density of common lambs quarters, purple nut sedge, and scarlet pimpernel by providing a weed smothering efficiency (WSE) of 45.7% and enhancing 76% seed yield (1610 kg/ha) of chickpea in comparison of non treated check (910 kg/ha). Transparent/black polythene sheet strips as mulches proved to be better with regards to weed suppression (57.7% WSE) and producing 116% more yield (1960 kg/ha) of chickpea seeds than of non treated check. Other covering material such as pigeon pea (*Cajanus cajan* L.), mango (*Mangifera indica* L.) tree litter, or rice straw also reduced weeds and enhanced chickpea yield. Among yield components, seed weight was reduced by weed infestation. The density and biomass of weeds declined with progressive increase in duration of the cover. Covering duration for 60 days proved significantly better than that of 30 days.

Key words: Covering materials, farm litters, farm wastes, mulches, tree litters, weed management

Introduction

Chickpea (*Cicer arietinum* L.) is one of the most widespread crops grown under rainfed conditions in India. Its yield is seriously reduced by accompanying weeds. Common lambs quarters (*Chenopodium album* L.), is one of the most common weeds found not only in chickpea but in most of the crops grown in winter in India and also in other South Asian countries. There are several farm wastes or material available with common farmers which could be utilized in a useful manner for enhancing productivity of a crop particularly when grown under rain fed conditions. These materials, if applied to the crop at planting reduces evaporation losses and do not allow the weed seeds to germinate.

Considerable evidence exists to show that weed populations are controlled with wheat straw in maize (Crutchfield *et al.* 1986). Singh *et al.* (1996) found that application of a grass cover at 6 t/ha resulted in significantly higher seed yield of mustard (*B. juncea*) than by other covers. Similarly, Sachan *et al.* (1997) found a significant increase in yield of rainfed mustard when paddy straw was used as mulch while Gupta *et al.* (1994) showed that seed yield of mungbean, (*V. radiata*) was higher with straw mulch than with a polythene mulch. Hepperly and Diaz (1983) reported that pigeon pea (*Cajanus cajan*) leaf litter significantly controlled the weed population for two months. Some tree litters from *Leucaena leucocephala* have been found to check the growth of a prominent weed - *Ageratum conyzoides*, probably because of the presence of mimosine, quercetin and 8 phytotoxic phenolics in its leaves (Chou and Kuo, 1987). Likewise, Jobidon (1986) investigated the allelopathic potential of weeds and their extract and found strong inhibitory effects on germination and growth of grass seedlings. Yaduraju and Ahuja (1990) reported 67% reduction in grass weed population by mulch of wet soil for 30 days. Similarly, Ghorai and Bera (1998) have also stated the beneficial effects of mulches. However, more information is necessary to ensure that use of such materials, provide a viable and sustainable option for farmers growing crops under dry conditions in India. With this in mind, a study was undertaken to determine if covering materials would suppress weed growth, reduce crop weed interference, and enhance yield of chickpea so that herbicide use could be reduced.

.Materials and Methods

The experiment was conducted at the experimental farm of Indian Institute of Pulses Research, Kanpur, India during 1995-97. The soil was sandy loam, poor in organic C (0.34 %), medium in available P (15.7 kg/ha) and available K (139.8 kg/ha). The soil pH was 7.4. Total rainfall received during the cropping season was 48.5 and 26.9 mm in 1995-96 and 1996-97, respectively. The treatment combinations comprised of seven mulches viz transparent polythene (0.4 mm thick), black polythene (0.4 mm thick), leaves of mango, *neem*, pigeon pea and rice straw each at 5000 kg/ha and a check (no mulch). The covers were applied for 30, 60 and 90 day durations. Treatments were arranged in a randomized block design with three replications. The plot size was 4.0 x 3.6 m^2.

Polythene strips and air-dried leaves/straw were spread in inter and intra row spaces immediately after emergence of chickpea. The experimental field was prepared by harrowing twice with a disc harrow followed by two cultivations with a tractor drawn cultivator. Planking (wood plank behind disc to level the field) was done after last tilling operation. Recommended dose of fertilizers (18:46:0 kg N: P: K/ha) for chickpea was applied at the time of sowing in the form of diammonium phosphate fertilizer. Sowing was done @ 80 kg/ha with a hand hoe by opening the furrow at 30 cm apart and placing the seeds at 10 cm in 3rd week of November and last week of October in 1995 and 1996, respectively. The harvesting of chickpea was done manually in the first week of April during both the years. Above ground weeds were clipped manually after placing a quadrat of 0.25 m^2 randomly at two places in each plot when common lambsquarters reached the inflorescence stage. The weed biomass was grouped and weighed by species in three category *i.e.* Common lambsquarters, purple nutsedge, and scarlet pimpernel. The weed smothering efficiency (WSE) was computed by following formula:

$$\text{WSE (\%)} = \frac{\text{Weed biomass in weedy check - Weed biomass in particular treatment}}{\text{Weed biomass in weedy check}}$$

Weed density data were subjected to square root transformation before statistical analysis to reduce the influence of non-normal data distributions. Statistical significance of treatment differences was detected by ‘F’ test at 5% level (Fisher, 1952).

Results and Discussion

Effect of covering materials on weed dynamics

Common lambs quarters (Chenopodium album L.): Covering materials influenced density and biomass of common lambsquarters infestation at the time of crop harvest (Table 1). Neem leaves applied plots had 8.4 and 7.1/m^2 common lambs quarters density and 35.06 and 28.30 g/m^2 biomass in comparison to 12.9 and 12.25 /m^2 density and 62.25 and 63.30 g/m^2 biomass in untreated check in first and second year, respectively. Neem leaves reduced mean density of common lambsquarters by 62.4% followed by pigeonpea, mango and straw leaves (58.7, 55.7 and 47.5% respectively) in comparison to 67.8% by black polythene strips. Similarly, a reduction in biomass was observed (49.5, 47.5, 41.9 and 34.9 due to neem, pigeonpea, mango and straw leaves, respectively, in comparison to 60.0 and 61.4% by the black and transparent polythene strips. Transparent polythene strips suppressed population of common lambsquarters by 70 and 80% in first and second year, respectively. No significant variation was observed in transparent or black polythene strips in this respect.

Table 1. Effect of covering materials and their duration on weed density and biomass of common lambs quarters, purple nut sedge and scarlet pimpernel over two years.

Treatment	common lambsquarters Density (plant/m^2)		common lambsquarters Biomass (g/m^2)		purple nutsedge Density (plant/m^2)		purple nutsedge Biomass (g/m^2)		scarlet pimpernel Density (plant/m^2)		scarlet pimpernel Biomass (g/m^2)	
Year	I	II	I	II	I	II	I	II	I	II	I	II
Covering materials												
Trans. polythene	7.10	5.50	26.30	22.20	6.36	6.00	9.93	9.35	6.50	6.02	22.05	19.00
	(70.0)*	(80.0)	(57.8)	(64.9)	(52.4)	(64.3)	(29.1)	(24.7)	(61.8)	(65.0)	(54.4)	(54.9)
Black polythene	7.90	6.45	27.22	22.90	7.35	6.90	11.32	10.00	6.98	6.02	23.77	19.22
	(62.9)	(72.7)	(56.3)	(63.8)	(35.7)	(51.0)	(19.1)	(19.5)	(56.4)	(65.0)	(50.8)	(54.4)
Mango leaves	8.85	7.95	38.50	34.45	7.35	8.00	11.45	11.25	8.40	7.25	32.86	24.45
	(53.3)	(58.0)	(38.2)	(45.6)	(35.7)	(33.7)	(18.2)	(9.4)	(36.4)	(49.5)	(32.0)	(41.9)
Neem leaves	8.40	7.10	35.06	28.30	7.78	7.38	12.12	11.20	6.96	6.80	24.00	23.77
	(58.1)	(66.7)	(43.7)	(55.3)	(28.6)	(44.9)	(13.4)	(9.8)	(56.4)	(55.3)	(50.4)	(43.6)
Pigeonpea leaves	8.85	7.35	36.90	28.95	7.90	7.78	12.25	11.28	8.15	6.80	29.90	24.05
	(53.3)	(64.0)	(40.7)	(54.3)	(26.2)	(38.8)	(12.5)	(9.2)	(40.0)	(56.3)	(38.2)	(42.9)
Rice straw	8.88	9.12	41.42	40.38	8.20	8.50	12.70	11.83	8.38	8.25	34.10	30.14
	(50.3)	(44.7)	(33.5)	(36.2)	(20.2)	(26.5)	(9.3)	(4.8)	(36.4)	(34.0)	(29.5)	(28.4)
No cover	12.90	12.25	62.25	63.30	9.20	9.90	14.00	12.42	10.50	10.15	48.35	42.11
LSD (p=0.05)	2.30	2.13	3.42	3.48	1.62	1.47	1.83	1.87	1.74	1.82	3.34	3.10
Covering period (days)												
30	11.66	11.02	55.50	58.45	9.61	9.20	13.44	11.88	10.17	9.35	45.21	32.02
60	8.63	7.10	31.48	23.34	6.50	6.18	9.85	9.25	7.25	6.67	24.97	23.00
	(45.6)**	(58.7)	(43.3)	(60.1)	(54.3)	(54.8)	(26.7)	(22.1)	(49.5)	(49.4)	(44.8)	(28.2)
90	6.52	5.61	27.75	21.26	5.52	4.73	8.63	8.90	6.29	5.96	21.95	19.32
	(69.1)	(74.4)	(50.0)	(63.6)	(67.4)	(73.8)	(35.8)	(25.1)	(62.1)	(59.8)	(51.4)	(39.7)
LSD (p=0.05)	2.13	1.97	3.48	3.54	1.80	1.68	1.95	1.90	1.66	1.63	3.22	3.42

*Per cent decline in population and biomass in comparison of untreated check

** Per cent decline in population and biomass in comparison of covering up to 30 days

Purple nutsedge (Cyperus rotundus L.): All covering materials significantly influenced the density and biomass of purple nutsedge by 4.8-64.30% during both the years, the effects being less in second year (Table 1). Amongst the farm litter, *neem* leaves recorded 7.78, 7.38 density/m^2 and, 12.12, 11.20 g/m^2 biomass in comparison to 9.2, 9.9 density/m^2, and 14.00,12.42g/m^2 biomass in uncovered plots in first and second year respectively. The maximum reduction in the mean density of this weed was 36.8% with *neem* leaves followed by 34.7, 32.5, and 17.9% in mango, pigeonpea leaves, and straw cover, respectively in comparison to 43.4% by black polythene. Transparent polythene provided 52.4, 64.3% less density, and 29.0, 24.7 % less biomass, than of uncovered plots in first and second year, respectively and was equivalent to that of black polythene covers.

Scarlet pimpernel (Anagallis arvensis L.): Different covering materials influenced scarlet pimpernel infestation (Table 1). Amongst the farm litter, *neem* leaves reduced mean density and biomass by 55.9 and 47.0% in comparison to the control uncovered treatment. Other farm litter (mango, pigeonpea and rice straw) reduced mean density by 42.3, 47.7, 35.1%, respectively and mean biomass by 37.0, 40.6 and 29.0% respectively when compared to 55.9 and 47.0 % by *neem* leaves. No significant variation was observed in either black or transparent polythene covers in respect to density and biomass of scarlet pimpernel

Transparent polythene had 61.8 and 65.0% less density and 54.4 and 54.9% less biomass than of the control (no cover) in first and second year, respectively.

Effect of covering duration on weed dynamics
Density and biomass of lambs quarters (Table 1) was minimum (6.52 and 5.61m^2) and (27.75 and 21.26 g/ m^2) when plots were covered for which was similar to the 60 day period but significantly superior over 30 day period. In purple nutsedge (Table 1), there was no significant variation in density and biomass of nutsedge with 60 and 90 days duration of cover in both the years. However, 60 days covering period significantly reduced density and biomass of nutsedge in comparison to 30 days in both the years. In scarlet pimpernel (Table 1), a cover for 90 days significantly lowered density (6.29 and 5.96/m^2) and biomass (21.95 and 19.32 g/m^2) as compared with 30 and 60 days cover during both the seasons. However, the differences in density and biomass between 90 and 60 days duration were non- significant.

Effects of covering materials on seed yield and yield components
Different covering materials and their duration influenced the yield of chickpea seeds significantly (Table 2). Transparent polythene and black polythene helped to attain maximum yields in comparison to the untreated check in both the years. *Neem* tree litter cover also enhanced seed yield of chickpea over the untreated check. Yields in plots with other tree litters were similar to that of the *neem* tree litter plots. Among the yield components (Table 2), number of pods/plant and 100 seed weight were significantly influenced by different covering materials.

Table 2. Effect of covering materials and their duration on seed yield and yield components of chickpea

Treatments	No. of pods/plant		No. of seeds/pod		Weight of 100 seeds (g)		Seed yield of chickpea (Kg/ha)		
Year	I	II	I	II	I	II	I	II	Mean
Covering materials									
Trans. Polythene	55.4	56.1	1.8	1.9	23.54	24.05	1900	2030	1960
Black polythene	50.9	50.8	1.7	1.8	21.92	22.00	1730	1990	1860
Mango leaves	37.9	44.2	1.6	1.7	17.78	18.04	1450	1550	1500
Neem leaves	46.4	48.0	1.7	1.7	18.75	20.12	1610	1610	1610
Pigeon pea leaves	41.3	45.3	1.7	1.7	18.05	19.35	1400	1580	1490
Rice/wheat straw	36.2	37.1	1.5	1.6	16.92	17.45	1190	1490	1340
No cover	31.7	32.3	1.5	1.5	15.15	15.08	840	980	910
LSD (p=0.05)	4.9	5.0	NS	NS	2.83	2.89	410	400	-
Covering period (days)									
30	38.3	37.9	1.6	1.7	19.72	21.30	1310	1470	1390
60	48.9	47.8	1.7	1.7	22.63	23.85	1430	1820	1630
90	50.0	50.3	1.7	1.8	23.00	23.94	1690	1980	1840
LSD (p=0.05)	4.9	4.8	NS	NS	2.73	2.76	340	360	-

Transparent polythene increased the number of pods/ plant by 42.6% and 100 seed weight by 36.5% when compared to plants in plots with no cover. There was no significant variation in number of pods /plant and 100 - seed weight due to black polythene and *neem* tree litter. Greater control of weeds by polythene cover not only increased the number of pods/plant and the seed weight but also enhanced the mean yield by 105.5% over that of plots without cover.

Neem leaves cover also increased the number of pods/plant and 100 - seed weight by 48.0 and 28.6% resulting in an overall increase in seed yield by 77.6% over the control respectively. These findings are in accord with Panwar and Malik (1996) who found that black and white polythene were effective covers for reducing the population of carpet weed and barnyard grass over weedy plots in cotton. Low density and biomass of weeds under transparent and black polythene might be due to better transmittance/ impregnation of heat. This may raise soil temperature just beneath the polythene strips, causing poor emergence, decay and death of new seedlings. Greater efficacy of weed control by tree litters or straw might be attributed to allelopathic effects.

There was no significant variation in 60 and 90 day covers, however, 60 days period caused a significant increase in number of pods/ plant and 100 - seed weight over 30 day durations. The low density and biomass of lambsquarters and other weeds with increase in duration of cover might be due to intense heating of soil up to 60 days affecting the germination and growth of weeds. Beyond 60 days there was no further damage to the germination and growth of new weed recruits.

It may be concluded from the above study that covers when applied in rainfed crop of chickpea as mulches reduce crop weed competition and increase the yields. Thus harnessing of farm wastes or tree litters such as *neem* leaves and others can be used to suppress weeds and increase yield without undue costs involved.

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INVESTIGATION ON WEEDS AS SYMPTOMLESS CARRIERS OF *Fusarium oxysporum* f.sp. *cubense* OF BANANA

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Abstract: Panama wilt caused by *Fusarium oxysporum* f.sp. *cubense* (FOC) is one of the major diseases in banana plantations throughout the world. Due to the survival of *Fusarium oxysporum* f.sp. *cubense* in various ways in banana fields, the crop cannot be economically grown. This study was undertaken to identify weed species associated with FOC in banana fields in southern Sri Lanka. Eleven banana fields were randomly selected and weed samples were collected from each field and brought in to the laboratory. Pathogens were isolated and identified using laboratory tests. Seventy four weed species were recorded in the eleven banana fields surveyed, where *Euphorbia heterophylla, Eclipta prostrata, E. hirta, Vernonia cinerea, Echinochloa* spp. were the most abundant species. Eleven *Fusarium oxysporum* isolates and six non-*Fusarium* species (*Curvularia* sp., *Trichoderma* sp., *Mucor* sp., *Backusella* sp., *Chaetomium* sp., and *Rhizoctonia* sp.) recorded and found to be associated with these weed species. The pathogenicity test proved none of the above *Fusarium* isolates of weeds was able to cause panama wilt symptoms on banana.

Key words: Banana, *Fusarium oxysporum* f.sp. *cubense,* panama wilt, weeds

Introduction

Banana (*Musa* spp) is considered world's most important fruit crop and it has also been identified as a top priority fruit crop in Sri Lanka. Panama wilt caused by *Fusarium oxysporum* f.sp. *cubense* (FOC) is one of the major diseases in banana plantations throughout the world. Due to the survival of *F. oxysporum* f.sp. *cubense* in soil and other sources in banana fields, the crop can not be economically grown (Ploetz, 1994). Weeds are considered as host for many *F. oxysporum* pathogens throughout the world and these weed host plants could be appeared without any typical symptoms of those diseases. No attempts have been made to study in detail about association of weed flora as careers of panama wilt pathogen. Such study is important to get further details about the survival of *F. oxysporum* f.sp *cubense* (FOC) in weeds, and thus further helps to provide information for effective management practices for increased production. The major objective of this study was to identify weed species associated with *F. oxysporum* f.sp *cubense* (FOC) in banana fields in southern Sri Lanka.

Materials and Methods

Eleven banana cultivated sites were randomly selected in southern region representing, three sites at Emblipitiya, two sites at Kiri Ibban Ara, three sites at Suriyawawe and three sites at Barawakumbuka.

Collection of weed samples
Weed samples were collected from randomly selected fields as the Z pattern (Witharama *et al.* 1997). Four locations were sampled in each arm of the Z pattern-giving a total of 12 locations. The number of individuals of each weed species were recorded and counted separately in a 50 cm x 50 cm ($0.25m^2$) quadrate placed randomly at each location. Each species of weeds was

tagged separately. Samples were brought in to the laboratory for isolation of fungi. The frequency of weed flora was determined using the below given qualitative measure (Thomas, 1985).

$$F_k = \frac{\sum^n Y_i \times 100}{n}$$

where, F_k = Frequency value of species K
Y_i = Presence (1) or absence (0) of species K in the field *i*
n = Number of field sites surveyed

Isolation of the pathogen

Weed samples were collected from these fields were brought in to the laboratory for identification of fungal pathogens. The root system was separated from the collected plants and washed thoroughly with water. Those roots were cut in to small pieces and kept in 1% Clorox for two minutes. Root pieces were then washed by distilled water and allowed to dry on filter papers. Surface sterilized root parts were placed on Pentachloronitronenzine (PCNB) medium. Four pieces of roots were placed in each Petri dish and incubated for growth. The fungus was sub cultured in potato sucrose agar (PSA) medium for further growth. Water agar (WA) medium was used for single spore culture technique. Spores on PSA medium was taken in to a needle and streak on water agar. After 8 hrs of growth, single spore was transferred to a PSA medium and allowed to grow. After 10-20 days these cultures were used for identification.

Pathogenicity studies

Healthy banana seedlings of 2 ½ to 3 months old growing in pots of methyl bromide fumigated soils were inoculated by digging a small trench of 0.5 inches from the base of the rhizome. *Fusarium* isolates were earlier maintained in 10% corn meal in sand. Two grams of a 2 week old 10% corn meal in sand *Fusarium* cultures were placed in the trench and the soil replaced. Inoculated pots were placed in green house and the after care operations were carried out regularly. Rhizomes were dug and examined one month after inoculation to observe symptoms.

Results and Discussion

The relative distribution of individual weed species in the region was recorded under different survey sites. Table 1 provides comprehensive list of the species of weeds found in banana fields in the area. According to the table, seventy-four weed species were recorded in eleven banana fields at four locations in the southern region. This indicates that there was wide diversity of weed flora within the study area. Among these weeds, *Euphorbia heterophylla, Eclipta prostrata, Euphorbia hirta* and *Echinochloa colonum* found in almost all banana fields surveyed. Thirty nine weed species occurred at or above the frequency value of 50% and observed in most lands. Therefore, these can be considered as common and important weed species of banana in the area. A total of 22 species had a frequency value of less than 25% while, 14 species occurred in between the frequency values of 25% and 50% (Table 1). The comparatively higher frequency values recorded by a few weed species indicates that the weed flora at the time of sampling was dominated by a few weed species. Therefore, the presence of weeds in abundance in the cropping area leads to crop-weed competition resulting in yield losses.

Table 1. Frequency of occurrence of different weed species in banana fields

Scientific name	Common name	Local name	Frequency of occurrence (%)
Euphorbia heterophylla	Milk weed	Wal Rubber	100.00
Ageratum conyzoides	Billy goat weed	Hulanthala	81.80
Cyperus rotundes	Purple Nut-sedge	Kalanduru	9.00
Sida cordifolia		Walbabila	45.40
Cenchrus echinatus	Sandbur	Kuwani grass	18.18
Ipomoea spp			45.45
Cleome rutidosperma			18.18
Phylanthus niruri		Pitawakka	72.72
Commelina indica		Girapala	36.36
Eclipta prostrata	White celipta	Kikiridiya	100.00
Euphorbia hirta			100.00
Cynodon spp.	Bermuda grass	Heen-Atora	36.36
Ocimum sanctum	Holly basil	Maduruthala	9.00
Ludwigia octovalvis		Diyakumbuk	63.63
Panicum maximum	Guniea grass	Guni thana	18.18
Eleutheranthera ruderalis			18.18
Minconta scandens		Wathupalu	27.27
Agyreia popalifolia		Grithilla	36.36
Imperata cylindrica	Lalany grass	Illuk	18.18
Mimosa pudica		Nidikumba	36.36
Vernonia cinerea	Sensitive plant	Monorakudumbiya	90.90
Echinochloa colonum		Girithana	100.00
Hediyotis auriculata			18.18
Emilia javanica		Kadupahara	63.63
Tridax procumbense	Tridax daisy	Wal-kapuru	54.54
Aeschynomene sp.	Joint vetches	Diyasiyabala	36.36
Paspalum conjugatum			36.36
Ipomoea pestigridis			18.18
Scoparia dulcis	Scoporia weed	Wal-kotthamalli	27.27
Fimbristylis globulosa			18.18
Acanthacea spp.			9.00
Fimbristylis dichotoma			18.18
Ipomoea aquatica		Kankun	45.45
Chromolaena odorata			18.18
Leptochoa filiformis			9.00
Eleusina indica			9.00
Sida acuta			18.18
Axonopus sp.			9.00
Boerhavia repande	Tar wine	Sarana	9.00
Panicum repense		Yapala	9.00
Amaranthus spinosus	Needle	Katuthampala	9.00
Axonopus sp .		Heen pothuthana	18.18
Calotropis spp.		Wara	9.00
Acalipa indica		Kuppamania	9.00
Mimosa invisa	Giant sensitiveplant	Mahanidikumba	9.00
Jacquemontia scandwicesis	Pau-o-hiaka	Wal-kollu	18.18
Mitracarpus hirtus	Small square weed	Gatakola	9.00
Brachiaria mutica	Para grass		27.27
Dactyloctenium aegyptium	Crowfoot grass		18.18
Borreria hispida		Galkura	18.18
Abutilon indicum		Wal-Anoda	9.90

Desmodium triflorus	Lanter flower	Heen-Udupiyaliya	45.45
Brachiaria decombense			9.00
Dicanthium aristatum	Signa grass		18.18
Gloriosa superba	Angleton grass	Niyagala	9.90
Aerva lanata		Polpala	18.18
Paspalum notatum			9.90
Tephrosia purpurea	Dallis grass	Pila	9.00
Galinsoga parviflora			9.00
Paspalum urville	Galin soga		18.18
Cucumis callosus	Vasey grass	Gonkakiri	18.18
Stachytarpheta jamaicensis		Balunaguta	9.90
Crotolaria verrucosa	Snake weed	Nil-Andanahiriya	9.00
Urena lobata		Patta-Apala	9.00
Cadiospermum halicacabum	Urena burr	Wal-penela	18.18
Eragrostis tenell	Baloon vine		9.00
Echinochloa sp.	Love grass		18.18
Cyperus esculenthus			9.00
Desmodium hetorophylla			9.00
Lubelia purpurascens			18.18
Digitaria scheamum		Maha-Udupiyaliya	9.00
Oxalis corniculata	Smooth grass	Ambul-Ebiliya	9.00
Cleome viscosa	Creeping oxalis	Wal-Aba	27.27
Cyperus spp.	Spider flower		9.00

Eleven *F. oxysporum* isolates and six non-*Fusarium* genera *(Curvularia* sp., *Trichoderma* sp., *Mucor* sp., *Backusella* sp., *Chaetomium* sp., and *Rhizoctonia* sp.*)* were recorded in the examined banana fields associated with weeds. *Fusarium oxysporum* was isolated from eleven species of weed plants in the surveying area (Table 2). However, the isolation frequency differed apparently in samples of weed species.

Table 2. Isolation frequency of *Fusarium oxysporum* from weed plants in banana fields.

Weed plants tested	No. of plants used for isolation (segments)	Isolation frequency of *Fusarium oxysporum* (%)
Ageratum conyzoides	9 (36)	50
Mikania scandens	3 (12)	33.3
Ludwijia octoralis	7 (28)	35.7
Paspalum conjugatum	4 (16)	68.7
Cenchrus spp.	2 (8)	62.5
Echinocloa colonum	4 (16)	31.5
Phylanthus niruri	6 (24)	62.5
Euphorbia hirta	7 (28)	53.57
Fimbristylis globulosa	2 (8)	62.5
Cleome rutidosperma	2 (8)	75
Ipomoea macaranthua	5 (20)	35

Fifty per cent or more isolation frequency was observed in weed species such as *Ageratum conyzoides, Paspalum conjugatum, Cenchrus sp, Phylanthus niruri, Euphorbia hirta, Fimbristylis globulosa,* and *Cleom rodosperma*. Among those weed species, isolation frequency of *Fusarium oxysporum* was highest in *Cleome urtidosperma* (75%). In general, all weed species listed in the table 2 had the isolation frequency of *Fusarium oxysporum* over 30

% indicates that *F. oxysporum* was prevalent in these weeds. Naiki and Morita (1983) isolated *F. oxysporum f. sp. spinaciae* from weed roots grown in spinach fields in Japan. It has been also reported that various wild and cultivated plants host to some fusarial wilt pathogens (Waite and Dunlap, 1953).

The pathogenicity tests with Fusarium isolates indicated that none of the above eleven isolates was pathogenic on banana as no symptoms developed in banana plants inoculated with the above isolates except *F. oxysporum* f.sp. *cubense* (control). This suggests that the weeds examined in the area do not harbour *F. oxysporum f.sp. cubense* as a symptomless career. The inoculation test for the weed plants harboring these pathogens were not carried out because of the limitation of time period in the study.

Conclusion

Fusarium spp., *Mucor* sp., *Curvularia* sp., *Rhizoctonia* sp., *Trichoderma* sp., *Bacckusella* sp. and *Chaetomium* sp. were found associated with the weed flora in examined banana fields. *Fusarium oxysporum* was able to isolate from *Ageratum conyzoides*, *Mikania scandens*, *Lodwijia octoralis*, *Paspalum conjugatum*, *Cenchrus spp*, *Echinochloa colonum*, *Phylanthus niruri*, *Euphorbia hirta*, *Fimbristylis globulosa*, *Cleome rutidosperma* and *Ipomoea macarantha* weed species in the area. However, none of the *Fusarium oxysporum* isolates of those weeds was pathogenic on banana.

Seventy-four weed species were recorded in eleven banana fields surveyed in the Southern region of Sri Lanka. *Euphorbia heterophylla*, *E. hirta*, *Echinochloa colonum*, and *Eclipta prostrata* were found in almost all the banana fields surveyed. There was a wide diversity of weed flora within the studied area suggesting any weed management programme should focus on the control of these different weed species.

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REMEDIATION OF COPPER CONTAMINATED SOIL USING POTENTIAL COPPER-ACCUMULATING PLANTS

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Abstract: Potential of moderate Cu accumulating plants to remediate Cu-contaminated soil was studied using common sorrel (*Rumex acetosa)* and southern crab grass (*Digitaria adscendens*), which are common weeds in agricultural fields. *Rumex acetosa* extracted a high amount of Cu from low Cu bioavailable soil because of high shoot Cu accumulation ability. *Digitaria adscendens* extracted significantly higher Cu from the high Cu bioavailable soil, because of high dry matter accumulation even under high Cu availability. Bioavailability of the soil Cu was increased by ethylenediaminetetra acetic acid (EDTA) application and cattle manure compost (CMC) reduced the soil Cu bioavailability and improved the plant dry matter accumulation but increased total Cu extraction. For efficient phytoremediation, improvement of growth conditions of potential Cu accumulating plants would be effective. Shoot Cu content was significantly higher compared to the range of Cu in average phyto-mass in some plant species sampled in Cu high soil of Minethipia. Potential Cu accumulating plants with external assistance to manipulate bioavailability of the soil Cu would remove reasonably high amounts of Cu from the contaminated soil s.

Key words: Copper, EDTA, phytoremediation, potential-plants

Introduction

Heavy metal contamination of environment is serious concern for agricultural land, ground water and ultimately animal and human health (Hammer and Keller, 2002). Phytoextraction of heavy metals using hyperaccumulating plants got more attention as remedial measures in present context. Hyperaccumulators are species capable of accumulating metal at levels 100 folds greater than typically measured in shoots of non-accumulator plants (Lasat, 2002). Most of the reported plants as Cu hyperaccumulators are not accumulate Cu up to that level in practical conditions. Despite subsequent reports, the existence of plants hyperaccumulating metals other than Cd, Ni, Se and Zn has been questioned and requires additional confirmation (Salt *et al*. 1995). Most of the hyperaccumulator plants produce very small shoot biomass and have extremely slow growth. Plant with high biomass with moderate metal uptake will remove the same amount of metal from the soil as the plant that has produced low shoot biomass. Characters of potential phytoaccumulator in that sense are accumulating particular metal in reasonably high level in shoot biomass and production of high shoot biomass, which can compensate the moderate accumulation level (Ruggiero *et al*. 2003). Use of potential accumulating plants for Cu phytoremediation got less attention, but availability of hyperaccumulator plants for Cu has been questioned. This experiment was conducted to identify level of shoot Cu accumulation in plants growing under high soil Cu content and to evaluate the potential of moderate Cu accumulating plants for soil Cu remediation and to estimate the effect of organic matter (OM) and ethylenediaminetetraacetic acid (EDTA) application on bioavailability of soil Cu.

Materials and Methods

Plant screening experiment

Minethopia in Ehime prefecture, Japan (33.50^{o}N, 132.45^{o}E) was selected to study Cu accumulation of plants in a naturally Cu contaminated site. Soil and different plant species were sampled. Soil total Cu, Diethylenetriaminepenta acetic acid (DTPA) extractable Cu (bioavailable) and plant shoot and root Cu contents were analyzed.

Pot experiment

Common sorrel (*Rumex acetosa)* and southern Crab grass (*Digitaria adscendens*) were grown in 4 types of soil treatments in 3 replicates. Soil treatments are given in Table 1. Fertilizer was applied to the soil at the rate of 200 kg N ha^{-1}, 150 kg P ha^{-1} and 200 kg K ha^{-1} for all the soils. Dolomite was applied at the rate of 3 t ha^{-1} to T1, and 2 t ha^{-1} to T2, T3 and T4.

Table 1. Soil treatments

Treatment No	Soil treatments
T1	Cu contaminated granitic regosol
T2	T1 + 5% (w/w) CMC (cattle manure compost)
T3	T1 + 5% (w/w) CMC + 6 m mol EDTA kg^{-1} soil
T4	Long term CMC amended granitic regosol

Soil was filled (1.5 kg) into 1l plastic pots. Four seedlings of *R. acetosa* and *D. adscendens* were introduced to each pot. Plants and soil samples were collected 60 days after plant growth. Plant shoot dry weight, plant shoot Cu content and soil Cu fractionation before and after plant growth were analyzed. Four types of soils were subjected to incubation in the same period of plant treatment and Cu fractionation was analyzed. Tessier's sequential extraction method (Tessier, *et al*. 1979) was used with some modifications to extract different Cu fractions. Easily exchangeable fraction (1 M $MgCl_2$) [F1], carbonate bound fraction (1 M NaOAc buffered at pH 5) [F2], Iron and manganese oxides bound fraction (1 M $NH_2OH{\cdot}HCl$ and 25% acetic acid) [F3], organic matter bound fraction (0.02 M HNO_3, 30% H_2O_2 adjusted to pH 2 and 3.2 M NH_4OAc in 20% HNO_3 acid) [F4] and residual fraction (digested with 1:1 H_2SO_4 and HNO_3) [F5] were extracted sequentially.

Results and Discussion

Plant Cu content in naturally Cu high soils

Total and bioavailable Cu was varying from 125.2 to 240.6 and 12.1 to 57.6 mg kg^{-1} soil, respectively in soil samples collected from Minethopia. The highest shoot Cu content was 66.8 mg kg^{-1} dry matter (DM) in Japanese mugwort (*Artemisia princes*). Thymeleaf sandwort (*Arenaria serphllifolia*), Japanese thistle (*Cirsium japonicum*), and Shepherd’s purse (*Capsella bursa-pastoris*) had accumulated 46.5, 38.1 and 31.4 mg Cu kg^{-1} DM, respectively (Table 2). Shoot Cu content was significantly higher as compared to the range of Cu in average phytomass, 0.004 – 0.02 mg kg^{-1} DM, when soil Cu content was higher than the natural level of 0.03 mg Cu kg^{-1} soil (Larcher, 2001).

Table 2. Cu fractionation in T1, T2, T3 and T4 soils

Soil	(F1)	(F2)	(F3)	(F4)	(F5)	Total
			Cu mg kg^{-1} Soil (SD)			
T1	3.41 (0.33)	10.71 (0.00)	20.93 (1.88)	0.60 (0.17)	2.84 (0.00)	38.48
T2	3.59 (0.08)	4.49 (0.07)	9.80 (0.00)	23.80 (0.00)	5.51 (1.11)	47.19
T3	26.46 (0.83)	1.68 (0.00)	2.06 (0.00)	14.64 (0.17)	5.04 (0.45)	49.88
T4	0.06 (0.00)	0.26 (0.00)	1.21 (0.00)	15.35 (0.17)	8.51 (0.89)	25.38

SD – Standard deviation.

Applicability of potential plants in the soil Cu remediation

Rumex acetosa is an erect perennial, native in a wide range of grassy habitats including grass leys. It is common on arable land and has been selected based on the available literature as a Cu tolerant plant (Jiang *et al*. 2002). *Digitaria. adscendens* has been introduced to many parts of the world and is often a troublesome weed (Ebinger, 1962). *Digitaria adscendens* was selected because of its tolerance of harsh environmental conditions and faster growth. Plants were subjected to soil conditions of high Cu bioavailability and very low Cu bioavailability in T1 and T4 treatments, respectively. The impact of CMC and EDTA to accelerate the soil Cu remediation by manipulating bioavailability of soil Cu was investigated by T2 and T3 treatments, respectively.

Shoot Cu concentration

Shoot Cu concentration in *R. acetosa* and *D. adscendens* was 21.8 and 1.8 mg kg^{-1} DM, respectively in T4 treatment, in which soil F1 was 0.06 mg kg^{-1} soil. Shoot Cu concentration in *R. acetosa* and *D. adscendens*, increased to 88.5 and 14.3 mg Cu kg^{-1} DM respectively in T1 treatment, in which F1 was 3.4 mg kg^{-1} soil. Both plants accumulated highest Cu in the T3 treatment, in which soil F1 Cu was most increased by EDTA. Bioavailable fraction of soil Cu was directly related to the shoot Cu concentration. Cu accumulation was high in *R. acetosa* compared to *D. adscendens* for all the soil treatments.

Shoot DM accumulation

The DM accumulation of *D. adscendens* was 31.8 g pot^{-1} in T1 but was only 0.2 g pot^{-1} in *R. acetosa*. DM accumulation of *R. acetosa* and *D. adscendens* were 2.2 g pot^{-1} and 32.9 g pot^{-1} respectively in T4. DM accumulation of *R. acetosa* was severely affected by high Cu availability in soil as compared to *D. adscendens*. DM accumulation of *D. adscendens* was significantly higher in all the treatments, because of its ability to tolerate unfavorable soil conditions as a grass species (Figure 1b). Growth of *D. adscendens* was improved from 31.8 to 47.8 g DM pot^{-1} by CMC amendment. Both plants accumulated the lowest dry matter in T3 because of Cu and EDTA toxicity. High DM accumulation of *D. adscendens* was a beneficial character as a Cu remediation plant.

Total Cu extraction

Total Cu extracted by *R. acetosa* was 0.02 and 0.05 mg Cu pot^{-1} in T1 and T4, respectively but *D. adscendens* extracted 0.45 and 0.06 mg Cu pot^{-1} in T1 and T4, respectively. *Digitaria adscendens* dominated in total Cu extraction from T1 because of high DM accumulation though it had a low shoot Cu content (Fig 1a). But Cu remediation potential of *R. acetosa* was closer to *D. adscendens* in T4, in which soil F1 was low. A plant's ability to produce high DM becomes a critical factor in total Cu extraction when soil Cu availability is high. If Cu availability is very low, high Cu accumulating ability is useful to extract more Cu from soil. CMC application increased total Cu extraction from 0.45 to 0.86 mg pot^{-1} by increasing DM accumulation, though OM reduced the shoot Cu content. EDTA increased F1 Cu, but plant DM accumulation was severely affected by toxicity.

Change of soil Cu bioavailability by CMC and EDTA

The F4 was low in the T1 soil, because of very low OM content. Nearly 50% of total Cu was F3 in the T1 soil. After CMC application into the T1 soil, F4 increased from 0.6 to 23.8 mg kg^{-1} soil. F3 was highly depleted and F2 was also depleted to increase F4 (Figure 2). The CMC application did not change F1 suddenly, but F1 was reduced from 3.6 to 0.8 mg kg^{-1} soil, after growth of *D. adscendens*. Residual Cu in CMC might be the reason for the slight increment of F5 in T2. The F1 increased significantly after EDTA amendment and F2, F3 and

F4 were highly depleted. Results indicated the potential of EDTA to increase F1 Cu in soil. But in T4 almost all Cu (94%) was in F4 and F5, which are hardly available for plants. Strong bindings of Cu ions to the organic matter fraction limited the Cu bioavailability.

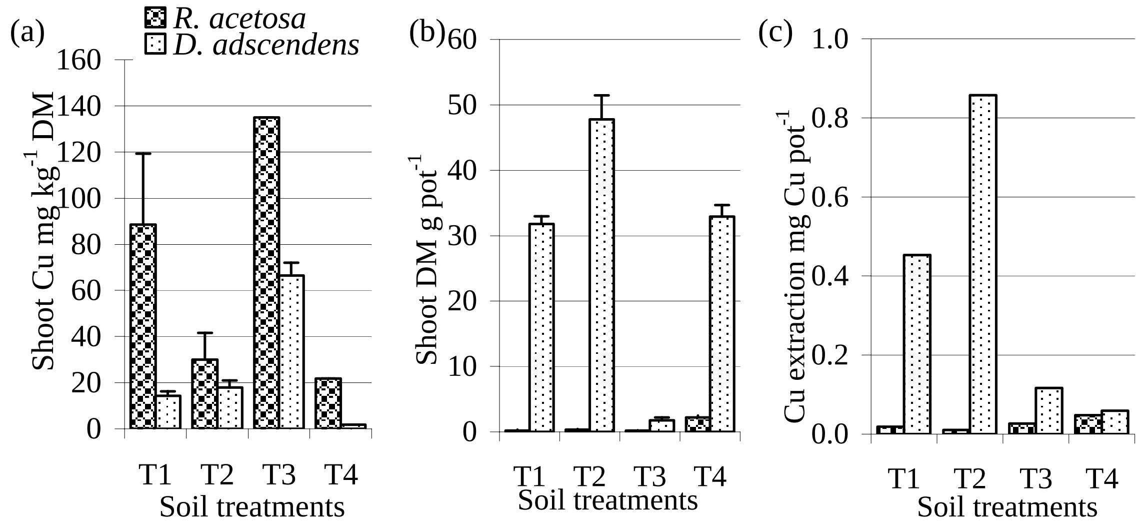

Figure 1. (a): Cu accumulation in plant shoots, (b): Dry matter accumulation in plant shoots and (c): Total Cu extraction from soil

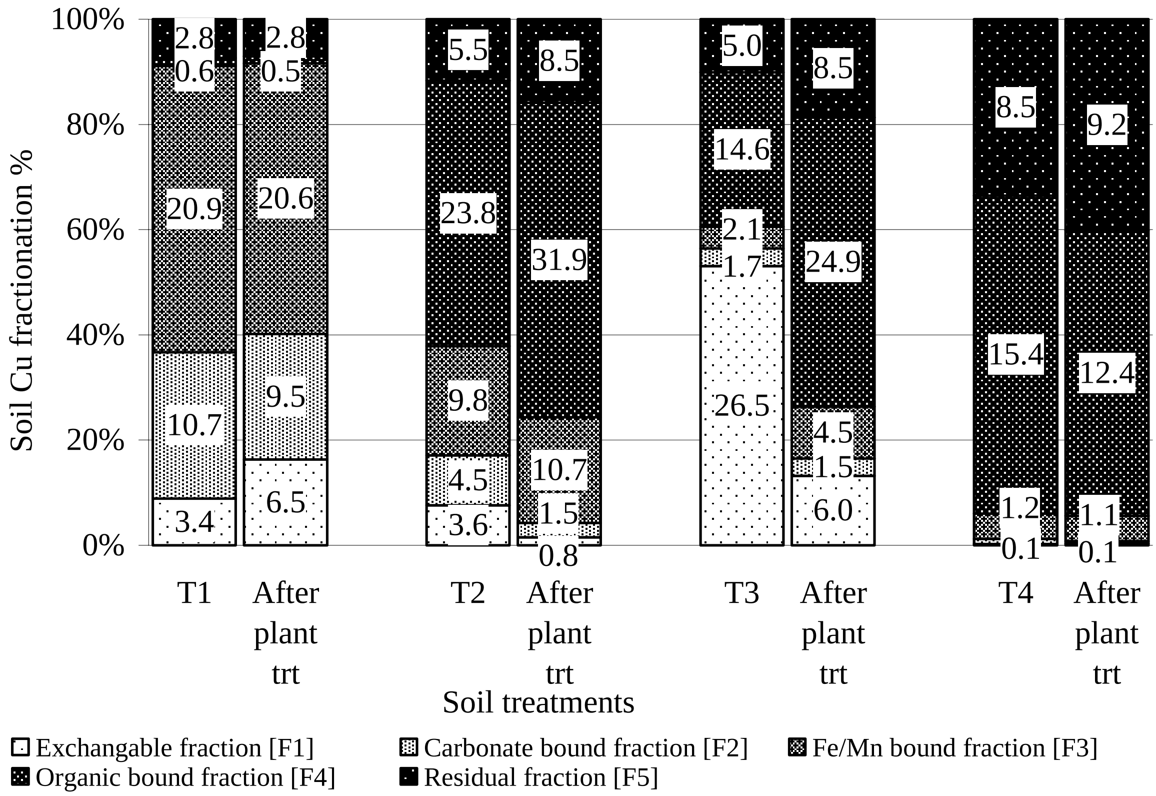

Figure 2. Initial Cu fractionation and change of Cu fractionation after plant growth in T1, T2, T3 and T4 soils

Plants grown in Cu high soil indicated high shoot Cu content than the natural level. *Artemisia princes*, *Arenaria serphllifolia*, *Cirsium japonicum* and *Capsella bursa-pastoris* were few plants which indicated more than 30 mg Cu kg^{-1} DM. Shoot Cu accumulation ability of *R.*

acetosa was high, as compared to *D. adscendens.* But vigorous growth of *D. adscendens* produced high shoot DM even in T1 soils in which soil F1 was high. Ability of a plant to maintain normal growth under high soil F1 is an important character to extract a high amount of Cu, when the soil Cu bioavailability is high. Total soil Cu extraction by *R. acetosa* was almost similar to *D. adscendens* in T4, in which F1 was very low. When soil F1 is low, a plant's ability for high shoot Cu accumulation becomes a critical factor to extract more Cu. Maintaining optimum plant density will further improve plant Cu remediation potential. Plants having high capacities for both shoot Cu accumulation and high DM production will be perfect as potential Cu accumulating plants. The CMC reduced the bioavailability of Cu in T1 and improved the growth of *D. adscendens*, which enabled higher Cu extraction from soil. Improving the conditions for plant growth would lead to more efficient phytoremediation, especially if potential Cu accumulating plants are used. High solubility of heavy metals-EDTA complexes (Grcman *et al*. 2001) effectively increased soil F1 in T3 soil and shoot Cu concentration. But, it decreased plant growth was significantly and limited the Cu extraction from soil. Similar growth retardation was reported by Shen (2002) on the growth of cabbage plant and increment of shoot Pb concentration when EDTA was applied. Potential Cu accumulating plants with external assistance to manipulate bioavailability of the soil Cu would remove reasonably high amounts of Cu from the contaminated soils.

Conclusions

Rumex acetosa indicated good soil Cu extraction potential in low Cu bioavailable soils and *D. adscendens* indicated significantly high soil Cu extraction potential in high Cu bioavailable soils. The CMC reduced the bioavailable fractions of Cu while EDTA increased the bioavailable fractions of Cu.

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BIOCHEMICAL ADAPTATION OF PURPLE NUTSEDGE (*Cyperus rotundus* L.) TO FLOODING: PYRUVATE DECARBOXYLASE ACTIVITY IN DRYLAND AND WETLAND ECOTYPES

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Abstract. Purple nutsedge (*Cyperus rotundus* L.), a major problem weed in dryland fields, is now increasing in populations and emerging as a problem weed in flooded rice grown in rotation with dryland crops. Apparently, the continuous wet-dry rotation pattern over the years has selected for an ecotype of purple nutsedge that can grow in flooded soil. Laboratory studies were conducted to determine the basis for its adaptation to flooded soil by comparing activities of pyruvate decarboxylase (PDC), one of the enzymes in alcoholic fermentation, in dryland and wetland ecotypes. At the onset of germination, PDC activity was detected in small amounts in wetland plants but was almost absent in dryland plants. Subjecting the seedlings to 24 hrs of hypoxia resulted in a significant increase in PDC activity in the wetland plants but leveled off as hypoxia was prolonged to 48 hrs. The dryland plants exhibited an increase in PDC activity after 24 hrs hypoxia following germination, which further increased as hypoxia was extended to 48 hrs. In addition, the amylase activity in the wetland tubers was twice higher than amylase activity in the dryland tubers during germination up to 24 and 48 hrs after germination. This ensured availability of enough free soluble sugars in the wetland plants, which was about thrice greater, than the level of soluble sugars in the dryland plants. Results indicate that wetland purple nutsedge adapted to flooded conditions by maintaining high levels of soluble sugars to fuel the ethanol fermentation pathway, and by utilizing low rates of ethanol fermentation by down-regulation of pyruvate decarboxylase to conserve energy sources throughout extended periods of flooding stress.

Key words: Anaerobic stress, ecotype, ethanol fermentation, pyruvate decarboxylase

Introduction

In the 1970s, purple nutsedge was a major weed in dryland crops and only a minor weed in flooded rice in the Philippines (Moody, 1992). Over the years, the continuous rotation pattern of flooded rice – dryland crop (usually vegetables) has selected for purple nutsedge that can grow in flooded rice. This resulted in increasing populations of purple nutsedge in flooded rice. A survey conducted in central Luzon, Philippines showed that purple nutsedge was the second most dominant weed in rainfed flooded rice in the late 1990s (Baltazar *et al.* 1999). This suggests that over the years, purple nutsedge has adapted to flooded soil in a continuous rice-vegetable rotation pattern.

A general response of organisms when exposed to anaerobic environment is the shift of energy production from aerobic respiration to alcoholic fermentation. The enzymes of glycolysis and ethanolic fermentation, pyruvate decarboxylase (PDC) and alcohol dehydrogenase (ADH) have been demonstrated to increase in rice (Ellis and Setter, 1999) and maize (Laszlo and St. Lawrence, 1983) under anaerobic conditions. Pyruvate decarboxylase catalyzes the decarboxylation of pyruvate to yield carbon dioxide and acetaldehyde, the latter being the substrate of alcohol dehydrogenase (ADH) in the pathway. PDC is considered as the key regulator of ethanol production under anaerobic conditions since it is present in lower amounts than ADH, and its activity is almost the same as the rate of ethanol fermentation *in vivo* (Tadege *et al*. 1998). With evidence supporting PDC as the regulatory enzyme in ethanol fermentation, PDC serves as an attractive target to study how *Cyperus rotundus* L. has adapted to flooded soil.

This research was conducted to determine the biochemical basis for the adaptation of purple nutsedge to flooding by (1) comparing PDC activities of wetland and dryland purple nutsedge; (2) determining the effect of hypoxic treatments on PDC activity and correlate PDC activity with hypoxia; and (3) measuring the total amylase activity in dryland and wetland tubers under different hypoxic treatments to assess their ability to sustain the ethanolic fermentation pathway. Results from this study can contribute to understanding how weeds adapt to their environment and thus help in determining ways to manage weeds through habitat manipulation. Data generated from this research can also be used in programs designed to develop tolerance in flood-sensitive crops. The studies were conducted at the laboratory and greenhouse facilities of the International Rice Research Institute (IRRI), Los Baños, Laguna, Philippines from June, 2005 to September, 2006.

Materials and Methods

Preparation of plant samples

Tubers were collected from wetland (also called lowland) and dryland (also called upland) fields in five provinces: Nueva Ecija, Laguna, Tarlac, Pangasinan, and Iloilo. The tubers were sprouted and grown in trays in the greenhouse in their respective habitats: flooded soil for wetland tubers and saturated soil for dryland tubers. At 7 days after sprouting, tubers with emerging roots and shoots but with the leaves not yet unfolded were used in the enzyme assays and in the hypoxia treatments, with dissolved oxygen content of 1.5 ppm or lower, for 24 and 48 h. Tubers from which the roots were cut off for the enzyme extraction and assays were used in determination of amylase activity and protein content.

Pyruvate decarboxylase extraction and activity assay

Enzyme extraction as optimized by Valdez (1995) was adapted with modifications. The PDC was extracted from 1 g roots using 6 ml of PDC extraction buffer. The roots were ground in a mortar and pestle in ice. Before centrifugation, the extracts were transferred to 1.5 ml Eppendorf tubes with enough bovine serum albumin (BSA) to give a final concentration of 1%. The pH was adjusted to 6.0 with 2-[(N-morpholino)ethanesulfonic acid] (MES) (1 ml of the extract requires 0.2 ml 250 mM MES containing 2.5 mM Mg^{+2} as cofactor to adjust its pH to 6.0), and thiamine pyrophosphate (TPP) was added to give a final concentration of 0.5 mM (1.2 ml of extract at pH 6.0 needs 0.06 ml of 10 mM TPP to give a final concentration of 0.5 mM TPP). The extracts were centrifuged at 13,000 rpm for 3 min at 4°C using a Beckman Coulter Allegra 21R centrifuge. The supernatant was transferred to clean Eppendorf tubes for enzyme assay and protein content determination.

The assay for PDC activity as optimized by Valdez (1995) was used with modifications. The enzyme extract (supernatant) was incubated at 25°C for 1 hr prior to assay. The assay mixture consisted of the following, which was introduced into a 1 ml-capacity cuvette in the following order: 500 µl 125 mM MES buffer (containing 2.5 mM $MgCl_2$), 100 µl 5 mM TPP, 100 µl 500 mM oxamate, 100 µl enzyme extract, 50 µl ADH (10 IU), 50 µl 3.4 mM ß-nicotinamide adenine dinucleotide, reduced form (NADH), and 100 µl 100 mM pyruvate. After the addition of NADH, the mixture was stirred (Vortex-2 Genie, Scientific Industries) and allowed to stand for 2 minutes for nonspecific oxidation to take place, after which pyruvate was added. The absorbance was read using a Beckman Coulter DU 800 spectrophotometer at 340 nm at 30°C continuously for 10 min. A time-course graph was provided by the spectrophotometer. The enzyme activity was estimated from the maximal slope of the decline in absorbance over time after addition of the substrate from which the rate of decline in absorbance over time before the addition of substrate was subtracted (Ellis and

Setter, 1999). One unit of enzyme activity (U) is defined as one micromole of NADH oxidized by 1 mg of protein per minute.

Amylase activity and protein content.
The tubers from which the roots were removed were peeled and chopped into small pieces. Extraction buffer (6 ml/gram sample) was added, then the mixture was ground in a mortar and pestle in ice. The extracts were centrifuged at 13,000 rpm for 3 min at 4°C using a Beckman Coulter Allegra 21R centrifuge. The supernatant was transferred to clean Eppendorf tubes for enzyme assay and protein content determination. The method of Bernfeld (1955) was adapted for total amylase activity assay. The samples were added with 1% starch as substrate and incubated at 25°C for 3 to 4 min to achieve temperature equilibration. At timed intervals, 0.5 ml of starch solution was added to each sample at 25°C and allowed to stand for 3 min for reaction to take place. One ml of dinitrosalicylic color reagent was added to each sample, incubated for 5 min in a water bath at 100°C, cooled to room temperature, then added with 10 ml distilled water. The solution was mixed well and the amount of maltose released from the reaction was measured by reading the absorbance at 540 nm against a standard curve. The amount of maltose (µmoles) released in each sample was calculated based on the standard curve. One unit of enzyme activity (U) is defined as one micromole of maltose released from starch by 1 mg of protein/min at 25°C and pH 6.9. The protein content of all enzyme preparations was measured using the Bradford method (Bradford, 1976) with BSA as the standard.

Statistical analysis.
A split-split plot design was used in the measurement of PDC and amylase activities, with the ecosystem (dryland and wetland) as the main plots, the areas within each ecosystem as the subplots, the hypoxia treatments as the sub-subplots, with each subplot having three replications. Each replication is an average of 3 repeat measurements. Experimental variables were analyzed in terms of ecosystem, area, treatment, ecosystem *x* area, ecosystem *x* treatment, area *x* treatment, ecosystem *x* area *x* treatment interaction effects. Values of the variables were analyzed using analysis of variance (ANOVA).

Results and Discussion

Morhphological characterization of wetland and dryland tubers
Wetland purple nutsedge plants were taller and bigger than the dryland plants (Plate 1). Differences in tuber size were also evident, with the wetland type almost twice bigger than the dryland type.

Plate 1: Wetland (left) and dryland (middle) fields where purple nutsedge was collected; left photo shows tubers from wetland (upper) and dryland (lower) purple nutsedge plants.

Upon germination, the roots of the dryland tubers were thinner and more fibrous while those of the wetland tubers were bigger and more succulent. Having bigger root systems for the wetland purple nutsedge could be one form of morphological adaptation to flooded conditions to allow the formation of aerenchyma networks for diffusion of toxic wastes generated from the ethanol fermentation pathway (Drew, 1997).

Preliminary screening of PDC activity

The_PDC activity was evident in the wetland seedlings during germination (Figures 1a and 1b). Subjecting the germinating wetland seedlings to hypoxia for 24 h led to higher rates of PDC activity. In the dryland seedlings, PDC activity was low but in detectable amounts during germination. When subjected to 24 h hypoxia following germination, dryland seedlings also exhibited an increase in PDC activity. Plants shift from aerobic respiration to the fermentation pathways when experiencing oxygen stress, with ethanol fermentation being the major pathway of anaerobic carbohydrate breakdown (Gibbs and Greenway, 2003). Many evidences have been presented to show that an increase in the enzymes of the ethanolic fermentation pathway, pyruvate decarboxylase (PDC) and alcohol dehydrogenase (ADH), takes place when plants are subjected to hypoxia or anoxia.

Pyruvate decarboxylase activity (U)

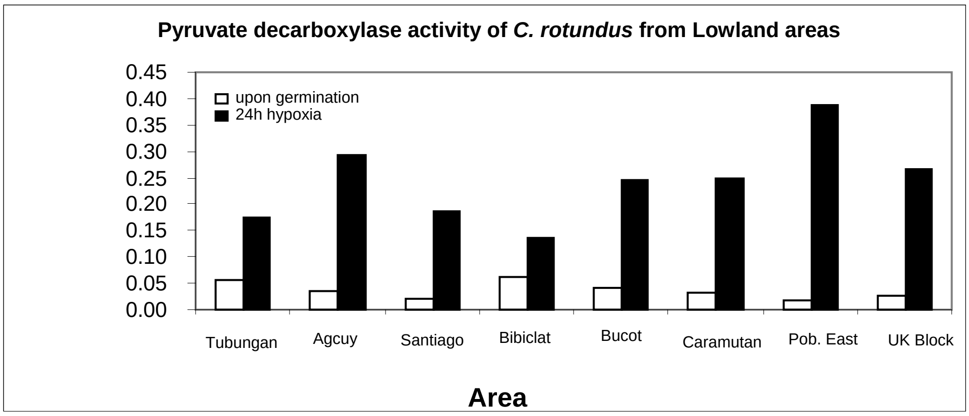
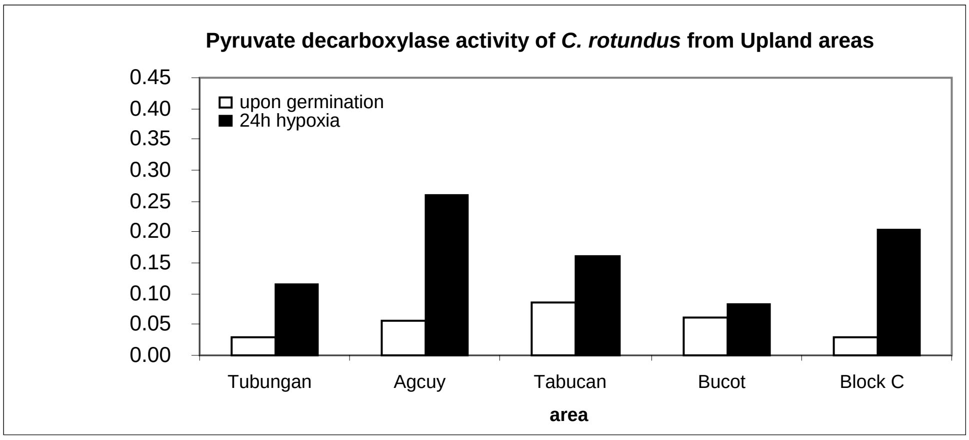


Figure 1a. Pyruvate decarboxylase activity of purple nutsedge from wetland (lowland) fields

Pyruvate decarboxylase activity (U)



Figur 1b. Pyruvate decarboxylase activity of purple nutsedge from dryland (upland) fields

The PDC activity of dryland and wetland seedlings subjected to 24 and 48 hrs hypoxia.
Subjecting the germinating seedlings to hypoxic stress for 24 hrs led to an increase in enzyme activities for both ecotypes (Figure 2). Hypoxia for 24 hrs led to an 84-fold to 1020-fold increase in PDC activity in the dryland seedlings. In contrast, there was only a 6-fold to 8-fold increase in PDC activity in the wetland seedlings. Further subjecting the seedlings to hypoxia for 48 h resulted in a leveling off of PDC activity in the wetland seedlings. In contrast, there was a continuous increase in PDC activity in the dryland seedlings (Figure 2). PDC activity of the dryland seedlings was significantly higher than PDC activity in the wetland seedlings after 48 h of hypoxic stress.

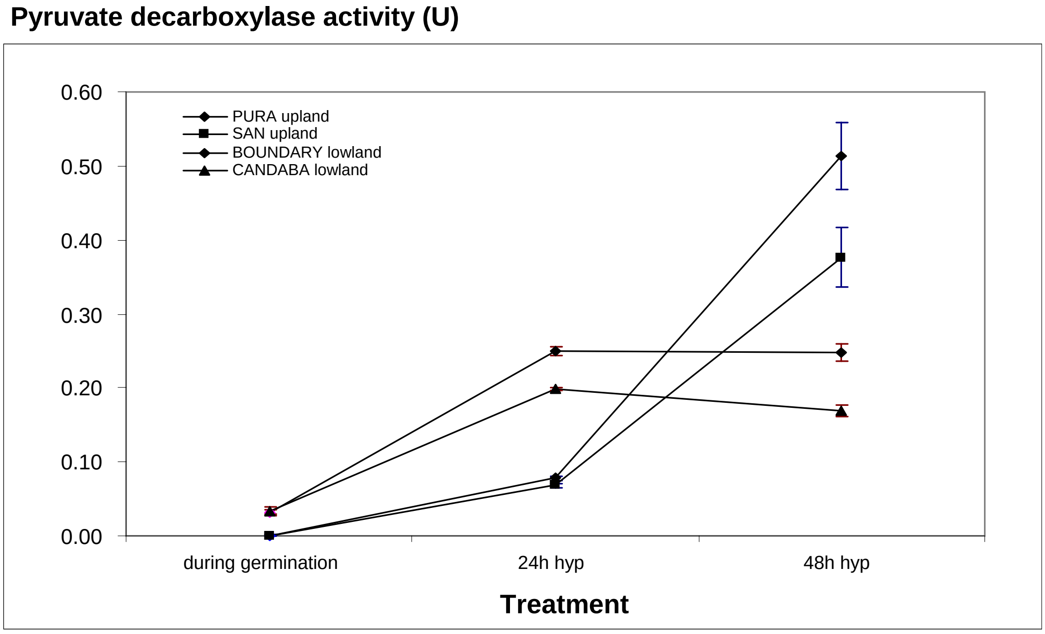


Figure 2. Pyruvate decarboxylase activity in dryland and wetland purple nutsedge seedlings subjected to 24 and 48 hrs hypoxia following germination.

Amylase activity under hypoxia.
Amylase activity was twice higher in non-germinated wetland tubers than in dryland tubers. This enzyme activity level remained constantly higher than amylase activity in dryland tubers before and during germination (data not shown). High amylase activity in the wetland tubers before and during germination ensures a constant supply of free soluble sugars to fuel the ethanol fermentation pathway. Perata *et al.* (1993) emphasized the importance of substrate availability during germination under anoxia. This observation is supported by results of a similar study (Peña, pers comm) who observed that soluble sugar levels are significantly higher in the tubers the lowland purple nutsedge compared to the dryland purple nutsedge before and during germination.

Our results suggest that both purple nutsedge ecotypes can induce PDC activities when experiencing hypoxic stress. The induction of enzyme activity was much greater in dryland plants with a continuous increase in PDC activity after prolonged hypoxic treatment. In the wetland plants, a significant increase in PDC activity was observed after 24 hrs of hypoxia following germination, but enzyme activity leveled off and was maintained at this level during prolonged hypoxia. These results indicate that the lowland ecotype utilizes low rates of ethanol fermentation as an adaptive mechanism to germinate and thrive under flooded environments. The leveling-off of PDC activity, coupled with high amylase activity to ensure enough supply of soluble free sugars to fuel the ethanol fermentation pathway, enables the lowland purple nutsedge to avoid depletion of substrates and energy source during long periods of exposure to oxygen stress hence avoid flooding injury and death of the plant (Gibbs and Greenway, 2003).

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RESPONSIVE CHOICES TO CONTROL *Mikania micrantha* H.B.K. IN NON-CULTIVATED LAND IN THAILAND

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Abstract: *Mikania micrantha* H.B.K. (Asteraceae) is native to tropical America. It is a noxious invasive weed in Southeast Asia, South Asia, and China. The weed grows fast and climbs up on top of trees and shrubs, forms a thick layer and eventually suffocates the support plants underneath. Large areas of primary vegetation in Thailand have been transformed into agricultural, agro-forests and non cultivated shrubs or secondary forests. The weed is currently widespread, growing uncontrolled on non-cultivated land and invading protected areas. The vast area of distribution makes manual removal impossible. Past experience indicates that selective herbicides should be applied and followed by measures ensuring that the vine cannot recover. Natural enemies of the weed are always promising. Efforts in searching for locally effective natural enemies, pathogens and insects, are strongly recommended. In China, two species of dodder were utilized and successful control was reported. Further investigation on the efficacy of local dodder species and the possible negative impacts on other species have to be done. Plant community reconstruction aims to create the environment unsuitable for the vine to grow. Several species have been successfully used for community reconstruction in China. The approach of selecting available local tree species for community reconstruction in Thailand is being investigated.

Key words: *Mikania micrantha*, herbicides, biological control, community reconstruction

Introduction

Alien species and their invasions occur worldwide and have become one of the most serious ecological and economical problems. *Mikania micrantha* H.B.K., which is a slightly woody vine species of Asteraceae, native to tropical America, is an alien invasive species in Thailand, where it has fast vegetative growth from March to August, flowering from December to January, and fruiting follows immediately. The production of seed closely relates to the environment. The production is higher in open, wet and fertile habitats.

Large area investigation

For the large area investigation, it was necessary to use a car instead of walking. Therefore, it was crucial to be able to spot *Mikania micrantha* from a moving car at medium speed. Since during most of the year, *M. micrantha* merged into the green background of other plants, an investigation should be set at the time when *M. micrantha* is easily distinguished. Thus, this investigation was set in December during *M. micrantha* flowering. The white color of the blooming inflorescence or the brown of mature fruit were clearly seen even at more than 40 m away. *Mikania micrantha* was particularly abundant along the road to the south from Bangkok, along the coastal area of southwest and eastern part of Thailand. The weed also appeared in Chiang Mai, the northern part of Thailand, even in nature reserves. *Mikania micrantha* grew rampant in areas where primary vegetation had been transformed to secondary forest or on abandoned land. The weed was usually under control in well managed agricultural and agro-forest lands since weeds were cleared regularly before they covered the crop. However, the amount of additional labor expenses for clearing *M. micrantha* was unknown. Some suggested that *M. micrantha* could be used as a kind of land covering in oil palm plantations.

Several clumps of *Cuscuta chinensis* were found parasitizing *M. micrantha* in Chiang Mai, and *C. reflexa* were found parasitizing this weed in Chumphon, a province in the south of Thailand. *Cuscuta* spp. appeared to naturally associate with *M. micrantha*.

Choices of responses
Based on experience in China several methods might be considered.

Spraying of herbicides
Carefully selected herbicides should be effective and leave the least impact on the environment. Spraying could keep the weed under control at least for several years. The crucial point is to determine the suitable timing for spraying. The two proper times recommended are the duration after the seedlings has grown to a certain size in order to be spotted easily and also before mass flowering starts, to avoid new seeds forming the new seed bank. Therefore, the life cycle of *Mikania micrantha* in Thailand should be investigated before mass spraying is recommended. Seed bank investigations in China indicated that the *M. micrantha* seeds were distributed 0-3 cm deep in the soil.

The viability of seed decreased from top to bottom (Zhang *et al.* 2005). Most of seed emergence was from 0.5-2.0 cm. No seedlings emerged from soil deeper than 5 cm (Li *et al.* 2002). Six months after the initial germination experiment, no new seedlings appeared. In the field, no seedlings appeared during June to February of the following year (Zhang *et al.* 2002). The seeds in Thailand are likely to follow a similar pattern, but this needs investigation. It should also be noted that herbicides should be applied in large areas at the same time. Otherwise seeds from the vicinity would invade the sprayed areas during fruiting season.

Manual / mechanical removal
This could be done in well managed areas such as parks. In mature oil palm plantations, as long as the weed was removed before climbing up and covering the top of the palm trees, it did not pose a severe threat to the plantation itself. But then, the seeds might be dispersed by wind or other means to other places and cause problems. The amount of possible additional labor cost, however, will be investigated.

Use of parasitic plants
The genus *Cuscuta* contain parasitic plants, belonging to the family Convolvulaceae. There are about 170 species in this genus. Two species of dodder, *C. chinensis and* giant dodder, *Cuscuta reflexa* were found parasitizing *M. micrantha* in Thailand. *Cuscuta* spp. could keep *M. micrantha* under control by deterring the growth of the weed. However, dodder’s growth lags behind *M. micrantha*.

The dodder develops well when the weed it is parasitizing grows well. Hence, it will be difficult for dodder to completely exclude *M. micrantha* from a large area. Dodder will serve as a reliever to the plant covered by *M. micrantha* to some extent. One concern of using parasitic plants is the ecological safety. The success of using them elsewhere does not guarantee the safety in Thailand. Therefore, the safe use of dodder should be investigated before using it as a biological control agent. In some areas in Thailand, the villagers eat *Cuscuta* spp. as indigenous vegetables. This might be considered as a deterrent for spreading of *Cuscuta* spp..

Use of natural enemies
Utilization of natural enemies is an attractive management measure. Unfortunately, promising natural enemies are not found at this stage. More efforts are needed to find an effective local natural enemy. The introduction of external control agents should be done in very carefully while preventing irreversible negative impacts.

Vegetation-ecological control

Considering that most invaded areas are the consequence of destruction of original plant communities (Cock, 1982b; Ipor,1991), it indicates that *Mikania micrantha* does not grow well under closed canopy (Bogidarmanti, 1989; Ipor, 1991; Huang *et al*. 2000; Zhou *et al*. 2005) and *M. micrantha* does not pose a threat in its native ranges in tropical America, this proves that the existence of *M. micrantha* does not necessarily cause damage.

Vegetation reconstruction by planting selected tree species with the least human intervention to suppress *M. micrantha* in China was successful. This might be a once and for all solution in the area where land is to be reverted to the original forest. The plant species selected was crucial for the success of reconstruction. The ideal species should be a regional, fast growing, broad canopy tall tree, although probably no single one species would meet all these criteria. Thus, investigations to select trees in Thailand are necessary.

Discussion

In Thailand, at present, emphasis should be placed on previously disturbed reserve areas where the intention was to let those areas undergo positive succession. Without human intervention, it is likely that the weed would cause degradation of the existing plant community, thus preventing the community to recover. The appearances of the weed in a reserve area in the north need to be closely monitored. The invasion should be terminated before it causes pronounced negative effects.

Crop plantations are under regular control. Therefore, the effect is usually terminated before *Mikania micrantha* grows on top of the crop. It does, however, become the source of propagules that might spread outside the plantations. Hence it is wise to clear the weed if it is close to an area, which needs exclusion of the weed, *e.g.* reserve areas. Chemical control is one of the important measures to manage *M. micrantha*. Large scale spraying would be able to clear *M. micrantha* for several years. The disadvantage of this is that spaying might need to be repeated for several years. The use of certain *Cuscuta* spp. would be helpful in suppressing *M. micrantha*. The use of *Cuscuta* spp. as a vegetable would become an additional protection for ecological safety if *Cuscuta* spp. did cause a negative effect.

Planting selected tree species to suppress *M. micrantha* is probably the best way. It takes advantage of natural processes to manage invasive species, but it is not universally applicable. For example, it is not suitable on plantation and other agricultural areas. Management measures might differ from place to place. With proper background research and careful planning, successful management of *M. micrantha* is possible. The invasion of a species might bring environmental and economical catastrophe. Therefore, it is crucial to manage the problem at national ecological security level. To prevent invasion from its source, it is necessary to establish related regulations and laws to raise public awareness and strengthen quarantine measures. It is known that preventing invasion is far more efficient than controlling after damage has occurred.

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EFFECT OF SPRAYER NOZZLES ON EFFICACY AND ECONOMY OF SPRAYING HERBICIDE IN SUGARCANE

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Abstract: Field experiments were conducted in 1999 and 2001 at the research farm of the Sugarcane Research Institute (SRI) of Sri Lanka to assess the efficacy and economy of controlling weeds in sugarcane after spraying herbicides with different types of sprayer nozzles. The nozzles had to be calibrated before spraying to apply the required amount of herbicide. Impact blue and flat fan are more suitable among the tested nozzles. Crop damage was significant by spraying with impact-red nozzle. More time was required for post-emergent spraying. As the operator had to swing the lance to cover the target area, spraying with nozzles having narrow swath width takes additional time. Post-emergent application with solid cone twin nozzle gave poor weed control.

Key words: Sugarcane, weeds, sprayer-nozzles

Introduction

Application of herbicide is a popular practice to control weeds in sugarcane. However, appropriate herbicide should be applied in the correct manner to obtain satisfactory results. The sprayer nozzles primarily determine the nature and pattern of spraying and hence the efficacy and economy of controlling weeds (Klingman, 1973). For example, discharge rate, application rate, droplet size, swath width, shape of spray etc. vary and depends on the type of nozzles. Thus, the nozzles must be chosen on the basis of satisfactory performance of a given job but information on efficacy and economy of controlling weeds after spraying with different types of sprayer-nozzles are lacking. This study was therefore planned to fill this gap in the knowledge.

Material and Methods

Field experiments were conducted in 1999 and in 2001 at the research farm of the Sugarcane Research Institute (SRI) of Sri Lanka (Latitude 6° 21’ N; Longitude 80° 48’ E and elevation 97 msl). Pre-emergent and post-emergent herbicides were sprayed in sugarcane by different types of sprayer nozzles (Figure 1). Fields were prepared to make ridges and furrows at 1.4 m spacing for both experiments. Sugarcane stem cuttings (setts) were planted in the furrows. Land preparation, planting and crop management were done as per SRI recommendation (SRI, 1991) and raised under supplementary irrigation. Nine meter long six cane rows were taken as a treatment plot. The operator walked on the ridges to spray herbicides. Each nozzle was calibrated before spraying.

In 1999, Impact – red, Impact – blue, Impact – green, Solid Cone, Hollow cone and Flat fan nozzles were tested in a Randomized Complete Block Design with three replications. Un-weeded and weed free plots were used as standards. A tank mixture of diuron 80% + paraquat 40EC (2.8 kg. a.i/ha + 400 g a.i/ha) was sprayed at 4 days after planting (DAP) as a pre-emergent application. Then the plots were weeded manually at 45 DAP. Paraquat 40EC (800g a.i/ha) was sprayed at 90 DAP as a post emergent application.

Weeds were sampled by using randomly placed 60 cm x 40 cm quadrat in each plot at 45 DAP and 90 DAP (45 days after manual weeding). Then the weeds were dried at 80°C to a constant weight to estimate weed bio-mass. Areas of damaged leaf of each treatment plot were measured by using automatic leaf area meter to assess crop damage.

Figure 1. Different types of sprayer nozzles tested in the experiments

In 2001, two identical experiments were conducted for pre- emergent and post-emergent spraying. Here, solid cone- twin nozzle was used instead of the flat fan nozzle tested previously. Experiments were treated in RCBD with three replications. Pre-emergent application was done at 4 DAP while post emergent application was done at 28 DAP with the same herbicide mixtures. Subsequent weeding was done manually as and when required.

Table 1. Characteristics of different nozzles used to spray herbicides

Type of nozzle	Swath width (m) at 45 cm height	Discharge rate l/h	Application rate l/ha
Impact – red	2.0	148	338
Impact – blue	1.5	99	271
Impact - green	1.0	74	175
Flat fan	1.5	57	172
Solid cone (single)	0.81	46	125
Hollow cone (single)	0.67	40	104
Solid cone (twin)	1.2	118	360

In contrast to the 1999 experiment, the operator swung the sprayer lance while spraying with single cone type nozzles, to cover the width in between sugarcane rows as practiced by farmers. Weed control, crop growth and the cost involved in each activity were recorded.

Results and Discussions

Different types of nozzles have different swath width, discharge rate and application rates (Table 1). Therefore, the sprayer should be calibrated with the respective nozzle before spraying to apply the required amount of herbicide.

Experiment in 1999

Application of pre-emergent herbicide: At 45 DAP, un-weeded plots recorded the highest weed biomass on ridges, followed by plots sprayed with impact-red nozzle. Weeds on ridges in plots sprayed with impact-blue, impact-green, hollow cone, solid cone and flat fan nozzles were significantly lower (P=0.05) and similar to the manually weeded plots and hence these nozzles are effective in controlling weeds on ridges in sugarcane (Table 2).

In the furrows, weed bio-mass at 45 DAP was more (P=0.05) and similar to the un-weeded plots in the plots sprayed with solid-cone and hollow cone nozzles. The operator walked on the ridges directing sprayer lance over the ridges to spray herbicide. Therefore, narrow swath widths (67 cm) of solid cone and hallow cone nozzles were not adequate to spray herbicide in-between the furrows spaced 1.4 meters apart. Therefore, weed control in the furrows of the plots sprayed with solid cone and hallow cone nozzles was less. Impact-red nozzle failed to control weeds satisfactorily on ridges and in furrows. Weed biomass was significantly lower (P=0.05) and similar to the manually weeded plots in plots sprayed with impact-blue, impact-green and flat-fan nozzles (Table 2). These three nozzles were effective in controlling weeds both on ridges and in furrows.

Application of post-emergent herbicide: The highest weed biomass ($p<0.05$) on ridges was observed in un-weeded plots and in plots sprayed with impact-red nozzles. Rest of the nozzles, except flat-fan controlled weeds as in manually weeded plots (Table 2).

In furrows, a level of weed control similar to the manually weeded plots was recorded only in the plots sprayed with impact-blue nozzles. Un-weeded plots and the plots sprayed with impact-red nozzle recorded higher weed bio-mass ($p<0.05$). In the furrows, weed control was moderate after spraying with impact-green, solid-cone, hollow -cone and flat-fan nozzles (Table 2). At this stage (90 DAP), sugarcane seedlings have grown to a height of 40 to 50 cm and the tiller density was15 to 20 per meter. Therefore, weed growth in the furrows has been partly checked due to mutual shading. This could be attributed with the lower amount of weeds in the furrows of the plots which were sprayed by nozzles with narrow swath width. Therefore, satisfactory weed control of the crop at this age could be achieved by inter-row spraying of post emergent herbicides even with nozzles having narrow swath width.

Table 2. Mean weed biomass (41 days after spraying pre-emergent herbicide- 45 DAP); and crop damage (damaged leaf cm^2/plant) after post-emergent herbicide application (45 days after manual weeding or 90 DAP) in 1999 experiment.

Type of nozzle	After pre-emergent herbicide application (45 DAP)		After Post-emergent herbicide application (90 DAP)		
	Weed biomass (g/m^2)		Weed biomass (g/m^2)		Damaged leaf (cm^2/plant)
	Ridges	Furrows	Ridges	Furrows	
Impact-red	18.1 ab	23.9 ab	11.0 b	10.8 b	148.1 a
Impact-blue	3.6 c	6.7 c	1.2 d	2.5 de	29.8 bc
Impact-green	10.9 bc	8.4 bc	3.9 cd	5.5 cd	47.8 b
Solid-cone	1.4 c	31.2 a	0.1 d	11.4 b	0
Hollow-cone	4.8 c	21.9 ab	1.8 d	8.6 bc	0
Flat-fan	9.0 bc	12.2 bc	8.7 bc	5.5 cd	0
Un-weeded	25.5 a	37.7 a	22.6 a	21.0 a	-
Manual weedin	1.7 c	4.0 c	1.5 d	0.7 e	-

Application of post emergent herbicide with impact-red nozzle not only gave poor weed control but also resulted in significant (P=0.05) crop damage (148 cm^2/plant). The plots sprayed with impact-blue and impact-green also showed some phytotoxicity, but damage levels were significantly lower. Use of solid-cone, hollow-cone and flat fan nozzle to spray herbicide did not cause phytotoxic symptoms on sugarcane leaves (Table 2).

Experiment in 2001

More time was required to spray post emergent herbicides than pre-emergent herbicides. Reduction of operator's walking speed in a weedy field compared to a bare land could be attributed to this. Also, the operator had to take some time to swing the sprayer lance while spraying to cover the targeted weeds adequately. Therefore, spraying time has been increased further by about 10% for pre-emergent and 22% for post-emergent application when using solid cone and hollow cone nozzles (Table 3).

Application of pre-emergent herbicide: All tested nozzles had similar weed control properties and hence there was no significant variation ($p<0.05$) in labor requirements for subsequent manual weeding between different treatment plots at 7 and 16 weeks after planting (WAP) and hence was equally effective to spray pre-emergent herbicide in sugarcane (The sprayer lance was swung to get required target coverage while spraying with the nozzles having narrow swath width) (Table 3).

Application of pre-emergent herbicide: Weed death on the ridges was highest in the plots sprayed with impact-green, solid cone and hollow cone nozzles. These nozzles generally have narrow swath width, which was less than sugarcane inter-row spacing. Therefore, the operator swung the sprayer lance while spraying and hence there were chances to spray more on the ridges. This could be attributed to the greater weed death on ridges by spraying nozzles with narrow swath width. No significant differences ($p<0.05$) in weed death in the furrows between difference types of nozzles. However, on the ridges, weed death was least in the plots sprayed by solid-cone twine nozzle. Also, more labor was required to weed plots sprayed by twin nozzle, perhaps due to poor weed control in these plots. Also labor requirement for manual weeding at 7 WAP was more in post emergence sprayed trial than pre-emergence sprayed trial (Table 3).

Paraquat was sprayed on grown up weeds but some of the mature weeds did not die. As such plots sprayed post-emergence required more labor for subsequent manual weeding. Labor requirement was twice (about 15.6 labor/ha) in the plots sprayed by single (impact and cone types) nozzles and thrice (24 labourers/ha) in the plots sprayed by twin nozzle than the pre-emergent herbicide applied plots. Labor requirement for manual weeding at 16 WAP did not differ much in both pre- and post-emergence sprayed trials (Table 3).

Table 3. Spraying time (hr/ha), dead weed after spraying, crop damages (CD), labour requirement for manual weeding (7 and 16 WAP) in pre-emergent and post emergent sprayed trials.

Type of nozzles*	Pre-emergence spraying			Post-emergence spraying					
	Time	Labour/ha		Time	Dead weeds (%)		CD	Labour/ha	
		7WAP	16WAP		Ridge	Furrow	(%)	7WAP	16WAP
Red	2.7 b	7.5	8.6	3.2 b	24.3 bc	20.7	55.5	15.6 ab	6.9 b
Blue	2.8 b	8.3	8.6	3.0 b	19.5 bc	24.9	60.0	15.6 ab	8.3 ab
Green	2.8 b	8.1	10.1	3.1 b	29.8 abc	22.0	60.8	15.0 b	7.7 ab
Solid	3.1 a	7.8	8.4	4.1 a	37.7 ab	23.4	49.2	17.6 ab	8.0 ab
Hollow	3.1 a	6.3	7.8	3.9 a	48.2 a	26.5	63.0	14.4 b	7.0 ab
Twin	2.7 b	8.6	9.5	3.7 a	12.9 c	14.5	47.9	24.0 a	8.7 a
Mean	2.9	7.8	8.8	3.5	28.7	22.0	56.1	17.0	7.8

Note: Red (impact-red); Blue (impact-blue); Green (impact-green); Solid (solid cone); Hollow (hollow-cone) and Twin (solid-cone twin).

Crop growth and yield did not differ significantly ($p<0.05$) due to spraying with different types of nozzles in both pre- and post-emergence sprayed experiments (Table 4). However, application of post emergent herbicides reduced crop growth ($p<0.05$) in early stages (e.g. germination, tillering, tiller height etc.). However, the number of millable stalks, cane yield, juice quality and sugar yield were not affected. This shows that initial set back of crop growth due to post-emergent application of paraquat had recovered at later stages without reducing the final yield.

Table 4. Mean differences of crop growth and yield in pre & post emergent experiments.

Crop growth Parameters	Pre-emergence spraying		Post-emergence spraying		t-statistic
	Mean value	SE	Mean value	SE	
Germination (no/m)	8.8	0.04	5.5	0.02	6.9**
Height-6wap (cm)	19.0	0.04	18.0	0.009	2.2*
Tiller-12wap (no/m)	21.1	0.03	14.2	0.10	9.8**
Height-12wap (cm)	57.1	0.21	38.5	0.22	14.1**
Millable stalk (no/m)	12.0	0.07	11.5	0.04	0.63
Cane yield (t/ha)	111.1	2.8	111	3.1	0.04
POCS (%)	10.7	0.02	11.2	0.01	-1.62
Sugar yield (t/ha)	11.8	0.05	12.4	0.03	-0.99
Weeding cost (Rs/ha)*	6670.78	269.49	6156.77	307.84	1.26

*Note: Prices of Diuron 852.25 Rs./kg., Gramoxane 452.86 Rs./l; in estimating labour ciost, labor wage was taken as 200 Rs./day and eight working hour/day with 60% efficiency, spraying labor worked four hrs/day.

Total cost of weed control did not differ ($p>0.05$) between these two types of spraying. However, in post emergence-sprayed trial, use of double cone type nozzle increased the weeding cost by about 33% (from 5834.24 to 7769.30 Rs./ha) than single nozzles. This may be due to poor weed control after spraying with this nozzle.

Conclusions

Nozzles should be calibrated before spraying to apply the required amount of herbicide. More time is required to cover the target adequately when spraying with cone type nozzles with narrow swath width. All tested nozzles except impact-red could be used to spray pre-emergent herbicides. Solid-cone twin nozzle and impact-red nozzle are not suitable to apply post-emergent herbicides. Post-emergent application of paraquat at about 3 to 4 WAP damaged young seedling but recovered later without affecting final yield under irrigation. Weed control cost did not differ between pre-emergent and post-emergent sprayed trial but more labor was required for subsequent manual weeding in post emergent sprayed trial. This is risky, because labor may not be available at time for manual weeding. Therefore, it is suggested to apply pre-emergent herbicides and follow up other operations accordingly to get higher yield with lesser risk.

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STRATEGY FOR WEED CONTROL IN GOLF TURFS

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Abstract: A study to develop a strategy for weed control in golf turfs was conducted in more than 58 golf courses in China during 2001~2006.The strategies were set up based on three turf grass climate zones in China. In zone1, the whole turf grass is warm season turf and includes Bermuda grass, *Zoysia spp*. or seashore paspalum. The strategy for weed control in this zone was sequential application of CHL and METS, RIM, BENS, CHM before or at weed emergence as pre-emergent control, twice or three times one year. In zone 2, most turf grass are warm season turf including Bermuda grass, *Zoysia* spp. or seashore paspalum in rough areas during the whole year and in fairways in summer and fall. Cool season turf such as perennial ryegrass are over sown in fairways in autumn, and grows from winter to early summer. The strategy for weed control in rough areas in this zone is same as in zone 1. In over-seeded fairways, pre emergent herbicide treatment is with BENS and PYR, 21 days after over-seeding as first application and in spring as the second application. The third application should be considered for both weed control and cool season turf with METS, RIM, BENS or CHM. In zone3, most turf grasses are cool season turf such as perennial ryegrass or bluegrass both in rough areas and fairways. Recently, *Zoysia* spp. became popular and was grown in rough areas instead of tall fescue. For weed control in rough areas, pre-emergent herbicide treatments are recommended with BENS and PYR in early spring and post treatment in later spring, or early summer in cool season turf with QUIN, TRIB, and in warm season turf with a high rate of METS,RIM, BENS,CHM. The pre-emergent treatment in fall to control winter weeds with BENS and PYR is also recommended.

Key words: Golf turf, weed control strategy
Abbreviations: CHL,Chlosulfuron; METS-metsulfuron; RIM-rimsulfuron; BENS- bensulfuron; CHM-chlorimuron; PYR-pyrazosulfuron; QUNI-quinclorac; TRIB-tribenuron

Introduction

Golf courses are being rapidly developed in China. More than 230 Golf courses has been set up by the end of 2006. At least 80 golf courses are being made. Weed control is a key component in any successful turf grass management program. Several herbicides can be used to control weeds as pre-emergence or post-emergence treatment. Bhowmik, (1995) indicated that weed control in turf can select several herbicides, according the weed species (Bhowmik, 1995; Johnson, 1996.). Most superintendents in golf courses try to control weed with chemicals, with little success, because they have not chosen the right strategy according the weed species to make suitable programs (Watschke and Hamilton. 1990; Xue and Ma 2001).

There are three turf grass zones in China, classified according to the climate zones. Warm season turf zone (zone1 includes Bermuda grass, *Zoysia* and seashore paspalum is in southern China. Cool season turf zone (zone2) includes perennial yyegrass, Kentucky bluegrass in fairways and tall fescue or *Zoysia* in rough areas in northern and western China. The rest is transition zone (zone3), which has warm season turf including Bermuda grass or *Zoysia* or seashore paspalum in rough areas all year but from later spring to late fall; cool season turf grass is over sown in fairways. This study was conducted in more than 58 golf courses in China from 2001~2006 to establish a set of strategies for weed control in golf turfs in three turf grass zones in China based on the climate zones and weed species.

Materials and Methods

The experiments were carried out with 1 to 4 pre-emergent weed controls per year at Dongshan golf, Zhaoqing golf, Fairy lake golf and Shenzhen tycoon golf respectively during

2001 and 2004 in zone 1. In transition zone 3, the experiments were carried out twice using pre emergence control in rough areas and one pre emergent control in fairways per year at Sanyang golf, 3 times pre-emergent control in rough areas and 2 times pre emergent control in fairways per year at Suzhou sunrise golf and 4 times pre emergent control in rough areas and 3 times pre emergent control in fairways per year at Grand shanghai golf respectively during 2002 and 2005. In zone 2, the experiments were carried out with 1 time pre emergent control both in rough areas and fairways. In addition, post emergent treatment was carried out in later spring or early summer at Qingdao huanshan golf in 2005, with 3 times pre emergent control in rough areas and fairways. The post treatment was carried out in later spring and summer at Tianjin warna golf course during 2004 and 2006 (Table 2).

At least 9 holes of turf including fairways and rough areas of every golf were used as experimental sites. The demonstration trials were conducted in more than 58 golf courses during 2001~2006 (Table 1). Chlosulfuron, metsulfuron, bensulfuron, rimsulfuron, chlorimuron and/or ethoxysulfuron were used as pre emergent control in warm season turf. Bensulfuron and pyrazosulfuron were the pre emergent control chemicals in cool season turf. Quinclorac and/or tribenuron were post emergent treatment in all zones. The application rate of herbicide was 15~30 over the recommended rate on the label. All herbicides were applied as a broadcast sprays in 1000 L ha^{-1} of water with a large machine sprayer. All the turf was mowed to the normal height 2 days before herbicide application and proper irrigation was supplied after the sprayings. Weed control rating were based on 0 to 100 where 0=no control ,>80% acceptable control, and 100=complete control. Weed control rating was made every10~20 days after application in one year. Turf injury rating were based on 0 to 100 where 0= no injury,1-15= minor leaf discoloration,16-30 =moderate discoloration with some necrosis,30=moderate to severe leave discoloration and necrosis, and 100=complete kill. Injury rating were made every 10-20 days after application for 100 days

Table 1. The location of golf courses in the demonstrate trial

Golf club	Zone
Tycoon golf(01~05) Xili golf(04~06) Green bay golf(03~06) Jiulong hill golf (02~05) Shenzhen airport golf (04~06) Century Seaview golf (03~04) Hill view Golf (04~06) Harbour plaza golf (04~06) Camdor harbour Golf (03~06) Zhuhai international golf (03~06) Gongzong golf (03~06)Chung Shan hot spring golf (02~04) Agile Golf (03~06) Royal orchid golf (02~04) Nansha Golf (04~05) Lotus hill golf (02~04) Luhu golf (01~04) Nanhu golf (04~06) Sino golf (02~05)Fosha golf (04~06) Zhaoqing golf (01~05)Palm island golf (02~05) Citic green golf (03~06) Donshan golf (01~04)Meishi golf (04~06) Kangle spa golf (04~06) Yalong bay golf (03~06)Ocean bay golf (01~04) West lake golf (05~06) Junan golf (06) Huizhou tangquan golf (06) Gaoming yinhai (05~06) Dragon lake (04~06) Orient (xiamen) golf (02~06) Southern wood golf (01~06) Amai golf (03~06)	1
Orient (Wuhan) golf (04~06) Jinyin lake golf (02~06) Merry land golf (04~06) Shanghai binhai golf (02~04) Shanghai reviera golf (03~06) Yin Tao golf (03~05) Tomson golf (02~06) Tianma golf (02~06) Hongqiao golf (05~06) Silport golf (02~06) Grand shanghai golf (01~06) Sunrise golf (01~06) Sanyang golf (02~06) Jinji lake Golf (06) West lake golf (03~05) Huangshan pine golf (03~06) Jianhu golf (05~06) Shuangshan golf (05~06) Nanjing zhongshan golf (05~06) Jingle lake golf (04~06)	2
Tianjin Warna golf (03~06), Qingdao huanshan golf (05), Daxina hot spring golf(04)	3

Results and Discussion

In zone 1, when pre-control measures were applied with the mixture of Chlosulfuron, metsulfuron, bensulfuron and ethoxysulfuron in May (1 time per year), weed control was of

95% for 60 days, 90 days of 85% weed control and 120 days of 75% weed control. With two pre-control applications in Apr and Aug per year, 200 days of 93% weed control, 240 days of 82% control and 300 days of 72% weed control were obtained. When three pre-control applications were made in Mar, Jun and Jul., 240 days of 93%weed control, 300 days of 88% weed control and 350 days of 80% weed control were obtained. Four pre-control applications on Feb, May, Aug and Oct per year, procured 270 days of 92% weed control, 310 days of 90%weed control and 360 days of 82% weed control. The result showed that the best strategy of weed control in this zone was 4 pre-control applications per year with Chlosulfuron, metsulfuron, bensulfuron and thoxysulfuron/chlorimuron The next was 3 times and 2 times per year (Table 2). When benefit are considered, a strategy with 2~3 times pre-control per year were suggested.

Table 2. Weed control methods and duration with pre-treatment in golf turfs of China

	Zone/Site	Treatment	Weed control and Effective period
1	Donshan golf	1 time (May)	60 d of 95%; 90 d of 85%;120 d of 75%
	Zhaoqing golf	2 times (Apr and Aug)	200 d of 93%; 240 d of 82%;300 d of 72%
	Fairy Lake golf	3 times (Mar, Jun and Jul)	240 d of 93%; 300 d of 88%;350 d of 80%
	Shenzhen Tycoon golf	4 times (Feb, May, Aug induct)	270 d of 92%; 310 d of 90%;360 d of 82%
2	Sanyang golf	2 times (Apr and Oct) 1 time (Oct) in Fairways	110 d of 92%; 150 d of 80%;180 d of 70%
	Suzhou sunrise golf	3 times (Feb, Jun and Oct) 2 time (Oct and May) in Fairways	240 d of 93%; 300 d of 85%;350 d of 80%
	Grand hanghai golf	4 times (Feb, May, Jul and Oct)) 3 times (Oct, Feb and May) in fairways	270 d of 95%; 310 d of 92%;360 d of 85%
3	Qingdao huashan golf	1 time (Apr) pre-; 1 time (Jun) post-	65 d of 93%; 90 d of 85%;110 d of 80%
	Tianjin warna golf	3 times (Apr, May and Jul) pre- 2 times (May and Jun) post-	180 d of 95%; 210 d of 92%;270 d of 85%

In zone2, the results indicated that the weed control strategy in rough areas in this zone was achieved with 2~3 times pre-control per year with Chlosulfuron, metsulfuron, bensulfuron and thoxysulfuron/chlorimuron on or before turf turn up or at the growing stage. In over seeded fairways, pre-control with bensulfuron and pyrazosulfuron 21 days after over seeding as first application and in spring as second application was the best for controlling weeds. The third application should be for both weed control and cool season turf suppression with metsulfuron, rimsulfuron, bensulfuron and/or chlorimuron. In this strategy, 240 days of 93% weed control, 300 days of 85% weed control and 350 days of 80% weed control was obtained. However, 4 times pre-control applied on Feb, May, Jul and Oct in rough areas and 3 times pre control applied on Oct, Feb and May in Fairways is not suggested for golf business even 85% of weed control lasted for 360 days (Table 2).

In zone3, most turf grass is cool season turf such as perennial ryegrass or bluegrass in rough areas and in fairways. Recently, *Zoysia* spp. began to be popular for rough areas instead of tall fescue in zone3. The strategy of weed control in rough areas was pre emergent treatment with bensulfuron and pyrazosulfuron in early spring and post emergent treatment in later spring or early summer in cool season turf with quinclorac and/or tribenuron. In warm season turf, metsulfuron, rimsulfuron, bensulfuron and chlorimuron at the rate which was 25~30% over the recommended rate on the label. The pre emergence treatment in fall to control winter weed with bensulfuron and pyrazosulfuron was also effective (Table 2).

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ORNAMENTAL AQUATICS: POTENTIAL WEEDS IN AQUATIC ECOSYSTEMS

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Abstract: A recent survey conducted to identify and document aquatic plant diversity of Sri Lanka has resulted in the identification of three exotic plant species, *Echinodorus* sp. (Alismataceae), *Mayaca flutivalis* (Myacaceae) and *Ludwigia sedioides* (Onagraceae) occurring extensively in the natural water bodies in Gampaha and Kurunegala districts in the country. All these three species are popular as aquarium plants that are currently being propagated and exported. Further, these species has not being recorded as naturally occurring in Sri Lanka. These plants has the potential in spreading into the other natural water bodies and becoming potential weeds while threatening the local flora that growing naturally in aquatic ecosystems. The threats by such alien species have being well documented and therefore adequate measures should be taken to control the escape of such exotics from control environments into the natural habitats.

Key words**:** Ornamental aquatic weeds, alien invasive species

Introduction

Ornamental aquatic plant industry has grown substantially during the last decade and has a potential to become one of the major foreign exchange generating ventures in Sri Lanka. These plants have become one of the essential components in aquaria and landscape industry. The developing interests in the landscape industry involving water as an element has contributed to the current demand of the aquatic plants. The private sector growers have started getting involved in the ornamental aquatic plant industry. Even though Sri Lanka harbors a large number of naturally occurring aquatics, there is a demand for certain exotic plants in the export market and this has resulted in the importation of alien plants into the country, where they are propagated and exported. According to the statistics of The National Aquaculture Development Authority of Sri Lanka (NAQDA), 180 plants have being listed under ornamental aquatics and 87 are being propagated and exported. Out of this 123 are exotic species. Once escaped from controlled environments, some of the exotic species, commonly referred as alien plants have the potential to invade extensively into the natural ecosystems thus causing detrimental damage to the local biodiversity and agriculture. Many such events have been encountered in the country in the past, and the plants involved have been identified and listed under the category of invasive alien plants (Amarasinghe, 2001; Marambe, 2001; Wijesundara, 2001). Sri Lanka itself has experienced many such plant introductions being noxious with time (Wijesundara, 1999; Marambe, 2001; Wijesundara, 2001). According to Marambe (2000), early detection and treatment of new infestations of invasive non-native plants is an effective and ecologically sound management approach. Further, strong inventory and monitoring programs provide the information on presence, distribution, and size of non-native plant populations. These inventories also provide essential information for planning, budgeting, and setting priorities. Harris *et al.* (2007), in a study on the introduction history and invasion success in exotic vines introduced to Australia, emphasizes the importance of basic information including inventorying exotic plants for future managements.

Therefore, identifying and documenting the aquatic plant diversity of Sri Lanka in order to identify recent introductions is of prime importance for future reference and also for management, if in case, any of these recent introductions become noxious weeds to the aquatic ecosystems.

Materials and Methods

A field survey was conducted in the natural aquatic ecosystems, covering the three major agro-ecological zones of the country, including Gampaha, Ratnapura, Kalutara and Kandy (wet zone), Kurunegala an Matale (intermediate zone), and Polonnaruwa and Anuradhapura (dry zone). districts. Field visits were made during the dry and wet seasons of the year. Plants were collected and identified using literature and the National Herbarium, Royal Botanical Gardens, Peradeniya, Sri Lanka. Whenever exotic species were recorded local people were interviewed in order to collect information regarding these plants.

Results and Discussion

The survey resulted in the identification of three exotic plant species occurring extensively in the natural water bodies in Gampaha and Kurunegala districts. The species included *Echinodorus* sp. (Alismataceae), *Mayaca flutivalis* (Myacaceae) and *Ludwigia sedioides* (Onagraceae). All three species were recorded in the Gampaha district while in the Kurunegala district only *Echinodorus* sp. was recorded. Further, species of *Echinodorus* and *Ludwigia sedioides* were observed extensively covering the respective water bodies. *Echinodorus* sp. were found growing on marsh areas of the water body. *Mayaca flutivalis* was found forming dense mats on wet soil during the dry periods. These mats when covered with water during rainy periods, showed extensive growth under the water. Further, they are also capable of withstanding slow water currents. *Ludwigia sedioides* was also found forming dense mats on the surface of the water bodies. During the dry period of the year, the plants that occurred on the drier part of the water body were forming dense mats on the moist soil. The interviews carried out with the local people in the Gampaha district provided information that these plants were recent introductions to the area. However, in the Kurunegala district such response was not encountered.

All these three species, *Echinodorus* sp., *Mayaca flutivalis* and *Ludwigia sedioides*, recorded during the present survey, as occurring in natural water bodies, are plants that have not been recorded as naturally occurring in Sri Lanka. They are popular as aquarium plants and are currently being propagated and exported by growers. All these plants are listed under the aquatic plants by the NAQDA. They have listed 19 different species of the genus *Echinodorus*, out of which 12 are listed as being exported.

Echinodorus Rich. ex. Engelm is a genus of aquatic plants found in the western hemisphere commonly known as Burhead (http://en.wikipedia.org/wiki/ Echinodorus). These species are popular as specimen plants in larger aquaria and in ponds. The plants produce long racemes or panicles of 1-18 whorls of flowers, which are eye-catching. They are marsh or bog plants by nature and are capable of growing submersed. Further, according to literature they prefer good light and deep, high nutrient substrates but most of the species are capable of growing in variable water conditions (http://en.wikipedia.org/wiki/Echinodorus). It was also noted that *Echinodorus* sp. produce a large number of plantlets attached to the inflorescence and kept attached to the mother plant for a certain period of time before getting established as an independent individual. They grow in marshlands and are capable of growing submerged to a certain extent, towering above the natural vegetation. These habitats are areas preferred by amphibious and shallowly submerged plants. The water bodies acquired by *Echinodorus* sp. in Gampaha district were almost dominated while in Kurunegala district only a small population was observed.

Mayaca flutivalis naturally occurs in the southeast U.S.A., central and tropical south America and in tropical southwest Africa. This plant with a moss like appearance has spirally arranged linear leaves. The plant is capable of growing fully submerged during floods, while it forms dense mats on marshy soils during the wet conditions. According to literature the means of propagation is readily by stem cuttings and branches (http://www.freshwater aquariumplants.com/amazonbiotope/amazon.html). Therefore, the plant has the potential in spreading in large scale with the water currents, w and becoming an aquatic weed, threatening the local flora.

Ludwigia sedioides, commonly known as Mosaic flower or False loosestrife, is a herbaceous perennial and is native to south America. This is also used as a floating aquatic plant in the landscape industry. The red and green diamond shape leaves occur in rounded clusters and gives an attractive appearance for the floating plant. Propagation of *L. sedoides* is by snipping off a rosette, with a section of stem attached, from the main stem (http://www. aquaticplantcentral.com/forumapc/ plantfinder/details.php?id=112). The main stem may then develop into another rosette.

Aquatic plants in general tend to propagate readily and the present observations and also literature supports this idea regarding the three-recorded alien aquatics. Their propagules have the ability to spread rapidly using water as the media of dispersal. The rapid spread can block irrigation canals, causing a threat to agricultural lands. In the case where these water bodies are connected to streams, the propagules can spread into the other areas causing detrimental affects. Since these plants are not native, there is a possibility of them becoming invasive weeds, and a threat to the natural flora.

Many of the aquatic invasive species recorded in Sri Lanka has been introduced into the horticulture industry as ornamentals. Many past records indicate the efforts made by the Sri Lankan scientists to control the spread of these invasive aquatic plants in our ecosystems (Amarasighe 2001, Kotalawaela 1993; Lester-Smith, 1927; Marambe 2000; 2001; Wijesundera 1999; 2001). With these past records, initiating adequate measures to monitor the introduction of such noxious plants that will affect the local flora needs priority. According to Marambe (2000), prevention of the introduction of these non-native plants is the first line of defense in protecting an ecosystem. Further, it is the most economical and efficient means of management. Even though, these plants have not been listed as invasive, the performance of a plant as an invasive depends on many conditions. Several factors contribute for the trigger. Therefore, monitoring the performance of these plants and their relationships with the native plants populations become important. As stated, the methods adopted must prevent introduction of non-native plant species into the country, vigilant detection of nascent populations and work with partners to develop national, regional and local prevention strategies (Marambe, 2000; Rejmánek, 2000). Further, it is to note that the alien invasive species problem is a national security issue, and should be tackled using holistic approach (Marambe, 2000). The problem of alien invasive species has grown, and increasing in a rapid rate and therefore, during policy making management of invasive alien species should be of high priority (Marambe, 2000).

According to the aquatic plant list documented by NQADA, two species of the genus *Egeria* have being listed, namely *E. densa* and *E. najas*. These are species that are not recorded as native species. Tthe USDA (www.invasivespeciesinfo.gov/aquat), *E. densa* is listed as an invasive plant species. In this context it is important to conduct surveys on aquatic flora throughout the country in view of identification of native as well as exotic plants exists in the natural areas, specially close monitoring should be done in the water bodies in the close vicinity to the aquatic plant nurseries in view of detecting any possibility of escaping the propagules of exotics. Further, regulations should be implemented preventing the use of exotics propagated locally for the export market, to be used in the local landscape industry.

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CLASSIFICATION OF SPECIES FROM THEIR SEEDS - CREATING A DATABASE OF INVASIVE PLANTS FOR RISK ASSESSMENT

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Abstract: Since the opening of diplomatic relations in the 19th century, the numbers of naturalized invasive alien plant species have been increasing rapidly in Japan. More than 1600 species have been recorded until now. One of the major routes of invasion is in the form of seed. For example, a lot of seeds can be unintentionally mixed in the imported grain and brought in from overseas. In Japan, however, there is no regulation by the Quarantine Act over accidental invasions by import. For preventing accidental invasions, it is necessary to not only revise the regulations, but also identify the taxa of the invasive plant seeds. Unfortunately, it is difficult to identify the seed by using previous illustrated Japanese wild plants seed books because the books did not have the key to identify the taxa based on morphological characters of the seed. Our laboratory has a history of collecting wild plants seed for about 100 years. We have been preserving the living seed of most weed species recorded in Japan since 1990. In the present study, we have utilized the collections and constructed a database of seed images of naturalized alien invasive plants in Japan for risk assessment. In our web site, photographs of the seed and the plant images of the naturalized weeds in Japan can be viewed. Also it enables one to search a seed by its morphological characters, such as shape and ornamentation of surface to narrow down your search alternatives. It is a new trial in the world to release the seed image-database with the search function covering most of the invasive plants in Japan. We hope that this database will be used not only for quarantine but also for other purposes.

Key words: Seed, naturalized plant, image-database

Introduction

In Japan, based on Kariyama (1987), Enomoto (1997) and Enomoto's unpublished data, the numbers of naturalized plants have been increasing to ca.1600 species since the 19th century. Except for rice, we rely strongly on imports for most of the grain that we eat, such as beans, wheat, and corn. A large quantity of grain is brought in by ship. Shimizu *et al.* (1996) clarified and Enomoto (1999) summarized that the various weed seeds were unintentionally brought into Japan from various countries as dusts mixed in the imported grains. Many of these plants had already become naturalized in Japan. It seems that such mixed plant seed is one of the causes of the increasing numbers of naturalized plants in Japan. Currently there is no regulation for unintentionally mixed plant seed in grain imported to Japan.

To prevent the invasion of alien plants in the form of seeds, it is necessary to identify seeds by their morphological features. For example, in Britain, there is a seed handbook for quarantine (Jones *et al.* 2004), and in China, some illustrated books on weeds seed with keys are available (Guo, 1998; Guan *et al.* 2000). In Japan, some illustrated books with the seed photographs which contain wild or naturalized plants have been published. Ishikawa (1994) and Nakayama *et al.* (2000) mainly contained native plants in their illustrated seed books, but not so many naturalized plants. Shimizu *et al.* (2001) and Japan Livestock Technology Association (1994) contained seed photographs of 260 and 165 species, respectively, in their illustrated books of naturalized plants. But, any of these books published in Japan have no key to identify a plant seed from its morphological features. Currently, there are no books to learn efficiently about morphological features and identification of the seeds in Japan.

In recent years, research institutes around the world have been launching homepages with plant seed-images. Many of these homepages are focused on weeds and naturalized plants in the respective countries or regions. It seems that these homepages have contributed to increase the public interest in the importance of plant seeds for weed prevention and Quarantine. Also, on some homepages, we can search the seeds by their morphological features.

Since 2005, we have been subsidized by the Ministry of Education, Culture, Sports, Science and Technology, for the research project on Risk Assessment and Prevention of Invasive plants spreading. As a part of this project, we have constructed a seed-image database of the naturalized alien plant species in Japan, and exhibited it on the Web. This database enables one to search the seeds by their morphological features. The purpose of this database is to provide efficient learning material of the seed morphological features to prevent invasion and spreading of naturalized alien plant seeds.

Materials and Methods

Plant seed collection and preservation

Since the founding of the Research Institute for Bioresources in 1914, our laboratory has been collecting and preserving wild plant seed focusing on weeds in Japan. Since the 1990's, we have been preserving seeds in the freezer to keep them alive.

When collecting plant seeds, we have gathered them from each population or individual plants. We have also collected voucher specimens from which the seeds were taken, and preserved them at the herbarium in our laboratory. Therefore, it is possible for us to identify or systematically reexamine the species. The collected seeds were dried at 45 and cleaned by removing impurities. Then they were preserved at -30°C. For photographing their image and morphological examinations, some of respective plant seed specimens are preserved in the dry room. We have collected 7478 seed specimens of naturalized plants (934 species of 95 families) till 21 February 2007. Using them as research material, we have obtained images of seeds.

Photographing the image

We have used the stereoscopic microscope to digitally photograph the seed-images, and stored the images on the hard disk in jpg format. We photographed fruit and false fruit as disseminules, for example, the achene (Compositae, Rosaceae and Ranunculaceae), caryopsis (Poaceae), involucral bracts (*Xanthium*), and so on. We have photographed 770 species of 88 families till 21 February 2007.

Secimen management database

Plant seed specimen and its voucher specimen are numbered and registered on the relational database for managing the detailed information of specimens. Each Seed-image file links to this database, and we can view the images from specimen data. The image files used for constructing the Seed-image database on the Web site were selected from this specimen management database.

Seed morphological features

We described the seed morphology of the following seven characteristics: ratio, shape, gloss, projection, mound, wing and size. The length ratio of short axis and long axis of the plant seeds were divided into 4 categories as 1:1, 1:1 ~ 2:3, 2:3 ~ 1:5 and 1:5 ~. Gloss and wing were judged "present" or "absent". The list of character states of shape, projection (including spine and hair) and mound (ridge and ditch) were presented as the tables in our homepage. The seed size was based on a measurement of the photographed seed specimens. We did not mention about the color of seeds.

Results and Discussion

The data base is hosted in the homepage of the Wild Plants Science Laboratory, Research Institute for Bioresources, Okayama University (http://www.rib.okayama-u.ac.jp/wild/index.html). In the homepage, click the button to enter the Seed-image database, and then proceed to the introduction page of this database (Figure 1). Our database mainly consists of 2 sections: (1) searching seed by scientific name, (2) searching seed by morphological features.

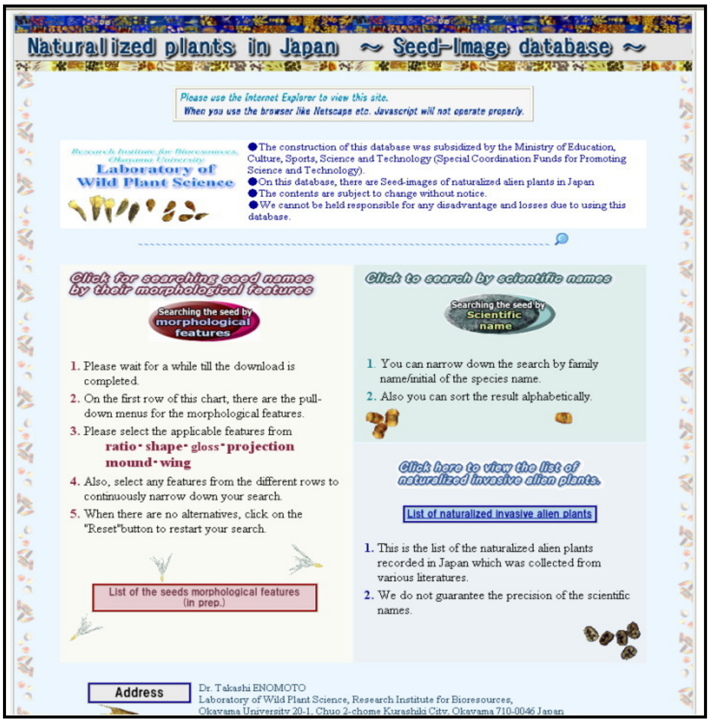

Figure 1. The introduiion page of our Seed-image Database

In the section for searching seed by scientific name (Figure 2), species names, family names, seed-images and plant-images will be displayed on the table. But the data sources of the plant images are not the same as those of seed specimens. A family name or the first letter of the name of the plant species could be used to search the seed- images. The regulated alien plant species by the Invasive Alien Species Act. Can be searched by clicking the each seed/plant-image.

Figure 2. The page for searching seed by scientific name

In the section for searching seed by the morphological features (Figure 3), the morphological characters (ratio, shape, gloss, projection, mound, wing and size), seed-images and plant species names will be displayed on the table. The seed characteristics at the top of the table are the pull-down menu. By clicking them, the list of seed characteristics will appear. Drag the mouse to select the applicable features from the menu and release the mouse button. Seeds with the selected characteristic will be listed. Furthermore, you can narrow down your search by selecting another feature among the other morphological characters. As the result, seeds with the two features of your choice will appear on the list. When there is no matching result, the list will not be displayed. When you start the new search, please click the reset button, otherwise you will not be able to correctly narrow down your search. By using above function, when you come across an unknown plant seed, you can list the seeds with the same morphological features. At present, we can narrow down our search to about less than 20 alternatives from the total records.

Figure 3. The page for searching the seed by morphological features

Some of Seed-image homepages which were exhibited from other countries in recent years have more plant species or better contents than ours. But there are not so many homepages with the search function of morphological features of the plant seed. At present, our Seed-image database is the only one in Japan as image database that contains the entire alien plant species of Japan. Also it is the only one with the search function of morphological features of the plant seed in Japan. Perhaps, our database is useful for learning about seed morphological features. We will not be responsible for any inconvenience or losses that may be caused by using this homepage.

At this stage, the seed morphological data of each species is based on one specimen, and we do not know about the variation range within the species. If we gather further information on the variation within the species, we will reflect them on the database. Besides, in future, for those species with the possibility of invasion due to artificial introduction into Japan, they should be added on to the seed-image database even if the same species are distributing as native species in Japan. To distinguish the alien species from the native Japanese plants, the scope to the wild plants in Japan should be widened.

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CROP PROTECTION PRODUCTS STEWARDSHIP PROGRAMS IN SRI LANKA

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Abstract: Crop protection products (CPP) enable farmers to produce high quality crops through control of pests and treatment of plant diseases. However, they must be used correctly to ensure safety. Syngenta's approach to product stewardship includes a commitment to support farmers through development and sales of effective and safe products, and promotion of their responsible use. A stepwise approach is used to develop programs targeted at specific needs of farmers in different countries. The first phase starts with an audit to understand the way products are used in practice, thereby allowing the selection of priority areas focused on activities associated with higher risks. Initial mitigation measures include new products and technologies with reduced risks, and rational product choice. The objective of the second phase is to define measures to minimize exposure and ensure their implementation. This is implemented in collaboration with a broad group of local partners ranging from agricultural communities to health organisations. Practical examples included farmers training activities and secure access initiatives. Finally the success of stewardship programs needs to be monitored, and one of the measures can be the development of adverse health incidents (AHI) recorded over time. This last step is important to continuously improve programme effectiveness and consequently the safe use of CPP. In Sri Lanka, Syngenta's commitments towards product stewardship are delivered through a number of measures, including introduction of a novel paraquat product with INTEON® technology (containing an alginate that converts to a gel under stomach acid conditions, increased emetic, and a purgative, hence with reduced oral toxicity), training in secure storage and provision of lockable boxes, and AHI monitoring and epidemiological study. The AHI records and study results both indicated that deliberate self-harm and accidental ingestion of CPP had been substantially reduced, and the new formulation improved overall survival of patients following paraquat ingestion.

Key words: Paraquat, stewardship, INTEON®

Introduction

The use of crop protection products (CPP) has enabled farmers to produce quality and sufficient crops through control of pests and treatment of plant diseases (Oerke and Dehne, 2004). Further, the use of CPP in many developing countries has freed women and children from lengthy, tedious and backbreaking manual weeding, with the time and energy released being used to advance education and the economic independence of rural communities (*e.g.* Srinivasan, 2003).

Yet agricultural production must be further increased considerably in the future to meet the food, feed and fuel demands of a rising human population. The United Nations estimated that by the year 2030, the world population will have increased to over 8 billion (UN, 2004). This growth is expected to be mostly urban and not rural, bringing with it a higher calorie demand per capita associated with dietary changes. This will further increase pressure on rural communities in the struggle to raise agricultural productivity to meet demand.

In addition, crops are increasingly being used to produce fuel (*e.g.* EC, 2007; US EPA, 2007). Hence, the challenge is how to meet this demand without further conversion of natural habitats of high biodiversity. The solution is to continue to increase the productivity of existing farmland. Syngenta helps farmers do this by reducing losses to pests and diseases and increasing yield through innovation in crop protection and seeds.

Need for Stewardship

However, CPP must be applied correctly to ensure safe, efficient and sustainable use. As a company, Syngenta are committed to ensuring that the sales and use of CPP are achieved in a sustainable way, for the long term future of our customers, the environment and our business. Hence, product stewardship – the ethical and responsible management of a CPP from its discovery through to its ultimate use and beyond – in compliance with the International Code of Conduct on the Distribution and Use of Pesticides (FAO 2005) forms a core element of Syngenta's corporate responsibility policy and commitments. Specifically, the FAO Code of Conduct notes that;-

> 3.4 Pesticide industry…should:
>
> 3.4.5 be capable of providing effective technical support, backed up by full product stewardship to field level, including advice on disposal of pesticides and used pesticide containers, if necessary;
>
> 3.4.6 retain an active interest in following their products to the end-user, keeping track of major uses and the occurrence of any problems arising from the use of their products, as a basis for determining the need for changes in labelling, directions for use, packaging, formulation or product availability.

In compliance with the FAO's Code of Conduct (2005), Syngenta's approach to product stewardship includes a commitment to support users – including growers and contract sprayers – through promotion of the safe and effective use of its products. A stepwise approach is used to develop programs targeted at specific needs of users in different countries and in different crop sectors.

The first phase starts with an audit to understand the way products are used in practice. The results enable us to select priority areas focused on activities associated with higher risks. Together with local partners solutions are defined which can be rapidly implemented. Initial mitigation measures include rational product choice and selection of targeted uses.

The second phase of the programme is implemented in collaboration with a broad group of local partners ranging from agricultural communities to health organisations. The objective of this second phase is to define measures to minimize the exposure and ensure their implementation. The Syngenta principles for the safe use of CPP are encapsulated in The 5 Golden Rules, and practical examples of ways to minimize exposure include farmers training activities, defining better working conditions, and developing improved application techniques. Although important, the use of personal protective equipment (PPE) is only factored in towards the end of the process when all other steps in risk reduction have already been addressed.

Finally, the lasting success of stewardship programs needs to be monitored, and one of the measures can be the development of adverse health incidents (AHI) recorded over time, provided it is done in a standardised way. This last step is important to continuously improve the programme effectiveness and consequently the safe use of CPP.

Product Stewardship by Syngenta

To examine the priorities set for stewardship programs, and to determine the safe use knowledge, attitudes and practices of those who spray crop protection products, Syngenta has commissioned independent market research since 2004. The surveys focused on users of knapsacks or hand held fixed line sprayers, moderate to very intensive use of CPP, and users generally considered at greatest risk, for example, smallholder farmers, contract sprayers, and plantation workers.

About 9,000 interviews in 26 countries were conducted with farmers who are growing a wide range of crops typical of their region. The questions ranged from purchase (transport and storage) and use (mixing and loading, spraying, knapsack maintenance) to pack disposal. Where best practice was not being observed, additional questions were asked to determine attitudes to safe practice.

There are variations across countries and certain practices are better developed than others. For example, Sri Lankan users were among the best in terms of personal hygiene practices while disposal of empty containers is a particular blind spot for some users in all countries. Sri Lanka was surveyed in 2005, and the key findings were;-

- Measuring practices were very good with 89% using a graduated container (78% global).
- Sri Lankan users were among the most likely to wash their sprayer, and among the best in terms of washing clothes and themselves after spraying.
- Disposal practices for empty containers were mixed. Many users punctured containers (18%, 4% global) or sold them rinsed for scrap (32%, 12% global). However 27% threw them away in the field or bushes (17% global).

In addition the surveys asked farmers about health incidents they had experienced. Most users had little or no health issues – 3% of users reported having experienced a health incident requiring medical attention in 2006 – and the majority of those effects occurred during spraying. Insecticides caused approximately 70% of these health incidents with a wide spread complaint on the level of smell. Among Sri Lankan users surveyed, their experience of CPP-related health incidents was similar to the global average for each severity of incident. This was also true for health incidents due to other causes such as machinery and wildlife.

Having identified the priority areas and issues – equipment maintenance and use of PPE especially during spraying, the second phase of the programme is to define measures to minimize the exposure and ensure their implementation. In 2006, Syngenta trained around 3.4 million farmers worldwide in safe handling, secure storage, and appropriate application of crop protection products. With extensive use of pictorial guides, simple-to-use training aids, and practical field demonstration, the comprehensive stewardship programs impart on growers best practices in CPP use in accordance with The 5 Golden Rules, but also with specific emphasis on better and safer knapsack spraying. For instance, the *Best Practice Guide for Knapsack Application* is widely distributed to ensure sprayer-operators know how to achieve better and safer results. The leaflet contains essential information on the “Do’s and Don’ts” – from sprayer maintenance, nozzle selection to correct application. The kit includes a Kalibottle to quickly and accurately calibrate correct sprayer output, along with a Dose Calculator to work out how much product to use.

The training sessions are conducted by Syngenta staff – all of whom have received appropriate safe use training beforehand – in collaboration with a wide range of partners, including government officials, extension workers, and distribution channel.

Finally, the lasting success of stewardship programs needs to be monitored, and one of the measures can be the development of adverse health incidents (AHI) recorded over time, provided it is done in a standardised way. To this end, Syngenta has initiated a new global medical stewardship campaign in 2004/05, to help standardize reporting of AHI relating to CPP use. The system is a key element in programs designed to address the root causes of such incidents through preventative education and training programs.

The Syngenta AHI Management initiative is focusing on representative areas in developed and developing countries and covers a broad range of users, from smallholder knapsack operators, to plantations and large-scale farming operations. The project involves establishing partnerships with hospitals, poison control centres and other authorities who already collect AHI data, and assisting them in gathering information in a consistent way. The system records not just the product involved, the type and severity of the incident, but also the possible link to exposure, the circumstances involved and the outcome. Improved reporting of AHIs should enable Syngenta to focus stewardship efforts on key issues, tailor and improve training programs and direct product development. By 2006, 42 countries are participating in this AHI Management initiative.

When the true risks of the use of CPP are put into the perspective of the enormous benefits to users, global food production and to soil conservation in sustainable agriculture, it is only possible to conclude that unique CPP give an enormous net benefit and that banning of the products would further disadvantage farmers in developing countries by removing valuable tools.

Misuse and Abuse

Unfortunately, deliberate self-harm (DSH), such as suicides by ingestion of CPP, is increasingly a public health problem across the developing world, including Sri Lanka (Eddleston and Phillips, 2004; Konradsen *et al.* 2006). The World Health Organization (WHO)/ IASP (International Association for Suicide Prevention) report of 2006 estimated that one third of all suicide deaths globally result from pesticide ingestion (Bertolote *et al.* 2006a), especially in rural communities where CPP are readily available in homes or in fields nearby (Bertolote *et al.* 2006b). The WHO has thus launched a global Pesticides and Health Initiative aimed at developing strategies to reduce the health impacts of CPP.

To this end, Syngenta has been implementing a number of stewardship programs, including introduction of a novel paraquat product with INTEON® technology, training in secure storage and provision of lockable boxes, and AHI monitoring and epidemiological study. The AHI records and study results both indicated that deliberate self-harm and accidental ingestion of CPP could be substantially reduced, and the new formulation improved overall survival of patients following paraquat ingestion.

Innovation and Partnerships

The INTEON® technology is essentially the introduction into the formulation of an alginate that converts to a gel under stomach acid conditions (Heyslings *et al.* in press). The alginate, plus increased emetic and a purgative reduce intestinal absorption of ingested paraquat, and hence enable reduced oral toxicity and safening of the paraquat formulation, sold as Gramoxone® by Syngenta. Gramoxone INTEON® has been launched in Sri Lanka in October 2004. Despite initial phase-separation issues, an on-going epidemiological survey there has found that INTEON® technology significantly improves the survival of patients following paraquat ingestion (Wilks *et al.* 2006). Survival was strongly associated with estimated ingestion volume, and the beneficial effect of INTEON® was apparent across the dose range.

In addition, Syngenta has supported and collaborated with a number of local and international organizations, such as Sumithrayo (a Sri Lankan suicide prevention NGO) and International Association of Suicide Prevention (IASP), in various secure access or safe storage initiatives (*e.g.* Pieris and Weerasinghe, 2005). The rationale behind is that intentional ingestion of CPP, as with all types of suicidal attempts, is largely an impulsive act. The availability of a means of suicide may be enough to trigger an attempt by an impulsive person who feels defeated and hopeless about the future (Williams and Pollock, 2000). Secure access of all chemical products is intuitively a method of reducing easy access and thus minimizing the risk of such a trigger. Indeed, WHO and IASP recommend secure access as a means of reducing deaths from intentional ingestion of agrochemicals (WHO, 2006).

Over the years there have been pilot projects in various parts of the world that indicate feasibility, acceptability, benefit and a likely reduction in incidents of intentional ingestion related to secure access of CPP; ensuring that products are safely stored away under lock and key when not in use. In one such project in Sri Lanka, there was a significant increase in the number of households keeping CPP from children between baseline and seven months after box distribution (64% vs. 89%) (Konradsen *et al.* 2007). Another example of a successful pilot secure access programme took place in 2002 in Samoa. There, various stakeholders including industry, the local suicide prevention agency Faatua La Ola (FLO), the ministries of agriculture and health, hospitals, media, retailers and farmers all worked together on a project that included the distribution of locked boxes and suicide helpline counseling. In the couple of years that followed the project, there was a halving in the number of deaths that arose from intentional ingestion of CPP (Salevao-Manutai, Director of FLO, *personal communication*). In Nicaragua, intensive farmer education projects from September 2002 to November 2004 where secure access amongst other safe use measures was emphasised also saw a great reduction in the number of reported cases of intentional CPP in Matagalpa and Jinotega (Nicaragua Poisons National Center 2005).

Conclusions

To conclude, excessive focus on issues of gross product misuse like intentional ingestion in suicide attempts can deflect attention away from the essential benefits that CPP bring. The CPP are an essential tool to maintain and enhance current farming yields. Therefore, the focus for everyone with the interests of farming and the needs of the world at heart must be the safe and effective use of CPP through responsible stewardship.

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UTILITY OF ALLELOPATHIC RICE CULTIVARS IN SUPPRESSING BARNYARDGRASS (*Echinochloa crus-galli*) AND REDUCING THE APPLICATION RATE OF MIXTURE OF BUTACHLOR + BENSULFURON-METHYL

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Abstract: A pot experiment and a herbicide bioassay were conducted to compare the barnyardgrass suppression by three rice varieties, *viz.* cv. Kouketsumochi (allelopathic), cv. Dongjinbyeo (non-allelopathic) and cv. K21 (newly bred potent allelopathic). The consequence of applying less herbicide to these cultivars was also evaluated. The results showed that the weed suppression potential varied among the three rice cultivars. Kouketsumochi was the most allelopathic rice cultivar; K21 was intermediate, while Dongjinbyeo was the least allelopathtic. The allelopathic effect of Koukishumuchi, K21 and Dongjinbyeo on barnyard grass height was about 45%, 31% and 20%, while on barnyard grass plant weight was about 35%, 30% and 20%, respectively. These effects were converted into herbicide rate according to the equation of toxicity regression. For the inhibition of plant height and weight, the value of Kouketsumochi is 10.6 mg/l and 8.4 mg/l, for K21 6.3 mg/l and 6.9 mg/l, and for Dongjinbyeo 3.7 mg/l and 4.0 mg/l respectively. By comparing the value of LD50 under conditions without rice, herbicide application rate has been reduced around 20-50%. Under conditions with rice growth, for Kouketsumochi and K21, herbicide rates can be reduced about 30% and 20% than Dongjinbyeo. The results suggested that rice cultivars with allelopathic potential can reduce herbicide application for weed management, and thus promote a more eco-friendly rice cropping system.

Key words: Rice allelopathy, barnyardgrass, reduce rates of herbicide

Introduction

The allelopathic potential of rice has received a great deal of attention since Dilday *et al.* (1991) identified rice cultivars exhibiting allelopathic potential against ducksalad [*Heteranthera limosa* (Sw.) Willd]. The use of allelopathic crops can definitely reduce the cost of weed control. This technology of allelopathy, which is seed-based technology is more easily transferable to the low-input management systems prevailing throughout most Asian rice-farming systems. In previous field and laboratory research at Daegu Korea, Kouketsumochi reduced density and growth of barnyard grass to a greater extent than the cultivar Donjinbyeo when herbicide was not applied (Kim, 2000). Similar efforts have been made to identify and develop weed-suppressive rice in upland rice and wild rice species (Dingkuhn *et al.* 1999; Fisher *et al.* 2001; Fofana and Rauber 2000; Garrity *et al.* 1992; Guo *et al.* 2005; Johnson *et al.* 1998; Jones *et al.* 1997; Ma *et al.* 2006).

Herbicide, mixture of bensulfuron-methyl and butachlor, has long been used to control a wide array of grasses and broadleaved weeds in rice production during recent times. Bensulfuronmethyl, as ALS inhibitor herbicide, is a sulfonylurea type which has been intensively used in the world since 1990. Currently, ALS inhibitor resistant weeds are increasing more rapidly than for any other herbicide mode of action (Source: Dr. Ian Heap, http://www.weedscience.com). On the other hand, butachlor is a chloroacetamide type herbicide, which is considered as a cell division inhibitor (Matthes *et al.* 1998).

Barnyardgrass evolved resistance to butachlor in China (Huang and Lin, 1993; Huang and Wang, 1998), but the resistance mechanism is unknown. Mixing of herbicides with different modes of action is one of the important methods to delay the development of resistance and may reduce the usage of herbicide. In this research, these three rice varieties, *viz*. cv. Kouketsumochi (allelopathic), cv. Dongjinbyeo (non-allelopathic) and cv. K21 have been used to compare the barnyard grass-suppressive abilities and to evaluate the resulting reduced rates of herbicide.

Materials and Methods

The three rice varieties viz. cv. Kouketsumochi (allelopathic), cv. Dongjinbyeo (non-allelopathic) and cv. K21 cultivars were used as donor plants, and the weed barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) as a target plant. The K21 is a newly bred potent allelopathic rice cultivar developed from Donjinbyeo (female parent), and Koukishumuchi (male parent) in Korea (Ma *et al.* 2006). Herbicide applied was the mixture of butachlor 2.5% and bensulfuronmethyl 0.17%, which is widely used in Korean rice culture at 800 g (a.i.) per hectare. Soil used was commercial paddy rice soil with full nutrients and pH of 4.5-5.8. The experiment was conducted in pots of 10 cm diameter and 7 cm deep.

Combined effect of rice varieties and herbicide on barnyard grass
The experimental design was a two-factor factorial randomized designs with 3 replications. The factor A was three rice cultivars and no rice pot growth with barnyard grass; the factor B was herbicide rates of 0, 15, 30 and 60 g/ha, which were applied at 2 days after sowing. There was a check in which barnyard grass grew alone without herbicide application.

The pot was filled with rice commercial soil. 15 rice seeds and 20 barnyard grass seeds were sown around 0.5 cm deep in each pot. Herbicide was applied on the surface of soil. The soil moisture was maintained around 50 % (w/w) during the first 5 d, and then 20 or 30 ml of water was added everyday. The pots were placed in a growth chamber at 25°C, 14 hours light and at 22°C, 10 hours dark condition. After 14 days of herbicide application, the height and fresh weight of 5 or 10 barnyardgrass plants per pot were measured to calculate the inhibitory rate.

Bioassay of herbicide to set up the equation of toxicity regression
Pots were filled with rice commercial soil. 20 pre-geminated weed seeds were sown in pots without or with 10 uniform pre-geminated rice seeds. Herbicide was applied on the surface of soil at the gradient of 0, 0.625, 1.25, 2.5, 5.0, 10 and 20 mg/l. The soil moisture was kept around 50% (W/W) during the first 5 days, and then 20 ml water was added everyday. The pots were placed in growth chamber at 25°C, 14 hours light and 22°C, 10 hours dark. After 14 d, height and fresh weight of 5 or 10 weeds were measured to calculate the inhibitory rate and to set up a toxicity regression equation. The experiment involved a complete randomized design with 3 replications.

Differentiation of allelopathic potential among rice varieties
The experiment was carried out in the same condition as above, which was a complete randomized design with 3 replications. Seeds of rice and weed were sown in pots as 15 rice seeds and 25 barnyardgrass seeds for each pot. After 5 days of sowing, the small or later germinated seedlings were removed to keep rice and weed as 10 seedlings per pot. Rice and barnyard grass were grown together for 2 weeks, and then the height and fresh weight of barnyard grass were measured. Barnyard grass grew alone as a check.

Analysis
The effect on barnyard grass at various herbicide gradients and rice cultivars were evaluated by IR (inhibitory rate), and analysis of variance was done by DPS (6.5) program.

$$IR = 100 \times (1 - tr/ck)$$

Where tr is barnyard grass grown with rice or herbicide applied; ck presents no herbicide applied and barnyard grass grown alone. The equation of toxicity regression was set up as Y=ax+b, where Y is inhibitory rate, X is dosage of herbicide. And then according to the equation of toxicity regression, LD_{50} was calculated to compare how much rate of herbicide was reduced. The allelopathic potential was evaluated by inhibitory rate (IR); calculation was similar to the herbicide inhibitory rate as mentioned above. IR was converted into the relative herbicide rate according to the toxicity regression equation of herbicide.

Results and Discussion

Combined effect of herbicide and rice cultivars on barnyard grass

From Tables 1 and 2, the results indicate that there significant differences existed among the three rice cultivars and herbicide rates. In addition, there was an interaction between herbicide rate and rice cultivars, which meant that the herbicide bioactivity varied when applied on different rice cultivars. The three rice cultivars had different suppressive abilities on barnyard grass and reduction of weed growth. Kouketsumochi was more suppressive than its descendant K21, while Dongjinbyeo was the least suppressive under both conditions of using or non-using herbicide (Figure 1).

Table 1. Analysis of variance for barnyard grass plant height inhibitory rate

Variance source	Square sum	df	Average of variance	F
Block	0.0010	2	0.0005	0.6745
A factor	1.1546	3	0.3849	494.6600**
B factor	1.9822	3	0.6607	848.7459**
AxB	0.1309	9	0.0145	18.6526**
Error	0.0233	30	0.0008	
Total	3.2920	47		

* Factor A was different rice cultivars; factor B was herbicide rates at 60g/ha, 30g/ha. ** p<0.01

Table 2. Analysis of variance for barnyard grass plant weight inhibitory rate.

Variance source	Square sum	df	Average of variance	F
Block	0.0218	2	0.0109	9.4336
A factor	0.3744	3	0.1248	107.9276**
B factor	3.3612	3	1.1204	968.9407**
AxB	0.0855	9	0.0095	8.2195**
Error	0.0347	30	0.0012	
Total	3.8776	47		

* Factor A was three rice cultivars; factor B was herbicide rates at 60g/ha, 30g/ha. ** p<0.01

Weed growth reduction by herbicide and allelopathic effect among rice cultivars

When herbicide was used with or without rice, weed growth reduction trends among the herbicide rates were similar. Decreased herbicide rates generally reduced the effect on weed height and weight, and the bioactivity reduced as the chemical rates decreased. But, all three cultivars reduced the barnyard grass height and weight over no-rice-treatment levels, especially under lower herbicide application, which suggested that rice cultivars are suppressing the weed. By comparing the value of LD_{50} obtained from toxicity regression equation, under no rice condition, herbicide values of LD_{50} on plant height and weight were

11.83mg/l and 12.6 mg/l, respectively. However, when herbicide was used with the growth of rice cultivars, the value was decreased to 5.1 mg/l and 6.48 mg/l for Koketsumochi, 5.87 mg/l and 8.51 mg/l for K21, 7.82 mg/l and 10.26 mg/l for Dongjinbyeo, respectively. Therefore, herbicide rate was lower by around 20-50% than that of without rice treatment, and lower by around 20-30% than that of the non-allelopathic rice cultivar, Dongjinbyeo (Tables 3 and 4). These data indicate that allelopathic rice cultivars can reduce the need for herbicide application. Thus, herbicide application rate for barnyard grass in these cultivars could be reduced.

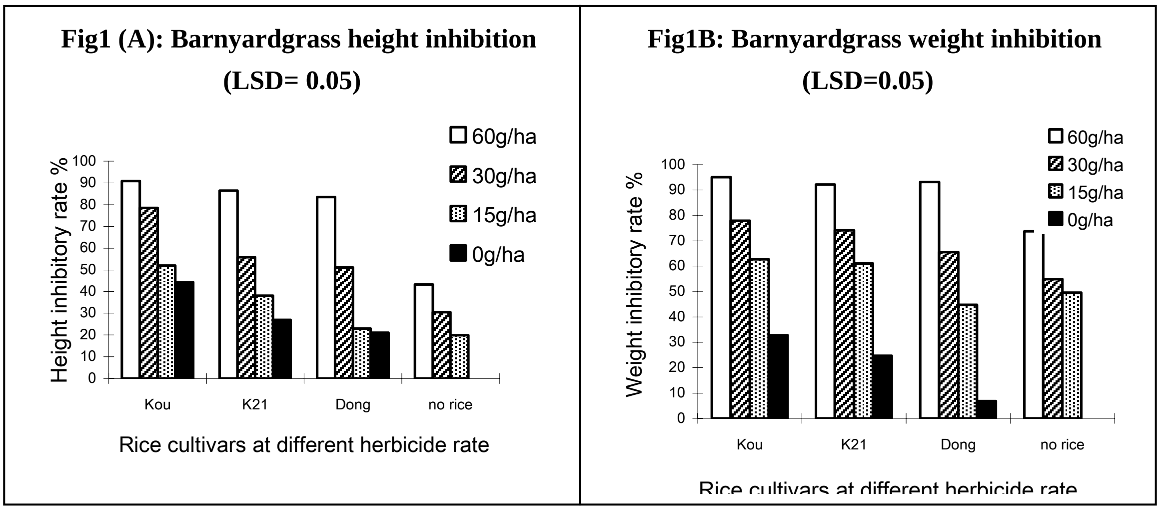

Figure 1 Growth parameter of barnyardgrass as influenced by rice cultivars and herbicide rates

Table 3. Herbicide bioactivity on barnyard grass (2 weeks, no rice).

Dosage a.i mg/l	Height (cm)	Inhibitory rate of height (%)	Weight (g)	Inhibitory rate of weight (%)
0	30.34	0	6.8536	0
0.625	25.29	16.64	6.1731	9.93
1.25	25.53	15.85	6.1220	10.67
2.5	25.14	17.14	5.9390	13.34
5.0	21.10	30.45	4.7610	30.53
10.0	17.34	42.85	3.7452	45.35
20.0	6.89	77.29	1.9292	71.85
Equation of toxicity regression	y = 3.4434x + 9.2337 R2= 0.9645 LD_{50}=11.83mg/l		y = 3.4459x + 6.5697 R2 0.9673 LD^{50}=12.60mg/l	

Table 4. Effect of allelopathic potential rice and herbicide on barnyard grass

Dosage ai mg/l	K21 Inhibitory rate %		Dongjinbyeo Inhibitory rate %		Kouketsumochi Inhibitory rate %	
	Height	Weight	Height	Weight	Height	Weight
0	0	0	0	0	0	0
1.25	17.19	0.89	11.27	-4.28	24.11	15.93
2.5	33.99	6.62	25.56	10.65	40.32	31.67
5.0	58.20	38.7	44.89	26.29	59.09	55.08
10.0	69.37	56.03	55.92	47.39	77.96	63.27
Equation	Y=6.7336x	Y=6.209x	Y=5.5278x	Y=5.223x	Y=7.207x	Y=6.158x

	+10.499	-2.8357	+6.7988	-3.5762	+13.27	+10.098
coefficient	R2=0.8693	R2 = 0.9299	R2=0.8951	R2 =0.954	R2=0.8876	R2=0.8508
LD^{50} mg/l	5.87	8.51	7.82	10.26	5.10	6.48
Reduce rate I	50.38	32.46	33.90	18.57	56.89	48.57
Reduce rate II	24.94	17.06	-	-	34.78	36.84

*Reduce rate I: Compared to herbicide effect on barnyardgrass: height LD50=11.83mg/l and weight LD50=12.60 mg/l, **Reduce rate II: Compared to the LD^{50} of herbicide effect on Donjingbyeo: height LD50=7.82 mg/l, and weight LD50=10.26 mg/l

Weed growth reduction by rice cultivars

When herbicide was not applied, weed suppression potential varied among the three rice cultivars. The results in Table 5 show that Kouketsumochi was the most allelopathic rice cultivar followed by K21, while Dongjinbyeo was the least allelopathic. The allelopathic effect of Koukishumuchi, K21 and Dongjinbyeo on barnyard grass height was about 45.8%, 31% and 22%, while on barnyard grass plant weight was about 35.4%, 30.5% and 20.4%, respectively. These allelopathice effects were converted into and compared to herbicide rates according to the toxicity regression equation in Table 3. For plant height and weight, the value of Kouketsumochi's effect has been equated to herbicide as 10.6 mg/l and 8.4 mg/l, for K21 as 6.3 mg/l and 6.9 mg/l and for Dongjinbyeo as 3.7 mg/land 4.0 mg/l respectively (see Table 5). The results have suggested that chemical control of barnyard grass in cultivars having allelopathic potential could be economical.

Table 5. Effect of allelopathic potential rice cultivars on barnyard grass.

Parameter	K21	Donjinbyeo	Kouketsumochi	Check *
Plant height (cm)	21.04	23.80	16.54	30.51
Inhibition of height (%)	31.04	21.99	45.79	-
Plant weight (g)	0.3218	0.3688	0.2993	0.4631
Inhibition of weight (%)	30.51	20.36	35.37	-
Amount of herbicide on plant height (mg/L)	6.33	3.70	10.61	-
Amount of herbicide on plant weight (mg/L)	6.95	4.00	8.36	-

*Without rice

Weed growth reduction by rice could be due to rice-weed competition and/or allelopathy. Plant height is one of the most important traits determining the competitive ability of plants, followed by early growth rate and rate of tillering (Bastiaans *et al.* 1997). Therefore, plant height and weight as investigative parameters is feasible.

Barnyard grass (*Echinochloa crus-galli*) is rated as the world's worst weed of rice (http:www.weedscience.com). Not only found in rice, but it is fairly common to all agricultural crops, especially on moist and rich soils. Barnyard grass can produce over 1 million seeds, and remove as much as 80 percent of nitrogen from the soil (Fred, 2000). Minimum lethal dosage (MLD) and 50% lethal dosage (LD50) is commonly used to evaluate the bioactivity of herbicide on target plants. Index of relative toxicity is used to indicate the bioactivity of two herbicides, which were reported by Sampford (1952), Blachman (1952) and Gowing (1959). In this study, barnyard grass was used as the target plant and a more efficient and sensitive dosage LD50 was introduced to determine rice cultivars with allelopathic effects, which is representative and has actual guidance to apply allelopathic cultivars for weed management in rice production.

Herbicides applied at rates below that recommended by the manufacturer can increase economic returns in rice production (Jordan, 1997a; 1997b). Gealy *et al.* (2003) reported that, with or without propanil, high competitive cultivars such as PI312777, Guichao and Teqing, suppressed barnyard grass more and consequently produced higher grain yields than did U.S. cultivars such as Kaybonner, Lemont and Cypress. This resulted in higher net returns as compared to the commercial cultivars. This experiment was conduced under laboratory conditions and the results suggest that weed-suppressive rice cultivars K21 and Kouketsumochi could reduce herbicide rates by 20-30%. Thus, combining weed-suppressive rice cultivars with reduced herbicide rates could be an economical alternative to the herbicide-intensive weed management programs, and promote a more eco-friendly rice cropping system.

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THE APPLICATION OF TRIFLURALIN FOR DRY SOWN RICE

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Abstract: Field trials were carried out to determine the phytotoxic effect of trifluralin in dry sown rice by applying herbicides into paddy fields during puddling or after puddling. Rice seeds were dry-sown into the plots 20 days after herbicide application. The minimal application rates of trifluralin resulting in significant reduction in the rice stand and grain yield were used as criteria for distinguishing phytotoxicity. Application of trifluralin at 375 g a.i/ha did not reduce the number of rice plants and grain yield in both treatments. In contrast, the rate of 750 g a.i./ha, caused a significant reduction of rice plants and grain yields when applied during puddling. At the highest rate of 3000 g a.i./ha, applied after puddling, the reduction in number of rice stands and grain yield was very significant. The normal application rate of trifluralin at 375-750 g a.i./ha after dilution into flooded fields after puddling could be used in dry sown rice for grass weed control.

Key words: Dry sown rice; phytotoxicity; trifluralin

Introduction

Trifluralin is a very effective herbicide with a considerable of control grass weeds when incorporated into soil (Bateman and Walker, 1985) especially to control barnyard grass (*Echinochloa crus-galli*). Due to its high phytotoxic effect on grain crops such as rice, farmers generally do not use trifluralin in paddy fields. In the past decades, direct seeded rice culture has become increasingly attractive as an alternative to transplanted rice (Ahmed and Azizul Islam, 1983). Hence studies were conducted to determine the phytotoxic effect of trifluralin on transplanted (Zhang, 1989) and direct seeded rice (Zhang, 1987). This study was a continuation of the program on direct seeded rice.

Materials and Methods

Experiments were carried out at the Beijing police army farm in 1992 using Trifluralin, an emulsifiable concentrate containing 480 g. a.i./l. produced by Elanco Products Co. In all field experiments, a hybrid rice cultivar (*Oryza sativa* var. *japonica*) “Liyou 57” was used and the soil type was a sandy clay loam.

The trials were laid out in randomized plots, each plot size was 5 m × 4 m. with five replications. Depending upon the application rates (g a.i./ha.), the diluted trifluralin emulsions in required concentration were uniformly sprayed by a knapsack sprayer with spray volume of 1000 l./ha. The experiments were separated into two application treatments . In treatment 1, the paddy fields were puddled, herbicide was applied into the plots and incorporated before leveling. In Treatment 2, the soil was puddled, leveled and the herbicide was applied into the flooded plots and was not incorporated. Each treatment received four rates of trifluralin application and also contained an untreated check plot. At 20 days after the herbicide had been applied into the plots, the rice seeds were dry sown in rows 25 cm apart at a seeding rate of 150 kg/ha and a seeding depth of 2-3 cm. When the rice plants were at the 2-3 leaf stage, plots were flooded. Thereafter, alternating flooding was adopted to maintain a shallow layer of water and drying and other cultural practices were followed for optimum crop growth until maturity.

The observations on crop growth and grain yield including the phytotoxic reduction in number of rice plants at 2-3 leaf stage were made within a 1 m^2 quadrat at 5 locations in each plot . Grain yields were also determined from these quadrats

The data were statistically analyzed and the minimal application rates of trifluralin that caused phytotoxic effects on rice in the treatments were compared with the untreated plots in terms of reduction in number of rice plants and grain yield.

Results and Discussion

In plots that were leveled after the application of the herbicide, trifluralin at 187 g or 375 g a.i./ha did not reduce the number of rice plants and grain yield. The application of trifluralin at the rate of 750 g a.i./ha, reduced the rice plants and grain yield significantly. The highest phytotoxic effect was observed at 1500 g a.i./ha (Table 1.).

Table 1. Effect of trifluralin applied during field puddling on dry sown rice

Rate of application g a.i./ha.	Seedling growth Plants/m^2	Grain yield kg/plot
187	336 a	9.64 a
375	340 a	9.40 a
750	299 b	7.80 b
1500	222 c	7.36 c
Untreated check	323 a	8.97 a

Note: The rice was direct seeded 20 days after trifluralin application. Thiobencarb was applied for barnyard grass control in untreated check plots. Within a column, means followed by the same letter are not significantly different (p=0.05).

The phytotoxic effect of trifluralin on direct seeded rice when applied after leveling was as follows: trifluralin at 375 or 750 g a.i./ha did not cause any significant reduction in number of rice plants and grain yield, while an application rate of 1500 g a.i./ha, reduced the plant numbers without a reduction in yield (Table 2). At the highest rate of 3000 g a.i./ha, the reduction in number of rice stands and grain yield was significant.

Table 2. Effect of trifluralin applied to flooded plots after puddling on dry sown rice

Rate of application a.i./ha.	Seedling growth Plants/m^2	Grain yield kg/plot
375	322 a	9.43 a
750	310 a	9.14 a
1500	274 b	9.18 a
3000	219 c	7.27 c
Untreated check	318 a	9.11 a

Note: The rice was direct seeded on the time of 20 days after trifluralin application. Thiobencarb was applied for barnyard grass control in untreated check plots. Within a column, means followed by the same letter are not significantly different (p=0.05).

The application rate of trifluralin at 375 g a.i./ha did not cause any phytotoxic effect on rice emergence and grain yield when applied during or after puddling the fields. The minimal application rate of trifluralin resulting in a significant phytotoxic reduction in rice stand and grain yield was at 750 g a.i./ha when trifluralin was applied during puddling. At a rate of trifluralin at 1500 g a.i./ha applied after puddling, there was significant in reduction of rice plants, while the minimum rate of trifluralin that caused a significant phytotoxic reduction in rice plants and grain yield was 3000 g a.i./ha, when trifluralin was applied after the fields had been puddled. The criteria for distinguishing phytotoxicity of trifluralin to direct seeded rice were clear from the trial plots. The normal application rate of trifluralin at 375-750 g a.i./ha after dilution in water into flooded fields after puddling could be used in dry sown rice for grass weed control. The rice should be seeded in the dry fields, 20 days after trifluralin application. The paddy fields should be irrigated again at the 2 – 3 leaf stage of rice seedlings and cultural practices followed to maintain optimum crop growth until rice maturing.

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THE IMPACT OF IMAZAPIC RESIDUE ON SUCCEEDING CROPS IN CHINA

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Abstract: The experiments were conducted in China from 1996 to 2000 to evaluate the safety of imazapic residue to succeeding crops. The peanut was sown in a sandy soil according to local practice, and the pre-emergent herbicide imazapic was applied at the rates of 0, 450, and 900 ml/ha, respectively. Four months after treatment (MAT), the peanut was harvested and wheat, canola and spinach were sown, and 12 MAT, maize, cotton, water melon and cucumber were sown in the specific pots in the specific plots. The emergence rate, plant height and yield were measured for each crop. Even at the dosage of 450 ml/ha, the imazapic residue could significantly affect the growth of the canola and spinach at 4 MAT, and cucumber and water melon at 12 MAT. Cotton did not show any sysmptoms of herbicide residue effects at 4 MAT under the dosage of 450 ml/ha. The herbicide residue did not have any detrimental impact on wheat under at dosage of 900 ml/ha at 4 MAT, and on at 12 MAT.

Key words: Herbicide residue, imazapic, succeeding crops

Introduction

Imazapic is a imidazoline herbicide with a high soil and foliar, and is registered for use in soybean and peanut cultivations. It controls many troublesome weeds such as *Cassia occidentalis* L., *Chenopodium album* L., *Ipomoea* spp., *Amaranthus* spp., *Cyperus rotundus* L. and *C. esculentus* L (Grichar *et al.* 1994, Wilcut *et al.* 1995). In addition, imazapic provides control and suppression of *Desmodium tortuosum*, *Sorghum halepense* L., *Panicum texanum* Buckl., and *Senna obrusifolia* L., which are not adequately controlled by imazethapyr (Grey *et al.* 2001). In crop rotation, imidazolinone herbicides must be applied cautiously. Monks and Banks (1991) observed slight corn injury and severe cotton injury due to the residue of imazaquin applied to soybean. Renner *et al.* (1988) observed significant corn injury from residues of imazaquin applied in the previous 1-2 years. Johnson *et al.* (1992) reported a significant injury to rice from residues of imazethapyr applied in the previous year to soybean. Rotational crops such as sugar beet, canola, cauliflower and lettuce can also be damaged when planted following imazethapyr (Tickes, 1991). There were some cases reported from northeast of China on the negative impact imazethapyr residues in rice.

The persistence of the imidazolinones in soil is influenced by the degree of adsorption to soil, soil moisture content, temperature, and amount of exposure to sunlight. The degree of soil adsorption increases as organic matter content increases and pH decreases. The primary mode of decomposition is by microbial degradation. Dissipation is most rapid in soils with temperature and moisture contents that favor microbial activity (Grichar, 2004).

Imazapic apply the registration in soybean in China, and it is very necessary to carry out some research to evaluate the safety to the following crops which may be susceptible to the imidazolinone herbicide. The objectives of this research are to evaluate the safety of imazapic to local following crops, and give some suggestions to farmers.

Materials and Methods

The seed of peanut, wheat, corn, sweet potato, cotton, water melon, cucumber, canola and spinach were supplied by the Shandong Seed Company, and the imazapic (BASF, Germany;

240 g/l aqueous solution) was used. The Linong sprayer with a span nozzle was used in the experiment.

The seed of the peanut was sowed in the sandy soil according to local practice, and the pre-emergent herbicide imazapic was applied at the 0, 450, and 900 ml/ha, respectively. The pot size was 74 m^2, and the spray volume was 600 l/ha. Four montha after treatment (MAT), peanut was harvested, and the wheat, canola and spinach were sown, and at12 MAT, maize, cotton, water melon and cucumber were sowed in separate pots. Watering, fertilization and cultivation were done according to local practice.

The emergence rate, plant height and yield were measured for every crop, and the experiment was repeated four times from 1996 to 2000 in Tai'an city of the Shandong province, China. The data were subjected to analysis of variance (ANOVA) using Statistical Analysis System (SAS) software.

Results and Discussion

Table 1 indicates that the treatments did not have a significant impact of imazapic on seedling emergence, plant growth and yield of wheat, even tough there was slight visual injury to the crop at the dosage of 900 ml/ha.

Table 1. The effect of imazapic on the growth of wheat

Treatment (ml/ha)	Emergence rate (%)	Plant height (cm)	Yield (kg/ha)	Visual injury
0	95 a	60.05 a	7300 a	No
450	96 a	59.50 a	7038 a	No
900	94 a	59.45 a	6808 a	slight

Within a column, means followed by the same letter are not significantly different (p=0.05).

There was a significant difference ($p<0.05$) among treatments on the measured parameters of canola, except the rate of seedling emergence (Table 2). The imazapic had a strong negative effect on the plant height and yield of the canola, and there was a serious visual injury to the crop at the dosage of 900 ml/ha. The imazapic at the dosage of 450 ml/ha was safe to canola when planted as a following-crop at 4 MAT. Imazapic residues significantly affected the emergence rate, plant height and yield of spinach, and there were middle and serious visual injury to the crop (Table 3). Table 4 shows that imazapic residues did not affect the growth of maize, when the crop was planted as a follow-crop.

Table 2. The effect of imazapic on the growth of canola.

Treatment (ml/ha)	Emergence rate (%)	Plant height (cm)	Yield (kg/ha)	Visual injury
0	95 a	8.29 a	6.3 a	No
450	96 a	7.25 b	5.2 a	slight
900	94 a	0 c	0 b	serious

Within a column, means followed by the same letter are not significantly different at p=0.05.

Table 3. The effect of imazapic on the growth of spinach.

Treatment (ml/ha)	Emergence rate (%)	Plant height (cm)	Yield (kg/ha)	Visual injury
0	95 a	8.79 a	633.33 a	No
450	63 b	8.05 a	626.67 a	middle
900	33 c	3.28 b	57.33 b	serious

Within a column, means followed by the same letter are not significantly different (p=0.05).

Table 4. The effect of imazapic on the growth of maize.

Treatment (ml/ha)	Emergence rate (%)	Plant height (cm)	Yield (kg/ha)	Visual injury
0	93 a	218.00 a	6427.5 a	No
450	94 a	213.17 a	6457.5 a	No
900	90 a	211.76 a	6375.0 a	No

Within a column, means followed by the same letter are not significantly different (p=0.05).

Imazapic residues significantly affected ($p<0.05$) the seedling emergence rate, plant height and yield of the cucumber, with middle and serious visual injury to the cucumber.

Table 5. The effect of imazapic on the growth of cucumber

Treatment (ml/ha)	Emergence rate (%)	Plant height (cm)	Yield (g/plant)	Visual injury
0	92 a	132.60 a	1505 a	No
450	41 b	100.00 b	1478 a	middle
900	33 c	74.40 c	1260 b	serious

Within a column, means followed by the same letter are not significantly different (p=0.05).

Table 6, indicates that imazapic residues did not have a strong negative effect on the plant height and yield of cotton, but the seedling emergence was affected with a slight visual injury to the cotton at the dosage of 450 ml/ha, where there was crop recovery, later.

Table 6. The effect of imazapic on the growth of cotton.

Treatment (ml/ha)	Emergence rate (%)	Plant height (cm)	Yield (g/plant)	Visual injury
0	89 a	135.76 a	553.6 a	No
450	71 b	130.68 a	531.6 a	slight
900	57 c	126.96 a	460.8 b	middle

Within a column, means followed by the same letter are not significantly different (p=0.05).

There was not significant difference of imazapic residues on the growth of the sweet potato (Table 7), even though there was slight visual injury to the cotton at the dosage of 450 ml/ha. However, imazapic residues had a significant effect on the growth of water melon even after 12 MAT.

Table 7. The effect of imazapic on the growth of sweet potato.

Treatment (ml/ha)	Emergence rate (%)	Plant height (cm)	Yield (kg/plant)	Visual injury
0	92 a	224.67 a	1.26 a	No
450	91 a	216.84 a	1.09 a	slight
900	78 b	80.42 b	0.71 b	middle

Within a column, means followed by the same letter are not significantly different (p=0.05).

Table 8. The effect of imazapic on the growth of water melon.

Treatment (ml/ha)	Emergence rate (%)	Plant height (cm)	Yield (g/plant)	Visual injury
0	89 a	193 a	900 a	No
450	68 b	162 b	710 b	middle
900	45 c	126 c	610 c	serious

Within a column, means followed by the same letter are not significantly different (p=0.05).

Conclusions

The persistence of the imazapic was not short. Even though the dosage was 450 ml/ha, the residue imazapic could affect the growth of the canola and spinach seriously after treatment four months. It was safe to wheat under the dosage of 900 ml/ha after treatment four months. Even though the dosage was 450 ml/ha, the residue imazapic could affect the growth of the cucumber and water melon significantly after treatment twelve months. It was safe to maize under the dosage of 900 ml/ha and to the cotton under the dosage of 450 ml/ha after treatment twelve months. It is not safe to plant the above susceptible crop after treatment of imazapic within the time. The degradation of the imazapic could be affected by many factors. It is very helpful if the residue periods of the imazapic could be studied under different types of soil and climates.

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ESTIMATION OF THE MINIMUM LETHAL DOSAGE OF BENTAZONE AND ATRAZINE USING THE PLANT PHOTOSYNTHETIC METER

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Abstract: The experiment was conducted in a green house of the Beijing, China in order to determine the Minimum Lethal Herbicide Dosage (MLHD) of photosynthesis-inhibiting herbicides, bentazone and atrazine, on three weed species and safety to the maize with Plant Photosynthetic Meter (PPM). The results showed that the diagnostics of weeds caused by bentazone could be observed 2 days and that by atrazine could be observed 6 days after application (DAA). Weed growth was inhibited later on. Both PPM values of weed leaves and weed biomass (shoot fresh weight) decreased as herbicide dosage increasing. However, the susceptibility of weed species to the mixture of bentazone and atrazine was different among weed species. *Amaranthus retroflexus* was very susceptible to the herbicide, and *Echinochloa crus-galli* was relatively less susceptible to the herbicide, and *Digitaria sanguinalis* was not susceptible to it. The MLHD could be calculated by weed biomass 14 DAA of the herbicide. There were close relationship between weed biomass reduction rate and PPM values of weed leaves 2-5 days after treatment. This indicates that the PPM value measured in early days after treatment could give reasonable prediction on weed control efficacy. The mixture of bentazone and atrazine usually provided 85-90% control by weed biomass when PPM value was about 20. The MLHD of bentazone and atrazine should be the dosage when PPM value is below 20 measured on the 2-5 day after treatment.

Keywords: MLHD, PPM, bentazone, atrazine, weed control

Introduction

The Minimum Lethal Herbicide Dose (MLHD) technology connected with the measurement of Plant Photosynthetic Meter (PPM) was developed by Plant Research International (PRI) in the Netherlands (Ketel and Lotz, 1997). It is a promising decision support system leading to the use of lower rates of photosynthesis-inhibiting herbicides. Based on simple and rapid measurements of plant photosynthetic activity, this method can predict whether a weed would be controlled or not with in a short time usually 2 to 4 days after application of herbicides and could help to calculate MLHD. The Plant Photosynthetic Meter (PPM) being developed by EARS Company is an electronic instrument for measuring the photosynthesis of plants. In the photosynthetic process carbon dioxide (CO_2) and water (H_2O) are converted into glucose (CH_2O) and oxygen (O_2). The energy required for the reaction is provided by light.

$$CO_2 + H_2O + \text{light}\text{-------}> CH_2O + O_2$$

The light reaction of photosynthesis takes place at the thylakoid membrane inside the leaf chloroplasts. This membrane consists of fats and proteins, in which clusters of some hundreds of chlorophyll molecules are embodied. These so-called photo systems consist of a reaction centre and an antenna. When the leaf is exposed to light, the chlorophyll antennae absorb photons. On absorption of a photon an electron is excited, *i.e.* is shot into a higher orbit. This "exciton" roams through the antenna. Its lifetime however, is short. The exciton can loose its excess energy in three ways (Figure 1). The exciton is swallowed by the "reaction centre" and participates in photochemistry. The exciton looses its extra energy while developing heat. The exciton returns to a lower orbit, emitting a photon (fluorescence).

These three possibilities together represent 100% of the absorbed light. The fraction of absorbed light that is used for photosynthesis is called the photosynthesis yield, photosynthetic light use efficiency or "quantum yield" (ΦP). The photosynthesis yield of a plant varies without visual sign. In the dark, the quantum yield of photosynthesis reaches the highest value, which is the same for almost all plants: 82%. At daytime this value decreases with increasing light level.

The plant photosynthetic meter (PPM) permits a prompt second herbicide application in case of failure. This last element ensures that even though minimal doses of herbicides have been employed, and there is a guarantee that the treatment will be successful in eliminating the weeds. Such a guarantee greatly contributed to the rapid and massive adoption of this MLHD methodology by Dutch farmers, agricultural contractors and others. The number of users is constantly increasing and demand for improvements and further developments is now coming, as well, from other European and Asiatic countries.

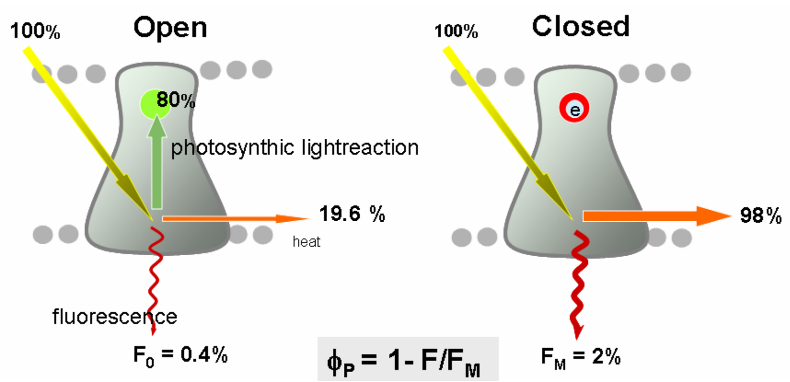

Figure 1. The PPM could predict the plant photosynthesis yield by measurement the fluorescence

The objectives of the experiment were to know whether PPM value of weed leaves could predict the efficacy of mixture of the bentazone and atrazine and the safety to the maize, and could determine the MLHD table for the three species of the weed.

Materials and Methods

The seed of *Echinochloa crus-galli*, *Digitaria sanguinalis* and *Amaranthus retroflexus* were supplied by the Shenyang Chemical Research Center of Liaoning province, and the Plant Photosynthesis Meter (PPM) was invented by EARS Company, the Netherlands. The sprayer 3WPSH-500D was made in China. The germinated seed of *Echinochloa crus-galli*, *Digitaria sanguinalis*, *Amaranthus retroflexus* and *Zea mays* were placed separately in the pots (10 cm diameters x 10 cm height), filled with loam soil and were incubated in a growth chamber. The cultivated temperature was about 25°C day/15°C night, and at 60% moisture.

Atrazine (385g/l, SC), and bentazone (480 g/l, SL) was used for the experiment. The proportion of the tank-mixed herbicide was 1 to 1, and the tank mixture was applied as shown in Table 1, and all treatments were repeated 8 times. Spray volume was 500 l/ha, and the spray pressure was controlled at 0.3 M Pa. At 0, 1, 2, 3, 4, 5, 6, 8, 10, and 12 DAA, the photosynthetic yields of weeds and maize were measured with the PPM. Weed fresh weights (biomass) were measured at 14 DAA. The maize plant heights were measured at 0, 3, 5, 7, 10 and 12 DAT. The ED_{90} for weeds were calculated using the herbicide dosage and weed biomass. The experimental data were subject to analysis of variance (ANOVA) using Statistical Analysis System (SAS) software.

Table 1. The treatment structure and dosages (g a.i/ha).

Treatment	2-leaf stage				4-leaf stage			
	*Ech.**	*Dig.*	*Ama.*	*Zea*	*Ech.*	*Dig.*	*Ama.*	*Zea*
1	0	0	0	0	0	0	0	0
2	175	175	25	175	280	280	40	280
3	225	225	75	225	360	360	120	360
4	325	325	125	325	520	520	200	520
5	425	425	175	425	650	650	280	650
6	600	600	225	600	900	900	360	900
7	750	750	325	750	1100	1100	520	1100

** Echi. – Echinochloa crus-galli, Dig. – Digitaria sanguinalis, Ama. – Amaranthus retroflexus, Zea. – Zea mays*

Results and Discussion

PPM-time curve analysis

<u>*PPM-time curve of weeds*</u>: The PPM value for weeds decreased sharply with the increase in the dosage of herbicides (Figures 2, 3, 4 and 5). Even though the visual injury symptoms could not be observed at 1 DAT. At the 4 leaf stage, the plants were stronger and less susceptible to the herbicides than at the 2 leaf stage plant. As a result of bentazone treatment, the visual injury was observed in red root (*Amaranthus retroflexus*) at 2 DAT, in barnyardgrass (*Echinochloa crus-galli*) at 4 DAT and in crabgrass (*Digitaria sanguinalis*) at 6 DAT. Withered leaves and necrotic spots were found on the top of red root leaves, while brown, discolored and withered leaves were observed in grass weeds.

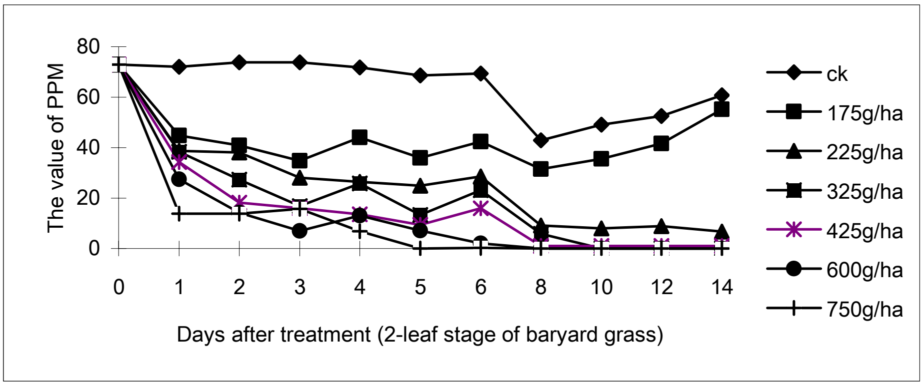

Figure 2. The PPM-time curve of 2-leaf stage of barnyard grass

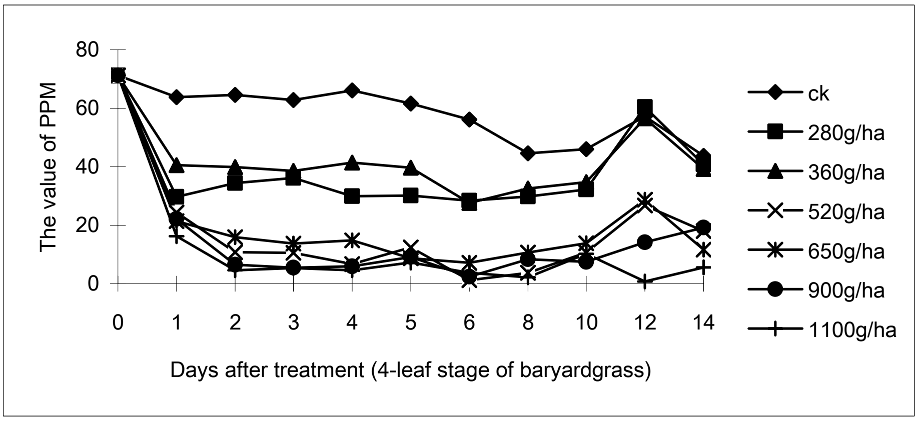

Figure 3. The PPM-time curve of 4-leaf stage of baryardgrass

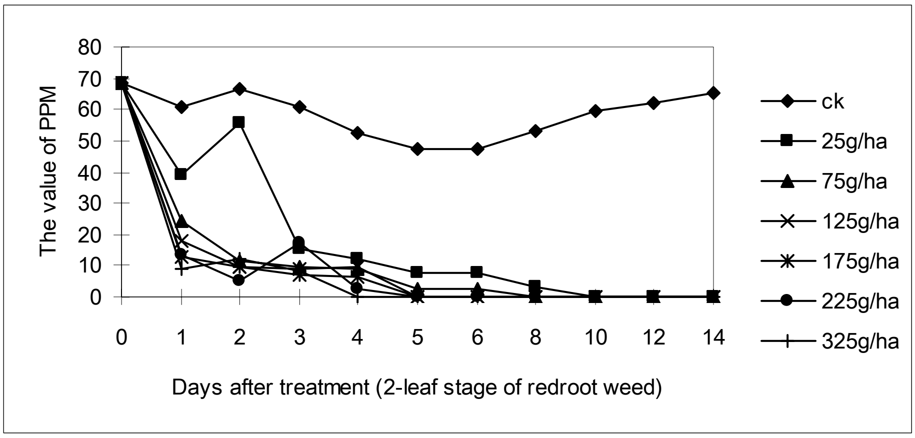

Figure 4. The PPM-time curve of 2-leaf stage of redroot weed

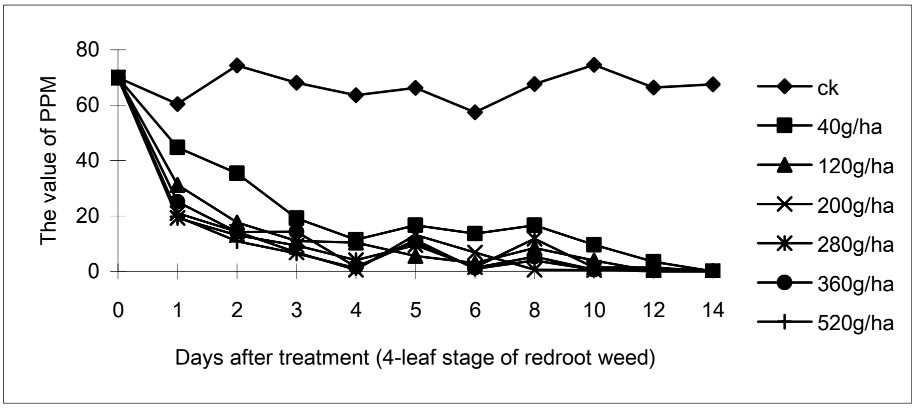

Figure 5. The PPM-time curve of 4-leaf stage of redroot weed

According to analysis of the PPM values, barnyardgrass and red root weed were highly susceptible to the herbicide, where the PPM value of some treatments was below 20, at 3 or 4 DAT. These weeds treated with the higher dosages died at 6 or 8 DAT.

The crab grass was not susceptible to the herbicide, where the PPM value was over 20 at 3 or 4 DAT. In this species, the PPM value increased gradually (Figures 6 and 7). The stem and leaf of the weed were, and hence could affect the spreading and absorption of the herbicide. Some special adjuvant could solve this problem.

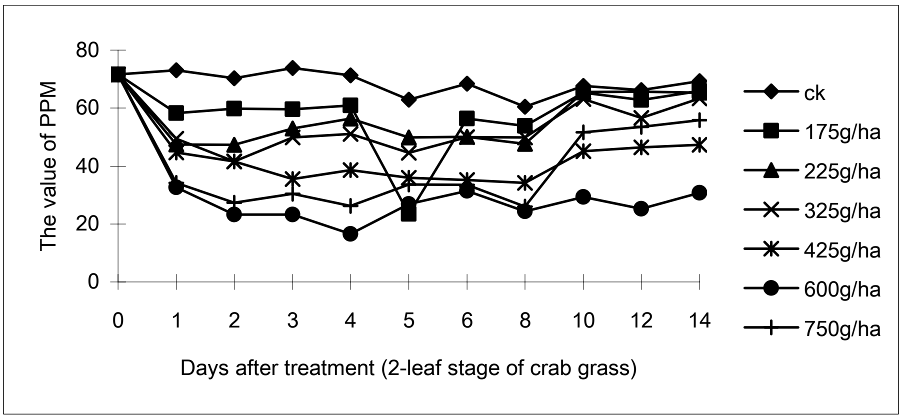

Figure 6. The PPM-time curve of 2-leaf stage of crabgrass

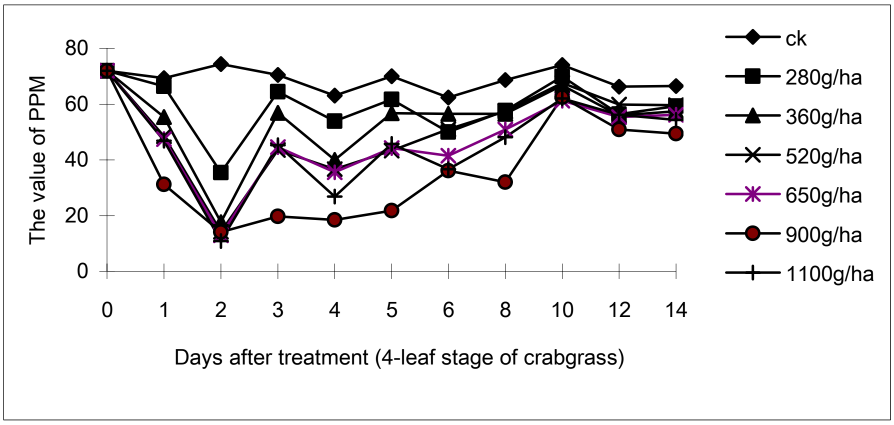

Figure 7. The PPM-time curve of 4-leaf stage of crab grass

<u>PPM-time curve of the maize</u>: The herbicides were safe to the maize crop, where visual injuries could not be observed. However, the PPM values decreased sharply at 1 DAT. Even though the herbicides could inhibit the photosynthetic yield, the PPM values were above 60 and the plants recovered at 12 DAT. There was no significant influence among the different treatments. The 4-leaf stage of maize was stronger and tolerant to the herbicides than the 2-leaf stage (Figures 8 and 9).

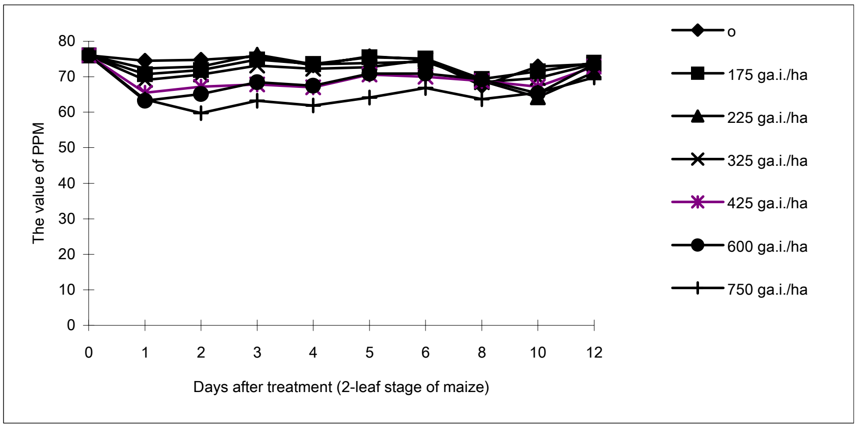

Figure 8. The PPM-time curve of 2-leaf stage of maize

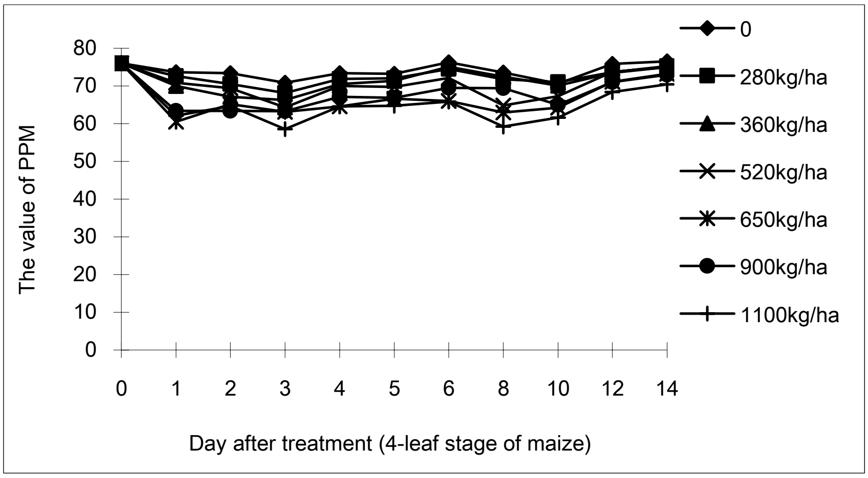

Figure 9. The PPM-time curve of 4-leaf stage of maize

The fresh weight reduction and sensitivity of the weed to the herbicide

Weed growth was inhibited after herbicide treatments. However, the susceptibility of weeds to the mixture of bentazone and atrazine was different among weed species. *Amaranthus retroflexus* was highly susceptible to the herbicide, and *E. crus-galli* was relatively less susceptible, while *D. sanguinalis* was not susceptible. The results of this experiment were not similar to a former study on weed sensitivity to the herbicide and MLHDs were calculated based on regression analysis between herbicide dosages and weed biomass (Table 2). Bentazone could control the broadleaf weed and had no effect on the two grass species, but atrazine could control the latter. However, atrazine with low foliar activity did not give a good efficacy without adequate amounts of the adjuvant. These were probably the reasons for the differences in results observed elsewhere

Table 2. Sensitivity of weed species to herbicides and MLHD.

Species of weed	MLHD study from PRI			Green house		
	Sensitivity	MLHD (g a.i./ha) bentazone	MLHD (g a.i./ha) atrazine	Sensitivity	ED_{90} (g a.i./ha) bentazone	ED_{90} (g a.i./ha) atrazine
2-leaf stage *Ama.*	+++	120	120	+++	9.36	9.36
4-leaf stage *Ama.*	+++	240	240	+++	23.28	23.28
2-leaf stage *Ech.*	++	200	200	++	No effect	208.69
4-leaf stage *Ech.*	++	400	400+oil	-	No effect	1013.02
2-leaf stage *Dig.*	+	400	400+oil	+	No effect	789.09
4-leaf stage *Dig.*	+	400	400+oil	-	No effect	>1100

Note: '+++' highly susceptible; '++' susceptible; '+' moderately sensitive; '-' not sensitive. * *Echi. – Echinochloa crus-galli, Dig. – Digitaria sanguinalis, Ama. – Amaranthus retroflexus*

The phytotoxicity of the herbicides to maize

The tank mixture of herbicides was safe to maize, and there were no significant differences among treatments (Figures 10 and 11).

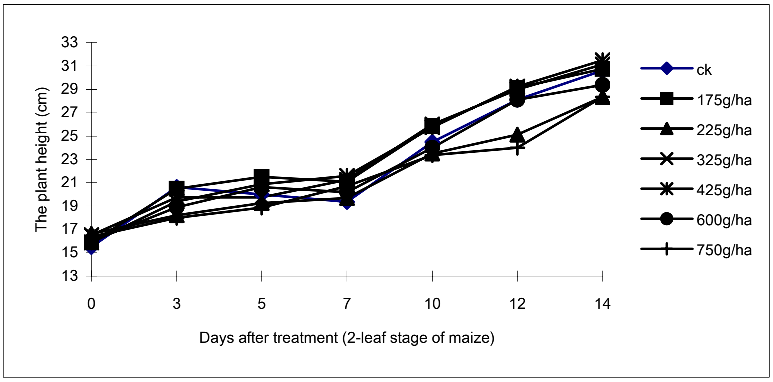

Figure 10. The plant height-time curve of 2-leaf stage of maize

The prediction of PPM value

Both of PPM value and weed fresh biomass decreased as dosages of the herbicide tank mixture increased. Table 3 shows how well the fresh weight reduction rate after treatment could be explained by PPM measurements at 2, 3, 4 or 5 DAT when a linear regression analyses could be done. Significant correlations ($p<0.05$) were observed between PPM measurements and the fresh weight reduction rate. The correlation coefficient for crabgrass was not satisfactory due to that the tank mixture of herbicides could not control it effectively. The hairy nature of the stem and leaves of this grass weed could not have resulted in less spread and absorption, without the presence of a good adjuvant. The PPM values of weed leaves at 2-5 DAT could give a good prediction for the biomass reduction rate.

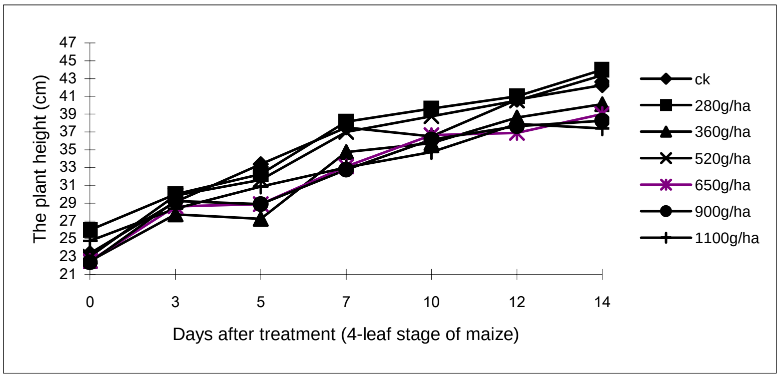

Figure 11. The plant height-time curve of 4-leaf stage of maize

Table 3. Correlation coefficient (R2 values) of regression of mean PPM values on 2, 3, 4 or 5 days after treatment and fresh weight reduction (%) 14 days after treatment.

Weed species	leaf stage	PPM measurements date (days after treatment)			
		2	3	4	5
Echinochloa crus-galli	2	0.87	0.97	0.86	0.89
	4	0.95	0.94	0.95	0.94
Digitaria sanguinalis	2	-	-	-	-
	4	0.57	0.51	0.66	0.56
Amaranthus retroflexus.	2	0.54	0.97	0.92	0.97
	4	0.88	0.97	0.97	0.97

Conclusions

The MLHD could be calculated by weed biomass 14 DAT of the tank mixture mixture of bentazone and atrazine. However, the susceptibility of weed species to the mixture was different and different dosages should be applied based on the dominant weed species in the field. There was a close relationship between the rate of reduction of weed biomass and the PPM values of weed leaves at 2-5 DAT. This indicates that the PPM value measured in early days after treatment could give a reasonable prediction on weed control efficacy of the herbicides. The tank mixture of bentazone and atrazine generally provided a 85-90% control of weed biomass when PPM value was about 20, and a relatively poor efficacy was obtained when the PPM value was below 40. The MLHD of bentazone and atrazine should be the dosage when PPM value is below 20 measured at 2-5 DAT.

The results of the present study indicated that determination of the MLHD for other herbicides would be useful exercise. If the weed could be controlled by MLHD technology, the damage to crops and contamination to environment could be ameliorated.

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SOME CHARACTERISTICS OF *Bidens pilosa* L. *var. radiata* Scheff., A NEW INVASIVE SPECIES IN THAILAND

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Abstract: Romerillo (*Bidens pilosa* L. var. *radiata* Schultz-Bip.), a native of Tropical America, was introduced to Thailand for apiculture around 1998-99 from Taiwan. The seeds were sown along road side in the Chiangmai province of northern Thailand. This species was detected 2 years after introduction. Seeds were collected and studied for weed potential such as seed germination, growth and seed production were studied in Bangkok. Other characteristics such as competitiveness including damage to crops were observed in the area of invasion in northern Thailand. It was found that the plant can survive in various habitats; on extreme dry land such as along cracks of concrete river banks, on flooded land and even in ditches. Each plant can produce about 41,000 seeds a year. The seeds harvested in Bangkok has about 72% germination rate, while the ones collected from the invasive area has 90-100% germination rate. The plant can complete its life cycle within 60 days. Propagation of this plant is both through reproductive and vegetative parts. The seed has small hooks and spines which assist in attaching to animal hairs, cloth or other materials aiding in dispersal far away. The plant has attractive flower heads with big ray flowers. Presently, this plant can be found in many locations such as in the north, northeast, south and central Thailand.

Key words: *Bidens pilosa*, invasive plants, biology, Thailand

Introduction

Invasive weeds that cause serious economic damage to farmers and the country in general are introduced weeds. These include water hyacinth, giant sensitive plant, pennisetum and Shama millet. Their introductions could be intentional introduction for specific purposes such as for production of fodder, for soil improvement or for ornamental purposes. They could be legal or illegal introductions, while some introductions were ignorant introductions or unintentional introductions such as seed contamination of agricultural imports.

Newly introduced species will adapt to the new environment. Establishment follows and if well established can become aggressive and turn into noxious weeds. Some species grow well together with native species, eventually become naturalized and no longer a problematic weed. Good examples of this instance are the water hyacinth (*Eichhornia crassipes* (Mart.) Solms) and Mexican fireplant (*Euphorbia heterophylla* (L.) Klotzsch & Garcke). The water hyacinth was introduced in 1901 for ornamental purpose and in 1913 the Water Hyacinth Control Act was promulgated. This law was ineffective as the weed continued to clog rivers, canals and waterways, impeding water traffic. However, a century after introduction, the water hyacinth is accepted as an naturalize plant.

Romerillo (*Bidens pilosa* L. var. *radiata* Schultz-Bip.), a native of Tropical America, is a member of Asteraceae or Compositae Family with about 200 species in the Genus *Bidens* scattered in tropical and sub-tropical regions of the world. Only 4-5 species are reported in South-east Asia, *Bidens pilosa* being the most common but due to wide variations, the taxonomy of this species is not yet satisfied (Alonzo and Hidebrand, 1999). In Thailand, *B. bipinnata* L., *B. pilosa* L. and *B. biternata* (Lour.) Merr. & Sherff were reported (Forestry Botany Section, 2001) which only *B. pilosa* L. *var pilosa*, *B. pilosa* L. *var minor* and *B. biternata* (Lour.) Merr. & Sherff were used as medicinal herb (Wit, 1996)

Seeds of Romerillo were brought to the Kingdom of Thailand around 1998 -1999 by a Taiwanese beekeeper as a food source as it flowers all year round, is a fast grower and needs very little attention. Seeds have been scattered in many nearby areas and the species eventually became known as Chiang Rai daisy, for its daisy-like flower. Some call it Taiwanese cosmos, after the introducer. The plant is similar to *B. pilosa* L. *var.* minor, a common weed in Thailand but the flower is more attractive than that of *B. pilosa* L. *var.* minor. So the purpose of this study was to compare the different of this plant from other varieties of the same species and the invasiveness character of this plant in Thailand.

Materials and Methods

Distribution

Since the plant was introduced without any record and it was first detected in year 2001, 2-3 years after introduction. So to clarify out the distribution and the habitat suitable of Romerillo, survey along roadside was done.

Seed germination

Matured seeds were collected from the northern region and from a net house in Bangkok in 2001. Seeds from each location were spread in a ∅90 mm Petri dish containing 1 piece of Whatman filter paper (No.4), at a rate of 50 seeds for each dish, with a total of 6 dishes per location. Five ml of distilled water was added to each Petri dish, and the Petri dishes were kept at room temperature to simulate the natural conditions in Bangkok. The numbers of germinated seed were recorded on the 7^{th} day from the commencement of the experiment.

<u>*Germination of seeds at different ages after development*</u>: Seeds of Romerillo were collected from flower head at (a) full bloom, petals of ray flower wilting, (b) all petals of ray flower dropped and partial wilting of disc flower, (c) all petals of disc flower wilted but seeds still green, (d) seeds darken and drying up, and (e) fully mature seeds. All the seeds were tested for germination at 1, 2, 3 and 4 day after removal. The seeds of each stage were spread in ∅ 90 mm Petri dish with 1 filter paper at a rate of 10 seeds per Petri dish, with 3 replicates. The number of germinated seed was record 7 days later.

Growth and other character

To study the growth habit and seed production of Romerillo, an experiment was set up in Bangkok, where 100 seeds of Romerillo were broadcasted in 1 x 1 m^2 plot, with a total of 12 plots. The number of seedlings in each plot was recorded until 1 month, and then the seedlings uprooted in randomly selected plots. The plant height, branches, flowering number and number of matured seeds were recorded once in two days for 4 months. The growth characteristics of Romerillo were compared with those of *B.s pilosa* L. *var. minor* and *B. biternata*, which already exist in Thailand.

Allelopathic potential of Romerillo

Leaves of Romerillo, *B. pilosa* L. *var*. *minor* and *B. pilosa* L. *var*. *pilosa* were collected from the experimental plots and about 10 g of each were dried in oven at 50°C for 24 hrs. Then the dried leaves at 0 (control), 0.01, 0.05 and 0.1 g were laid between 2 layers of 10-10 ml of 0.5% agar at ϕ29 mm x 130 mm height. Six seeds of *Mimosa pigra* L. were placed on the top of the agar, sealed with transparent film. Another set was done following the same procedure, but using fresh leaves at the same weight. All the tubes were placed in a growth chamber at 30°C with 24 hrs light. Root length and plant height were recorded at 7 days later.

Results and Discussion

Distribution

Romerillo has its origins in tropical South America. There are reports that it is a serious weed in American Samoa (IPM Plans of Work – Alabama to Hawaii), very wide spread weed in Okinawa of Japan and similarly in Taiwan. In the USA, it is found in Alabama, Connecticut, Florida, Hawaii, Louisiana, Missouri and Pennsylvania.

Surveyed areas in the northern Thailand include Chiang Mai, Chiangrai, Payao, Phrae, Nan, Lampang and Lampoon where the weed grows in all sorts of conditions. Along the road edge, along the rim of the canal, mid-road islands and even in cracks of hot and dry concrete banks along the Mekong River. It has spread into vegetable growing areas, citrus groves, mango and longan orchards as well as maize land. Farmers in the area indicated that they have applied herbicides in the maize plots to control Romerillo, however, the type of herbicide used could not be determined.

Seed germination

Germination of matured seeds collected from the northern region of Thailand showed high percentage of germination (94%). The seeds harvested from the net house in Bangkok gave a lower percentage of germination (72.3%). Various stages of seeds, which developed from flower, are shown in Figure 1 and their germination at each growth stage is shown in Figure 2. About 53-100% matured seeds germinated without dormancy. The seeds at stage 3-4 germinated at a rate of more than 50 % even when they are not completely matured at harvesting. The seeds, which were not well developed at the stage 1, did not geminate at any time of testing. However, the young seeds that were collected when the ray flowers were dropped (stage 2), germinated after keeping for 3 days, but the germinations rate was less than 50%. This implied that even the seeds are not completely mature at the time of cutting may develop further if left for few days, and geminate later.

Figure 1. Stages of seeds, which use for the germination test (1 = Bloom, petals of ray flower wilting, 2 = All petals of ray flower dropped and partial wilting of disc flower, 3 = All petals of disc flower wilted but seeds still green, 4 = Seeds darken and drying up and 5 = Fully mature seeds)

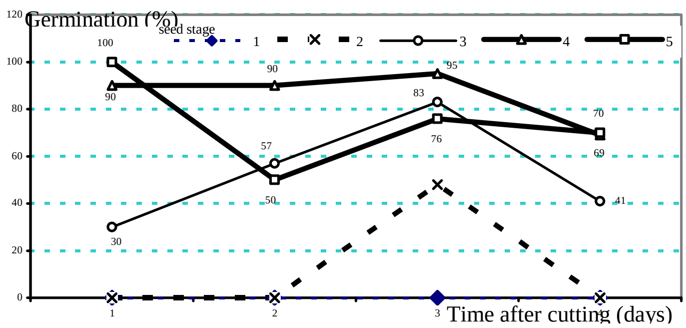

Figure 2. Germination of the seeds of various stages

Growth and other characters

Seed germination in soil: About 65% of the seeds germinated within the first 10 days of the experiment, but the rate of germination increased gradually thereafter (Figure 3).

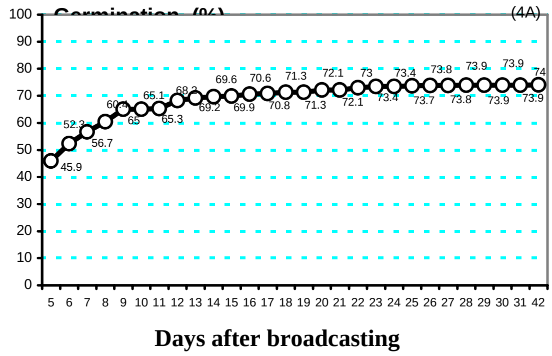

Figure 3. Germination of seeds in 1 x 1 m^2 plots

Growth and development: The plant height during the first 25 days averaged only 10 cm. At 36 days, plots of all planting densities produced plants of similar height. A difference in plant height was observed at 52 days, where the plots with 1 plant/m^2 produced plants at a height of 48 cm while those in plots with a density of 58 plants/m^2 resulted in an average height of 26 cm. No further gain in plant height was observed after 75 days (Figure 4). The plots with a single plant/ m^2 recorded a height of 120 cm while those with 58 plants/m^2, recorded a height of 95 cm. This is as expected due to severe competition for resources when the population density increases.

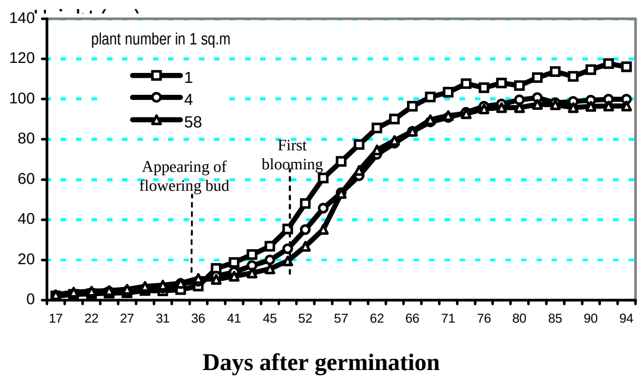

Figure 4. Height of plants at varying density per m^2

The number of branches per plant will influence the number of flowers as they are formed apically. Therefore, more branches would result in more seeds being produced. At 120 days, the plants still continued to branch, especially in the plots with a single plant/ m^2, resulting in 246 branches. The plots with a density of 58 plants/m^2 produced only 57 branches per plant (Figure 5)

Number of leaves per plant showed a similar trend to that of plant height and number of branches. At 61 days after seeding, the plots with a single plant/ m^2 had an average of 403 leaves while in the plot with 58 plants/ m^2, it was 132 leaves per plant (Figure 6). It is worthy to note that after 35 days, apical floral buds were formed, followed by new branching of the central stem and then by the side stems.

Seed production of this species is phenomenal, meeting the criteria that major weeds of the world are prolific seed producers. The number of flowers per plant was high, maturing with white petals of ray flowers that are clearly visible at 59 days after germination (Figure

7). This is considered a complete flower and the plots with single plant/m^2 produced about 89 blooms per day (Figure 6) while those with 58 plants/m^2 produced only 1 new bloom per day. Flower production increased gradually as the plant become taller.

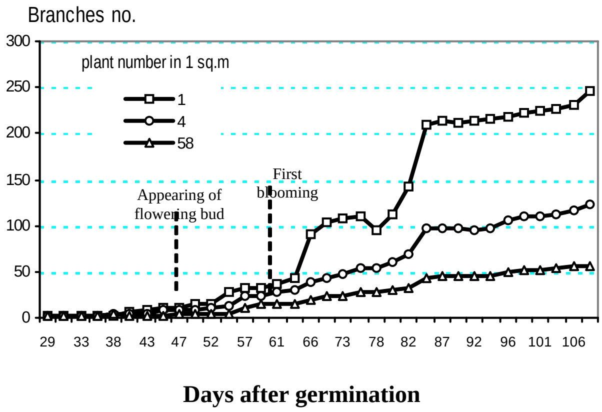

Figure 5. Number of branches per plant

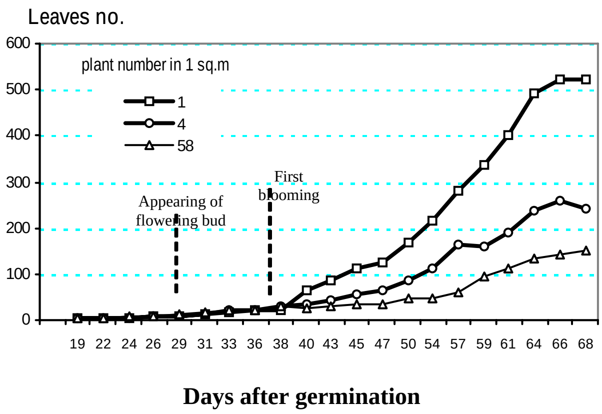

Figure 6. Number of leaves per plant

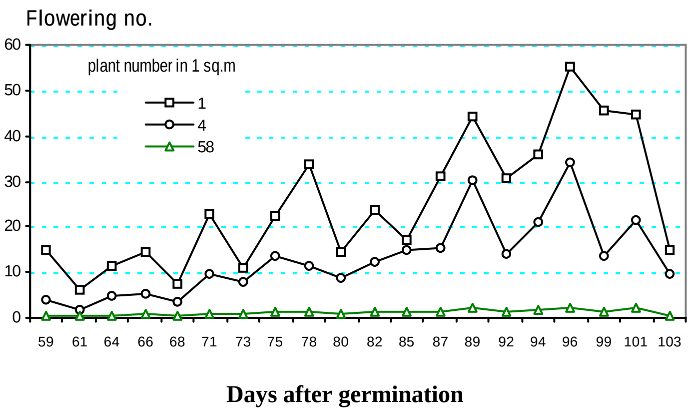

Figure 7. Number of flowers in each plot size of varying density

The number of seeds per plant was highly variable, depending upon planting density/m^2. The cumulative number of seeds formed on the 96th day after germination was 13,929 per plant for the treatment with a single plant/m^2 while the lowest was in the plots with 58 plants/ m^2 (761 seeds per plant). The estimated seed production for the plant densities of 1, 4 and 58 plants/ m^2 is 13,929, 33619 and 44,142, respectively (Figure 8).

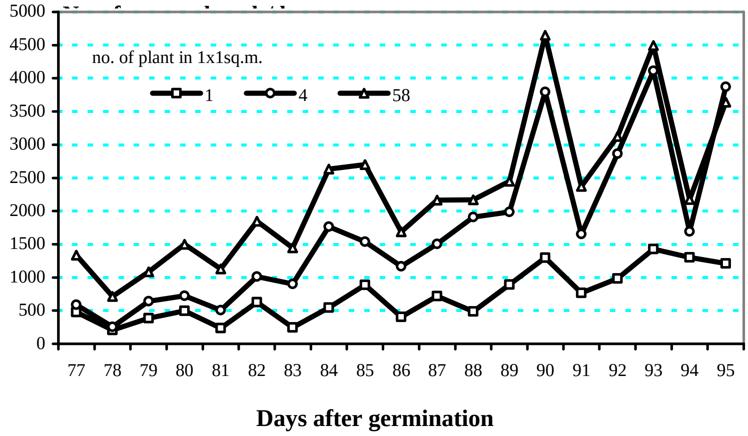

Figure 8. Number of Romerillo seeds produced m² under varying planting densities

Allelopathic potential of Romerillo

In comparison with *B. pilosa* L. *var. pilosa* and *B. pilosa* L. *var. minor*, leaves of all three species at 0.1 and 0.5 g were able to inhibit growth of young *Mimosa pigra*, with Romerillo demonstrating the highest ipact (Figure 9). Hence, in its natural habitat, it is usually seen growing in pure stands.

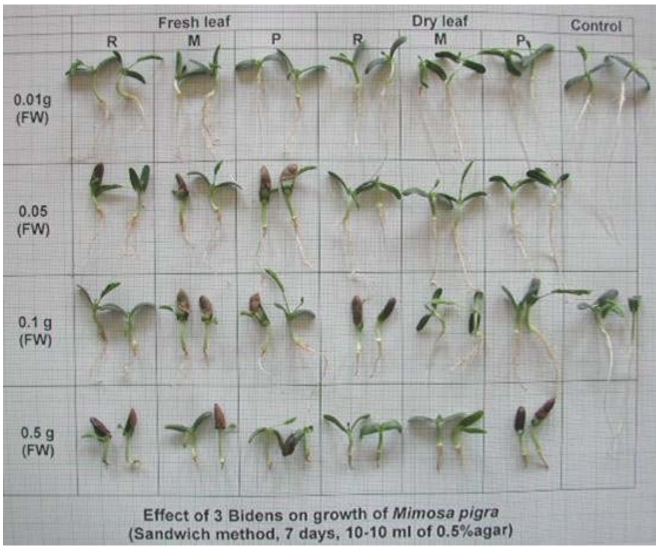

Figure 9. Growth of *Mimosa pigra* in fresh and dried leaves of 3 varieties of *Bidens pilosa* L.; var *radiata* (R), var. *minor* (M), and var. *pilosa* (P)

In Thailand the *B. pilosa var. minor* and *var. pilosa* are common weeds, especially in the northern region. The *var.* minor has ray flowers with petals while the var. *pilosa* does not. Moreover, the field surveys also indicated that cuttings of mature stems sealed in polyethylene bags for 7 days with adequate moisture were still with green leaves, and many adventitious roots forming from the base up to top 4-5 leaves. When these cuttings were planted in the soil, they established well and continued to grow and compete against other species. The large white-ray flowers are attractive when compared to other plants. As they grow well without much attention, people harvest seeds and plant them on vacant land, helping the spread of the species. The seeds have 2 – 6 hooks at one end, which help the get attached to clothing, animal hairs, etc. supporting their dispersal. This study revealed that the seeds of Romerillo have no dormancy, with 80 – 90% germination rate in the laboratory and

72% in the field. Among the newly emerged plants, 81% produced seeds after 35 – 40 days and completed the full cycle within 57 – 70 days. In Thailand, the climatic conditions enable Romerillo to go through 5 – 6 cycles per year, with the original plant still reproducing.

Estimation done based on these figures resulted in a minimum of 31 million plants after 2 years of growth from one single plant (Table 1), which is worse than *B. pilosa* Linn. that has already established in Thailand. A single plant of *B. pilosa* Linn. can produce about 3,000 - 6,000 mature seeds, which are ready to germinate. *Bidens pilosa* Linn completes about 3 – 4 cycles in a given year with 95% germination rate, and even after 3 – 5 years of storage, the seeds still have about 80% germination rate. The Romerillo's capabilities surpass this species. In its natural habitat in the north, Romerillo does not grow in mixed stand, most likely due to its stronger alleopathic effects than *B. pilosa*.. The summarized properties of the species when compared to the characteristics of an ideal weed Muenscher (1981) are shown in Table 2.

Table 1. Number of seeds produced

Year	No. of Seeds Available	72% Germination	80% Survived	No. of seeds produced
1			1	41,787
2	41,787	30,087	24,069	54,950,239
3	54,950,239	39,564,172	31,651,338	72,260,004,276
4	72,260,004,276	52,027,203,079	41,621,762,463	95,022,483,703,177

Table 2. Important properties of weeds as summarized by Muenscher (1981)

Some Important Properties of Weeds	Characteristics of Romerillo
Growth	
Grow well under unsuitable conditions and suitable conditions similar to that required by the crop.	Grow well in moist and drought conditions, even in concrete cracks and in the shade.
Contain undesirable odor or unpalatable taste, covered with hairs, thorns or slime, enabling it to survive attacks by insects and animals.	Contain allelochemicals inhibiting growth of other plants. Leaves release odor when crushed. Leaves not damaged by insects and no known natural enemies in Thailand.
Seed Production	
Able to produce many seeds per year.	1, 4 and 58 plants/m^2 produce 41,787, 25,215 and 2,283 seeds/year, respectively
Seed viability is long, able to germinate even if in the soil for many years	Seeds known to survive up to 6 years
Seeds able to develop into maturity, even if removed from plant early, some species seeds mature at flowering stage	Seeds mature 10-15 days after bloom and can germinate at only 6-7 days later. And it can germinate even not mature.
Seeds usually difficult to separate from seeds of crop as usually of similar size and shape	
Fruit or seeds usually have dispersal aids	2 – 6 hooks found at one end of seeds and along the seed length there are small thorns, which aid in seeds attaching to animal hairs or human clothing
Propagation	
Able to propagate with stem and roots	Cut stem can form roots and grow given adequate moisture

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INTEGRATED WEED MANAGEMENT TO CONTROL *Pteridium* spp AND *Polygonum chinens* IN YOUNG TEA PLANTATIONS OF INDIA.

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Abstract: Ferns (*Pteridium* spp) and *Polygonum chinens* are considered to be among the few very hard-to-kill weed species in Tea Plantations in north east India. Chemical control alone could not effectively manage these obnoxious weed species. Hence, manual weed control (hand weeding and hoeing) during the cold weather when free mandays are available more in number, followed by a scheduling of chemical herbicides have proven to be the best way of managing these weed species. Ammonium salt of glyphosate 71% (SG), a broad spectrum post-emergent water soluble herbicide, was found to be the most effective against different other herbicides and herbicide mixtures such as glyphosate 41% IPA salt, paraquat, glufosinate ammonium and glyphosate 41%+, and oxyfluorfen. The most effective results were achieved by using the ammonium salt of glyphosate 71% (SG) at 1.5 kg per ha followed by a cocktail of glyphosate 41% at 2 l per ha + oxyfluorfen at 0.5 l per ha.

Key words: Ammonium salt of glyphosate 71% (SG), *Pteridium* spp., *Polygonum chinens*

Introduction

Weeds are a serious problem in Tea just as much as in other crops. India rank first in Tea production in the world. *Pteridium* spp. and *Polygonum chinens* are very hard to kill weed species in major Tea growing areas in India. It can result in crop loss to the extent of 50% (Rao, 1978). It also hampers the frame formation in young teas and thereby affects their long term productivity. These weeds are predominant in the areas where drainage is inadequate and water table is high. Polygonum is a perennial scrambler that covers the ground very fast and on good support it climbs up to a considerable height. Different herbicides were tried out to control polygonum but it always proved to be very difficult to control by one round or rounds of a single herbicide. The present study was focused at a round the year scheduling of herbicides along with a combination of manual weed control during cold weather so that these weeds do not create a recurrent problem in the following years .

Materials and Methods

The experiment was conducted at Chalouni Tea Garden of Goodricke Group Ltd during April to October, 2005. The garden is located in Dooars, West Bengal, India. The trial was laid out in a Randomized Complete Block Design (RCBD) with 3 replications in a young tea (5 years old) section. The section was under Light pruning done in the month of January. The treatments used were, T1 - glyphostae 41 SL (Glycel®) at 3 l/ha, T2 - glyphosate 41 SL (Glycel®) at 2 l/ha + 2,4 D amine salt at 0.6 l/ha, T3 - ammonium salt of glyphosate 71% SG (Speed®) at 2.5 kg/ha, T4 - paraquat at 1.5 l/ha + 2,4 D amine salt at 0.6 l/ha, T5 - ammonium salt of glyphosate 71% SG (Speed®) at 3 kg/ha, T6 - 2,4 D amine salt at 1 l/ha and T7 - control. The entire ground was cheeled (scraped with mechanical process) and leveled after pruning in February and two rounds of irrigation (5 cm of water) were given at 21-day interval in February and March. The area received around 310 cm rainfall between April to October, 2005.

All herbicides were sprayed with ASPEE BAKPAK sprayer with ULV 100 nozzle. The first round of herbicide was applied to all treatments by last week of April, 2005 when about 90% ground was covered by weeds. The weed count showed dominance of *Polygonum sp*, Ferns (*Pteridium* spp), *Cynodon dactylon, Borreria hispida.* The second round of herbicide application was given in July 1st week. The bio-efficacy was assessed against the control plot and also as compared with other treatments. The yield/ha was also recorded at the end of the Trial from all plots.

Results and Discussion

The degree of weed control in treated plots is shown in Tables 1 and 2. The second round of spray was given after 60days of first application. It was noticed that T5 had excellent control even after 60 days. The treatments, T2 and T3, also showed fairly good control up to 45 days after spraying.

Table 1. Effect of different herbicides and their combinations (mean of 3 replicates) after the first round of spray.

Treatment	Days after spraying					
	15	30	45	60	75	Mean
T1	83	87	75	48	0	73.25
T2	95	85	72	60	0	78.00
T3	98	95	90	75	0	89.50
T4	80	55	48	34	0	54.25
T5	96	96	95	86	0	93.25
T6	75	68	53	32	0	57.00
T7	-	-	-	-	-	

Table 2. Effect of different herbicides and their combinations (mean of 3 replicates) after the second round of spray.

Treatment	Days after spraying					
	15	30	45	60	75	Mean
T1	85	85	79	51	35	67.00
T2	96	95	78	63	58	78.00
T3	98	98	90	76	68	86.00
T4	82	60	53	40	18	50.60
T5	97	99	95	88	72	90.20
T6	75	66	55	37	20	50.60
T7	-	-	-	-	-	

The overall best result was achieved by T5 where the ammonium salt of glyphosate 71% SG (at 3 kg/ha) was used followed by T3 (ammonium salt of glyphosate 71% SG at 2.5 kg/ha) and T2 (isopropyl amine salt of glyphosate 41% SL + 2,4 D amine salt) was used. It was evident that all these 3 treatments performed better than other treatments on a mixed weed flora dominated by broad leaf weeds. T4 and T6 almost showed similar results after 60 days while T4 showed quicker burning effect during first 15 days. It was also noticed that the effect of weed control had a direct effect on the productivity in terms of yield per hectare narrated in table 3.

The trial clearly highlighted that Ammonium salt of glyphosate SG performed better than all other herbicides applied alone or in combination with other herbicides. It was observed that the overall population of *Polygonum* and *Pteridium* spp. were reduced after second round of application to the tune of 55 % and 52%, respectively. The yield /ha also showed the best positive response in T5 followed by T2 and T1.

The result of the present study will definitely help tea planters to plan their annual herbicide schedule to control obnoxious weeds like *Polygonum chinens* and *Pteridium* spp., which are definitely a menace in the Tea Growing areas in North Eastern part of India.

Table 3. Yield of Tea in different treatments of herbicide application

Treatment	Yield from Apr-Nov 2005 (kg/ha)*
T1	2304
T2	2322
T3	2700
T4	2028
T5	2915
T6	2010
T7	1914

*Means of three replicates

Literature cited

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AN AGGRESSIVE INVADER PLANT *Andropogon virginicus* L. IN JAPAN AND HAWAII ISLANDS

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Abstract: *Andropogon virginicus* is a native from the southeastern United States to northern south America, and it was introduced to Hawaii prior to 1924. It is considered as one of the three most invasive grasses in Hawaii pasture systems. According to the Australian risk assessment system, it scored 13, a score for a plant not allowed to be imported, and likely to be a pest. The specimens of *A. virginicus* from major herbariums in Japan were checked in the laboratory. The oldest specimen was collected in 1940, in Kyoto Prefecture, and they have spread into central part of Japan. The distribution status of *A. virginicus* was surveyed in 2006, and was found to have widely distributed from Miyagi prefecture to Kagoshima prefecture (about 1300 km). Most of the invasive plant species in Japan grow where the original vegetation is disturbed by human activities. *Andropogon virginicus* grows at open sites such as reclaimed land and housing sites. This is a unique feature of the species that grows on non-disturbed places like the Rocky Mountains. It grows along the roadsides, railroad tracks, on the lawns at the golf courses, and gardens. The plant was also found to grow on the granite terrains but very few survives on the lime stone areas. The extension of a root under the different aluminum concentration of the soil was examined and found that it can survive under high aluminum concentrations in soil. The seed has dormancy and germinate at the temperature of 20 to 40°C. *A. virginicus* seeds germinated under natural conditions in April, bloomed and bear fruits in October of the same year.

Key words: *Andropogon virginicus*, distribution, invasive species

Introduction

The present study was carried out to identify the distribution of *Andropogon virginicus* L., an alien plant found in Japan, to locate their growing sites, to investigate the environmental factors affecting their germination and growth and relate it to their botanical information, and to disseminate information on the distribution of the species in Japan on the homepage.

Materials and Methods

The specimens of the *Andropogon virginicus* L available at the 16 major herbariums in Japan were investigated to identify the locations where the plant has invaded. The homepage of the Research Institute of Bioresouces, Okayama University, Japan, was used to explain the species distribution where updated information on *A. virginicus* L. has been made available. The information on the distribution of this species was collected with the help of 1200 associates of Kurashiki museum of Natural History. Field surveys were carried out to record the growing locations by using Geographic Information System (GPS), and gather the information of the surrounding environment. The survey was carried out in two ways. Firstly, the investigators visited many places in Japan where *A. virginicus* L. was found, and recorded the latitude and longitude of their growing locations were recorded by using a GPS. Most of the time, height above the sea level was recorded, too, however, this parameter was estimated from the locations. Concurrently, the authors requested assistance through the internet, gathered information through e-mail regarding the growing locations of the plant species, including the name of the site. New sites where plant has invaded over the past year were visited. In areas where new growth was recorded, the relationship with the environment and the coexistence plant species were investigated.

Results and Discussion

Investigation of the herbarium specimens

The authors have investigated 434 specimens from the 16 herbariums around Japan, and the oldest specimen has been recorded in Kyoto in 1940, from the Department of Botany of the Faculty of Science, Kyoto University (KYO). In Okayama, the first record of the specimen was dated back to 1955. According to Figures 1.1 to 1.7, the species has been reported from Keihanshin and Nagoya areas in1940s, and later it has spread to Toukai, Sanyo, Shikoku, Kyushu, Kanto, and to the areas along Japan Sea.

Literature survey

The authors have investigated 121 literatures, and found that northern limit of the spread of the plant from the side of Sea of Japan was Niigata and from the side of Pacific Ocean was Fukushima, and Kagoshima has been the southern limit of spread.

Field survey

Through the field visits, and internet and e-mail surveys, 11,067 growing locations of the species was recorded (Figure 1.8). The distribution point was identified to be further north when compared to the reports of the literature survey. The present study revealed that the northern limit of the distribution point is Miyagi, where as the southern limit is the Yakushima Island.

In Okayama Prefecture, investigations were conducted thoroughly, and 3,349 locations that the plant grows were identified. The species grew well in the sunny places, along the sides of the railroad, roadsides, newly developed residential sites, and on the lawns. The plant species also existed on places where it is impossible for the wind to carry seeds. The seeds of *A. virginicus* L. have been transported together with mountain soil that was moved by people to establish housing sites, which has facilitated its invasion. The seeds of the species have spread into lawns from nearby areas, again carried by human beings. The seeds are often mixed with the garden trees when they are transplanting.

Invasion of *A. virginicus* L. has been identified from new environments when compared to that recorded in the previous year. Currently, the soil condition in the newly invaded areas and the construction species are investigated. The species seems to be quite tolerant and adapts itself to the wide moisture ranges of the soil, from the dry granite area to the rice field conditions.

The authors also surveyed the spread of *A. virginicus* L. in Hawaii Island where they have first invaded in 1924. The species was recorded along the roadsides, and on the farmland. Invasion was seen also in the crater in Kilauea Volcano and lava fields (Figure 2). *A. virginicus* L. has invaded into sparse vegetation with many endemic species, and as a result, it is clear that the rare native species are in danger of extinction.

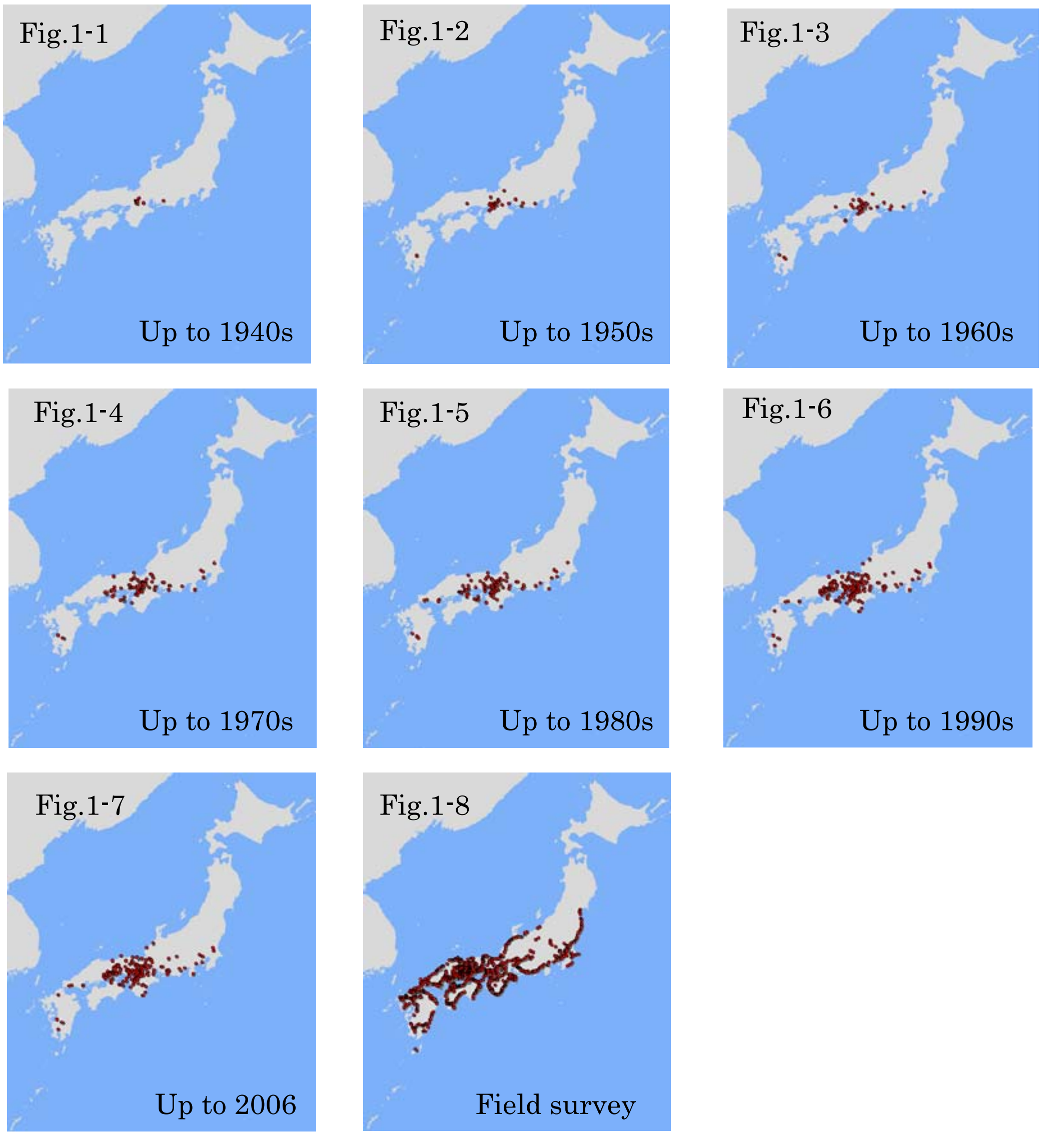

Figure 1. The history of distribution pattern of *A. virginicus* L. in Japan.

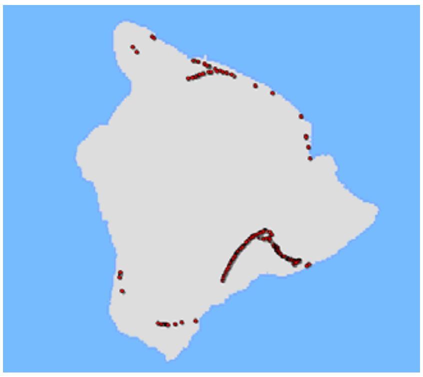

Figure 2. The distribution of *A. virginicus* L. in Hawaii Island

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COMPARATIVE ABSORPTION, TRANSLOCATION AND METABOLISM OF THE HERBICIDE FLUCETOSULFURON IN RICE AND BARNYARDGRASS

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Abstract: Flucetosulfuron is a sulfonylurea herbicide that controls barnyardgrass (*Echinochloa crus-galli*) in paddy rice. In the present study, we investigated the physiological basis of selectivity between barnyardgrass and rice (*Oryza sativa*) in terms of absorption, translocation, and metabolism of foliar-applied flucetosulfuron. The initial absorption of [^{14}C]flucetosulfuron at 6 hours after treatment (HAT) was >70% in barnyardgrass, while that in rice was only about 20% of the applied radioactivity. At the prolonged period, barnyardgrass absorbed 3.6, 2.8, 1.7, and 1.6-times more than in rice at 12, 24, 48, and 72 HAT, respectively. Due to greater absorption, translocation out of the treated leaf was about 3-fold greater in barnyardgrass that in rice. [^{14}C]Flucetosulfuron was metabolized very rapidly to metabolite-1 (herbicidally active) and metabolite-3 in both species, and the rate of metabolism was similar in both cases. Flucetosulfuron and metabolite-1 remained at <3% of the applied dosage at 144 HAT in both test plant species. These results indicate that the selectivity to flucetosulfuron between rice and barnyardgrass may be primarily due to the differential absorption rates between plant species.

Key words: Absorption, barnyardgrass, flucetosulfuron, metabolism, rice, selectivity, translocation

Introduction

Flucetosulfuron is a sulfonylurea herbicide for paddy rice developed by LG Life Sciences, Ltd (Koo *et al*., 2003). Unlike other sulfonylurea rice herbicides, the herbicide controls barnyardgrass very effectively as well as other annual and perennial weeds. The herbicide can be applied as both foliar or soil applications, as a pre- to post-emergent herbicide (Kim *et al.* 2003).

Kim *et al.* (2006) reported that when flucetosulfuron was used as a foliar applied herbicide, the selectivity ratio to rice, which is calculated by GR_{50} (rate that decreased plant fresh weight by 50%) was 49-fold when compared to that of barnyardgrass (GR_{50} for rice divided by that for barnyardgrass). In terms of biochemical mode of action, flucetosulfuron inhibited Acetolactate synthase (ALS), similar to other sulfonylurea herbicides (Hwang *et al.* 2003). The I_{50} (concentration that inhibited enzyme activity by 50%) in *in vitro* ALS assay was 83.8 and 93.8 μM for rice and barnyardgrass, respectively, showing a marginal difference. In *in vivo* ALS assay, the ALS activity was inhibited strongly within 24 hrs after application in both the plant species, however, ALS activity in rice began to recover after 24 hrs reaching about 70% of that of the untreated plants. This recovery occurred selectively in rice only (Hwang *et al.* 2003). These results suggested that the selectivity of rice to the herbicide could be due to faster detoxification.

In the present study, further investigations on the physiological basis of selectivity between rice and barnyardgrass was carried out in terms of absorption, translocation, and metabolism, using radiolabelled flucetosulfuron.

Materials and Methods

Radiolabelled herbicide

Radiolabelled [α-^{14}C-pyridine] flucetosulfuron (Figure 1) with a specific activity of 57.18 mCimmol^{-1} and purity of >98% was synthesized by Korea Radiochemical Research Center (Suwon, Korea).

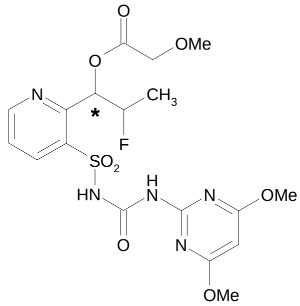

Figure 1. Structures of radiolabelled [α-^{14}C-pyridine]flucetosulfuron. *Site of the ^{14}C label.

Plant material

Seeds of rice (*Oryza sativa*, cv. Ilpum) and barnyardgrass (*Echinochloa crus-galli*) were germinated in flats in a commercial greenhouse soil mix (Boo-Nong Soil Mix, Korea) and watered with tap water. Plants were grown in a greenhouse maintained at 25 ± 3/15 ± 3°C day/night, with a 14-hrs photoperiod for 6 days. Both plant species were between the 3 to 3.5 leaf stage at the time of application. After application with the [^{14}C]flucetosulfuron, all the plants were returned to the glasshouse until harvest.

Foliar absorption and translocation studies

A 10 μl of 50% acetone solution (with 0.1% Tween 20®) containing 4.34 kBq of [^{14}C] flucetosulfuron was applied to the adaxial leaf surface of the middle of the third fully expanded leaf of both rice and barnyardgrass in 10 droplets. Plants were harvested at 1, 3, 6, 12, 24, 48, 72 hrs after application. Uptake and translocation of [^{14}C]flucetosulfuron were quantified by scintillation counting of plant parts. The level of radio activity in the plants was determined after combustion of four separate parts of the plant, *i.e.* the treated leaf, the first and second leaves, the fourth leaf, and root. At harvest, the treated 3rd leaf was washed three times sequentially for 20 sec. with 5 ml of acetonitrile:water (20:80, v:v). The washes were then combined. A 1 ml aliquot of each leaf wash was counted by a liquid scintillation counter (LS6000TA, Beckman, USA). Plant samples were combusted in a sample oxidizer (Model 307, Packard Instruments, USA). Permafluor® Scintillation Cocktail and Carbo-Sorb® Cocktail (Packard) were used to analyze these samples.

Metabolism study

Rice and barnyardgrass at the stage of 3 ~ 4 leaf stage were sprayed with [^{14}C]flucetosulfuron (0.52 MBq) dissolved in a 50:50 solution of acetone and distilled water containing 0.1% Tween 20®, and used at the rate of 40 g a.i. ha^{-1}, which is the field used rate of the herbicide. Herbicide application was made with a CO_2-pressurized belt-driven sprayer equipped with an 8002E flat fan nozzle adjusted to deliver 500 l ha^{-1}. Plants were harvested at the soil surface level at 0, 1, 3, 6 days after treatment. The treated foliage was washed with acetonitrile for 3 times to remove the unabsorbed herbicide, and 4 ml of each wash samples was counted by liquid scintillation counter.

Surface-washed plants were stored at -80℃ prior to analysis. For extraction, plants were chopped with dry ice and then the chopped plants were homogenized in 20ml of acetonitrile by a sample homogenizer (POLYTRON®, Switzerland) for 1 min. The homogenates were centrifuged at 10,000*g* for 30 min, and the supernatant was saved. The pellets were extracted two more times with 20 ml of acetonitrile, followed by 10 ml of 50% acetone solution (pH 4), and acetone:water:*c*-HCl solution (35:35:1, v:v). One ml sample of each combined supernatants was counted by liquid scintillation counter to determine the total radioactivity extracted from the leaf. Ten ml of the combined supernatants was concentrated to 1 ml under nitrogen gas stream, and the concentrated supernatant was filtered through 2-µm nylon disposable filters (Gelman Sciences) to separate and to quantify the parent herbicide and metabolites by HPLC equipped with Radiomatic 610TR radio active detector (3 ml liquid cell, PerkinElmer, USA). Ultraviolet chromatograms were obtained for non-radiolabelled flucetosulfuron and metabolite standards at 254 nm and co-chromatographed with radiolabelled flucetosulfuron and metabolites from plant extracts using the radiodetectors. Binary mobile phases were A = acetonitrile and B = water (0.1% trifluoracetic acid). Separation method (Table 1) was operated at a flow rate of 1 ml min^{-1} and a column temperature of 25°C using a CapcellPak-C_{18} reverse-phase column (4.6 mm i.d. × 250 mm).

Table 1. The separation method employed in the experiment.

Time (min)	% A*	% B
0	10	90
30	100	0
35	100	0
40	10	90

*A – Acetontrile, B – water (0.1% trifluroacetic acid)

Results and Discussion

Foliar absorption and translocation studies

Absorption of [^{14}C]flucetosulfuron increased with time in both rice and barnyardgrass. The initial absorption of [^{14}C]flucetosulfuron in barnyardgrass was relatively fast, where higher than 71% of the radioactivity was absorbed in 6 hours after treatment (HAT). The maximum absorption of >90% was achieved after 48 HAT in barnyardgrass. In contrast, absorption of [^{14}C]flucetosulfuron in rice was initially about 20% at 6 HAT, and the maximum was about 60% after 48 HAT (Figure 2). Consequently, translocation was greater in barnyardgrass, where the weed translocated about 9% of the total applied from the treated leaf to the higher leaf and roots, while rice translocated only about 3% (Figure 3).

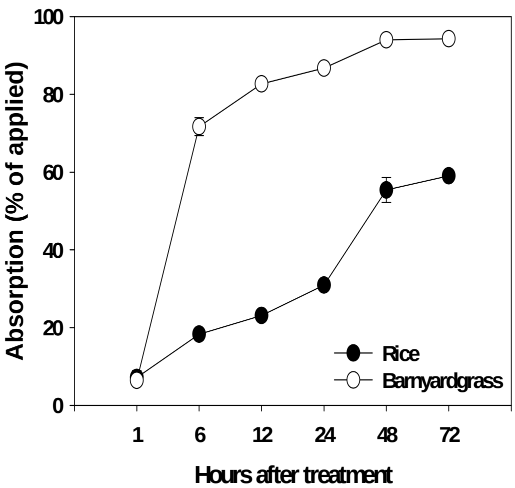

Figure 2. Foliar absorption of [^{14}C]flucetosulfuron in rice and barnyardgrass through 72 h. Vertical bars represent the standard errors of five replicates.

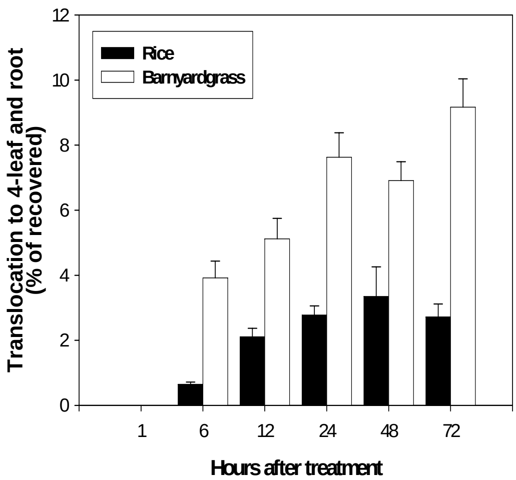

Figure 3. Translocation of [^{14}C]fluceotosulfuron to fourth-leaf and root in rice and barnyardgrass through 72 h. Vertical bars represent the standard errors of five replicates.

Metabolism studies

Flucetosulfuron and its metabolites were analyzed by HPLC using soluble extract concentration. The parent flucetosulfuron eluted at 19.3 min. Two major metabolites of flucetosulfuron eluted at 16.3 min (metabolite-1) and 8.1 min (metabolite-3), and one of minor metabolite eluted at 3.3 min (metabolite-2). Flucetosulfuron was metabolized very rapidly to metabolite-1 and metabolite-3 in rice and barnyardgrass and the rate of metabolism was not different between the both plants. At 1 HAT, metabolite-1 constituted 43.5 and 47.0% and metabolite-3 constituted about 7.4 and 6.0% of applied flucetosulfuron in rice and barnyardgrass, respectively. Flucetosulfuron and metabolite-1 in rice and barnyardgrass decreased with elapsed time and reached below about 2% at 144 HAT. Chronologically, metabolite-3 increased with time and reached at above 67% of treated flucetosulfuron. Metabolite-2 increased after 24 HAT, and consisted of 4.8 and 3.2% of applied flucetosulfuron, at 144 HAT in rice and barnyardgrass, respectively. These results showed that the rate of metabolism and the kinds of metabolites of flucetosulfuron was not different between in rice and barnyardgrass. The proposed metabolism pathway of flucetosulfuron in rice and barnyargrass is shown at Figure 4 and the metabolic profile is given in Table 2.

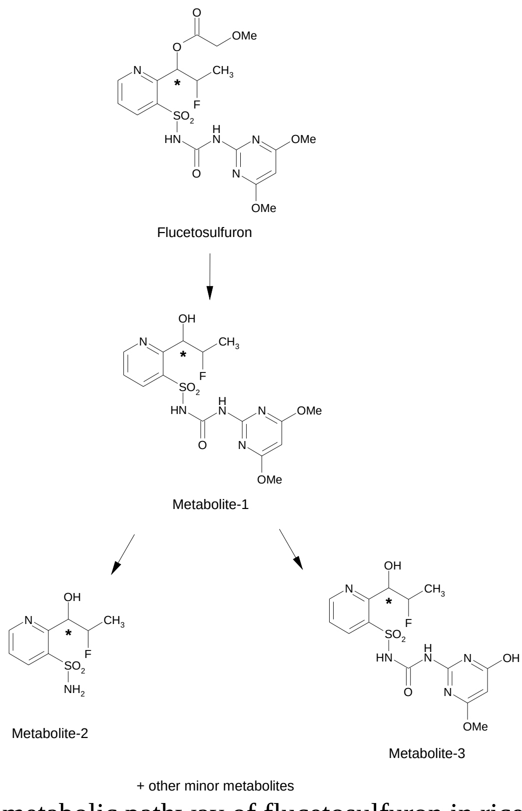

Figure 4. Proposed metabolic pathway of flucetosulfuron in rice and barnyardgrass.

Table 2. Metabolic profile of flucetosulfuron in rice and barnyardgrass

Time (h)	Rice					Barnyardgrass				
	Fluceto-sulfuron	M-1	M-2	M-3	Un known	Fluceto-sulfuron	M-1	M-2	M-3	Un known
	% of total extractable ^{14}C									
1	47.1	43.5	0.0	7.4	2.0	31.8	47.0	0.0	6.0	15.3
24	13.1	40.1	1.9	43.3	1.6	9.4	45.0	1.6	25.1	19.0
72	5.0	10.2	3.5	67.1	14.3	0.8	12.3	1.5	66.8	18.6
144	1.4	2.0	4.8	71.7	20.1	0.3	2.1	3.2	67.8	26.6

The proposed metabolic pathway of flucetosulfuron in rice and barnyardgrass is shown in Figure 4.In conclusion, the results indicated that the selectivity of rice to flucetosulfuron was primarily due to the lower foliar absorption and resulting in lower translocation, but not due to differential metabolism when compared to that in barnyardgrass.

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A STRATEGY ADOPTED BY WOODY PLANTS TO INVADE A WEED COMMUNITY

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Abstract: To clarify the mechanism of succession, we conducted a fixed point survey for 17 years on the school ground of reclaimed land of the Tokyo Bay coast to identify species composition in processes from an early secondary succession stage of a weedy community to a woody plant community. The dominant plant species at each stage in the secondary succession were *Eragrostis cilianensis*, *Digitaria ciliaris* , *Oenothera laciniata* (weedy stage 1) → *Erigeron sumatrensis*, *Erigeron canadensis* (weedy stage 2) → *Imperata cylindrica*, *Solidago altissima*, *Phragmites australis, Miscanthus sinensis* var. *sinensis* (weedy stage 3) → *Ligustrum lucidum*, *Pyracantha angustifolia*, *Zelkova serrata*, *Prunus speciosa*, *Quercus phillyraeoides*, *Daphniphyllum macropodum* var. *macropodum*, *Celtis sinensis* var. *japonica*, (woody stage). Shade tolerance and photosynthetic rate of these dominant species were examined to clarify the mechanism of succession and competition. Weedy stage 1 species grew poorly in 90% shade. Woody stage species were tolerant to this condition. In high sunlight, the photosynthetic rate of weedy stage 1 species was higher than that of the woody stage and weedy stage 2 and 3 species, the latter three groups of species sharing a similar photosynthetic rate. In addition, woody plants have shade and sun leaves on the same individual, depending on the habitat. The light compensation-point of the weedy stage 1, 2 and 3 species was higher than that of the woody stage species grown in the shade. The woody plant seedlings possessed strong shade tolerance because of their low light compensation-point, and woody plants could make sun leaves under full sunlight. These results suggest a strategy by woody plant seedlings to invade and to establish in a weedy community even under low light intensity.

Key words: Dominant species, light compensation-point, photosynthetic rate, secondary succession, weedy community, woody plant

Introduction

The management of abandoned land around cultivation areas and farmland will never be perfect. Despite the luxuriant growth of weeds and wild plants, woody plants invade the area before long, and a coexisting plant community of woody plants and weeds is formed (Aldrich and Kremer, 1997; Bard, 1952; Bazzaz, 1968; Clemennts, 1928; Hayashi, 1977; Iiizumi, 1968; Keever, 1952; Kobayashi *et al*. 2003; Lee, 2000; Numata, 1955; Numata and Suzuki, 1958; Numata and Yamai, 1955; Numata *et al*. 1964; Oosting, 1942; Pikett, 1982; Quatermann, 1957; Schmidt, 1981; Shimizu, 1969; Sugawara, 1978). This poses a big problem in agricultural production since the constitution of plant communities changes in meadows resulting in the predominance of perennial plants. As a result the management of an orchard becomes the management of a grassy plain over time, gradually reducing productivity of the former. The constitution of such a plant community suffers changes caused by environmental water, nutrients and physiological factors that define the interaction of a plant with its neighbours and with the plant community. This allows one to predict the change in dominant species and community composition, making it easy to manage this change positively; this has positive repercussions in vegetation management (Ito, 1993). Moreover, there is increasing attention to management technology that considers the choice of woody plants as the climax community, especially in residential areas (Chiba Prefectural Government, 2001).

Studies on the succession of a plant community are time-consuming, and there are few examples of continuous studies of a single area of land in Japan (Numata, 1987). Many studies have investigated a particular spot after a number of years following denudation, which can not be used to estimate a fixed point survey. Therefore, in order to better understand the shift from a weedy to woody plant community, we thought that it was extremely important to investigate the same location over a long term, in particular focusing on the vegetation of secondary succession in a plant community. The study, carried out over 17 years, was a school ground on reclaimed land of the Tokyo Bay coast.

There are strengths and weaknesses of tolerance shading of seedlings, in which photosynthetic rate largely influences the shift from a weed to a woody plant community, which is caused by seed germination, and whose resulting seedlings become firmly established, and then grow (Keever,1955). Therefore we chose a representative dominant species which was chosen based on results of a prior vegetation study of secondary succession of the plant community in this study; woody plants invaded the site and we assessed two factors, shade tolerance and photosynthetic rate of the weed community by examining its dominant species.

Materials and Methods

The dominant species in secondary succession of the plant community

The study was undertaken on reclaimed land of the Tokyo Bay coast on school grounds reclaimed from the sea between 1950 and 1980. We made a 3 m × 10 m permanent quadrats which was denuded of all vegetation in 1987 and investigated the rate at which plant communities covered the land and also plant height over a maximum of 17 years. Woody plants such as *Ligustrum lucidum* Ait., *Pinus thunbergii* Parl., *Lithocarpus edulis* Nakai, and *Prunus speciosa* Nakai were planted as a hedge and tree-planting in the school ground. In addition, *Castanopsis cuspidata* var. *sieboldii* Nakai, *Daphniphyllum macropodum* Miq., *Myrica rubra* Sieb. *et* Zucc. were planted in a tree-planting forest and *Ginkgo biloba* L. was planted as a roadside tree outside of the school grounds. An expressway passes about 50 m away from the school grounds. At the beginning of the study in 1987, we chose a vacant land on the school grounds, on which weeds such as *Solidago altissima* L. formed.

Shade tolerance of dominant species

Ten seeds of dominant species at each stage of succession were sown in 6 plastic pots per species, 13 cm high and 11 cm in diameter filled with field soil and placed in an unheated greenhouse. After germination, five seedlings 1 cm tall and with two leaves were placed per pot. These pots were divided into two treatments: shade and no shade (control). Shading strength was about 90% which measured the mean illumination five times above the topsoil of the sampling site. We measured plant height and fresh weight of the above ground part in the shade on the 50th, 65th or 90th day. Seedlings, which were germinated outdoors in plastic pots as done for the woody plants, were placed under shading and we measured fresh weight of the above-ground part on the 65th or 210th day.

Photosynthetic rate of dominant species

Erigeron species which were used for measurement of photosynthesis were sown in pots in May and were cultivated in a greenhouse. Other weedy stage species which were used for measurement of photosynthesis were collected from the field. woody species' seedlings were transplanted into pots and were cultivated under 90% shading and no shading condition in the greenhouse for more than one year. We measured photosynthetic rate at the center of each adult's leaves under different light strengths in the laboratory in August. Photosynthetic rate was measured for fresh air and the CO_2 density in the chamber was calculated with a hand-held Shimadzu-type photosynthesis transpiration meter SPB-H3 that could measure the quantity of absorption. The light source could be changed to emit photosynthetic active radiation from 0 to 600 µmol/m^2/s under natural light; using a HAYASHI Leminar Ace LA-150SX we measured light intensity and photosynthetic rate.

Results

Dominant species in secondary succession of a plant community

Table 1 shows dominant species on stage of succession in the study site. The dominant species of a secondary succession of the plant community in the study site were *Eragrostis cilianensis* (Th), *Digitaria ciliaris* , *Oenothera laciniata* (Th(w)) (Th) (weedy stage 1) → *Erigeron sumatrensis* (Th(w)), *Erigeron canadensis* (Th(w)) (weedy stage 2) → *Imperata cylindrica* (G), *Solidago altissima* (Ch,Th(v)), *Phragmites australis* (HH) → *Ligustrum lucidum* (M), *Phragmites australis* (HH), *Solidago altissima* (Ch,Th(v)), *Phragmites australis* (HH), *Miscanthus sinensis* var. *sinensis* (H) (weedy stage 3) → *Ligustrum lucidum* (M), *Pyracantha angustifolia* (M), *Prunus speciosa* (MM), *Zelkova serrata* (MM), *Quercus phillyraeoides* (M), *Daphniphyllum macropodum* var. *macropodum* (MM), *Celtis sinensis* var. *japonica* (MM) (woody stage). Observing the change in dominant species of a dormancy type, the following change could be observed: summer type annual plants (Th) → winter type annual plants (Th(w)) → perennial plants (Ch, H, HH, G, Th(v)) → woody plants (M, MM). Woody plants invaded the weed community at the third year after investigation.

Table 1. Dominant species on stage of succession

Stage of succession	Species (dormancy forms)	Dominance*					
		Years after investigation					
		1	2	3	5	10	15
Weedy stage 1	*Eragrostis cilianensis* (Th)	2	0	r	0	0	0
	Digitaria ciliaris (Th)	2	r	0	0	0	0
	Oenothera laciniata (Th(w))	1	r	r	0	0	0
Weedy stage 2	*Erigeron sumatrensis* (Th(w))	0	1	1	0	0	0
	Erigeron canadensis (Th(w))	0	1	2	0	0	0
Weedy stage 3	*Imperata cylindrica* (G)	0	r	2	2	0	0
	Solidago altissima (Ch,Th(v))	0	0	2	2	3	2
	Phragmites australis (HH)	0	r	1	3	3	2
	Miscanthus sinensis var. *sinensis* (H)	0	0	0	0	2	0
Woody stage	*Ligustrum lucidum* (M)	0	0	r	0	r	3
	Pyracantha angustifolia (M)	0	0	0	0	1	2
	Zelkova serrata (MM)	0	0	0	0	r	2
	Prunus speciosa (MM)	0	0	0	0	r	2
	Quercus phillyraeoide (M)	0	0	0	0	r	r
	Daphniphyllum macropodum	0	0	0	0	r	r
	Celtis sinensis var. *japonica* (MM)	0	0	0	0	r	r

(* height ×cover rate r :1-100, 1:100-1000, 2 :1000-10000, 3:10000-)

Shade tolerance of dominant species

Table 2 shows 90% shading tolerance and survival rate of dominant species at different stages of succession. There are substantial differences between 90% shaded and control plot in fresh weight of *D. ciliallis*, *E. phyladelphicus*, *E. sumatrensis*, *E. canadensis* and *S. altissima* which are weedy stage 1-3 species. The survival rate of *D. ciliallis* within the shaded plot was 76%. However the woody stage species showed similar fresh weights between 90% shading and control plot. It was clear that these woody stage species were strongly shade-tolerant.

Table 2. The 90% shading tolerance and survival rate of dominant species at different stages of succession

Stage of succession	Species	Shading period (days)	Fresh weight (% of control)	Survival rate (% of control)
Weedy stage 1	*Digitaria ciliaris*	65	1	76
Weedy stage 2	*Erigeron phyladelphicus*	50	10	100
	Erigeron sumatrensis	90	1	100
	Erigeron canadensis	90	23	100
Weedy stage 3	*Solidago altissima*	50	21	100
Woody stage	*Ligustrum lucidum*	65	91	100
	Celtis sinensis	210	115	100
	Daphniphyllum macropodum	210	124	94

Photosynthetic rate of dominant species

Table 3 shows photosynthetic rate and light compensation point of dominant species of secondary succession of the plant community. The photosynthetic rate of *D. cilialis*, *E. crus-galli* which are weedy stage 1 dominant species, was higher than that of weedy stage 2-3 and woody stage species at 400 µmol/m^2/s light intensity. Woody stage plants have sun and shade leaves on the same individual, depending on the habitat. The photosynthetic rate of the dominant species in the weedy stage 2-3 was similar to that of the woody stage plants. The photosynthetic rate of sun leaves of woody stage plants was higher than that of shade leaves. The light compensation point of sun leaves was also higher than that of shade leaves.

Table 3. Photosynthesis rate and light compensation point on dominant species at different stages of secondary succession of a plant community

Stage of succession	Species	Photosynthesis rate at 400 µmol/m^2/s (µmol/m^2/s)	Light compensation point (µmol/m^2/s)
Weedy stage 1	*Digitaria ciliaris*	14.5	36.1
	Echinochloa crus-galli	15.5	33.1
Weedy stage 2	*Erigeron annuus*	9.5	27.5
	Erigeron philadelphicus	8.1	36.7
	Erigeron canadensis	12.6	41.1
	Erigeron sumatrensis	13.2	35.9
Weedy stage 3	*Imperata cylindrica*	15.7	35.1
	Solidago altissima	8.2	34.9
	Phragmites australis	6.2	67.9
Woody stage	*Ligustrum lucidum* (sun leaf)	2.9	34.0
	Ligustrum lucidum (shade leaf)	1.0	6.6
	Celtis sinensis (sun leaf)	8.4	27.2
	Celtis sinensis (shade leaf)	3.2	2.0
	Daphniphyllum macropodum (sun leaf)	8.8	20.3
	Daphniphyllum macropodum (shade leaf)	3.8	5.7

Discussion

We chose the maximum photosynthetic rate and shade tolerance to be the factors that indicate the invasion of woody plants into a weed community in succession and performed an experiment to examine this hypothesis using the dominant species of a plant community. The seedlings of the dominant species of the woody stage showed high shade tolerance. In addition, the maximum photosynthetic rate of woody stage species was lower than the maximum photosynthetic rate of weedy stages 1 species. Furthermore, woody stage species have shade and sun leaves even if the leaves originate from the same species. In other words, the photosynthetic rate and light compensation point of seedlings under low light intensity was lower than sun leaves; subsequently, woody plants increased in height resulting in an increased photosynthetic rate as a result of increased light capture.

These results suggest that woody plants can invade a weed community and ultimately become the dominant species. Due to shade tolerance of the seedling, woody plants – which invade the weed community – are strong, for some reason, their seeds germinate and seedlings grow even in conditions of low light intensity in the lower layers of the weed community, a zone in which other weeds cannot grow. Eventually, the trees rapidly increase in height and adapts from a state of low light intensity to high light intensity with a corresponding rise in photosynthetic rate. As a result, these woody species become the dominant species of the plant community before long.

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UTILIZATION OF ALLELOPATHIC CROP WATER EXTRACTS FOR REDUCING HERBICIDE USE IN CANOLA (*Brassica napus* L.)

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Abstract: A field experiment was conducted at Agronomic Research Farm, University of Agriculture, Faisalabad, during the winter of year 2005-06 to investigate the allelopathic effects of crop water extracts as sorghum (*Sorghum bicolor* L.), sunflower (*Helianthus annus* L.), brassica (*Brassica campestris* L.) and rice (*Oryza sativa* L.) combined with low rates of pendimethalin for weed control in canola (*Brassica napus* L.). Crop water extracts at 15 l ha^{-1} each in combination with pendimethalin (Stomp® 33 EC) at 600 g a.i ha^{-1} and 400 g a.i ha^{-1} were sprayed immediately after sowing. Full dose of pendimethalin 1200 g a.i ha^{-1} was used as standard. A weedy check was also maintained for comparison. The treatments significantly decreased total weed density and dry weight over control. Sorghum and rice water extracts each at 15 l ha^{-1} combined with half dose of pendimethalin 600 g a.i. ha^{-1} showed maximum reduction in total weed density by 67.58 and 66.21% at 40 and 60 DAS respectively. All the treatments offered more than 80% reduction in total weed dry weight over control at 40 DAS while 44.93 to 63.99% reduction in total weed dry weight over control at 60 DAS were recorded in all the treatments. The plots treated with sorghum and sunflower water extracts each at 15 l ha^{-1} combined with half (600 g a.i. ha^{-1}) the recommended dose of pendimethalin produced maximum seed yield 2.604 Mg ha^{-1} which was 39.99% more as compared to control. In crux, herbicide dose may be reduced by 50-67% in combination with allelopathic water extracts for effective weed control and higher yields in canola.

Key words**:** Allelopathy, canola, herbicide reduction, pendimethalin, weed management.

Introduction

Food and oil processing industries have a great interest in canola (*Brassica napus* L.) production due to its premium quality oil. In Pakistan, canola is grown on area of 323,000 acres (130,713 ha) producing 162,000 tons of canola seeds and 58,000 tons of canola oil. The average yield of canola in Pakistan is 501 kg per acre (approx 203 kg per ha), which is very low compared to its potential yield (Govt. of Pakistan, 2006). Heavy weed infestation is an important reason for low yields in canola. Weeds not only compete with crop plants for nutrients, water, space and light but also give refuge to pests and diseases; interfere with crop growth by releasing allelopathic substances into the rhizosphere of the crop plants (Rice, 1984), interfere with harvest operations, and increase the processing costs and significantly reduce (21-45%) the crop yields (Ashiq and Ata, 2005). Herbicides offer a promising option to increase crop yields through effective weed control. However non-judicious use of herbicides has resulted in environmental and health problems. Surface and ground water used for human and livestock consumption is being continuously contaminated with toxic herbicides (Judith *et al.* 2001), and their entry to the food chain may prove hazardous even to future generations (Clarkson, 1995). These toxic herbicide residues in the environment and food chain have resulted in dangerous diseases not only in humans but also animals (Judith *et al.,* 2001). It has also been found that in some cases herbicide use can cause some weed species to dominate fields as the weeds develop resistance to herbicides (Heap, 2007). Environmental, health and resistance development issues, therefore, have a significant influence on the development of economical and environment friendly technologies for weed control which may reduce the use of chemical herbicides.

Allelopathy seems to be an effective, economical and natural method as well as alternative of herbicides for weed control. Although use of herbicides cannot be eliminated, their use can be reduced by exploiting allelopathy as an alternate weed management tool**.** There is a tremendous scope for exploring allelopathic phenomena for natural weed management (Bhowmik *et al.* 2003). Cheema *et al.* (2003a) proposed the use of allelopathic crop water extracts combined with lower herbicide rates as an economically viable and environment friendly weed control technique. Herbicides and allelopathic products can work complementary. As a result, the herbicide dose might be reduced when applied in combination with allelopathic products and effectiveness of the allelopathic products could be enhanced by using lower rates of herbicides (Cheema *et al.* 2005). Hence, the present studies were designed with the objectives to search the possibilities of reducing herbicide use in combination with allelopathic crop water extracts, exploit the efficacy of allelopathic crop water extracts by mixing them with a common pre-emergence herbicide pendimethalin and evaluate the combined effects of allelopathic crop water extracts of sorghum, sunflower, brassica and rice on weed growth in canola.

Materials and Methods

The experiment was conducted at Agronomic Research Farm, University of Agriculture, Faisalabad, (31.5°N, 73.09°S) during the winter season 2005-2006 to investigate the allelopathic effects of the crop water extracts from sorghum (*Sorghum bicolor* L.), sunflower (*Helianthus annus* L.), rice (*Oryza sativa* L.) and brassica (*Brassica campestris* L.) together with low dosages of pendimethalin (Stomp® 33 EC) for weed control in canola. Seeds of canola (*Brassica napus* L.) hybrid “Hyola 401” were obtained from Monsanto Pakistan Agri. Tech. (Pvt) Ltd., and planted in a row arrangement with a hand drill, having 30 cm row space on 6th of October 2005. Experiment was laid out in a Randomized Complete Block Design with a net plot size of 2.4 m x 5 m. The recommended seed rate of canola (5 kg ha^{-1}) was sown. Nitrogen was applied at 90 kg ha^{-1} while phosphorus was applied at 60 kg ha^{-1}. All phosphorus fertilizer was drilled at sowing, while nitrogen was applied in three equal splits at sowing, first irrigation and second irrigation.

The crop water extracts were prepared by harvesting the respective allelopathic crops at maturity, drying for few days under shade, and chopping them into 2 cm pieces with the help of an electric fodder cutter. The chopped material was soaked in the water at 1:10 (w/v) ratio for 24 hrs. Water extracts were collected by passing through sieves. The filtrates were boiled at 100 $^{\circ}$C for reducing the volume by 20 times. The concentrated crop water extracts were stored at room temperature (Cheema *et al*. 2000). Crop water extracts at 15 l ha^{-1} each in combination with pendimethalin at 600 g a.i ha^{-1} and 400 g a.i ha^{-1} were sprayed immediately after sowing by a knapsack hand sprayer fitted with a flat fan nozzle after calibrations using 320 liters of water per hectare. Recommended dose of pendimethalin (1200 g a.i ha^{-1}) was used as the standard. A weedy check was also maintained for comparison. Weed density and dry weight were recorded at 40 and 60 days after sowing (DAS) using a quadrate of 0.25 m^2. Weeds were cleaned, air dried under shade for 24 hrs and then oven dried at 70^{o}C for 72 hrs before recording their dry weight. Crop was harvested on 21st March 2006. Harvested crop was kept under sunlight for one week to dry. The biological yield of each plot was recorded through a spring balance and then converted to Mg ha^{-1}. The crop was threshed manually and the seeds obtained were weighed and then converted into seed yield (Mg ha^{-1}). The harvest index (HI) was calculated as the ratio of seed yield to biological yield and was expressed in %.

Economic and marginal analyses were performed according to procedure laid out by CIMMYT (1988). Data collected were subjected to Fisher s’ Analysis of Variance technique. Least Significant Difference test (p=0.05) was used to compare differences among the treatments means (Steel *et al.* 1997).

Results and Discussion

Weed flora of the experimental field consisted of purple nutsedge (*Cyperus rotundus* L.), horse purslane (*Trianthema portulacastrum* L.), lambsquarters (*Chenopodium album* L.) and swine cress (*Cronopus didymus* L.). Table 1 shows that all the treatments significantly suppressed total weed density when compared to the control.

Table 1. Effect of various allelopathic crop water extracts in combination with reduced rates of pendimethalin on total weed density (0.25 m^2) and total weed dry weight (g/0.25 m^2) in canola.

Treatments	Rate (Extract/Herbicide)	Total weed density (0.25m^2)		Total weed dry weight (g/0.25m^2)	
		40 DAS[1]	60 DAS	40 DAS	60 DAS
Control (weedy check)	-	86.0 a	46.88 a	7.037 a	4.258 a
Pendimethalin	1200g a.i. ha^{-1}	43.25 a (49.7)	25.38 c (46)	1.285 b (81.73)	2.325 b (45.39)
Sorghum WE[2] + Sunflower WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 600g a.i. ha^{-1}	40.0 cd (53.49)	27.0 c (42.55)	1.095 b (84.43)	1.855 c (56.43)
Sorghum WE + Sunflower WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 400g a.i. ha^{-1}	45.88 bc (46.65)	33.13 b (29.51)	1.11 b (84.23)	2.345 b (44.93)
Sorghum WE + Brassica WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 600g a.i. ha^{-1}	35.25 d (59.01)	17.0 e (63.83)	1.29 b (81.67)	1.548 e (63.66)
Sorghum WE + Brassica WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 400g a.i. ha^{-1}	44.0 c (48.84)	22.75 d (51.59)	1.09 b (84.51)	1.75 d (58.9)
Sorghum WE + Rice WE+ Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 600g a.i. ha^{-1}	27.88 e (67.58)	15.88 e (66.21)	1.005 b (85.71)	1.533 e (63.99)
Sorghum WE + Rice WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 400g a.i. ha^{-1}	51.5 b (40.12)	26.88 c (42.81)	1.197 b (82.99)	1.803 cd (57.65)

[1]Days after sowing; [2]Water extract. Within a column, means followed by the same letter are not significantly different (p=0.05). The figures in the parentheses show percent decrease over control.

The combined spray of sorghum and rice water extracts each at 15 l ha^{-1} with (half dose) pendimethalin (Stomp 33EC) at 600 g a.i. ha^{-1} gave inhibition by 68% of total weed density over control and it was followed by the sorghum and brassica water extracts each at 15 l ha^{-1} combined with half the recommended rate of pendimethalin (600 g a.i. ha^{-1}), where 59% reduction in total weed density was recorded at 40 DAS. Application of the water extracts of sorghum and rice water extracts (each at 15 l ha^{-1}) with half the recommended dose (600 g a.i. ha^{-1}) of pendimethalin and the combined spray of the sorghum and brassica water extracts with 600 g a.i.ha^{-1} of pendimethalin decreased total weed density by 66 & 64%, respectively, when compared to the control, at 60 DAS. All the treatments showed more than 80%

reduction in total weed dry weight at 40 DAS. Minimum weed dry weight at 60 DAS was recorded in plots applied with the water extracts of sorghum and brassica in combination with pendimethalin at 600 g a.i. ha^{-1}, and the water extracts of sorghum and rice in combination with pendimethalin at 600 g a.i. ha^{-1}. The results of both treatments were statistically similar. The recommended dosage of pendimethalin (1200 g a.i. ha^{-1}) was comparatively less effective at 60 DAS resulting in a 45% reduction in total weed dry weight when compared to the control. Reduction in total weed dry weight at 60 DAS was less as compared to reduction in total weed dry weight at 40 DAS, which may be due to the loss in efficacy of herbicide and water extracts over time.

These results indicated that the half of the recommended dosage of herbicide tank mixed with allelopathic crop water extracts offer better weed control than the recommended dosage of the herbicide, probably due to the strong allelopathic influence of different crop water extracts and the increased efficacy in herbicides when combined with allelopathic crop water extracts. Similar results have been reported by Cheema and Irshad (2004) on allelopathic effect of sorghum for control of barnyard grass in rice. The results also infer that pendimethalin dosage can be reduced up to 50-67% when combined with allelopathic water extracts for weed control in canola. Similarly, Cheema *et al*. (2002) suggested that pendimethalin dose can be reduced (more than 50%) when combined with concentrated sorgaab. Reducing herbicide dose in combination with allelopathic products for controlling weeds in field crops has been previously suggested by Cheema *et al.* (2003b) who reported 67.85 % reduction in total weed dry weight by the application sorghum water extract at 10 l ha^{-1} combined with three fourth of the recommended dose (667g a.i ha^{-1}) of pendimethalin for weed control in cotton. Similarly, Cheema *et al.* (2005) reported 81.72% reduction in total weed dry weight over control by application of sorghum water extract at 12 l ha^{-1} combined with 1/3rd rate of pendimethalin.

Data on seed yield are given in Table 2. The minimum seed yield was recorded in the weedy check, which was mainly due to the competition for resources among the weeds and crop plants. All the treatments increased seed yield over the control. The maximum seed yield of 2.604 Mg ha^{-1} was recorded in plots treated with the water extracts of sorghum and sunflower combined with half dose of pendimethalin, which was 40% more when compared to the control and 3% more when compared to the recommended dose (1200 g a.i. ha^{-1}) of pendimethalin. The water extracts if sorghum and brassica with half the recommended dose of pendimethalin produced grains equal to the recommended of pendimethalin. Cheema (1988) reported of increase in wheat grain yield by incorporation of sorghum residues in the soil for weed management. Khaliq *et al*. (2002) reported an increase in mungbean grain yield over control when treated with sorghum water extract tank mixed with reduced dose of pendimethalin. Cheema *et al.* (2003b) reported that the application of sorghum water extract at 10 l ha^{-1} combined with pendimethalin 333 g a.i. ha^{-1} increased seed cotton yield by 25.4 % over control.

The maximum increase in biological yield (Table 2) over control was recorded when the water extracts of sorghum and rice were applied in combination with 1/3rd rate of pendimethalin (400 g a.i. ha^{-1}). The Minimum biological yield (9.077 Mg ha^{-1}) was recorded in the weedy check. The increase in biological yield in different treatments over control was a result of the effective weed control. The results are supported by the findings of Erman *et al*. (2003) who reported that effective weed control increases biological yield. Different treatments significantly improved harvest index over control (Table 2). The minimum harvest index (20.61%) was recorded in weedy check. The maximum harvest index (25.14%) was recorded in plots treated with the water extracts of sorghum and brassica combined with half the recommended dose of pendimethalin, and was followed by the treatment of sorghum and sunflower water extracts combined with the recommended and half the recommended dosage of pendimethalin. The results are supported by findings of Cheema (1988) and Khaliq *et al.* (2002) who reported an increase in harvest index by incorporating sorghum as an allelopathic material as a weed control.

Table 2. Effect of various allelopathic crop water extracts in combination with reduced rates of pendimethalin on biological yield (Mg ha^{-1}), seed yield (Mg ha^{-1}) and harvest index (%) of canola.

Treatments	Rate (Extract/Herbicide)	Biological yield (Mg ha^{-1})	Seed yield (Mg ha^{-1})	Harvest index (%)
Control (weedy check)		9.077 d	1.86 f	20.61 e
Pendimethalin	1200g a.i.ha^{-1}	10.56 bc	2.529 b (35.99)	23.95 ab
Sorghum WE[1] + Sunflower WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 600g a.i. ha^{-1}	10.79 ab	2.604 a (39.99)	24.14 ab
Sorghum WE + Sunflower WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 400g a.i. ha^{-1}	10.71 b	2.455 c (32.0)	23.54 bc
Sorghum WE + Brassica WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 600g a.i. ha^{-1}	9.97 c	2.529 b (35.99)	25.14 a
Sorghum WE + Brassica WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 400g a.i. ha^{-1}	11.09 ab	2.381 d (28.0)	21.5 de
Sorghum WE + Rice WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 600g a.i. ha^{-1}	10.04 c	2.232 e (19.99)	22.26 cd
Sorghum WE + Rice WE + Pendimethalin	15 L ha^{-1} + 15 L ha^{-1} + 400g a.i. ha^{-1}	11.38 a	2.455 c (32.0)	21.6 de

[1]Days after sowing. [2]Water extract. Means followed by the same letter are not significantly different (p=0.05). The figures in the parentheses show percent decrease over control.

The usefulness of a weed control method is evaluated on the basis of its economics. Economic and marginal analyses of the experiment are given in Table 3 and 4, respectively. The maximum net benefits (Rs. 69375 ha^{-1}) were obtained from the treatment consisting of the water extracts of sorghum and sunflower combined with half the recommended dose of pendimethalin (600 g a.i. ha^{-1}). The water extracts of sorghum and sunflower combined with 1/3rd the recommended dose of pendimethalin (400 g a.i. ha^{-1}), sorghum and rice water extracts combined with 1/3rd the recommended dose of pendimethalin, sorghum and sunflower water extracts combined with half the recommended dose of pendimethalin (600 g a.i. ha^{-1}) were the economical treatments with 2059%, 2059% and 1793% marginal rates of returns, respectively. The results are supported by the findings of Cheema *et al*. (2002) who reported a Rs. 44445 net benefit and 427.7% marginal rate of return by application of sorghum water extract at 12 l ha^{-1} combined with 500 g a.i. ha^{-1} of pendimethalin for weed control in cotton.

Table 3. Economic analysis

	T_1*	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Remarks
Total seed yield	1.86	2.529	2.604	2.455	2.529	2.381	2.232	2.455	Mg ha^{-1}
Adjusted yield	1.674	2.276	2.343	2.209	2.276	2.142	2.008	2.209	Mg ha^{-1}
Gross income	50222	68302	70311	66293	68302	64287	60267	66293	Rs: 1200 per 40 kg
Cost of herbicide	0	1272.7	636.4	424.2	636.4	424.2	636.4	424.2	Rs: 350 l^{-1}
Cost of extracts	0		120	120	120	120	120	120	Rs: 60 per 15 l
Spray rent	0	60	60	60	60	60	60	60	
Spray application	0	120	120	120	120	120	120	120	
Cost that vary	0	1452.7	936.4	724.2	936.4	724.2	936.4	724.2	
Net benefit	50222	66850	69375	65569	67366	63562	59330	65569	Rs ha^{-1}

*T1= Control (weedy check); T2 = Pendimethalin (Stomp, 33 EC) at 1.2 kg a.i ha^{-1} (Pre-emergence); T3 = Sorghum WE at 15 l ha^{-1} + sunflower WE at 15 l ha^{-1} + pendimethalin at 0.6 kg a.i. ha^{-1} (Pre-emergence); T4 = Sorghum WE at 15 l ha^{-1} + sunflower WE at 15 l ha^{-1} + pendimethalin at 0.4 kg a.i. ha^{-1} (Pre-emergence); T5 = Sorghum WE at 15 l ha^{-1} + brassica WE at 15 l ha^{-1} + pendimethalin at 0.6 kg a.i. ha^{-1} (Pre-emergence); T6 = Sorghum WE at 15 l ha^{-1} + brassica WE 15 l ha^{-1} + pendimethalin at 0.4 kg a.i. ha^{-1} (Pre-emergence); T7 = Sorghum WE at 15 l ha^{-1} + rice WE at 15 l ha^{-1} + pendimethalin at 0.6 kg a.i. ha^{-1} (Pre-emergence); T8 = Sorghum WE at 15 l ha^{-1} + rice WE at 15 l ha^{-1} + pendimethalin at 0.4 kg a.i ha^{-1} (Pre-emergence); *Note*: Stomp® 33 EC was used as the source of pendimethalin.

Table 4. Marginal analysis

Treatments	Cost that vary	Net benefits Rs.	Change in cost	Change in net benefit	Marginal rate of return %
T_1 = Control	0	50222.7	-	-	-
T4 = Sorghum + sunflower WE each at 15 l ha^{-1} + pendimethalin (Stomp® 33 EC) at 400 g a.i. ha^{-1} (Pre-emergence)	724.2	65569.5	724.2	15346.8	2059.5
T8 = Sorghum + rice WE each at 15 l ha^{-1} + pendimethalin (Stomp® 33 EC) at 400 g a.i ha^{-1} (Pre-emergence)	724.2	65569.5	724.2	15346.8	2059.5
T6 = Sorghum + brassica WE each 15 l ha^{-1} + pendimethalin (Stomp® 33 EC) at 400 g a.i. ha^{-1} (Pre-emergence)	724.2	63562.8	-	-	D
T3 = Sorghum + sunflower WE each at 15 l ha^{-1} + pendimethalin (Stomp® 33 EC) at 600 g a.i. ha^{-1} (Pre-emergence)	936.4	69375.2	212.2	3805.7	1793.45
T5 = Sorghum + brassica WE each at 15 l ha^{-1} + pendimethalin (Stomp® 33 EC) at 600 g a.i. ha^{-1} (Pre-emergence)	936.4	67366.4	-	-	D
T7 = Sorghum + rice WE each at 15 l ha^{-1} + pendimethalin (Stomp® 33EC) at 600 g a.i. ha^{-1} (Pre-emergence)	936.4	59330.6	-	-	D
T2 = Pendimethalin (Stomp® 33 EC) at 1200 g a.i ha^{-1} (Pre-emergence)	1452.73	66850.07	-	-	D

D = Dominated due to less benefits than preceding treatments; Variable cost = the cost of purchase of inputs, labour and machinery ha^{-1} that vary between the experimental treatments; Net benefit = Gross income-variable cost; Marginal Rate of Return (%) = (Change in net benefits/Change in cost) x 100.

Conclusions

The findings of this study suggest that sorghum + sunflower combinations were better than sorghum + brassica or rice in terms of using water extracts in combination with different dosages of pendimethalin for weed control. Moreover, it is also noted that herbicide dose may be reduced by 50-67% in combination with allelopathic crop water extracts.

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WILD FLOWERING PLANTS AS HOSTS OF PARASITOIDS OF VEGETABLE PESTS

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Abstract: The agrochemical-based pest management strategies in the modern tropical agriculture systems are no longer effective and sustainable. As a part of the process of reverting to the nature's way, habitat enhancement of natural enemies by providing alternate weed hosts is a major area under research. A study was done to identify the most abundant parasitoids in an organically grown vegetable system (cabbage, bitter gourd, bean, brinjal) and the wild flowering plants that can act as alternative host plants of parasitoids of vegetable pests. Four species of parasitoids (*Braconid* sp., *Tachinid* sp., *Psytallia* sp. and *Opius* sp.) visited the wild flowering plants namely, *Bidens pilosa* var. *minor*, *Synedrella nodiflora, Tagetes erecta*, *Doronicum tenuifolium, Tridax Procumbens*, *Mimosa pudica,* and *Ipomoea triloba*, established adjacent to the vegetable plots. *Cleome rutidosperma, Commelina benghalensis,* and *Vernonia cinerea* were not identified as hosts of these parasitoids.

Key words: Vegetables, parasitoids, wild flowering plants

Introduction

At present, pest management strategies have been dominated by the use of chemical methods, due to quick results. The natural balance between insect pests and their natural enemies is often broken down by the use of chemicals. Therefore, there is an emerging trend to use long-term, eco-friendly pest control methods through the use of natural enemies (Driesche and Bellows, 1996; Lampkin, 1990).

Materials and Methods

Ten species of wild flowering plants were established in border strips surrounding the plots consisting of organically grown vegetables namely, cabbage, brinjal, bitter gourd, and bean. At the flowering stage of the wild flowering plants, 1 m x 1 m areas from six border strips representing ten plants were selected for observations. The field observations with respect to the visit of parasitoids were done each day between 07:30 – 08:30 hrs. Part of the plant where the parasitoids settled and duration of their resting time were recorded. All six squares were observed in 6 consecutive days and the procedure was repeated 5 times (30 days).

Results and Discussion

Four parasitoids, including 3 hymenopterans and 1 dipteran, visited the experimental plots. As the number of dipterans observed was low, the study focused mainly on the Braconid spp. (Hymenoptera). Out of the ten wild flowering plants, the parasitoids visited *Bidens pilosa* var. *minor*, *Synedrella nodiflora*, *Tagetes erecta*, *Doronicum tenuifolium*, *Tridax procumbens*, *Mimosa pudica,* and *Ipomoea triloba.* Parasitoids did not visit *Cleome rutidosperma, Commelina benghalensis,* and *Vernonia cinerea*. The frequency of visits made by the Braconid wasps to *Bidens pilosa* (70 visits per 30 hr period) was significantly higher than that made by the parasitoid to other host weeds (Figure 1). The duration of the visits made by the Broconids to *Bidens pilosa* plants indicated a strong stability of this host-plant relationship (Figure 2) as stated by Gautham (1994) and Maredia *et al.* (2003)

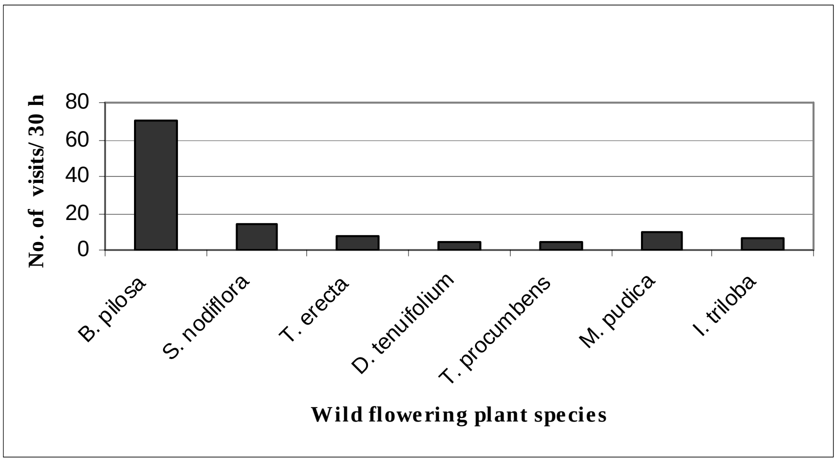

Figure 1. Number of visits done by Braconoids to wild flowering plants

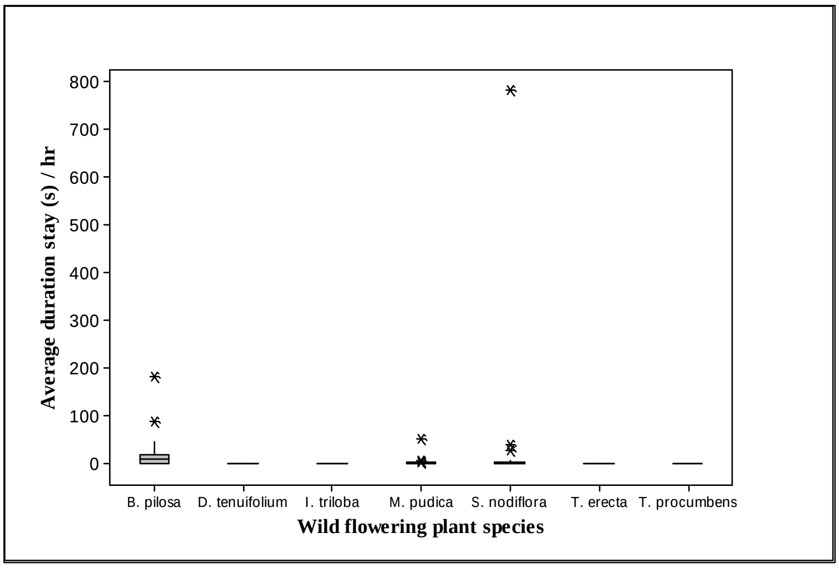

Figure 2. Average duration of stay of Braconids on wild flowering plants

Conclusions

Four species of parasitoids were found to visit seven out of ten wild flowering plants established adjacent to plots growing vegetables (Cabbage, Brinjal, Bitter Gourd and Beans). The parasitoids observed were *Braconidae* sp., *Tachinidae* sp., *Psyttalia* sp. and *Opius* sp. The alternate host plants were namely; *Bidens pilosa* var. *minor*, *Synedrella nodiflora*, *Tagetes erecta*, *Doronicum tenuifolium*, *Tridax Procumbens*, *Mimosa pudica*, and *Ipomoea triloba. Bidens pilosa* var. *minor* was the preferred host that enhanced the parasitoid population in the field.

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LABORATORY SIMULATED STUDIES ON THE PERSISTENCE BEHAVIOUR OF "PENOXSULAM 24 SC" IN SOILS OF DIFFERENT AGROCLIMATIC ZONES IN INDIA

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Abstract: Penoxsulam [2-(2,2-difluoroethoxy)–N-(5,8-dimethoxy) [1,2,4] triazolo [1,5-C] pyrimidin-2—yl) – 6 - (trifluoromethyl) benzene sulfonamide] a newly introduced post-emergent triazolopyrimidine sulfonamide group of herbicide is used to control important grasses (*Echinochloa colona*, and *E.crus-galli*) broadleaved weeds and sedges in transplanted, dry seeded and water seeded rice. In order to find the dissipation pattern, one laboratory simulated experiment was conducted taking different types of soils i.e. saline soil (pH 7.6) from Canning, neutral soil (pH 7.02) from Kalyani, acidic soil (pH 5.45) from Jhargram and black soil (pH 8.14) from Nasik at Pesticide Residue Laboratory, B.C.K.V., Mohanpur. The soil samples were fortified at 2 ppm (T_1) and 4 ppm (T_2) levels along with untreated controls (T_3). Soil samples were extracted with acetonitrile: water (8:2, v/v) followed by partitioning thrice with dichloromethane. The organic layer was concentrated in a rotary vacuum evaporator at 40°C .The final volume was made up with methanol for HPLC analysis (model JASCO PU 1580, coupled with UV/VIS detector and thermo hypersil (250 mm x 4.6 mm) 5 μ hypersil ® ODS column). HPLC analysis was done with the following parameters: wavelength 230 nm; mobile phase 100% Methanol; the flow rate 1 ml/min. Retention time of this compound was 2.25 min. The dissipation of penoxsulam follows first order kinetics irrespective of the doses and soil type. Initial deposit of penoxsulam varies from 1.73 - 1.89 ppm (T_1) and 3.41-3.58 ppm (T_2) irrespective of soil type. The half life ($T_{1/2}$) values were found to be in the range of 39.09 - 43.00 days (Acidic soil), 26.64 - 32.57days (Neutral soil) and 21.50 - 28.94 days (Saline soil) and 19.93 - 27.12 days (Black soil). No residue was detected in the untreated control throughout the study. The present study revealed that the penoxsulam dissipated faster in Black soil followed by Saline soil and Neutral soil. Penoxsulam is more stable in acidic soil.

Key words: Dissipation, herbicide, penoxsulam, soil.

Introduction

In the Indian sub-continent as well as in the whole world rice (*Oryza sativa*) is one of the most important cereal crops which suffer yield reduction by the infestation of weeds throughout its life cycle. The loss of yield due to weeds ranges from 25-30% (Upadhyay and Gagoi, 1993) and yearly loss of rice grain production in India is 32.5% (Bhattacharyya *et al.*, 2004). Environmental conditions favorable for rice production are also suitable for growth of weeds. Only 16% herbicide among the total pesticide consumption, is used to control weeds in India. (Yaduraju *et al.* 2005). Because of indiscriminate high doses pesticides, weeds of plantation and cereal crops (mainly wheat and rice) in developing countries have developed resistance against herbicides. Penoxsulam [2-(2,2-difluoroethoxy)–N-(5,8-dimethoxy) [1,2,4] triazolo [1,5-C] pyrimidin-2—yl)-6-(trifluoromethyl) benzene sulfonamide] a newly introduced post-emergent triazolopyrimidine sulfonamide group of herbicide is used to control important grass, broadleaved and sedge weeds in transplanted, dry seeded and water seeded rice (EPA Pesticide fact sheet, 2004). It has outstanding effects on all *Echinochloa* grasses, the major weed in rice around the world and the major broad-leaved and sedge weeds when applied between the two-leaf and mid-tillering stage of rice (Larelle *et al*. 2003) .Research recommendation on chemical control of pests is considered incomplete if data on toxic residue of the herbicide are not provided. However, a literature survey reveals that no systematic work has yet been done to study the nature of dissipation and residual fate of this herbicide in soils of different agro climatic conditions in India as well as in the world. In this context, the present investigation was undertaken to determine the dissipation pattern and the residual level of penoxsulam in soils under four different agro climatic regions of India.

Materials and Methods

Analytical grade(98.4% pure) of penoxsulam [2-(2,2-difluoroethoxy)–N-(5,8-dimethoxy) [1,2,4] triazolo [1,5-C] pyrimidin-2—yl) – 6 - (trifluoromethyl) benzene sulfonamide] was obtained from M/S Dow Agro Science India Pvt. Ltd. The experiment was conducted at the Pesticide Residue Laboratory, Kalyani, BCKV, during the year 2005-06 with the 100 gm of air dried, finely sieved soil (20 mesh). Soil samples were collected from four different agro climatic zones of India, *i.e.* Lateritic soil (Jhargram, pH 5.45), Gangetic alluvial soil (Kalyani, pH 7.02), coastal saline soil (Canning, pH 7.6) and black soil (Nasik, pH 8.14). The Physico-chemical properties of the soils are given in the Table 1.

Table1. Physico-chemical characteristics of the soils.

Physico-chemical properties of soil	Lateritic soil (Jhargram)	Gangetic alluvial soil (Kalyani)	Coastal saline soil (Canning)	Black soil (Nasik)
pH	5.45	7.02	7.6	8.14
Organic Carbon (%)	0.64	1.00	1.03	0.67
Percentage of sand	54	12	50	16
Percentage of slit	16	65	23	24
Percentage of clay	30	23	27	60

The stock solution of penoxsulam (100 ppm) was prepared in methanol with the analytical grade (98.4% pure) and applied to the soils at 2 ppm (T_1) and 4 ppm (T_2) along with untreated control (T_3). In control samples, 2 ml and 4 ml of methanol was added. Water was added to maintain the moisture level at 80% of water holding capacity. The conicals were covered with few holes and kept at room temperature.

A portion of water was lost everyday through evaporation and hence that quantity of water was added after every 24 hrs to maintain the specific moisture status. Three replicates for each treatment were maintained in 250 ml conical flasks (Borosil). Soil samples for each treatment and soil type were taken replication-wise at different day intervals, *i.e.* 0 (after 2 hrs of spiking), 3, 7, 15, 30, 45, 60 and 90 days after spiking of penoxsulam. All the reagents used were of analytical reagent grade and all the solvents were redistilled before use. Water used was double glass distilled. The soil collected from each treatment replicate was dissolved in 100 ml of acetonitrile: water (8:2, v/v) in a 250 ml conical flask and kept overnight. Then the content was shaken with mechanical shaker for one and half hours. The content was filtered through Buchner funnel and rinsed thoroughly with acetonitrile: water (8:2, v/v). The filtrate was concentrated in a rotary vacuum evaporator at 40°C. The concentrated extract was transferred in a separatory funnel with 100 ml of water and 20 ml of saturated sodium chloride solution. Then it was partitioned thrice with (100 + 50 + 50) ml dichloromethane. The organic layer was collected over anhydrous sodium sulphate and concentrated in a rotary vacuum evaporator at 40°C. The final volume was made up to 10 ml with methanol for analysis by HPLC (JASCO-JAPAN; Model: UV-1575) equipped with UV-VIS detector coupled with Chemito 5000 data processor. The C-18 reverse phase column [Thermo Hypersil (250 mm x 4.6 mm i.d.) 5 µ Hypersil® ODS] along with guard column was used. The 100% methanol was used as mobile phase for the detection of penoxsulam residue. The other parameters like flow, wave length (λ max), retention time, Limit of detection (LOD) and

Limit of quantification (LOQ) were 1 ml/min, 230 nm, 2.25±0.2 min, 0.01 ppm and 0.05 ppm, respectively. In order to establish the reliability of the analytical method adopted and to know the efficiency of the steps employed for the determination of dissipation of penoxsulam in soil of different agroclimatic regions, soil samples were fortified with 0.1, 1 and 5 ppm of analytical grade penoxsulam standard and the average recoveries were found to be in the range of 86.47 to 89.93%.

Results and Discussion

The residue data, dissipation percentage, regression equation and half life values of penoxsulam occurring in soil are presented in Tables, 2, 3, 4 and 5.

Table 2. Persistence of penoxsulam in Jhargram soil.

Treatment applied	DAT	M ± S.D. (% Dissipation)	Regression Equation [Half-life($T_{1/2}$)]
T_1 (2 µg/g)	0	1.89 ± 0.03 (-)	Y = 3.2430-0.0077X [39.09 days]
	3	1.64 ± 0.04 (13.23)	
	7	1.41 ± 0.03 (25.39)	
	15	1.24 ± 0.03 (34.39)	
	30	1.12 ± 0.04 (40.74)	
	45	0.82 ± 0.03 (56.61)	
	60	0.58 ± 0.034 (69.31)	
	90	0.37 ± 0.05 (80.95)	
T_2 (4 µg/ g)	0	3.58 ± 0.05 (-)	Y = 3.5450 -0.0070X [43.00 days]
	3	3.24 ± 0.04 (09.49)	
	7	3.09 ± 0.06 (13.69)	
	15	2.59 ± 0.07 (27.65)	
	30	2.24 ± 0.06 (37.43)	
	45	1.96 ± 0.04 (45.25)	
	60	1.39 ± 0.05 (61.17)	
	90	0.71 ± 0.05 (80.17)	

Table 3. Persistence of penoxsulam in Kalyani soil.

Treatment applied	DAT	M ± S.D. (% Dissipation)	Regression Equation [Half-life($T_{1/2}$)]
T_1 (2 µg/ g)	0	1.81 ± 0.07 (-)	Y = 3.2468 – 0.0113X [26.64 days]
	3	1.69 ± 0.04 (06.63)	
	7	1.25 ± 0.04 (30.94)	
	15	1.16 ± 0.03 (35.91)	
	30	0.89 ± 0.05 (50.83)	
	45	0.52 ± 0.04 (71.27)	
	60	0.31 ± 0.04 (82.27)	
	90	0.19 ± 0.04 (89.50)	
T_2 (4 µg/ g)	0	3.52 ± 0.05 (-)	Y = 3.5671 -0.0093X [32.57days]
	3	3.09 ± 0.05 (12.22)	
	7	2.78 ± 0.04 (21.02)	
	15	2.57 ± 0.05 (26.99)	
	30	2.29 ± 0.04 (34.94)	
	45	1.63 ± 0.04 (53.69)	
	60	1.19 ± 0.04 (66.19)	
	90	0.44 ± 0.05 (87.50)	

Table 4. Persistence of penoxsulam in Canning soil.

Treatment applied	DAT	M ± S.D. (% Dissipation)	Regression Equation [Half-life($T_{1/2}$)]
T_1 (2 µg/ g)	0	1.75 ± 0.04 (-)	Y = 3.2383 -0.0140X [21.50days]
	3	1.61 ± 0.05 (08.00)	
	7	1.18 ± 0.04 (32.57)	
	15	0.98 ± 0.04 (44.00)	
	30	0.75 ± 0.05 (57.14)	
	45	0.36 ± 0.04 (79.43)	
	60	0.21 ± 0.04 (88.00)	
	90	0.11 ± 0.04 (93.71)	
T_2 (4 µg/ g)	0	3.46 ± 0.09 (-)	Y = 3.5415 -0.0104X [28.94 days]
	3	3.01 ± 0.05 (13.01)	
	7	2.85 ± 0.09 (17.63)	
	15	2.21 ± 0.08 (36.13)	
	30	1.99 ± 0.04 (42.49)	
	45	1.47 ± 0.05 (57.51)	
	60	0.84 ± 0.06 (75.72)	
	90	0.35 ± 0.07 (89.84)	

Table 5. Persistence of penoxsulam in Black soil.

Treatment applied	DAT	M ± S.D. (% Dissipation)	Regression Equation [Half-life($T_{1/2}$)]
T_1 (2 µg/ g)	0	1.73 ± 0.04 (-)	Y = 3.2330 - 0.0151X [19.93 days]
	3	1.58 ± 0.03 (08.67)	
	7	1.11 ± 0.05 (35.84)	
	15	0.89 ± 0.05 (48.55)	
	30	0.70 ± 0.05 (59.54)	
	45	0.32 ± 0.05 (81.50)	
	60	0.17 ± 0.04 (90.17)	
	90	0.09 ± 0.03 (94.79)	
T_2 (4 µg/ g)	0	3.41 ± 0.06 (-)	Y = 3.5037 -0.0111X [27.12 days]
	3	3.08 ± 0.04 (09.68)	
	7	2.45 ± 0.05 (28.15)	
	15	2.01 ± 0.04 (41.06)	
	30	1.53 ± 0.06 (55.13)	
	45	1.11 ± 0.05 (67.45)	
	60	0.68 ± 0.04 (80.06)	
	90	0.31 ± 0.05 (90.91)	

From the results, it was found that the penoxsulam residue was significantly decreased with the increment of time and followed first order kinetics irrespective of soil types and treatments. No residue was obtained in the untreated control throughout the study. Initial deposit of penoxsulam varied from 1.73- 1.89 ppm (T1) and 3.41-3.58 ppm (T2) irrespective of soil types.

The half life ($T_{1/2}$) values were found to be in the ranges of 39.09-43.00 days (Acidic soil), 26.64-32.57days (Neutral soil) and 21.50-28.94 days (Saline soil) and 19.93-27.12 days

(Black soil). The present study revealed that the penoxsulam dissipated faster in Black soil followed by Saline soil and Neutral soil. Penoxsulam is more stable in acidic soil. The present findings are also comparable with the earlier studies of Roberts *et al.* (2003) who found that penoxsulam was rapidly dissipated in water and soil following the first-order kinetics. Residues in soil were low with a half life value of 13 to 16 days.

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IMPACT OF DIFFERENT HERBICIDES ON *Amaranthus spinosus* IN KOREA

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Abstract: In Korea, 315 species of exotic weeds were found from 37 families. Some exotic weeds need systematic management because weeds such as *Amaranthus spinosus*, inflicted enormous damages on environment and/or human beings and livestock. Therefore, selecting herbicides and establishment of effective weed control methods of *Amaranthus spinosus* in pasture of Korea, has been set up. Results of field testing of pendimethalin 31.7% EC in treated plots showed a weeding effect of 10%. However, pendimethalin 31.7% EC+MCPP 50% SL, MCPP 50% SL and dicamba 48.2% SL treated plots showed weeding effect of 95%, 95% and 100%, respectively. The optimum treatment period is from early June to mid July.

Key words: *Amaranthus spinosus*, exotic weeds, herbicides, weed control.

Introduction

The results of a survey showed 315 exotic weed species from 37 families during 2001 in Korea (Oh *et al.* 2002). Among exotic weeds found in the country, some weeds were adapted completely to the inland environment, while the others are adjusting and was were found in some regions of Korea. As trade of agricultural products is increased with recent liberalization of import, many seeds of exotic plants mixed with imported grain were brought in to the country. Some exotic weeds were being managed by government because of environmental and human hazards. Therefore, the spread of exotic weeds needs to be controlled at an earlier stage. *Amaranthus spinosus* was introduced in 1970’s in Jeju island, Korea (Ryang *et al.* 2004; Park, 1995). This is an annual weed, mainly occurring in Jeju pasture causing damage to human and livestock as prickles and nitrate in plant, Livestock especially get poisoned by nitrate resulting in excessive cattle death. So, this study was conducted to control the troublesome exotic weed, *A. spinosus,* by several herbicides. Results of this experiment provide guidelines to effective management and prevention of spreading.

Materials and Methods

This experiment was conducted in pasture, which were densely covered with *Amaranthus spinosus* in Jeju Island, Korea. The list of herbicides to control *A. spinosus* in the field is shown in Table 1.

Table 1. The list of herbicides to control of *A. spinosus* in the field

Herbicides	Content (%)	Dosage (10a)	Remarks
Pendimetnalin EC	4.5	300 ml	Soil treatment
Dicamba SL	48.2	400 ml	Foliar treatment
MCPP SL	50.0	500 ml	Foliar treatment
Triclopyr-TEA SL	30.0	500 ml	Foliar treatment
Pendi + MCPP	-	300+500 ml	-

Herbicides were treated on 24th May, 3rd June and 18th July. Weeding effect was investigated by visual estimation at 30 days after treatment (DAT).

Results and Discussion

A pre-test was conducted in greenhouse to control *A. spinosus* in pasture. Some herbicides were selected based on results of the pre-test. These were soil treatment with pendimethalin EC, and foliar treatment with dicamba SL, MCPP SL and triclopyr-TEA SL (Table 2).

Table 2. Weeding efficacy to control *A. spinosus* in a pre-test in greenhouse

Herbicide	Weeding efficacy (%) by *A. spinosus* growth stage		
	1st Step (Plant height was less than 15 ㎝)	2nd Step (Plant height was less than 30 ㎝)	3rd Step (Plant height was less than 50 ㎝)
Alachlor EC	75 [a]	-	-
Pendimethalin EC	80 [a]	-	-
Dicamba SL	95	90	80
MCPP SL	100	100	100
Triclopyr-TEA SL	100	100	100

a - Alachlor and pendimethalin were soil treatment herbicides.

A. spinosus was controlled by pendimethalin EC + MCPP SL, dicamba SL and MCPP SL in pasture (Table 2). Pedimethalin EC, showed 80% weeding effect in greenhouse, but in open field showed a very low weeding effect (*i.e.* 10%). Because of variation of treatment time, *A. spinosus* did not germinate in greenhouse. But, in open field it did germinate.

Table 3 shows that weeding efficacy was highest from 3rd June to 18th July in treated plots. Hence, it is recommended that control of *A. spinosus* by foliar herbicides must be done from early June to early July.

Table 3. Weeding effect of *A. spinosus* by several herbicides in pasture by visual estimation at 30 days after treatment

Herbicides	Weeding effect (%) by treated time		
	24th May	3rd June	18th July
Pendimethalin EC	10	-	-
Dicamba SL	80	95	95
MCPP SL	92	100	95
Triclopyr-TEA SL	20	-	-
Pendimethalin + MCPP SL	90	100	95

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MODE OF ACTION OF A NEW ISOXAZOLINE COMPOUND

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Abstract: A new isoxazoline compound [5-(2,6-difluorobenzyl)oxymethyl-5-methyl-3-(3-methylthiophen-2-yl)-1,2-isoxazoline; code name, EK-5229] was reported to have safety in rice and control barnyardgrass (*Echinochloa crus-galli*), but its mode of action was unknown. The compound did not inhibit germination of sensitive grasses, but it inhibited further growth. The symptom was primarily stunting and discoloration, but did not resemble those of existing herbicides. In whole plant studies, EK-5229 completely inhibited root elongation of corn, susceptible grass, at 1μM within 6 h of application. EK-5229 inhibited incorporation of [^{14}C]glucose into hemicellulose and cellulose of corn root cell walls by 50 to 70% at 1μM, after 24 h. The effect of EK-5229 on cell wall biosynthesis was dose-dependent in the range between 0.01 and 1μM, and corresponded with the dose-dependency of growth of corn root. However, in terms of time-course response, inhibition of [^{14}C]glucose incorporation into the cell wall constituents was evident after 12 h of treatment, which was after the growth inhibition occurred. This result suggested that the mechanism of action of EK-5229 might be directly or indirectly associated with cell wall biosynthesis in susceptible plants.

Key words: EK-5229, cell wall biosynthesis, isoxazoline

Introduction

Herbicidal new molecules in 5-benzyloxymethyl-1,2-isoxazoline derivatives were published by Ryu *et al*. (2002). These compounds were known to have the herbicidal activity and selectivity to rice with an unknown mechanism of action. The EK-5229, 5-(2,6-difluorobenzyl)oxymethyl-5-methyl-3-3(3-methylthiophen-2-yl)-1,2-isoxazoline, was a representative compound of 5-benzyloxymethyl-1,2-isoxazoline derivatives, and demonstrated good rice selectivity and potent herbicidal activity against annual weeds include barnyardgrass at 125 g a.i. ha^{-1} (Hwang *et al*. 2003). But the mode of action of this molecule has been unknown (Hwang *et al*. 2003). In this paper, we report herbicidal characteristics of EK-5229 in terms of symptoms and whole plant growth response, and possible involvement of its mechanism in cell wall biosynthesis metabolism.

Materials and Methods

Biological symptom test

Seeds of barnyardgrass (*Echinochloa crus-galli*) were used in the biological symptom test. Seeds were placed on 9 cm diameter Petri dishes containing 10 ml of the herbicide solutions namely, EK-5229 (purity 95%, synthesized at Korea Research Institute of Chemical Technology), dichlobenil (purity 97%, Aldrich), butachlor (33% suspension concentrate formulation, Dongbu Hannong Chemical, Korea) and pendimethalin (31.7% EC, Dongbu Hannong Chemical, Korea), at the concentrations 0.1, 1, 10 and 100 μM. The Petri dishes were kept in a growth chamber controlled at 25°C. The germination rate, length of leaves and roots were measured at 7 day after treatment.

Corn root response

Seeds of water-soaked corn were planted in moist vermiculite. When the seeds germinated and the primary roots elongated to about 3 cm, the seedlings were transferred to a hydroponic culture in a solution containing 5 mM Mes-Tris buffer (pH 5.0) and 0.2 mM $CaCl_2$. After adjustment period for 1 h, EK-5229 was applied by replacing the solution with the same buffer solution containing the EK-5229 in various concentrations. For dose response study, concentrations of EK-5229 tested varied from 0.0001 to 10 µM, and the treatment duration was 24 h. Net growth of each seedling during 24 h was measured. For time-course study, root growth was measured at 0, 1, 2, 4, 6 and 24 h after herbicide treatment, and the concentrations of EK-5229 were 1 and 10 µM. All treatments consisted of 8 replicates of corn root.

[^{14}C]Glucose incorporation into the cell wall
Plant materials were prepared according to the corn root response experiment described above. The roots of corn was treated with EK-5229 (1 µM) and other known cell wall biosynthesis inhibitors, dichlobenil (1µM) and quinclorac (10 µM) for 24 h. [^{14}C]Glucose (1 mCi/ml, ARC) was added in the final 1 h of the herbicide treatment period. After 24 h, 15 mm root tips were cut, blotted and placed into a vial. Incorporation of glucose into the cell wall was determined as described by Koo *et al.* (1997). The tissues were sequentially extracted in 50% methanol (2 h), 100% DMSO (24 h), 50 mM EDTA (pH 6.5) (1 h in boiling water bath), 1 N KOH (24 h room temperature), and acetic acid:nitric acid: water (8:1:2) (2 h in boiling water bath). Between each extraction procedure, the plant materials were washed with excess quantity of 50% methanol using a micro-filter system on a glass filter (Millipore, Durapore® 0.45µm HV).

The volume of all extracts was 1 ml and an aliquot (0.5 ml) of each extract was counted by liquid scintillation spectroscopy (LSS). Resulting insoluble cell wall materials (cellulose) were analyzed by sample oxidizer. The methanol-soluble and DMSO-soluble fractions represented cytoplasmic unincorporated glucose and its derivatives, and starch incorporated glucose, respectively. The EDTA fraction represented pectin (Li *et al.* 2006) One N KOH- and acid-soluble fractions represented hemicellulose mainly composed of glucuronoarabinoxylan (GAX) with different levels of substitution (Koo *et al.* 1997).

Dose response
Corn roots were treated with EK-5229 at 0, 0.001, 0.01, 0.1 and 1 µM for 24 hr, and [^{14}C]glucose (1 mCi/ml, ARC) was added during the final 1 h treatment as above.

Time-course response
Corn roots were treated with EK-5229 at 1µM for 1, 2, 6, 12, 24 h, and [^{14}C]glucose (1 mCi/ml, ARC) was added during the final 1 h treatment as above. Analysis was the same as above.

Results and Discussion

Biological symptom
The EK-5229 did not inhibit germination of barnyardgrass, but caused severe inhibition of leaf and root length even at 1 µM (Table 1). The biological symptoms such as germination, leaf length and root length were observed with the treatment of dichlobenil, butachlor and pendimethalin. However, the morphological symptoms were different. In case of EK-5229, coleoptiles changed into a thin and broad shape, and showed violet color. Dichlobenil and pendimethalin similarly resulted in thick leaves and roots, but the color of the coleoptile treated with pendimethalin was dark green than when treated with dichlobenil. The color of root treated with pendimethalin also changed into green but there was no change of root color treated with dichlobenil. Butachlor stopped the growth of coleoptiles, the color of coleoptiles changed into dark, and there was necrosis of leaf. Some other possible modes of action were recognized as unlikely on the basis of the biological symptoms observed. EK-5229 did not

bleach plants (and thus was unlikely to inhibit the biosynthesis of carotenoids), did not cause rapid scorching (and thus was unlikely to act in a similar way to inhibitors of protoporphyrinogen oxidase or redox mediators of photosystem I), and did not show any hormonal symptoms.

Table 1. Herbicidal activity of each herbicide on germination, leaf and root length.

Herbicide	Dose (µM)	Germination (%)	Leaf length	Root length
			- % of control -	
EK-5229	100	81.8	31.1	4.4
	10	92.7	31.5	6.1
	1	93.2	66.3	10.1
	0.1	92.9	115.6	74.6
Dichlobenil	100	81.8	3.2	0.1
	10	80.9	9.1	2.2
	1	88.4	102.0	68.4
	0.1	93.1	101.5	115.2
Butachlor	100	92.9	16.0	16.7
	10	87.5	21.3	57.5
	1	90.5	80.2	87.6
	0.1	95.8	100.2	97.9
Pendimethalin	100	84.0	13.4	4.7
	10	91.0	17.9	6.5
	1	88.9	63.3	35.0
	0.1	87.5	83.7	31.8
Untreated	-	95.9	100.0	100.0

Corn root response

Corn has been identified as a susceptible plant to the test herbicide (data not shown). The corn root was highly susceptible as shown in Table 1, and hence the corn root response was characterized. The root growth of corn was inhibited by EK-5229 with an IC_{50} (concentration that causes growth inhibition by 50%) of 0.03 µM (Figure 1A). Complete inhibition was obtained at 1 µM. In terms of time-course, growth was completely inhibited from 6 h after treatment, and there was no difference between 1 and 10 µM (Figure 1B).

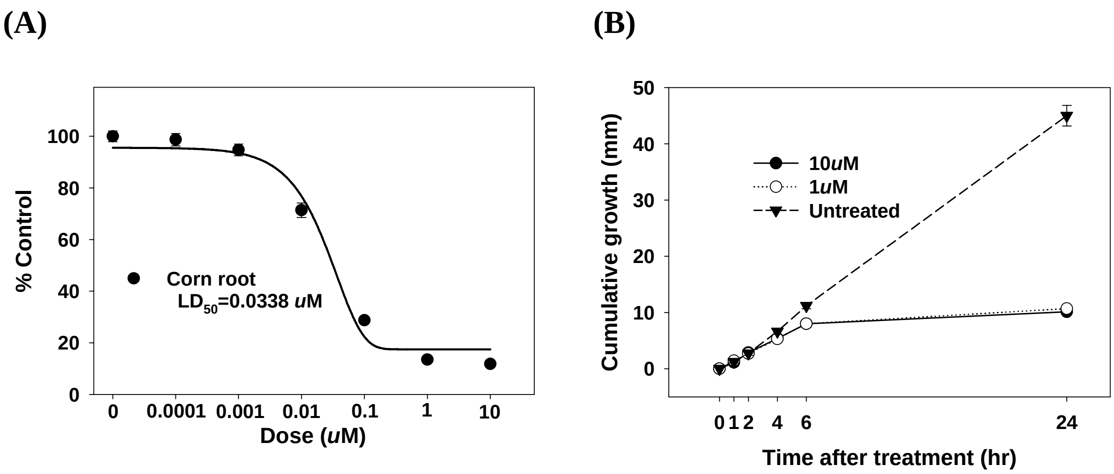

Figure 1. Corn root response to EK-5229 in terms of dose (A) and time-course (B). Each point is the mean of 8 replicates ± standard deviation.

[^{14}C]Glucose incorporation into the cell wall

The EK-5229 inhibited glucose incorporation into both hemicellulose (GAX 1 and GAX 2) and cellulose (Figure 2). Dichlobenil was known to be a selective inhibitor of cellulose (Hogetsu *et al*. 1974). In this study, selective inhibition of cellulose was reconfirmed but the extent of inhibition was rather small (<20%). Quinclorac was reported to inhibit both hemicelluose and cellulose involving barnyardgrass and crabgrass (Koo *et al*. 1997). In the present study, strong inhibition of cellulose was reconfirmed, but inhibition of hemicellulose was not such notable. This difference might be due to plant material differences. Anyhow, inhibition by EK-5229 of glucose incorporation into the cell wall was much greater than those by dichlobenil and quinclorac. Incorporation of glucose into pectin increased as with unincorporated radiolabels in cytoplasm.

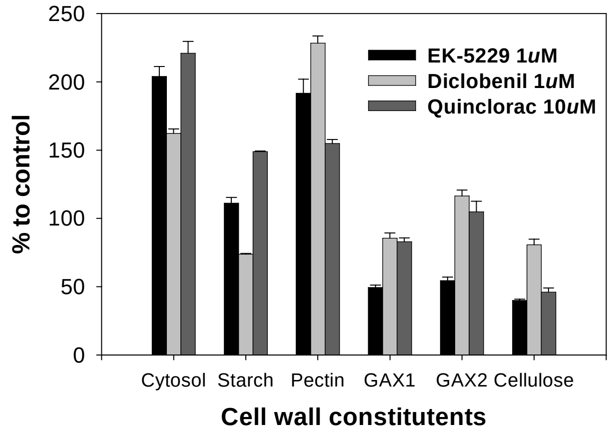

Figure 2. Inhibition of [^{14}C]glucose incorporation into cell wall of corn primary roots at each concentration of EK-5229, dichlobenil and quinclorac.

In dose response, inhibition of glucose incorporation into the cell wall was evident from 0.1 µM (Table 3). In the time-course response, inhibition of glucose incorporation into the cell wall was evident from 6 h after treatment (Table 4).

Table 3. Dose response of EK-5229 on the incorporation of [^{14}C]glucose into corn cell wall constituents.

Concentration (µM)	Cellulose	GAX1	GAX2	Pectin	Starch	Cytoplasm
			- % to control -			
Untreated	100.0	100.0	100.0	100.0	100.0	100.0
0.001	96.5	100.8	115.9	97.1	92.0	119.5
0.01	109.9	102.7	119.4	103.0	99.7	109.3
0.1	42.0	79.6	69.5	128.6	117.4	141.7
1	29.8	45.1	51.9	167.2	115.4	165.0
LSD (p=0.05)	26.04	17.23	26.33	19.05	12.56	22.82

From these results, it was clear that EK-5229 inhibited glucose incorporation into the cell wall dramatically, in particular, both hemicellulose and cellulose similarly. This activity was quite consistent with the corn root growth response, but not completely correspondent. For example, corn root growth was sensitive at 0.01µM, but at this concentration glucose incorporation into the cell wall was not inhibited.

Table 4. Time-course effect of 1μM EK-5229 on the incorporation of [^{14}C]glucose into corn cell wall constituents.

Hours after treatment	Cellulose	GAX1	GAX2	Pectin	Starch	Cytoplasm
			- % to control -			
Untreated	100.0	100.0	100.0	100.0	100.0	100.0
2	125.3	79.8	110.9	105.6	94.2	83.1
4	124.5	103.6	105.3	100.4	100.9	102.2
6	149.2	158.9	132.2	136.6	128.8	117.2
12	59.1	80.8	62.4	161.4	124.5	105.4
24	18.3	45.2	26.9	164.1	105.3	99.7
LSD (p=0.05)	13.64	15.21	18.38	43.01	15.73	15.95

In time-course, corn root growth was inhibited as fast as 6 h, but glucose incorporation was inhibited after 12 h. Despite some inconsistency, glucose incorporation into the cell wall by EK-5229 was very dramatic. Therefore, it is clear that the mechanism of EK-5229 and related isoxazoline compounds is associated with the cell wall biosynthesis in susceptible plants. However, whether the inhibitory effect is a primary or indirect one remains to be investigated.

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WHITE LEAFHOPPER (*Balclutha saltuella* Baum): A POTENT INSECT FOR BIOLOGICAL CONTROL OF SPRANGLETOP [*Leptochloa chinensis* (L.) Nees] IN RICE CULTIVATION

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Abstract: Sprangletop (*Leptochloa chinensis* L. Nees) is a noxious grass weed in pre-germinated wet-seeded rice cultivation in central Thailand. It could reduce rice yields by 20-100% depending on the level of infestation. In some areas, herbicides have failed to control this weed species as a result of the development of herbicide resistance. Hence, the white leafhopper (*Balclutha saltuella* Kirschbaum) (Homoptera: Cicadellidae), a grass feeding species, has been identified as an alternative control method. Prior to utilization of white leafhopper, biology, ecology, crop selectivity, host plants, potential of weed damage, response to insecticides and being a possible vector of *rice ragged stunt virus* (RRSV) were investigated. The results showed high host-specificity of white plant hopper on sprangletop, with no damage on rice. The insect could not complete life cycle and found dead within 7 days after feeding on six rice varieties. However, Crabgrass (*Digitaria sanguinalis* L. Scop.) was an alternate host. All insecticides affecting brown planthoppers, were also effective on this species. Forty adults could reduce seed formation of sprangletop by 60% resulting in the depletion of seed bank in soil. In addition, RRSV was unlikely to be transmitted by this insect as no symptom of RRSV disease appeared in a disease-sensitive rice variety, Taichung native 1. Hence, white leafhopper is a potent biological control method for the control of sprangletop in the rice fields.

Key words: *Balclutha saltuella*, *Leptochloa chinensis*, biological control

Introduction

Sprangletop (*Leptochloa chinensis* L. Nees) is a troublesome grass weed in the direct wet-seeded rice in Thailand. To control this species, herbicides have been continuously applied resulting in the development of resistance to ACCase-inhibiting herbicides (Maneechote *et al.* 2005). In 2003, white planthopper (*Balclutha saltuella* Kirschbuam), Family Cicadellidae, was firstly reported as a potent insect for biological control of sprangletop in Thailand. The preliminary study showed that 33-87 white plant hoppers on one panicle of the grass reduced seed formation by 99% (Maneechote *et al.* 2003), where the insect attacked only sprangletop with no impact on rice. Interestingly, *B. saltuella* was not recorded as a pest in any crops, and is a grass-feeding species. In the past, the population densities of *B. saltuella* were small in the rice fields.

To-date, sprangletop has been widespread as a result of the development of herbicide resistance (Maneechote, unpublished data). There may be a correlation between the distribution of sprangletop and the presence of white plant hopper, as some sprangletop plants showed symptoms of *rice ragged stunt virus* (RRSV) disease, which is normally transferred by brown planthopper (*Nilaparvata lugens*) (Wilson and Claridge, 1991). More investigations are needed to check whether *B. saltuella* could be a vector of this disease in both sprangletop and rice. Hence, a study was carried out understand the biology, ecology, crop selectivity, host plants, potential of weed damage, response to insecticides and being a possible vector of *rice ragged stunt* virus (RRSV) of white leafhopper (*B. saltuella*) prior to making use of this species as a biological control agent sprangletop.

Materials and Methods

Rice yield loss by sprangletop (L. chinensis) infestation

During dry season 2006, crop-cut data were collected in 21 rice fields cultivated to the variety Supanburi1, in the Supanburi province, Thailand. The number of sprangletop and rice plants in the field was also counted in 1 x 1 m^2 area of 4 replicates. The percentage of sprangletop density was calculated from the proportion of sprangletop plants available to the total numbers of rice and sprangletop plants. The correlation between the density of sprangletop and rice yield was also demonstrated

Life cycle of white leafhopper (B. saltuella)

Laboratory study was conducted at the Department of Rice, Bangkok during June- August 2005. Twenty female white leafhoppers were placed on the panicles of sprangletop for six hours before removal. Hatching period of 50 eggs were observed. After hatching, 40 of the first instar nymphs were collected and fed on a panicle of sprangletop in a 100 ml test tube fitted with cotton stopper. The molting periods of nymphs were daily recorded.

Selectivity of white plant hopper on rice varieties

To examine the impact of the insect on cultivated rice, six popular varieties grown in Central Plain namely, Hom Klong Luang, Pathum Thani1, Chainat1, Bang Taen, Pitsanoloke 2 and Supanburi 2, were grown in the pots under glasshouse condition. Pots were arranged in a Randomized Complete Block Design (RCBD) with 5 replicates. One plant per pot was used as one replicate. Sprangletop was also grown as a check. At the flowering stage of the main stem, each variety were separately kept in an insect rearing case sized 50 x 50 x1 50 cm^3 and five males and five females of white leafhoppers were released in each case. Rice plants free of white leafhoppers were used as the untreated control. At 1, 3, 5, 7 and 30 days after treatment, the number of nymph and adult of white leafhoppers on rice and sprangletop panicles were counted.

Alternate host plants of white leaf hopper

Flowers of six weed species commonly found nearby the rice fields *i.e*. *Echinochloa colona* L. Link., *Digitaria sanguinalis* L. Scop., *Panicum repens* L., *Chloris barbata* L., *Cyperus iria* L. and *C. difformis* L. were separately placed in 15 ml test tubes moistened with distilled water. Ten adults of white plant hoppers were placed in one tube with 5 tubes per weed species. Number of white plant hoppers (adults and nymphs) was recorded at 7, 14 and 21 days after feeding.

Rice ragged stunt virus transmitted by white leafhopper

Two experiments were conducted in laboratory in the Department of Rice, Bangkok during 2005-2006 to investigate the ability of the white leafhopper to transmit the *rice ragged stunt virus* (RRSV). Firstly, eight samples of sprangletop showing symptoms of RRSV disease were collected from the fields in Supanburi province and examined by using Dot Immuno-binding Assay (DIBA) using antiserum against RRSV (Sanimthong *et al*. 1994; Omura *et al.* 1992). Secondly, the second-instar nymphs of the white leafhopper were fed on rice plants infected with RRSV for 2, 4 and 6 days. Then they were transferred on to 7 day old seedlings of a disease-sensitive rice variety, Taichung Native 1 (TN1). Those rice seedlings were transplanted in a plastic box filled with clay soil for 30 days in order to observe the expression of disease symptoms on rice plants.

Potential of white leafhopper on the reduction of sprangletop seed bank
A glasshouse experiment was conducted to investigate the effects of number of white leafhoppers on seed formation of sprangletop, during November 2005- January 2006. Sprangletop plants were individually grown in 30 cm plastic pots. At the flowering stage, pairs of males and females (10, 20, 30 and 40 pairs) of white leafhopper were released in each pot placed in the insect rearing case. An un-treated pot was used as the control. Two weeks later, the panicles of sprangletop were collected and the number of filled and unfilled seeds was counted. Ten panicles were used for each treatment. Data represented the average (one panicle =one replicate) ± standard errors of ten replicates.

Influence of some insecticides on white leafhoppers
Fifty panicles of sprangletop with nymphs and adults of white leafhoppers were taken from the field and transported to the laboratory. Twenty panicles were placed in 100 ml beaker and used as one replicate. Seven treatments were arranged in a complete randomized design (CRD) with 4 replicates. Treatments were buprofezin 10%WP, isoprocarb 50%WP, fenobucarb 50%EC, ethofenprox 10%EC, carbosulfan 20%EC, imidacloprid 10%SL and distilled water. Insecticides were sprayed on panicles of sprangletop using revolving turntable sprayer with 12.5 ml spray volume. Three hours after application, panicles was individually covered with plastic bags and placed in water to keep them fresh. The number of dead and alive insects was counted after 24 h from treatment. The experiment was repeated in the following week.

Results and Discussion

Rice yield loss by sprangletop infestation
Rice yield was about 5 t ha^{-1}, without sprangletop infestation. When density of sprangletop was increased by 40%, the rice yield was reduced by > 50% (Figure 1). Since 2003, the number of herbicide-resistant sprangletop populations has been increasing in the rice growing areas in central Thailand, thus resulting in higher competitive pressure of this grass weed on the rice crop.

Biology and Ecology of white leafhopper
The presence of sprangletop in higher population densities may be the reasons why white leafhoppers were abundantly appearing on sprangletop plants at the flowering and seed formation stages (Maneechote *et al.* 2003). Consequently, seeds of sprangletop plants were mostly steriled after insect invasion.

The life cycle of white leafhopper was 20-24 days. Duration of the egg stage was 6 ± 0.40 days (n = 50), first-instar nymphs 3 ±0.46 days (n = 40), second-instar nymphs 3 ± 0.47 days (n = 39), third-instar nymphs 3 ±0.23 days (n = 36), fourth-instar nymphs 4 ± 0.28 days (n = 35) and fifth-instar nymphs 4 ± 0.24 days (n = 34). The suitable mating time was in the early morning or late evening. Once mated, females laid their eggs within the young spikelets before the formation of seeds. First-instar nymphs are greenish brown or black with approximately 0.4 mm in length. The body length of male and female (n = 20) adults were 1.69 x 0.11 and 2.08 x 0.14 mm, respectively.

White leafhoppers (*B. saltuella*) were usually found at the flowering stage of sprangletop. They stayed only on flowers of this grass species but did not appear on the stems or leaves. Interestingly, they did not appear on any parts of the rice plants. Nymphs were less active than adults and stayed on the peduncles and pedicels of the panicles sucking for the honey dews of sprangletop. Adults would sit on the panicles or moved onto other panicles nearby. However, they become more active in the evening.

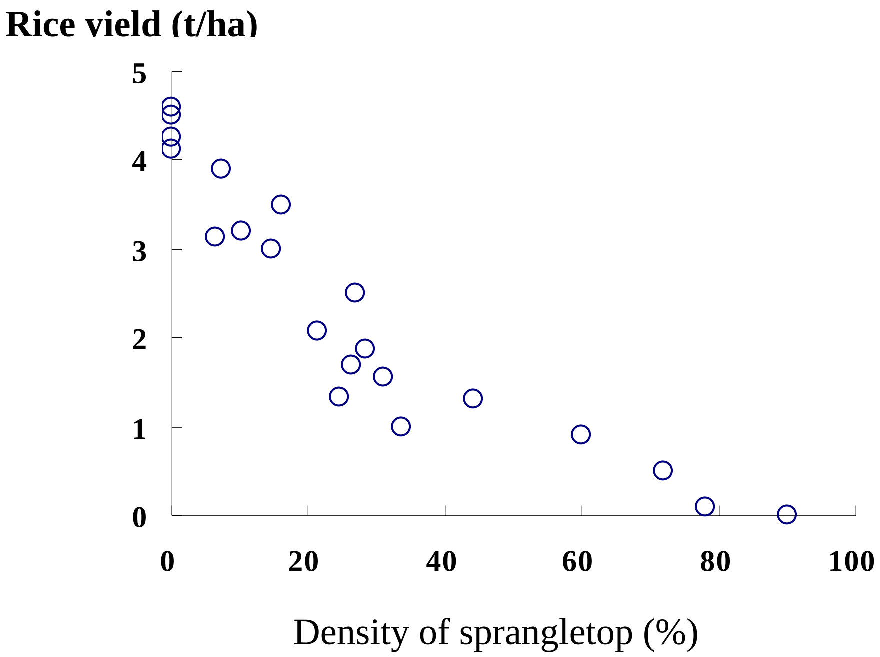

Figure 1. Correlation between the density of sprangletop (*L. chinensis*) and yield of Supanburi1 rice variety.

Selectivity of white leafhopper on rice varieties

At 1, 3, 5 and 7 days after treatment, all white leafhoppers disappeared in all tested rice varieties and sprangletop. However, at 30 days after treatment, a number of nymphs were found on the panicles of sprangletop but none was found on panicles in any of the rice varieties tested. It was clear that white leafhoppers could not live on rice but it was highly specific predator on sprangletop.

Alternative host plants of white leafhopper

Five weed species namely, *Echinochloa colona* L. Link., *Panicum repens* L., *Chloris barbata* L., *Cyperus iria* L. and *C. difformis* L. were not found to be preferred alternative host plants of white leafhoppers as all insects were dead within 7 days after feeding. In contrast, they could live and complete the life cycle when crabgrass (*Digitaria sanguinalis* L. Scop.) was the host. This grass species was also reported to be a host plant of another species of leafhopper (*B. incisa* Matsumara) (Narhardiyati and Bailey, 2005). Crabgrass is a common upland weed distributed throughout Thailand and its spikelets are similar to those of sprangletop. Frequently, both nymphs and adults of *B. saltuella* were found on the panicles of crabgrass nearby the rice fields. When panicles of sprangletop were not available, white leafhopper could live on crabgrass as an alternate host plant.

Rice ragged stunt virus transmitted by white leafhopper

Positive reaction of *rice ragged stunt virus* (RRSV) was observed in all samples of sprangletop plants expressing symptom of RRSV disease in the field. However, further investigations showed that the disease-sensitive rice variety TN1, did not show any RRSV symptoms after transmitted by white leafhopper, for 2, 4 and 6 days. In contrast, this rice variety would develop RRSV symptom within 30 days, after the virus is transmitted by brown planthoppers for 2-7 days. Sanimthong *et al.* (1994) reported that sprangletop was a host plant of RRSV transmitted by brown plant hopper. Thus, the sprangletop plants expressing RRSV symptom may be previously attacked by brown planthopper at the early stage and the symptom was shown when white leafhoppers appeared in the fields. In addition, glasshouse experiment showed that rice plants did not appeared to be a host plant of white leafhopper as they stayed only on sprangletop panicles.

In 1978, *rice ragged stunt virus* (RRSV), transmitted by brown planthopper (BPH), was firstly documented as a cause of severe damage to rice yields in many provinces in central Thailand (Chettanachit *et al*. 1978). During the last three years, there were no RRSV reported from the Supanburi province. Hence, it was unlikely that white leafhopper was a vector of RRSV on the tested rice varieties. More investigations are needed clarify this in the other susceptible rice varieties before the insect is used as a bio-control agent of sprangletop in rice fields.

Potential of white leafhopper on the reduction of sprangletop seed bank

Without white leafhopper, seed sterility of sprangletop was very low at 10 ± 2 % (Figure 2).

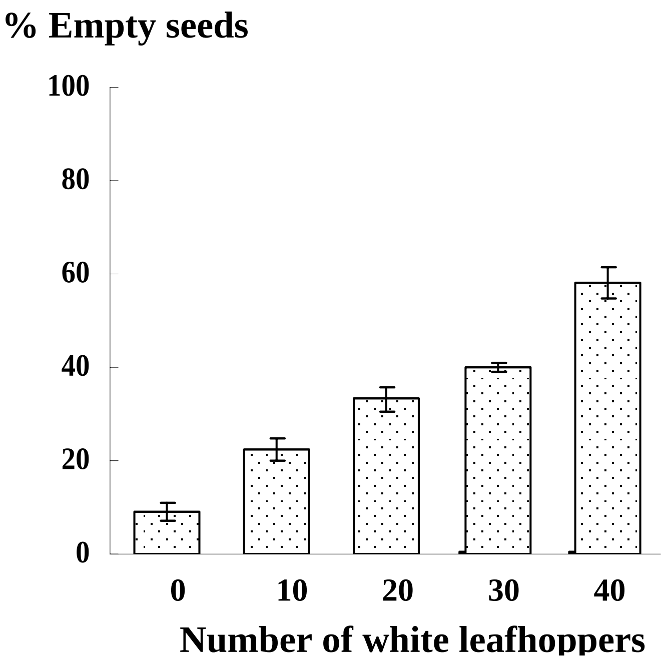

Figure 2. Effect of number of white plant hoppers (*Balclutha saltuella*) on empty seeds (%) in sprangletop (*Leptochloa chinenesis*) at two weeks after feeding. Vertical bars represented standard error of ten replicates.

The percentage of seed sterility increased when the panicles were fed with 10-40 insects per plant. Seed formation was inhibited by 60% when 40 insects were released on one plant. Previous study showed that 33-87 white leafhoppers (adult and nymph) naturally found on one grass panicle could cause seed sterility by 99% (Maneechote *et al.* 2003). Hence, 40 insects were effective to reduce seed formation in this grass species resulting in the reduction of seed bank in the following season.

Influence of some insecticides on the mortality of white leafhoppers

All insecticides, except buprofezin, used in this study at the recommended dosage killed white leafhopper (Table 1). Buprofezin resulted in only 66.4% mortality. As buprofezin is chitin inhibitor, it did not show a fast action like the other contact and systemic insecticides used in this study.

These insecticides were generally sprayed at 15-30 days after sowing rice to kill BPH. The flowering period of sprangletop is 30-45 days after sowing rice. To preserve white leafhopper in the rice fields, farmers will have to stop spraying BPH insecticides prior to the emergence of sprangletop panicles. However, the mass production of this insect may be required if the populations of white leafhopper was lower than 10 insects per panicle (Figure 2).

Table 1. Effect of insecticidal applications on the mortality of white leafhoppers (*B. saltuella*)

Treatment	Trade name	% mortality of *B. saltuella*		Average
		Exp I	Exp II	(%)
buprofezin	Applaud 10% WP	79.0	53.8	66.4
isoprocarb	Mipcin 50%WP	100.0	99.4	99.7
fenobucarb	Bassa 50% EC	100	99.5	99.7
ethofenprox	Treton 10% EC	100	100.0	100.0
carbosulfan	Posse 20% EC	100	100	100.0
imidacloprid	Confidor 10% SL	99.7	100	99.8
Control (water)	-	17.05	4.35	10.7

The species of genus *Balclutha* are distributed worldwide and known as the grass-feeding species with no report of becoming pests of important crops (Knight, 1987). In Thailand, there are 11 species reported in many crops such as rice, eggplant, soybean, cotton and orange (Hongsapruek, 1992). In 2000, a field survey indicated that only four species of *Balclutha* namely, *B. incisa* (Matsumura), *B. rosea* (Scott), *B. rubrostriata* Melichar and *B. viridinervis* Matsumara, were found in the rice fields (Hongsapruek, 2000), which was the first report on widespread of *B. saltuella* in the rice fields in Thailand without crop damage. As white leafhopper (*B. saltuella*) was not likely to be a vector of RRSV disease in rice and has a highly specific damage on sprangletop with no attention on rice plants, it is a potent insect for biological control of sprangletop (*L. chinensis*) in the rice fields.

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CURRENT STATUS ON INFLUX AND HABITAT OF EXOTIC WEEDS IN KOREA

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Abstract: For investigation on the present status of exotic weeds in Korea, the number of exotic weed species, their inflow time and native regions were examined with literature. Field surveys were also conducted for seven years from 1995 to 2001. Exotic weeds naturalized in Korea are composed of 315 species in 37 families until 2001. Among the weed flora, 135 exotic weed species have been reported since 1980. The number of exotic weeds by the native regions was 72, 32, 122 and 29 in North America, South America, Europe and Asia, respectively. Many species of exotic weeds are distributed near the ports, roadside for transport of imported cereals, circumference of highway and reclaimed land with wastes. 72 and 56 exotic weeds are found in farmland and pasture, respectively. Also, exotic weed newly found in cultivated area from 1995 to 1999 was reported as 34 species from 13 families.

Key words: Exotic weeds, in-flow time, native regions, habitat.

Introduction

Weeds found in Korea can be discriminated as native weeds and exotic ones. However, all weeds not originating in the country are called foreign weeds. In addition, naturalized weeds are those exotic ones that are completely adapted to the domestic environment. However, because influx time of weeds for naturalization is not determined clearly, all the weeds inhabiting the country from foreign lands are considered proper to be called exotic weeds. A study on inland exotic weeds was initiated first about plants native to the American continent (Lee and Kim, 1961). Lee and Oh (1974) did a research on naturalized plants of Korea and a study on distribution of naturalized plants of the Korean peninsula was carried out (Lim and Jean, 1980). Park reported to the Plant Taxonomy Association of Korea that exotic species had not been recorded inland from 1992 to 2001. Among exotic weeds found in the country, some weeds were adapted completely to the inland environment, and the others were adjusting and found in some regions of Korea. As trade of agricultural products increased with recent liberalization of import, many seeds of exotic plants mixed with import grain were brought in the country. Accordingly, this study was carried out to elucidate the kind of exotic weeds flown in to the country, inflow time, native origin and main places of occurrence and obtain basic information for management of exotic weeds

Materials and Methods

Survey on kinds of exotic weed through different documents
In order to survey exotic weeds distributed in the country, weedy plants of alien native origin based on plant picture book published inland and an article, were chosen and summarized by family. Exotic weeds investigated were rearranged by species and only the legitimate name was written using rule of international plant nomenclature after identifying basionym, synonym and misidentified name.

Influx time and native place of exotic weed
Date of first entry to the country was surmised based on reference and plant books pre-reported, even if that may not be completely accurate. Depending on references when exotic weeds were reported firstly, influx periods were sorted by 5 periods (before 1910; before the opening of port, 1911~1945; governing period of Japan, 1946~1960; the restoration after war,

1961~1980; growing period of economy, 1981~1999; period of free trade). We used several references such as Flora Korean (1911 Nakai), Plant name of Josun (1949 Biology Society of Josun), Plant name of Korea (Lee and An, 1963), Plant name of Daehan (Lee, 1982) and Color picture of Naturalized plant of Korea (Lee, 1995) to obtain information on influx time of exotic weeds. Native region by species was investigated through many relevant articles.

Survey on distribution and inhabitation of exotic weeds
In order to obtain information on distribution and inhabitation of exotic weeds, we usually surveyed the surroundings of main harbors such as Inchun, Pusan, Gunsan, Mokpoe and Yeosu from which grain is usually imported, near roadside for transport of imported grain and around dairy farms and pasture land. In addition, we surveyed exotic weeds troublesome in cultivated areas including crops like feed corn and grains. Newly found species were identified based on domestic and foreign plant picture books.

Results and Discussion

Exotic weeds found domestically by late 2001 were identified as belonging to 315 species of 37 families and 18 orders (Table 1). Table 8 shows the species by different families and native regions and inflow time to Korea. In Japan, naturalized plants of 403 species were identified in Colored Illustration of Naturalized Plants of Japan (Takemasa, 1992) and 165 species were recorded as exotic weeds of Japan (Livestock Society of Japan, 1994). Park (1995) published picture books including 261 exotic weed species and Kang and Shim (2002) reported 471 exotic weed species. However, it is difficult to compare these with our results because Kang and Shim (2002) includes some flowering grass and cultivated crops in their results.

Table 1. Number of exotic weed species by family.

Family	No. of species	Family	No. of species
Poaceae	51	Malvaceae	9
Polygonaceae	12	Convolvulaceae	8
Chenopodiaceae	7	Solanaceae	13
Amaranthaceae	14	Scrophulariaceae	9
Caryophyllaceae	13	Asteraceae	65
Brassicaceae	31	Others 25 Family	63
Fabaceae	20	Total 37 Family	315

When 315 exotic weeds inhabiting domestically were categorized according to plant families, the family Asteraceae 65 had species, Poaceae 51 species, Brassicaceae 31 species and Fabaceae 20 species. These families formed 53% of total exotic weeds and the rest were Polygonaceae, Amaranthaceae, Solanaceae and Caryophyllaceae having 12 to 14 species.

Influx time and native region of exotic weeds
The species number by influx periods of exotic weeds is shown in Table 2. Before 1910, even if that time is very long ago, 54 species were reported as exotic weeds.

Table 2. Number of exotic weed species by introduction period

Introduction Period	~’10	’11~’45	’46~’60	’61~’80	’81~’01	Total
No. of Species (%)	54 (17)	46 (15)	16 (5)	64 (20)	135 (43)	315 (100)

If we are to determine the period before 1910 to be least 200 years, this period conforms to about 1711~1910 during which some trade to foreign countries was carried out. Most of 55 exotic weeds are considered as be flown at that time. After this period, 46 species and 16 species were introduced during 1911~1945 (the period of Japanese annexation of Korea to independence of Korea) and 1946~1960 (early period after the independence), respectively. Influx of exotic weeds during the latter period is reported as little even though much American agricultural products were coming in. This is considered as due to the low interest about new plant species and exotic weeds considering the situation at that time. With the advent of real industrialization from 1961 to 1980, 64 exotic weed species are reported. Since 1980, as a lot of alien agricultural products have been imported with opening of importation, many exotic weeds have flown during this period, with 135 species being identified amounting to 43% of total exotic weeds. Increase of exotic weeds was because of the influx of seeds of exotic weeds coming as contaminations of imported grains.

Native region of exotic weeds

Because exotic weeds were not only confined to the native region, the native region of exotic weeds was not the country from which they were flown to Korea. However, native regions provide important information on ecological characters of the species. Table 3 shows the division of exotic weeds based on native regions and introduction period.

Table 3. Number of exotic weed species by native regions and introduction period.

Division	As.*	Eu.	Eu-As	nA	sA	Others	Total
Before 1960	19	45	17	14	10	10	115
1961~2001	10	77	18	58	21	16	200
Total	29	122	35	72	31	23	315

* As.; Asia, Af.; Africa, Aus.; Australia, Eu.; Europe, nA.; north America, sA.; south America, tA.; tropical America, Chi.; China, Ind.; india, cAs.; central Asia, sEu.; south Europe, Tro.; Tropical, wAs.; west Asia, Mex.; Mexico, Jap.; Japan)

Weed species native to Europe consisted for 122 species forming 39% of the total followed by South America (31 species and 10%), Asia (29 species and 9%) and Eurasia (35 species and 11%) and Australia and Africa (9 species). Considering adjacency and native region of exotic weeds, weeds native to Asia, Europe and Eurasia for 56% and 176 weed species followed by North and South America (33% and 103 species). The results implied that influx of exotic weeds have much connection to these native regions. If comparing native regions of exotic weeds before and after 1960 (Table 3), weeds native to Asia were identified as 19 and 10 species before and after 1960, respectively, followed by North and South America and Europe with 24 and 79 and 45 and 77 species during corresponding periods. Though the native region of weeds is not always the nation from which exotic weeds were introduced, kinds of exotic weed surveyed were greatly influenced by the change of nation for agricultural trade. Since

1960, exotic weeds were judged to be introduced through imported agricultural products considering increased trade of agricultural products from North and South America and Europe.

Distribution and habitation situation of exotic weeds

Results of the survey carried out near the habours in terms of distribution and species of domestic exotic weeds are shown in Table 4.

Table 4. Number of exotic weed species distributed near different ports.

Inchun	Pusan	Ulsan	Kunsan	Mokpo	Yeosu
38	32	35	27	23	23

Near Incheon harbor forming over 60% of total domestic import grains 38 exotic weed species were found followed by Ulsan and Pusan harbors (35 and 32 species respectively). The main reason for many exotic weeds to be found near harbor was the close relation with diffusion of weed seeds when imported grains were loaded and unloaded. In the future, as import of agricultural products is continuously increased, new exotic weeds are predicted to be found more and more around harbors. Imported grain is unloaded to the harbor, stored in storage house and then transported to a processing plant. Accordingly, the region of first occurrence of a new exotic weed is apt determined depending on transferring roads and destination of imported grains. Many new exotic weeds are found along the roads towards industries and express highways (Table 5).

Table 5. Number of exotic weed species by major distribution site.

Near by ports	Roadside highway	Pasture	Reclaimed land with wastes
54	98	56	31

Much of the grain dropped by roadside is often observed on the way to transfer of imported grains. Weed seeds are dropped together with falling grains and new exotic weeds come into existence and spread rapidly. In addition, much exotic weeds are found around cultivated areas of forage crops and pasture of dairy farms. 31 weed species were found in reclaimed and wasteland near Seoul. Main habitation area of exotic weed is greatly related to the imported grain. For example, forage grain used for feed in dairy farms entirely depends on imported one. Accordingly, weed seeds mixed in imported forage grain were a main reason for the increased occurrence of new exotic weeds around dairy farms. Reclaimed and waste land near Seoul also becomes the main occurrence region of exotic weeds because waste after selecting carefully imported forage grain and sweepings of cleaning roadside were put in to reclaimed land. In the future, it is necessary to continuously survey the regions in which new exotic weeds are likely to occur. If new weeds are discovered, management counter measures to prevent a spread of these noxious weeds should be prepared rapidly.

Current aspects of exotic weeds occurring in domestic cultivated area

There were 72 species occurring in cultivated areas among total exotic weeds. Classification of weeds found by crops is shown in Table 6.

Table 6. Exotic weed species in farmland in Korea.

Class		Weed species
Paddy filed (8)		*Diplachne fusca, Alopecurus japonicus, Panicum dichotomiflorum, Paspalum distichum, Paspalum distichum* var *indutum, Lindernia attenuata, Lindernia dubia* var. *anagallidea, Bidens frondosa*
Upland	Corn (31)	*Panicum dichotomiflorum, Lolium perenne* ssp. *multiflorum, Dactylis glomerata, Polygonum convolvulus, Polygonum orientale, Rumex crispus, Rumex obtusifolius, Chenopodium album* var. *album, Chenopodium ficifolium, Chenopodium glaucum, Amaranthus hybridus, Amaranthus blitum, Amaranthus viridis, Thlaspi arvense, Abutilon theophrasti, Malva neglecta, Ipomoea hederacea, Ipomoea purpurea, Datura stramonium, Solanum carolinense, Solanum americanun, Ambrosia artemisiifolia* var. *elatior, Ambrosia trifida, Bidens frondosa, Galinsoga parviflora, Galinsoga quadriradiata, Senecio vulgaris, Sonchus asper, Sonchus oleraceus, Xanthium strumarium* var. *canadense, Xanthium strumarium* var. *glabratum*
	Soybean (12)	*Panicum dichotomiflorum, Polygonum orientale, Chenopodium album* var. *album, Chenopodium ficifolium, Amaranthus hybridus, Amaranthus blitum, Abutilon theophrasti, Ambrosia artemisiifolia* var. *elatior, Bidens frondosa, Senecio vulgaris, Xanthium strumarium* var. *canadense, Xanthium strumarium* var. *glabratum*
Pasture land (59)		*Panicum dichotomiflorum, Bromus tectorum, Polygonum orientale, Rumex acetosella, Rumex crispus, Rumex obtusifolius, Rumex conglomeratus, Chenopodium album* var. *album, Chenopodium ficifolium, Chenopodium glaucum, Amaranthus hybridus, Amaranthus blitum, Amaranthus spinosus, Amaranthus viridis, Phytolacca americana, Cerastium glomeratum, Spergula arvense, Silene latifolia* ssp.*alba, Ranunculus muricatus, Barbarea vulgaris, Thlaspi arvense, Descurainia pinnata, Descurainia sophia, Trifolium pratense, Trifolium repens, Abutilon theophrasti, Malva parviflora, Malva neglecta, Lamium purpureum, Datura stramonium, Solanum carolinense, Solanum americanun, Veronica didyma* var. *lilacina, Veronica persica, Ambrosia artemisiifolia* var. *elatior, Anthemis arvensis, Anthemis cotula, Aster pilosus, Aster subulatus, Aster subulatus* var. *ligulatus, Carduus crispus, Conyza bonariensis* var. *leiotheca, Conyza canadensis* var. *canadensis, Erigeron annuus, Erigeron strigosus, Crassocephalum crepidioides, Erechtites hieracifolia, Senecio vulgaris, Galinsoga parviflora, Galinsoga quadriradiata, Sonchus asper, Sonchus oleraceus, Tagetes minuta, Taraxacum officinale, Hypochaeris radicata, Taraxacum laevigatum, Xanthium strumarium, Xanthium strumarium* var. *canadense, Xanthium strumarium* var. *glabratum*

Number of weeds occurring in cultivated areas was found to be 8 species including *Diplachne fusca* in paddy field, 12 species including *Amaranthus hybridus* in soybean upland field, 31 species containing *Abutilon theophrasti* in forage corn upland paddy field and 59 species such as *Rumex acetosella*, *Rumex obtusifolius* and *Solanum carolinense* in pasture. These weeds, which occurred in cultivated areas were not always noxious to crops. Kinds of exotic weeds greatly troublesome to crops are shown in Table 7. These weeds have rapid growth characteristics and cause great damage to crops. Because some species also have offensive odor and livestock was reluctant to eat them, occurrence of these species became the main reason for the declining quality of feed for livestock.

Table 7. Troublesome exotic weed species in cultivated fields in Korea.

Crops	Weed species
Soybean (4)	*Amaranthus hybridus*, *Chenopodium album* var. *album*, *Polygonum orientale*, *Bidens frondosa*
Corn (10)	*Abutilon theophrasti*, *Amaranthus hybridus*, *Rumex obtusifolius*, *Polygonum orientale*, *Chenopodium album* var. *album*, *Datura stramonium*, *Xanthium strumarium* var. *canadense*, *Xanthium strumarium* var. *glabratum*, *Solanum carolinense*, *Ambrosia trifida*
Pasture land (19)	*Rumex acetosella*, *Rumex obtusifolius*, *Solanum carolinense*, *Malva neglecta*, *Amaranthus spinosus*, *Lamium purpureum*, *Datura stramonium*, *Xanthium strumarium* var. *canadense*, *Xanthium strumarium* var. *glabratum*, *Abutilon theophrasti*, *Anthemis arvensis*, *Veronica persica*, *Carduus crispus*, *Erechtites hieracifolia*, *Crassocephalum crepidioides*, *Hypochaeris radicata*, *Sonchus asper*, *Sonchus oleraceus*, *Tagetes minuta*
Paddy filed (2)	*Diplachne fusca*(reclaimed paddy), *Bidens frondosa*

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PRE-BURIED APPLICATION OF DIQUAT-PARAQUAT MIXTURE TO WEED SEEDS: ITS' IMPACT ON SEED BANK REGULATION IN PADDY FIELDS

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Abstract: The inhibition of seed germination and emergence of seventeen weed species and four rice cultivars by the herbicide PreegloxL® (Syngenta Japan K.K; DP) containing diquat (diquat dibromide, 7%) and paraquat (paraquat dichloride, 5%) was investigated. The seeds were soaked in 100-fold diluted diquat-paraquat mixture (DP) for 60 min and buried into paddy soil put in Petri dishes. This treatment with DP caused significant death of the seedlings after the emergence on the weeds which had specific morphological features such as rough surface (*Bidens frondosa* and *B. tripartite*) and/or envelop structure (*Echinochloa oryzicola* and *Oryza sativa*). On the other hand, the seeds with smooth surface without envelop structure (*Scirpus juncoides*, *Lindernia procumbens, etc.*) were not affected by DP. In the second experiment, seeds of four weed species (*Scirpus juncoides, E. oryzicola*, *B. frondosa* and *B. tripartite*) and two rice cultivars (Manamusume and Yumeaoba) were treated with 100-fold diluted DP and then buried into the soil in paddy field after paddling and transplanting of rice. On 107th days after burying the seeds, the test plants were collected from the soil. The DP treatment strongly suppressed the emergence of *E. oryzicola* and *Oryza sativa*, but there was no effect on *S. juncoides*. Irrespective of the treatment, *B. frondosa* and *B. tripartite* did not germinate at all in the paddy field condition although it normally germinated in Petri dishes. These results suggest that application of DP to weed seeds inhibits the emergence by the adhesion of the chemicals on the seed surface or the infiltration into envelop structure of the seeds, although such inhibitory effect is significantly reduced by soil contact in some weed species. However, it is also supposed that pre-buried DP application to seeds should be useful for seed bank regulation in paddy field for at least two species of Gramineae (*E. oryzicola* and *O. sativa*).

Key words: diquat-paraquat, pre-buried application, seed bank

Introduction

A number of attempts have currently been made to prevent the growth of weedy rice by application of diquat-paraquat (PreegloxL®, Syngenta Japan K.K; DP) to fallen seed before field tilling. Such attempts are based on the findings that an application of the reagent to rice seed strongly suppresses the emergence of the seedlings even when their seeds were buried in the ground. Recently, the effect of diquat-paraquat (DP) on several weeds was investigated in order to regulate net amount of emergence from seed bank (Asai *et al.* 2005; Uchino *et al.* 2006). In the previous study, we have demonstrated an evidence of the inhibition of the growth of *Scirpus juncoides* by DP treatment just after germination irrespective of existence of light. Further, it was suggested that the lethal effect could be largely reduced by soil contact (Okawa *et al.* 2006). For practical regulation of weed seed bank by pre-buried DP application, we had to clarify the effect of soil on the efficacy of DP to the seeds and define applicable weed species. So in this study, we characterized the DP effect on several weed species and rice cultivars using soil medium put in Petri dish (Experiment 1). The effect of pre-buried DP treatment on the possible weed species and rice cultivars was investigated in the open paddy field (Experiment 2).

Materials and Methods

Experiment 1

Plant material: Weed species and rice cultivars used in the experiments are given in Table 1. The seeds of weeds were collected from the paddy filed of Miyagi Prefectural Furukawa Agricultural Experiment Station (MFAES) from September to October in 2005. Seeds of edible rice cultivars were harvested at MFAES in 2005, and seeds of feeding rice were obtained from Japan Grassland Agriculture and Forage Seed Association (GAFSA). Weed seeds were awaked dormancy in water at 5°C for more than three months, and rice seeds were soaked for 10 days at 5°C before the treatments.

Table 1. Weed species and rice cultivars used in the experiments

Family	Species ; Use or Common name
Gramineae	*Echinochloa oryzicola* Vasing. ; Early watergrass
	Oryza sativa L. cv. Hitomebore and Manamusume ; Edible
	Oryza sativa L. cv. Hoshiaoba, Kusayutaka and Yumeaoba ; Feeding
Cyperaceae	*Scirpus juncoides* Roxb. var. ohwianus T. Koyama
	Cyperus brevifolius (Rottb.) Hassk. var. *leiolepis* (Franch. et Savat.) T. Koyama
	Cyperus difformis L. ; smallflower umbrella sedge
	Eleocharis congesta D. Don
Alismataceae	*Alisma canaliculatum* A. Br. et Bouche
	Sagittaria trifolia L. ; Arrowhead
Pontederiaceae	*Monochoria vaginalis* (Burm. f.) Presl var. *plantaginea* (Roxb.) Solms-Laub.
Scrophulariaceae	*Lindernia procumbens* (Krock.) Borbas ; Common falsepimpernel
Lythraceae	*Rotala indica* (Willd.) Koehne var. *uliginosa* (Miq.) Koehne ; Indian toothcup
Compositae	*Centipeda minima* (L.) A.Braun. et Aschers. ; Spreeding sneezeweed
	Bidens frondosa L. ; Devils beggarticks
	Bidens tripartita L. ; Bur beggarticks
Polygonaceae	*Persicaria lapathifolia* (L.) S. F. Gray
	Persicaria hydropiper (L.) Spach ; Marshpepper smartweed
Chenopodiaceae	*Ludwigia epilobioides* Maxim.
Leguminosae	*Aeschynomene indica* L. ; Indian jointvetch

DP treatment: The seeds were soaked in 100-fold diluted diquat-paraquat (PreegloxL®, Syngenta Japan K.K; DP) for 60 min, and washed with distilled water for 60 min to remove the excess reagent. Those seeds were semidried on a filer paper within 12 hrs. As a control treatment, seeds were soaked in distilled water instead of the reagent.

Culture condition: DP treated seeds were sowed on wet filter paper or into paddy field soil sheeted in Petri dish at 1 mm depth. The medium soil was taken from paddy field in MFAES and filtrated into 2 mm mesh. Forty to fifty seeds were lined in a Petri dish with three replicates. The Petri dishes were placed in growth chamber maintained at 30°C with a continuous illumination.

Observation: Seed germination rate on wet filer medium, seedling emergence rate on soil medium and seedling death rate on each medium were observed at 21 days after sowing.

Experiment 2

Plant material: Four weed species and two rice cultivars were prepared as in Experiment 1.

DP treatment: Total of fifty seeds were put in a plastic net, soaked in 100-fold diluted DP for 60 min, then in tap water for 60 min to remove the excess reagent. For the control treatment, the seeds placed in the nets were soaked in tap water. Treated seeds were kept wet and preserved for 18 hrs at 5°C.

Burying seeds into field: On the 13th of May, 2006, the nets containing DP treated seeds were submerged into paddled soil of open field in MFAES at 1 to 2 cm depth from soil surface between lines of rice seedling transplanted in two days before the burial.

Observation and TTC test: At 107 days after burying seeds, the test plants were dug out from the soil and washed with tap water. The rate of emergence and the percentage of the seeds which germinated but did not emerged, were assessed. Non-germinated seeds in the ground were sowed on wet filter in Petri dishes and incubated in a growth chamber maintained at 30 °C with a continuous illumination for other 14 days to determine additional germination rate. Furthermore, viability of the seeds which even that did not germinate at after the additional culture was examined by staining their vertical section with 0.5 % (w/v) of TTC (2, 3, 5-triphenyl tetrazolium chloride) for 18 hrs (TTC test).

Results and Discussion

Effect of DP treatment on the seed germination, emergence and subsequent death

Figure 1 shows the rates of seed germination on filter paper and seedling emergence from soil medium at 21 days after sowing. Seed germination rates varied with the weeds and it might be depended on the suitability of the test condition to the weeds. However, there were significant differences in germination rates between DP treatment and the control on three species. On the other hand, emergence rate of six species on paddy soil was significantly suppressed by DP treatment especially on *E. oryzicola*.

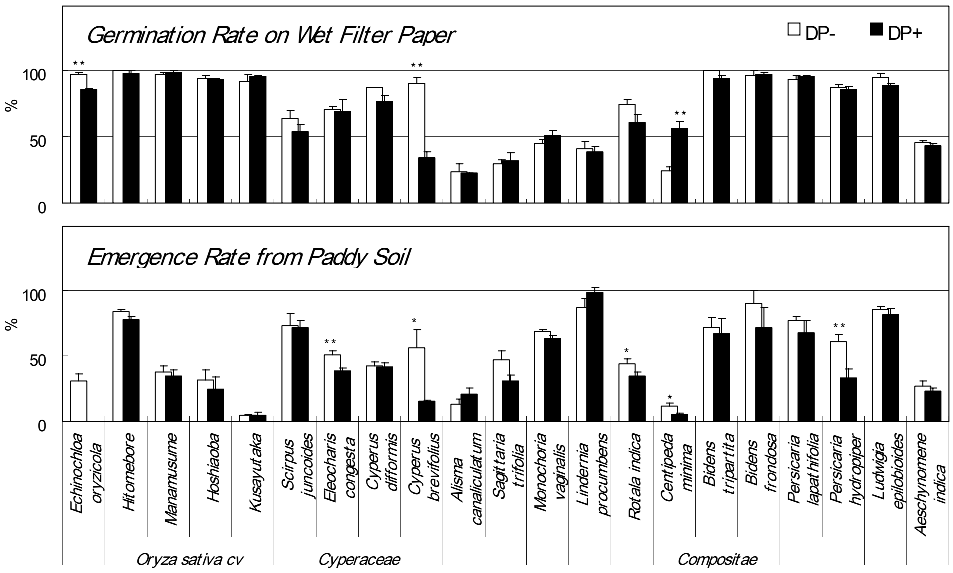

Figure 1. Effect of DP treatment on the seed germination on filter paper medium and seedling emergence from soil medium at 21 days after sowing. Values are the mean of three replicates and bars indicate ± s.e. *and ** indicate statically significant differences between the treatments at p=0.10 and p=0.05, respectively.

Figure 2 shows the rates of dead seedlings to the number of germinated or emerged seedling. Most species except for three (*Sagittaria trifolia*, *Alisma canaliculatum* and *Lindernia procumbens*) were significantly or absolutely led to seedling death after germinating by DP treatment on the filter medium. Otherwise in soil medium, all the seedlings of *Oryza sativa* cultivars and approximately 50% seedlings of *B. tripartita*, *B. frondosa* and *Persicaria lapathifolia* were led to death by DP after the emergence.

Table 2 shows relationship between the effect of DP treatment and morphological features of the seed of weeds and rice cultivars tested in the Experiment 1. Influence of pre-sowing DP treatment on germination on filter medium and emergence and subsequent death of seedling in soil medium was clearly observed on the weeds which seeds had specific morphological features such as rough surface (*B. frondosa*, *B. tripartite*) and/or an envelop structure (*e.g.* glume: *O. sativa* and *E. oryzicola*, scale: *C. brevifolius var. leiolepis*, pericarp: *P. hydropiper*). On the other hand, the seeds with smooth surface without envelop structure (*S. juncoides*, *L. procumbens*) were not affected by DP treatment in the soil condition. These results suggest that application of DP to weed seeds inhibits the seedling emergence by the adhesion of chemicals on seed surface and/or the infiltration into envelop structure, although such inhibitory effect is significantly reduced by soil contact in some species.

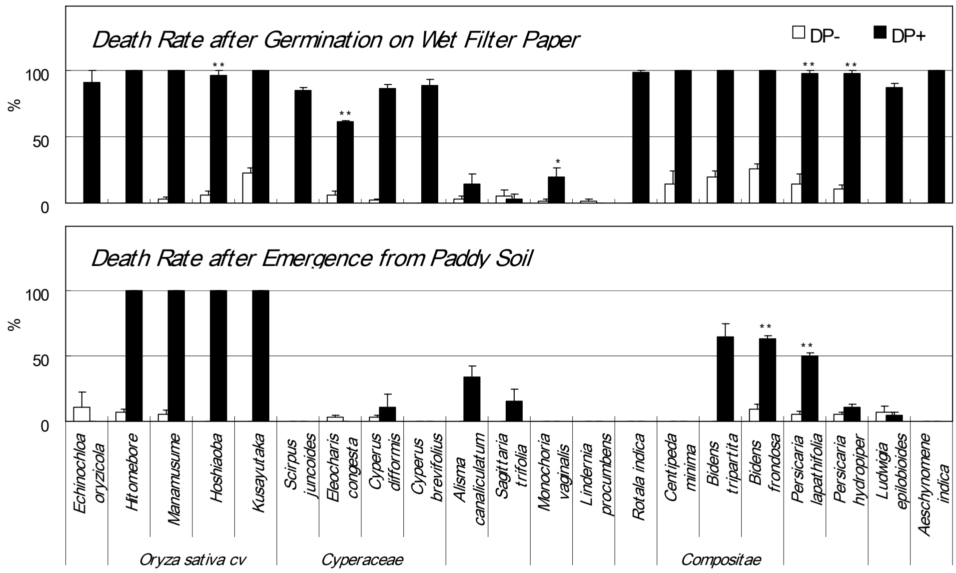

Figure 2. Effect of DP treatment on the seedlings after germination on filter paper or after emergence from soil medium at 21 days after sowing. Values are the mean of three replicates and bars indicate ± s.e. *and ** indicate statically significant differences between the treatments at p=0.10 and p=0.05, respectively.

Effect of pre-buried DP treatment on weed seeds in paddy field

Figure 3 shows the rate of seed germination in soil and rate of seedling emergence from at 107 days after burying the seeds. *S. juncoides*, which was not affected by DP in soil medium in Experiment 1, was also not influenced by DP under the experimental field conditions. Rate of germination of *E. oryzicola* and two cultivars of *O. sativa* in the field tended to decrease in comparison with the control treatment. Irrespective of the treatments, *B. frondosa* and *B. tripartite* did not germinate at all. These results show that the pre-buried DP application would be applicable to two species of Gramineae (*E. oryzicola* and *O. sativa*) and it would not be effective to the species which were not influenced by DP in soil medium in the Experiment 1.

Table 2. Relationship between DP effects and morphological feature of seeds

Family - Species		Effects of DP treatment: Filter Paper, Germinate Repression	Filter Paper, Seedling Death	Paddy Soil, Emergence Repression	Paddy Soil, Seedling Death	Morphological Feature of Seed: Envelope structure	Envelope Closeness	Roughness of Surface
Gramineae	*Echinochloa oryzicola*	±	++	++	·	+	±	+
Oryza sativa cv	Hitomebore	-	++	-	++	+	+	+
	Manamusume	-	++	-	++	+	+	+
	Hoshiaoba	-	++	-	++	+	+	+
	Kusayutaka	-	++	-	++	+	+	+
Cyperaceae	*Scirpus juncoides*	-	++	-	-	-	·	-
	Eleocharis congesta	-	++	±	-	-	·	-
	Cyperus difformis	-	++	-	-	-	·	-
	Cyperus brevifolius	+	++	±	-	+	±	-
Alismataceae	*Alisma canaliculatum*	-	-	-	±	+	+	-
	Sagittaria trifolia	-	-	-	±	+	+	-
Pontederiaceae	*Monochoria vaginalis*	-	±	-	-	-	·	-
Scrophulariaceae	*Lindernia procumbens*	-	-	-	-	-	·	-
Lythraceae	*Rotala indica*	-	++	±	-	-	·	-
Compositae	*Centipeda minima*	-	++	±	-	+	+	+
	Bidens tripartita	-	++	-	+	+	+	+
	Bidens frondosa	-	++	-	+	+	+	+
Polygonaceae	*Persicaria lapathifolia*	-	++	-	+	+	±	+
	Persicaria hydropiper	-	++	+	-	+	+	+
Chenopodiaceae	*Ludwigia epilobioides*	-	++	-	-	+	-	+
Leguminosae	*Aeschynomene indica*	-	++	-	-	+	-	+

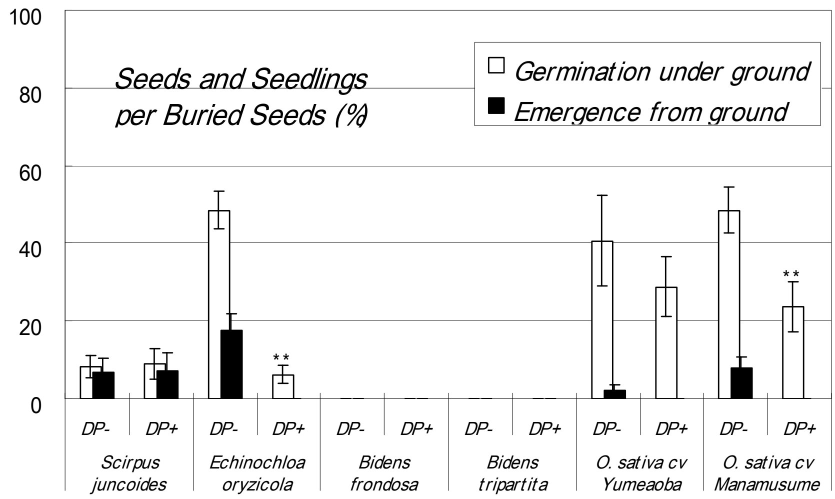

Figure 3. Effect of pre-buried DP treatment to seeds on the germination and seedling emergence in paddy field at 107 days after burying the seeds. Values are the mean of four replicates and bars indicate ± s.e. *and ** indicate statically significant differences between the treatments at p=0.10 and p=0.05, respectively.

Figure 4 shows the proportion of germinated seeds in the field, germinated seeds on Petri dish, viable and nonviable seeds among non-germinated ones based on TTC-test, and albumen-perished empty seeds, which were thought to die in the filed after burial. Just after digging and washing out, *B. frondosa* and *B. tripartite* immediately started germinating showing no symptoms such as whitening and abortion as a result of DP treatment in the culture on filter medium in Experiment 1. It is suggesting that competitive absorption of active ingredient from seed surface to soil particles happened, then absence of DP on/within seed occurred during longer term contact with soil. Large number of empty seeds of *E. oryzicola* and non-germinated TTC-negative seeds of two species from family Gramineae and *S. juncoides* were seen in pre-buried DP treatment. This result suggests that embryo activity is depressed within seed by DP infiltration under submerged conditions or during the post-digging culture.

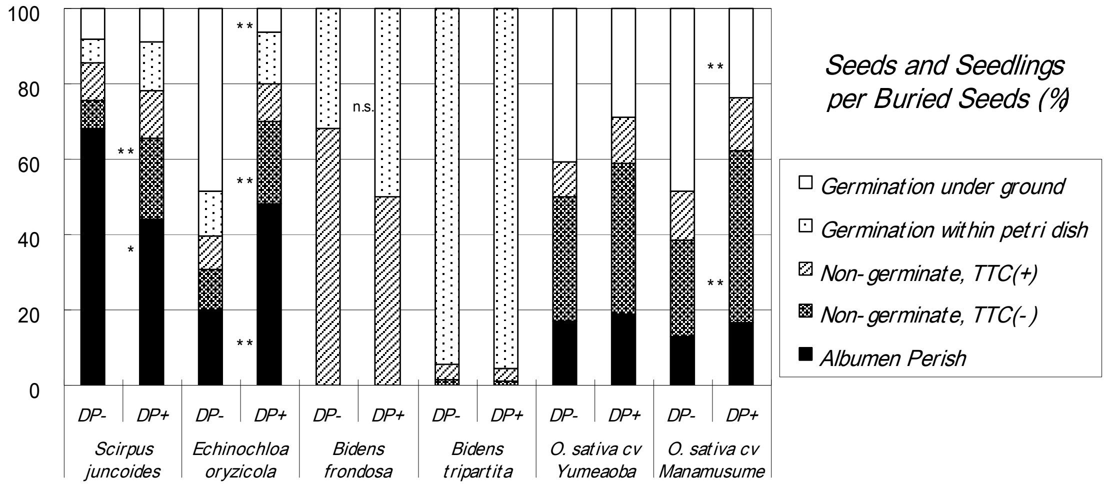

Figure 4. Effect of pre-buried DP treatment to seeds on the viability of the seeds in the paddy field and on the additional culture. Values are the mean of four replicates and bars indicate ± s.e. *and ** indicate statically significant differences between the treatments at p=0.10 and p=0.05, respectively. n.s. indicates that there is not a statically significant difference at p=0.10.

In conclusion, a pre-buried application of DP to weed seeds inhibits their seedling emergence due to suppression of germination and growth after germination as a result of the adhesion of the active ingredients to seed surface and the infiltration into seed envelope. However, such inhibitory effects were significantly reduced by soil contact in some species. Furthermore, it pre-buried DP application should be useful for seed bank regulation in paddy fields for at least two species of family Gramineae *i.e. E. oryzicola* and *O. sativa*.

Acknowledgements

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COMPETITION OF PURPLE AMMANIA (*Ammannia coccinea* Rottb.) WITH RICE IN PADDY FIELDS

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Abstract: To estimate the critical period of competition between the *A. coccinea* and rice, *A. coccinea* was weeded by every two weeks until 14 weeks after emergence (WAE) in one set. In the other set, respective plots were weeded out and *A. coccinea* was left to grow and compete with rice after controlling by 2-week intervals until 14 WAE. Gompertz and logistic models were used to analyze the non-linear change of yield affected by different treatments and critical period of weed competition by *A. coccinea* was analyzed. From 2 WAE to 4 WAE, number of *A. coccinea* increased from 126 to 640 plant m^{-2}. Although it kept increasing until harvest, it appeared that the group emerged in the first 4 weeks provided most competition with rice. Until 6 WAE, dry weight of *A. coccinea* was 76.5 g m^{-2} and it increased more than 3 times to 241.3 g m^{-2} in the two following weeks. The yield reductions by *A. coccinea* increased with longer competition periods. In comparison with the control, yield of rice was to 4,127 kg ha^{-1} (a 21% decrease) after the 14-week competition period. Response to the sharply increasing of biomass of *A. coccinea* 6 WAE, rice yield decreased significantly to a low level from 8 weeks of competition. As weed-free maintenance period of *A. coccinea* increased, rice yields increased. *A. coccinea*, which emerged 8 WAE had less competition on the rice yield. Rice yields were constant after 8 weeks of weed-free periods comparing with those with shorter weed-free periods.

Key words: Competition, *Ammannia coccinea,* rice

Introduction

In Korea, *A. coccinea* was first found in Changwon and Yeonggwang in 1981 (NIER, 2001; RDA, 2005), much later than the first appearance of *A. multiflora* reported in 1966. Recently, *A. coccinea* was also found in a paddy field in Daejeon, Korea in 2002 (Shen *et al*. 2003).

Some efforts have been made to investigate eco-physiological characteristics of Ammannia species in terms of their germination and seedling emergence. Nakayama and Takabayashi (1987) reported high germination of air-dried seed of *A. coccinea* around 94.7% and no germination of *A. coccinea* and *A. multiflora* under dark condition, indicating that they are photoblastic. Germination of *A. multiflora* peaked at 32/28°C and required a prolonged period for its germination at 21/17 °C (day/night). For the first seedling emergence of *A. multiflora*, about 10 and 3 days were required at 21/17 and 32/28°C (day/night), respectively, under flooded conditions (Chiang and Chiang, 2004). However, most of studies have focused on *A. multiflora* and not many studies have been done to investigate eco-physiological characteristics of *A. coccinea* as a paddy rice weed. In weed management, it is important to understand early establishment of weeds, which will provide us practical information for weed control. Little information is available, particularly about competition of *A. coccinea* with rice. Therefore, this study was conducted to understand competition aspects of *A. coccinea* with rice in paddy fields.

Materials and Methods

Effect of weed competition period of A. coccinea on rice yield
Experiments were conducted during 2004. Soil was a sandy loam with a pH of 6.3 and 1.2% organic matter. In order to evaluate the onset of the critical period of *A. coccinea* removal,

plots were left weedy and weed-free for 4, 6, 8, 10, 12 and 14 weeks, and all season by periodic hand-hoeing. The 14 treatments were repeated two times in a randomized complete block. Fertilizer was applied 110, 45 and 57 kg ha^{-1} of N, P and K, respectively. Each plot was 2 × 3 m^2. Rice was transplanted on May 17, 2004 after 30 days in the nursery, at 30 × 15 cm^2 spacing. *A. coccinea* was sampled each week in 30 × 30 cm^2, weighed after oven-drying at 70°C for 3 days.

Effect of weed-free period of A. coccinea on rice yield

Seeds of *A. coccinea* were sown carefully after harrowing the paddy field. Rice was transplanted on May 17, 2004 after 30 days in the nursery at 30 × 15 cm^2 spacing. Plots were left weedy and weed-free for 4, 6, 8, 10, 12, 14 and 16 weeks, and all season by periodic hand-hoeing. The 14 treatments were repeated two times in a randomized complete block design. Each plot was 2 × 3 m^2. Fertilizer was applied 110, 45 and 57 kg ha^{-1} of N, P and K, respectively. The plant height of *A. coccinea*, rice and biomass of shoot of *A. coccinea* were recorded simultaneously every two weeks. Yield of rice was investigated at harvest. In all experiments, data were analyzed by ANOVA followed by Fisher's protected LSD test to identify homogenous groups within the means.

Critical period of weed competition in rice paddy field

Based on the former experiments carried out in 2004, plant height, number of plants and fresh weight of *A. coccinea* were collected every week after emergence of *A. coccinea*. Plant height and the number of tillers of rice was investigated at the same time. Rice yield of each plot was measured after harvest. Gampertz model was used to analyze the linear change of yield affected by different treatments and the critical period of weed competition by *A. coccinea* was determined.

All measurements were initially subjected to analysis of variance (ANOVA). Non-linear regression was used to fit the Gompertz model (Cussans *et al*. 1996) and the logistic model. All statistical analyses were carried out using Genstat (Genstat Committee, 1997)

Result and Discussion

Effects of weed competition period of *A. coccinea* on rice yield

The first emergence of *A. coccinea* was observed on May 28, 2004. Generally, the yield reduction increased with increasing competition periods by *A. coccinea* (Table 1). Panicle length of rice was significantly shortened after 12 weeks of competition from *A. coccinea*.

Table 1. Plant height and yield components of rice as affected by different periods of competition with *A. coccinea*.

Weed competition period (week)	Plant height (cm)	Panicle length (cm)	No. of panicles $hill^{-1}$	No. of spikelets $panicle^{-1}$	Weight of 1,000-seed (g)	Grain weight/plant (g)
0	101.3	18.2	16.3	304	24.5	34.1
2	103.2	18.6	16.5	296	24.8	33.5
4	101.1	17.0	16.2	291	25.1	32.4
6	102.8	17.2	15.6	268	23.1	34.2
8	103.1	16.9	15.2	274	24.6	31.2
10	101.4	17.1	14.2	261	23.5	32.5
12	97.8	15.8	15.6	253	24.3	30.2
14	101.9	15.2	15.8	258	23.1	31.3

Number of panicles per hill decreased sharply from 8 weeks after emergence (WAE) of *A. coccinea*. Differences between plant height, panicle length, and 1,000-grain weight were relatively smaller than those between number of panicle per hills and spikelets per panicle. Yield of rice was 4,127 kg ha^{-1} (21% decreased in comparison to the control) after a 14-week competition period (Table 1). Response to the sharply increasing of biomass of *A. coccinea* 6 WAE, rice yield decreased significantly to a low level from 8 weeks of competition.

Effects of weed-free period of A. coccinea on rice yield

In the weed-free maintenance period experiment, plant height of *A. coccinea* was 94.6 cm at harvest. A total of 1,315 plants of *A. coccinea* were observed throughout the growth season (Table 2). Number of *A. coccinea* decreased to 1,001 and 863 at weeding 2 and 4 WAE. Number of plants of *A. coccinea* was as low as 568 when weeding 6 WAE. Dry weight of *A. coccinea* after removal until harvest significantly decreased after 8 WAE.

Table 2. Growth of *A. coccinea* and plant height and yield of rice as affected by different weed-free maintenance periods of *A. coccinea*.

Weed free maintenance period (week)	*A. coccinea**			Rice		
	Plant height (cm)	Plant number (No.m^{-2})	Dry weight (g m^{-2})	Plant height (cm)	Milled rice yield (kg ha^{-1})	Yield index (%)
0	85.6	1,315.6	412.6	101.3	4,550	83
2	94.6	1,001.2	396.5	101.9	4,915	90
4	84.6	863.2	313.6	102.8	4,935	90
6	56.2	568.6	256.3	103.1	5,265	96
8	32.3	363.5	89.6	101.4	5,320	97
10	15.5	217.5	40.0	97.8	5,175	95
12	7.4	165.3	32.9	97.8	5,198	95
14	2.3	53.6	21.1	101.9	5,467	100

***Weed samples were collected just before rice harvest in 2004.**

As weed-free maintenance period of *A. coccinea* increased, rice yields increased (Table 2). *Ammania coccinea*, which emerged 8 WAE had less competition on the rice yield. Rice yields were constant after 8 weeks of weed-free period as compared to those with shorter weed-free periods. In comparison with 0 weeks of weed-free period, 14 weeks of weed-free maintenance increased rice yield by 21%, from 4,550 kg ha^{-1} to 5,467 kg ha^{-1} (Table 2). Following the sharp decrease in the biomass of *A. coccinea* with increasing weed-free period, rice yield increased significantly.

Critical period of weed competition of A. coccinea for rice paddy field

To determine the critical period of *A. coccinea* competition with rice, non-linear regression analysis was conducted by fitting Gompertz and logistic models to rice yields resulting from competition with *A. coccinea* for different periods of time.

Gompertz and logistic models adequately described the relationship between competition periods of *A. coccinea* and rice yield. They estimated the parameter C (rice yield for 0 week competition with *A. coccinea*) to be 5,228 kg and 5,251 kg ha^{-1} of rice grain yield, respectively (Table 3). The parameter M values estimated by Gompertz and logistic models were 22.05 and 23.61 weeks, respectively, much longer than the whole period of rice cultivation, indicating that the competition effect of *A. coccinea* on rice was not so high.

Considering the size of residual mean square of regression analysis, both models were not significantly different in describing the relationship.

Table 3. Summary of parameters estimated for the relationship between the competition periods and the weed- free maintenance periods of *A. coccinea* and rice yields.

	Model	Parameters estimates				
		C**	B	M	RMS*	R^2
Competition period	Gompertz	5,251 (376)	-0.150 (0.110)	23.61 (5.89)	933.0	0.530
	Logistic	5,228 (339)	-0.166 (0.113)	22.05 (4.50)	934.3	0.606
Weed-free maintenance period	Gompertz	5,412 (217)	0.185 (0.123)	-9.35 (5.86)	613.2	0.570
	Logistic	5,398 (198)	0.201 (0.127)	-8.19 (4.90)	613.1	0.570

*RMS is residual mean square of regression analysis. **C: Estimated maximum rice yield. B: Parameter indicating the slope of curve. M: Competition period and weed- free period required to inhibit 50% of the estimated maximum rice yield.

For the relationship between the weed-free maintenance periods of *A. coccinea* and rice yield, Gompertz and logistic models were also fitted to rice yield data. They described the relationship satisfactorily and estimated the parameter C (rice yield for 14 weeks of *A. coccinea* free condition) to be 5,412 kg and 5,398 kg ha^{-1} of rice grain, respectively (Table 3). Considering the size of the residual mean square of regression analysis, both models were not significantly different in describing the relationship.

Using the logistic model, rice grain yields were predicted for increasing periods of weed-free or competition with *A. coccinea* (Figure 1). To restrict yield loss by 5%, the model suggested a weed-free period of 7 weeks from the first emergence of *A. coccinea*, which is equivalent to 60 DAT. Hence, 4 weeks of weed competition periods from the first emergence of *A. coccinea*, equivalent to 39 DAT can be allowed. This suggestion implies that *A. coccinea* should be controlled from 24 June in Daejon (39 DAT) by any means and this control effect must be maintained until July 15 (60 DAT). Therefore, the period between 39 and 60 DAT is the critical period of *A. coccinea* competition with rice.

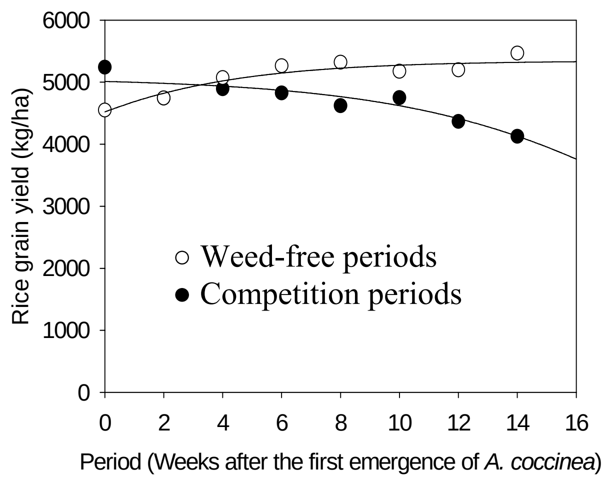

Figure 1. Rice grain yield as affected by competition from *A. coccinea*. The continuous line is the predicted rice yield calculated using the logistic model and the parameter estimates in Table 3.

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INFLUENCE OF SELECTED ENVIRONMENTAL FACTORS ON GERMINATION AND EMERGENCE OF *Hyptis suaveolens* OF COCONUT PLANTATIONS IN SRI LANKA

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Abstract: *Hyptis suaveolens* is a prominent and dominant weed species in the coconut plantations of Sri Lanka. Hence, laboratory and greenhouse studies were conducted to determine the effects of pH, osmotic stress, planting depth and submerged time period on *H. suaveolens* seed germination and emergence. Seed germination was significantly reduced in solutions with osmotic potentials below -0.6MPa. The highest germination was at 0 Mpa, and seedling emergence was not observed at -1.3MPa. The optimum pH range for germination of *H. suaveolens* seed was between 5 and 8, although some seeds germinated at pH levels of 4 and 9. The maximum *H. suaveolens* emergence occurred when seeds were planted on the soil surface. Seedlings did not emerge when seeds were planted at a depth of 7 cm. No seedlings emerged when seed was exposed to flooding for 13 days or more. The highest percentage (95%) of seeds emerged from a depth of 1cm when the soil was flooded for one day. These results suggest that *H. suaveolens* seed is capable of germinating and surviving in a range of climatic and edaphic conditions, and that flooding is not a viable management option for controlling this weed after emergence.

Key words: Coconut, weed seed, *Hyptis suaveolens*, germination

Introduction

The growth habit and canopy structure of the coconut palm requires a wide spacing between palms, which permits abundant sunlight to the ground vegetation. Thus, a wide range of perennial and annual weed species invade the unutilized space beneath the palms (Senarathne *et al.* 2003). Among these weed species (*Hyptis suaveolens*) is one of the most problematic dicotyledonous weed in coconut plantations in Sri Lanka. It is a major weed of the family *Lamiaceae* and is a perennial, broad leaved erect strongly aromatic herb (Stone, 1970). The weed grows on all types of well drained soils in areas that receive between 250 mm – 3000 mm of rainfall per annum (Stone, 1970). It resists drought very well and is tolerant to salt spray. Aerial portion of the plant are killed by temperature of -2°C, but quickly grows back (Wagner *et al.* 1999). Invasion of *H. suaveolens* has been a problem in areas such as distributed pastures, overgrazed areas, open forests, plantations and slash and burn agricultural fields. The shrub is reported to highly allelopathic to vegetation (Smith, 2002). It may grow as a single plant and it grows from near sea level up to 1300 m in elevation (Wanger *et al.* 1999). *Hyptis suaveolens* plant flowers and fruits throughout the year (Smith, 2002). Seeds are dispersed by animals, humans, field machinery and when attached to fur, clothing and mud (Smith, 2002).

Seed germination is the key event in determining the success of a weed in an agro ecosystem. Several environmental factors such as temperature, light, soil salinity, moisture and pH are known to affect weed seed germination (Chachalis and Reddy, 2000; Koger *et al.* 2004). Burial depth of seeds also affects germination and seedling emergence (Koger *et al.* 2004; Shaw *et al*. 1991) and the availability of moisture, diurnal temperature fluctuation, and light exposure varies with depth. All of these attributes of the microenvironment potentially influences the behavior of weed seeds. Therefore, it is critical to procure information on seed germination, persistence and seedling emergence to improve the management systems for specific weed species (Mennan and Ngouajio, 2006). There is little information on the seed biology of *H. suaveolens* which affects the development of strategies for the management of this weed. Therefore, the objective of this study was to determine the effect of osmotic stress, pH, flooding and burial depth on germination of this specie using laboratory and green house experiments.

Materials and Methods

Seeds of *Hyptis suaveolens* were collected from five different locations in the major coconut growing region of Sri Lanka between February to March 2005 and were stored at 5^{o}C under dark conditions. The selected treatments of the experiments were arranged in a Complete Randomized Design (CRD) with ten replicates (each Petri dish and pot representing one replication in each trial) in the respective studies.

Effect of moisture stress
Aqueous solutions of polyethylene glycol (PEG) (average molecular weight of 6000) were prepared to obtain osmotic potentials of 0, -0.3, -0.4, -0.6, -0.9 and -1.3 MPa by dissolving 0, 154, 191, 230, 297 or 350 g of PEG in 1 liter of deionized water (Michel and Kaufmann, 1973). Thereafter, 50 seeds of *H. suaveolens* were placed in individual 9 cm diameter Petri dishes containing two filter papers. The filter papers were moistened with 5 ml deionized water or the test solutions and the Petri dishes were placed in a green house. Seed germination was counted once in two days for a period of 30 days.

Effect of pH
Hyptis suaveolens seeds were placed in Buffer solutions having pH values of 4, 5, 6, 7, 8 and 9 with distilled water as a control. The buffer solutions were prepared as described by Reddy and Singh (1992), using Potassium hydrogen pthalate, and were adjusted to pH 4 with HCl. The 2(N-morpholino)ethanesulfonic acid solutions were adjusted to pH 5 and 6, N-(2 Hydroxymethyl) piperazine-N-(2-ethanesulfonic acid) solution was adjusted to pH 7 and 8 and N-Tris(hydroxymethyl)methyglycine (Tricine) solution was adjusted to pH 9 with NaOH. Fifty seeds were placed on two sheets of filter paper placed in 9 cm diameter Petri dishes and moistened with 5 ml of the respective pH solutions. Seed germination was counted once in two days for 30 days.

Effect of planting depth
Fifty *Hyptis suaveolens* seeds were planted in a sandy clay loam soil in polythene pots (15 cm diameter) at depths of 0, 1, 3, 5, 7, 9 and 11 cm, and placed in a green house, where the temperatures were 30°C ± 4°C during the day and 26°C ± 4°C during the night. Pots were watered as needed to maintain between available moisture 70% - 80% soil moisture. Geminated seedlings were counted once in 7 days for 30 days Seedlings were considered emerged when the cotyledons could be visually discerned and were removed after counting at 7 day intervals.

Effect of submerged time period (flooding)
Fifty *Hyptis suaveolens* seeds were planted 1 cm deep in a sandy clay loam soil in plastic pots (9 cm diameter). Flooding was imposed by maintaining water at a height of 1.5 cm above the soil surface. The water level was maintained for 1, 3, 5, 7, 9, 11, 13 and 15 days after planting. Flooding was discontinued after the indicated period and seeds were watered as needed to maintain adequate moisture. Emergence was recorded weekly for a period of 28 days after planting.

Statistical analysis
In all experiments except the pH test, percentage of germination data were transformed using the log (X + 1) transformation, where X is the percentage of germination, to improve homogeneity. The ANOVA and regression analysis were performed on both transformed and non transformed percentages of germination. Means from experiments were separated using, the Fishers protected LSD test at p=0.05.

Results and Discussion

Effect of moisture stress
The impact of osmotic potential on the germination of *H. suaveolens* seeds presented as a quadratic equation (Figure 1).

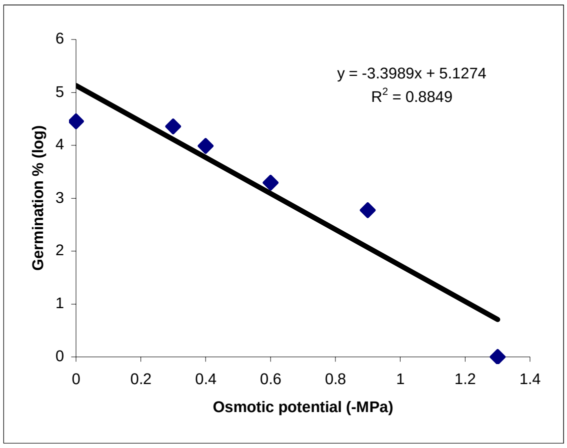

Figure 1. Effect of moisture stress (osmotic potential – Mpa) on germination of *Hyptis suaveolens* seeds.

Germination of *H. suaveolens* seeds decreased when the osmotic potential increased from 0 MPa to - 1.3 MPa indicating that these seeds can germinate under high water stress conditions. Seed germination was highest at osmotic potential of 0 and -0.3 MPa. Seeds (16%) germinated at an osmotic potential of -0.9 MPa. No germination occurred at an osmotic potential of -1.3 MPa. Germination over a broad range of osmotic potential indicates that *H. suaveolens* could pose a weed threat under both low and high moisture conditions and could grow in wet, intermediate and dry zone coconut lands of Sri Lanka. In general, germination of *H. suaveolens* seed is inhibited under high water stress conditions. Such conditions can occur temporarily between periods of rainfall at the start of the growing seasons in Sri Lanka. However, weed species such as Red Vine (*Brunnichia ovata* (Walt) Shinners) are highly sensitive to low water potential (Shaw *et al.* 1991). The rate of decline in *H. suaveolens* seed germination due to the different moisture stress conditions was 3.3989. The results of the study suggest that *H. suaveolens* is highly tolerant to conditions of water stress, and germination is likely to be favored by a moderately moist environment.

Effect of planting depth
Seedling emergency decreased with increased planting depth. No seedlings emerged from seeds placed at a depth of 7cm. The highest seed germination (85%) was at the soil surface (Figure 2). How ever, reduced seedling emergence of seeds placed on the soil surface has been reported previously for many weeds (Singh and Achhireddy, 1984; Balyan and Bhan, 1986; Shaw *et al.*1991; Horak and Sweat, 1994).

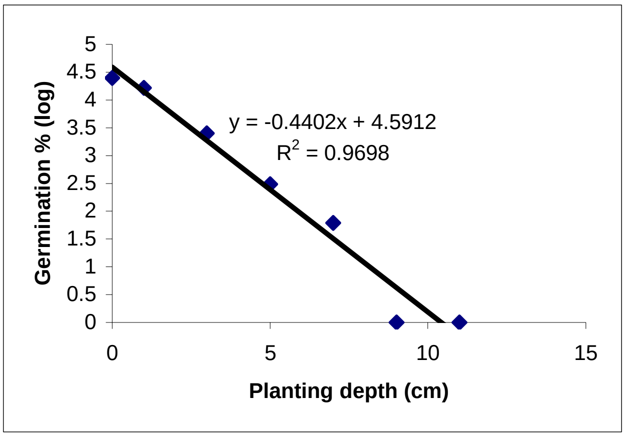

Figure 2. Effect of planting depth on germination of *Hyptis suaveolens* seeds

In many weed species, seedling emergence decreased with increased depth of seed burial (Cussans *et al*. 1996; Qi and Upadhyaya, 1993; Shaw *et al*. 1997). Emergence after burial in soil depends on seed size and light conditions. Large seeds with sufficient reserves can emerge from greater depths (Baskin and Baskin, 1998). *Hyptis suaveolens* seed is a relatively large seed and does not require light for germination and seedlings emerge well from moderate depths up to 4cm.

Depth mediated *H. suaveolens* emergence (expressed as a percentage of maximum emergences) is well described by a logistic model, with the greatest emergence occurring from the soil surface. This trend has been observed in other weed species, where greater emergence is often from the soil surface when conditions are favorable for germination (Benvenuti *et al*. 2001). The decreasing rate of seed germination due to the different planting depths was 0.4402.

Effect of pH

Hyptis suaveolens seeds germinated over a wide pH range, indicating that pH is not likely to be a limiting factor for germination in most soil types. Seed germination of *H. suaveolens* seeds followed a quadratic response to increasing pH with enhanced germination between pH 4 and pH 7 and decreasing germination at pH levels of 7 to 9 (Figure 3). The maximum germination in *H. suaveolens* was at pH 6, although seeds of *H. suaveolens* germinated at pH 4 and pH 9 which were acid and alkaline conditions.

The percentage of seeds of *H. suaveolens* that germinated at pH 4 and pH 9 were 5% and 34%, respectively. Therefore, the ability to germinate over a wide pH range supports the view that *H. suaveolens* weed is adapted to a wide range of soil conditions. This characteristic is common in many weed species such as *Asclepias syriaca* L. and *Scoparia dulcis* (Evetts and Burnside, 1972; Jain and Singh, 1989). Other weed species such as *Avena fatua* L may require a specific pH to break dormancy (Adkins *et al*. 1985). Germination under a variety of soil conditions aids the ability of a plant to invade diverse habitats.

Effect of submerged time period (flooding)

Emergence of seedlings of *Hyptis suaveolens* decreased with increasing duration of submergence. The highest percentage of seedlings (95%) emerged from a depth of 1 cm when not exposed to the flood conditions or duration of flooding was one day. Seedling emergence decreased gradually when duration of flooding was three days or longer (Figure 4).

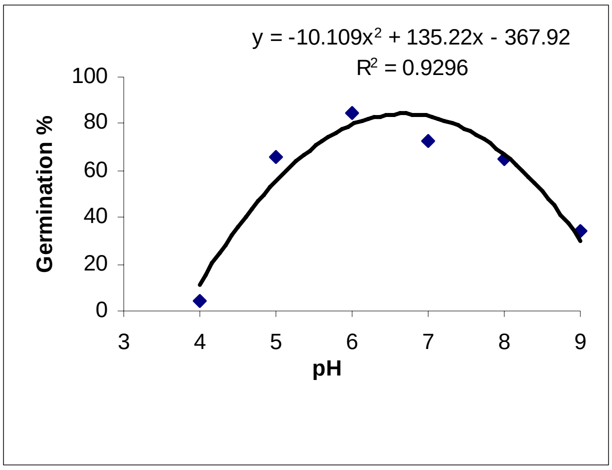

Figure 3. Effect of pH on germination of *Hyptis suaveolens* seed

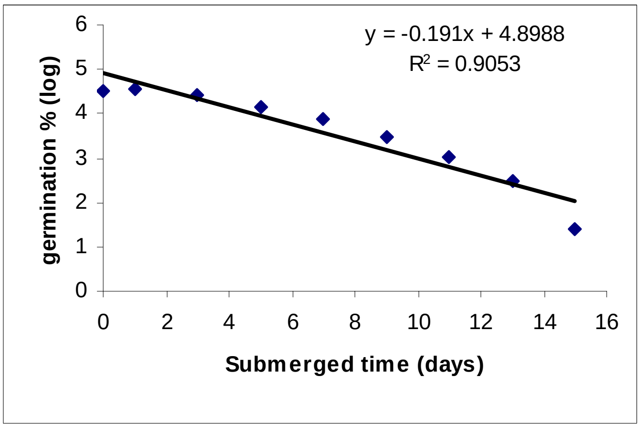

Figure 4. Germination of *Hyptis suaveolens* seeds as affected by submerged times

The rate of decline in *H. suaveolens* seed germination due to the different submergence time periods was 0.1910. However, seedling emergence (81%) was observed when flooding was maintained 15 days. Hence, germination may be favored in areas prone to even short period of flooding. Activation of the physiologically process necessary for seed germination requires an O_2 supply, however, soil inundation restricts O_2 availability to the embryo and thereby prevents or delays seed germination in many species (Kozlowski and Pallardy, 1997). In general, soaking seeds of upland species is recommended for several hours to few days to accelerate germination, whereas soaking for long periods inhibits this process (Kozlowski and Pallardy, 1997).

Conclusions

Hyptis suaveolens seed germination was observed over wide range of soil pH values. This suggests that soil acidity or alkalinity is not likely to be a limiting factor for seeds of this species to germinate in most coconut growing soil types in Sri Lanka. Seedling emergence was optimal at shallow burial depths, which indicate that agricultural practices such as no till and minimum tillage that promotes shallow burial of weed seeds may stimulate greater seedling emergence of *Hyptis suaveolens*. When seeds were buried in deep layers of the soil, seed germination percentage was reduced, suggesting that ploughing and harrowing can be used to suppress this species. Generally seed germination is severely restricted under both osmotic stress and flooded conditions. However, *Hyptis suaveolens* seeds could germinate at over a broad range of osmotic potentials indicating that this weed could pose a weed threat under both low and high moisture stress conditions. The overall results suggest that *Hyptis suaveolens* has the ability to germinate under a broad range of environmental conditions. Prevailing ecological conditions could determine the extent of germination, subsequent emergence, and severity of *H. suaveolens* infestations in coconut plantations. The warm climate and well and moderately drained soil in coconut lands coupled with abundant moisture (rainfall) and frequent fertilization are ideal for *Hyptis suaveolens s*eed germination and establishment. Therefore, it is fast becoming a prominent weed species in wet, intermediate and dry zone coconut plantations in Sri Lanka, and results from this study could become useful in developing possible control measures.

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CHARACTERIZATION OF RED RICE (*Oryza sativa* L.) BASED ON INDICA/JAPONICA AND WILD/CULTIVATED TYPIFICATION

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Abstract: Studies were carried out on 141 red rice accessions collected from 14 countries to characterize them based on indica/japonica and wild/cultivated typification with 6 morpho-physiological characteristics. The indica/japonica types of the common rice varieties were also surveyed in these countries to compare the genetic background between the red rice accessions and common rice varieties. The most of red rice accessions collected from north/south America and Italy were characterized as indica type though those from other European countries ware characterized as japonica. It is interesting that the most cultivated rice in United States were japonica type though the most red rice accessions collected from U.S. were characterized as indica type. The accessions from South/South East/East Asia had wide variation of characteristics between indica/japonica and wild/cultivated type. Weedy red rice accessions collected from Japan recently were characterized as japonica and had characteristics very close to those of wild type.

Key words: Weedy rice, ferality, biotype, alien weed, micro-evolution

Introduction

Red rice (*Oryza sativa* L.) is generic term used to refer to rice with red pericarp; however it often means weedy rice in certain countries. Some red rice occurs in paddy field as a noxious weed, and its grains contaminate the harvested rice varieties. The weedy red rice has become a serious problem in United States, Brazil, Korea, Japan and other rice production countries and it reduces the yield and market grade of cultivated rice (FAO, 1999, IRRI, 2000). The problem of the red rice has been deepened by spread of direct seeding cultivation in recent years. It is difficult to control the red rice with herbicides or other cultural methods because it is same as cultivated rice taxonomically and have morpho-physiological similarity with cultivated rice. On the other hand, some red rice has specific characteristics for example seed dormancy and seed shattering.

In this study, we compared 141 red rice accessions collected from 14 rice-producing countries to reveal the genetic and morpho-physiological characteristics of red rice by countries/regions. We categorized these accessions into indica/japonica type and compared the regional breakdown of indica/japonica type of the red rice accessions and common rice varieties. We also evaluated the wild/cultivated type of these red rice accessions based on morpho-physiological characteristics, for example seed dormancy and seed shattering, by field experiments in Japan. The relationship between the red rice characteristics and cultivation/weeding system used in these regions was discussed.

Materials and Methods

One hundred and forty one accessions of red rice were provided by the National Small Grains Research Facility, USDA-ARS, USA, The Institute of Genetic Resources, Kyushu University, National Institute of Agro biological Sciences Genebank, The Japan Association for Advancement of Phyto-regulators, and Nagano Agricultural Experiment Station, Japan. These red rice accessions collected from North and South America (United States and Brazil), Europe (Italy, Hungary, and Russia), South and Southeast Asia (Pakistan, India, Nepal, Sri Lanka, Indonesia, and the Philippines), and East Asia (China, Korea, and Japan) were studied.

These red rice accessions were cultivated in Tsukuba, Japan (lat 36° 0'29" N, long 140° 1'18" E, 50 m ASL, annual mean temperature 14.3°C, monthly mean temperature max. 25.7°C and min. 3.6°C, annual rainfall 1400 mm, monthly rainfall max. 184 mm and min. 51 mm) in 2005. Six morpho-physiological characteristics of these red rice were examined: three indica/japonica diagnostic characteristics (resistance to $KClO_3$, glume hair length, phenol reaction) and three wild /cultivated characteristics (100-grain weight, degree of seed dormancy and degree of seed shattering). The methods described by Tang and Morishima (1997) were used to examine these characteristics. We surveyed on indica/japonica type of common rice variety in related countries by the database “World’s Planted Rice Varieties” which is based on the survey during 1981-97 and was released on the following website by International Rice Commission, FAO (http://www.fao.org/ag/agp/agpc/doc/riceinfo/plantvar/intro.htm.).

Results and Discussion

The most red rice accessions collected from North and South America (United States and Brazil) were characterized as indica type though they had variation on wild/cultivated characteristics. The red rice accessions collected from Europe were split into different types, the accessions collected from Italy were characterized as indica type, and the accessions collected from Hungary and Russia were characterized as japonica types. Most of the accessions collected from South Asia (Pakistan, India, and Nepal) were characterized as indica and wild type. The accessions from South East Asia (Indonesia and the Philippines) had characteristics closer to those of cultivated rice. The accessions collected from South East Asia and East Asia (China and Korea) were divided into japonica and indica types independently of the countries. The accessions collected from Sri Lanka had intermediate characteristics between those of japonica/indica and wild/cultivated types. The red rice collected from Japan in past years was split up into japonica and indica type and they have wide variation on wild/cultivated characteristics. However, the weedy red rice recently collected from Nagano, Japan were characterized as typical japonica and had characteristics very close to those of wild type.

These countries were categorized in four groups based on indica/japonica type of red rice; indica type group (U.S., Brazil, Italy, Pakistan, India, and Nepal), japonica type group (Hungary, Russia, and Nagano, Japan), both indica/japonica types group (Indonesia, Philippines, China, Korea and Japan except Nagano), indica/japonica intermediate type group (Sri Lanka). It is interesting that indica/japonica type of common rice variety in these countries did not necessarily match with that of red rice. For example, the most common cultivated rice in the U.S. were a japonica type though the most red rice accessions collected from the U.S. were characterized as indica types. This discrepancy between rice variety and red rice suggest that they did not have a genetic relationship and the red rice were smuggled in from abroad because U.S. has no wild rice geographically. On the other hand, Japan has both indica/japonica red rice cultivars historically though it does not have wild rice geographically. However, only japonica red rice is occurring as a weed in Japan, especially in Nagano. The weedy red rice has most wild-like characteristics, for example strong seed dormancy and shattering habit, and were quite different from the red rice collected in the past. This difference might suggest the weedy red rice in Japan had changed its characteristics to survive in the fields under intensive weed control and selective pressure.

In conclusion, the indica/japonica characteristics of red rice vary from region to region, and the variation may reflect the cultivated rice varieties introduced in the region, however, smuggled red rice also may exist in certain regions independently. The variation of wild/cultivated characteristics of red rice may reflect not only the existence of wild rice in these regions but also micro-evolution of red rice by the selective pressure with cultivation systems (for example rotation or continuous cropping) and weeding systems (such as the practice of hand weeding) used in these regions. We should take great care when cultivating red rice, especially when using direct-seeding system, because these strains could easily become weedy type if they have characteristics that allow them to survive in the field, such as shattering.

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BAMBOOS AS A WEED AT URBAN FRINGE IN JAPAN

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Abstract: This study investigates how bamboos as weeds have expanded at urban fringes in Japan. The number of bamboo patches and patch sizes were monitored in relation to surrounding land by using a GIS system at four different years: 1968, 1978, 1992, and 2002. The total area of bamboo patches increased after 1968, in contrast to after 1992 when the number of patches decreased. The relative expansion rate estimated by applying a logistic equation showed its peak in 1978, was maintained at a high level until 1992, and then decreased. The relative expansion rate of bamboo patches was high in non-human-disturbed areas, while it was low in disturbed areas. The decreased bamboo use in this decade due to development of Japan has led to unmanaged bamboo patches and promoted bamboo weediness.

Key words: Bamboo forests, GIS, land use, sustainable management

Introduction

Bamboos, all representatives belonging to Bambuseae, Gramineae (Clayton and Renvoize, 1986) are widely distributed throughout Asian, African, and south American continents. There are 111 bamboo genera in the world, where 1030 to 1100 species are assumed to exist (Ohrnberger, 1983). In Japan there are 300 or 500 species/taxa including the dwarf bamboos (Suzuki, 1978, Okamura *et al.* 1991). Three major useful bamboo species, Japanese Timber Bamboo (*Phyllostachys bambusoides*), Moso bamboo (*P. pubesense*), and Henon bamboo (*P. nigra.* var. *henonis*) are frequently planted throughout Japan. Due to its vigorous nature and growing capacity, tens of centimeters of elongation per day, the bamboo is considered to have the spirits, and then used as a sacred plant at belief and ethnic festive occasions, as well as being used for timber in the tea ceremony and the art of flower arrangement, and other traditional Japanese arts. Bamboo shoots are pleasant vegetables, and as bamboo timbers are able to be divided easily and are lighter and more elastic than wood timbers, they are excellent materials for miscellaneous daily goods and for farming tools such as netted sieves and baskets. In contrast to such traditional historical uses in Japan, bamboo utilization drastically changed after 1960 during a time of high economic growth.

Consumption of bamboo has decreased in Japan as cheaper bamboo shoots and timbers have been imported from developing Asian countries and due to the reduction of industrial materials that are competitive with bamboo materials. Moreover, bamboos conventionally cultivated in rural areas are unmanaged due to reducing of agriculture activity. Consequently, unmanaged bamboo forests have expanded in a given region or area. It is feared that the expansion of bamboo forests expels the surrounding vegetation, makes a monotone landscape composed of bamboo, and may decrease the level of biodiversity. The establishment of continuous maintenance and administrative plans for bamboo forests has not been achieved although the management activity such as cutting of bamboo forests has recently been conducted as a temporary approach to control the expansion of bamboo forests in several parts of Japan. It is necessary to clarify the expansion process of bamboo forest and its characteristics and to estimate the utilization of bamboo for sustainable management of bamboo forests. In this review the authors have integrated the current state and expansion characteristic of bamboo forests according to our current research.

Survey Sites and Methods

Survey Site

The survey site is Kishiwada City located in the southwest part of Osaka Prefecture, Japan. The site is rich in natural environmental elements such as sea and mountains, and has a traditional and long history. Its urban areas, farmland, and mountainous districts are almost similar in proportion to their land use; the urban areas extend to the plains facing the sea, and agriculture is active in the hilly areas and in the valley lines at the feet of mountains.

The mountainous part was for forestry but is not so active. The annual average temperature is 15℃-16℃, and annual precipitation is approximately 1000 mm-1250 mm (Anonymous, 1976). Recent production of bamboo shoot was 179 t (in 1998; Anonymous, 1998) although the highest production of bamboo shoots in this region occurred in Osaka Prefecture (859t in 1969; Anonymous,1969). The exploration of 78 patches in 2000 indicates that bamboo forests are composed of pure forests of Moso bamboo, pure forests of Henon bamboo, and mixed forests of Moso, Henon and Japanese timber bamboos. Pure Moso bamboo forests accounted for about 70 per cent of the entire forests, and the bamboo forests including Moso bamboo accounted for 90 per cent or more. In the composition of bamboo forests in this region, the Moso bamboo is the dominant species.

Methods

A topographical map and digital mapping data were basically used to determine the location of bamboo forests. In addition, QuickBird satellite images were used to obtain the latest location data. An analytical year is 2002, 1992, 1978 and 1968. A Geographic Information System (GIS) was used for a temporal, spatial analysis for bamboo patches in relation to artificial influence on distribution, structure change, geomorphology and the surrounding land use of bamboo forests.

Results and Discussion

Transition of patch

Bamboo in the survey area is distributed in many small patches of forests in the plains in the northern area, and large patches exist in mountainous areas from hilly sites in the central area (Ohno *et al*. 2002A). Drastic expansion of the bamboo patches was found in the hilly and the mountainous areas (Figure 1). Analyses done in 2002, 1992, 1978 and 1968 on the structure change in each patch of bamboo indicated that bamboo patches before 1992 showed a remarkable patchy structure in expanding areas (Ohno *et al*. 2004). Although the area occupied has expanded after 1992, small patchy fragmented bamboo forests increased greatly. Thus it is clear that the expansion of bamboo forests with reduction of patch size involved synchronized events (Ohno *et al*. 2004). The area of the bamboo forests has increased since 1968, 213.46 ha in 1968, 253.50 ha in 1978, 421.07 ha in 1992, and 484.91 ha in 2002. The patch size remarkably increased from 1978 to 1992, however, it was relatively stable from 1992-2002 (Ohno *et al*. 2004). The number of patches in the area studied increased from 773 in 1992 to 738 in 2002, and the maximum patch size decreased from 65.09 ha in 1992 to 26.11 ha in 2002 (Table 1).

The reduction of the patch size is considered to be a patch extinction and fragmentation caused by urbanization of the area investigated. The small expansion of bamboo forests from 1992 to 2002 implies a saturation of bamboo forest expansion in the area surveyed.

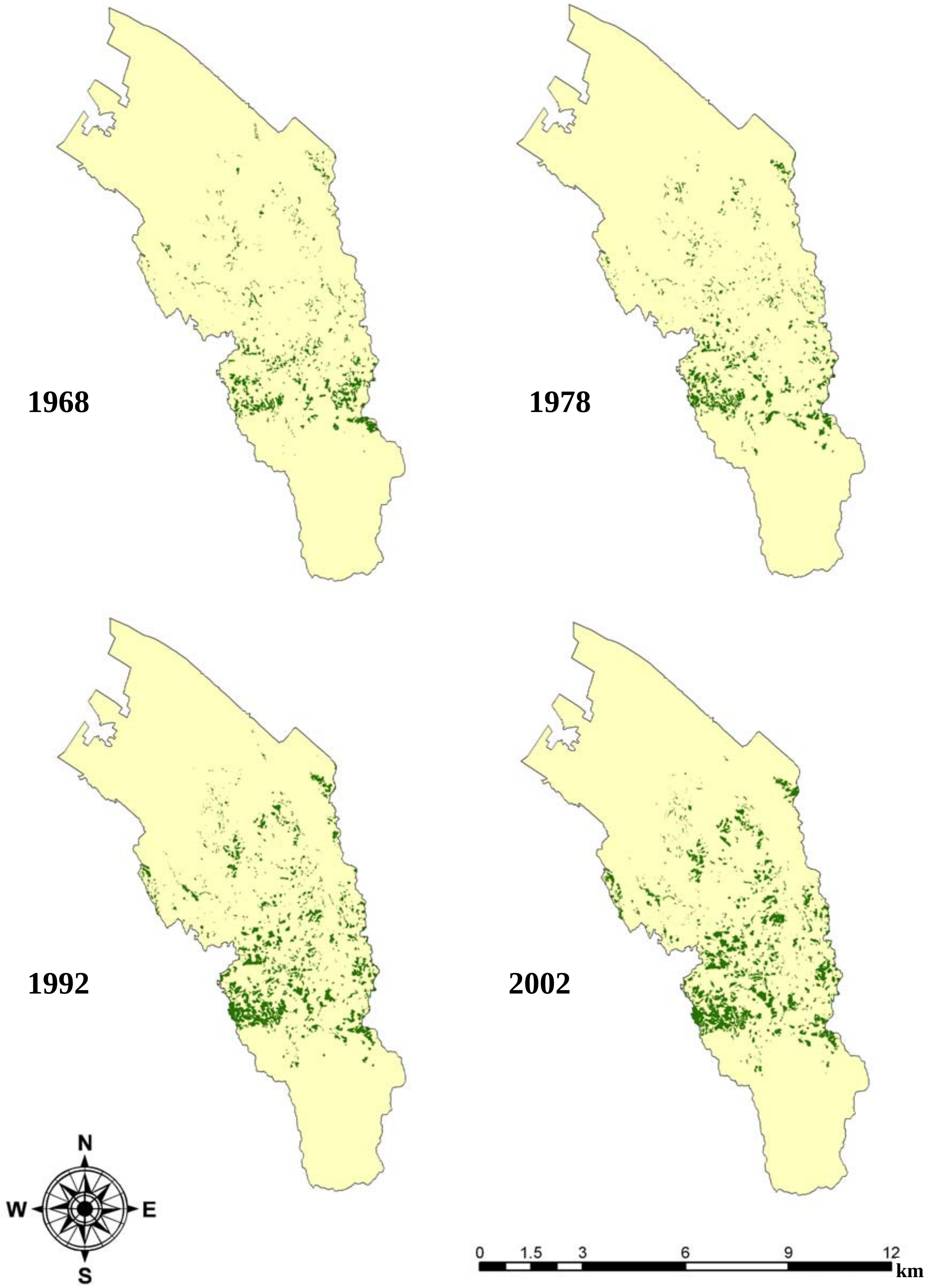

Figure 1. Distribution of bamboo forests at four different years in survey site

Distribution of bamboo forests in relation to altitude

The area occupied by bamboo showed different transition patterns at different altitudes. The occupation of the bamboo forests is very low, 1% of the total area 2988.79 ha at 0-50 m in altitude (Ohno *et al*. 2005). There are no bamboo forests at 300 m high or more, however, the occupation of the forest exceeded 30 % or more at 200 m to 300 m (Ohno *et al*. 2005). The total area of the bamboo forests changed remarkably at an altitude of 50-100 m, in particular from 1978-1992 (Table 2) (Ohno *et al*. 2005). The increasing rate of bamboo forests was highest in 1992-1978 among all altitudes. The increasing rate decreased after 1992 though the total area increased at all altitude levels in 1992 to 2002 (Table 3) (Ohno *et al*. 2005).

Table 1. Number of patches and total area of bamboo forests in 1968, 1978, 1992 and 2002 (Ohno *et al*. 2004)

Items	year			
	1968	1978	1992	2002
Number of patches	567	619	773	738
Total area of bamboo forest (ha)	213.46	253.5	421.07	484.91
Average patch size of bamboo forest (ha)	0.38	0.41	0.54	0.66
Max	14.19	13.66	65.09	26.11
Minimum	0.01	0.01	0.01	0.01
Standard deviation	0.98	0.92	2.55	1.77

Table 2. Occupation of bamboo forests in each altitudinal level (Ohno *et al*. 2005)

Altitude	year								
	1968		1978		1992		2002		survey area
	ha	%*1	ha	%	ha	%	ha	%	ha
0 ~ 50m	14.25	0.5	15.22	0.5	26.39	0.9	31.28	1.0	2988.79
50 ~ 100m	34.81	6.2	41.9	7.4	97.56	17.3	118.05	20.9	564.22
100 ~ 150m	31.72	7.8	41.82	10.2	56.76	13.9	64.45	15.8	408.48
150 ~ 200m	37.82	7.7	59.04	12.0	82.7	16.9	99.2	20.2	490.34
200 ~ 250m	58.66	15.1	65.52	16.8	117.88	30.3	131.6	33.8	389.61
250 ~ 300m	36.2	28.9	29.49	23.6	39.35	31.4	39.82	31.8	125.18
300m ~	0	0.0	0.51	0.0	0.43	0.0	0.51	0.0	1410.95

*1.percentage to area surveyed

Table 3. The increasing rate to bamboo forests in each altitude level (Ohno *et al*. 2005)

Altitude	increasing rate (year^{-1})		
	year		
	1968- 1978	1978 - 1992	1922 - 2002
0 ~ 50m	0.007	0.039	0.017
50 ~ 100m	0.019	0.060	0.019
100 ~ 150m	0.028	0.022	0.013
150 ~ 200m	0.045	0.024	0.018
200 ~ 250m	0.011	0.042	0.011
250 ~ 300m	- 0.021	0.021	0.001
300m ~	-	- 0.012	0.017

increasing rate= r   r =(log(S2)-log(S1))/t

r=The increasing rate   S1=Area before change

t=Years of period   S2=Area after changes

The relative expansion rate

The relative expansion rate of bamboo forest is proportional to the square root of the area size at previous estimation (Ohno *et al*. 2004). The relative expansion rate decreased after a peak dueirng 1978-1992. Since that time the relative expansion rates at three periods were compared by using this relational expression (Ohno *et al*. 2004). In comparison of the expansion rate of patch area, and the increased rate of patches at a given term, the results indicate the expansion rate of patch area increased, but the increased rate of patches decreased after 1992. When the relation of the surrounding land use and the relative expansion rate was examined in 2002, the relative expansion rate of bamboo forests was high in non-human-disturbed areas such as broad-leaved forests, coniferous forests, and wasteland (Ohno *et al*. 2002B) while the expansion rate was low in disturbed areas such as urban areas, road areas, and waters. No relationship was observed between the relative expansion rate and the land use, where land uses in the surrounding area consisted of rice fields, upland common fields, and orchards, because the major land uses of the sites investigated were periodically managed. This tendency was confirmed by an applied ecological analysis (Ohno *et al*. 2004).

Conclusion

The expansion of bamboo forests is increasing now in Japan, but the relative expansion rate of bamboo forests changes with time, and has become gradual after peaking in 1978-1992. Bamboo forests require maintenance and management, or they are diverted to other land uses and decrease if the land use is strongly influenced by human activity. Conversely, land that has been managed up to now becomes a place for expansion of bamboo forests if the use of the land is not controlled, and if the bamboo forests increase. The expansion of bamboo forests is dependent on the intensity of human activity rather than limitation caused by geomorphology. Our studies will provide management skill of bamboo expansion as weed.

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SEED DEVELOPMENT AND GERMINATION ABILITY OF *Alopecurus aequalis* L.

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Abstract: *Alopecurus aequalis* L. (Water foxtail) is one of the common winter-annual weeds of upland fields in Korea. Formation of embryo and endosperm of a species affects the germination ability of seeds. This study was conducted to define seed development and germination ability of *Alopecurus aequalis* L. after anthesis. The seed dry weight was affected by the number of days after anthesis (DAA). The quadratic regression analysis indicated that seed weight increased sharply up to 19 DAA from 0.136 to 0.471 mg grain^{-1}, and decreased thereafter. The length and the width of seed increased with increasing DAA. Seed thickness increased moderately up to 16 DAA, but did not show a distinct increase after 17 days. Germination ability of the seeds was low up to 6 DAA, but it sharply increased from 11 to 14 DAA (9.3% to 83.3%). However, it moderately increased after 15 days (94%). This pattern was similar to the weight increase observed in the developing seeds..

Keywords: Seed development, germination ability, *Alopecurus aequalis* L.

Introduction

Alopecurus aequalis L. is a common winter-annual weed of upland fields in Korea (Ku *et al*. 2004). Seeds of common weeds are ripened early than that of crops, and fall on the ground before harvesting the crop, enriching the soil seed bank and sustaining the generations. *Alopecurus aequalis* L. blooms in late April to middle May, where the plant is able to produce about 2,000 seeds (Lee, 1985). Seeds germinate best in early fall or spring; however germination can occur throughout the year in Korea. Seed development requires the formation of embryo and accumulation of endosperm tissues, giving the seed the germination ability during maturity. In crop plants, the length and the width of seeds are decided before heading. However, the thickness of seed is depended upon conditions such as nutrition and weather especially after heading (Park and Lee, 2004). Increase of seed dry weight continues until physiological maturity stage which is about 30 days after heading, with no observable increase thereafter (Ju *et al.* 2007). Recent research has shown that germination of grape and *Hibiscus syriacus* with larger embryo size was higher than those with smaller sized embryos (Park and Kim, 2001; Kim *et al*. 1996). Expression of germination ability seems to be associated with seed development. However, the process and identity of seed developments in weeds remains unknown. Information and knowledge on seed development in major weeds would help in estimating the potential of their spread to new crop lands. Therefore, a study was conducted with the objective of identifying the seed development and germination ability of *Alopecurus aequalis* L during different days after anthesis.

Materials and Methods

Plant material

Naturally occurring populations of *Alopecurus aequalis* L. in Korea, on sandy-loam soil, were selected for the study.

Sampling times and measurements

Heads of the main stem of 150 plants of *A. aequalis* L. were tagged when they were in full anthesis. The heads of main stem from 5 plants were sampled everyday from 1st to 26th days after anthesis (DAA). Heads were separated into seeds and pods. Seeds were dried at 60 °C for 72 hrs in a dryer and were weighed. The pericarps of the weighed seeds were removed, and the width, length and thickness of seed were measured using a microscope, holding with tweezers due to the smaller size of seeds. Germination ability of the seeds sampled at each DAA was studied using Petri dishes that were incubated at 15°C for 20 days under dark condition.

Results and Discussion

Seed is composed of endosperm and embryo, which is surrounded and nourished by starch. Endosperm development is essential for seed development, thus increasing the seed weight. The dry weight of seeds was different with number of days after anthesis (DAA). The quadratic regression best described the increasing of seed weight response to getting DAA. The regression analysis indicated that seed weight increased sharply up to 19 DAA, from 0.136 to 0.471 mg grain^{-1}, and then decreased moderately (Figure 1). Increase of seed dry weight continued up to the physiological maturity stage, which was 30 days after heading (DAH), but the seed weight did not show any increase after the physiological maturity (Ju *et al*. 2007).

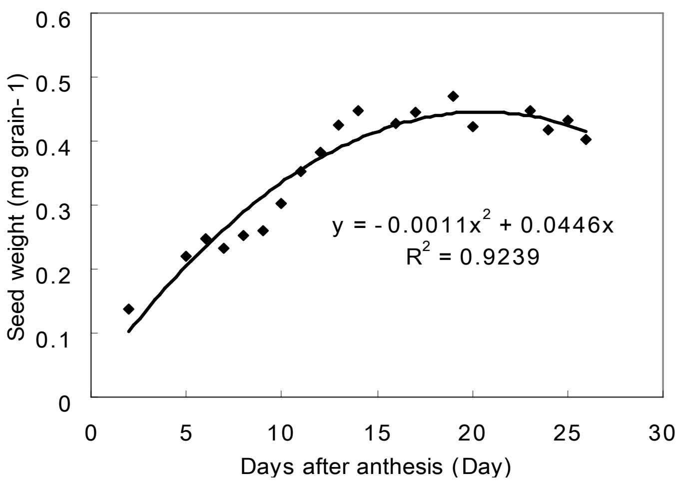
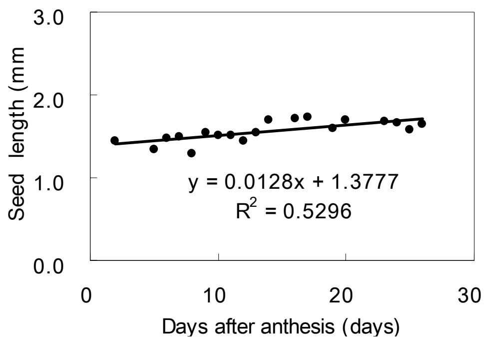


Figure 1. Seed weight response of *Alopecurus aequalis* L. to days after anthesis.

In *Oryza sativa*, the seeds accumulate protein and starch during thirty five days after heading, and reach the maximum weight (Park and Lee, 2004). Seog and Kim(1993) reported that the dry weight in barley sharply increased from 20 to 36 DAH, and did not increased thereafter, demonstrating a S-shaped growth. The results of the present study were to similar to previous reports and it can be concluded that the maximum weight stage of *Alopecurus aequalis* L. was 19 DAA.

Seed development requires the formation of embryo and accumulation of endosperm tissues that eventually increase the seed weight. The protein synthesis varies according to the stage of seed development (Byeon *et al,* 2005). In the present study, the length and width of seed was increased slightly with the increasing DAA (Figure 2). Seed thickness increased moderately up to 16 DAA, but no increase in the thickness was observed thereafter (Figure 3), similar to the increasing of seed weight observed (Figure 1). Previous studies carried out with *Oryza sativa* L. have also shown that seed length does not show any considerable increase from 5 days after heading (DAH), But the width, and the thickness increased to 15 and 21 DAH, respectively (Park and Lee, 2004). The results this study suggested that seed thickness of *Alopecurus aequalis* L. sharply increased, and the length and width of the seed did not show a marked increase with DAH. The results are similar to the reports that accumulation of endosperm continued up to the maturing stage (Lee and Lee, 1994), but in barley the synthesis of starch did not increase (Kim *et al*.,1993).



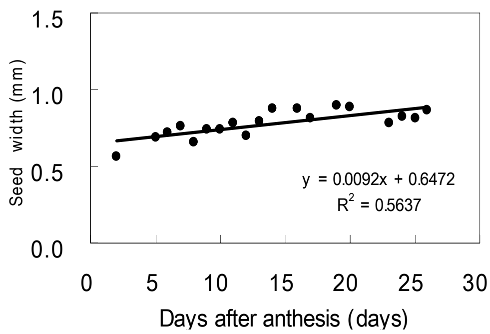


Figure 2. Seed length (left) and wide (right) response of *Alopecurus aequalis* L. to days after anthesis.

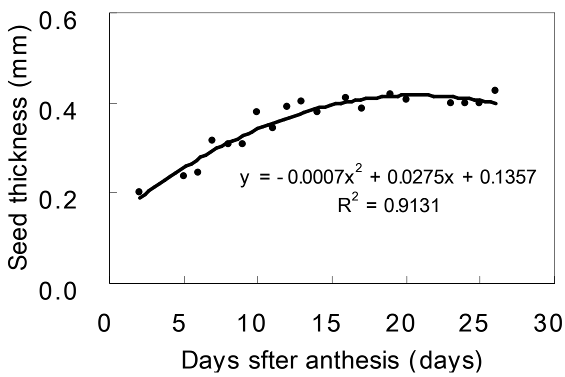


Figure 3. Seed thickness response of *Alopecurus aequalis* L. to days after anthesis.

Seed maturity has an effect on the germination ability of the seeds. Germination ability of the seeds were different based on the number of DAA, where it was low up to 6 DAA, and sharply increased from 11 to 14 DAA showing an increase from 9.3% to 83.3%. The germination ability of the seeds moderately increased after 15 days to 94%, following a pattern similar to that observed in the increase of seed weight.

The results indicate that there are three stages of seed development where germination ability of the seed varies (Figure 4); the first stage was from 6 to 10 DDA with a marginal increase in germination ability of seeds, the second stage was from 11 to 14 DAA with a sharp increase in germination ability, and the third stage from 15 DAA with a marginal increase. Recent research results have shown that *Oryza sativa* L. seeds had a germination ability at 10 days after pollination (Park and Lee, 2004), and seeds of *Hibiscus syriacus* with larger embryos germinated well compared to those with smaller embryos (Park and Kim, 2001, Kim *et al.* 1996). The results of the present study suggested that *Alopecurus aequalis* L. had the germination ability from 6 DAA with a larger embryo.

Various external conditions, such as pH, temperature and osmotic potential affects germination (Chachalis and Reddy, 2000; Koger *et al*. 2004; Nandula *et al*. 2006) of seeds self- and cross-pollinated plants (Pounders *et al.* 2006). In the present study the seed size and the maturity of *A. aequalis* L. had a significant influence on the germination ability of seeds.

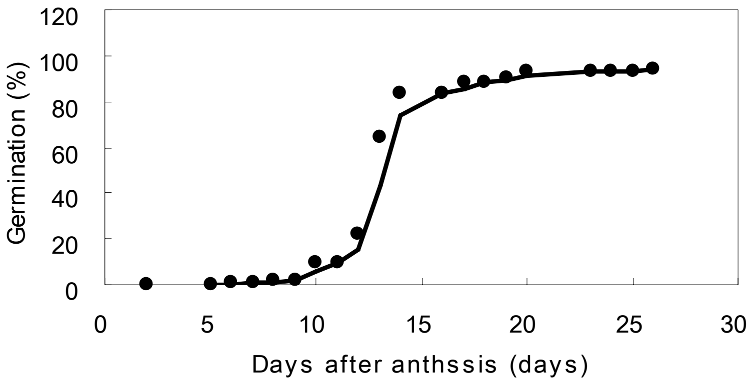


Figure 4. Seed germination response of *Alopecurus aequalis* L. to days after anthesis.

The results conclude that *A. aequalis* L. seeds have the ability to germinate from 6 DAA. The width and the length did not show a significant change with increasing DAA. The dropped seed of *A. aequalis* L. on the ground after 6 DAA has ability to germinate, allowing the chance of propagating weed seeds to next year.

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A HIDDEN VARIETY OF BARNYARD GRASS (*Echinochloa crus-galli* var. *riukiuensis* Ohwi) FOUND IN OKINAWA, JAPAN

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Abstract: The genus *Echinochloa* includes the world's worst weeds and important field crops. However, due to its wide morphological and ecological variations, taxonomic confusion still remains. For clarifying the present living status of the genus, the author has conducted many field surveys in East Asia. In the winter of 2005, the author discovered a representative of *Echinochloa* in a vacant paddy field on Iriomote Island, Okinawa Prefecture. The representative is similar to *E. crus-galli* in almost all of its plant morphological features. The plants were collected and maintained in a glass house, mature seeds were sown and plants were cultured from seedlings in Sakai. The plants at full maturity showed scattered ripening of shoots with good seed set. Nearly green whole plants were stumpy clumped and did not have extended horizontal stems (stolon). New shoots continuously emerged from the axially part of the stem base over a one year period, although mature stems had withered up. The author's observation indicated that this species is closely similar to features of caryopsis with *E. crus-galli* but not to spikelet features with *E. oryzicola*. Panicle and raceme shapes resemble those of perennial *E. stagnina* (Asian tetraploid form), which fits the taxon described earlier as *E. crus-galli* var. *riukiuensis*. Molecular phylogenetic analysis indicated that this representative shared the same sequences of non-coding regions of *trn T-F* genes with those of *E. crus-galli* var. *crus-galli*. The intermediate life form between perennial and annual found in this representative implies that *E. crus-galli* has an original perennial habit, or as a result of rapid adaptation to aquatic habitat, is seen frequently in typical *E. crus-galli* growing in aquatic habitats of tropical regions.

Key words: *Echinochloa crus-galli* var. *riukiuensis*, annual *vs*. perennial, morphological feature, ecological habit

Introduction

The genus *Echinochloa* includes three harmful weedy species and is important for cultivated species for cereals and pastures (Yabuno and Yamaguchi, 2003). Recently, several *Echinochloa* species have invaded human disturbed sites and waterfront areas causing loss of biodiversity as invaders (Michael 2001, 2003). Due to the lack of surveys on the living status of the genus, confusion of the taxonomic treatise still remains. A better understanding of ecological habit and diversity for the genus at the global level is necessary. In East Asia, there are three annual species, *E. crus-galli* (L.) Beauv., *E. oryzicola* Vasinger, and *E. colona* (L.) Link, as well as two perennial species, *E. picta* (Koen) Michael and *E. stagnina* (Retz.) P. Beauv. In annual species, each wild species has a cultivated counterpart. *E. crus-galli* has a temperate domesticate Japanese millet, *E. esculenta* (A. Braun) H. Scholz, *E. oryzicola* has Mosou barnyard millet, *E. oryzicola*, *E. colona* has semiarid species Indian millet, *E. frumentacea* (Roxb.) Link (Yabuno and Yamaguchi, 2003). These domesticated species are useful for animal feed and wine beverage. Every wild species includes several taxonomical varieties and morpho-ecological forms, however, in weed control these taxa are less discriminated, thus the ecological information accumulated so far is not universal.

Materials and Methods

During surveys conducted on *Echinochloa* occurring in unknown places, a representative was found in December 2005 growing in a vacant paddy field on Iriomote Island, Okinawa

Prefecture, Japan (Plate 1).Seeds and plants collected from this representative were sown and maintained in the laboratory and in a glass house on University campus (Sakai city, Osaka, Japan). Since the collection was done during the fall, the plants were all maintained in warm water. Seeds were germinated under natural environment, and seedlings and the plants were established in the natural paddy were grown vigorously in the glass house. From winter through spring to summer, the plants were grown as an ordinary annual species. Flowering culms were found from August in transplanted materials and seedlings. In the fall, every plant produced new stems and bear flowering scapes continuously. Fertile culms thrived due to the maturation of spikelets, however, plant stumps did not wither because of continuous production of new stems. New shoots could be produced in basal nodes of culms and from second and third nodes. The plants were kept in the glass house to avoid low temperature damage through winter, and thus no plants died over the 12 month period. A part of each stump with 2-3 culms was transplanted into new pots.

Plate 1. *Echinochloa crus-glli* var. *riukiuensis* in natural habitats.

Results and Discussion

On Ryukyu Island, two varieties of *Echonchloa crus-galli* were described in the 1940's (Ohwi 1942) namely, *E. crus-galli* var. *riukiuensis* Ohwi and var. *austro-japonensis* Ohwi. The representative collected in this study fits to the description of the former. The plant is green in color and has shorter leaves than the normal *E. crus-galli*. The length of the Inflorescence (panicles) ranges from 8 cm to 10 cm with 5 to 8 branches at less than 4 cm. Spikelets are arranged as one sided, two or three rows on each raceme. Spilelets bear an awn at a length of *ca*. 7 mm. The first glume is one third of the spikelet length. The tip of the spikelet is acute similar to that of var. *formosensis*. The lemma of the first floret is flat and rough. The caryopsis is 4.8 mm to 5.2 mm in length, 1.8 mm to 2.1 mm in width, nearly round, and the embryo scutellum is relatively large，80％ of the caryopsis length. Panicles are green and straw color at maturity. Internodes at the stump base are frequently reddish purple. All the transplanted individuals regenerated and showed good growth. The outline of the panicle and racemes are likely similar to that of *E. oryzicola* or *E. satgnina* (Plate 2).

Since the perennial habit and panicle shape of this representative are similar to *E. stagnina*, the *trnL-F* region sequences of this representative have been determined; however its sequences are the same as the sequences of normal Asian *E. crus-galli* (Yamaguchi *et al.*

2005, Yasuda *et al*. 2002). This finding agrees with the representative of *Echinochloa crus-galli* var. *riukiuensis* Ohwi.

Plate 2. Regeneration of young shoot (Left) and panicles (Center and Right)

Echinochloa crus-galli is conventionally treated as annual species, not perennial. It is well known that *Oryza sativa* is basically perennial although it is cultivated as an annual. Similarly, *E. cus-galli* might be perennial, not true annual, as var. *riukiuensis* shows a perennial habit. A similar case has been seen in some tropical countries. In 2006, a perennial habit in several *E. crus-galli* plants was identified in Khon Kaen in Thailand. The plant shown in Plate 3 has the regenerated shoots at nodes on the middle portion of the stem which were found at the edge of a creek at the beginning of dry season. In addition, several individuals of *E. crus-galli* (which may be var. *oryzoides* or non-shattering form) showed long extended stoloniferous stem with spongy pith.

Figure 3. Vegetative regeneration from stem nods in *E. crus-galli* in Thailand (Left); Fertile *E. picta* in Mindanao, Philippines. Vegetative stems expand in paddy, grazed shoots on the levee bear full mature panicles (Right).

These individuals showed a similar ecological feature with *Oryza rufipogon* or *O. nivara*. In other cases, the perennial *E. picta* usually shows clonally growth at waterfronts of streams or creeks along tropical rice paddies. Most of them bear sterile spikelets, however, the *E. picta* plants at drier places along rice paddies have high fertile habit with many ripen seeds. Fertile *E. picta* shows frequently small elegant plant stature due to grazing from buffalo or animals in the paddy sides (Plate 3). The fertile *E. picta* can have an annual life cycle depending on the dry *vs.* wet season circulation. Almost all perennial *E. picta* are aneuploid, which leads to sterile seeds, however, fertile *E. picta* is euploid (Tanesaka, 1991) and produces many seeds at human-disturbed sites. These all imply that *Echinochloa* species can be annual although the genus is basically perennial.

As shown in Plate 1, var. *riukiuensis* shows both seed and vegetative reproduction. The habitat of var. *riukiuensis* is usually wet, even during at non-cropped seasons, and traditional weed control of paddy has been by animal trampling on Iriomote Island (Ankei, 2007). The intermediate life form between perennial and annual found in var. *riukiuensis* implies that *E. crus-galli* has an original perennial habit, or as a result of rapid adaptation to aquatic habitat, it is seen frequently in typical *E. crus-galli* growing in aquatic habitats of tropical regions, or it is an adaptation to such traditional weeding practices. Consequently, varieties of *E. crus-galli* (L.) Beauv. in temperate and tropical East Asia can be classified as real representatives as var. *crus-galli*, var. *praticola* Ohwi, var. *formosensis* Ohwi, var. *oryzoides* (Ard.) Lindm., and var. *riukiuensis* Ohwi.

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SIMULTANEOUS EXCHANGE OF INFORMATION ON NATURALIZED PLANTS THROUGH MAILING LIST IN JAPAN

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Abstract: Co-ownership of information on naturalized plant species, which is generally reported in local journals is indispensable to understand plant invasion in Japan. Internet made exchange of plant information among different localities possible at the same time. In order to exchange situation of naturalized plants simultaneously, mailing-list for naturalized plant was established in web site of affrc.go.jp on January 2002, using Japanese language. As of January 2007, 2900 articles on taxonomy, distribution, ecology etc. about naturalized plants have been sent by around 500 members including professional and amateur botanists, weed scientists, technicians of environmental companies and lovers of plants from all over Japan. Activities of this mailing-list on new records and distribution such as *Silene nocturna* L., *Brassica tournefortii* Gouanl, *Gymnocoronis spilanthodes* DC. and *Linaria canadensis* Dum., keys for identification for *Physalis*, *Solanum*, *Cenchrus* etc., meeting for observation and so on, are introduced in this paper.

Key words: Naturalized plants, mailing list, keys for identification, Japan

Introduction

Social interest on invasive plant species is increasing in Japan, taking the opportunity of Invasive Alien Species Act (Law No.78, 2004). Records on invasion and distribution of naturalized plants were published occasionally in local plant Journals. It causes the difficulty in co-ownership of information on naturalized plant species. Efforts to exchange information on plants had been made through papers among professional and amateur botanists to contribute co-ownership of knowledge on plants including naturalized species in Japan. Exchanging information on invasion and distribution simultaneously throughout the country is indispensable to understand the dynamics of naturalized plant species, because their situations are affected greatly by human disturbance. In order to activate the exchange of situations on naturalized plants, simultaneous exchange system through mailing list was established. Outline of the activity of mailing list is introduced in this paper.

Materials and Methods

Mailing list for naturalized plants has been established in January 2002 with following concept and system.

Concept: Mailing list for naturalized plant naturplant@ml.affrc.go.jp is for the exchange of information on naturalized plants which affect agriculture, urban life, landscape and environment, throughout Japan. Mailing list consists of the scientists on botany, weed and ecology, the experts on environmental industries and the amateur plant lovers. It will be useful to exchange the information on naturalized plant on time in Japan through E-mail system.

System

1: E-mail address of mailing system, hereafter ML, is naturplant@ml.affrc.go.jp. science
2: Contribution and delivery of articles in ML is completely limited to the members registered.
3: Contact to enter or leave should be sent to the administrator, naturplant-admin@ml.affrc.go.jp.
4: Article should be text only without any kind of file attachment to avoid the computer virus problem.

Results and Discussion

Result of articles contributed

As of May 2007, member of ML is around five hundreds though it was around a hundred when started in 2002. Number of articles contributed exceeded three thousands at May 2007 as shown in Figure 1.

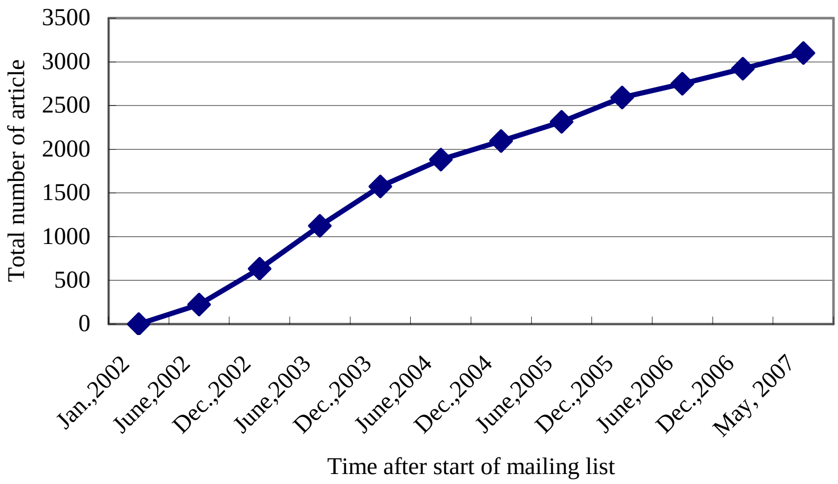

Figure 1. Number of articles in mailing list for naturalized plants

Outline of activities of ML

Naturalized plants newly recorded in Japan were introduced through ML for such as *Silene nocturna* L., *Brassica tournefortii* Gouanl, *Gymnocoronis spilanthodes* DC. (Plate 1), *Cleome rutidosperma* DC., *Desmodium elegans* DC., *Agalinis heterophylla* Small ex Britton, *Dryopteris intermedia* A. Gray and so on. Because of prohibition of file attachment, images of those plants are shown in Home Page of each contributor.

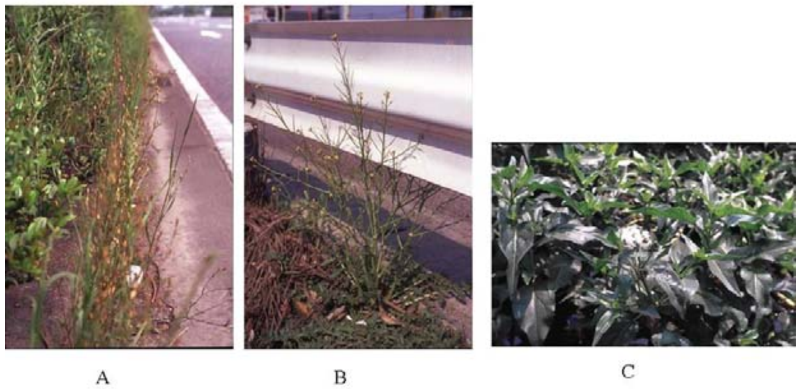

Plate 1. Newly introduced naturalized plant species in ML. A: *Silene nocturna* L., B: *Brassica tournefortii* Gouanl, C: *Gymnocoronis spilanthodes* DC.

Information on distribution of particular species can be provided from different prefectures or districts for such as *Linaria canadensis* Dum., *Veronica hederaefolia* L., *Gymnocoronis spilanthodes* DC., *Alternanthera philoxeroides* Griseb.(Figure 3), *Papaver dubium* L. and so on. Significant information on *Cyperus esculentus* L. was provided from ML members when Weed Science Society of Japan compiled the distribution map of *C. esculentus* (Shibuya and Morita, 2005).

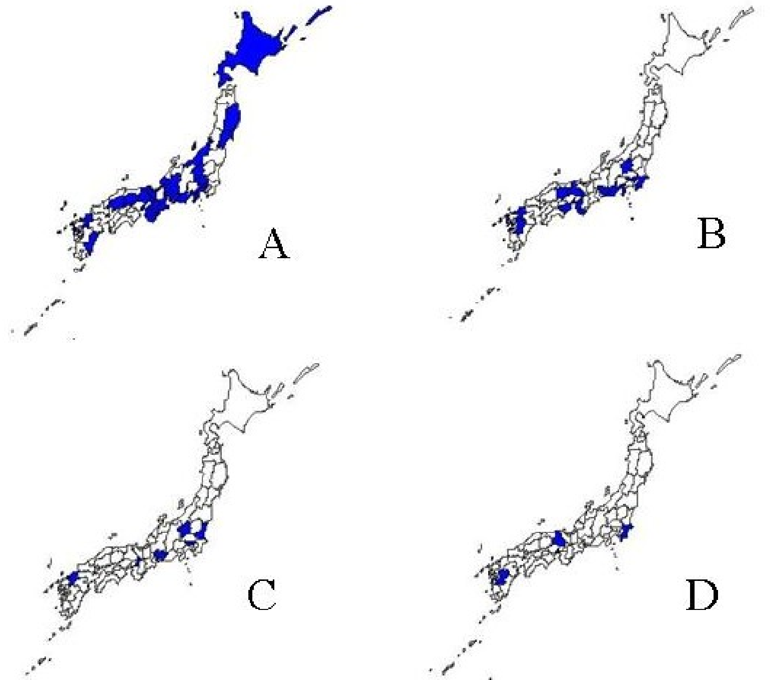

Figure 2. Distribution map in Japan compiled by information of ML. A: *Linaria canadensis* Dum., B: *Veronica hederaefolia* L., C: *Gymnocoronis spilanthodes* DC., D: *Alternanthera philoxeroides* Griseb.

Information for correct identification of naturalized plants which have close relatives has been provided through ML, because the detailed keys to identify are insufficient usually in Japan. Keys for correct identification have been adjusted through ML in *Xanthium*, *Tragopogon* (Table 1), relatives of *S. nigrum* in *Solanum*, *Physalis* (Table 2), *Cenchrus* (Table 3) and *Verbena* reported in Japan.

Table 1. Keys for *Tragopogon* species naturalized in Japan revised with information exchanged in mailing list

A: Flower bluish purple -----------	*Tragopogon porrifolius*
A: Flower yellow	
B: Head peduncle not thickened -------------	*T. pratensis*
B: Head peduncle thickened -------------	*T. dubius*

Based on T. Katsuyama [naturplant:1153] Tragopogon dubius

Table 2. Keys for *Physalis* species naturalized in Japan distributed by mailing list

A: Corolla less than 8mm in length, anther 1 – 2 mm in length	
B: Glabrous to puberulent, leaf margin acutely serrate, dark purple spot not obvious at center of corolla	
C: Leaf ovate, corolla 6 - 8mm in length, calyx 3 - 4mm in length at flowering, calyx lobe 2 - 2.5mm ----------------------	*P. angulata* L. var. angulata
C: Leaf ovate - lanceolate, corolla 6 - 8mm in length, calyx 4 - 5mm in length at flowering, calyx lobe 1mm in length ---------	*P. angulata* L. var. pendula (Rydb.) Waterfall
C: Leaf lanceolate, corolla 4 - 5mm in length, calyx 3 - 4mm in length at flowering, calyx lobe 1mm in length ------------------------	*P. angulata* L. var. lanceifolia (Nees) Waterfall
B: Pubescent, leaf margin entire or obtusely serrate, dark purple spot obvious at the center of corolla	
C: Glandular hair absent, leaf margin entire or several obtusely serrate -----------------	*P. pubescens* L. var. pubescens
C: Densely pubescent with white hair and glandular hair, leaf margin obtusely irregularly serrate -----------------	*P. pubescens* L. var. grisea Waterfall

Compiled from Kimura and Katsuyama (2000)

Table 3. Keys for Cenchrus species naturalized in Japan distributed by mailing list

A: Involucre more than 15 in an inflorescence, with thick and hispid prickles, thick prickles erect or ascending when matured	
B: Involucre dense on axis, 4 – 5mm in length, pubescent ----------	*C. brownii* Roem.
B: Involucre thin on axis, 5 – 6mm in length, covered with white pubescent hair -----------	*C. echinatus* L.
A: Involucre 6 – 10 in an inflorescence, with thick prickles only, divaricate or reflex when matured	
B: Involucre 7 – 8 mm in length	
C: Prickles on involucre around 30, base of prickle 1.5 – 2 mm in width -----------	*C. incertus* M.A.Crutis
C: Prickles on involucre 45 – 75, base of prickle less than 1 mm, basal short prickles reflex downward -----------	*C. lingispinus* Fernald
B: Involucre 10 – 15 mm in length -----------	*C. tribuloides* L.

11 Apr 2002 T. Katsuyama [naturplant:102] on genus of *Cenchrus*

Meetings for observation of naturalized plants were organized by the active members of ML. The first observation meeting was held on April 2002 at Kobe, Hyogo Prefecture where many kinds of naturalized plant species were collected, because of the location near the international port. The second meeting was held on August 2003, at a naturalized plant garden in Koto Ward, Tokyo. The participants of meeting could learn the point of identification with the intact plants.

In conclusion, this mailing list is playing a central role to exchange information on naturalized plants simultaneously in Japan. And also, it shows that this system is useful to grasp the present situation of plant invasion in the country.

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NOXIOUS WEEDS IN WHEAT, BARLEY, SOYBEAN AND DIRECT SEEDED RICE, AND NARO'S RESEARCH PROJECT FOR INTEGRATED WEED MANAGEMENT

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Abstract: Many herbicides have been developed and popularly used in crop fields in Japan. Arable noxious weeds, however, still cost farmers much to control them; *Avena fatua*, *Lolium multiflorum*, *Alopecurus aequalis* and *Beckmannia syzigachne* in winter cereals, *Chenopodium album* and *Amaranthus* spp. in soybean, and *Murdannia keisak* and *Echinochloa crus-galli* in direct seeded rice. These noxious weeds are often tolerant to control measures and adaptable to various habitats, threatening sustainability in crop production. The noxious weeds need to be managed by integration of several control measures including cultural practices, crop rotation, cover plants cropping, dead/living mulch, weeding machines, safer herbicides etc. National Agriculture and Food Research Organization (NARO) conducts the research project to develop innovative technology for integrated weed management (IWM) to solve the noxious weed problems. The project contains studies on weed seed bank, weed population dynamics, influence of insect seed predators on weed population, developed weeding machine, cover plants cropping and confirmation of economic optimum threshold for sustainable weed control.

Key words: Integrated weed management, weed seed bank, herbicide, direct seeding, noxious weeds, cover plants cropping

Introduction

In Japan rice planted area is decreasing year by year, one third of lowland fields are used for upland crop production; soybean, wheat, barley, forage crops etc., or fallowed. Introducing of upland crops into lowland areas resulted in various crop rotations. Although paddy-upland crop rotation usually contributed to reduce weed emergence, recently noxious weeds became serious in these lowland fields. It was due to very limited herbicides had been registered to control grass weeds for winter cereals and broadleaved weed for soybean. Noxious weed are often resistant, tolerant or avoidant to herbicide activity. Furthermore infestation with invasive exotic weeds made the problems complicated. To solve the problems, various weed control measures including mechanical, ecological, biological and chemical methods should be integrated. National Agriculture and Food Research Organization (NARO) has just started the research project for Integrated Weed Management (IWM) in 2007. The target weeds and research contents of the project are described in this paper.

Infestation and Problems of Several Noxious Weeds

Avena fatua L. and Lolium multiflorum Lam. in winter cereals

Occurrence of wild oat (*A. fatua*) and Italian ryegrass (*L. multiflorum*) was investigated in Kanto-Tokai region (Asai and Yogo, 2005), and found that wheat or barley fields in every prefectures were infested with these two grasses. Wild oat was more serious in continuous winter cereals cropping compared to rice-wheat/barley rotation. Water irrigation in summer season was supposed to reduce buried seeds viability of wild oat during rice cultivation. No-tillage during summer caused early emergence and reducing seed viability of wild oat in the next wheat season (Asai and Shibuya, 2006). Italian ryegrass was adaptable to wider range of soil moisture in lowland crop-rotation fields. The seed was more tolerant to summer water irrigation than wild oat seed.

Table 1. Recent noxious weeds in paddy-upland rotation in Japan.

Crops	Noxious weeds	Remarks
Winter cereals (Wheat, Barley)	*Avena fatua* L.	Low sensitive to herbicides
	Lolium multiflorum Lam.	Low sensitive to herbicides
	Beckmannia syzigachne (Steud.) Fernald	Low sensitive to herbicides
	Alopecurus aequalis Sobol. var. *amurensis* (Komar.) Ohwi	Multi-Resistant biotypes*
Soybean	*Chenopodium album* L.	Low sensitive to bentazone
	Amaranthus spp.	Low sensitive to bentazone
	Bidens frondosa L.	Invasive naturalized weeds
	Ipomoea spp.	Invasive naturalized weeds**
Rice	*Echinochloa crus-galli* (L.) Beauv. var. *crus-galli*	Wet/Dry seeding
	Murdannia keisak (Hassk.) Hand-Mazz.	Wet seeding
	Scirpus juncoides Roxb. var. *ohwianus* T. Koyama	SU-Resistant biotypes***
	Monochoria vaginalis Presl var. *plantaginea* Solms-Laub.	SU-Resistant biotypes****

* Uchikawa *et al*. (2005), Ohdan *et al.* (2005), ** Hukumi and Yamashita (2005), Hiraiwa *et al.* (2005),
*** Kohara *et al.* (1999), **** Koarai and Morita (2002)

Chenopodium album L. in soybean

Infestation of broadleaved weeds is more serious in northern part of Japan where the competitive grass weeds is not very much dominated in cool region. In 2005 foliar application of bentazone was registered to control broadleaved weeds for soybean production. Many weeds including *Persicaria* species, *Portuloca oleracea* L. and *Cyperus* species were highly sensitive to the herbicide, but it was not very effective on several weeds such as common lambsqarters (*Chenopodium album* L) and *Amaranthus* species (Shibuya *et al.* 2006). Since minimum temperature for seed germination of *C. album* is very low, around 6 degrees centigrade, the weed emerges earlier after soybean seeding in the cool temperate region. Adult plants produced so many small size seeds that the management of the weed seed bank was very difficult.

Murudannia keisak (Hassk.) Hand-Mazz. in dry seeded rice

Dry direct seeding of rice is one of the cost effective seedling establish method. It saves labor cost especially in practiced using multi-low seeder in large scale fields. Weed infestation, however, is usually more serious compared to transplanted or wet-seeded rice, three or four times of herbicide application are necessary. Marsh dayflower (*Murudannia keisak*) is one of levee weeds which emerge under dry or drained conditions. In dry seeded rice it emerged during rice seedling establishment period and grows vigorously in standing water after irrigation. The weed was effectively controlled by foliar application of bispyribac-sodium in rice (Shimamune and Hanzawa, 2005). The weed plants those had emerged at levee elongated their procumbent stems and invaded into rice fields producing a lot of seeds in the fields. Integration of field and levee management would be very important to control the weed.

Echinochloa crus-galli (L.) Beauv. in wet seeded rice

Two species of barnyardgrass grows in lowland rice fields, *E. oryzicola* and *E. crus-galli* (Yabuno, 1975). The former is very mimic to rice and well emerges in standing water condition, which has been the most important grass weed in traditional and modern transplanted rice in Japan (Yamasue, 2001). Recently, however, *E. crus-galli* often became dominant in wet direct seeded rice. It may be due to water drainage treatment after rice seeding which is practiced for stable rice establishment. In wet seeded rice *E. crus-galli* emerged and grew 1 or 2 leaf stage earlier than rice seedlings under drained fields, resulting in insufficient herbicidal efficacy; the bigger plants often recovered after herbicide application. Proper land preparation before/at rice seeding is necessary to stabilize control effect of herbicides on *E, crus-galli*.

NARO's Research Project on IWM

NARO has just started research project on integrated weed management (IWM). The objective is to solve the noxious weed problems without increase of herbicide use in several paddy-upland rotations. In the project, ecological and mechanical weed control technique would be integrated with minimum use of chemical herbicides in winter cereals, soybean and direct seeded rice respectively. Sustainability of weed control effects in each IWM are evaluated in farmers' fields where the economical benefit would be also estimated.

Table 2. Research components of the NARO's IWM research project

(1) Diagnosis and Management of weed seed bank	
i)	Diagnosis of weed seed bank and its threshold for IWM practice
ii)	Evaluation of insect seed predators for management of weed seed bank
iii)	Effects of cover plants cropping and flooding treatment on reducing buried weed seeds
iv)	Proper living mulch cultivation in soybean based on growth analysis of mulch plant barley
v)	Levee management to suppress weed invasion into fields
(2) Simulation model for population dynamics of noxious weed species and evaluation of economical benefit of IWM for farmers	
i)	Population dynamics of *Avena fatua* and *Lolium multiflorum* in winter cereals
ii)	Population dynamics of noxious broadleaved weeds in soybean
iii)	Population dynamics of *Murdannia keisak* in dry seeded rice
iv)	Population dynamics of *Aeschynomene indica* and *Beckmannia syzigachne* in paddy-upland rotation
v)	Population dynamics of *Echinochloa cruss-galli* in wet seeded rice
vi)	Evaluation of farmers' benefit and confirmation of economic optimum threshold for sustainable weed control
(3) Proof and establishment of integrated weed management in paddy-upland rotation	
i)	Cultural practices based IWM for winter cereals production
ii)	Developed weeding machine (so-called "hybrid-weeder") based IWM for soybean production
iii)	Cover plants cropping and winter irrigation based IWM for soybean production
iv)	Levee/field management based IWM for stable dry seeding rice culture
v)	Developed rice seeder based IWM for stable wet seeding rice culture

Development of IWM will be supported by basic studies especially on monitoring and diagnosis of weed seed bank, simulation of weed population dynamics, development of new weeding machine, utilization of cover plants, evaluation of cultural practices and confirmation of economic optimum threshold for sustainable weed control. Performance of insect seed predators will also be investigated, since seed-feeding by insects was expected to be a very important factor that influenced weed seed bank (Yamashita and Kobayashi, 2007, Ichihara *et al.* 2007).

These research targets are studied by collaboration of eight research institutes; National Agricultural Research Center (NARC), National Agricultural Research Center for Tohoku Region (NARCT), National Agricultural Research Center for Kyushu Okinawa Region (KONARC), Miyagi Pref. Furukawa Agricultural Experiment Station (MFAES), Fukui Pref. Agricultural Experiment Station (FAES), Shizuoka Pref. Research Institute of Agriculture and

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STATISTICAL ASSESSMENT ON THE GENETIC DIVERSITY OF *Echinochloa* spp. BY USING AFLP AND MORPHOLOGICAL ANALYSIS IN KOREA

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Abstract: *Echinochloa* spp. is causing serious problems in both of paddy and upland. In this study, for the systematic classification of *Echinochloa* spp, various morphological and molecular genetic methodologies were developed and applied on 5 species and 42 samples of *Echinochloa* spp. collected in Korea. Six phenotypic characteristics were selected as main factors, such as awn length, 1000 grains weight, plant height, leaf length, 1st husk rate, and grain shape. But the phenotypic classification could not provide efficient way of classification for ecotype differences from existing species, especially intermediate species. AFLP (Amplified Fragment Length Polymorphism) that was commonly used for plant classification was applied to classify *Echinochloa* collections. A total of 685 PCR products were generated using 11 primer pairs. Among the products, 352 polymorphic fragments were evaluated for their association with each of 25 phenotypic characteristics. Analysis using combination of AFLP products related to plant height and 1000 grains weight performed successfully in classification of collected *Echinochloa* spp.

Key words: *Echinochloa* spp., AFLP, genetic diversity.

Introduction

Echinochloa spp. is the most serious paddy weed in Korea. The species is classified to 3 to 6 sub-species according to its morphology and ecology. However, the taxonomical classification of *Echinochloa* spp. is not yet scientifically established due to its genetic mutation and hybridization. The first record of *Echinochloa* in Korea is written by Nakai (1911), and published by Mori (1921) and Jung (1927). Ehara and Abe (1952) classified *Echinochloa* spp. by root, shoot, leaf, inflorescence, and shape of spikelet. They insisted 6 characters as key factors of *Echinochloa* classification such as leaf shape, stem length, ear length, ear number, heading date and hair of leaf sheath. Yabuno (1975) classified *E. crus-galli* var. *oryzicola* as different species because *E. oryzicola* is triploid (2n = 4X =36) and others is hexaploid (2n = 6X = 54). Kim (1989) was classified as *E. crus-galli* var. *crus-galli*, *E. crus-galli* var. *praticola*, *E. oryzicola* by its shape of first husk. But it was difficult to classify between *E. crus-galli* var. *crus-galli* and *E. crus-galli* var. *praticola.*

The morphological characteristics are changed due to environmental adaptation and genetic mutation. In this reason, molecular-biological classification has been tried. Plant taxonomy have been developed recently using RFLP (Restriction Fragment Length Polymorphism) and AFLP (Amplified Fragment Length Polymorphism). Danquah *et al*. (2002) classified *Echinochloa* spp. using AFLP. *Echinochloa colona* was clearly classified and *E. crus-pavonis* classified into 4 groups. Kentaro *et al*. (2002) classified *E. crus-galli* var *praticola* and *E. oryzicola* by AFLP, using trn-c, trn-d primer and restrict enzyme *Dra* I. The present study used AFLP to clarify taxonomical position of collected *Echinochloa* spp. and evaluate their taxonomical relationship

Materials and Methods

Plant material and DNA isolation

For these experiments, a total of 42 *Echinochloa* spp. were collected from various regions (Table 1). These collected plants were cultivated in paddy field and the morphological traits were examined. Young leaves were collected at the beginning of May from a single plant of each variety and stored at -45℃. DNA was isolated from 1 g of fresh material by Murray and Thomson (1980) method.

Table 1. List of the 42 *Echinochloa* spp. accession used in this study.

Accession no.	*Description*	Collection sites
1	*E. crusgalli*	Gyeonggi
2	*E. crusgalli* var. *oryzicola*	Gyeonggi
3	*E. crusgalli* var. *frumentaceae*	Jeju
4	*E. crusgalli* var. *praticola*	Goyang
5	*E. crusgalli* var. *oryzicola*	Gyeonggi
6	*E. crusgalli* var. *echinata*	Jeju
7	*E. crusgalli* var. *frumentaceae*	Suwon
8	*E. crusgalli* var. *oryzicola*	Gyeonggi
9	*E. crusgalli* var. *oryzicola*	Gyeonggi
10	*E. crusgalli* var. *echinata*	Gyeonggi
11	*E. crusgalli*	Namyang
12	*E. crusgalli* var. *oryzicola*	Deokso
13	*E. crusgalli* var. *echinata*	Goyang
14	*E. crusgalli* var. *frumentaceae*	Suwon
15	*E. crusgalli*	Yeoncheon
16	*E. crusgalli* var. *oryzicola*	Goyang
17	*E. crusgalli* var. *echinata*	Goyang
18	*E. crusgalli*	Jeju
19	*E. crusgalli*	Gyeonggi
20	*E. crusgalli* var. *oryzicola*	Yangpyeong
21	*E. crusgalli* var. *echinata*	Seoul
22	*E. crusgalli* var. *frumentaceae*	Namyang
23	*E. crusgalli*	Deokso
24	*E. crusgalli* var. *oryzicola*	Deokso
25	*E. crusgalli*	Deokso
26	*E. crusgalli*	Deokso
27	*E. crusgalli*	Deokso
28	*E. crusgalli* var. *echinata*	Gyeonggi
29	*E. crusgalli*	Deokso
30	*E. crusgalli* var. echinata	Goyang
31	*E. crusgalli*	Deokso
32	*E. crusgalli* var. *oryzicola*	Yeoncheon
33	*E. crusgalli* var. echinata	Goyang
34	*E. crusgalli*	Deokso
35	*E. crusgalli* var. echinata	Goyang
36	*E. crusgalli*	Deokso
37	*E. crusgalli* var. *oryzicola*	Goyang
38	*E. crusgalli* var. *praticola*	Jeju
39	*E. crusgalli* var. echinata	Goyang
40	*E. crusgalli* var. echinata	Gyeonggi
41	*E. crusgalli* var. *praticola*	Jeju
42	*E. crusgalli*	Deokso

AFLP assays

The AFLP analysis was performed according to Vos *et al*. (1995) with some modifications. The adaptor oligonucleotide sequences were AFPA-1 (5'-CTCGTAGAC TGCGT-ACATGCA-3') and AFPA-2 (5'-TGTACGCAGTCTAC-3') for PstI and AFMA-1(5'-CATGCGACGATGAGTCCTGAG-3') and AFMA-2(5'-TACT CAGGACTCATCG- TCG-3') for MseI. Preamplification was conducted with 5 pmole each AFPP-0 (5'-CGTAGACTGCGTACATGCAG-3') and AFMP-A (5'-GCGACGA-TGAGTCCTG-AGTAAA-3') primers, 6µl of 1:10 diluted ligated DNA, 1x reaction buffer, 1.5 mM $MgCl_2$, 0.2 mM of each dNTP, and 1U of Taq polymerase (Nurotics, Korea) in a 30ul reaction volume. The PCR condition was 95°C for 1 min, 25 cycles of 95°C for 20s, 56°C for 30 sec, and 72°C for 1 min, with a final extension step of 72°C for 10 min. Two µl of 1:20 diluted pre-amplified DNA was selectively amplified using 5pmole of each of the two (PstI ends) and three (MseI ends) selective nucleotides with 0.5 U of Taq polymerase in a 20ul reaction volume.

Fluorescent AFLPs were detected using an ABI 3100 Genetic Analyzer (Applied Biosystems, California, USA). The AFLP fingerprinting patters were analyzed using the software package Genographer (Benham *et al*. 1999). The AFLPs were named based on the primer combinations used for specific amplifications and the band size. All PCR amplifications were performed using a MJ PTC-100 Thermal Cycler (Waltham, Massachusetts, USA) and the sequences of the primers used in this study.

Data analysis

The presence and absence ratings of interpretable bands of the amplification products were computed. We estimated 11 marker information indices for AFLP. Similarity among tested genotypes was computed as the simple matching coefficient of genetic distance. The cluster and principal coordinate analyses were performed using NTSYS PC version 2.0 software (Rohlf, 1998).

Result and Discussion

Morphological classification of Echinochloa spp.

Total 25 morphological traits were examined. In these traits, we focused large CV traits, because those traits changed a lot within collected plants (Table 2). Total 25 morphological traits could be contracted by Principle Components Analysis into 5 clusters (Figure 1). After these analysis, 5 morphological traits (T6, T7, T13, T24, T25) were selected, and the UPGMA cluster dendrogram was developed (Figure 2). Some collected plants could not be classified as the correct species (red circle), suggesting that morphological classification has its limitations due to the presence of intermediate ecotypes of collected plants.

Amplified DNA polymorphism of Echinochloa spp.

A total of 11 primer combinations were used for AFLP. The size of amplified products ranged from 70 bp to 750 bp. Using EcoR1-AAg and MseI-AcG combination, 74 fragments were obtained, and 29 fragments (39%) showed polymorphism. These polymorphic fragments marked their band strength (Table 3).

Table 2. Characters and their descriptions morphometric analyses of forty two *Echinochloa* spp. collected in Korea.

Character ID	Descriptions	CV*
T1	Growth stage at 30-day seedling barnyard grass (leaf number)	21.2
T2	Plant height at 30-day seedling barnyard grass (Cm)	25.7
T3	Average Tiller number (ea)	39.3
T4	Stem color (1 = at red colored, 0 = at green)	77.9
T5	Awn (1 = at present, 0 = at absent)	153.7
T6	Awn length (Cm)	175.3
T7	1000 grains weight (g)	29.9
T8	Spike length (Cm)	25.7
T9	Spike width (Cm)	139.5
T10	Spike weight (g)	142.9
T11	Spike shape (=width/length)	51.6
T12	Heading date (Julian day)	6.5
T13	Plant height (Cm)	24.3
T14	Leaf width (mm)	38.9
T15	Erectness (of stems, °)	45.8
T16	Total nitrogen contents (%/plant)	17.4
T17	Plant mineral (K) contents (%)	21.8
T18	Plant mineral (Ca) contents (%)	23.2
T19	Plant mineral (Mg) contents (%)	26.2
T20	Plant mineral (Na) contents (%)	153.7
T21	Transpiration rate	19.2
T22	Stomatal conductance	25.6
T23	Photosynthetic rate	16.3
T24	1st husk rate (1st husk/grain length)	11.4
T25	Grain shape (grain length/grain width)	13.7

CV* : Coefficient of Variation

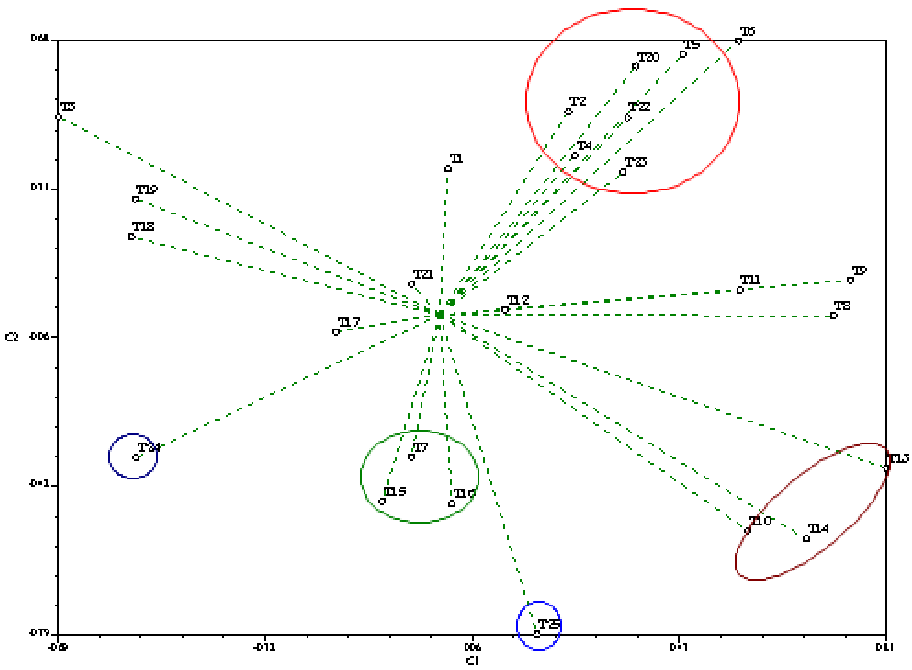

Figure 1. Scatter diagram of 25 morphological characters of *Echinochloa* spp. from a principal component analysis. The proportion of total variance along the first two axes was 20.75 and 18.27% respectively.

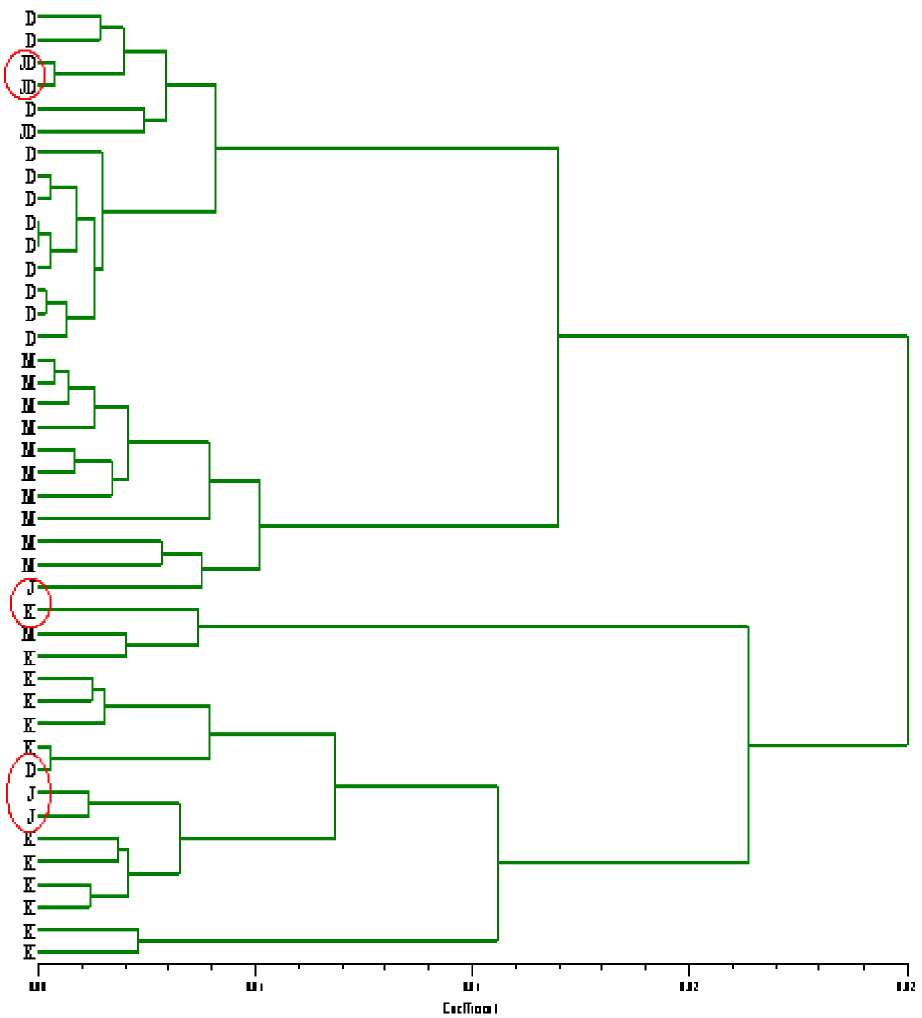

Figure 2. UPGMA cluster dendrogram of *Echinochloa* spp. using 5 morphological characters (T6+7+13+24+25). **D**: *Echinochloa crus-galli*, **J**: *E. crus-galli* var. *frumentacea*, **K**: *E. crus-galli* var. *oryzicola*, **JD**: *E. crus-galli* var. *praticola*, **M**: *E. crus-galli* var. *echinata*

Table 3. AFLP markers of 42 collected *Echinochloa* species obtained from 11 primer combinations and there marker information.

Primer combination	Polymorphic Strength			Information‡	
	S	A	B	TL	NP
E†-AAG + M†-ACG	8	12	9	29	74
E-AAG + M-AGT	14	13	29	56	76
E-AGA + M-ACG	3	6	19	28	69
E-AGA + M-AGT	2	3	16	21	52
E-AGA + M-ATC	5	13	18	36	71
P†-AAG + M-ACG	9	5	11	25	66
P-AAG + M-AGT	5	8	12	25	65
P-AAG + M-ATC	14	8	10	32	98
P-AGA + M-ACG	3	7	10	20	51
P-AGA + M-AGT	2	5	29	36	71
P-AGA + M-ATC	10	11	23	44	78
Total	75	91	186	352	771

†E : EcoR I, M : Mse I, P : Pst I, ‡TL : Total number of loci detected per assay unit, NP : Number of polymorphic bands per assay unit

A cluster analysis was performed with UPGMA using the mean Jaccard similarity (Figure 3). It is apparent that considerable genetic diversity exists in the populations of *Echinochloa* spp. and that there is introgression of this species into material. As found in other plant species, AFLPs proved highly informative in assessing diversity in *Echinochloa* spp. from geographic to field levels. Because of the importance of the *Echinochloa* spp., and *E. crus-galli* in particular, rice production would be appropriate to conduct a study on genetic diversity together with full morphological descriptors on a much wider selection of materials to elucidate the extent of the diversity in the *Echinochloa* spp. This would provide a sound basis from which appropriate management strategies could be developed.

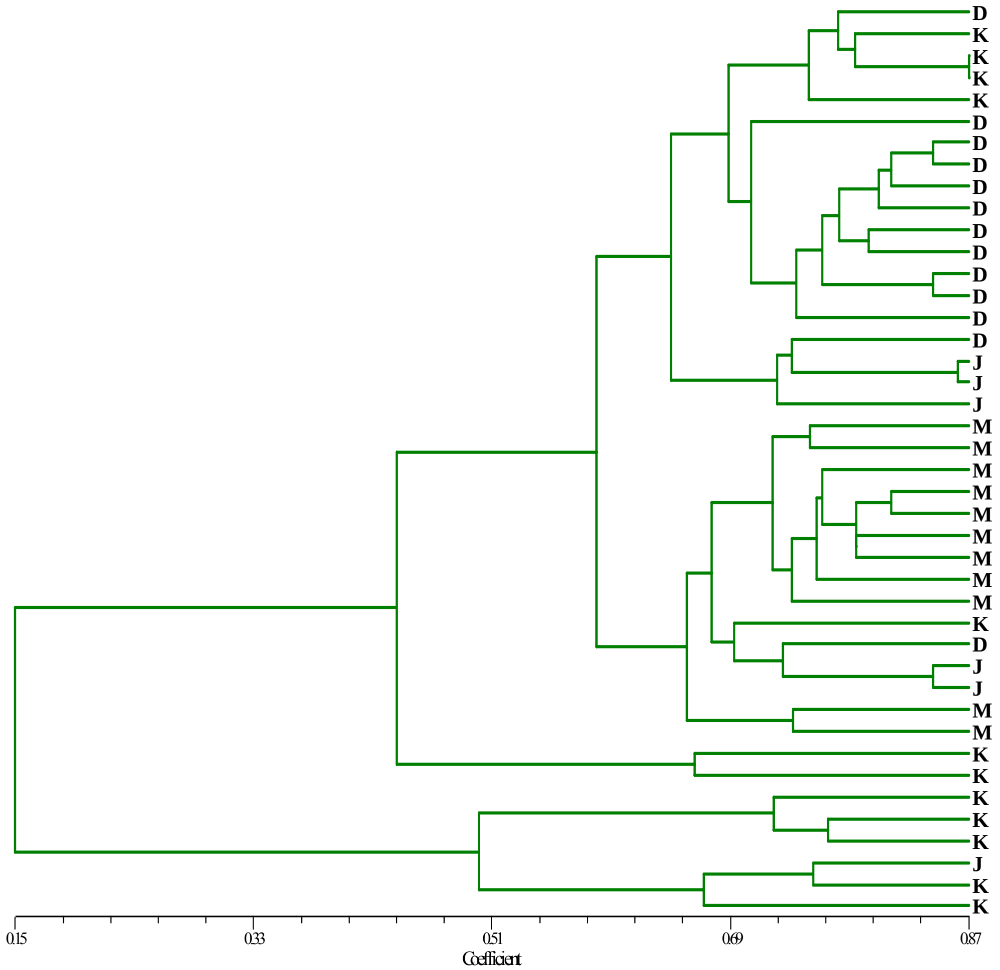

Figure 3. Forty-three collected *Echinochloa* species clustered by un-weighted pair-group method, arithmetic average (UPGMA) procedure applied to 352 polymorphic AFLP markers.

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DETERMINATION OF COMPETITIVE ABILITY OF DIFFERENT RICE VARIETIES (*Oryza sativa*) AGAINST *Ludwigia octovalvis* (Jacq.) Raven.

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Abstract: Weeds have continued to be one of the most important factors reducing the rice yields mainly due to competition. The degree of weed competition varies among weed species, growth stage and crop age. *Ludwigia octovalvis* (Jacq.) Raven. is an abundant weed than others in the low country wet zone of Sri Lanka. Hence, this study was carried out to investigate the Ludwigia- rice competition using three rice varieties *i.e.* Bg 250, Bg 300 and Bw 267-3 representing 2.5, 3 and 3.5 months age groups. The experiment was conducted at the Rice Research and Development Centre, Bombuwela, Sri Lanka in 2005. Seven different proportions of each rice variety and Ludwigia were used as treatments, in a factorial arrangement in RCBD with three replicates. Rice plant dry weight and LAI decreased significantly with increase in Ludwigia density up to 40 plants m^{-2}. However, further significant reduction in LAI and crop dry weight was not observed with higher Ludwigia densities. Rice yield decreased significantly with increasing Ludwigia population densities, which amounted to 29% and 30% reduction in Bg 250 and Bg 300, respectively. The yield of Bw 267-3 was not affected when Ludwigia density was increased from 0 to 25 plants m^{-2}. The highest weed density (85 Ludwigia plants m^{-2}) resulted in 60%, 80% and 68.8% reduction of yield in Bg 250, Bg 300 Bw 267-3, respectively. Among the tested rice varieties, Bw 267-3 was the most competitive variety, which maintained a land equivalent ratio of > 1, up to 55 Ludwigia plants m^{-2}.

Key words: Weed competition, *Oryza sativa*, *Ludwigia octovalvis*

Introduction

Rice is vital as the main food crop in Sri Lanka and will continue to be our primary source of food for the next centuries. Hence the rice production should be increased to accomplish the increasing populations' needs via overcoming the confines. On this mission weeds have been identified as the most critical biotic factor that limits the rice production (De Data, 1981). This is due mainly, weeds are nourished by the same nutrients and environmental elements needed by the crops. The limited supply of these vital elements reluctantly leads to a competition. The crop-weed competition at its severe levels, above the critical threshold level leads to reduce the crop yield by 30-40% or more. Thus it causes a tremendous financial loss to farmers. It was recorded that yield losses from all biotic and abiotic constraints was above 2t/ha which is about 54% of current farmers' yield (Herath Banda *et al.* 1998)

The degree of competition is different among different species and within the growth stage as well (Mercado, 1979). During the cropping period there is a specific duration of crop-weed competition which is known as the critical period. The duration of this critical period varies in different crops and is determined by allowing the crop to compete with weeds. The same scenario furthermore could be seen in rice, as the rice-weed competition differs according to the varieties as well as the age groups. Generally morphological characters of high yielding varieties negatively correlated with the competitive ability against weeds. However, newly improved rice varieties have shown some capabilities to suppress weeds during their vegetative phase.

Similarly, crop-weed competition also differs based on the weed species and its biology. Hence competition studies are conducted to determine the potential damage of weeds, the period when a weed is capable of causing damage to the crop, the competitive power of the weeds against the crop and vise versa.

Weed species are highly varied according to the location under wet-seeded rice cultivation in Sri Lanka. Obviously grasses are dominant in dry zone whereas broad-leaved and sedges are dominant in wet zone (Chandrasena, 1988). Of all the broad-leaved weeds, *Ludwigia octovalvis* syn. *Ludwigia hyssopifolia* (G.Don) Exell. was reported as the most wide spread and abundant species in the low country wet zone of Sri Lanka (Chandrasena,1988). However, there is a scarcity of studies to describe the limiting resources and threshold level of its competition. Thus, this study was conducted to quantify the rice - Ludwigia competition.

Materials and Methods

The experiment was carried out in lowland field at the Regional Rice Research and Development Centre, Bombuwela, Sri Lanka during 2005/2006 Maha season. The experiment site was located in low county wet zone. Its annual rainfall ranges from 2500 to 3250 mm. The daily mean temperature ranges from 24°C to 31°C.The soil of the experimental location was containing mixture of low humic Glay and red yellow podsolic soil. Three different rice (*Oryza sativa*) varieties *i.e.* Bg 250, Bg 300 and Bw 267-3 representing 2.5, 3 and 3.5 month age groups respectively were used for the experiment. Seven different densities of Ludwigia were used to get the varying proportions in weed and crop mixture as treatments for each rice variety (Table 1). The total density of weed and crop was maintained constant, at each density level of weed. Treatments were arranged as two factor factorial arrangement in a Randomized Complete Block Design with three replicates.

Table 1. Proportions of Ludwigia and rice plants in crop weed mixtures

Ludwigia (No./m^2)	Rice (No./m^2)	Total density (No./m^2)
0	100	100
25	75	100
40	60	100
55	45	100
70	30	100
85	15	100
100	0	100

The experimental area was ploughed twice followed by a single harrowing. The plots, each in size of 1m^2 and surrounded with ditches of 45 cm wide, were then puddled and finally leveled before planting. The fertilizer mixture consisting of 10 kg N, 30 kg P_2O_5 and 15 kg K_2O ha^{-1} as Urea, Triple Supper Phosphate and Muriate of Potash were applied as the basal application for all three varieties, and 20 kg N and 22 kg K_2O ha^{-1} were top dressed at 3 and 6 weeks after planting (WAP) for Bg 250 and Bg 300 varieties. Nitrogen was top dressed at 15 kg ha^{-1} at 2, 5 and 7 WAP and 20 kg K_2O ha^{-1} was top dressed at 2 and 5 WAP for the Bw 267-3 variety. Fourteen-day old rice seedlings together with Ludwigia seedlings at a uniform growth stage (2-3 leaf stage) were randomly transplanted according to the correct proportions of the crop and weed.

Rice plant height was recorded from randomly selected ten plants from each plot at 2, 4, and 6 WAP. An area of 50 cm *x* 50 cm was left as harvested area and other observations were made from the rest of the plot area. Rice and Ludwigia were sampled at 6 WAP by using a 30 cm *x* 30 cm quadrat to measure the crop and weed dry weights, and leaf area. The leaf area index (LAI) and the land equivalent ratio (LER) based on crop and weed dry weights were calculated by using following formula. The rice grain yield was recorded at 14% moisture content.

$$\text{LAI} = \frac{\text{Leaf area}}{\text{Land area}}$$

$$\text{LER} = \frac{\text{crop dry weight}}{\text{(crop+weed) dry weights}} + \frac{\text{weed dry weight}}{\text{(crop+weed) dry weights}}$$

Results and Discussion

A significant interaction effect ($p<0.05$) was observed between rice varieties and weed densities, in plant dry weight and LAI of rice at 6 WAP. In each variety, the rice plant dry weight and LAI decreased with increasing Ludwigia density (Table 2) up to 40 plants of Ludwigia m^{-2}. There was no significant difference either in crop dry weight or in LAI with further increase in the density of the weed. A rapid decrease in LAI was observed in variety Bg 300 than other two varieties (Table3) with the increase in weed densities. Increasing the Ludwigia plant density from 0 to 25 plants m^{-2} decreased the LAI by 56, 30 and 24% in Bg 300, Bw 267-3 and Bg 250 varieties, respectively. Rice plant height in each variety was not affected by different densities of Ludwigia at 2, 4 and 6 WAP (data not shown).

Table 2. Effect of Ludwigia density on plant dry weight of rice varieties.

Ludwigia (No./m^2)	Rice plant dry weight (t/ha)		
	Bg 250	Bg 300	Bw 267/3
0	312.86 ab	400.05 a	221.89 a
25	189.79 b	179.79 b	147.89 b
40	143.01 c	81.69 c	91.68 c
55	68.80 d	80.77 c	66.96 cd
70	79.53 d	43.09 c	38.29 d
85	40.96 cd	35.67 c	34.43 d
CV (%)	17.46	23.43	26.18

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05)

Table 3. Effect of Ludwigia density on LAI of rice

Ludwigia (No./m^2)	Rice plant dry weight (t/ha)		
	Bg 250	Bg 300	Bw 267/3
0	3.00 a	4.45 a	3.31 a
25	2.29 ab	1.95 b	2.32 ab
40	1.93 c	1.23 c	1.47 c
55	1.07 cd	1.02 cd	1.09 cd
70	1.01 cd	0.87 cd	0.43 d
85	0.55 d	0.50 d	0.41 d
CV (%)	33.3	22.66	36.09

Within a column, means followed by the same letter are not significantly different by the DMRT (p=0.05)

A significant interaction effect ($p<0.05$) was also observed between the rice varieties and weed densities, on crop yield. In each rice variety tested, the rice yield decreased with increasing weed density levels (Table 4). The yield was reduced by 29% and 30% in Bg 250 and Bg 300, respectively, at a weed density of 25 Ludwigia plants m^{-2}, however the yield of Bw 267-3 was not significantly different ($p>0.05$) from its pure crop stand. At higher density levels of the weed, the yield loss in Bw 267-3 was lower than that in other two varieties. The highest weed density (85 Ludwigia plants m^{-2}) resulted in 61%, 80% and 68.8% yield reductions in Bg 250, Bg 300 and Bw 267/3, respectively.

Table 4. Effect of Ludwigia density on yield of rice

Ludwigia (No./m^2)	Rice plant dry weight (t/ha)		
	Bg 250	Bg 300	Bw 267/3
0	3.08 a	3.58 a	2.50 a
25	22.18 b	2.48 b	2.76 a
40	1.64 c	2.38 b	1.66 b
55	1.72 c	1.24 c	1.47 b
70	1.40 cd	1.12 c	1.29 bc
85	1.20 d	0.71 c	0.78 c
CV (%)	10.59	16.82	19.26

Within a column, means followed by the same letter are not significantly different by the DMRT ($p=0.05$)

The land equivalent ratio (LER) in Bg 250 and Bg 300 was < 1, when the weed density was above 25 plants m^{-2} (Table 4) indicating their lower competitive ability against the higher densities of Ludwigia. In contrast, the rice variety Bw 267-3 showed an LER of 1 or > 1 up to (55 Ludwigia plants m^{-2}) suggesting its higher competitive ability against the weed. Zimdahal (1981) also reported similar findings on LER in the competitive crops.

Table 5. The Land Equivalent Ratio (LER)

Ludwigia (No./m^2)	Rice plant dry weight (t/ha)		
	Bg 250	Bg 300	Bw 267/3
25	1.10	1.09	1.46
40	0.94	0.86	1.01
55	0.85	0.98	1.00
70	0.61	1.02	0.77
85	1.22	0.90	0.77

Conclusions

Ludwigia should be controlled when it reaches density levels of 25 plants m^{-2} in field cultivated to Bg250 and Bg300 varieties under the low country wet zone climatic conditions. However, Bw267-3 was able to withstand with up to 30 Ludwigia plants m^{-2} without affecting its yield. Therefore, among the three rice varieties tested, Bw 267-3 was the most competitive maintaining an LER of > 1 one up to 55 plants of Ludwigia m^{-2}

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HERBICIDAL EFFICACY OF BENZOFENAP AGAINST SULFONYLUREA-RESISTANT BIOTYPES OF *Sagittaria trifolia* L.

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Abstract: Sulfonylurea-resistant (SUR) biotypes of the *Sagittaria trifolia* L. (arrowhead weed) has been reported from the paddy fields of Japan. It has caused serious problems in Japan as sulfonylureas have been used as the most effective herbicide among soil applied herbicides. In this study, the efficacy of the other soil-applied herbicides was investigated against the SUR biotypes, and benzofenap was found as a good substitute for sulfonylureas.

Key words: benzofenap, sulfonylurea-resistance, *Sagittaria trifolia*L.

Introduction

Sagittaria trifolia L. (arrowhead weed) is a paddy weed that has sagittate leaf blades and propagates mainly by tubers, with severe damage to rice in Japanese paddy fields (Yamakawa and Itoh, 2004). This weed has been controlled mainly by soil-applied sulfonylurea herbicides (Itoh, 2005). However, sulfonylurea-resistant (SUR) biotypes of the *Sagittaria* were found in several paddy fields at Akita Prefecture (Uchino and Watanabe, 2002). The occurrence of SUR biotypes is now increasing and has caused serious problems in the paddy fields of Japan. In this report, the effect of the other herbicides investigated on their efficacy to control SUR *Sagittaria* with a view to find alternatives for sulfonylureas.

Materials and Methods

Plant materials

The tubers of *Sagittaria trifolia* L. were dug out in 2003 in a paddy field at Iwanuma City, Miyagi Prefecture, where severe infestation of this weed had been found, and stored at 5℃ in the moist soil. The tubers collected at Ushiku City at Ibaraki Prefecture, were kindly provided by the Japan Association for Advancement of Phyto-regulators (JAPR) and used as the representative of susceptible biotypes.

Response to sulfonylurea application

The tubers we replanted in 1/50 m² Wagner pots in a growth chamber, and bensulfuron-methyl was applied to the pots at the primary or second leaf stage of the weed.

Efficacy of several herbicides on SUR biotypes

Quadrats (PVC sheet 3 m × 3 m) were set up in the paddy field at Iwanuma City after rice was transplanted. Several herbicides were applied to the quadrates at 15th day after transplanting (DAT) rice, and efficacy on SUR *Sagittaria* was investigated at 36th DAT

Results and Discussion

Application of bensulfuron-methyl did not control the biotype of Iwanuma even at 600 g a.i./ha, which is eight times of the recommended dose in paddy fields (Plate 1). This shows that the Iwanuma biotype has high resistance to a sulfonylurea. Among several herbicides, benzofenap showed high efficacy on the SUR biotype of Iwanuma. More than 1000 g a.i./ha benzofenap strongly inhibited the growth of SUR *Sagittaria* in the paddy field (Plate 2). Ther herbicide could be a useful tool to control SUR *Sagittaria* in rice cultivation.

Plate 1. Effect of bensulfuron-methyl on the biotypes of *Sagittaria trifolia* The pots in the front row are the Ushiku biotype and those in the back row are Iwanuma biotype. The doses of bensulfuron-methyl are 2, 400, 600, 150, 75, 37.5 and 0 g a.i./ha from left to right. The recommended dose for paddy fields is 75 g a.i./ha. The photograph was taken at 57 days after application of the herbicide.

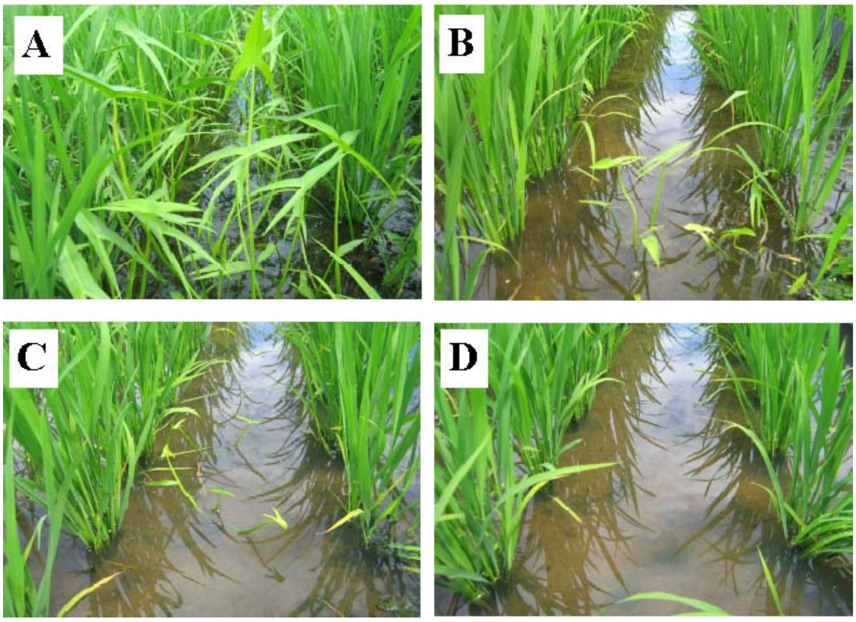

Plate 2. Effect of benzofenap on *Sagittaria trifolia* in a paddy field at Iwanuma. Benzofenap was applied at 500 (A), 1,000 (B), 1,500 (C) and 2,000 (D) g a.i./ha.

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NON CHEMICAL MANAGEMENT OF WEEDS IN SELECTED ARABLE CROPS

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Abstract: Field experiments were conducted in certified organic farms in Canterbury, New Zealand over several years to test new weed control options and to optimize farmers' practices. The study found that timing was important in getting the best results with tine weeding. In both crops, a pre-emergence pass of spring tine harrow controlled weed seedlings in the soil and small weeds. Post-emergence tine weeding was best at the two- to three-leaf stage of wheat or peas. A third pass later in the season was not always effective and could damage the crop. In linseed, experiments conducted over two seasons did not show yield increases from tine weeding, even though reductions in weed density were measured. Flame or steam weeding damaged linseed and was not considered an option. Sheep grazing in linseed was tested at two different growth stages and two intensities and caused reductions in both weed and linseed populations and biomass. However, weeds took advantage of the opened canopy and grew faster. None of the grazing treatments improved linseed yield.

Key words: Organic weed control, mechanical weed control, tine weeding, thermal weeding, grazing

Introduction

In organic arable crops in New Zealand, weed control during the season relies mainly on tine weeding with spring-tine harrows. However, the impact of tine weeding on weeds and crops is not well defined. Information about the response of crops and weeds to harrows at different stages of growth can be used to optimise their use for mechanical weed control in organic systems or even as part of integrated weed management in conventional systems. Very few studies are reported on mechanical weed control in New Zealand (Stiefel and Popay, 1990). Reddiex *et al.* (2001) compared tine weeder, spoon weeder and inter-row hoe in four crops and found that the performance of these tools depended on the crop. Overseas reports highlight the need for more research on timing and frequency of harrowing for mechanical weed management (Rasmussen, 1991; Welsh *et al.*, 1997). Barberi *et al*. (1999) studied the effect of finger harrowing in durum wheat (*Triticum durum* L.) and found that it gave comparable yield with chemical control only when weed pressure was low. This study was undertaken to compare different times and numbers of tine weeding in commercially grown organic wheat (*T. aestivum* L.), process peas (*Pisum sativum* L.) and linseed *(Linum usitatissimum* L.*)*. Moreover, the study examined thermal weeding and sheep grazing as potential methods for weed management in linseed.

Materials and Methods

Five field experiments in wheat, six in pea crops and four in linseed were carried out between 2001 and 2005 on certified organic farms in mid-Canterbury, New Zealand. Experiments received the same crop management applied by the farmers. Details of experimental design, measurements and analysis are described in Dastgheib (2003). In general, plot sizes were between 10 to 20 m long and one pass of tine weeder wide. Tine weeder used in the experiments consisted of four rows of spring tines on a metal frame and was 6-m wide. Tine weeding treatments were designed according to the crop-weed situation as outlined in the Results section. Early post-emergence weeding was conducted at 2-3 leaf stage and late post-emergence at the 5-leaf stage of either wheat or peas.

For linseed, tine weeding treatments were imposed at pre-emergence, cotyledon stage, early post-emergence (linseed 5-7 cm, with 4 leaf nodes), late post-emergence (linseed 10-15 cm, with >7 leaf nodes) and combinations of these. Flame weeder used LPG fuel and delivered flame close to the soil surface through burners under a metal panel hood about 130-cm wide. Steam weeder was a local prototype design and used diesel to generate steam which was delivered to plants through nozzles under a metal panel hood about 180 cm wide. Flame or steam weeders were mounted on a tractor and driven at a speed of 5 km/h. In the grazing experiment on linseed, two grazing times were compared: early grazing at approximately 12-cm crop height and late grazing at approximately 35-cm crop height. At each time two grazing intensities, namely lax grazing (4 sheep per plot) and hard grazing (8 sheep per plot) were compared. Plots (12 m x 12 m) were blocked off by electric flexi-fence to contain the sheep during a 20 hour grazing period.

All experiments were laid out in randomised complete blocks with three or four replicates. All data were analysed by Microsoft Excel through ANOVA and where the F test was significant, $LSD_{0.05}$ values were calculated for mean comparisons.

Results

Wheat

<u>*Effect of tine on weeds*</u>: Despite variability in weed population and crop vigour, most experiments showed similar results from tine weeding treatments. As an example, the site of the main experiment in the first year was very weedy with an average of 444 weeds/m^2 in the control plots. Pre-emergence tine weeding alone caused 66% reduction in weed density, while the combination of pre-emergence + late post-emergence was the most effective treatment giving a reduction of 84% (Table 1). The other two experiments in the first year showed that early post-emergence (2-3 leaf-stage of wheat) tine weeding was more effective in weed control than late (5 leaf stage) post-emergence.

Table 1. Weed measurements in different tine weeding treatments in the first two years.

	First year		Second year
Tine weeding	No. weeds /m^2*	Weed DM**	Weed DM**
Nil	444	22.9	52.4
Pre	149	8.9	7.25
Early post	210	13.1	13.3
Late post	188	7.8	23.1
Pre + Early	--	--	3.5
Pre + Late	71	2.5	9.8
Early + Late	--	--	21.2
LSD (p=0.05)	283	16.8	15.4

* measured eight weeks after the final weeding treatment, ** Weed dry matter (g/m^2) measured at the end of the season

In the second year, significant reductions in weed biomass were measured by all tine weeding treatments (Table 1) with the greatest reduction of 93% in plots receiving both pre-emergence and early post-emergence tine weeding. In most experiments, late post-emergence tine weeding did not result in weed control on its own, but when performed on plots with a previous tine weeding, it gave a significant reduction in weed density.

<u>*Growth and yield of wheat crops*</u>: Depending on site and year, post-emergence tine weeding at 2-3 leaf stage caused between 12 to 18% mortality in wheat plants, while tine weeding at 5-leaf stage showed only 5% crop mortality. In none of the experiments carried out during these years did grain yield show significant differences between treatments. Only in one

experiment, there was a yield increase of 1.7 t/ha in pre-emergence + early post-emergence tine weeding compared to the control.

Peas

<u>*Effect of tine on weeds*</u>: Pooled results from three trials carried out in the first year are presented in Table 2. A single tine weeding at early post-emergence (2-leaf stage) was the most successful treatment and gave more than 70% reduction in weed numbers. Trials in the second year showed that the best treatment was two passes of tine, pre-emergence + early post at 3-leaf stage. Tine weeding at 5-leaf stage was not an effective treatment.

Table 2. Per cent weed control, weed dry matter (DM, g/m^2) and pea yield (t/ha) in tine weeding treatments.

	First year[1]		Third year	
Tine weeding	% weed control[2]	Pea yield	Weed DM	Pea yield
Nil	0.0	4.19	311	1.692
Pre-emergence	40.2	6.47	153	2.675
2-leaf	70.5	5.89	62	3.195
3-leaf	--	--	140	2.006
2 + 3-leaf	--	--	13	3.161
5-leaf	53.2	6.10	130	1.952
Pre + 2-leaf	--	--	31	3.293
Pre + 3-leaf	--	--	93	2.431
Pre + 5-leaf	58.3	5.45	82	3.001
LSD (p=0.05)	15.4	1.56	73.7	0.893

[1] Values are the mean of three sites. [2] Weed control percentages are based on reductions in weed density relative to the nil treatment.

Tine weeding treatments in the third year looked into timing in more detail. One pass of tine weeding pre-emergence caused a significant reduction of 57% in weed density while post-emergence tine at 2-leaf stage gave 88% and at 3-leaf stage gave 74% reduction in weed numbers (data not presented). The greatest reduction in weed density (98%) and weed DM (96%) were obtained by two passes of tine at 2- and 3-leaf stage (Table 2).

<u>*Growth and yield of pea crops*</u>: Most tine weeding treatments gave significant increases in pea yield. The greatest yield increase of 1.6 t/ha (95%) was obtained in plots with a pre-emergence and a post-emergence tine weeding at 2-leaf stage (Table 2). Pea yield was not significantly different from the control in treatments with one pass of tine at 3- or 5-leaf stage or two passes at pre-emergence and 3-leaf stage.

Linseed

<u>*Mechanical weeding experiments*</u>: Results from both sites were similar. Weed density was significantly reduced by all tine weeding treatments with the largest reduction (85%) in plots receiving two passes of tines at cotyledon and early post-emergence stage (Table 3). Moreover, all treatments showed significant reductions in linseed populations with the highest reduction of 50% in cotyledon + early post-emergence tine weeding.

In the other experiment, plots which received tine weeding at cotyledon + early post-emergence stage and those with three passes of tine weeder were behind in maturity compared to the control plots. No significant difference in yield was observed between treatments at either of these experiments (data not presented). However, there was a trend for tine weeding at cotyledon + early post-emergence stage of linseed to improve yield (10% increase at one site and 32% increase at another).

Table 3. Linseed and weed density (plants/m^2) and weed dry matter (DM, g/m^2) in tine weeding treatments in the first year

Tine weeding	Weed density	Weed DM	Linseed population
Control	175	145	737
Cotyledon (C)	92	76	536
Early	92	132	512
C + Early	33	38	370
C + Late	37	52	425
Early + Late	36	106	457
LSD (p=0.05)	30.6	60.1	120.4

Thermal weeding experiment

The trial site had a low weed population and the main weed species was fathen (*Chenopodium album* L.). Visual assessments did not show significant differences in weed control between treatments. At the same time, linseed showed great sensitivity to heat at its early growth stage and post-emergence flame or steam left only a few surviving plants. Even pre-emergence passes of flame or steam weeder reduced linseed population by 16-24% (data not presented).

Grazing experiment

Sheep did not show a particular preference and grazed both weeds and linseed. One day after the first grazing, the biomass of weeds left behind was significantly lower in grazed plots than in the un-grazed ones, irrespective of the grazing level (data not presented). The second grazing did not result in significant reductions in weed dry weight.

Linseed height, population and biomass were significantly reduced by sheep grazing irrespective of timing or level (data not presented). The damage was in a haphazard manner causing the grazed plots to look patchy. Linseed showed signs of recovery from sheep damage but remained shorter, less vigorous with more open spaces in the canopy compared to the control plots. The dry weight of linseed was significantly reduced in all grazing treatments, but more so for the first grazing (Figure 1).

Weeds took advantage of the open canopy produced by sheep and grew faster, especially in the first grazing treatments (Figure 1). Weeds in the control plots were shaded by linseed and were less vigorous than weeds in grazed plots. Harvest assessment showed non-significant yield reductions due to grazing in both levels and times (data not presented). The second grazing, when the crop was 35-cm tall was more damaging than the first grazing and caused yield reductions in excess of 17%.

Discussion

For wheat, the results over three years showed that early passes of tine weeding control weeds better but also cause more crop damage. To compensate for the loss, higher sowing rates should be considered. However, a good wheat crop is able to tolerate some loss without a yield penalty. Pre-emergence tine weeding usually control some weeds but new flushes of weeds may appear later. When pre-emergence tine was followed by an early post tine at 3-leaf stage, the low weed density was maintained throughout the season.

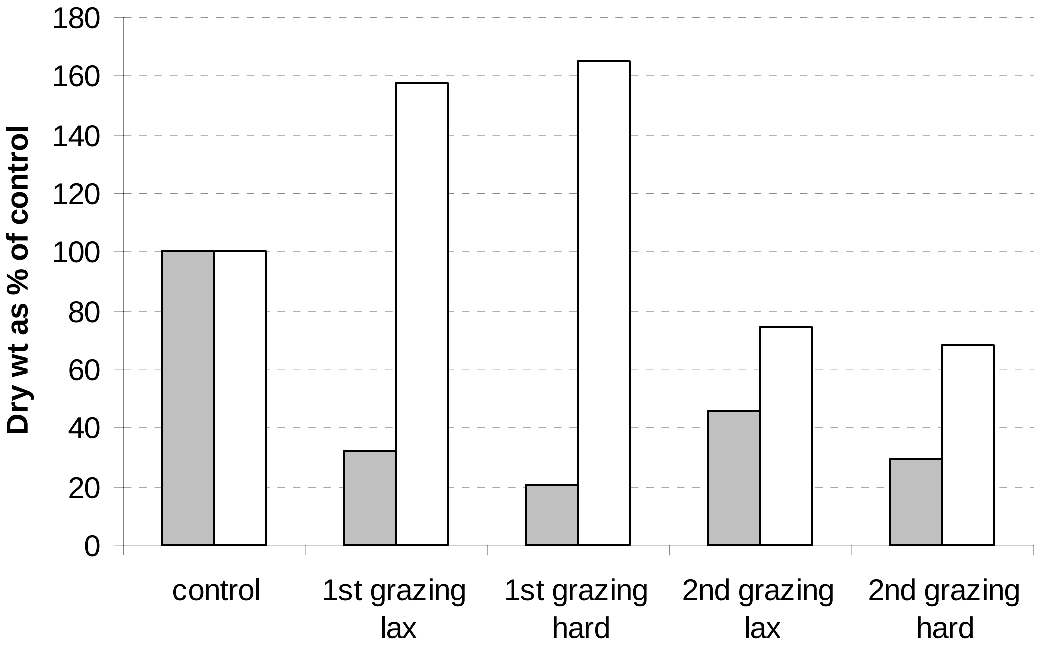

Figure 1. Dry weights of linseed (grey bars) and weeds (white bars) as percentages of non-grazed control. $LSD_{0.05}$ values for weeds 49.8 and for linseed 15.5.

There were significant increases in wheat growth due to early tine weeding when measured in mid-season (Dastgheib, 2003). However, the results from all experiments showed that in most cases tine weeding did not improve grain yield. This is mainly due to the excellent ability of wheat to compete with annual weeds once the canopy closure starts. Previously, Stiefel and Popay (1990) and Welsh *et al.* (1997) reported no yield increases in wheat following one or two passes of tine weeding.

For peas, all experiments showed that no benefit was obtained when tine weeding was delayed until 5-leaf stage of peas. Tine weeding is not useful at this stage mainly because it allows weeds to grow during the early period of pea growth. Although the critical period of weed competition in peas is not well defined, most research suggest early removal of weeds is critical for optimal yield (Harker *et al.* 2001). Moreover, passing through the crop at 5-leaf stage is likely to damage the plants.

In most experiments, significant yield increases were obtained with certain tine weeding treatments. The best treatments, both for reduction in weed biomass and improving pea yield, were two passes of tine either at pre-emergence and 2-leaf stage, or at both 2- and 3-leaf stage of the crop. Previous reports by Stiefel and Popay (1990) and Reddiex *et al.* (2001) found no yield increase in peas from tine weeding. This was mainly due to low weed populations in those studies. Apparently, peas are more vulnerable than wheat to weed competition as they showed significant yield response to tine weeding treatments.

The study also focused on three methods of weed management in linseed namely: mechanical, thermal and sheep grazing. The results showed that two tine weeding passes at the cotyledon and early post-emergence stage of linseed gave effective reduction in weed density but did not give significant yield increase. Another study reported 22% increase in linseed yield as a result of tine weeding (Reddiex *et al.* 2001). In experiments reported here, weed pressure was not very high and linseed competition suppressed their growth. Linseed seems to be more sensitive than wheat or peas to tine weeding as it suffered between 27 to 31% mortality after one pass of tines at cotyledon or early post-emergence stage, respectively (Table 3).

Both flame and steam weeding were lethal to linseed when performed early post-emergence. At pre-emergence or later growth stages (height 15-20 cm), linseed showed more tolerance to thermal weeding with better survival rates but its growth and yield were reduced.

Sheep grazing has been utilized in cereals as a way to supply feed and control weeds. Even in wheat, the benefits of grazing depend on several conditions and yield reduction is likely. Grazing treatments used in this study damaged linseed, did not control weeds and opened the crop for more vigorous weed growth. In conclusion, the study showed no benefit from tine weeding in linseed is likely if weed pressure is low. Under high weed pressure, a maximum of two tine weeding passes, one at cotyledon stage and another when average crop height is 5-7 cm, should be adequate. Higher sowing rate is recommended if tine weeding is in the plan to compensate for population loss.

Acknowledgements

The project was jointly funded by Agricultural and Marketing Research and Development Trust (AGMARDT), Foundation for Arable Research (FAR), Heinz Wattie's Ltd. and Biological Husbandry Unit (BHU) of Lincoln University. The author wishes to thank farmers who provided sites and machinery for the experiments.

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EFFECT OF TIMING AND NUMBER OF TILLAGE ON THE EMERGENCE PATTERN OF WEEDS IN IRRIGATED AREAS OF ETHIOPIA

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Abstract: Five schedules of tillage ranging from a single to six times repeated tillage and a no tillage control treatment were investigated at Melka Werer Research Center under irrigation to determine the natural emergence pattern of locally important weed species. The treatments were arranged in a randomized complete block design with four replications in a plot size of 18 m^2 with 60 cm spacing between ridges. The method of irrigation was furrow where by a 10-12 cm of water volume was applied at 15 day intervals. Eight weed species were identified and recorded among which *Portulaca oleraecae, Boerhavvia erecta, Sorghum arundinaceaum* and *Cyperus spp*. were the dominant once. They were the most frequently emerging, starting at the beginning of the season in November and at times as with for instance, *Cyperus spp*. among them continue to flourish was beyond the end of the season as long as water was available. The other four weed species each constituted at a range of 0.6-6.2% of the total weed seedling population and showed little or no germination on the first irrigation but exhibited prolonged emergence from November onwards.

Keywords: Tillage, *Portulaca oleraceae, Boerhavvia erecta, Sorghum arundinaceum, Cyperus spp*

Introduction

In the Awash River Basin, the Ministry of State Farms Development cultivates cotton on only 40,000 ha or 1.3% of total irrigable land during mid- April to September, despite the year round availability of irrigation facilities and conducive climatic conditions. Research results have shown that wheat is one of the candidate crops that could fit in an irrigated double cropping program in the Awash Valley (Jammal, 1987; 1994; Tanner *et al.* 1991, Kassahun, 1995).

There are however, lessons from earlier experiences that there will be a tremendous build up of weed populations of both stay home and new species because of the continuous supply of irrigation water (Kassahun, 1993; 1998). It is apparent that a proper and cost-effective weed control system is essential for irrigated wheat production in this area. It is therefore important to acquire a better knowledge on the types of weed species presents their level of infestations and behaviours, and their relative importance in order to launch a sound weed management scheme, which is virtually absent of the area under questions. The main objective of this study was to address this information gap by determining the natural pattern of emergence of the locally important weed species, under various tillage operations and to facilitate the prediction of infestations in crops planted at different dates and the design of agronomic approaches to control problematic weed species.

Materials and Methods

The trial was conducted for four consecutive years (1997-2002) at Melka Wearer Research Centre (90^o 16' N and 40^o 9'E) at an altitude of 750 m asl with a climate of semi-arid, the mean maximum and minimum temperatures of 31.4 and 18.2^{o}C, respectively, and a soil texture of silty clay. One month after the first irrigation in each year, all weeds were counted as required for specific treatments, taking 4 quadrat counts per plot (*i.e.* the quadrant measured 0.5 m *x* 0.5 m). Weed species were counted separately, considering only the newly germinated seedlings. When it was impossible to immediately identify certain seedlings, they were temporarily coded and identified after sufficiently matured. Treatments were arranged in a randomized complete block design with four replications. Plot size of 18 m^2 with 60 cm pacing between ridges was used. The method of irrigation was furrow, whereby a 10-12 cm volume of water was applied at 15 days interval. Each weed species was identified and recorded. No crop was planted through out each growing cycle. Six treatments of tillage were investigated in the experiment, namely T 1: No tillage, germinating weed seedlings were recorded one month after the first irrigation, T 2: Tillage practiced six times, where the first tillage was done one month after the first irrigation and followed by tillage at one month intervals, T 3: One tillage at one month after the first irrigation, T 4: One tillage at two months after the first irrigation, T 5: One tillage three months after first irrigation, T 6: Tillage practiced four times, where the first tillage was done two months after irrigation followed by tillage at one month intervals until the end of irrigation season

Results and Discussion

Species composition and emergence characteristics

Among 15 weed species identified, eight were registered as important (defined as >0.1% of total population) and accounted for 85.1 % of the total emerged species. The predominant weeds identified were *Portulaca oleraceae, Boerhavvia erecta, Corchurus olitorius, Lactuca taraxcifolia, Gaynandropsis gaynandra, Sorghum arundinaceum, Echinochloa colona* and *Cyperus spp* (Table 1). Some weed species such as *Lactuca taraxcifolia, Gaynandropsis gaynandra* and *Echinochloa colona* showed little or no germination on the first irrigation but had prolonged emergence from November onwards. The other weed species*, P. oleracea, B. erecta, C. olitorius, S. arundinaceum*, and *Cyperus spp*. continued to flourish even in March or April (Figure1).

Table 1. Comparison of weed emergence (No./m^2) in the different time and number of tillage for preceding intervals during (1997 –2007)

Date of count	*P oleraceae*	*B. erecta*	*C. olitorius*	*L. taraxcifolia*	*G. gaynandra*	*S. arundinaceum*	*E. colona*	*Cyperus spp.*
Nov. 15	177 a	22 bc	40 a	3 b	0 b	482 a	0 b	182a
Dec .14	70 b	27 b	11 b	5 ab	6 a	135 b	17 a	164 a
Jan. 15	57 b	18 bc	8 bc	4 b	3 b	12 c	11 b	94 b
Feb. 15	41 bc	38 a	5 bc	10 a	2 b	13 c	11 b	63 bc
Mar. 16	6 c	11 c	2 c	2 b	0 b	2 c	9 b	48 bc
Apr. 15	3 c	0 d	0 c	1 b	0 b	4 c	7 b	32 c
CV (%	34.1	30.5	39.5	67.9	36.5	40.3	25.6	27.9

Within a column, means followed by the same letter are not significantly different by the LSD (p=0.05).

Effect of tillage on emergence

There were significant differences in weed seedling densities between the control (no tilled) and monthly repeated tilled treatments. The number of weeds emerged on the repeatedly tilled plots was generally higher than on plots tilled only once. There was evidence on the depletion of certain species later in the season on these plots. *Sorghum arundinaceum, Cyperus spp* and *P. oleraceae* had the highest level of emergence in the no-till treatment (Figure 2). Of all weed species, 48.7% emerged with in one month after the first irrigation. *Boerhavvia erecta* and *Cyperus spp*. had relatively lower number of emergence under zero tillage (Table 1). The highest seedling densities of *L. taraxcifolia* and *B. erecta* were observed in February, however in all the other months, there was almost the same level of emergence. *Gaynandropsis* *gaynandra*, and *E. colona* did not emerge on no till plots but a lower degree of germination was observed on the other treatments. The density of *E. colona*, which was not found in the no- till plots, sharply increased with repeated ploughing and consistently decreased as the season advanced. The results suggest that weed control practices must aim at important weeds starting from November or at early stages.

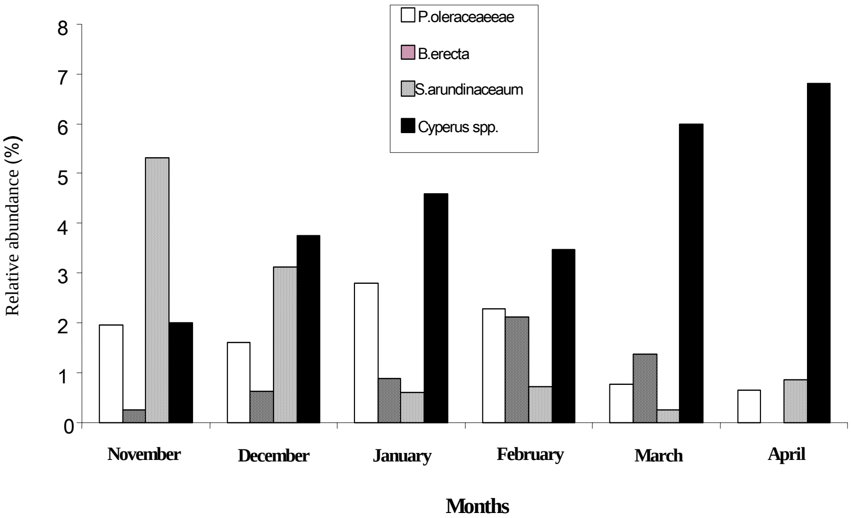

Figure 1. Emergence pattern (%) of the most predominant weed species as affected by tillage

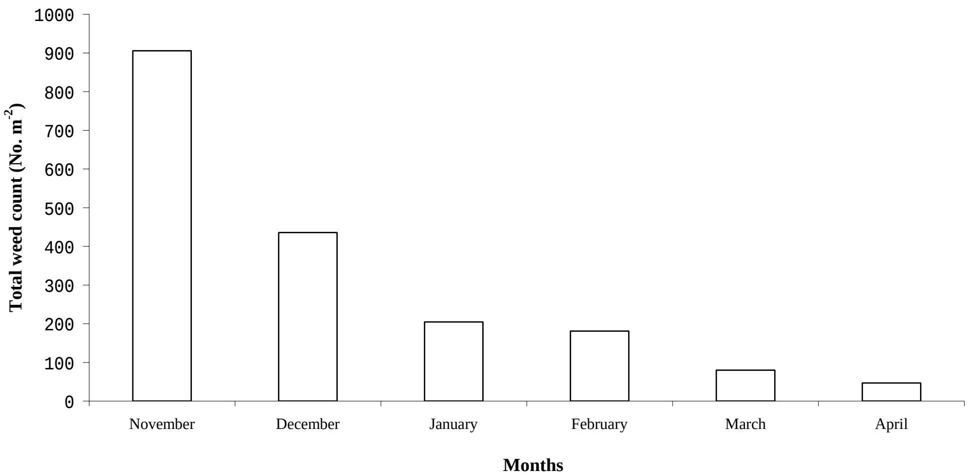

Figure 2. The total emergence of weeds during the experimental period.

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SCREENING OF ALLELOPATHY BY BIOASSAY AND EVALUATION OF ALLELOCHEMICALS IN THE FIELD USING THE CONCEPT OF TOTAL ACTIVITY

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Abstract: Allelopathic activity of more than 4000 plants were evaluated by specific bioassays, namely “Plant Box Method”, “Sandwich Method”, “Dish-pack Method”, and “Rhizosphere Soil Method”. Hairy vetch (*Vicia villosa*), velvet bean (*Mucuna pruriens*), buckwheat (*Fagopyrum esculentum*), spider lily (*Lycoris radiata*), common locust (*Robinia pseudo-acacia*), and Thunberg spirea (*Spiraea thunbergii)* were strongly allelopathic. Hairy vetch, velvet bean, buckwheat and spider lily were practically useful to suppress weeds in the field. Allelochemicals namely, L-DOPA, cyanamide, rutin and lycorine were isolated from these plants. In order to evaluate the contribution of allelochemicals on the field, a new concept of “Total Activity” has been developed and evaluated to investigate whether the above chemicals are the main allelochemicals in action.

Key Words: Allelopathy, bioassay, Total Activity, Plant Box Method, Sandwich Method, Dish-pack Method, Rhizosphere Soil Method

Plant-Box Method: A Bioassay to Evaluate Allelopathy through Root Exudates

The “Plant Box Method” is a bioassay to evaluate allelopathy through root exudates in situ. By using this method, the authors have examined the allelopathic activity of crop, vegetables and weeds grown in Japan.

Materials and Methods

Preparation of plants

River sand was passed through a 3 mm-sieve and washed with water, and filled into Black vinyl plastic pots measuring 6 x 6 (width) x 7cm (height). Four to eight seeds of each plant species were seeded in each pot. Nutrient solution was applied according to the need of plants. A vinyl acetate tube for tap water pipe with 32 and 25 mm, outer and inner diameter, respectively, was cut into 65 mm length. Then, a window was opened leaving a 90°angle wall (Figure 1) and was covered with Polyester gauze (Toray Tetron, Japan, #C-119 Skylark) using an adhesive agent (Ethron).

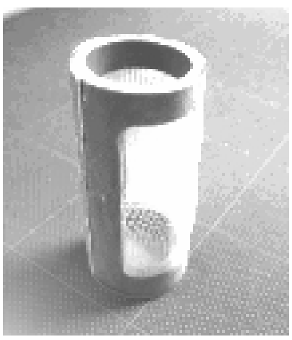


Figure 1. Plastic pots used in the experiment.

A 2-mm plate was attached and glued by the same agent at its bottom. The constituents of this adhesive agent were cyclohexane (40%), methy ethyl ketone (35%) and acetone (25%). The tube was oven dried at 60ºC to eliminate all solvents and washed with distilled water.

Root zone-separating tube Tube for Plant Box Method.
Agar preparation: Low-temperature gelatinizing agar (Nacalai Tesque, Kyoto, Japan), concentration of 0.5 to 0.75 % (W/V) and gelatinizing temperature of 30- 32ºC was autoclaved at 115ºC for 15 min and kept at 40ºC in a water bath until used.

Transplanting of test plants into plant-box: Donor plants grown in sand culture with estimated root dry weight of 100 to 300 mg were washed with distilled water and placed into the root zone-separating tubes of the plant-box (Magenta Box, Magenta Corporation, Chicago, USA, 60 x 60 (width) x 100 mm (height)). Five guide-dots at 10 mm distance were marked on 2 upper sides of the box at about 65 mm from the base for precise positioning of seeds (Figure 2). The tubes containing the test plants were fixed at the corner of the box through a cellophane tape attached to the upper part of the plant (Figure 2). Agar was poured into the box up to the level indicated with dots. The boxes were then cooled immediately in ice water and allowed to stand for 15 min until the agar gelatinized. Seeding of lettuce, the acceptor plant species, followed after one hour.

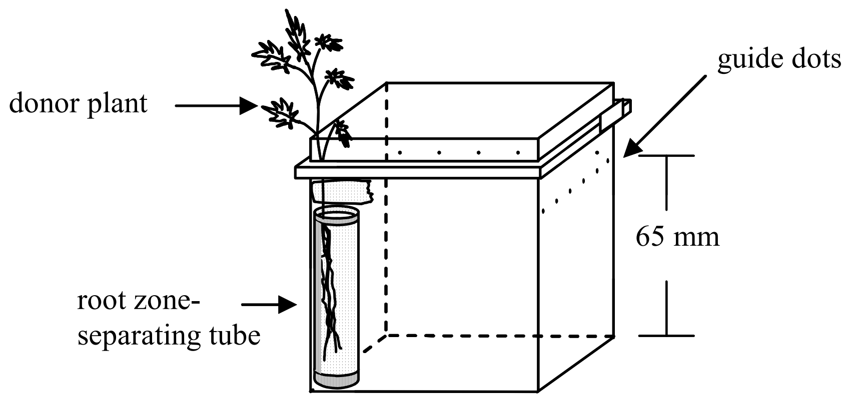


Figure 2. Diagram of a Plant-Box (Lateral View)

Seeding process: Lettuce seeds (*Lactuca sativa*, cv. Great Lakes 366) were used because of their high sensitivity, simultaneous and rapid germination, reliable germination percentage and homogeneity. Seeding was done (narrow tip downward) at distances of 1 cm apart as shown in Figure 3.

Plant growth conditions: The plant-boxes were fully covered (except the upper surface) with black plastic film in order to avoid root phototropism, and the top surface with polyethylene film to avoid possible bacterial contamination. They were set in the incubator and their daily conditions were checked. The incubation temperature was at 25/20ºC (12/12) and the agar was kept moist by adding appropriate amount of distilled water everyday. The radicle and hypocotyl lengths were measured after 5 d of incubation. Dry weights and heights of donor plant were also calculated. Usually, radicle length was affected more than the hypocotyl length, radicle growth inhibition was compared with the control.

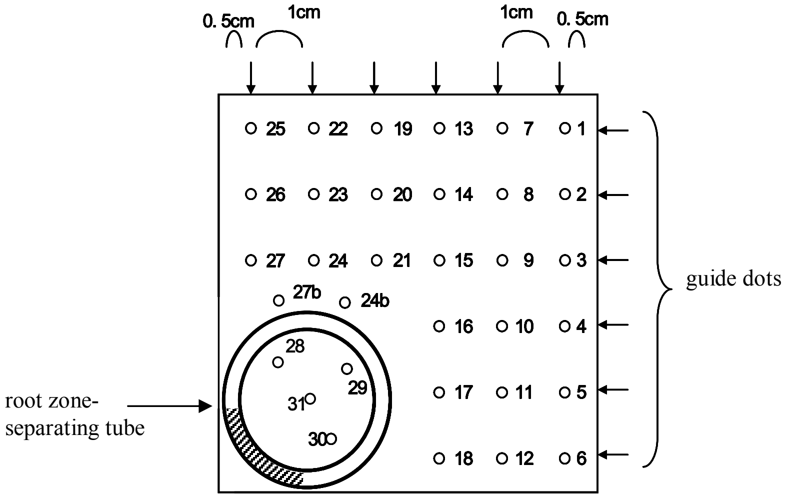


Figure 3. Top View of Plant Box (Seeding Diagram)

With the guide-dots followed, a lattice distribution of seeds had resulted.

Calculation of allelopathic activity: With the plant-box apparatus transparent characteristic, the effect of donor plant against lettuce, acceptor plant, (expressed by their radicle lengths) became highly visible. Radicle lengths of lettuce seedlings in close proximity to the donor plant tended to be shorter for allelopathic plants. As shown in Figure 4, distances from the root zone-separating tube and the radicle length were plotted and the linear regression curve was fitted with a computer calculation. From this calculated curve, the radicle length of seedlings inside root zone (zero distance) was compared to the length of control. This length shows the inhibitory effect of allelochemicals released from the root of the donor plant. The decline of each regression equation shows the migration speed of allelochemicals in agar medium. The change of migration speed may be due to their solubility in water, molecular weight and the shape of molecules.

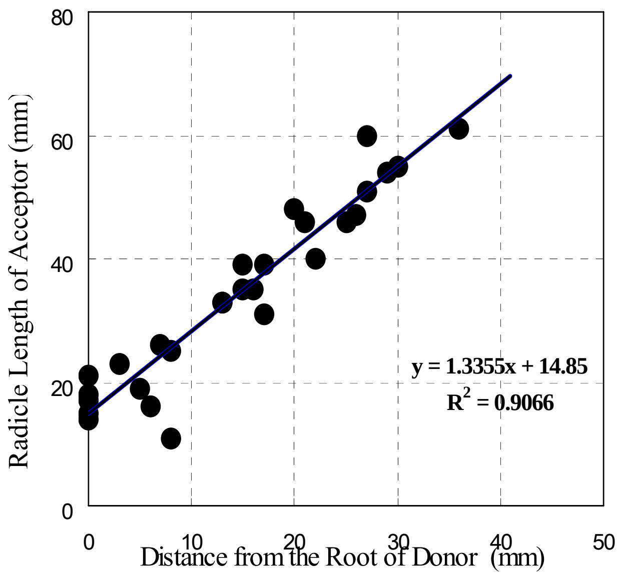


Figure 4. Calculation of allelopathic activity

Results

Assessment of allelopathic activity by the plant-box method

Table 1 shows the allelopathic activity between species and cultivars against lettuce as the acceptor plant. In Table 1 for *Leguminous* species, among 70 species tested, velvetbean showed the strongest inhibitory activity. We have already isolated L-3,4-dihydroxypenyl alanine (L-DOPA) as a major allelochemical from velvetbean (Fujii *et al*. 1990). Among its cultivars, 'hassjo', semi-dwarf type from Japan was the strongest. *Vicia* species such as *Vicia faba*, a winter legume crop cultivated for food and cover crop and *Vicia villosa*, known as hairy vetch or woolly pod vetch, also used as a cover crop was found to be strongly allelopathic. We isolated cyanamide as a new allelochemical from *Vicia villosa* species (Kamo *et al*. 2003).

Toxic legume such as *Pachyrhizus, Mimosa, Galega* were also allelopathic. It is better to observe the invasion of these legumes. Other promising legumes are *Medicago* spp*, Leucaena leucosephala, Canavalia ensiformis, Melilotus* spp*, Puerarila lobata,* and *Vigna* spp.

Table 2 shows that the species of family *Gramineae* show lesser inhibitory effects than the leguminous species. *Avena* spp like Oat (*Avena sativa*) known for its allelopathic activity since ancient times (Schreiner and Reed, 1907) showed the strongest inhibition with wild oats, *A. sterillis, A. murphy, and A. barbata* showing the highest activity. Other Gramineae species that seem promising are *Setaria italica, Panicum miliaceum, Anthoxanthum odoratum, Triticum* spp*, Sorghum* spp, *and Hordeum* spp. Most of these species have been reported as allelopathic plants (Rice, 1984).

In conclusion, plants such as *Avena, Mucuna, Abtilon, Erigeron, Brassica* and *Vicia* that have been reported allelopathic, have likewise shown strong inhibitory activity by the plant-box method. This method used lettuce as an acceptor plant, but it could be substituted with other plants such as weeds. If weeds are used, more replications and pre-incubation of seedlings should be done.

There are many bioassays reported during the past 30 years (Leather and Einhellig, 1986) but most of them are only tested for germination using extracts, exudates or leachates from plants. There are some reports that aim at standardization (Lehle and Putnam, 1982; Fujii *et al*. 1990a, b) but few bioassays use intact plants (Anderson, 1985) as our method does. This method makes it possible to assess the direct allelopathic activity of a donor plant and its direct effect to an acceptor plant.

Presently, more tests using more plant species and different cultivars are being evaluated. Our hope is to find plants that are highly allelopathic and to identify and assess the presence of possible allelochemicals in the field where the interaction takes place. By a combination of this method and field tests (Fujii *et al*. 1990a, b, 1991a-i;Yasuda *et al*. 1991) our team could separate the effects of allelopathy from those due to competition for light, nutrients, and water and possibly answer the questions proposed by Harper (1977).

Sandwich Method: A bioassay to evaluate Allelopathy through Leaf Leachates

Sandwich Method is a bioassay to evaluate allelopathy through leaf leachates *in situ*. By using this method, we have examined the allelopathic activity of crop, vegetables and weeds grown in Japan.

Table 1. Allelopathic activity Leguminosae Family by Plant Box Method

Scientific name	English name	activity
Pachyrhizus erosus	Jicama	6
Mucuna pruriens var. *utilis*	Velvetbean	7
Mimosa invisa		9
Crotalaria zanzibarica	Sunn hemp	12
Medicago scutellata	Snail medic	13
Galega orientalis	Galega	14
Coronilla varia	Crown vetch	16
Medicago arabica		16
Trifolium album		16
Medicago lupulina	Hop clover	19
Phaseolus vulgaris	Kidney bean (Old cv.)	19
Vicia villosa var. dasycarpa	Woolly pod vetch	19
Abrus praecatorius	Rosary pea	20
Canavalia ensiformis	Jack bean	20
Lupinus bicolor		20
Vicia villosa var. villosa	Hairy vetch	20
Melilotus albus	White sweet clover	23
Dolicos lablab	Lablab bean	25
Vicia sativa	Common vetch	25
Pueraria lobata	Kudzu	28
Crotalaria juncea	Sunn hemp	30
Trigonella foenum-graecum	Fenugreek	30
Vigna unguiculata	Cowpea	33
Cassia tora	Sickle senna	34
Medicago sativa	Alfalfa	34
Vicia faba	Broad bean	34
Onobrychis viciifolia	Esparcette	35
Pachyrhizus tuberosus	Ahipa	36
Phaseolus vulgaris	Kidney bean (Modrn cv.)	37
Vigna angularis	Adzuki bean	38
Trifolium incarnatum	Crimson clover	39
Cajanus cajan	Pigeon pea	40
Crotaralia spectabilis	Sunn hemp	40
Pisum sativum	Pea	42
Cicer arietinum	Chickpea	44
Lathyrus odoratus	Sweet pea	44
Amorpha fruticosa	False indigo	45
Latyrus sativus	Grass pea	46
Lupinus albus	White lupine	46
Arachis hypogaea	Peanut	49
Calopogonium mucunoides	Calopogonio	49
Desmodium paniculatum	Tick trefoil	49
Vigna unguiculata subsp. sesquipedalis	Asparagus pea	49
Mimosa pudica	Sensitive plant	55
Vicia angustifolia var. segetalis	Karasunoendou	55
Aeschynomeue rudis	Joint vetch	58
Lotus corniculatus	Bird's foot trefoil	59
Cytisus scoparius	Common broom	60
Lathyrus latifolius	Perennial pea	60
Sesbania cannabina	Sesbania	61
Trifolium repens	White clover	61
Astragalus sinicus	Chinese milk vetch	62
Glycine max	Soybean, cv. Tachi-nagaha	65
Trifolium pratense	Red clover	72
Trifolium hybridum	Alsike clover	118

Table 2. Allelopathic activity Gramineae Family by Plant Box Method

Scientific name	English name	activity
Avena sterilis	Wild Oat	12
Triticum polonicum	Polish Wheat	13
Setaria italica	Foxtail Millet	18
Avena barbata	Wild Oat	18
Triticum spelta	Spelt Wheat	22
Triticum sp. x Secale sp.	Triticale	23
Chloris gayana	Rhodesgrass	24
Avena byzantina	Byzantine Oat	25
Avena hirtula	(Wild Oat)	25
Anthoxanthum odoratum	Sweet Vernalgrass	28
Secale cereale	Rye	29
Avena sativa	Oat	30
Festuca rubra	Chewing Fescue	30
Panicum coloratum	Coloured Guinea Grass	33
Triticum aestivum	Common Wheat	34
Coix lacryma	Coix	37
Hordeum vulgare	Barley	38
Panicum maximum	Guinea Grass	38
Avena abyssinica	Ethiopian Oat	39
Avena brevis	Brevis Oat	39
Bromus catharticus	Rescuegrass	40
Triticum dicoccum	Emmer Wheat	41
Dactyloctenium aegyptiacum	Coast Button Grass	41
Triticum turgidum	Rivet Wheat	41
Sorghum halepense	Johnson Grass	43
Oryzopsis miliacea	Smilo Grass	44
Festuca arundinacea	Tall Fescue	45
Sorghum sudanense	Sudan Grass	45
Agropyron repens	Quackgrass	46
Digitaria sanguinalis var. *ciliaris*		49
Setaria viridis	Foxtail Grass	53
Poa trivialis	Rough-stalked Meadowgrass	54
Pennisetum glaucum	Pearl Millet	54
Euchlaena mexicana	Teosinte	56
Chloris distichophylla	Weeping Chloris	57
Poa ampla	Big Bluegrass	57
Zoycia japonica	Japanese Lawn Grass	58
Poa pratensis	Kentucky Bluegrass	60
Echinochloa utilis	Japanese Barnyard Millet	60
Zea mays	Corn	60
Dactylis glomerata	Orchard Grass	62
Eragrostis curvula var. *valida*	African Lovegrass	62
Agrostis alba	Red Top	62
Lolium perenne	Perennial Ryegrass	64
Alopecurus aequalis var. *amurensis*		64
Cenchrus ciliaris	Buffel Grass	71
Paspalum dilatatum	Dallis Grass	72
Paspalum notatum	Bahia grass	73
Eleusine indica	Goose Grass	74
Lolium multiflorum	Italian Ryegrass	74
Phalaris arundinacea	Reed Canary grass	76
Festuca elatior	Meadow Fescue	87
Phleum pratense	Timothy	96
Imperata cylindrica var. *koenigii*	Cogon grass	115

Materials and Methods

Main protocol for Sandwich Method

Fifty mg or ten mg of dried leaves was added in between the two layers of agar (Figure 5) in each well (10 cm2) of the multi-dish plate, based on the data that the average amount of fallen leaves of a particular species per tree per year remained in constant, and about 3 t/ha/year (Numata, 1969). Three tons is equivalent to 30mg of leaves on 10 cm2. Then 10 and 50 mg was selected. In case the leaves were hydrophobic and floated on the surface of the agar gels, a pin-set was used to insert the leaves into the gel. Prior to the bioassay, we calculated the migration speed of known compounds by the same condition. Riboflavin (0.2 mg/ml), molecular weight is 376, and Blue Dextran 2000 (2mg/ml), molecular weight is 2000, was used as model compounds with visible color. By corkscrew of diameter 6 mm, agar of the corner of dish was removed and filled by the solution of each model compounds. Then migration speed was measured optically. The results show that migration speed of both compounds is not so different and about 1 cm per day. Then, we assumed that the allelochemicals exuded from leaves of donor plants will reach at the end of the dish within a day, and thus the incubation period of 3 to 4 days is enough for the elution of allelochemicals for this bioassay. Lettuce (*Lactuca sativa* L. Great Lakes 366, Takii Seed Co. Ltd., Japan) was used as test plant material.

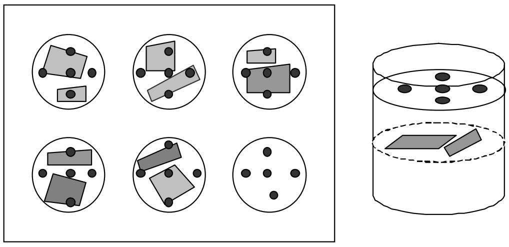


Figure 5. Rough scheme of Sandwich method

This plant has been used as test material universally for ease of handling, and a rapid and uniform growth response. After gelatinizing the agar, 5 lettuce seeds were placed on the surface of each agar-containing well of the multi-dish plastic plate. Length of both radicle and hypocotyl of lettuce seedlings was measured every day after imbibition onset for 4.5 days. In addition, the germination percentage of seeds was recorded. Each experiment was conducted three times and results are presented as the mean of three replicates. After seeding, the multi-dish plastic plate was covered with plastic tape, labeled appropriately, and kept in an incubator (BITEC 300-L) (Shimadzu Rica Inst. Co. Ltd., Japan) for 4.5 days at 25ºC under complete darkness.

Results

We have evaluated the allelopathic activity of about 4,000 plant species. Distribution of these data is summarized in Figure 6. Compiling all data, the results show normal distribution. An example of the results by sandwich method for the evaluation of leaf materials of 20 different tree and herbal species in Asia is shown in Table 3. For the evaluation of allelopathic activity, we introduced the concept of “standard deviation value”. As to the statistical analysis, we calculated the mean (M) and standard deviation (σ), and evaluated from the criteria of standard deviation value (SDV). Criteria (**) in the column in the Table 2 means stronger inhibitory activity more than the mean value minus 1.0σ, that means SDV = 60. Criteria (*) in the column means inhibitory activity more than the mean value minus 0.5σ, that means SDV = 55.

Among the species tested, lemongrass (*Cymbopogon citratus*) and derris (*Derris scandens*) showed the strongest inhibitory activity and caused 100% growth inhibition in the radicle and hypocotyl in lettuce seedlings. Betel pepper (*Piper betle*), tamarind (*Tamarindus indica*), and gliricidia (*Gliricidia*) also showed very strong inhibitory activity. All these plants are known to have specific natural chemicals and used as herb, medicine, or special use for insect control. Swamp Pea (*Sesbania grandiflora*), acacia (*Acacia farnesiana*), golden dewdrop (*Duranta repens*) and persimmon (*Diospyros mollis*) showed moderate inhibitory activity. These plants are known to possess special natural chemicals and famous for resistance against insect and disease attack. We have already isolated new natural bioactive substances named - durantanin that function as potent allelochemicals from golden dewdrop (Hiradate *et al*. 1999). Compared to these plants, Alexandrian laurel (*Calophyllum inophyllum*), queen of flowering tree (*Amherstia nobilis*), Lamta (*Cynometra cauliflora*) and litchi (*Litchi chinensis*) showed either no inhibitory or slightly promotive activity.

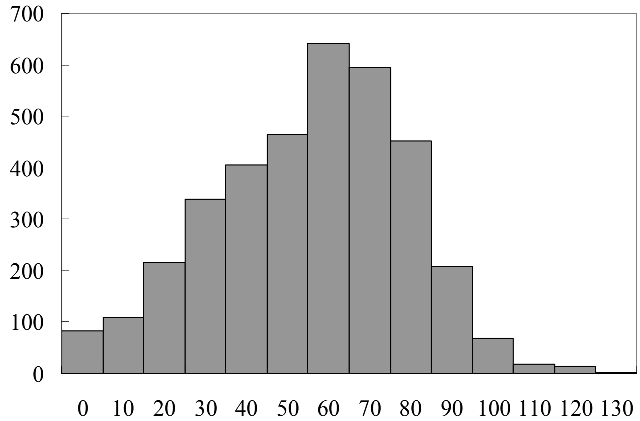


Figure 6. Distribution of the results of the sandwich method

Table 3. Allelopathic activity by Sandwich Method

Scientific name	Radicle	Hypocotyl
Cymbopogon citratus	0**	0**
Derris scandens	0**	0**
Piper betle	3**	18**
Tamarindus indica	3**	26*
Gliricidia sepium	5**	19*
Sesbania grandiflora	11*	45*
Acacia farnesiana	15*	37
Duranta repens	17*	51
Diospyros mollis	23*	22
Afgekia sericea	38	120
Ipomea pes-caprae	39	90
Jatropha integerrima	44	122
Melia azedarach	52	115
Citharexylum spinosum	70	60
Molineria latifolia	75	167
Passiflora coccinea	86	117
Calophyllum inophyllum	94	146
Amhersita nobilis	96	127
Cynometra cauliflora	101	113
Litchi chinensis	113	115

Radicle and hypocotyl means % growth of lettuce seedling to the control (in agar medium). ** after the data means inhibitory activity stronger than standard deviation value of 60, and * means 55.

Discussion

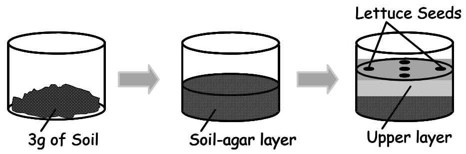
The merit of using agar is to transfer the water soluble chemicals from leaves. This idea is already used for the bioassay for plant hormones. It has been reported that with a particular species, the average amount of fallen leaves per tree per year remained constant. For example, broad leaf deciduous trees shed leaves of ca 3 to 5 t/ha/year, while the amount is twice in coniferous tress and thrice in tropical rainforest. Since, one of the three routes of allelopathic phenomenon is through leaching by leaf litter, as described in the introduction, therefore, it is very important to evaluate the allelopathic potentiality of leaf litter leachates.

Dish Pack Method: A Bioassay to Evaluate Allelopathy through Volatile Chemicals

Dish Pack Method is a bioassay to evaluate allelopathy through volatile chemicals *in situ*. By using this method, we have examined the allelopathic activity of crop, vegetables and weeds grown in Japan.

Materials and Methods

Test plants were cultivated for about one month by sand or soil culture, supplying water containing the nutrient solution in greenhouses (Figure 7). The receiver plant used for bioassay was lettuce (Great Lakes 366), since it is highly sensitive to inhibition by allelochemicals. Standards of volatile compounds used in this experiment were commercial products. Multi-dishes with 6 holes made in Nunc Company (diameter of holes; 3.5 cm), and gas chromatograph-mass spectrometer (GC-MS) model QP-5000 made in Shimadzu Company were used. Capillary column for GC-MS was HR-Thermon600T (Shin-etsu Chemical Company). The column was 25m long and its inside diameter was 0.25 mm. Pressure of Helium gas as carrier was 37 kPa/min. Injection temperature was 250℃. The column was produced an increase in temperature at the rate of 4℃/min from 50℃ to 110℃. We checked 35 test species mainly of cover plants. Used multi-dishes with 6 holes were same with those used in Sandwich method, except that the area of the cover corresponding to each holes were drilled in the center with a drill. Silicon septums were fitted into the drilled holes. 2 g of fresh leaves were cut by scissors and placed into one of the holes in the multi-dishes. Filter paper and 0.7ml of distilled water were put into the other 5 holes, and 7 seeds of lettuce were put on the filter paper. Each side of the multi-dishes were sealed by cellophane tape and packed in aluminum foil, then placed in incubator under 25℃. Radicle and hypocotyl lengths of lettuce were checked after 4 days. As just described, we could check volatile compounds activities in non-contact system. Speed of diffusion and intensity of activity of volatile compounds were estimated based on the relation of the distance between test plants and lettuce seeds.



1. Soil-agar layer: 5mL of agar (5%) cooled at 42°C was added into multi-dish (6 dish) containing 3g of the fresh soil in the dry weight. We amended 5 replications both rhizosphere soil and root-zone soil.
2. Upper Layer: After solidification, 3.2mL of agar (5%) cooled at 42°C was added on soil-agar layer. 5 lettuce seeds were placed on the agar culture medium.
3. The multi-dishes were incubated at 25°C in the dark. After 3 days, the length of their radicles were measured.

Figure 7. Dish Pack Method.and Concise Protocol of Dish Pack Method

The internal volatile gas was analyzed with time by GC-MS. Headspace gas (0.5ml) was collected from the hole with a gas tight syringe through the septum set up on the holes. To identify the volatile compounds in test plants, we then used glass bottles (100ml) with holes drilled in the center of their top covers as test instruments. 2g of ground test plants were put in it, then 15 min. after, internal gas (0.5ml) was collected from the bottles with a gas tight syringe and analyzed by GC-MS. Additionally volatile standard compounds were tested with the same method.

Results

The screening results for allelopathic cover plants by Dish Pack Method, Plant Box Method and Sandwich Method were shown in Table 4. In the case of Plant Box Method and Sandwich Method, allelopathic activities were high in legume. At the same time in the case of Dish pack Method and Sandwich Method, allelopathic activities of *Cleome* were the highest of all test plants.

Table 3. Allelopathic activity by Dish Pack Method, Sandwich Method and Plant Box Method

Scientific name	Dish Pack	Sandwich	Plant Box	major vilatile chemicals
Cleome spinosa	0	0	57	methyl isothiocyanate
Papaver rhoeas	16	39	10	2-hexenal
Pueraria thunbergiana	22	65	20	2-hexenal, hexenal, 4-pentenal
Hibiscus cannabinus	31			2-hexenal, 3-hexenal
Solidago altissima	32	65	79	α-pinene, limonene, myrcene, ocimene
Vicia villosa	34	15	30	2-hexenal
Ficus carica	34			2-hexenal, 4-pentenal
Rosmarinus officinalis	36	94		α-pinene, camphor, cineole
Crotalaria agatiflora	39	36	23	2-hexenal, *trans*-3-hexenol
Hieracium aurantiacum	39	81		
Artemisia princeps	41		85	β-pinene, cineol, 2-octenal
Vinca major	45	34		*cis*-3-hexenyl acetate, *trans*-3-hexenol
Ipomoea aquatica	54	22	98	2-hexenal, 4-pentenal
Mucuna pruriens	60	10	11	hexenal, 2-hexenal, 3-hexenal
Fagopyrum esculentum	66	19	74	2-hexenal, 3-hexenal
Arctotheca calendula	66	68		β-pinene, 2-hexenal
Phlox subulata	68	23	29	limonene
Phacelia tanacetifolia	71	26	32	myrcene, limonene, 2-hexenal
Potentilla verna	71	69	23	
Thymus serphyllum	74	44		terpinen, cymene, isocaryophyllene
Oxalis articulata	75	23	28	3-hexen-1-ol acetate
Chamomilla nobilis	79	83	60	ocimene, cyclopropanecarboxylic acid
Festuca myuros	82	27	27	*cis*-3-hexenyl acetate
Lampranthus spectabilis	83	16		
Coreopsis tinctoria	83	81	93	limonene, α-phellandrene, α-pinene
Sedum sarmentosum	85	32	58	
Mentha pulegium	87	50	66	pulegone, myrcene, limonene
Zoysia japonica	89			
Cymbopogon citratus	92			myrcene, citral,
Houttuynia cordata	93		74	myrcene, β-pinene, ocimene, limonene
Lycoris radidata	94	94	65	2-hexenal
Ocimum basilicum	97	83	83	linalol, cineole

Volatile compounds from these test plants were analyzed by GC-MS. From legume, carbonyl compounds (*trans*-2-hexenal, et al.) were mainly identified. From oxeye and labiate, monoterpenoids (α-pinene, limonene, myrcene et al.) were mainly identified. And from *Cleome*, MITC (methyl isothiocyanate) was identified.

In another test by Dish Pack Method, MITC completely inhibited the germination and growth of lettuce at 18 ppm (v/v). In the same method, leaf aldehyde (*trans*-2-hexenal) inhibited growth of lettuce radicle 64% at 21ppm. Inhibitory activity by leaf alcohols (*cis*-3-henenol et al.), monoterpenoids (myrcene, limonene, pinene, et al.) are not strong. We analysed the concentration of MITC in Cleome. MITC concentration increased in injured leaves, but less from injured roots. MITC was also released in the germination process. Growth inhibitory activity in Dish Pack could be explained by MITC concentration contained in each part of *Cleome*.

Discussion

Usually volatile compounds from leaves are diffused by wind and usually difficult to be concentrated enough to have potent inhibitory activity in the field. But in case of cover crops or their fallen leaves thickly covering the ground surface, it is possible that volatile compounds could reach enough effective concentration to inhibit the germination and growth of other plants. It is possible that MITC from *Cleome* protect plants from enemies in case of injury or germination.

Rhizosphere Soil Method: A Bioassay to Evaluate Allelopathy through Rhizosphere Soil

Rhizosphere Soil Method is a bioassay to evaluate allelopathy through rhizosphere soil *in situ*. By using this method, we have examined the allelopathic activity of crop, vegetables and weeds grown in Japan.

Materials and Methods

Each plant was grown in plastic pot using mixed soil (Andosol and commercially available soil), and cultivated at the green house at 25℃ for several weeks. The plants were taken out from the plastic pot without disturbance, then plant root were shaken softly to remove the root-zone soils. We collected the soil adhering to the surroundings of a root, called "rhizosphere soil" in general, and the soil shaken off is “root-zone soil”. Collected soils were sieved in a 1mm mesh removing root hair as much as possible. Fresh soil was used for the experiment. Because of the difference in growing of the root hair or soil moisture content, the amount of the rhizosphere soil from one pot differed between plants. In order to obtain the amount of soil required for sandwich method, rhizosphere soils were collected from several pots of same plant species.

Results and Discussion

The results are shown in Table 5. *Prosopis juliflora, Mucuna pruriens* and and *Vicia villosa*, belonging to the family Leguminosae, inhibited radicle growth of lettuce strongly in the Rhizosphere Soil Method. Rhizosphere soils of *Solidago altissima* also showed strong inhibitory activity. *Solidago altissima* is an invader plant from North America. *Mucuna* and *Vicia* are well-known allelopathic cover crop in Japan (Fujii 2001, Fujii *et al*. 2003), and our group identified L-DOPA and cyanamide as the allelochemical from these plants. The results from Rhizosphere Soil Method showed the allelopathic activity of allelochemicals exudated into soil. The inhibitory activities of rhizosphere soils were stronger than those of root-zone soils in all cases. This suggests that substances released from roots of plants are adsorbed on soil, or decomposed by soil microorganisms.

Table 5. Allelopathic activity by the Rghyzosphere Soil Method

Scientific name	Rhizosphere	Root zone
Prosopis juliflora	15	14
Paspalum notatum	17	89
Mucuna pruriens	18	75
Robinia pseudoacacia	26	50
Vicia villosa	26	71
Solidago altissima	47	56
Eucalyptus pellita	48	73
Eucalyptus grandis	49	60
Bischofia javanica	50	57
Tamarindus indicus	50	98
Eucalyptus citriodora	54	67
Cuphea hyssopifolia	59	88
Duranta repens	61	90
Cassia grandis	62	90
Cuphea hyssopifolia	63	61
Cordyline stricta	65	85
Cycas revoluta	71	77
Enterolobium contortosiliquum	74	71
Artocarpus heterophyllus	75	81
Albizia guachapel	80	90
Cassia siamea	83	89
Delonix regia	86	87
Caesalpinia pelfophoroides	88	89

Total Activity Method: A Bioassay to Evaluate Allelopathy and Isolate Allelochemicals in action *in situ*.

Total Activity Method is a concept to evaluate allelopathy and isolate allelochemicals really acting in the field. By using this method, it is possible to evaluate the allelopathic activity of plants in the field.

Materials and Methods

In the course of screening of allelochemicals in action in the field (*in situ*), we introduced a new strategy of "Total Activity" concept according to the isolation method of enzymes in plants instead of "Specific Activity" method used in many reports ever published. For the isolation of natural agrochemicals in order to isolate new bioactive compounds with high specific activity, and use these chemicals as lead compounds for agrochemicals, specific activity is of course useful. But for the search of allelochemicals in action in the field, "Total Activity Method" is more important to isolate such allelochemicals. By this strategy, we could isolate unique compounds and important allelochemicals in the field.

As shown in Table 6, the specific activity means biological activity per unit weight of the compound, and total activity means biological activity per unit weight of the organism. Specific activity is easy to calculate, and in our case, we calculate EC50. For the isolation of allelochemicals candidates from crude extract, each activity was measured on "g" base, as shown in Table 5. For the comparison of Total Activity to other reports, Index on molar base was calculated after the isolation and identification of allelochemicals.

In order to show the importance of Total Activity, Table 7 explains and compares the explanatory value of Specific Activity and Total Activity. In this example, as for Specific Activity, compound A is ten times stronger than compound B. But concentration of compound B is hundred times higher. Thus, Total Activity of compound B is ten times stronger than compound A. In this example, we will conclude that compound B is ten times important to contribute for the allelopathic phenomena in the field and more important as allelochemicals.

In order to explain how to separate allelochemicals candidates by "Total Activity" concept, Scheme explain the strategy. In each step of isolation, Specific Activity of each fraction (Fr. 1, 2, 3, …) was measured each time. Weight of each fraction was also measured and based on these data, Total Activity was calculated. In the standpoint of total activity, Fraction with the highest total activity was selected for further fractionation.

Table 6. Comparison of total activity and specific activity

Target	Compounds	Index (g-weight base)
Direct use of Allelopathy	Allelochemicals in action *in situ*	Total Activity = 1/EC_{50}× Concentration in plants 1/ (kg/L) x (kg/L in fresh weight) =(L/kg) x (kg/L) = (no unit) high value ~ high activity
Use of natural Chemicals	Bioactive chemicals with high specific activity	Specific Activity = EC_{50} (50% inhibitory Concentration) (kg/L) ~ ppm low ppm ~ high activity

Table 7. Concept and calculation of Total Activity

	Specific Activity (EC_{50}, μ M)	Concentration (μ M)	Total Activity (no unit)
Compound A	1	1	1
Compound B	10	100	10

Specific Activity : A > B (10 times)
Concentration : B > A (100 times)
Total activity : B > A (10 times)
(B is more important as allelochemicals)

Total activity= 1 / Specific Activity (EC_{50})× Concentration

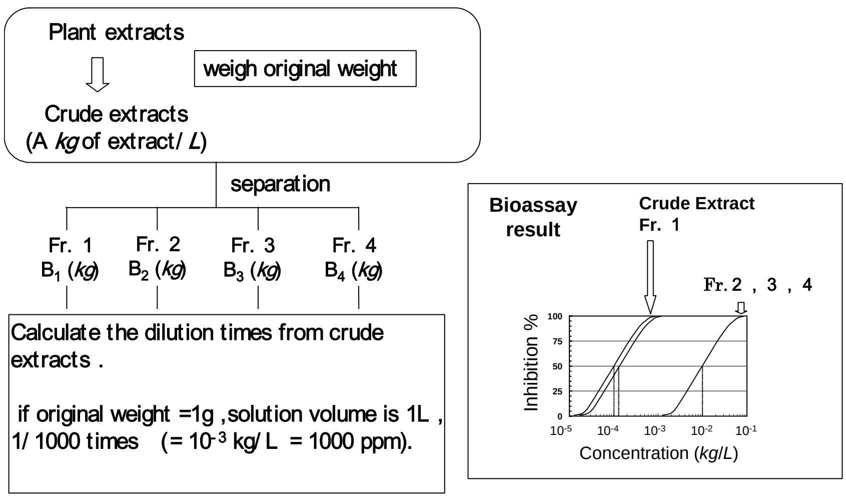


Figure 8. Scheme of isolation of allelochemicals by Total Activity Method

Results and Discussion

By means of Total Activity Methid, L-DOPA from velvet bean (*Mucuna pruriens*). The concentration of L-DOPA in the plant is extremely high and about 1 % in fresh weight in the leaves and roots (Fujii *et al.* 1991, 1992). Specific activity of L-DOPA is 4x10-4M, Total Activity is 200. This value is the highest before the isolation of *cis*-cinnamic acid form *Spiraea thunbergii*, with value of 1000. The Specific Activity of *cis*-cinnamic acid is 1000 times stronger than *trans*-cinnamic acid and about 3x10-6 M. This activity is equivalent to *cis*-ABA, one of the plant hormone (Hiradate *et al.* 2004, 2005).

Using the Total Activity method, a potent natural chemicals from *Duranta repens* has been isolated. As these compounds were new saponins, they are named as "durantanins"(Figure 7). These compounds were contained about 4 % in dry weight in *Duranta* (Hiradate *et al.* 1999).

Hairy vetch is a cover crop. We have studied the practical weed suppression activity of hairy vetch from 1990, and demonstrated its use in the field (Fujii 1999, 2000, 2001). As a result of our recommendation, hairy vetch is now cultivated as organic weed control in orchard and fallow of rice field in Japan. Isolation of plant growth inhibitory chemical from hairy vetch was very difficult and after ten years of research, we had isolated cyanamide as a natural chemical found in it (Kamo *et al.* 2003). Cyanamide is a well- known synthetic fertilizer and herbicide synthesized about 100 years ago, but there has been no report of cyanamide being a natural product before we reported. Cyanamide is a very simple compound with low molecular weight. As cyanamide has only one simple signal by NMR and MS, it is usually very difficult to identify it in the course of isolation. Without the strategy of Total Activity, it could have been difficult to isolate this compound.

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DISTRIBUTION AND ECOLOGICAL IMPACTS OF *Parthenium hysterophorus* L. IN ETHIOPIA

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Abstract: Parthenium (*Parthenium hysterophorus* L.) is an alien weed species introduced in Ethiopia some twenty-five years back. It has aggressive characters invading cropland, pastureland and irrigation canals causing an irreversible displacement of natural pasture grasses and has an effect on animal husbandry, human health and biodiversity. Parthenium is one of the major biotic limiting factors to the production of different crops and grazing lands in Ethiopia. To collect accurate information on the distribution, spread and impact of parthenium, field survey was conducted during 2006 and 2007 in central and southern parts of Ethiopia. The survey results indicated that it has a wider spread, infesting most of the surveyed areas. The infestation levels of parthenium varied from low (60% of the infested fields), moderate or high (5% of the infested fields) and to very high (35% of the infested fields). From the interviews done and data recorded, the infestation with parthenium is considered by 75% of the farmers as a very serious problem and 16% of them observed an increasing infestation over time, while 9 % of them did not know the weed. In general the weed was found in varying dimensions in waste lands, and along roadsides, railway tracks, and cultivated fields, attaining the status of a worst weed among the terrestrial weeds in the country.

Key words: *Parthenium hysterophorus*, ecological impacts.

Introduction

Congress grass /fever few/ wild fire (*Parthenium hysterophorus* L.) is alien or exotic weed to Ethiopia. There mere speculation that the weed was introduced to Ethiopia through aid food grain. The occurrence of the weed was first reported in eastern parts of the country, *i.e.* around Dire Dewa in 1967 (Frew *et al*. 1996). Since then it has spread to central and northern parts of the country especially along roadsides, and has demonstrated its' ability of having an extraordinary capacity to spread rapidly. *Parthenium hysterophorus* L. is a noxious weed that competes strongly with different food crops for nutrients, water and sunlight. Hence, it affects the production of many crops.

Parthenium weed has evolved specificity to crops and plants in the natural vegetation. Other plant species could hardly establish themselves in a field infested with parthenium due to the antagonistic effect of the toxins produced by the roots of the weed. Parthenium is not only damaging the crop plants, but also poses a serious human and animal health risk due to the dangerous toxins that all parts of the weed can produce. It also lowers the quality of dairy products if dairy cows are feed to the weed (Rezene *et al*. 2005; Taye, 2002).

A considerable loss in growth and yield of many food and fodder crops is caused by Parthenium weed. Nowadays, parthenium is considered as the greatest single biotic constraint to food production in Ethiopia, where the livelihood of 70 million people is adversely affected. In infested areas, yield losses associated with Parthenium damage are often significant, ranging from 40-100 per cent (Kassahun, 1999). Moreover, it is predicted that the grain production in Ethiopia is potentially at even increasing risk in the future. The study was conducted to collect information on the distribution and spread of parthenium in central Ethiopia.

Materials and Methods

Field survey was conducted in central Ethiopia: Central, East, and North Shoa, North Wolo and East Hararge during July-Augst 2006 and 2007. The survey covered grazing land, fallow land, roadside, residential area, riverside and in the land where different type of crops were grown. The major crops in the survey area are: Tef (*Eragrostis tef*), Wheat (*Triticum aestivum* L.), Sorghum (*Sorghum bicolor* L.) and Maize (*Zea mays* L.). Quantitative weed survey was undertaken. Five samples were collected from each locality with a quadrate of 1m^2 sampling area, within 25 km distance and the infestation was converted to a scaleaccording to the density levels of the weed, namely, 1 plant/m^2 = low abundance, 2-3 plants/m^2 = medium abundance , and > 3 plants/m^2 = high abundance. Observation and counting of other weed species was done. The coordinates were recorded for each locality. By developing questionnaire that covers all general issues about exotic weeds were recorded. A total of 250 sample sites assessed. The infestation level observed from the sampled quadrants was ranged from low to high.

Results and Discussions

In all surveyed areas from Adama up to Boset (Figures 1 and 2), the weed was recorded at “high abundance”. The infestation observed was nearly equally distributed and indicated that all were beside the high way, residential areas, grazing areas and in the land where different type of crops were grown. From the interview done in these areas among farmers, *P. hysterophorus* ranked the top most problematic weed, as it was replacing other beneficial vegetations like grasses, and also spoils animal products such as milk giving bitter tests. It causes yield reduction of crops by draining the moisture, reducing soil fertility, and harbouring insect pests. The farmers in the countryside and some urban dweller responded that they often used hand pulling, and slashing and burning method to control the weed.

In Boset Woroda, 38 kebeles were totally invaded out of 42, and were hotspots for parthenium infestation. In this area, the highways at about 2 - 3 km radius were covered by the weed, besides most grazing areas. The other most endangered area was the national park of Awash (Figure 2).

Near and in Addis Abeba (Figure 1), the weed is spreading from Kaliti to Akaki at a very high coverage along the roadside in the field and in the fenced areas that are left for construction. In the Arsi zone, the invasion of parthenium was observed only at few locations. Its’ coverage in the area ranged from low to high (Figure 2). However, from Dera to Kofele, parthenium was not observed, and farmers and urban dweller were also unaware of the weed. Parthenium has being infesting the roadside at different spots; however it is moving near the farm land and replacing important grass species in the grazing land. The infestation level is very high at the roadside up to a 100 - 500 m radius and at low level in the farm land. In general the infestation levels of parthenium, varied, from low (60% of the infested fields), moderate or high (5% of the infested fields) and to very high (35% of the infested fields). From the interview made and data recorded the infestation with parthenium is considered by 75% of the farmers as a very serious problem and 16% of them observed an increasing infestation over time, while 9% did not know the weed. Thus, the extent of problem caused by *P. hysterophorus* was very much severe as compared to other weeds in most of the sampled localities.

Partheinum distribution

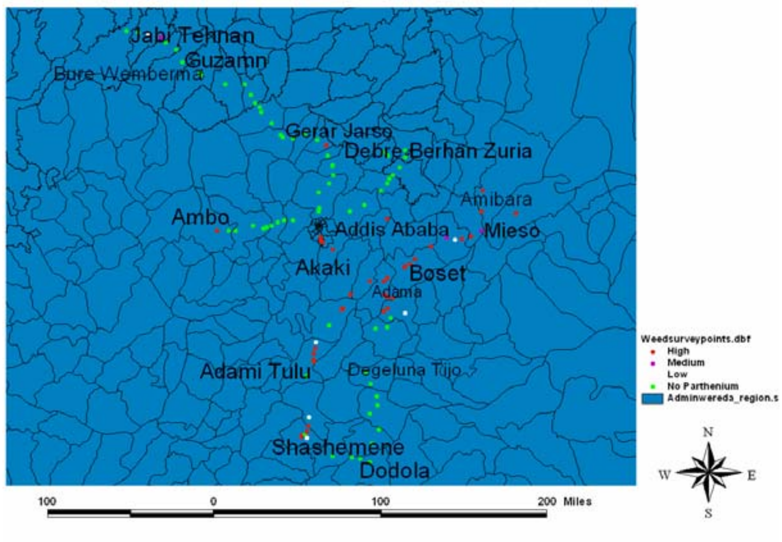

Figure 1. Areas of partheinum weed surveyed in central Ethiopia

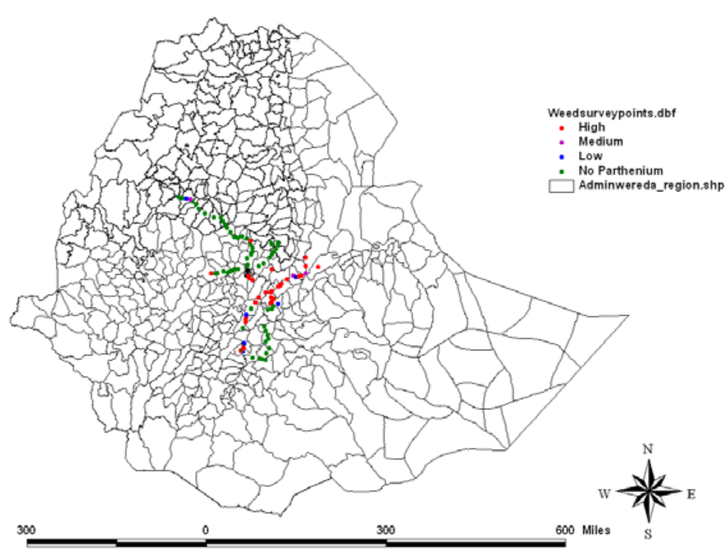

Figure 2. Level of infestation of partheinum weed in central Ethiopia

Conclusions

The survey results clearly indicated that education and awareness creation, establishing community based local rules and regulation, application of preventive measures, regular assessment and eradication measures, and local quarantine regulation to minimize spread to other regions are important measures to limit the spread of *P. hysterophorus* in Ethiopia. The possible control options available to the farmers would be mechanical, chemical, restoration of land by useful tree species, rehabilitation of biodiversity growing in the area, and classical biological control and control by utilization.

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