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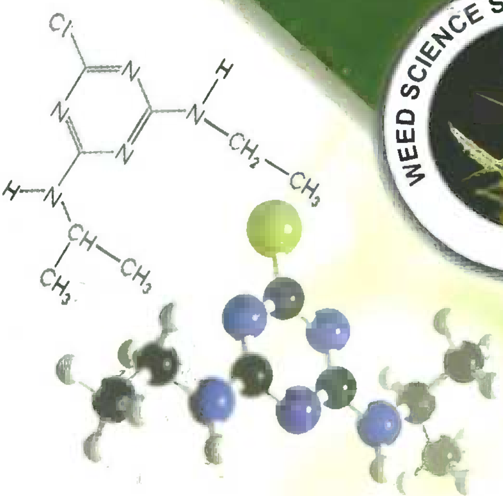

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1. Contributors do not have to be members, however, copies of journal and reprints will be provided free of cost to the members only.
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    Prasad, K. 1985. Efficacy of post-emergence weedicides applied at different stages in control of annual weeds of irrigated wheat under mid-Himalayan conditions. Pesticides 19(6): 20-22. [Weed Absts., 35(5): 1431;1986].

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WEED MANAGEMENT IN WHEAT THROUGH COMBINATION OF ALLELOPATHIC WATER EXTRACT WITH REDUCED DOSES OF HERBICIDES

A. Razzaq, Z.A. Cheema, K. Jabran, M. Farooq, A. Khaliq, G. Haider and S.M.A. Basra

ABSTRACT

Allelopathy has potential to tackle concerns associated with indiscriminate use of synthetic herbicides. In order to reduce herbicide usage, allelopathic crop water extracts with reduced herbicides doses were tested for weed management in wheat during the year 2005-06 at research farm of the Department of Agronomy, University of Agriculture, Faisalabad. Sorghum and sunflower water extracts (WE) combinations each at 18 L ha^{-1} with reduced doses by 70% of mesosulfuron + idosulfuron (Atlantis 3.6 WG), mesosulfuron + idosulfuron (Atlantis 12 EC), metribuzin + fenoxaprop (Bullet 38 SC), bensulfuron + isoproturon (Cleaner 70 WP) and metribuzin (Sencor 70 WP) were compared at their label doses which were sprayed alone 30 days after sowing for weed control in wheat. Label dose of isoproturon was used as a standard treatment and a weedy check was also maintained. Overall weed density, dry weed biomass and wheat grain yield were significantly influenced by different combination of plant water extracts with reduced (70%) doses of synthetic herbicides. Combination of sorghum + sunflower WE each at 18 L ha^{-1} with reduced dose of metribuzin + fenoxaprop significantly reduced dry weed biomass by 92% comparable to label dose of mesosulfuron + idosulfuron (93%). Treatment combination of sorghum + sunflower WE each at 18 L ha^{-1} with reduced dose of metribuzin + fenoxaprop by 70% produced maximum (2.82 t ha^{-1}) grain yield with 34 % increase over control and it was significantly higher than its label dose to the extent of 17% increase in grain yield.

Key words: Wheat, allelopathic crop water extracts, weed management.

INTRODUCTION

Allelopathy is existing in nature since centuries, but it has drawn special attention over couple of decades. Recently, it is exploited as a weed control strategy, alternative to the synthetic chemical herbicide

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(Bhowmik and Inderjit, 2003; Jabran *et al.*, 2008). It is environmentally safe, can conserve the available resources and also may mitigate the problems raised by synthetic chemicals (Rizvi and Rizvi, 1992; Duke *et al.*, 2001).

Sorghum (*Sorghum bicolor* L.) is one of the potential allelopathic crop (Jabran *et al.*, 2010a,b) containing a large number of secondary metabolites. Like sorghum, sunflower (*Helianthus annuus* L.) also has a strong allelopathic potential by which it can actively influence the growth of surrounding plants (Azania *et al.*, 2003). About 125 allelochemicals from different cultivars of sunflower, phytotoxic towards many weed species were isolated by Macias *et al.*, (2002). These chemicals are highly water soluble, can be released through root exudation, leaching from plants by rain, or decomposition of residues (Rice, 1984) effects of which depend upon species; concentration (Cheema and Ahmad, 1992) their movement, fate and persistence in soil (Inderjit *et al.*, 2001). These are also selective like synthetic herbicides (Weston, 1996). This characteristic could be used in weed management programmes and allelopathic crops may help in the development of biological herbicides.

Weeds must be controlled up to 80% or more depending upon the nature of the weeds and the extent of infestation to reduce the losses caused by weeds and to manage it to the economic threshold level. On average weeds cause 20-30% losses in different crops in Pakistan. Losses due to weeds exceed beyond Rs. 120 billion on national level, whereas wheat alone accounts for more than 30 billion. Losses due to weeds in major cereal crops of Pakistan exist in the range of Rs.40, 30 and 4 billion for rice, wheat and maize, respectively (Anonymous, 2005). Increasing problems related to weed control, such as herbicide resistant weed biotypes and residual effects of herbicides, increase the demand for organic farming, and increasing public concerns on environmental pollution issues require alternative farming systems which are less dependent on pesticide or based on naturally occurring compounds (Singh *et al.*, 2003; Waller, 2004; Jabran *et al.*, 2010a,b).

Allelopathy is an approach, being utilized in agriculture in various ways i.e. intercropping, allelopathic stubble mulches, allelopathic crops in rotation and inter/mixed cropping systems and aqueous extracts of allelopathic crops as spray (Fortney and Foy, 1985; Cheema, 1988; Narwal, 2000). An important approach is to use foliar sprays of different allelopathic crop water extracts for inhibiting weeds in field crops (Iqbal, 1997; Cheema and Ahmad, 1992; Jabran *et al.*, 2008 & 2010a, b). Extracts of many plant species can be successfully used as bioherbicide (Marwat *et al.* 2008) and thus the herbicides use could be reduced by combining the allelopathic water extracts with lower doses of herbicides (Jabran *et al.*, 2008 & 2010a,b). Mesosulfuron methyl @ 10.8 g a.i. ha^{-1} and ½ dose of isoproturon @ 500 g a.i. ha^{-1} + sorgaab @ 12 L ha^{-1} at

30 DAS were found most effective in reducing total weed density by 57-85% and 73-81%, respectively (Sharif *et al.*, 2005). Reduction in weed biomass and increase in wheat yield was observed by the application of sorghum (*Sorghum bicolor* L.) water extracts (Cheema *et al.*, 2003). Similar observations were made in other crops (Jabran *et al.*, 2008 & 2010a).

The recommended dose of isoproturon, for weed control in wheat, has been reduced by 50-60% if used in combination with sorgaab at 12 L ha^{-1} (Cheema *et al.*, 2003). Sorgaab @ 6 or 12 L ha^{-1} applied in combination with a low rate of sulfosulfuron (15 g a.i. ha^{-1}) at 30 DAS reduced *Avena fatua* L. density by 78-92% and dry weed biomass by 72-98% (Jamil *et al.*, 2007). One spray of sorgaab @ 12 L ha^{-1} mixed with sulfosulfuron (½ dose, @ 15 g a.i. ha^{-1}) at 30 DAS suppressed *Phalaris minor* L. density by 82-91% and dry weed biomass by 86-87%.

The corresponding suppression in weed density and dry weed biomass with a full dose of sulfosulfuron was 91-95% and 97-100%. Grain yields of wheat from treatments with sorgaab in combination with low doses of sulfosulfuron were the same as the recommended dose of sulfosulfuron (Jamil *et al.*, 2007). A large number of studies on allelopathy have been conducted in Pakistan; however, activities related to weed control in wheat by reduced doses of herbicides in combination with allelopathic water extracts are still not well documented internationally. Therefore, the aim of this study was to evaluate the effect of sorghum and sunflower water extracts with reduced doses of some early post emergence herbicides for weed control in wheat.

MATERIALS AND METHODS

Site and Soil

A one year field study was conducted at research farm, Department of Agronomy, University of Agriculture, Faisalabad (31.25° N, 73.09° E, and 184 m), Pakistan, during the year 2005-06. The soil type at the experimental site was sandy clay loam.

Crop husbandry

Wheat variety "Inqalab-91" was planted on a well prepared seed bed in 22 cm spaced rows with single row hand hoe using seed rate at 125 kg ha^{-1}. A basal dose of fertilizer @ 100 kg ha^{-1} N and 85 kg ha^{-1} P_2O_5 was used in the form of urea and diammonium phosphate, respectively. Half of the nitrogen and full dose of the phosphorus were applied at the time of sowing, while remaining half was applied at first irrigation. First irrigation was given after twenty days after sowing and subsequent irrigations were adjusted according to the climatic conditions and need of the crop.

Experimental details

The experiment was laid out in randomized complete block design (RCBD) with four replications in plots measuring 7 x 1.98 m^2. The study comprised different combinations of allelopathic water extracts and reduced doses of herbicides (70%) viz. Sorghum and sunflower WE tank mixed with bensulfuron + isoproturon (Cleaner 70 WP) @ 315 g a. i. ha^{-1}, metribuzin (Sencor 70 WP) @ 52.5 g a.i. ha^{-1}, metribuzin + fenoxaprop (Bullet 38 SC) @ 57 g a.i. ha^{-1}, mesosulfuron + idosulfuron (Atlantis 12 EC) @ 36 g a.i. ha^{-1}, and mesosulfuron + idosulfuron (Atlantis 3.6 WG) at 4.32 g a.i. ha^{-1}. The label doses of isoproturon (50 WP) @ 1000 g a.i. ha^{-1}, mesosulfuron + idosulfuron (Atlantis 3.6 WG) @ 14.40 g a.i. ha^{-1}, mesosulfuron + idosulfuron (Atlantis 12 EC) @ 120 g a.i. ha^{-1}, metribuzin + fenoxaprop @ 190 g a.i. ha^{-1}, bensulfuron + idosulfuron @ 1050 g a.i. ha^{-1} and metribuzin @ 175 g a.i. ha^{-1} alone were sprayed as early post emergence spray 30 days after sowing using knapsack hand sprayer fitted with T-jet nozzle. Isoproturon was used as standard treatment and a weedy check was also maintained.

Measurements

The data on weed density and dry weed biomass were recorded with the help of quadrate measuring 0.25 x 0.25 m^2 randomly placed at two places in respective experimental units at 70 days after sowing. Weeds were counted individually and then by cutting just above the ground for recording fresh weed biomass. Weeds were dried in an oven at 70 $^{\circ}$C for 48 hours for recording dry weed biomass. Ten plants were randomly selected from each plot and their plant height, sterile tillers, fertile tillers, spikelets per spike and grains per spike were noted. Three samples of 1000-grain each were taken randomly from the seed lot of each plot, weighed and then averaged to measure 1000-grain weight. Straw and grain yield were recorded on plot basis by mechanical threshing and converted into t ha^{-1}.

Statistical analysis

Data obtained were analyzed statistically by using statistical package MSTATC (Freed and Scott, 1986). Analysis of variance (ANOVA) was performed by using Fisher's analysis of variance technique while multiple comparisons among treatment means were made using least significant difference (LSD) test at $P \leq 0.05$ (Steel *et al.*, 1997).

RESULTS AND DISCUSSION

All the combination of water extracts and reduced doses (by 70%) of herbicides significantly reduced the weeds density (Table-1). Sorghum + sunflower water extracts (WE) each at 18 L ha^{-1} with 70% reduced dose of metribuzin + fenoxaprop Bullet 38 SC, mesosulfuron + idosulfuron (Atlantis 3.6 WG) full dose, metribuzin (Sencor 70 WP) full dose, sorghum + sunflower WE each @ 18 L ha^{-1} with 70% reduced dose of Atlantis 3.6 WG, sorghum + sunflower WE each @ 18 L ha^{-1} with 70% reduced dose of bensulfuron + isoproturon (Cleaner 70 WP), sorghum + sunflower WE + 70% reduced dose of mesosulfuron + idosulfuron (Atlantis 12EC), Bullet 38 SC full dose, and Cleaner 70 WP full dose reduced the *Phalaris minor* density by 92.31, 92.31, 92.31, 88.46, 88.46, 88.46, 88.46 and 84.62%, respectively at 70 DAS over control. All of these treatments of label doses of herbicides and their reduced doses up to 70% were statistically at par with each other. *Coronopus didymus* L. population was also significantly influenced by water extracts in combination with reduced doses of herbicides.

Full dose of Atlantis 3.6 WG at 70 DAS inhibited *Coronopus didymus* L. density by 94% followed by full dose of Sencor 70 WP, Atlantis 12 EC full dose, sorghum + sunflower WE each at 18 L ha^{-1} with 70% reduced dose of Cleaner 70 WP, Bullet 38 SC full dose, sorghum + sunflower WE each @ 18 L ha^{-1} with 70% reduced dose of Bullet 38 SC, sorghum + sunflower WE each @ 18 L ha^{-1} with 70% reduced dose of Atlantis 12 EC and sorghum + sunflower WE each @ 18 L ha^{-1} with 70% reduced dose of Atlantis 3.6 WG, which reduces the *Coronopus didymus* density by 90.6, 90.6, 88.24, 87.25, 87.25, 87.25 and 87.25% respectively, over control, and these were statistically at par with each other.

The population of *Phalaris minor* L. was significantly inhibited by all the treatments, (Table-1) but sorghum + sunflower WE each @ 18 L ha^{-1} with reduced dose of (Bullet 38 SC 70%) and Atlantis 3.6 WG full dose were the best treatments to reduce weed density. Bullet 38 SC reduced dose by 70% in combination with water extracts was statistically at par with full dose of (Atlantis 3.6 WG) due to synergistic effect of allelochemicals with synthetic herbicide.

The range of population reduction was 42 to 92% at 70 DAS over control. Similar results were found in case of *Phalaris minor* L. dry weed biomass (Table-2). Sorghum and sunflower water extracts each @ 18 L ha^{-1} combined with reduced doses by 70% of Cleaner 70WP and Atlantis 12EC gave the maximum reduction in dry weed biomass by 97.96% each, over control and these were followed by combination of allelopathic crop water extracts with reduced dose by 70% of Bullet 38SC which reduced the *Phalaris minor* L. dry weed biomass at 70 DAS, by 96.94% over control.

The results are supported by Cheema *et al.*, (2002a) that herbicide dose can be reduced in combination with allelopathic crop water extracts and by Cheema *et al.*, (2003) who reported that sorghum water extract can be used with reduced dose of herbicide for effective weed control in cotton.

Table-1. Effect of sorghum and sunflower water extracts (70 DAS) with reduced doses of herbicides on weed density in wheat.

Treatments	Rates ha^{-1}	Density of *Phalaris minor*	Density of *Coronopus didymus*
Control (weedy check)	-	6.50 a †	25.5 a
Isoproturon 50 WP	1000 g a.i.	2.0 bc (-69.23)	6.00 b (-76.47)
Bensulfuron+isoproturon (Cleaner 70 WP)	1050 g a.i.	1.0 c (-84.62)	4.75 c (-81.37)
Metribuzin (Sencor 70 WP)	175 g a.i.	0.50 c (-92.31)	2.50 d (-90.6)
Metribuzin + fenoxaprop (Bullet 38 SC)	190 g a.i.	0.75 c (-88.46)	3.25 d (-87.25)
Mesosulfuron + idosulfuron (Atlantis 12 EC)	120 g a.i.	3.75 b (-42.31)	2.50 d (-90.6)
Mesosulfuron + idosulfuron (Atlantis 3.6 WG)	14.4 g a.i.	0.50 c (-92.31)	1.00 e (-96.08)
Sorghum + sunflower WE + Sencor 70 WP	18 L each + 52.5 g a.i.	2.0 bc (-69.23)	4.25 c (-83.33)
Sorghum + sunflower WE + Cleaner 70 WP	18 L each + 315 g a.i.	0.75 c (-88.46)	3.00 d (-88.24)
Sorghum + sunflower WE + Bullet 38 SC	18 L each + 57 g a.i.	0.50 c (-92.31)	3.25 d (-87.25)
Sorghum + sunflower WE + Atlantis 12 EC	18 L each + 36 g a.i.	0.75 c (-88.46)	3.25 d (-87.25)
Sorghum + sunflower WE + Atlantis 3.6 WG	18 L each + 4.32 g a.i.	0.75 c (-88.46)	3.25 d (-87.25)
LSD value at P≤0.05		**2.106**	**0.840**

DAS= Days after sowing; WE = Water extracts.
†=Means not sharing a letter in common differ significantly by LSD at p≤0.05.
Figures in parenthesis show percent decrease in weeds density and dry weed biomass over control.

Dry weed biomass (Table-2) was also significantly influenced by different post emergence herbicides alone and in combination with allelopathic crop water extracts. Sorghum + sunflower WE each @ 18 L ha^{-1} combined with 70% reduced doses of Cleaner 70 WP and Atlantis 12 EC gave the maximum reduction in dry weed biomass by 98 and 98%, respectively, over control, followed by label doses of Sencor 70 WP and allelopathic crop water extracts of sorghum + sunflower each @ 18 L ha^{-1} combined with 70% reduced doses of Bullet 38 SC which reduced the *Phalaris minor* L. dry weed biomass at 70 DAS, by 96, and 95%, respectively, over control, and both of these were statistically at par with each other.

Similarly crop water extracts of sorghum and sunflower each @ 18 L ha^{-1} combined with 70% reduced doses of Sencor 70 WP, mesosulfuron + idosulfuron gave the maximum reduction by 94% in dry weed biomass of *Coronopus didymus* L., and these were followed by label dose of mesosulfuron + idosulfuron and its reduced dose 70% combined with crop water extracts controlled the dry weed biomass by 93% respectively, over control. Dry weed biomass was significantly influenced by all the treatment combinations.

Maximum reduction (94.29%) was found by label dose of Atlantis 3.6 WG and it was statistically at par with reduced Bullet 38SC reduced dose by 70% in combination with plant water extracts (94.29%). Dry weed biomass reduction by the different treatments ranged between 64.29 to 94.29%. These results support the hypothesis that allelopathic water extracts can be used to inhibit weeds or in combination with lower herbicide rate, thereby can considerably decrease their dose by 70 %. More over the concept of having additive or complementarily effects of allelopathic materials in mixture were noted as compared to their separate use. Cheema *et al.*, (2002b) reported that herbicide dose can be reduced in combination with allelopathic crop water extracts.

Grain yield was significantly enhanced by controlling weeds through post emergence herbicides alone and in combination with allelopathic crop water extracts (Table-2). Treatment of sorghum + sunflower WE each @ 18 L ha^{-1} with 70% reduced doses of Bullet 38SC produced maximum (2.82 t ha^{-1}) grain yield with 34.29 % increase over control and it was significantly more than its label dose. The grain yield in other treatments either at their label doses or with allelopathic water extracts were higher than the control and statistically at par with each others. The reason for increase in grain yield over control was the control of weeds and probably the allelopathic effects of crop water extracts promoted the wheat growth which ultimately increases grain yield (Jabran *et al.*, 2008).

The study suggests that reduction of herbicide doses by adding allelopathic crop water extracts would reduce the reliance on the synthetic herbicides. The combination of sorghum + sunflower WE each @ 18 L ha^{-1} with reduced dose by 70% of Bullet 38SC was found to be best treatment which produced maximum grain yield (2.82 t ha^{-1}) with 34.29% increase over control and synthetic herbicide dose reduced was 70%. Reduced herbicide doses in combination with allelopathic crop water extracts would minimize the use of synthetic pesticides, decrease the cost of production and increase the farm income.

Table-2. Effect of sorghum and sunflower water extracts with reduced doses of herbicides on dry weed biomass 70 DAS and grain yield of wheat.

Treatments	**Rates ha^{-1}**	**Dry weed biomass of *Phalaris minor***	**Dry weed biomass of *Coronopus didymus***	**Grain yield (t ha^{-1})**
Control (weedy check)	-	†1.96 a	0.70 a	2.10 c
Isoproturon 50 WP	1000 g a.i.	0.44 b (-77.56)	0.20 bc (-71.43)	2.40 b (+14.29)
Bensulfuron+isoproturon (Cleaner 70 WP)	1050 g a.i.	0.13 def (-93.37)	0.25 b (-64.29)	2.42 b (+15.24)
Metribuzin (Sencor 70 WP)	175 g a.i.	0.07 efg (-46.43)	0.16 bcde (-77.14)	2.55 b (+21.43)
Metribuzin + fenoxaprop (Bullet 38 SC)	190 g a.i.	0.15 d (-92.35)	0.06 def (-91.43)	2.46 b (+17.14)
Mesosulfuron + idosulfuron (Atlantis 12 EC)	120 g a.i.	0.29 c (-85.21)	0.18 bcd (-74.29)	2.38 b (+13.33)
Mesosulfuron + idosulfuron (Atlantis 3.6 WG)	14.4 g a.i.	0.34 c (-82.66)	0.04 f (-94.29)	2.62 ab (+24.76)
Sorghum + sunflower WE + Sencor 70 WP	18 L each + 52.5 g a.i.	0.04 g (-97.96)	0.12 cdef (-82.86)	2.52 b (+20.00)
Sorghum + sunflower WE + Cleaner 70 WP	18 L each + 315 g a.i.	0.10 defg (-94.90)	0.09 cdef (-87.14)	2.60 ab (+23.81)
Sorghum + sunflower WE + Bullet 38 SC	18 L each + 57 g a.i.	0.06 fg (-96.94)	0.04 f (-94.29)	2.82 a (+34.29)
Sorghum + sunflower WE + Atlantis 12 EC	18 L each + 36 g a.i.	0.04 g (-97.96)	0.05 ef (-92.86)	2.56 b (+21.90)
Sorghum + sunflower WE + Atlantis 3.6 WG	18 L each + 4.32 g a.i.	0.14 de (-92.87)	0.05 ef (-92.86)	2.53b (+20.48)
LSD value at P≤0.05		**0.788**	**0.120**	**0.241**

DAS= Days after sowing; WE = Water extracts.
†=Means not sharing a letter in common differ significantly by LSD at p≤0.05.
Figures in parenthesis show percent decrease of dry weed biomass and increase of grain yield over control.

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EVALUATION OF INTEGRATED WEED MANAGEMENT PRACTICES FOR SUGARCANE

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ABSTRACT

A field study was conducted at Sugarcane Research Institute, Faisalabad during 2008-2009 to evaluate integrated weed management for sugarcane, Randomized Complete Block Design, having three replications was used in the experiment. The treatments including (1) Ametryn + atrazine @ 3.75 kg ha^{-1} (2) Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows + inter-row cultivation, (3) hand weeding in cane rows + inter-row cultivation (4) inter-row cultivation only (5) hand hoeing twice and (6) weedy check. Statistical analysis of the data showed that weed density as well as yield related parameters were significantly affected by different treatments. In general weed management practices suppressed the weeds and increased the yield related traits. However, Ametryn + atrazine @ 3.75 kg ha^{-1} pre-emergence on cane rows only + inter-row cultivation was the most effective and economical than hand hoeing or inter-row cultivation by tractor. It was further concluded that chemical weed control along with one inter-row cultivation during tillering gave higher cane yield and cost benefit ratio (1:12.85) while the least cost benefit ratio (1:7.25) was observed in hand weeding alone.

Key Words: *Saccharum officinarum*, integrated weed management, chemicals, intercropping.

INTRODUCTION

Weed control is essential for economical crop production. Weeds reduce sugarcane yields by competing for moisture, nutrients, and light during its growing period. Khan *et al.* (2004) reported that cane yield is reduced to the extent of 20-25% due to weed infestation. Weed control prior to crop canopy spread is crucial. Heavy weed infestation hinders sugarcane harvesting by adding unnecessary harvesting expenses. Even a single weed plant growing to maturity may produce seeds that create problems for many years to come (Srivastava *et al.*, 2003). A good and uniform stand of sugarcane crop develops complete canopy that shades the spaces between the cane

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rows, which is very helpful in reducing weed competition. Cultivation can be an economical measure of suppressing weed growth. To ensure that the sugarcane plants get early advantage in the competition for sunlight, a height differential must be established between cane plants and weeds.

Herbicides can be useful and economical tools in increased sugarcane production. It is important that sugarcane crop has the initial competitive advantage against weeds. Pre-emergence herbicide applications, in conjunction with mechanical cultivation, helps to ensure the early season advantage (Chattha *et al.*, 2004), but the development of resistance in weeds and environmental safety are the major concerns against herbicide use.

Sugarcane has a prolonged growing season varying from 10-12 months, thus effective and timely control of weeds is an important component of its management. In its early stages, sugarcane germinates and grows very slowly, while weeds show a rapid growth due to the lack competition from the crop. If not checked timely, early tillering and growth of sugarcane is likely to be affected by weed competition. Singh *et al*. (1980) reported that critical period of weed control was between 30 and 120 days after planting sugarcane in Spring. Punzelan and Cruzz (1981) obtained maximum yield of cane when the crop was kept weed free from one to three months after planting, controlling weeds for longer periods did not enhance yields. It was further observed that weeds competition for one month from planting had no adverse effect on cane yields, whereas competition for two months reduced yield by 15% and for the whole season by 55%.

In Pakistan, traditional cultural practices, like hand weeding and inter-row cultivation by bullock drawn implements, are employed to eradicate weeds from cane fields. This practice is more often common with small growers using family labor for cultural operations. In recent past the chemical weed control has been introduced in the country. Several pre- and post-emergence weedicides have been introduced, of which Gesapax Combi (amtry+atrazine) is getting widely popular. Singh *et al*. (2008) observed that simazine (chemical name) and atrazine (chemical name) gave best control of weeds in cane fields and increased tillering. Luke (2007) revealed that Gesapax Combi, effectively controlled weeds in sugarcane and thus resulted in substantial increase in the yield of sugarcane. Beg (1975) reported that Gesapax Combi applied at 2.2-2.8 kg ha^{-1} controlled all types of broad leaved and narrow leaved weeds in sugarcane. The residual effect of pre-emergence application of Gesapax was observed to last for three months and it was no longer necessary to hoe the crop until the lines had closed up at the advanced stage of growth. Gill (1978) compared the efficiency of different herbicides and mechanical weed control. Gesapax Combi gave better check on weed population and produced the highest yield of cane and was found economical than hand weeding. Any chemical or mechanical treatment of weed control did not affect the juice quality. Fayadomi (1983) did not find the effect of weedicide on juice quality. There is no sound data available in Pakistan on the comparative efficiency of different weed control methods in sugarcane. The present work was therefore, undertaken to study the economic efficiency of different methods for reducing weeds in sugar-cane fields as well as their effect on crop yield.

MATERIALS AND METHODS

The study was conducted to ascertain the effect of integrated weed management practices on sugarcane yield and yield related parameters in the research area of Sugarcane Research Institute, Faisalabad. The study was conducted during 2008-09 on cane variety CPF-246. The experiment was planted during March and laid out in randomized complete block design with three replications having a plot size of 10 x 4.8 m^2 keeping 1.2 meter apart double row strips. A seed rate of 50,000 triple budded setts (TBS) ha^{-1} and NPK fertilizer dose of 168-112-112 kg ha^{-1} was used. The treatments included in the experiment were Ametryn + atrazine @ 3.75 kg ha^{-1} + earthing up
Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up, Hand weeding in cane rows + inter-row cultivation + earthing up, Inter-row cultivation only + earthing up, Hand weeding twice + earthing up and the Weedy check

The Ametryn + atrazine @ 3.75 kg ha^{-1} was applied as pre-emergence weedicide with knapsack sprayer within 14-15 days after planting, spray volume was approximately 350 L ha^{-1}. Weed control in each treatment was evaluated by recording the weed density data Four quadrats (1 m^2) were randomly placed in each treatment and the weeds were counted and pooled. Subsequently, mean m^{-2} was obtained for individual weed species. Data were recorded on germination, tillers per plant, number of millable canes and cane yield per plot. Cane juice was analyzed for Brix, Pol, CCS and recovery %age. The data were analyzed statistically by analysis of variance and least significance difference test was applied to compare the differences in treatment means (Steel & Torrie, 1980). The input costs of weed control treatments were also computed to compare the economic efficiency of each treatment.

RESULTS AND DISCUSSION

Weed density (m^{-2})

The data on weed density and biomass (Table-1) showed significant differences among different treatments. The weedy check showed the largest weed density. Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up offered an effective control of weeds followed by Ametryn + atrazine @ 3.75 kg ha^{-1} + earthing up. The data also showed that the *Cyperus rotundus* was not completely controlled by Ametryn + atrazine and re-sprouted after hand weeding or cultivation. Weeds other than nutsedge were eliminated to variable extent. These include mostly dicots like *Trianthema portulcastrum*, *Convolvulus arvensis* and *Euphorbia helioscopia*. It was however, observed that weed density of nutsedge though not completely checked but its growth was curtailed. Plants turned pale and weak and did not compete with the growth of cane plants. Hand weeding alone could not control weeds as effectively as herbicide alone. Hand weeding also could not control or check the growth of new weeds after irrigation. Ametryn + atrazine suppressed the further growth of weeds till a complete canopy cover was achieved. These findings are in a great analogy with the work of Khan *et al.* (2001) who reported an effective control of weeds with application of Gesapax Combi and reported better control of *Conyza stricta* with 2,4-D as compared to Gesapax Combi.

Table-1. Effect of different weed management practices on the weed density (m^{-2}) and biomass in sugarcane crop during 2008-09.

Treatment	*Cyperus rotundus*	*Trianthema portulcastrum*	*Convolvulus arvensis*	*Euphorbia helioscopia*	Weed Biomass 75 DAS(g m^{-2})
Ametryn + atrazine @ 3.75 kg ha^{-1} + earthing up	72d	8cd	6c	10b	255 d
Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up	78c	10c	5cd	5c	248 d
Hand weeding in cane rows + inter-row cultivation + earthing up	80c	8cd	5cd	3c	286 c
Inter-row cultivation only + earthing up	87b	29b	10b	10b	358 b
Hand hoeing twice + earthing up.	70d	5d	3d	0d	258 d
Weedy check	120a	70a	15a	15a	405 a
$LSD_{0.05}$	4.52	3.38	3.38	2.29	35

Dry biomass (g m^{-2}) 75 days after sowing

Weed biomass was significantly affected by different weed management practices (Table-1). Maximum dry weed biomass (405 g m^{-2}) was recorded in weedy check followed by inter-row cultivation and no weeding in cane rows by producing 358 g m^{-2} while third position with respect of dry biomass was occupied by hand weeding in cane rows + inter-row cultivation. Minimum weed biomass was in T_1 and T_2 and T_5 treatments which were statistically at par with respect to dry biomass production and also produced better cane yield than rest of the weed management practices under study (Table-2).

Germination %

Germination %age data in Table-2 showed that germination was not affected by different weed control practices hence the differences among the different treatments are non-significant. The application of herbicides and other treatments thus, had no effect on germination of sugarcane.

Number of tillers $plant^{-1}$

The data presented in Table-2 revealed that high weed density depressed the tillering significantly in the weedy check, which produced the lowest No. of tillers (2.39) $plant^{-1}$. However, the variation among the various treatments could reach the statistical significance level.

Number of millable canes

The data on millable canes (Table-2) showed that cane density was almost inversely proportional to weed density. Weeds have direct bearing on tillering and ultimately production of millable canes. Thus weedy check produced the minimum number of millable canes. Millable canes were also reduced when only inter-row cultivation was given. The weeds left in intra-row spaces affected the millable canes adversely. The highest millable canes were produced by Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up, but at par with all other treatments included in the study except weedy check and Inter-row cultivation only + earthing up. These finding are corroborated with the previous work of Raskar (2004) who obtained highest cane yield with the sequential application of 2,4-D as post after the pre-emergence application of metribuzin.

Cane yield tons ha^{-1}

The cane yield data presented in Table-2 revealed significant differences among different management practices as compared to the weedy check which produced the lowest yield of cane. The highest cane yield was determined where Ametryn + atrazine was applied followed by inter-row cultivation at tillering while cane yield in rest of the treatments was higher than the weedy check, but non-significant to one another.

Yield data showed that highest percentage of 44.6 of Gain in yield was obtained in treatment Ametryn + atrazine @ 3.75 kg ha^{-1} + inter-row cultivation and earthing up in May, while hand hoeing twice + earthing-up gave 41.6% increase as compared to weedy check. While Ametryn + atrazine @ 3.75 kg ha^{-1} + earthing up and hand hoeing in cane rows + inter-row cultivation and earthing up both gave 35% increase over weedy check. Thus combination of herbicide Ametryn + atrazine as pre-emergence and inter-row cultivation at tillering + earthing in May was found to be the most effective for control of weeds in sugarcane. The findings of Beg (1975), Gill (1978), Luke (2007) and Singh *et al*., (2008) also support the effectiveness of Gesapax Combi (ametryn + atrazine) as selective herbicides for sugarcane.

Table-2. Effect of different weed management practices on cane density and yield of sugarcane during 2008-09.

Treatment	Germination %	No. of Tillers plant^{-1}	No. of Mill able canes (000 ha^{-1})	Cane Yield in tons ha^{-1}	% Gain in yield.
Ametryn + atrazine @ 3.75 kg ha^{-1} + earthing up	40	2.79	91,400 ab	75.60 ab	35
Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up	42	3.01	1,06,667 a	81.00 a	44.6
Hand weeding in cane rows + inter-row cultivation + earthing up	39.5	2.75	91,445 ab	75.56 ab	34.9
Inter-row cultivation only + earthing up	39.5	2.61	83,593 b	70.45 b	25.8
Hand hoeing twice + earthing up.	42	2.67	93,757 ab	79.30 ab	41.6
Weedy check	39	2.39	67,026 c	56.00 c	-
LSD $_{0.05}$	**NS**	**NS**	**15,270**	**9.3**	

Cane juice quality

The cane juice analysis for Brix, Pol, purity, CCS and recovery CCS are presented in Table-3. The data in respect of different management practices on cane juice quality revealed that various treatments did not show marked differences in quality of cane. However, cane yield variation, if any would deficiently affect the sugarcane yield per hectare. The results are in conformity with those of Fayadomi (1983) and Gill (1978).

Table-3. Effect of different weed management practices on cane juice quality of sugarcane variety CPF-246 during 2008-09 (cane juice analysis).

Treatment	Brix %	Pol %	Purity %	Fiber %	CCS %	Recovery CCS %
Ametryn + atrazine @ 3.75 kg ha^{-1} + earthing up	20.93	17.74	84.77	12.75	13.85	13.01
Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up	20.87	17.44	83.56	12.85	13.90	13.07
Hand weeding in cane rows + inter-row cultivation + earthing up	21.00	17.45	83.10	12.99	13.90	13.07
Inter-row cultivation only + earthing up	22.00	18.60	84.49	13.01	13.80	12.97
Hand hoeing twice + earthing up.	21.60	17.90	82.87	12.87	13.95	13.11
Weedy check	20.80	17.50	84.13	13.10	12.93	12.15
$LSD_{0.05}$	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**

Cost: benefit ratio of different weed management practices

The gross value of increased yield over weedy check and cost: benefit ratio of various weed control treatments are presented in Table-4. The data revealed that hand hoeing is more expensive than the use of herbicide alone. The highest cash return were obtain in Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up. Cost benefit ratio of 1:12.85 and 1:11.46 was observed in Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up and Ametryn + atrazine @ 3.75 kg ha^{-1} + earthing up treatments, respectively as against 1:7.25 in hand weeding twice alone which shows economic efficiency of herbicide application for control of weeds and the yield increment in sugarcane.

CONCLUSION

From the foregoing discussion it could be concluded that use of Ametryn and atrazine @ 3.75 kg ha^{-1} as pre-emergence herbicide is most effective means of weed control in sugarcane fields. The chemical control of weeds is also cheaper than manual / mechanical control measures as the former method showed high cost benefit ratio than the later ones.

Table-4. Economics of different weed management practices and their cost benefit ratio in sugarcane crop during 2008-09.

Treatment	Mean yield t ha^{-1}	Increase over check plot in t ha^{-1}	Gross value of increased yield ($Rs.ha^{-1}$)	Added Cost in Rs. ha^{-1}	Increase in cash return over weedy check	Cost benefit ratio
Ametryn + atrazine @ 3.75 kg ha^{-1} + earthing up	75.60	19.60	39200	3145	36055	1:11.46
Ametryn + atrazine @ 3.75 kg ha^{-1} on cane rows only + inter-row cultivation + earthing up	81.00	25.00	50000	3608	46392	1:12.85
Hand weeding in cane rows + inter-row cultivation + earthing up	75.56	19.56	39120	4700	34420	1:7.32
Inter-row cultivation only + earthing up	70.45	14.45	28900	2500	26400	1:10.56
Hand hoeing twice + earthing up.	79.30	23.30	46600	5650	40940	1:7.25
Weedy check	**56.00**	-	-	-	-	-

Cane @ Rs. 80/ 40 Kg or Rs. 2000 /ton.
Ametryn + Atrazin @ 3.750 kg ha^{-1} @ Rs. 420/ kg + 1 man day for application charges @ Rs. 220 ha^{-1} & rent of sprayer Rs. 100 ha^{-1}
One hoeing with 10 men day ha^{-1} @ Rs. 220 per man day
One inter row cultivation with tractor Rs. 1250 ha^{-1}
Hand weeding 10 men day @ Rs. 220 per man day.

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RESPONSE OF CHICKPEA (*Cicer arietinum*) AND *Euphorbia dracunculoides* TO PRE AND POST-EMERGENCE HERBICIDES

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ABSTRACT

A pot trial was conducted at the Agronomic Research Farm, University of Agriculture, Faisalabad, Pakistan in 2007-08 to evaluate nine pre and post emergence herbicides. Subsequently, a field experiment was conducted during winter 2008-09 to study the effect of herbicides selected from pot trial on weeds and growth and yield of chickpea at farmer's field in district Bhakar, Punjab, Pakistan. Stomp-455CS at 1875, 2000mL ha^{-1}, Dualgold-960 EC at 1500 mL ha^{-1}, Buctril super-60 EC at 1125 mL ha^{-1}, Aim-40 DF at 75 g ha^{-1}, Starane-M 60 EC at 1125 mL ha^{-1}, 1250 mL ha^{-1}, Sencor-70 WDG at 375 and Topgrow-90 WDG at 700 g ha^{-1} were applied as pre-emergence spray while aim-40 DF at 75 g ha^{-1} was also applied as pre-emergence sand mix broadcast application. Basagran-48 SL at 1500 mL ha^{-1} was applied as a post-emergence spray. The results revealed that all the herbicides (except Aim-40 DF) applied as sand mixed broadcast application caused 100% crop mortality in pot trial. Generally control of Euphorbia dracunculoides was 30 to 92%. In field trial all herbicides except Dualgold-960 EC gave 82 to 100% control of Asphodelus tenuifolius and control of Carthamus oxycantha ranging from 75 to 100% with all herbicides except Dualgold-960 EC, Starane-M60EC at 1125 mL ha^{-1} both as pre-emergence spray and Aim-40 DF at 75 g ha^{-1} as sand mix broadcast application. Whereas, control of Euphorbia dracunculoides with these herbicides except Dualgold-960 EC was 72 to 100%. Basagran-48 SL at 1500 mL ha^{-1} caused 100% crop mortality followed by 79, 46 and 43% in Sencor-70 WDG at 375 g ha^{-1}, Starane-M60 EC at 1125mL ha^{-1} and 1250 mL ha^{-1}, respectively. Pre-emergence application of Stomp-455 CS, Aim-40 DF and Top grow-90 WDG each as pre-emergence spray recorded more than 40% increase in grain yield of chickpea over check.

Key words: Weed management, herbicides crop injury, chemical control.

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INTRODUCTION

Chickpea (*Cicer arietinum*) the major grain legume of Pakistan, suffers from yield losses of 24-63% (Tanveer *et al*., 1998) and 38% (Aslam *et al*., 2007) on account of infestation with weeds. Yield losses due to weeds in chickpea depend on the level of weed infestation, weed species prevailing, competition duration, management practices and climatic conditions. No doubt cultural and mechanical methods of weed control are effective but can not be adopted on large scale being labour intensive and costly. This necessitates provision of suitable options of herbicides to the farmers that can be cost effective and cope with the scarcity of labour at the time of need. Weed control with herbicides would be advantageous for optimizing input efficiency in a particular crop, by reducing the population of weeds. Substantial control of weeds and significant increase in grain yield of chickpea with different herbicides has been reported by Yadav *et al*. (2006), Malik *et al*. (2001), Marwat *et al*. (2004), Ghosheh and Shatnawi (2005) and Yadav *et al*. (2007).

According to Willoughby *et al.,* (1996) use of herbicides must provide adequate weed control without adversely affecting seedling emergence and survival. Tolerance of seedling to herbicides is dependent on the herbicide dose, cultivar and environmental conditions (VanGessel *et al*., 2000). The earlier research results indicated the option of using pre-emergence pendimethalin (Aslam *et al*., 2007) in chickpea. However, there is a need to identify and test newer herbicides over a wide range of rates for weed control in chickpea. The literature related to the effect of herbicides on weed and growth parameters of chickpea is, however quite meager in Pakistan. *E. dracunculoides* L. (Green Spurge: family Euphorbiaceae) is a much branched annual winter weed. In Pakistan it grows in October-Novembar in rainfed areas of chickpea-chickpea cropping system and matures in April. It is one of the most serious weeds of chickpea in addition to *A. tenuifolius.*

Therefore, an investigation was undertaken to evaluate different herbicides for effective and broad spectrum weed control in chickpea. This information will also help in evaluating the selectivity of a particular herbicide in chickpea.

MATERIALS AND METHODS

A. Herbicide Screening Experiment

A pot experiment was carried out at the Agronomic Research Farm, University of Agriculture, Faisalabad, Pakistan to evaluate the response of chickpea (*Cicer arietinum*) and *E. dracunculoides* to various pre- and post-emergence herbicides. The experiment was laid out in completely randomized design (CRD) with four replications. The variety sown was Bital-98. Ten seeds each of chickpea and *E.*

dracunculoides were sown on 20th November, 2007 in each pot. Pot size was 27.5 cm × 25 cm^2. Herbicides applied along with dose, time and method of application are given in Table-1.

Table-1. Herbicides used, time and their method of application in pot experiment on chickpea.

Herbicide	Dose ha^{-1}	Time of application	Method of application
Weedy check	-	-	-
Stomp-455 CS (pendimethalin)	1875 mL	Pre	Spray
Stomp-455 CS (pendimethalin)	2000 mL	Pre	Spray
Stomp-455 CS (pendimethalin)	1875 mL	Pre	Sand mix Broad
Stomp-455 CS (pendimethalin)	2000 mL	Pre	Sand mix Broad
Dualgold-960 EC (S-metolachlor)	1500 mL	Pre	Spray
Dualgold-960EC (S-Metolachlor)	1500 mL	Pre	Sand mix Broad
Bestrazine-38 SC (atrazin)	1875 mL	Pre	Spray
Bestrazine-38 SC (atrazin)	1875 mL	Pre	Sand mix Broad
Bestrazine-38 SC (atrazin)	1250 mL	Post	Spray
Starane-M 60 EC (fluroxypyr+MCPA)	1125 mL	Pre	Spray
Starane-M 60 EC (fluroxypyr+MCPA)	1125 mL	Pre	Sand mix Broad
Starane-M 60 EC (fluroxypyr+MCPA)	1250 mL	Pre	Spray
Starane-M 60 EC (fluroxypyr+MCPA)	1250 mL	Pre	Sand mix Broad
Buctril-super-60EC (bromoxynil+MCPA)	1125 mL	Pre	Spray
Buctril-super-60EC (bromoxynil+MCPA)	1125 mL	Pre	Sand mix Broad
Sencor-70WDG (metribuzin)	375 g	Pre	Spray
Sencor-70WDG (metribuzin)	325 g	Pre	Sand mix Broad
Top grow- 90 WDG (terbutryn)	700 g	Pre	Spray
Top grow- 90 WDG (terbutryn)	700 g	Pre	Sand mix Broad
Aim-40 DF (carfentrazone ethyl)	75 g	Pre	Spray
Aim-40 DF (carfentrazone ethyl)	75 g	Pre	Sand mix Broad
Basagran-48SL (bentazon)	1500 mL	Post	Spray
Basagran-48SL (bentazon)	2000 mL	Post	Spray

Pre = Pre-emergence, Post = Post-emergence, Broad = Broadcast

Calibration was done to know the exact volume of water and sand to spray and broadcast herbicide, respectively. The area of pot was compared with the area of an acre then the exact volume of herbicide was applied in each pot.

B. Field experiment

The field experiment was conducted during November 2008-May 2009, to study the effect of different herbicides selected from preliminary screening in pot trial, on *E. dracunculoides* and yield of chickpea at farmer's field in district Bhakar. The experiment was quadruplicated in a randomized complete block design measuring a plot size of 4.0 x 1.2 m^2 (Table-2). Chickpea variety Bital-98 was sown in October with a tractor driven drill in 30 cm apart rows.

Table-2. Herbicides used and their time and method of application in field experiment on chickpea.

Herbicide	Dose ha^{-1}	Time of application	Method of Application
Weedy check	-	-	-
Stomp-455 CS (pendimethalin)	1875 mL	Pre	Spray
Stomp-455 CS (pendimethalin)	2000 mL	Pre	Spray
Basagran-48SL (Bentazon)	1500 mL	Post	Spray
Dualgold-960EC (S-Metolachlor)	1500 mL	Pre	Spray
Buctril-super-60EC (Bromoxynil+MCPA)	1125 mL	Pre	Spray
Aim-40 DF (Carfentrazone ethyl)	75 g	Pre	Spray
Aim-40 DF (Carfentrazone ethyl)	75 g	Pre	Sand mix Broadcast
Starane-M 60 EC (Fluroxypyr+MCPA)	1125 mL	Pre	Spray
Starane-M 60 EC (Fluroxypyr+MCPA)	1250 mL	Pre	Spray
Zencor-70WDG (Metribuzin)	375 g	Pre	Spray
Top grow-90 WDG (Terbutryn)	700 g	Pre	Spray

Pre = Pre-emergence, Post = Post-emergence

Statistical Analysis

The data collected were analyzed statistically using Fisher's analysis of variances technique and treatment means showing F-values significant were compared using least significant difference (LSD) test at 0.05 probability level (Steel *et al.,* 1997).

RESULTS AND DISCUSSION

Pot experiment Emergence (%)

Seed emergence is the fundamental necessity for achieving a desirable plant population and ultimately a good yield. Data regarding emergence of chickpea as influenced by different pre and post emergence herbicides at different rates along with their application methods are given in Table-3. It revealed that maximum emergence

(100 %) was recorded in pots where Basagran-48 SL at 2000 mL ha^{-1} was sprayed as post-emergence. It was followed by Stomp-455 CS at 1875 mL ha^{-1} pre-emergence spray, Stomp-455 CS at 2000 mL ha^{-1} pre-emergence spray, Dualgold-960 EC at 1500 mL ha^{-1} pre-emergence spray, Bestrazin-38 SC at 1875 mL ha^{-1} as pre-emergence spray and at 1250 ml ha^{-1} as post-emergence spray and Aim-40 DF at 75 g ha^{-1} as pre-emergence spray. All these treatments showed exactly similar chickpea emergence (97.50%). No chickpea emergence was observed in pots where sand mix broadcast application of herbicides except Aim-40 DF at 75 g ha^{-1} as pre-emergence sand mix broadcast (92.50%) was done. These results are in confirmation with those of Yadav *et al.* (2006) who reported that herbicides affected the germination of chickpea.

Euphorbia dracunculoides **control (%)**

The data related to control of *E. dracunculoides* are given in Table-3 and demonstrated that all herbicide applications at different stages and rates along with their application methods gave varying control of *E. dracunculoides.* Maximum control (92.50 %) was observed in pot where Bestrazin-38 SC at 1875 mL ha^{-1} was broadcasted after mixing with sand. It was followed by Sencor-70 WDG at 375 g ha^{-1} sand mixed broadcast (90.00 %) and Stomp-455 CS at 2000 mL ha^{-1} sand mixed broadcast (85.00 %), Starane M-60 EC at 1125 mL ha^{-1} pre-emergence spray (85.00 %), Dualgold-960 EC at 1500 mL ha^{-1} sand mixed broadcast (82.50 %), Starane M-50 EC at 1125 mL ha^{-1} sand mixed broadcast (80.00), Topgrow-90 WDG at 700 g ha^{-1} sand mixed broadcast (80.00), and Buctril Super-60 EC at 1125 mL ha^{-1} sand mixed broadcast (77.50 %), Stomp-455 CS at 1875 mL ha^{-1} sand mixed broadcast (77.50 %) and Aim-40 DF at 75 g ha^{-1} pre-emergence spray (75.00 %). Minimum control (30.00 %) of *E. dracunculoides* was recorded in Starane M-60 EC at 1250 mL ha^{-1} pre-emergence spray. Bestrazin-38 SC at 1875 mL ha^{-1} pre-emergence spray and Basagran-48 SL at 1500 mL ha^{-1} as post-emergence spray gave 45 and 60 % control, respectively.

Field experiment

Weed flora encountered in the field was *E. dracunculoides*, *Asphodelus tenuifolius* and *Carthamus oxyacantha* with *E. dracunculoides* as a predominant weed. Application of Basagran-48 SL at 1500 mL ha^{-1} as post-emergence spray showed 100% control of *E. dracunculoides* which was followed by Starane-M-60 EC at 1125 and 1250 mL ha^{-1} as pre-emergence application (85%). In other treatments control of *E. dracunculoides* ranged between 60-82.50%. Control of *A. tenuifolius* was also maximum (100%) with Basagran-48 SL at 1500 mL ha^{-1} as post-emergence spray and minimum (31.46%) with Dual gold-960 EC as a pre-emergence spray. In all other treatments control of *A. tenuifolius* varied from 82 to 89%. Best

(100%) control of *C. oxyacantha* was recorded again with Basagran-48 SL at 1500 mL ha^{-1} as post-emergence spray followed by 88.63% in Stomp-455 CS at 2000 mL ha^{-1}. Minimum control of *C. oxyacantha* (43.18%) was recorded with pre-emergence spray of Starane-M-60 EC at 1125 mL ha^{-1} (Table-4). Application of Stomp-455 CS at 1875 and 2000 mL ha^{-1} and Top-grow at 700 g ha^{-1} both as pre-emergence spray did not affect the plant population of chickpea (Table-5). Whereas all other herbicides caused crop mortality ranging from 0.80 to 100% with maximum (100%) in Basagran-48 SL at 1500 mL ha^{-1} and minimum (0.80) in Aim-40-DF at 75 g ha^{-1} as sand mixed broadcast application compared with weedy check.

The application of Stomp 455-CS at 1815 mL ha^{-1} as a pre-emergence spray gave maximum seed yield of chickpea (2234 kg ha^{-1}) and remained at par with Aim -40- DF at 75 g ha^{-1}, Stomp 455-CS at 2000 mL ha^{-1} and Top-grow both applied as pre-emergence spray with grain yields of 2187, 2192 and 2213 kg ha^{-1}, respectively. These were followed by Aim-40 DF at 75 g ha^{-1}as sand mix broadcast application with grain yield of 2119 kg ha^{-1}. Starane-M-60 EC at 1250 mL ha^{-1} as a pre-emergence spray gave grain yield (1505 kg ha^{-1}) statistically similar to weedy check (1552 kg ha^{-1}). Starane-M-60 EC at 1125 mL ha^{-1} and Sencor-70 WDG at 375 g ha^{-1} as pre-emergence spray significantly decreased grain yield over weedy check and this decrease was 18.12 and 66.76 %, respectively due to significant decrease in plant population of chickpea in these treatments. On the basis of one year results, it could be concluded that application of Stomp-455 CS at 1875, 2000 mL ha^{-1}, Aim-40-DF at 75 g $ha^{-1}$and Top-grow-90 WDG at 700 g ha^{-1} as a pre-emergence spray provide an option to farmers to manage *E. dracunculoides* and other weeds effectively along with improved growth leading to higher grain yield of chickpea.

The weed control achieved through Dualgold-960EC was not adequate. Less effect of Dualgold-960EC could be attributed to its photodecomposition which is a major contributor to its dissipation in the dry field condition (EXTOXNET, 2000). Inadequate control of *C. oxyacantha* with Starane M 60EC at 1125 mL ha^{-1} might be due to lower dose for this weed or erratic behavior of herbicide due to soil moisture compared with other weeds control (Martin, 1995). Severe crop mortality with Basagran-48SL, Starane-M 60EC and Sencor-70WDG might have resulted from drought stress and fluctuating temperatures (Anonymous, 1995). The highest yield from pre-emergence application of Stomp-455CS, Dualgold-960EC, Buctril super-60EC, Aim-40DF and Top grow-90WDG was in accordance with highest values of plant population which is the main yield attributing character, compared with other herbicides. The role of yield attributing factors and enhanced yield on account of chemical control of weeds has been documented earlier (Patel *et al*., 1997; Malik *et al*., 2001, Marwat *et al*., 2004; Ghosheh and Shatnawi,

Table-3. Effect of pre and post-emergence herbicides on emergence (%) of chickpea and control (%) of *Euphorbia dracunculoides* in pot experiment.

Herbicide	Dose ha^{-1}	Time of application	Method of application	Emergence (%) of Chickpea	Control (%) *E. dracunculoides*
Weedy check	-	-	-	95.00	-
Stomp-455CS	1875 mL	Pre	Spray	97.50	55.00
Stomp-455CS	2000 mL	Pre	Spray	97.50	60.00
Stomp-455CS	1875 mL		Sand mix Broad.	0.00	77.50
Stomp-455CS	2000 mL		Sand mix Broad.	0.00	85.00
Dualgold-960EC	1500 mL	Pre	Spray	97.50	62.50
Dualgold-960EC	1500 mL		Sand mix Broad.	0.00	82.50
Bestrazine-38 SC	1875 mL	Pre	Spray	97.50	45.00
Bestrazine-38 SC	1875 mL		Sand mix Broad.	0.00	92.50
Bestrazine-38 SC	1250 mL	Post	Spray	97.50	70.00
Starane-M 60 EC	1125 mL	Pre	Spray	57.50	85.00
Starane-M 60 EC	1125 mL		Sand mix Broad.	0.00	80.00
Starane-M 60 EC	1250 mL	Pre	Spray	80.00	30.00
Starane-M 60 EC	1250 mL		Sand mix Broad.	0.00	82.50
Buctril-super-60EC	1125 mL	Pre	Spray	95.00	72.50
Buctril-super-60EC	1125 mL		Sand mix Broad.	0.00	77.50
Sencor-70WDG	375 g	Pre	Spray	92.50	77.50
Sencor-70WDG	325 g		Sand mix Broad.	0.00	90.00
Top grow-90 WDG	700 g	Pre	Spray	92.50	82.50
Top grow- 90 WDG	700 g		Sand mix Broad.	0.00	80.00
Aim-40 DF	75 g	Pre	Spray	97.50	75.00
Aim-40 DF	75 g		Sand mix Broad.	92.50	72.50
Basagran-48SL	1500 mL	Post	Spray	95.00	60.00
Basagran-48SL	2000 mL	Post	Spray	100.00	70.00

Pre = Pre-emergence, Post = Post-emergence

Table-4. Species wise weed control efficiency (%) as affected by herbicides and application methods.

Treatment	Dose ha^{-1}	Time of application	Method of application	*Asphodelus tenuifolius*	*Carthamus oxyacantha*	*Euphorbia dracunculoides*
Weedy check	-	-	-	-	-	-
Stomp-455 CS	1875 mL	Pre	Spray	89.33	84.09	75.0
Stom-455(CS)	2000 mL	Pre	Spray	86.80	88.63	80.0
Basagran-48SL	1500 mL	Post	Spray	100	100	100.0
Dualgold-960EC	1500 mL	Pre	Spray	31.46	45.45	62.0
Buctril-super-60EC	1125 mL	Pre	Spray	82.36	88.63	72.50
Aim-40 DF	75 g	Pre	Spray	88.38	77.27	75.0
Aim-40 DF	75 g	Pre	Sand mix Broad cast	85.20	65.90	72.50
Starane-M 60 EC	1125 mL	Pre	Spray	86.27	43.18	85.0
Starane-M 60 EC	1250 mL	Pre	Spray	85.85	86.36	85.0
Zencor-70WDG	375 g	Pre	Spray	87.32	77.27	77.50
Top grow- 90 WDG	700 g	Pre	Spray	88.38	75.0	82.50

Pre = Pre-emergence, Post = Post-emergence

Table-5. Plant population and yield of chickpea as influenced by herbicides and application methods under field conditions.

Herbicide	Dose ha^{-1}	Time of application	Method of application	Plant Population (1.2m x 4m)	Crop Mortality %	Grain yield Kg ha^{-1}	% yield decrease (-) / % yield increase (+) over weedy check
Weedy check	-	-	-	124.50 a	-	1552.0d	-
Stomp-455CS	1875 mL	Pre	Spray	124.75a	-	2234.3a	+ 43.94
do	2000 mL	do	do	125.25a	-	2192.5a	+ 41.26
Basagran-48SL	1500 mL	Post	Spray	0.00g	100	0.00g	-100
Dualgold-960EC	1500 mL	Pre	Spray	122.75b	1.40	1848.8c	+ 19.12
Buctril-super-60EC	1125 mL	do	Spray	121.75c	2.21	1869.5c	+ 20.45
Aim-40 DF	75 g	do	Spray	121.00c	2.81	2187.3a	+ 40.93
do	do	do	Sand mix broadcast	123.50b	0.80	2119.7b	+ 36.58
Starane-M 60 EC	1125 mL	do	Spray	66.25e	46.25	1270.8e	-18.12
do	1250 mL	do	Spray	69.25d	43.50	1505.0d	-3.02
Zencor-70WDG	375 g	do	Spray	25.75f	79.31	515.75f	-66.76
Top grow- 90 WDG	700 g	do	Spray	125.25a	-	2213.3a	+ 42.60
LSD				**0.931**		**49.40**	

Pre = Pre-emergence, Post = Post-emergence

Means sharing the same letter in a column do not differ significantly at $P < 0.05$.

2005; Yadav *et al*., 2006). Decrease in grain yield with pre-emergence application of Basagran-48SL, Starane-M60EC and Sencor-70WDG was due to crop injury which caused severe reduction in plant population. These results are supported by the findings of Singh and Wright (1999) and Ghosheh and Shatnawi (2005).

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copiesBIO-EFFICACY OF SAND MIX APPLICATION OF PRE-EMERGENCE HERBICIDES ALONE AND IN SEQUENCE WITH IMAZETHAPYR ON WEED CONTROL IN RELAY CROP OF BLACKGRAM

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ABSTRACT

A field experiment was conducted during winter seasons of 2007-08 and 2008-09 to study the bio-efficacy of sand mix application of pre emergence herbicides, pendimethalin 1000 g ha^{-1}, pretilachlor 500 g ha^{-1}, oxyfluorfen 120 g ha^{-1}, imazethapyr 63 g ha^{-1} alone and in sequence with post emergence application of imazethapyr 50 g ha^{-1} on weed control in black gram grown as relay crop. Results indicated that sequential treatments were found to be superior to individual applications. Among the sequential treatments, pre emergence sand mix application of pendimethalin 1000 g ha^{-1} followed by imazethapyr 50 g ha^{-1} at 20 days after sowing (DAS) significantly reduced weed growth and recorded the highest seed yield (1113 kg ha^{-1}), net monetary returns (Rs.2255 ha^{-1}) and B:C ratio (1.33) and was at par with other sequential treatment, oxyfluorfen 120 g ha^{-1} followed by imazethapyr 50 g ha^{-1} and also with hand weeding at 15 and 30 DAS. Uncontrolled weed growth caused 61 percent reduction in seed yield of blackgram.

Key Words: Sand mix application, pre emergence herbicide, relay crop.

INTRODUCTION

Cultivation of black gram as a relay crop in rice fallows is a common practice in coastal districts of Andhra Pradesh, India, wherein sprouted seeds of black gram are broadcasted in standing crop of rice 2 to3 days before its harvest. The black gram sown in this system survives entirely on residual moisture and fertility only. As there is no field preparation, weed growth particularly of *Echinochloa* spp. is severe and effectively competitive with the crop for residual moisture, nutrients and reduces the black gram yield to the extent of 75 percent (Rao and Rao, 2003; Rao, 2008).

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The continuous use of post emergence grassy herbicides (ACCase inhibitors) resulted in shifting of weed flora towards broad leaf weeds (BLW) with the dominance of *Grangea maderaspatana* in relay crop of black gram. The broad spectrum herbicide imazethapyr causes slight injury to relay crop of black gram, thus farmers are reluctant to use this herbicide Therefore, in the present investigation an attempt has been made to find out the efficacy of applying pre emergence herbicides along with post emergence herbicide imazethapyr alone and in sequence, as normal method of pre emergence application is not viable in the existing system.

MATERIALS AND METHODS

A field experiment was conducted during winter seasons of 2007-08 and 2008-09 at Water Management Pilot area at Modukur Village of Guntur District, Andhra Pradesh, India. The soil texture of the experimental field was clay having lower available nitrogen, medium available phosphorus and higher available potassium with soil pH of 7.8. The experiment consisting of ten treatments (Table-1) was laid out in randomized complete block design with three replications.

The sprouted seeds of black gram (cv. LBG 645) were broadcasted uniformly two days before harvest of rice crop. Immediately after removal of paddy sheaves (one week after harvest) the pre emergence herbicides were thoroughly mixed in dry sand at 50 kg ha^{-1} and broadcasted uniformly over the field and then water was sprayed by using a spray volume of 500 L ha^{-1} in order to create normal pre emergence situation.

The post emergence herbicide was sprayed at 20 DAS using a spray volume of 500 L ha^{-1}. The crop survived entirely on the residual moisture and fertility only. Density of different weed groups and total weed dry weight were recorded at various stages with the help of a quadrate and then converted to m^{-2} and the data on weed parameters were subjected to square root transformation (x + 0.5) before statistical analysis (Panse and Sukhatme, 1978).

RESULTS AND DISCUSSION

Effect on weeds

The experimental field was dominated by natural infestation of BLW like *Grangea maderaspatana*, *Gnaphalium polycaulon*, *Nasturtium indicum, Chrozophora rottleri, Cardanthera uliginosa, Xanthium strumarium* and grasses like *Echinochloa colona, Dinebra retroflexa, Leptochloa chinensis.*

All the weed control treatments significantly reduced the density of grasses, BLW and total weed dry weight over unweeded check at all stages of observation (Tables-1 & 2). Among the treatments, pre emergence sand mix application of pendimethalin 1.0 kg followed by sequential application of imazethapyr 50 g ha^{-1} at 20 DAS recorded higher weed control efficiency (WCE) of 70 % at 60DAS and was at par with other sequential treatments and also with hand weeding at 15 and 30 DAS. Similar trend was observed at harvest also. Unweeded check recorded the highest weed growth. In general, sequential treatments were found to be superior to individual application of herbicides.

Effect on crop

Sand mix application of pre emergence herbicide oxyfluorfen 120 g ha^{-1} and pretilachlor 500 g ha^{-1} and post emergence application of imazethapyr 50 g ha^{-1} resulted in slight injury to black gram for about a week and then the crop resumed to normal condition. All the weed control treatments significantly influenced the yield and yield attributes over unweeded check (Table-3).

Among the treatments, pre emergence sand mix application of pendimethalin 1.0 kg ha^{-1} followed by imazethapyr 50 g ha^{-1} at 20 DAS recorded significantly the highest seed yield (1000 kg ha^{-1}) over all other treatments, except with the treatments, oxyfluorfen 120 g ha^{-1} followed by imazethapyr (50 g ha^{-1}) and hand weeding at 15 and 30 DAS (1201 kg ha^{-1}).

The increased yield in these treatments might be due to effective control of weeds in early stage by pre emergence sand mix application and late emerged weeds by post emergence application of imazethapyr, which resulted in reduced weed growth and increased crop growth and yield components. The results are akin to those reported by Rao and Murthy (2004) and Begum and Rao (2006). The uncontrolled weed growth resulted in 61 % yield reduction.

Economics

The highest net monetary return (Rs.22,255 ha^{-1}) and benefit cost ratio of 1.33 was obtained with pre emergence sand mix application of pendimethalin 1000 g ha^{-1} fb. imazethapyr 50 g ha^{-1} at 20 DAS (Table-3). This was closely followed by pre sand mix application of pretilachlor 500 g ha^{-1} followed by imazethapyr 50 g ha^{-1} with net monetary return of Rs.18,270 and benefit cost ratio of 1.13 which may be due to higher WCE and lower cost of treatment.

From the results it can be concluded that pre emergence sand mix application of pendimethalin 1.0 kg ha^{-1} followed by imazethpyr 50 g ha^{-1} at 20 DAS appears to be effective, economically and a good substitute for hand weeding in rice fallow black gram, when grown as relay crop.

Table-1. Effect of different treatments on density of different weed groups in black gram (pooled data).

Treatments	Dose (g ha^{-1})	Time of application (DAS)	Weed density (plants m^{-2}) at 30 DAS				Weed density (plants m^{-2}) at 60 DAS			
			Grasses	BLW	WCE (%)		Grasses	BLW	WCE (%)	
					Grasses	BLW			Grasses	BLW
T1- Unweeded check	-	-	14.5 (216.7)	18.6 (274.7)	-	-	13.7 (196.0)	18.9 (410.7)	-	-
T2- Hand weeding	-	15&30	6.0 (44.0)	12.8 (180.7)	59	32	3.8 (16.0)	4.5 (24.0)	72	76
T3- Pendimethalin	1000	10 (SMA)	4.7 (26.7)	10.5 (123.3)	68	44	7.0 (58.7)	9.0 (121.3)	49	52
T4- Oxyfluorfen	120	10 (SMA)	7.2 (42.0)	9.5 (93.3)	50	49	7.4 (61.3)	9.4 (132.0)	46	50
T5- Pretilachor	500	10 (SMA)	8.1 (66.0)	12.9 (172.7)	44	31	9.6 (98.7)	11.9 (172.0)	30	37
T6- Imazethapyr	68	10 (SMA)	8.7 (79.3)	11.5 (137.3)	40	41	10.3 (110.7)	13.2 (194.7)	25	30
T7- Imazethapyr	50	20 (SMA)	5.4 (37.3)	9.7 (97.3)	63	48	8.0 (68.0)	10.3 (128.0)	42	44
T8- T3 fb. T7	1000 fb. 50	10(SMA) fb.20	3.6 (15.0)	5.4 (34.7)	75	71	4.1 (20.0)	5.2 (35.3)	70	72
T9- T4 fb. T7	120 fb. 50	10(SMA) fb.20	4.5 (70.7)	5.1 (32.0)	69	73	4.8 (30.7)	5.6 (39.3)	65	70
T10- T5 fb. T7	500 fb. 50	10(SMA) fb.20	4.5 (49.3)	6.1 (42.0)	69	67	6.4 (38.0)	6.4 (47.7)	53	66
CD at 5%			**2.1**	**2.3**			**1.9**	**2.6**		

Note SMA: Sand mix application, DAS: Days after sowing. Data transformed to $\sqrt{x+0.5}$ transformation. Figures in parentheses are original values

Table-2. Effect of different treatments on density of different weed groups and total weed dry weight in blackgram (pooled data).

Treatments	Dose (g ha^{-1})	Time of application (DAS)	Weed density (plants m^{-2}) at Harvest				Total weed dry weight (g m^{-2}) at		WCE (%) at	
			Grasses	BLW	WCE (%)		60 DAS	Harvest	60 DAS	Harvest
					Grasses	BLW				
T1- Unweeded check	-	-	12.5 (161.3)	19.4 (383.3)	-	-	7.6 (62.9)	11.6 (140.4)	-	-
T2- Hand weeding	-	15&30	5.3 (29.3)	5.4 (34.0)	58	72	2.0 (4.5)	4.5 (21.6)	74	61
T3- Pendimethalin	1000	10 (SMA)	7.8 (63.3)	10.0 (114.7)	38	49	4.3 (18.8)	6.4 (46.0)	43	45
T4- Oxyfluorfen	120	10 (SMA)	7.9 (64.0)	9.8 (109.3)	37	50	4.6 (20.7)	6.6 (44.8)	40	43
T5- Pretilachor	500	10 (SMA)	8.6 (80.3)	12.9 (185.3)	31	34	5.3 (27.9)	7.5 (57.7)	30	35
T6- Imazethapyr	68	10 (SMA)	8.2 (75.3)	12.7 (168.7)	34	35	4.7 (22.6)	7.6 (60.2)	38	34
T7- Imazethapyr	50	20 (SMA)	8.3 (71.3)	11.0 (126.0)	34	43	3.8 (14.0)	6.5 (43.3)	50	44
T8- T3 fb. T7	1000 fb.50	10 (SMA) fb.20	4.6 (26.8)	7.2 (57.3)	63	63	2.8 (7.4)	4.8 (23.1)	63	59
T9- T4 fb. T7	120 fb.50	10(SMA) fb.20	4.9 (34.7)	7.6 (64.0)	61	61	3.1 (8.8)	5.1 (25.8)	59	56
T10- T5 fb. T7	500 fb.50	10(SMA) fb.20	5.6 (39.0)	7.9 (69.3)	55	59	3.4 (10.9)	5.3 (27.78)	55	54
CD at 5%			1.8	1.8			0.8	1.2		

Note; SMA: Sand mix application DAS: Days after sowing. Data transformed to $\sqrt{x+0.5}$ transformation. Figures in parentheses are original values.

Table-3. Effect of different treatments on yield and yield components of black gram (pooled data).

Treatments	Dose (g ha^{-1})	Time of application (DAS)	Crop injury score (%) days after application		Crop dry weight (g m-^{2}) at		No. of Pods Plant^{-1}	No. of seeds pod^{-1}	100 seed weight (g)	Seed yield (kg ha^{-1})	Net return (Rs ha^{-1})	BCR (Rs)
			7	14	60 DAS	Harvest						
T1- Unweeded check	-	-			90.0	201.1	11.3	5.5	4.55	464	1240	0.08
T2- Hand weeding	-	15&30			117.1	435.6	27.0	6.8	5.55	1201	24,035	1.33
T3- Pendimethalin	1000	10 (SMA)			126.7	319.6	17.9	6.2	5.17	840	13,300	0.83
T4- Oxyfluorfen	120	10 (SMA)	7.5	0	119.1	330.1	17.8	6.2	5.14	880	14,884	0.94
T5- Pretilachor	500	10 (SMA)	10	0	114.0	273.2	16.7	6.0	4.90	688	8,580	0.55
T6- Imazethapyr	68	10 (SMA)			117.1	274.8	16.7	6.1	5.19	669	7,307	0.45
T7- Imazethapyr	50	20 (SMA)			120.9	302.2	17.6	6.2	5.10	728	9,580	0.60
T8- T3 fb. T7	1000 fb. 50	10 (SMA) fb. 20			165.8	406.8	25.4	6.6	5.36	1113	22,255	1.33
T9- T4 fb. T7	120 fb. 50	10 (SMA) fb. 20	5	0	155.3	367.3	22.9	6.4	5.12	1000	18,484	1.12
T10- T5 fb. T7	500 fb. 50	10 (SMA) fb. 20	10	0	149.3	368.4	23.0	6.6	4.98	982	18,484	1.12
CD at 5%					18.6	46.4	2.9	0.36	0.41	115.0		

Fb. = Followed by

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DIVERSITY OF EPIPHYTIC AND ENDOPHYTIC MICROORGANISMS IN SOME DOMINANT WEEDS

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ABSTRACT

Diversity in epi and endophytic microorganisms from the local weeds is thoroughly studied in this paper. For this purpose, 46 fungal and 19 bacterial strains were isolated from the surfaces and the inner tissues of four dominant agricultural weeds. Leaf wash and homogenized leaf mixture solution were used for the isolations from healthy leaves of four weeds viz. Chenopodium album, Euphorbia helioscopia, Parthenium hysterophorus and Convolvulus arvensis. Our study indicated that complex interactions existed between the host and their epi and endophytic microflora. Each weed has specific bacterial community with the reference of epi and endo phyllosphere. The number and species of bacterial strains varied not only with their host weed plants but also in epi and endo phyllospere. Sørensen's QS of all tested weeds for the endophytic and epiphytic bacterial assemblages was 0.00 that indicated no species overlap / similarity between the communities. Five fungal genera were common as epi and endophytes in all weeds samples: Aspergillus (56% of all isolates), Drechslera (10%), Alternaria (10%) Penicillium (6%) and Cladosporium (4%). Frequency of all five common genera differed significantly among weeds. It was also noted that endophytic fungal communities were not noticeably less speciose than epiphyte communities. Sørensen's QS of E. helipscopia (0.23), C. album (0.37) and C. arvensis (0.46) for the endophytic and epiphytic fungal assemblages was intermediate in the range (0.12–0.79) of previous studies. In case of P. hysterophorus, the value for Sørensen's QS was 0.00 indicating no species similarity. The other identified genera were rare, such as Absidia, Cuvularia, Phoma and Trichoderma.

Key words: Epi and endophytic, fungi, microorganism, phyllosphere, *Aspergillus.*

INTRODUCTION

The phyllosphere of a leaf includes its surface (phylloplane) and the internal tissues colonized by a variety of epiphytic as well as

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endophytic microorganisms respectively, thereby occupying two distinct habitats on the leaf (Andrews, 1996; Carroll *et al.,* 1977; Petrini, 1991). These flora coexist within millimeters of each other, but are usually studied separately and may have important implications for plant health and plant protection (Andrews and Harris, 2000; Arnold *et al.,* 2003; Strobel and Long, 1995; Sturz *et al.*, 2000), microbial biodiversity (Arnold *et al*., 2001; Carroll 1995; Gamboa *et al*., 2002; Hawksworth and Rossman 1997; Petrini *et al.,* 1995) and drug discovery (Strobel and Long, 1995).

Relationships between epiphytes and endophytes have important implications for micro-organisms biodiversity and plant health. It is unclear to what extent plants control which endophytes are able to enter the leaf, and to what extent epiphytes may affect this process (Lebro *et al.,* 2001). The interest shown in the last few years in the study of phyllosphere microbes is due to their interactions with plants, herbivores and pathogens on living leaves which may be involved in the plant immunity system, reabsorption of organic and mineral matters from leachates, redistribution of nutrients prior to leaf fall and participation in the primary degradation of plant tissues (Carroll *et al*., 1977; Cabral, 1985; Lindow and Brandl, 2003; Osono, 2006). Comparison of endophytic and epiphytic flora may help to determine the basis for selectivity. Therefore, the purpose of this study was (1) to search for association of epi and endophytic microorganisms in the weeds habitats and (2) epiphytes and endophytes may potentially be useful for biocontrol study of these weeds. Therefore, with these perspectives it was thought desirable to undertake a preliminary study on the diversity of phyllosphere fungal community of four dominant weed species in agricultural crops of Pakistan.

METERIALS AND METHODS

Sample collection and microbial isolation

The fresh and healthy leaves of four weeds viz. *Chenopodium album, Euphorbia helioscopia, Parthenium hysterophorus* and *Convolvulus arvensis*, were collected from the premises of University of the Punjab, Lahore, Pakistan. Five Leaves of each of 4 weeds were collected from different locations in the university and put separately into sterile bags then taken back to laboratory in less than 2 hours for isolation of epiphytic and endophytic phyllosphere microorganisms.

To analyze epiphytic microflora, leaf washings were used for the isolation. A leaf sample (was shaken for about 1h in 100 ml of sterile distilled water. An aliquot of 1ml from leaf wash was plated on 2% Malt Extract Agar (MEA) medium (g/L): Malt, 20.0; Agar, 20.0 for fungal isolation and LB Medium (g/L): Peptone, 5.0; Beef Extract, 3.0; Agar, 15.0, was used for bacterial isolation.

For endophytic microflora, leaves of each weed were washed through in running water followed by surface-sterilization in 70% ethyl alcohol (1 min), 2.6% $NaClO_2$ (3 min), and 70% EtOH (1 min). Sterile leaves were ground in blender with 100ml of sterile distilled water to form a homogenized leaf solution mixture. Leaf mixture (1 ml) was then plated on 2% Malt Extract and on LB Medium for fungal bacterial isolation, respectively.

The Petri dishes were incubated for 3-4 days at 25-28^{o}C for the fungal colony count. Bacterial colonies were counted after 24 hours at 37^{o}C and purified for further identification.

Sørensen's quotient of similarity (*QS*) was calculated to examine the similarity of fungal /bacterial assemblages in leaf interiors and on leaf surfaces:

$$QS = 2a \;/\; (2a+b+c)$$

Where *a* is the number of common species and *b* and *c* are the numbers of species specific to the interior and the surface, respectively (Osono and Mori*,* 2004). The Sørensen index is a very simple measure of beta diversity, ranging from a value of 0 where there is no species overlap between the communities, to a value of 1 when exactly the same species are found in both communities. The relative abundance/ frequency (%) of each fungal/bacterial species isolated by dilution plating was also calculated as: (Number of colonies of a fungal species/ Total number of fungal colonies) × 100.

Morphological taxonomy of fungal and bacterial isolates

Isolated fungal species were plated onto MEA Petri dishes and incubated for 5 days at 25- 28°C in darkness to observe the colonies' morphology and measure their diameters. A small portion of fungal colony was used to identify the fungal isolates under the microscope on their morphological characters using various mycological keys (Ellis, 1971, Domsch *et al*., 1980, Pitt, 2000). Bacterial strains were identified including pigment, colony form, elevation, margin, texture and opacity (Smibert and Krieg, 1981). In addition, bacterial strains were tested with respect to Gram reaction and biochemical characteristics (Holt *et al.,* 1994).

RESULTS AND DISCUSSION

Diversity of bacteria

Nineteen bacterial species were isolated and identified from phyllosphere of weeds (Table-1). The number and species of bacterial strains varied not only with their host plants but also in epi and endo phyllospere. From *Parthenium hysterophorus* leaves, three ecto (*Peptococcus* sp., *Kurthia gibsonii, Acidovorax facilis*) and two endo

(*Ensifer adhaerens, Acinetobacter calcoaceticus*) bacterial species were isolated. *Chenopodium album* supported *Bacillus farraginis* and *Enterobacter agglomerans* as endophytic bacteria whereas *Curtobacterium albidum* and *Acinetobacter lwoffii* were isolated from epiphyllosphere. *Klebsiella* sp. and *Burkholderia pseudomallei* were only purified from epi phyllospher and *Yersinia ruckeri* and *Corynebacterium minutissimum* from endophyllosphere of *Convolvulus arvensis* (Table-1). In the phyllosphere of *Euphorbia helioscopia,* three bacterial species (*Bacillus farraginis, Kurthia* sp., *Enterobacter agglomerans*) were recorded as epiphytic and three (*Azospirillum lipoferum, Acinetobacter lwoffii*, *Cedecea davisae*) as edophytic bacteria.

Diversity of fungi

We found that all leaves of weed species contained fungal endophytes and epiphytes. A total of 46 fungi were isolated as endo and epiphytic fungal isolates from phyllosphere of four weeds (Table-2). Five fungal genera were common as epi and endophytes from more than one site: *Aspergillus* (56% of all isolates), *Drechslera* (10%), *Alternaria* (10%), *Penicillium* (6%) and *Cladosporium* (4%). The other identified genera were rare, such as *Absidia*, *Cuvularia*, *Phoma* and *Trichoderma.* Frequency of all five common genera differed significantly among weeds (Table-2). For example, *Aspergillus* was a common epiphyte as well endophyte in all test weeds where as *Absidia, Cuvularia*, *Phoma* were only isolated as endophyte in *Chenopodium album* and *Parthenium hysterophoru*s. Sørensen's *QS* of *Euphorbia helioscopia* (0.23)*, Chenopodium album* (0.37) and *Convolvulus arvensis* (0.46) for the endophytic and epiphytic fungal assemblages was intermediate in the range (0.12–0.79) of previous studies. In case of *P. hysterophorus,* the value for Sørensen's *QS* was 0.00 indicating no species similarity.

The aerial parts of living plants including leaves, stems, buds, flowers and fruits provide a habitat for microorganisms termed the phyllosphere. Current study indicated that complex interactions existed among the tested species in epi and endophytic microflora with relation to their hosts. The microscopic examination of the endo and epiphytic phyllosphere gave valuable information on the development, distribution and frequency of the natural mycoflora of weeds leaves surfaces (Table-1 & 2). Total number of microorganisms isolates of phyllosphere differed significantly among tested weeds. Nineteen bacterial species were isolated and identified from phyllospher of weeds (Table-1). The number and species of bacterial strains varied not only with their host plants, but also in epi and endo phyllospere.

Bacteria are considered to be the dominant microbial inhabitants of the phyllosphere, although archaea, filamentous fungi, and yeasts may also be important. These microbes can be found both as epiphytes on the plant surface and as endophytes within plant tissues (Arnold, *et al.* 2000; Inacio *et al.* 2002; Lindow and Brandl 2003; Stapleton and Simmons 2006). Adams and Kloepper (2002) showed that endophytic bacterial population sizes and structure differed between cotton cultivars, and in cultivars were found to contain endophytic bacteria with one showing a higher colonization level than the others (Elvira-Recuendo and van-Vuurde, 2000). It was concluded from the result that each weed has specific bacterial community with the reference of epi and endo phyllospere. Sørensen's *QS* of all tested weeds for the endophytic and epiphytic bacterial assemblages was 0.00 that indicated no species overlap/ similarity between the communities. Each of the five most common fungal genera [*Aspergillus* (56% of all isolates), *Drechslera* (10%), *Alternaria* (10%), *Penicillium* (6%) and *Cladosporium* (4%)] was more common either outside or inside the leaves, and differences were highly significant in all cases (Table-2). Results also supported that these fungi were predominant in epiphyte and endophytic communities. The presence of these common taxa suggests that either endophytic fungi produce some fast growing spores/or hyphae which come out to the outer leaf surface or epiphytic fungi may have penetrated the host tissues and have colonized internal tissues as endophytes.

Furthermore, unexpectedly, endophytic communities were not noticeably lesser than epiphyte communities. However, in case of *Chenopodium album* and *Convolvulus arvensis*, the number of fungal species found was similar between epiphytes and endophytes (Table-2). However, the possibility of a chance occurrence of certain fungal species on a particular weed cannot be overruled. Sørensen's *QS* of *Euphorbia helioscopia* (0.23), *Chenopodium album* (0.37) and *Convolvulus arvensis* (0.46) for the endophytic and epiphytic fungal assemblages was intermediate in the range (0.12–0.79) of previous studies on forest tree leaves (Osono and Mori, 2004), while it varies significantly than that of *Pinus resinosa* (0.120) (Legault *et al.* 1989) and *Nothofagus truncata* (0.788) [Ruscoe, 1971].

Abundance of species in both communities approximated a log-normal distribution, as is typical for fungal communities (Dix and Webster, 1995; Gamboa and Bayman, 2001). Frequently recovered fungal species like *Alternaria, Aspergillus, Cladosporium, Penicillium,* and *Trichoderma* spp. from four weeds leaf samples, grow quickly and produce large number of conidia which are easily dispersed and exhibit wide ecological spectrum (Christensen, 1981). Some of these species are able to utilize cellulosic components and gallic acid (Kjøller and Struwe, 1987; Rai *et al.,* 1988) and also found to play important role in primary degradation of plant tissues. *Alternaria alternata, Cladosporium cladosporioides* along with *Fusarium oxysporum* and

Table-1. List of bacterial species isolated from different weeds.

Name of weeds	Epiphytic species	Colony Frequency	Colony %	Endophytic species	Colony Frequency	Colony %	QS
Convolvulus arvensis	*Klebsiella* sp.	4	26	*Yersinia ruckeri*	5	33	0.00
	Burkholderia pseudomallei	3	20	*Corynebacterium minutissimum*	3	20	
Euphorbia helioscopia	*Bacillus farraginis*	8	25	*Azospirillum lipoferum*	4	12	0.00
	Kurthia sp.	7	22	*Acinetobacter lwoffii*	2	6.4	
	Enterobacter agglomerans	7	22	*Cedecea davisae*	3	9.6	
Parthenium hysterophorus	*Peptococcus* sp.	3	18	*Ensifer adhaerens*	3	18	0.00
	Kurthia gibsonii	4	25	*Acinetobacter calcoaceticus*	4	25	
	Acidovorax facilis	2	12				
Chenopodium album	*Bacillus farraginis*	4	25	*Curtobacterium albidum*	6	37	0.00
	Enterobacter agglomerans	3	28	*Acinetobacter lwoffii*	3	18	

Table-2. List of fungal species isolated from different weeds.

Weeds	Epiphtic Fungi	Freq.	Freq. %	Endophtic Fungi	Freq.	Freq. %	QS
Parthenium hysterophorus	*Aspergillus flavus*	6	37	*Aspergillus niger*	2	11	0.00
	Alternaria alternata	3	18	*Aspergillus parasiticus*	6	35	
	Drechslera biseptata	5	31	*Aspergillus reperi*	5	29	
	Mucor sp.	2	37	*Absidia ramosa*	1	5	
				Curvularia clavata	1	5	
				Phoma sp.	2	11	
Convolvulus arvensis	*Aspergillus flavus*	5	23	*Aspergillus flavus*	5	23	0.46
	Aspergillus niger	2	9	*Aspergillus niger*	2	9	
	Aspergillus reperi	4	19	*Aspergillus reperi*	1	7	
	Aspergillus fumigatus	2	9	*Aspergillus fumigatus*	2	15	
	Aspergillus terreus	1	4	*Aspergillus terreus*	2	15	
	Trichoderma sp.	1	4	*Drechslera biseptata*	1	7	
	Drechslera australiensis	1	4	*Drechslera australiensis*	2	15	
Euphorbia helioscopia	*Aspergillus aculeatus*	2	15	*Aspergillus japonicus*	1	9	0.23
	Aspergillus niger	5	38	*Aspergillus aculeatus*	3	27	
	Aspergillus terreus	3	23	*Cladosporium cladosporioides*	1	9	
	Alternaria alternata	1	7	*Aspergillus sydowi*	2	18	
	Drechslera biseptata	2	15	*Alternaria alternata*	1	9	
				Alternaria dianthi	1	9	
				Penicillium spp.	2	18	
Chenopodium album	*Aspergillus phoenicis*	3	18	*Aspergillus phoenicis*	1	10	0.37
	Aspergillus flavus	2	12	*Aspergillus flavus*	4	40	
	Alternaria alternata	9	56	*Alternaria alternata*	1	10	
	Cladosporium sp.	1	6	*Cuvularia clavata*	2	20	
	Penicillium oxalicum	1	6	*Aspergillus reperi*	2	20	

Freq = Frequency

Pestalotiopsis sp. are also recorded as dominant surface and interior colonizers of different tree species leaves (Kayini and Pandey, 2010). In general, these species are extensively reported as common primary saprobes and ubiquitous hyphomycetes from attached leaf surfaces of wide variety of plants throughout the world (Breeze and Dix, 1981; Mishra and Dickinson, 1981; Pandey, 1990; Andrews, 1996; Osono, 2006).

The phyllosphere represents a niche with great agricultural and environmental significance. There is growing evidence for important interactions of phyllosphere microbial inhabitants which may affect the fitness of natural plant populations and the quality and productivity of agricultural crops. Phyllosphere bacteria can promote plant growth and both suppress and stimulate the colonization and infection of tissues by plant pathogens (Lindow and Brandl, 2003; Rasche *et al.*, 2006). Similarly, fungal endophytes of leaves may deter herbivores, protect against pathogens and increase drought tolerance (Arnold *et al.,* 2003; Schweitzer *et al.,* 2006). Epiphytic and endophytic microflora presumably interacts and connects in cross-talk in ways that affect the host plant. Interactions within each community are poorly understood, and interactions between endophytes and epiphytes are completely unexplored. Fungi and bacteria make a complicate linkage of endophytes and epiphytes, and their interactions are also poorly understood. Understanding these phyllospheric communities and their interactions in weeds can improve crops health. Study of phyllosphere microbial communities in weeds, represents one of the most promising and poorly understood areas of agriculture. Understanding the microbial communities and interactions in weeds phyllosphere, can be helpful to improve crop health in sustainable agriculture.

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HERBICIDES AND THEIR DOSES EFFECTS ON WILD ONION (*Asphodelus tenuifolius* Cav.) IN CHICKPEA

Muhammad Ishfaq Khan[1], Gul Hassan and Imtiaz Khan

ABSTRACT

This study aims to determine dose requirements of herbicides for controlling Asphodlus tenuifolius on farmer's field in chickpea during 2005-06 and 2006-07. Five herbicides pendimethalin, s-metolachlor, fenoxaprop-p-ethyl, MCPA and isoproturon with four doses were studied in the trials. MCPA produced phytotoxic effect on both weed and crop and also completely inhibited crop and weed growth. Isoproturon was less effective on A. tenuifolius with less phytotoxicity on crop. Best seed yield (1164 and 1150 kg ha^{-1}) was recorded in pre-emergence herbicides at high dose as compared to fenoxaprop-p-ethyl (1088 kg ha^{-1}). Next year (2006-2007) again the same herbicides were tested while MCPA was replaced by clodinafop propargyl (post-emergence) due to its phytotoxicity on crop. Almost similar results were recorded with the only difference of herbicides clodinafop propargyl. The highest seed yield of 1109 kg ha^{-1} was recorded each for pendimethalin and s-metolachlor while it was statistically similar with the yield of fenoxaprop-p-ethyl (1107 kg ha^{-1}). Lower 0.5x and 1.0X doses of post emergence herbicides produced good results as compared to 1.5X. Pre emergence herbicides were found to be effective only at 1.5X dose.

Key words: Chickpea, *A. tenuifolius*, herbicides, doses, interaction.

INTRODUCTION

Chickpea (*Cicer arietinum* L.) is a major food legume and an important source of protein in many countries in Asia and Africa. This species is the second most consumed and the third most cultivated grain legume in Asia (Dodak *et al.,* 1993). It is an ancient crop and is grown in tropical, subtropical and temperate regions. India, Pakistan and Mexico were recorded as major producers of chickpea (Badshah *et al.,* 2003). In India and Pakistan, chickpeas are consumed locally; furthermore about 56% of the crop is retained by growers. Chickpea is valued for its nutritive seeds with high-protein content 17–22% and 25.3–28.9%, before and after dehulling, respectively (Hulse, 1991; Badshah *et al.,* 2003).

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Weeds are a serious constraint of production and easy harvesting of chickpea. Chickpea, however, is a poor competitor to weeds because of its slow growth rate and limited leaf area development at early stages of crop growth and establishment. Yield losses due to weed competition vary considerably depending on the level of weed infestation and weed species prevailing. Nevertheless, almost all values reflect the seriousness of the weed problem. Yield losses were observed to vary from 40-94% in the Indian subcontinent (ICARDA, 1985; Bhan and Kukula, 1987), 40-75% in West Asia (ICARDA, 1982, 1986), 13-98% in North Africa (El-Brahli, 1988; Knott and Halila, 1988), and 35% in Italy (Calcagno *et al.,* 1987). Effective weed control may increase yield in chickpea by 17-105%.

There are more than 75 weed species that were reported to infest chickpea fields in the Mediterranean region (Calcagno *et al.,* 1987; El-Brahli, 1988). These species are mostly dicotyledonous and belong to 26 different families. Post emergence application of herbicides can be, indeed, substantially reduced if the "minimum dose requirement for a satisfactory efficacy" (MDRE) is known with respect to the most common "herbicide-weed species" combinations (Davies *et al*., 1993, Kudsk, 1989, Onofri *et al*., 1997; Pannacci and Covarelli, 2003). Effective pre-planting and soil incorporated herbicides include fluchloralin, oxyfluorfen, trifluralin and triallate. Whereas effective pre-emergent herbicides are alachlor, chlorobromuron, cyanazine, dinoseb amine, methabenzthiazuron, metribuzin, pronamide, prometryne and terbutryne. Post-emergent herbicides include dinosebacetate, fluazifop-butyl and fenoxprop-ethyl. Post emergent applications need great care with respect to stage of growth and air temperature to avoid phytotoxicity (Bhan and Kukula, 1987).

Chemical weed control with pre-emergence terbutryne at 2.0 kg a.i. ha^{-1} and pronamide at 0.5 kg a.i. ha^{-1} increased yield by 26% and 6% in winter and spring sowing, respectively, compared to control. *Cuscuta campestris* was selectively controlled by pre emergence application of pronamide with chlorthal dimethyl (Graf *et al.,* 1982). Some crops are likely to be more amenable than others to the use of reduced herbicide doses. Kirkland *et al.* (2000) reported that good crop yields and the highest net returns could be attained with a 50% herbicide dose in barley but that a 100% herbicide dose was required to attain the highest yields and net returns in lentil (*Lens culinaris* L.). Keeping in view the economic importance of wild onion infestation in chickpea crop the present studies were conducted to:

1. Figure out the most economical herbicide for the control of *A. tenuifolius.*
2. Test the efficacy of herbicides at varying doses and its effects on crop.
3. Identify the minimum dose requirement of tested herbicides.

MATERIALS AND METHODS

Experiments were conducted in chickpea on farmer's field in district Lakki Marwat, Khyber Pukhtunkhwa Province, Pakistan during rabi season 2005-06 and 2006-07. The experiments were laid out in Randomized Complete Block (RCB) design with split plot arrangements with three replications. The herbicides were assigned to main plots, while herbicides doses were kept in the sub plots. KC-98 chickpea cultivar was seeded during the second week of October, in each year of study. Each sub plot measured 5 × 2 m^2. Two pre-emergence and three post-emergence herbicides each with four doses were used. The herbicidal treatments were the pre emergence application of pendimethalin and s-metolchlor at 0, 0.41 (0.5x), 0.82 (1X) and 1.20 (1.5X). While the post emergence herbicides were isoproturon at 0, 2.0 (0.5x), 4.0 (1X) and 6.0 (1.5X), fenoxaprop-p-ethyl at 0, 0.47 (0.5x), 0.94 (1X) and 1.30 (1.5X), MCPA at 0, 0.28 (½x), 0.56 (1X) and 0.90 (1.5X), and clodinafop propargyl 0, 0.48 (0.5x), 0.98 (1X) and 1.50 (1.5X) and kg a.i. ha^{-1}. MCPA was replaced by clodinafop propargyl in the second season due to its phytotoxic effects on the crop. Herbicides were sprayed using knapsack sprayer. All the weeds in the field except *A. tenuifolius* were manually uprooted.

Data collected from both field experiments were recorded on the following parameters as follow:

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

The plants of *A. tenuifolius* were collected with the help of quadrate of 25 × 25 cm^2 from each treatment and weighed in kg. Three random quadrates were taken in each treatment. The data was subsequently converted to kg ha^{-1}.

100 seed weight (g)

A random sun dried and clean seeds sample of 100 grains from each treatment was taken and weighted (g) using electronic balance.

Seed yield (kg ha^{-1})

Two central rows were harvested in each plot and then the grain yield (kg ha^{-1}) was obtained by the following formula:

$$\text{Grain yield (kg ha}^{-1}\text{)} = \frac{\text{Grain yield (kg)} \times 10000}{\text{Area harvested (m}^2\text{)}}$$

Statistical analysis

The data recorded for each trait was subjected to the ANOVA technique using MSTATC computer software and the means were separated by using Fisher's protected LSD test (Steel and Torrie, 1980).

RESULTS AND DISCUSSION
Season 1 (2005-06)
Fresh weed biomass (kg ha^{-1})

Analysis of variance of the data showed that herbicides, herbicides doses and their interaction had significant effect on the fresh weight of *A. tenuifolius* (Fig. 1). The data exhibited that minimum fresh weight (4.98 kg ha^{-1}) was recorded for MCPA followed by pendimethalin (8.27 kg ha^{-1}), s-metolachlor (8.42 kg ha^{-1}) and fenoxaprop-p-ethyl (8.57 kg ha^{-1})respectively. While maximum fresh weight (11.0 kg ha^{-1}) was recorded in untreated control. Among the doses of herbicides used, the lowest fresh biomass (6.41 kg ha^{-1}) was observed at 1.5X, while maximum fresh weight (11.61 kg ha^{-1}) was recorded in untreated control. In the interaction of herbicides and doses maximum fresh weight (11.61 kg ha^{-1}) was observed in untreated control, followed by isoproturon at all the tested doses while minimum fresh weight (2.5 kg ha^{-1}) in the interaction was observed in MCPA at high dose which was statistically at par with rest of the doses of the same herbicide. Pendimethalin, s-metolachlor and fenoxaprop-p-ethyl produced statistically similar results at 1.5X dose. Herbicides doses have great influence on fresh and dry biomass of wild onion and consequently use in the management of wild onion (Khan *et al.* 2009).

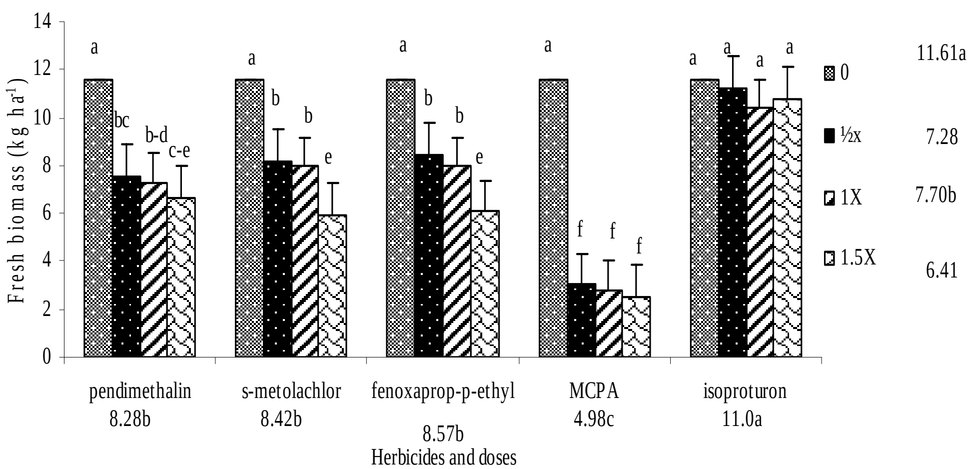

Figure 1. Effect of different herbicides on fresh biomass of *A. tenuifolius* during 2005-06.

Means followed by different letters indicate significant differeces among different doses in the same herbicides (Fisher's protected LSD test, p>0.05). The mean value of 100 seed weight was significantly affected by the herbicides, doses and their interaction (Fig. 2). Among the herbicides maximum seed weight of 26.96 and 26.82g was recorded in the pre-emergence treatment of pendimethalin and s-metolachlor respectively. The minimum 100 seed weight was observed in isoproturon (23.76 g) and fenoxaprop-p-ethyl (24.88 g). Untreated control produced highest seed weight (24.10 g) while 1.5X dose produced lowest one (19.88 g). In the interaction of herbicides and doses highest 100 seed weight was observed in pendimethalin (31.90 g) and s-metolachlor (31.43 g) respectively at 1X dose. While the minimum 100 seed weight was observed in plots treated with isoproturon however, it was statistically at par with the rest of the doses of the same herbicide. MCPA produce 0.00 seed weight due to its phytotoxic effects on the crop.

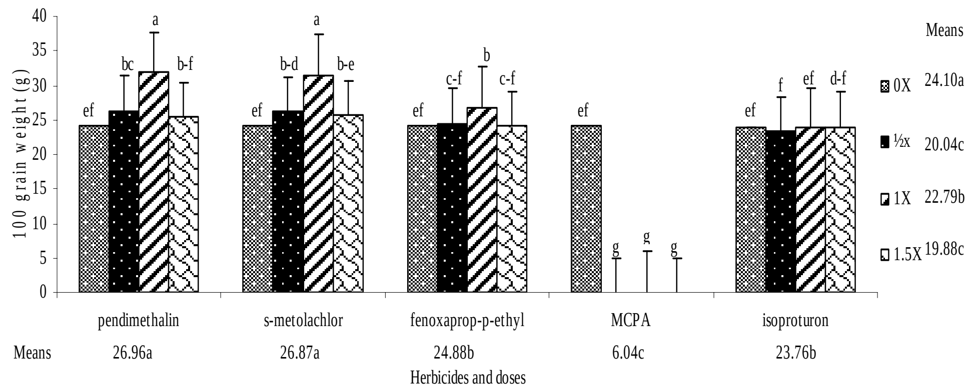

Figure 2. Effect of different herbicides on 100 seeds weight (g) of chickpea during 2005-06.

Means followed by different letters indicate significant differences among different doses in the same herbicides (Fisher's protected LSD test, $p>0.05$).

Seed yield (kg ha^{-1})

Same as fresh biomass and weight of 100 seeds, seed yield was significantly affected by herbicides, their doses and interaction (Fig. 3). The results indicated that the main effects of herbicides produced 1164 kg ha^{-1} seed yield in pendimethalin treated plots however it was statistically at par with s-metolachlor (1150 kg ha^{-1}) followed by fenoxaprop-p-ethyl (1088 kg ha^{-1}) while minimum yield (991.6 kg ha^{-1}) was recorded in isoproturon and MCPA treated plots. Among the tested doses maximum seed yield of 984.0 kg ha^{-1} was recorded in untreated plots followed by the 1X dose (930.5 kg ha^{-1}) while minimum seed yield (910.0 kg ha^{-1}) was observed at the highest dose (1.5X). In the interaction of herbicides and doses highest yield 1270.0 and 1233.0 kg ha^{-1} were observed in pendimethalin at doses

1.5 and 1X respectively. While the preemergence herbicide s-metolachlor gave almost statistically similar results at all tested doses.

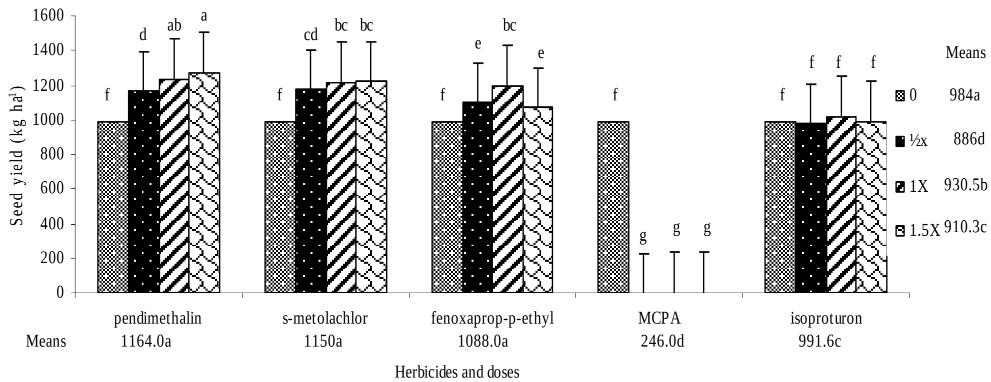

Figure 3. Effect of different herbicides on seed yield kg ha^{-1} of chickpea during 2005-06.

Means followed by different letters indicate significant differences between different doses in the same herbicides (Fisher's protected LSD test, $p>0.05$).

Season 2 (2006-07)

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

Herbicides, herbicide doses and their interactions had different patterns on the fresh biomass of *A. tenuifolius* (Fig. 4). The main effects of herbicides showed that lowest fresh weight (9.47 kg ha^{-1}) was recorded for pendimethalin followed by the s-metolachlor (9.47 kg ha^{-1}), fenoxaprop-p-ethyl (9.69 kg ha^{-1}) and clodinafop propargyl (10.02 kg ha^{-1}) respectively. While highest fresh weight (11.80 kg ha^{-1}) was recorded in plots treated with isoproturon. Among the herbicides doses minimum fresh weight (8.40 kg ha^{-1}) was recorded at the higher dose. Whereas maximum fresh weight (12.50 kg ha^{-1}) was recorded in untreated control. In the interaction of herbicides and doses minimum fresh weight (7.05 kg ha^{-1}) was observed in s-metolachlor at 1.5X dose however it was statistically at par with pendimethalin, fenoxaprop-p-ethyl and clodinafop propargyl at 1.5X dose and pendimethalin at 1X dose as well. While maximum fresh weight (12.63 kg ha^{-1}) was observed in untreated control in all the herbicides followed by isoproturon at all tested doses.

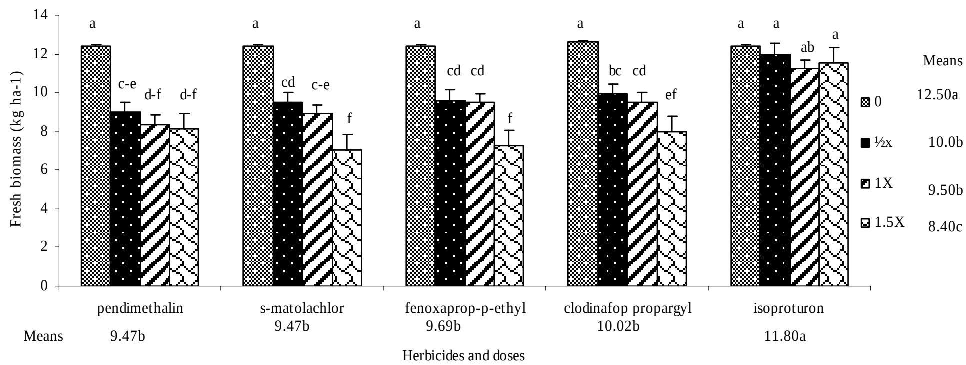

Figure 4. Effect of different herbicides on fresh biomass of *A. tenuifolius* during 2006-07.

Means followed by different letters indicate significant different between different doses in the same herbicides (Fisher's protected LSD test, $p>0.05$).

100 seed weight (g)

The 100 seed weight was significantly affected by the herbicides, herbicide doses and their interaction (Fig. 5). Among the herbicides, maximum seed weight of 26.96 and 26.31 g were recorded in the pre-emergence treatment of s-metolachlor and pendimethalin respectively. The minimum 100 seed weight of 22.18 and 23.14 g were observed in clodinafop propargyl and isoproturon respectively. Among the herbicides doses maximum 100 seed weight (26.41 g) was recorded at 1X dose while the other two herbicide doses produced statistically similar results. In the interaction of herbicides and doses the highest 100 seed weight (31.18 g) was observed in pendimethalin at 1X dose while the minimum seed weight (21.55, 21.70 and 21.85 g) was observed in clodinafop propargyl at all tested herbicidal doses.However, it was statistically at par with the all herbicides doses of isoproturon and all doses of fenoxaprop-p-ethyl except that for 1X dose.

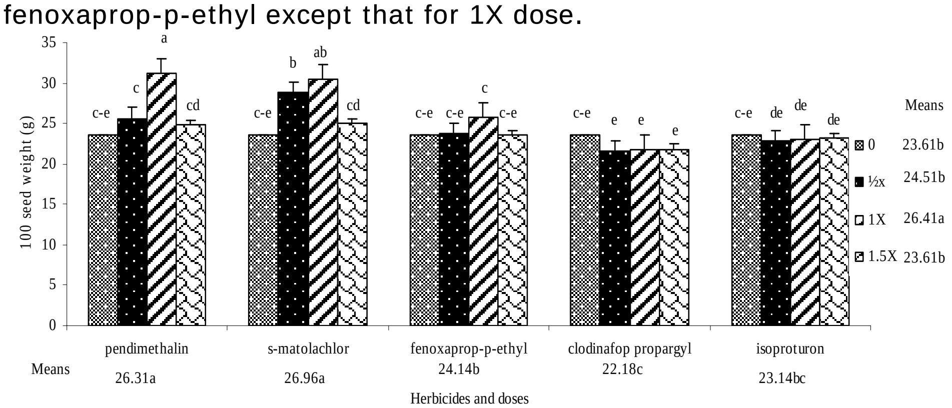

Figure 5. Effect of different herbicides on weight (g) of 100 seeds chickpea during 2006-07.

Seed yield (kg ha^{-1})

Seed yield was also differently affected by herbicides, herbicide doses and their interaction (Fig. 6). The results indicated that among the different herbicides, highest seed yield (1109 kg ha^{-1}) was recorded in pendimethalin and s-metolachlor treated plots, followed by fenoxaprop-p-ethyl (1004.52 kg ha^{-1}) while the minimum yield was recorded in isoproturon (943.4 kg ha^{-1}) and clodinafop propargyl (955.3 kg ha^{-1}). The main effects of herbicides doses indicated that maximum seed yield (1111 kg ha^{-1})was recorded at 1X dose however it was statistically at par with ½x doses (1043 kg ha^{-1}) of the herbicides while minimum seed yield was observed in untreated control (931.3 kg ha^{-1}). In the interaction of herbicides and doses, highest yield was observed in s-metolachlor (1111 kg ha^{-1}) at 1X dose however it was statistically at par with pendimethalin at 1.5X dose.

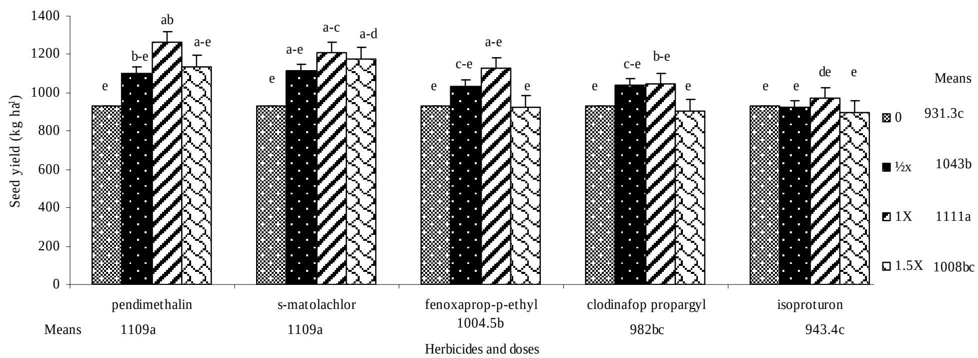

Figure 6. Effect of different herbicides on seed yield kg ha^{-1} of chickpea as during 2006-07.

Means followed by different letters indicate significant different among different doses in the same herbicides (Fisher's protected LSD test, $p>0.05$).

Herbicides, doses, and their interaction affected almost all the tested parameters during both years of the studies. During the first year, MCPA reduced the fresh and dry biomass of weed more than that of the other herbicides although MCPA had exerted phytotoxic effect on the crop as well. So MCPA was replaced by clodinafop propargyl for the next year. Pre-emergence herbicides pendimethalin and s-metolachlor and the post emergence herbicide fenoxaprop-p-ethyl reduced the fresh weed biomass significantly with no injury to the crop. In case of fresh weed biomass pendimethalin, s-metolachlor and fenoxaprop-p-ethyl were the best without injury to the crop. Malik *et al.,* (2003) reported that herbicides decreased the dry weight of weeds significantly. These results are also in a great analogy with the work of Iqbal *et al.,* (1991) and Poonia *et al.,* (1993). The authors reported that herbicides decreased the dry weight of weed significantly. The probable reason for the positive performance of these herbicides is their efficacious control of *A. tenuifolius.* 100 grain weight (g) and grain yield (kg ha^{-1}) were also increased by the application of the pre-emergence herbicides and the post emergence fenoxaprop-p-ethyl as well. The increments of the 100 seed weight and seed yield in this study were probably due to maximum inhibition of wild onion consequently the crop was flourished and efficiently utilized all the available resources.

In both the experiments 1X and 1.5X doses of pre emergence herbicides produced good results when compared to the lower dose. Whereas in post emergence herbicides dose 0.5x and 1X produced good results when compared to 1.5X dose.

Thus it is concluded that the doses of the pre-emergence herbicides pendimethalin and s-metalchlor could not be curtailed for harvesting good yield of chickpea.

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IMPACT OF OXADIAZON AND PYRAZOSULFURON-ETHYL ON RICE AND ASSOCIATED WEEDS IN DRY SEASON RICE CULTIVATION

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ABSTRACT

An experiment was conducted at Bangladesh Rice Research Institute (BRRI), Gazipur Bangladesh in the dry season of 2009 (January-May) to study the efficacy of different postemergence herbicides and select the cost-effective treatment as an option for weed control in dry season rice. Ten different weed control treatments viz. T_1 = Zealus 10 WP at 125 g ha^{-1}, T_2 = Amaraj 10 WP at 150 g ha^{-1}, T_3 = Siniron 10WP at 187 g ha^{-1}, T_4 = Herbikill 10 WP at 150 g ha^{-1}, T_5 = Res Q 25 EC at 1.2 L ha^{-1}, T_6 = Remover 10 WP at 187 g ha^{-1}, T_7 = Safety 10 WP at 200g ha^{-1}, T_8 = Laser 10 WP at 125 g ha^{-1}, T_9 = hand weeding at 15, 30, and 45 DAT and T_{10} = weedy check were compared. The active ingredient of Res Q 25 EC is oxadiazon, whereas the active ingredient of the other products is pyrazosulfuron-ethyl. Eight different weed species were present in the experimental field among which Scirpus maritimus followed by Echinochloa crus-galli were the most dominant in terms of density and importance value. Among the treatments, T_6 (Remover 10 WP) gave the lowest weed density, dry weed biomass and weed index, and the highest weed control efficiency. The yield and yield components of rice (e.g. No. of panicles m^{-2}, No. of grains per panicle, grain and straw yield) were greatly influenced by the treatments. Herbicide treatment T_6 (Remover 10 WP) produced similar yield to hand weeding (T_9), but the weeding cost of T_6 was almost one-sixth of T_9. Maximum marginal return rate with T_6 (Remover 10 WP) suggests that this treatment could be used as alternative tool when labor is a limiting factor in dry season rice cultivation.

Key words: Oxadiazon and pyrazosulfuron-ethyl, herbicide, dry season, weed and rice.

INTRODUCTION

Rice (*Oryza sativa* L.) is the vital food for more than two billion people in Asia and four hundreds millions of people in Africa and Latin

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America (IRRI, 2006). The people in Bangladesh depend on rice as staple food with a tremendous influence on the agrarian economy of Bangladesh. Rice alone constitutes 95% of the food grain production in Bangladesh (Julfiquar *et al*., 1998). Among different groups of dry season rice, *Boro* rice covers about 43.6% of the total rice area and it contributes to 61.3% of the total rice production in Bangladesh (BBS, 2008). *Boro* covers the second largest area of 4.61 million hectares with a production of 17.72 million metric tonnes and the average yield is about 3.84 t ha^{-1} (BBS, 2008). The average yield of rice in Bangladesh is 2.73 t ha^{-1} (BRRI, 2006), which is almost 50% of the average rice grain yield per ha worldwide.

Weed infestation and interference is a serious problem in rice fields that significantly decreases yield. In Bangladesh, weeds reduce rice grain yield by 70-80% in *Aus* rice (early summer), by 30-40% for transplanted (T) *Aman* rice (late summer) and by 22-36% in modern *Boro* rice cultivars (winter rice) (BRRI, 2006; Mamun, 1990). The prevailing climatic and edaphic conditions are highly favorable for numerous weed species that strongly compete with the rice crop. In Bangladesh, the traditional methods of weed control include preparatory land tillage and hand weeding. Usually two or three hand weedings are done in the growing season depending on the nature of weeds, their intensity, and the vigor of rice plants. Mechanical and cultural weed control methods in transplant *Boro* rice are expensive. Especially during periods of labor shortage, late weeding can cause drastic losses in grain yield, while chemical weed control is available, easy to implement, and efficient.

Nowadays the use of herbicides is gaining popularity in rice fields due to their rapid effects and the lower costs compared with the traditional methods (Karim, 2008). The available herbicides for weed control in rice are of overseas origin. The country depends on foreign multinational companies for the supply of herbicides, but usually the companies do not supply the same brand of herbicides for long time. Thus, continuous evaluation of the available herbicides in rice is necessary for the benefit of the farmers of this country. Therefore, the objective of this study was to examine the performance of different postemergence herbicides in comparison with manual weeding for the control of weeds in *Boro* rice.

MATERIALS AND METHODS

An experiment was conducted at Bangladesh Rice Research Institute, Bangladesh during *Boro* season rice (January-May) of 2009. The soil of the experimental field was clay loam with pH of 5.47-5.63. The experiment was laid out in a Randomized Complete Block Design (RCBD) with 3 replications. The plot size was 4m X 4m. Ten different weed control treatments were applied: T_1=Zealus 10 WP at 125 g ha^{-1}, T_2=Amaraj 10 WP at 150 g ha^{-1}, T_3=Siniron 10WP at 187 g ha^{-1}, T_4=Herbikill 10 WP at 150 g ha^{-1}, T_5=Res Q 25 EC at 1.2 L ha^{-1}, T_6=Remover 10 WP at 187 g ha^{-1}, T_7=Safety 10 WP at 200g ha^{-1}, T_8=Laser 10 WP at125 g ha^{-1}, T_9=three hand weedings at 15, 30 and 45 DAT (days after transplanting) and T_{10}=weedy check. The active ingredient of Res Q 25 EC is oxadiazon, whereas the active ingredient of the other products is pyrazosulfuron-ethyl. All the tested commercial herbicides were postemergence and applied at 2 to 3-leaf stage of weeds. Seeds of *Boro* rice cv. 'BRRI dhan29' were sown in the seedbed on December 20, 2008 and transplanted in the main field on January 28, 2009. The planting distance in the field was 20 cm (row-row) × 20 cm (hill-hill). The field was fertilized with urea, triple super phosphate, potassium chloride, gypsum, and zinc sulphate at 220, 100, 60, 60 and 10 kg ha^{-1}, respectively. Except urea, all fertilizers were added during land preparation. Urea was top dressed in three rates at 15, 30 and 45 DAT. Other cultural operations such as gap filling, irrigation, and plant protection were carried out as required. Data regarding weeds were recorded at 50 DAT. Dry weed biomass was determined by drying them in an electric oven at 60^{o}C for 72 hours. Relative weed density (RWD), importance value of weed (IVW), weed control efficiency (WCE), weed index (WI) and marginal rate of return (MRR) were calculated according to the following formulae:

$$\text{RWD} = \frac{\text{Density of individual weed species in the community}}{\text{Total density of all weed species in the community}} \times 100$$

$$\text{IVW} = \frac{\text{Dry weight of a given oven dried weed species}}{\text{Dry weight of all oven dried weed species}} \times 100$$

$$\text{WCE} = \frac{\text{Dry weight of weeds in weedy check plots - Dry weight of weeds in treated plots}}{\text{Dry weight of weeds in weedy check plots}} \times 100$$

$$\text{WI} = \frac{\text{Grain yield in weed free plot- Grain yield in treated plot}}{\text{Grain yield in treated plot}} \times 100$$

$$\text{MRR} = \frac{\text{Marginal gross margin of a treatment}}{\text{Marginal variable cost of that treatment}} \times 100$$

At harvest, various characters of rice plants and yield data were recorded. The data were analyzed following analysis of variance (ANOVA) and mean separations were made by the Multiple Comparison test (Gomez and Gomez, 1984) using the statistical computer program MSTAT-C v.1.2 (Russel, 1986).

RESULTS AND DISCUSSION

Weed infestation

The favorable conditions for dry season rice cultivation are also favorable for the growth of numerous weed species that compete with crop plants. Different weed species from various botanical families infested the experimental plots. The weed species that were present in the experimental field were grasses, broadleaf weeds, and sedges. Most of them belonged to the families Poaceae, Cyperaceae, Pontederiaceae and Oxalidaceae (Table-1). The relative density and importance value of these weed species were also different. The most important weeds in the experimental field were *Scirpus maritimus* followed by *Echinochloa crus-galli*, whereas the least important was *Leptochloa chinensis*. Among the existed weed species, the maximum relative weed density was observed for *Scirpus maritimus*, whereas the minimum relative weed density was observed in the case of *Leptochloa chinensis*. As regards broadleaf weeds, it was observed that these were not dominant in this study.

Table-1. Importance value and relative weed density of the weed flora in the dry season rice.

Weed species	Family	Weed type	Relative weed density (%)	Importance value (%)
Scirpus maritimus L.	Cyperaceae	Sedge	25.21	24.70
Echinochloa crus-galli L.	Poaceae	Grass	21.14	17.60
Monochoria vaginalis L.	Pontederiaceae	Broadleaf	13.65	16.31
Oxalis europea L.	Oxalidaceae	Broadleaf	10.24	14.55
Cynodon dactylon (L.) Pers.	Poaceae	Grass	9.58	10.43
Cyperus difformis L.	Cyperaceae	Sedge	14.09	9.86
Leersia hexandra L.	Poaceae	Grass	3.96	5.25
Leptochloa chinensis L.	Poaceae	Grass	2.13	1.30

Weed control

Weed density was significantly affected by different herbicidal treatments (Fig. 1). Weed density was highest in weedy check plots (T_{10}). Different treatments significantly reduced weed population. Among the treatments, T_8 exhibited the highest reduction (93.6%) of weed density m^{-2}. T_5 and T_9 also showed the same result. Similarly, T_6 and T_9 gave identical results in relation to weed density. Al-Kothayri and Hasan (1990) found that all herbicide treatments significantly reduced the weed populations compared with the weedy check. Similar results were obtained by Hasanuzzaman *et al*. (2008). Significant differences in weed dry weight were observed due to the different weeding treatments (Table-2). Among the treatments, T_6 produced the lowest weed dry matter, which was identical to the other treatment effect. This shows that use of the studied postemergence herbicides reduced the weed biomass effectively. The second lowest weed dry matter was recorded in T_8. The highest weed dry matter (79.6 g m^{-2}) was produced by the weedy check (T_{10}). Weed control efficiency above 80% was found in each treatment. Among the treatments, T_6 showed the best result (92%), which was superior to all the other treatments (Table-2). This may be due to the emergence of fewer weed species. The treatments T_3, and T_4 produced similar results. The lowest weed control efficiency (81%) was shown in T_5. This result was partially supported by the findings of Hasanuzzaman *et al*. (2008). A significant effect on weed index (%) was found due to the different herbicide treatments (Table-2). The lowest weed index (11.6%) was found in T_5, which was identical to the other herbicide treatments. This was due to efficient control of weeds by the herbicide treatments. The highest weed index (88.9%) was found in case of the weedy check.

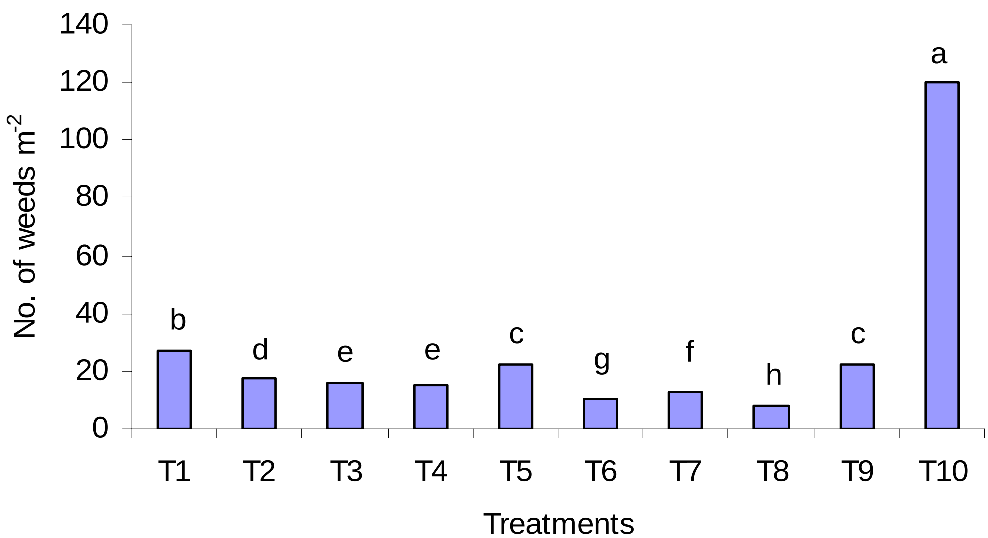

Figure 1. Weed density in rice field as affected by weed control options.

[T_1=Zealus 10 WP at 125 g ha^{-1}, T_2=Amaraj 10 WP at 150 g ha^{-1}, T_3=Siniron 10WP at 187 g ha^{-1}, T_4=Herbikill 10 WP at 150 g ha^{-1}, T_5=Res Q 25 EC at 1.2 L ha^{-1}, T_6=Remover 10 WP at 187 g ha^{-1}, T_7=Safety 10 WP at 200g ha^{-1}, T_8=Laser 10 WP at125 g ha^{-1}, T_9=Three hand weeding at 15, 30 and 45 DAT and T_{10}=Weedy check].

Table-2. Weed dry matter, weed control efficiency, and weed index as affected by the different weed control treatments.

Treatments	Total weed biomass (g m^{-2})	Weed control efficiency (%)	Weed index (%)
T_1=Zealus 10 WP at 125 g ha^{-1}	9.7 b	88.0 e	31.0 b
T_2=Amaraj 10 WP at 150 g ha^{-1}	10.6 b	87.0 f	20.2 b
T_3=Siniron 10WP at 187 g ha^{-1}	8.6 b	89.0 d	27.2 b
T_4=Herbikill 10 WP at 150 g ha^{-1}	8.3 b	90.0 cd	27.1 b
T_5=Res Q 25 EC at 1.2 L ha^{-1}	15.3 b	81.0 h	11.6 b
T_6=Remover 10 WP at 187 g ha^{-1}	6.6 b	92.0 a	14.3 b
T_7=Safety 10 WP at 200g ha^{-1}	11.2 b	86.0 g	23.6 b
T_8=Laser 10 WP at125 g ha^{-1}	7.5 b	90.6 b	23.5 b
T_9=Three hand weeding	8.3 b	89.8 c	-
T_{10}=Weedy check	79.6 a	-	88.9 a
$LSD_{0.05}$	12.9	0.54	20.7

In a column the values having common letter(s) do not differ significantly by LSD test at $P \leq 0.05$.

Yield components

Yield components such as the number of panicles m^{-2} and the number of grains per panicle were significantly influenced by the different weed control treatments used in this experiment (Table-3). Variables such as plant height, panicle length, sterility percent and 1000-grain weight did not differ significantly in the different weeding methods. Among the weeding methods, T_7 had the tallest rice plants (96.5 cm), whereas the non-weeded control plots had the shortest. Apparently, plant height reduction in the non-weeded check was due to competition for longer period of time which prevented rice plants from becoming taller. Higher number of panicles m^{-2} (234) were found in T_5, although statistically similar to T_2, T_4, T_6, T_7, and T_9. This was because the proper control of weeds reduced the weed density and allowed crop plants to have sufficient space, light, nutrients and moisture, which resulted in increased number of panicles m^{-2}. The lowest number of panicles m^{-2} (129.67) were recorded in the weedy check (T_{10}). The treatment T_9 produced the highest panicle length followed by T_5. The shortest panicle length was found in the weedy check (T_{10}). Weeds always compete with crops for the available resources like light, water, nutrients, etc. which are necessary for plant growth to produce more grains (Antigua *et al*., 1988). In this study, the highest weed infestation in the non-weeded plots resulted in the lowest number of grains per panicle. The treatment T_9 produced the maximum number of grains per panicle, although statistically similar to T_5, T_6, T_7, and T_8 (Table-3). This was mainly due to the weed free conditions in these treatments. Hasanuzzaman *et al.* (2008) and Ahmed *et al*. (2005) found that the application of any herbicide resulted in similar number of grains per panicle. In this study, 1000-grain weight was not significantly affected by the weeding treatments. The highest grain weight was observed in T_9 followed by T_3, whereas the lowest in T_{10}. Also, sterility (%) was not significantly affected by the weeding treatments. However, the highest percentage of grain sterility was observed with T_{10} and the lowest with T_1.

Table-3. Plant characters and yield components of transplanted rice as affected by the different weed control treatments.

Treatments	Plant height (cm)	Panicle m^{-2}	Panicle length (cm)	Grains $panicle^{-1}$	1000-grain weight (g)	Sterility (%)
T_1=Zealus 10 WP at 125 g ha^{-1}	96.2	165.7 bc	23.9	99.3 abc	23.8	12.7
T_2=Amaraj 10 WP at 150 g ha^{-1}	96.3	201.7 ab	23.7	100.3 abc	23.1	14.5
T_3=Siniron 10WP at 187 g ha^{-1}	93.1	178.7 b	23.7	91.0 bc	24.1	13.8
T_4=Herbikill 10 WP at 150 g ha^{-1}	93.3	197.3 ab	23.3	83.3 c	24.2	14.5
T_5=Res Q 25 EC at 1.2 L ha^{-1}	95.9	233.7 a	24.0	103.7 ab	23.8	13.1
T_6=Remover 10 WP at 187 g ha^{-1}	95.3	197.3 ab	23.3	103.7 ab	23.6	14.5
T_7=Safety 10 WP at 200g ha^{-1}	96.5	207.3 ab	23.9	99.7 ab	23.6	14.1
T_8=Laser 10 WP at 125 g ha^{-1}	95.8	174.7 bc	23.1	108.7 ab	23.3	14.3
T_9=Three hand weeding	96.0	192.3 ab	24.3	111.3 a	24.1	13.9
T_{10}=Weedy check	91.8	129.7 c	23.1	66.3 d	23.06	17.40
$LSD_{0.05}$		44.94		16.10		

In a column the values having common letter(s) do not differ significantly.

Yield and harvest index

The maximum level of weed control provided by T_9 (three hand weedings) was reflected on the maximum grain yield (5.52 t ha^{-1}) of the transplanted rice cultivation in the dry season (Table-4). However, yield in T_9 did not differ statistically from T_5 and T_6, which might be due to the higher number of panicles m^{-2} and the higher number of grains per panicle in those treatments (Table-3). T_5, T_6, and T_9 produced 40.6%, 39.3, and 46.7% higher yield than the non-weeded control, respectively. This finding is partially supported by Hasanuzzaman *et al.* (2008). Straw yield was also significantly affected by the weeding treatments (Table-4). The highest straw yield was observed with T_9, which however was statistically similar to T_5 and T_6. This shows that the herbicide application was equally effective to the hand weeding treatments. Weedy check plots produced the lowest straw yield. No significant differences in terms of harvest index were found in this study. However, the highest harvest index was observed in T_1 (Zealus 10 WP), where the lowest harvest index was observed in T_{10} (weedy check).

Table-4. Yield and harvest index of dry season rice as affected by the different weed control treatments.

Treatments	Grain yield (t ha^{-1})	Straw yield (t ha^{-1})	Harvest index (%)
T_1=Zealus 10 WP at 125 g ha^{-1}	4.27 b	5.66 c	43.0
T_2=Amaraj 10 WP at 150 g ha^{-1}	4.64 b	6.54 abc	41.44
T_3=Siniron 10WP at 187 g ha^{-1}	4.37 b	6.05 bc	42.1
T_4=Herbikill 10 WP at 150 g ha^{-1}	4.37 b	6.30 bc	41.0
T_5=Res Q 25 EC at 1.2 L ha^{-1}	4.95 ab	6.77 ab	42.2
T_6=Remover 10 WP at 187 g ha^{-1}	4.84 ab	6.75 ab	41.8
T_7=Safety 10 WP at 200g ha^{-1}	4.53 b	6.60 abc	40.6
T_8=Laser 10 WP at125 g ha^{-1}	4.50 b	6.57 abc	40.6
T_9=Three hand weeding	5.52 a	7.39 a	42.9
T_{10}=Weedy check	2.94 c	4.40 d	40.0
$LSD_{0.05}$	0.72	0.96	

In a column the values having common letter(s) do not differ significantly.

Economic analysis of weed control options in dry season rice

Different weed control treatments involved different weed control costs, which affected the total production cost in dry season rice cultivation (Table-5). The economic analysis indicated that the maximum cost of weeding was hand weeding (T_9) due to increased labour requirement. This finding is also supported by Hasanuzzaman *et al.* (2008). The treatment T_5 recorded the second highest cost, which was almost one-third of T_9. The maximum gross return from dry season rice cultivation was found in T_9 (three hand weedings) followed by T_5 (Res Q 25 EC) and T_6 (Remover 10 WP). The lowest gross return was obtained from the weedy check due to its lowest production of grain and straw. The highest gross margin was received from the treatment T_6 (Remover 10 WP) which was even higher than T_9 (Table-5). By cost dominant analysis, it was found that five treatments T_2, T_3, T_5, T_7, and T_9 were cost dominated. In these treatments the cost was higher, but the gross margin was lower than that of many other treatments (Table-6). The marginal analysis of non-dominated treatments showed that the highest marginal rate of return (2630.02%) was found in T_6 (Remover 10 WP) (Table-7). This finding indicates that the highest marginal rate of return (MRR) on investment was obtained by the herbicide Remover 10 WP, which means that this herbicide treatment was more profitable than hand weeding.

Table-5. Treatment wise variable cost (herbicide and labor), gross return, and gross margin of the dry season rice.

Treatments	Variable cost (US $ ha^{-1})	Gross return (US $ ha^{-1})	Gross margin (US $ ha^{-1})
T_1=Zealus 10 WP at 125 g ha^{-1}	16.27	833.4	817.2
T_2=Amaraj 10 WP at 150 g ha^{-1}	29.63	908.4	878.8
T_3=Siniron 10WP at 187 g ha^{-1}	35.15	854.8	819.6
T_4=Herbikill 10 WP at 150 g ha^{-1}	19.45	856.6	837.1
T_5=Res Q 25 EC at 1.2 L ha^{-1}	49.36	967.6	918.3
T_6=Remover 10 WP at 187 g ha^{-1}	26.75	947.1	920.3
T_7=Safety 10 WP at 200g ha^{-1}	37.64	888.4	850.8
T_8=Laser 10 WP at125 g ha^{-1}	24.39	882.6	858.3
T_9=Three hand weeding	158.36	1077.9	919.6
T_{10}=Weedy check	00.00	577.4	577.4

Table-6. Treatment wise cost dominant analysis.

Treatments	Gross margin (US $ ha^{-1})	Variable cost (US $ ha^{-1})	Cost dominated treatments
T_6=Remover 10 WP at 187 g ha^{-1}	920.32	26.75	
T_9=Three hand weeding	919.57	158.36	*
T_5=Res Q 25 EC at 1.2 L ha^{-1}	818.28	49.36	*
T_2=Amaraj 10 WP at 150 g ha^{-1}	878.80	29.63	*
T_8=Laser 10 WP at125 g ha^{-1}	858.25	24.39	
T_7=Safety 10 WP at 200g ha^{-1}	850.79	37.64	*
T_4=Herbikill 10 WP at 150 g ha^{-1}	837.12	19.45	
T_3=Siniron 10WP at 187 g ha^{-1}	819.64	35.15	*
T_1=Zealus 10 WP at 125 g ha^{-1}	817.16	16.27	
T_{10}=Weedy check	577.43	0.00	

Table-7. Marginal analysis of non-dominated treatments.

Treatments	Gross margin (US$ ha^{-1})	Variable cost (US $ ha^{-1})	Marginal variable cost (US $ ha^{-1})	Marginal gross margin (US $ ha^{-1})	Marginal rate of return (%)
T_6=Remover 10 WP at 187 g ha^{-1}	920.32	26.75	2.36	62.07	2630.02
T_8=Laser 10 WP at125 g ha^{-1}	858.25	24.39	4.94	21.13	427.76
T_4=Herbikill 10 WP at 150 g ha^{-1}	837.12	19.45	3.18	19.96	627.76
T_1=Zealus 10 WP at 125 g ha^{-1}	817.16	16.27	16.27	239.73	1473.45
T_{10}=Weedy check	577.43	0.00			

CONCLUSION

It might be concluded that the use of post-emergence herbicides may be an easy and cost-effective alternative for weed control. Application of Remover 10 WP for weed control in dry season rice cultivation maximized the rate of return to capital and can be used as an alternative weed control option in dry season rice cultivation.

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POTENTIAL OF DUCKWEED (*Wolffia arrhiza*)-AN INVASIVE AQUATIC PLANT AS FISH FEED IN TILAPIA (*Oreochromis niloticus*) FRY REARING

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ABSTRACT

Duckweed (Wolffia arrhiza) is a tiny aquatic plant, abundant in nutrient rich wetlands which contain up to 43% protein by dry weight and could be used as feed for rearing Tilapia (Oreochromis niloticus) fry. Tilapia fingerlings are in high demand in the development of inland fish production in the country. The aim of this study was to evaluate the potential of duckweed as feed in fry rearing of Tilapia and to utilise this plant in a profitable manner. The trial was carried out at National Aquatic Resources Research & Development Agency, Sri Lanka for a period of 41 days. Six cement tanks (0.75 m^3) were stocked with Tilapia fry (0.0754±0.0441g) according to the stocking density of 500 fry m^{-3}. Fresh duckweed (Dw) and commercial feed (Cf) were provided daily at a rate of 5% of body weight. Fish were sampled weekly and the feed amounts were adjusted according to total biomass in respective tanks, assuming there was no mortality. The pH and temperature were measured daily. Nitrite (NO^{-2}), Nitrate (NO^{-3}) and Total Ammonia Nitrogen (TAN) were measured weekly. Average Daily Growth (ADG), Specific Growth Rate (SGR-W), Condition Factor (CF), Final body weight (BW_{final}), Feed Conversion Ratio (FCR), % survival rates of Tilapia and Toxic Ammonia (NH_3) were determined. The Mean ADG (0.0261±0.0077, 0.0039 ± 0.0020 g day^{-1}), BW_{final} (1.1450±0.7390, 0.2880 ± 0.2070 g) and CF (1.7727±1.18, 2.655±2.06) of Tilapia fed on Cf and Dw were significantly different ($p<0.05$), while SGR-W (6.7324±1.7879, 3.9035±0.4696) and % survival rates (44.6±16.35, 25.5± 6.74) were not significant statistically. The mean temperature, pH, (NO^{-2}), (NO^{-3}) and NH_3 in Cf and Dw provided tanks were not significantly different ($P>0.05$). FCR of Cf and Dw were 1.003±0.6097and 4.2066±0.2462. Wolffia arrhiza could be used as feed for Tilapia fry rearing but the amount Dw provided (5% of body weight) is not sufficient. Further research is needed to determine optimal amounts. The use of W. arrhiza as fish feed would reduce its nuisance value considerably.

Key words: Tilapia fry rearing, Duckweed, aquatic plant

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INTRODUCTION

Tilapia *(Oreochromis niloticus)* is an exotic fish, well established in Sri Lankan water bodies and it is the mainstay of reservoir fisheries in Sri Lanka (Amarasinghe, 1995). Though it is a naturally bred fish species with parental care, fry rearing with special attention on feeding would be much benefited to produce more fingerlings. These fingerlings could be used to stock perennial reservoirs and seasonal tanks to promote inland fisheries and culture based fisheries in the country. However, it is necessary to use a supplementary feed in the seed production of Tilapia, for better results. According to De Silva and Anderson (1995), the cost of feed is the highest recurrent cost of the aquaculture industry, which often ranges from 30% to 60% of the total cost depending on the intensity of the practice. As such, alternative feeds should be used to reduce the cost of feed in the aquaculture industry.

According to Islam *et al.* (2004) a tiny, fragile, free-floating, aquatic plant, Duckweed (Dw) (*Wolffia* spp.) could be used as an alternative fish feed. As cited by Ferdoushi *et al.* (2008), *Lemna minor, L. gibba, Wolffia arrhiza* and *Azolla pinnata* are free floating duckweeds included in Lemnaceae family. Mbagwu *et al.* (1990) have shown that fresh Dw is a good food source for Tilapia, as it contains about 35-45% crude protein (CP) with good amino acid and mineral profiles. Likewise Hassan and Edwards (1992) have shown that Dw is converted efficiently to live weight by certain fish, which include carp and Tilapia. According to Yilmaz *et al.* (2004), Dw occurs naturally in heavily fertilized/nourished wetlands. Mazid *et al.* (1992) has shown that the Dw grows abundantly in warm climatic zones in fresh water with rapid rate of vegetative reproduction *i.e.* double their quantity in three days or less.

As the size of Dw is very tiny, small fish (fry) can easily consume it. As such, newly harvested duckweed plants contain up to 43% protein by dry weight and can be used without further processing as a complete feed for fish (Mbagwu *et al.*, 1990). As cited by Islam *et al.* (2004), Dw has long been recognized as a potential source of protein for animals and studies conducted in various countries of the world, e.g. the former U.S.S.R., U.S.A. and Canada have demonstrated the nutritional benefits of Dw for both livestock and fish. Accordingly, the attempt of this study is to evaluate the efficacy of duckweed (*Wolffia arrhiza*) as fish feed in the fry rearing of Tilapia and reduce the nuisance value of duckweed.

MATERIALS AND METHODS

The trial was carried out at National Aquatic Resources Research & Development Agency (NARA), Colombo, Sri Lanka for a period of 41 days from 23rd August, 2005 to 4th October, 2005. Six rectangular cement tanks (1.6m × 1.0m × 0.5m; 0.75 m^3) were cleaned, washed and filled with tap water up to 45 cm that was the maximum depth of the tanks. After 5 days, Tilapia fry (initial weight 0.0754±0.0441g) were stocked according to a stocking density of 500 fry m^{-3}. Tilapia fry were obtained from the Aquaculture Development Centre in Udawalawe, Sri Lanka.

Two feed types i.e. fresh duckweed (Dw) (*W. arrhiza*) and commercial feed (Cf) were used as feed for Tilapia fry and tested in triplicate in randomly selected tanks. Cf was the control feed and was purchased from Ceylon Aquatech (Pvt.) Limited, Rock House Lane, Colombo-15, Sri Lanka. Dw was cultured in two cement tanks using organic manure, cow dung, and collected using hand nets, washed with tap water and left to drain for 45 min. before being used as feed.

During the experimental period fish were fed twice per day, once in the morning (0930 hrs) and once in the afternoon (1500 hrs) at a daily rate of 5% of body weight. Twenty fish in each tank were sampled weekly to determine average weight of the fish in respective tanks and calculated the total biomass accordingly. The feed amount was adjusted according to the total biomass in respective tanks, assuming there was no mortality.

The water temperature and the pH of the tanks were measured using glass mercury thermometer and the pH meter (Model: GENWAY-3051) daily around 0900 hrs. Nitrite-nitrogen (NO^{-2}), Nitrate-nitrogen (NO^{-3}) and Total Ammonia Nitrogen (TAN) were measured once a week using DR 4000 spectro photometer. Toxic un-ionized ammonia (NH_3) was calculated using the SRAC publication No.463 (Durborow *et al.*, 1997). After 41days of rearing, the fish were harvested, counted and % survivals were determined in each tank, respectively. As such, the final sampling (determine weight and total length of fish respectively) was carried out with 20 fish collected randomly from each tank.

Proximate analysis of Cf was carried out using standard method described in APHA (1985). The moisture content was determined by oven drying a weighed sample in porcelain crucibles at 105 ^oC for 24 hrs. The total volatile matter lost at this temperature was taken as the moisture content. The Ash content was determined by incinerating the dried samples 1-2 g of flesh overnight in a Muffle furnace at 550 ^oC. The percentage of protein (N × 6.25) was estimated by semi-micro Kjeldahl digestion, distillation and titration. The % of fat was determined using the chloroform method (Bligh and Dyer, 1959).

The Average Daily Growth (ADG), Specific Growth Rate in weight (SGR-W), Feed Conversion Ratio (FCR), Condition factor (CF) and % survival of the fish in each tank were determined according to the following equations.

$$ADG\ (g/day) = \frac{\text{Final weight of fish- Initial weight of fish}}{\text{Days of rearing}}$$

$$CF = \frac{\text{Total weight of fish(W)}}{\text{Standard length }(L)^3} \times 100 \quad \text{Bagenal, 1978}$$

$$\%\ Survival = \frac{\text{No. of fish harvested}}{\text{No. of fish stocked}} \times 100$$

$$SGR = \frac{Ln_{\text{Final weight}} - Ln_{\text{Initial weight}}}{\text{Experimental duration}} \times 100 \quad \text{Ricker, 1979}$$

$$FCR = \frac{\text{Dry weight of feed given(g)}}{\text{Wet weight gain(g)}} \times 100 \quad \text{Hepher, 1988}$$

Two- Sample t-tests were applied to determine the significance of the calculated indices in two variables. Statistical significance was assessed using a probability level of $P=0.05$.

RESULTS AND DISCUSSION

Mean final body weight (BW_{final}), CF and FCR of the fish that was fed on Cf and Dw were significantly different ($P<0.05$), respectively. However, SGR-W and % survivals of the fish in these two feed types were not significantly different ($P>0.05$), respectively (Table-1). Nevertheless Fasakin *et al.* (1999) found that the difference in final weights, specific growth rates, feed conversion of tilapia that were fed on diets containing up to 20% of Dw were not significantly different from those of tilapia that were fed on the control diet.

Average Daily growth (ADG)

The mean ADG of the fish fed on Dw was 0.0039±0.0020 g/day and it was significantly lower than the ADG of the fish fed on Cf (0.0261±0.0077 g/day). It may be due to the insufficient amount of Dw provided for the fish. Accordingly, the provided feed amount (5% of body weight) should be increased up to optimal levels to improve the growth of fish.

Final Body weight (BW_{final})

The BW_{final} of Tilapia fry fed on Cf and Dw were significantly different and lower in the fish fed on Dw than the fish fed on Cf (Table-1). The low growth of Tilapia fry may be due to insufficient amounts of Dw (5% of body weight) that was provided for fish. Nevertheless, polyculture of Tilapia, Common carp (*Cyprinus carpio*) and Mrigal

(*Cirrhinus mrigala*) in mud ponds for food fish culture has recorded fast growth with Dw (Thy *et al.*, 2008). However the SGR-W of the fish was not significantly different in these two feeds. It has shown the suitability of the two feeds provided.

Table-1. Mean values of Average Daily Growth (ADG), Final Body Weight (BW_{final}), Specific Growth Rate in weight (SGR-W), Feed conversion ratio (FCR) and % survival rates of Tilapia (*Oreochromis niloticus*) fry fed on commercial feed (Cf) and duckweed (Dw)

Feed type	ADG ($g\ day^{-1}$)	SGR-W	% Survival	CF	BW_{final} (g)	FCR
Cf	0.0261[a] ±0.0077	6.7324[c] ±1.7879	44.6[d] ±16.35	1.7727[e] ±1.18	1.1450[g] ±0.739	1.0030[j] ±0.6097
DW	0.0039[b] ±0.0020	3.9035[c] ±0.4696	2 5.5[d] ±6.74	2.6550[f] ±2.06	0.2880[h] ±0.207	4.2066[k] ±0.2462

All values over Mean±S.D. Mean with the same letter in the same column is not statistically significant (P<0.05)

Condition factor (CF)

CF, which is related to both growth and feeding, was another variable checked in the study. It was significantly different ($p<0.05$) in these two feed types and significantly higher in Dw than in Cf (Table-1). In fisheries science, the CF is used to compare the "condition" "fatness" or wellbeing of fish (Anene, 2005). It has been shown that the wellbeing and health of Tilapia fry that were fed on Dw were significantly higher than that fed on Cf. Accordingly, the use of Dw as feed in fry rearing of Tilapia has shown good indicator for better growth.

Feed conversion ratio (FCR)

The FCR of Dw and Cf with Tilapia fry were significantly different ($p<0.05$) and significantly higher in Dw than the Cf. The Cf is a complete commercial feed that was manufactured to fulfill the nutritional requirement of the fish (Table-1). Accordingly the FCR value should be low. The correlation of ADG and FCR in two feed types has shown negative relationship and the relationship in tanks provided with Dw was very strong ($R^2=0.9107$) (Fig.1). It was shown that Dw supported the growth of Tilapia fry more than Cf. It may be due to the nourishment of Dw.

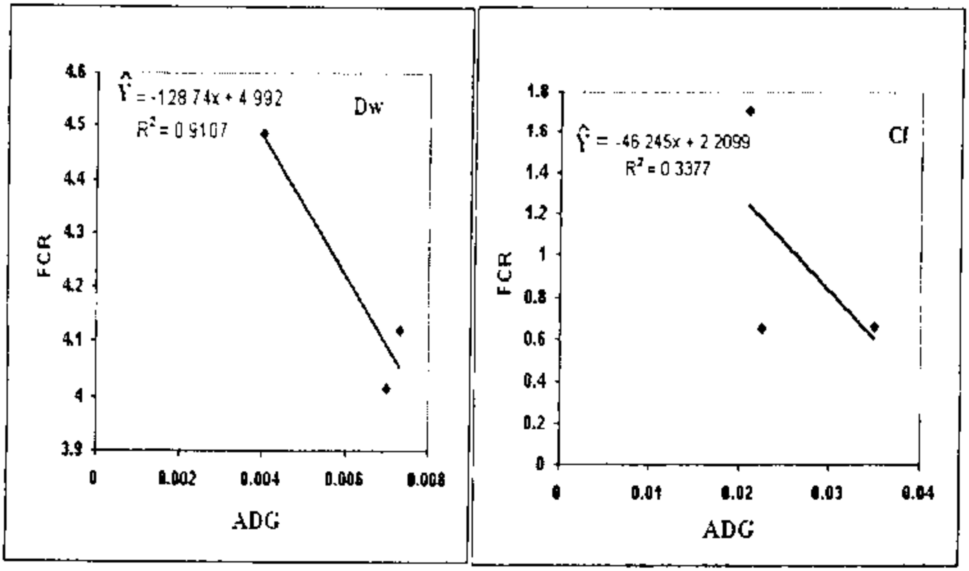
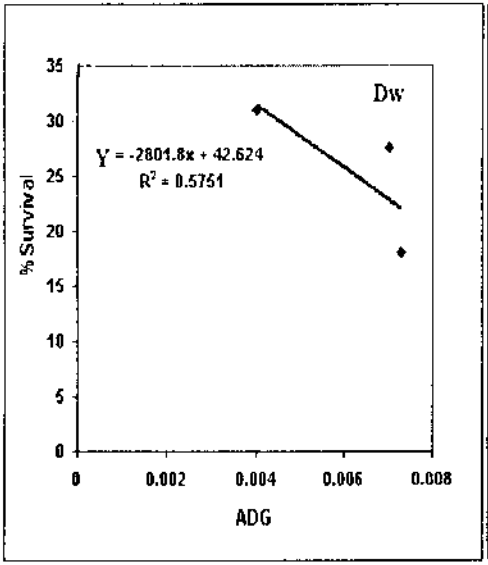


Figure 1. Relationship of feed conversion ratio (FCR) and average daily growth (ADG) of Tilapia *Oreochromis niloticus*) fry fed on duckweed (Dw) and commercial feed (Cf).

Fish survival rate (%)

The % survivals of the fish fed on Cf and Dw were not significantly different (P>0.05). However, the % survival of the fish that was fed on Dw was recorded as 25.5±6.74 which was lesser than the % survival of the fish fed on Cf (44.6±16.35). It could be due to the higher standard deviation of the respective mean values (Table-1). Thy *et al.* (2008) have observed the high % survival (89.6) of Tilapia than Common carp (75) and Mrigal (75) while fed on Dw in polyculture grow out trials.

The correlation between % survival and ADG of the fish in Cf provided tanks was positive and in Dw provided tanks was negative (Fig. 2). It could be due to the insufficient amount of feed that provided as Dw. However, the correlation was not strong in Dw provided tanks as well as Cf provided tanks. Due to the insufficient feed amount that provided Dw as 5% of body weight was not sufficient, the cannibalism could have happened. Katavic *et al.* (1989) have observed the size variation of the fish and food availability are the primary causes of cannibalism. As such cannibalism among tilapia fry and fingerlings has been identified as one of the major problems by small-scale hatchery operators (Pantastico *et al.*, 1988). Accordingly cannibalism may be the cause for the low % survival of Tilapia fry fed on Dw and for the negative relationship between ADG and % survival.



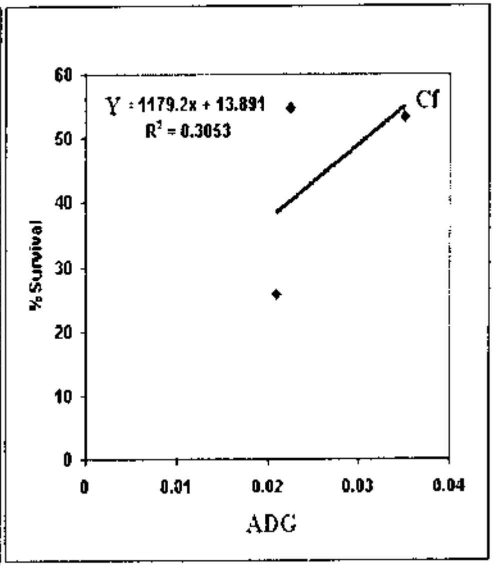


Figure 2. Relationship of % survival and average daily growth (ADG) of *Oreochromis niloticus* fry with commercial feed (Cf) and duckweed (Dw).

Water quality

There were no differences on water quality parameters (pH, water temperature, NO^{-2}, NO^{-3} and TAN (Table-2).Water quality parameters measured over the 41 days rearing period are shown in Table-2. During the experimental period, the values of water temperature were more or less similar in all six tanks for two treatments and fluctuated from 28.0 ºC to 33.0 ºC. It may have therefore affected fish growth similarly in these two feed types.

The nitrate levels in Cf provided and Dw provided tanks were 1.214±0.765 and 1.679±0.649, respectively. According to Durborow *et al.* (1997) and Floyd and Watson, (2005) nitrate is not toxic to fish except at extremely high levels, and can be considered harmless and it is used by plants, including algae, for food. As cited by Boyd (1992), concentration of nitrite as low in 0.5 mg l^{-1} was toxic to certain cold water fish. However, the levels of nitrite in this trial were 0.0043±0.0029 mg l^{-1} in Cf provided tanks and 0.0114±0.0101 mg l^{-1} in Dw provided tanks and these are very low levels than the toxic levels.

Ammonia is the major end product in the breakdown of proteins in fish (Durborow *et al.* 1997). The toxic Ammonia (NH_3) levels in Dw provided tanks (0.0403±0.0307 mg l^{-1}) was lower than Cf provided tanks (0.0587±0.0596 mg l^{-1}) and not significantly different (P>0.05) (Table-2). As such the amount of NH_3 in Dw provided tanks were lower than the NH_3 in Cf provided tanks throughout the rearing period (Fig. 3). According to Pompa and Masser (1999) the NH_3 levels in these NH_3 tanks were not lethal (>2 mg l^{-1}), sub-lethal (>3 mg l^{-1}) or depress food consumption of the fish (<0.08 mg l^{-1}). According to Durborow *et al.* (1997) 0.6 mg/l are capable of killing fish and 0.06 mg/l can cause gill and kidney damages. However, the toxic NH_3 levels in Cf provided tanks and Dw provided tanks were not reached this level. The toxic NH_3 could be produced in fish tanks in two ways. (1) through the deteriorating of remaining feed and (2) through the excreta of fish. Toxic ammonia in Cf provided tanks were produced in these two ways but in Dw provided tanks were produced through the fish excreta only, because the Dw did not deteriorate, as it is a live aquatic plant. Accordingly the water quality in Dw provided tanks would be more favourable than in the Cf provided tanks for Tilapia fry rearing. This is one of the advantages in feeding fish with Dw.

Table-2. Water quality parameters measured over the 41 days rearing period of Tilapia (*Oreochromis niloticus*) fry with commercial feed (Cf) and duckweed (Dw).

Feed type	Water quality parameters				
	pH	Water Temp °C	Nitrite-nitrogen (NO^{-2}) (mg l^{-1})	Nitrate-nitrogen (NO^{-3}) (mg l^{-1})	Toxic Ammonia (NH_3) (mg l^{-1})
Cf	8.56^{a} ±0.5361	29.26^{b} ±1.3936	0.0043^{d} ±0.0029	1.214^{e} ±0.765	0.0587^{f} ±0.0596
Dw	8.72^{a} ±0.6206	29.26^{b} ±1.3936	0.0114^{d} ±0.0101	1.679^{e} ±0.649	0.0403^{f} ±0.0307

All values over Mean ±S.D. Mean with the same letter in the same column is not statistically significant (P<0.05).

Proximate composition and nutritional value

The Cf used in this trial has provided 32.5±1.13 % of crude protein (Table-3). The Dw is rich in crude protein and it will provide 43% of crude protein in dry weight (Yilmaz *et al.*, 2004). However, the estimated dietary protein requirement for maximal growth of juvenile Tilapia is 30 % (NRC, 1993). This amount of protein is provided through the fishmeal in Cf which is expensive. Nevertheless, Dw could provide this amount of protein without any cost and could be obtained from the paddy fields as it is an invasive plant for paddy farming. Accordingly, use of Dw as fish feed is much benefited for Tilapia fry rearing as well as this intervention will result in the management of duckweed.

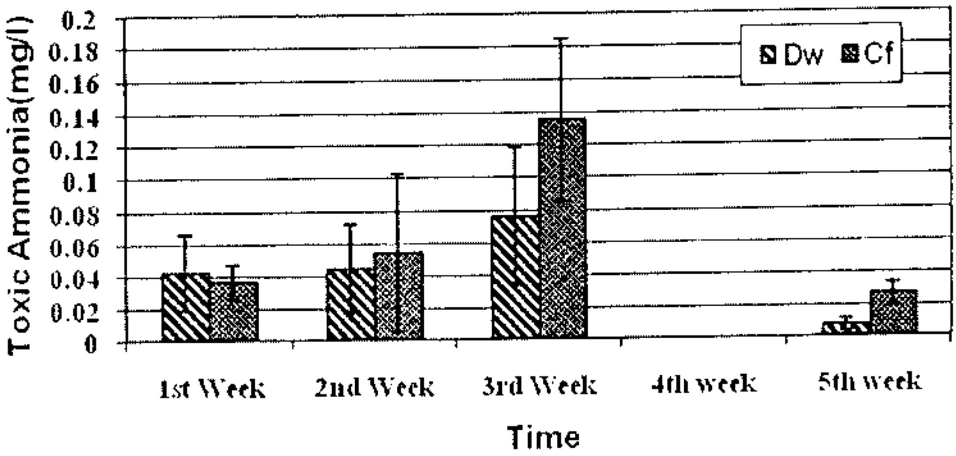


Figure 3. The toxic Ammonia (NH_3) levels in duckweed (Dw) and commercial feed (Cf) provided tanks.

Table-3. Ingredient composition and proximate composition of commercial feed (Cf) used in Tilapia fry rearing trials (APHA, 1985).

Ingredient composition	Proximate composition (%)	
Fish products	Dry matter	90.23±0.17
Other animal by-products	Moisture	9.77±0.17
Cereals & Cereal by-product	Ash	8.81±0.14
Oilseed meals	Protein	32.5±1.13
Fish oil	Fat	8.64±0.09
Minerals	Carbohydrate	49.97±1.64
Vitamins		
Permitted additives		

According to Hilman and Culley (1978), Dw as a natural protein source has a better array of essential amino acids (EAA) than most other vegetable proteins and more closely resembles animal protein including high concentration of Lysine (10.9% in dry weight) (Table-4). It is higher than the lysine requirement (1.3g/kg) of Nile Tilapia (Santiago and Lovel, 1988). According to Palavesam *et al.* (2008) Lysine is one among the ten indispensable amino acids that are required in dietary protein and it is the most limiting amino acid in plant proteins and the most critical amino acid in fish feed. Accordingly, use of Dw as fish feed in Tilapia fry rearing is much benefited.

Table-4. Proximate and amino acid compositions of duck weed (adopted from Yilmaz *et al.*, 2004) and amino acid requirement of juvenile Tilapia (*Oreochromis niloticus*) (NRC, 1993).

Proximate composition	g $100g^{-1}$ dry weight	Amino acid requirement of juvenile Tilapia (NRC, 1993)
Crude protein	**18.38**	
Lipid	2.32	
Ash	23.71	
Amino acids:		
Aspartic acid	1.096	
Serine	0.471	
Glutamic acid	1.216	
Glycine	0.438	
Alanine	0.747	
Tyrosine	0.287	
Essential Amino Acids		
Arginine		1.18
Histidine	0.224	0.48
Isoleucine	0.518	0.87
Leucine	0.696	0.95
Lysine	10.900	1.43
Methionine	0.179	0.75
Phenylalanine	0.623	1.05
Threonine	0.494	1.05
Tryptophan		0.28
Valine	0.646	0.78

Yilmaz *et al.* (2004) have reported that Dw has no anti-nutrients as the most plant-derived nutrient sources like soybean meal, rapeseed meal, lupin seed meal, pea seed meal, sunflower oil cake, cottonseed meal, leucaena leaf meal contain a wide variety of anti-nutritional substances that affect the growth and health of fish. Therefore, Dw provides an easy, practical and cheaper fish feed as it requires no processing to destroy any anti-nutrients. Furthermore, according to Leng *et al.* (1995) Dw grown in nutrient-rich water has a high concentration of trace minerals, Potassium (K), Phosphorus (P) and pigments, particularly carotene and xanthophylls which make Dw especially valuable as a dietary supplement for fish. According to Chaturvedi *et al.* (2003) compared with most other plants, Dw leaves contain little fibre (5% in dry matter for cultivated plants) and little or no indigestible material even for monogastric animals.

However, Dw is an unwanted invasive aquatic plant for paddy cultivation in Sri Lanka. In the meantime it is nutritious feed with no cost for fish farmer. To optimize the use of natural resources, the integration of paddy cultivation and fish culture is an ideal strategy. Then more attention should be paid on the integration of rice farming and fish culture with Dw in Sri Lanka.

The aquatic plant *Wolffia arrhiza* could be used as supplementary feed for Tilapia fry rearing. Nevertheless, the amount provided (5% of body weight) was not sufficient. Further investigation, therefore, is necessary to determine the optimal amounts of Dw needed for the rearing of fry of Tilapia.

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