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INVASIVE WEED *Parthenium hysterophorus:* RESPONSES TO CHEMICAL AND ALLELOPATHIC EXTRACTS AT DIFFERENT STAGES

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ABSTRACT

Parthenium hysterophorus L. is a noxious alien invasive weed species rapidly spreading in Bangladesh. *P. hysterophorus* examined the prospects of using allelopathic extracts during the period of July 2021 to June 2022. Experiments were conducted in the net house at Sher-e-Bangla Agricultural University, in non-cropped areas of Khustia and cropped areas of Chudanga. The net house experiments were laid out in complete randomized design (CRD) and replicated three times separately for each growth stage. Results showed that by using pretilachlor, pendimethalin, bensulfuron methyl + pretilachlor, bensulfuron methyl + acetachlor, oxadiazon, and pyrazosulfuron-ethyl + pretilachlor decreased the seed germination of parthenium at pot bioassay under both puddle and dry sown condition. At rosette stage of parthenium weed, glyphosate and carfentrazone ethyl (5%) + glyphosate with ½, ¾, and full doses gave 100% mortality at three weeks after treatment (WAT). In cropped areas carfentrazone Ethyl (5%) + glyphosate (36%) and quizalofop-p-ethyl resulted in a 100% and 90% mortality rate at 21 DAT. Overall, the efficacy of herbicides was more effective on rosette parthenium than bolted plants. A phytotoxic response to weed growth was induced by aqueous extracts of *Oryza sativa* L. var. Boteswar, *Triticum aestivum* L. var. BARI gom-21, *Helianthus annus* L., *Datura metel*, *Mangifera indica* L., *Delonix regia*, and *Acacia nilotica*. *Delonix regia* was the most effective for inhibition of germination (29%). Whereas *Datura metel* extracts and *Oryza sativa* var. Boteswar straw extracts significantly reduced root (1.5 mm) and shoot (3.8 mm) length.

Keywords: Parthenium, Control, Herbicides, Plant extracts, Herbicidal activity

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INTRODUCTION

Parthenium hysterophorus, an alien invasive species, is an annual ephemeral herb. In Bangladesh, this weed spreads aggressively in wastelands, degraded areas, along water channels, roadsides, and railway tracks. It has also been reported in cultivated lands (Masum et al., 2022). This noxious weed affects crop production, animal husbandry, human health, and biodiversity. Fast growth rate, high reproductive potential, adaptive nature, and interference by allelopathy are the major strategies to this weed's rapid spread and successful establishment in any ecosystem. Above all, the lack of natural enemies of this weed in Bangladesh and many parts of the world away from its natural homeland is also contributing to a large extent to the rapid spread of this weed (Masum et al., 2013). Among various weed control measures, herbicides are widely preferred by farmers for the control of *P. hysterophorus* due to cost- and time effectiveness. Earlier Kanchan and Jayachandra (1977) found that bromocil, diuron, and terbacil @1.5 kg ha^{-1} were very effective against parthenium. Mishra and Bhan (1994) have recommended the sulphonylurea herbicides like chlorimuron ethyl and metsulfuron-methyl to control parthenium in non-cropped areas. Javaid (2007) found that Bromoxynil + MCPA was the most effective, where all the employed doses of the herbicide completely killed the weed within 7 days at both the growth stages. Atrazine and glyphosate killed 5 and 8 weeks old parthenium plants by 5-10, and 12 days, respectively. When the weed was in its fifth week of growth, ametryn + atrazine took 11 days to control it, whereas it took 14 days in its eighth week. Butachlor was found to be the least effective herbicide. Bajwa et al. (2019) reported that the sequential application of pendimethalin and bispyribac-sodium plus bensulfuron methyl provided an excellent way to control (90%) parthenium weed and enhanced the productivity and profitability of rice varieties under direct-seeded conditions. In contrast, several well-known herbicides such as paraquat, trifularin, diphenamid, napropamide, acetanilides, alachlor, metochlor, and propachor are ineffective against parthenium weed (Khan et al., 2012).

Managing parthenium weed to limit its negative impacts on crop growth and yields is extremely important. Since chemical herbicides also pose health risks, the use of these chemicals should be reduced to a minimum. However, the increased use of herbicides poses severe environmental and public health concerns. Moreover, the evolution of resistance to several herbicides, including atrazine, fenoxaprop-p, flupyrsulfuron, isoproturon, and triazine, has been reported (Chhokar et al., 2008). Therefore, alternative weed management systems are required, which are based on naturally occurring compounds (Cuthbertson and Murchie, 2005). Thus plant derivatives are more environmentally safe than synthetic chemicals (Hashim and Devi, 2003). During the past two decades, much work has been done on plant-derived compounds as ecologically safe alternatives to herbicides for weed control (Shafique et al., 2011). Hence, there is a need to test well-known allelopathic plants to explore their potential phytotoxic activity against *Parthenium* weed. Considering the above-mentioned facts, therefore, the present research work was carried out to evaluate the potential of different chemical herbicides against this weed and some herbaceous plant species were evaluated as well for their herbicidal activity against *Parthenium* weed.

MATERIALS AND METHODS

A. Chemical control of parthenium weed (*Parthenium hysterophorus* L.)

Seeds of *P. hysterophorus* were collected from the parthenium infestation areas. Forty-eight clay pots (30 cm diameter and 35 cm height) were filled with approximately 5 kg of sandy loam soil and was placed in a plastic shade house approximately 30/18 ±2 (day/night) with 60% relative humidity. Five seeds of P. *hysterophorus* were sown in each pot at about 2 cm depth and were watered regularly at field capacity.

Pre-emergence control

Commercially registered pre-emergence herbicides were applied to check pre-emergence control. The required herbicide quantity was calculated based on the recommendation and the pot's diameter. Six ml of the solution was then carefully sprayed in each pot using a hand sprayer. The efficacy of pre-emergence herbicides was studied under both dry sown and puddle sown conditions. In dry sown conditions, *P. hysterophorus* seeds were sown in pots filled with soil and irrigated lightly without flooding, simulating rainfall. Herbicides were sprayed on the day after sowing. In puddle-sown conditions, soil in the pots was puddled by hand, and excess water was removed after settling the clay. Twenty-five seeds were sown on the top layer and, after one day, again flooded to a depth of three centimeters. Water was drained after one day, and herbicides were sprayed on the third day after sowing. Flooding was again done after 24 hours and continued for the study.

The preemergence herbicidal treatments were: pretilachlor @ 0.75 kg a.i. ha^{-1}, pyrazosulfuron ethyl @ 1.25 kg a.i. ha^{-1}, pendimethalin @1.5 kg a.i. ha^{-1}, bensulfuron methyl + pretilachlor @ (0.18+ 0.4) kg a.i. ha^{-1}, bensulfuron methyl + acetachlor (0.04 + 0.14) kg a.i. ha^{-1}, butachlor @ 1.25 kg a.i. ha^{-1}, oxadiazon @ 0.10 kg a.i. ha^{-1}, pyrazosulfuron-ethyl + pretilachlor @ (0.0015 + 0.06) kg a.i. ha^{-1}, along with untreated control.

Weed control efficiency (WCE) was calculated based on the number of surviving weed seedlings in each pot (Mani *et al*., 1973).

WCE = x-y/x × 100, where, x - No. of *P. hysterophorus* seedlings in unweeded control and y - No. of *P. hysterophorus* seedlings in the treatment plot

Post-emergence control

Fourteen days after sowing, when the maximum germination was attained, the number of plants was reduced to ten $tray^{-1}$ to have a uniform density. Seven commercially registered post emergence herbicides against the broad-leaved weeds i.e., 2,4-D Amine, Quizalofop-p-Ethyl, Paraquat, Glyphosate, Bispyribac sodium, Carfentrazone Ethyl (5%) + Glyphosate (36%), Atrazine 50% + Mesotrione 5% were selected and applied for the chemical control of *P. hysterophorus* in two stages (Table 1). Three doses of each herbicide were formulated viz., the recommended dose (full dose), ¾ and ½ of the full dose. Each treatment was replicated three times. The quantity of each herbicide required for a single pot was estimated by calculating the area (0.05 m^2) of the pot used (dose with trade name given in Table 3). The required amount of each herbicide was applied to all pots except the control on three-week-old *P. hysterophorus* plants. In the control treatment, a similar amount of distilled water was applied to the plant. Data on the mortality rate was recorded at 7, 14, and 21 days after the herbicide application. The bolted stage experiments were done on the roadsides of Solokhanda, Bhermara, Khustia and, Akandabaria, Damurhuda, Chudanga in Bangladesh.

Table 1. Parthenium weed morphology and density at the time of herbicide application at Khustia and Chuadanga District in 2021-2022

Growth stage	Density (Nos.)	Plant height (cm)	No. of Leaves $plant^{-1}$	Flowering
Rosette	15	16-20	8-13	No
Bolted	15-20	55-90	22-29	Yes

Chemical control under field conditions

One suitable field site (5 m × 10 m) with a high and even density of *P. hysterophorus* was selected for the chemical control. These sites were in the jute field at Joyrampur, Damurhuda, Chudanga. Within the selected sites, twelve plots (1 × 1 m) were created and marked with the help of wire tags. Four herbicides viz., pendamethylen at 25.00 kg ha^{-1}, bispyribac sodium (Xtra Power 20 WP) at 150 L ha^{-1}, quizalofop-p-ethyl (Bikash 5EC) at 2.5 L ha^{-1}, and Carfentrazone Ethyl (5%) + Glyphosate (36%) (Trigger 365EW) at 2.00 L ha^{-1} were selected. Each treatment was replicated three times. A surfactant (1mL $1L^{-1}$) was added to enhance the herbicides' dispersion and penetration and promote residual effects on the *P. hysterophorus* plants. The formulated doses of these herbicides were applied to *P. hysterophorus* plants using a 5L manual pump sprayer early in the morning on a clear day. The plants were carefully observed throughout the experiment, and data on the mortality of the plants were taken at an interval of 7, 14, and 21 days after spray (DAS). Mortality rate (%) was calculated based on the number of weed plant kills at 7, 14, and 21 DAS. A visual criterion was developed to assign mortality rates (%) in selected plots in the field. For example, if 1/5 of the plot area is dead, 20% mortality was assigned; if ½ is dead, 50% is assigned, and so on. Due to many plants present in $plot^{-1}$ and time constraints, the number of individual plants $plot^{-1}$ was not counted.

B. Phytotoxic effects of allelopathic plant extracts against *P. hysterophorus* L.

Preparation of aqueous extracts

Rice (*Oryza sativa* L.) straw (VarietyBoteswar, wheat (*Triticum aestivum* L.) straw (variety BARI gom-21), stover of sunflower (*Helianthus annus* L.), plant of Datura (*Datura metel*), leaf of mango (*Mangifera indica* L.), leaf of krishnachura (*Delonix regia*) , leaf of acacia (*Acacia nilotica*) were collected and kept at room temperature for drying and then oven-dried at 70ºC for 48 h. These were then cut into small (2 cm) pieces and separately soaked in distilled water at a ratio of 1: 20 w/v (herbage: water) for 24 h at room temperature. Materials were filtered through a muslin cloth followed by Whatman No. 1 filter paper.

Germination studies

In a laboratory bioassay, the effect of different concentrations of the extracts on germination and early seedling growth of parthenium was studied. Petri plates of 9 cm diameter were sterilized at 150 °C for two hours. Ten seeds of parthenium were placed in each Petri plate lined with filter paper. Four ml of different concentrations of extracts of each studied species was added to each Petri plate. Treatment in a similar manner but with distilled water was served as control. Each treatment was replicated four times. Petri plates were arranged in a completely randomized design with a factorial arrangement of species extract concentration in a growth room at 25°C. Germination was recorded daily for 20 days.

Seedling growth studies

Extracts of all collected species were tested for their post-emergence herbicidal potential against parthenium weed through seedling growth bioassay in pots kept in the laboratory. Twenty seeds of parthenium weed were sown in plastic pots (10 cm dia. and 10 cm depth) filled with 650 g sandy loam soil per pot initially moistened with 200 mL of distilled water. Each treatment was replicated three times, and the experimental units were arranged in a completely randomized design. Three seedlings were maintained per pot. Six mL aqueous extract of each plant species' was sprayed over 15 days-old parthenium weed seedlings. Seedlings in the control treatment were also similarly sprayed with distilled water. Twenty-five old seedlings (10 days after spraying) were uprooted from pots, and their fresh weights were recorded. Germination percentage and seedling length were calculated as described by Masum et al. (2019).

Statistical analysis

The data collected were statistically analyzed following ANOVA, and the means were compared based on the least significant difference (LSD) at a 0.05 level of significance. The statistical software ‘STATIX 10’ was used for the analysis.

RESULTS AND DISCUSSION

Chemical herbicidal effects on *P. hysterophorus*

Pre-emergence control

There was no seed germination in pots treated with pretilachlor, pendimethalin, bensulfuron methyl + pretilachlor, bensulfuron methyl + acetachlor, oxadiazon, and Pyrazosulfuron-ethyl + pretilachlor in both puddle sown and dry sown conditions (Table 2). These pre-emergence herbicides were thus 100 percent efficient in controlling all four types of *P. hysterophorus*. However, seedling emergence was observed in pots treated with butachlor in dry sown condition, whereas, in puddle condition, no seedling emergence was found. In the case of pyrazosulfuron-ethyl, seedling emergence was observed in both conditions. Butachlor is recommended to be applied in stagnant water, and pyrazosulfuron-ethyl is recommended to be used at 6 – 9 days after sowing, which could be the reason for their poor performance in the present study where they were applied in dry conditions and 3 – 6 days earlier, respectively. However, plants grown in dry sown conditions were more susceptible to pyrazosulfuron-ethyl than those in wet sown conditions.

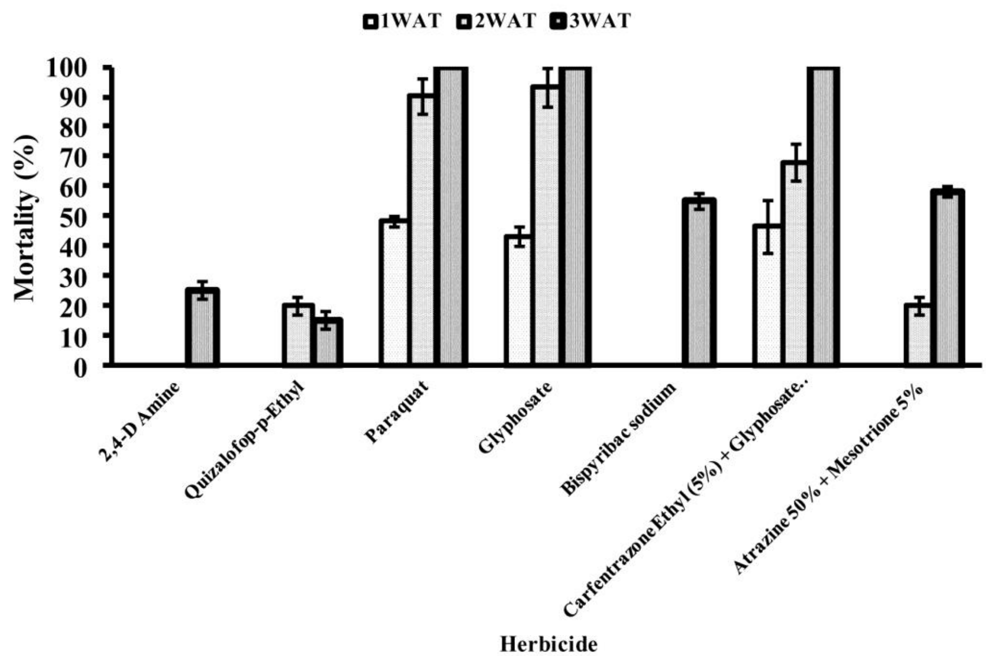
Table 2. Efficacy of butachlor and pyrazosulfuron ethyl on *P. hysterophorus*.

Treatments	Dry sown		Puddle sown	
	Emergence (%)	WCE (%)	Emergence (%)	WCE (%)
butachlor	8.0	87.29	-	-
Control	61.33		-	-
pyrazosulfuron ethyl	22.67	71.99	21.33	66.75
Control	60.0		66.67	

Post-emergence control at rosette stage

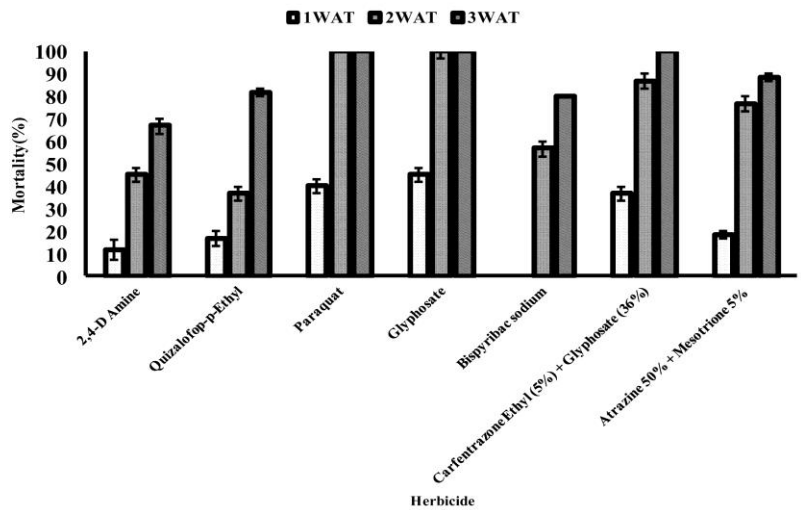
The statistical analysis of the data showed that different herbicidal treatments significantly affected parthenium weed mortality in non-cropped conditions. The ½ dose of herbicidal treatments provided 0% mortality at 1 WAT, 3.3 to 25% mortality at 2 WAT, and 39 to 61% mortality at 3 WAT (Fig. 1). The results exhibited that maximum weed mortality (61%) at 3 WAT was recorded in glyphosate, followed by paraquat-treated plants scoring 45% mortality. The ¾ dose herbicidal treatments gave 0 to 48.33% mortality at 1 WAT, 0 to 93% mortality at 2 WAT, and 15 to 100% mortality at 3 WAT (Fig. 2). The results exhibited that maximum weed mortality of 48.33% at 1 WAT was recorded in paraquat which was followed by carfentrazone ethyl (5%) + glyphosate (36%) treated plants scoring 46.67% mortality. However, glyphosate-treated plants gave maximum mortality (93%) at 2 WAT which was followed by paraquat (90%). At 3 WAT paraquat, glyphosate and carfentrazone ethyl

(5%) + glyphosate gave 100% mortality (Fig. 3). Among the full strength herbicidal treatments glyphosate, paraquat, and carfentrazone ethyl (5%) + glyphosate resulted in the maximum mortality (45, 40, and 36.67%, respectively) at 1 WAT. However, in the case of glyphosate and paraquat, it was increased to 100% in both 2 WAT and 3 WAT where carfentrazone ethyl (5%) + glyphosate resulted in 86.67 and 100% at 2 and 3 WAT, respectively. Parthenium sprayed with glyphosate, and carfentrazone ethyl (5%) + glyphosate started getting pale from the day after spraying, and complete mortality was achieved after one week with their lower doses. In contrast, the plant regenerated after the death of the top with the same doses of paraquat. Therefore, glyphosate and carfentrazone ethyl (5%) + glyphosate were the most effective as these treatments could able to completely controlled parthenium. These findings are in line with Shabbir (2014) and Khan et al. (2012). Krishna et al. (2007) affirmed that glyphosate provided greater than 93% control of parthenium weed at the rosette stage at 3 WAT.



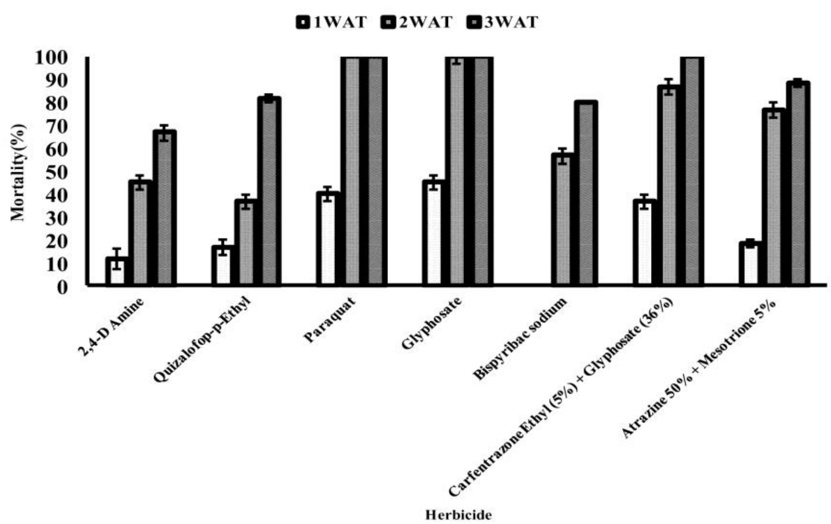
Vertical bars show standard errors of means of three replications.

Figure 1. Parthenium weed control at rosette stage with the half doses of different herbicidal application at 1, 2 and 3 WAT in non-cropped condition



Vertical bars show standard errors of means of three replication.

Figure 2. Parthenium weed control at rosette stage with the three-quarters (¾) doses of different herbicidal applications at 1, 2, and 3 WAT in non-cropped condition



Vertical bars show standard errors of means of three replication.

Figure 3. Parthenium weed control at rosette stage with full doses of different herbicidal applications at 1, 2, and 3 WAT in non-cropped condition

Post-emergence control at the bolted stage

Application of recommended dose of paraquat, glyphosate, and carfentrazone ethyl (5%) + glyphosate (36%) completely killed the bolted parthenium plants in 10, 8, and 12 days after treatment (DAT), respectively (Table 3). Generally, parthenium plants regenerate after a few days once their top is killed by the herbicide application (Mahaderappa, 1999). However, in contrast to that, there was no regeneration of the plants in glyphosate among the test herbicidal treatment (Fig. 4).

Table 3. No. of days required for the complete killing of parthenium by the application of different herbicides and percentage mortality.

Herbicide Common name	Trade Name	Dose (ha)	Days taken for complete death of Parthenium	Mortality (%)
2,4-D Amine	Fielder	2.80 L	18 ± 0.58	65
Quizalofop-p-Ethyl	Weednil 5 EC	650 ml	22 ± 1.2	76
Paraquat	Paraxone	2.80 L	10 ± 0.88	100
Glyphosate	Sun Up	3.70 L	8 ± 0.88	100
Bispyribac sodium	Xtra Power 20 WP	150 L	22 ± 1.2	75
Carfentrazone Ethyl (5%) + Glyphosate (36%)	Trigger 365EW	2.00 L	12 ± 0	100
Atrazine 50% + Mesotrione 5%	Triozine 55 SC	2.00 L	21 ± 1.33	86

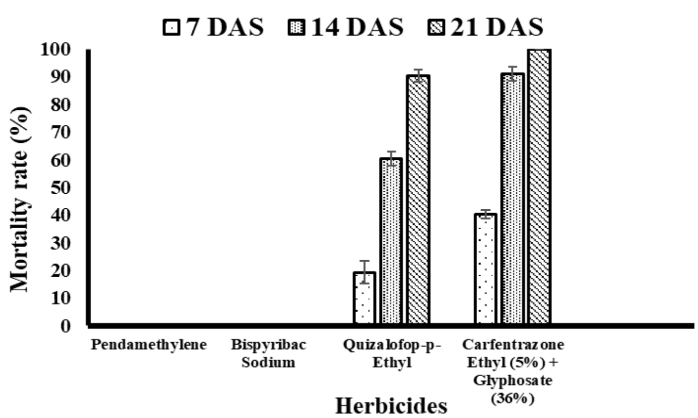
Figure 4. Effect of herbicides at bolted stage of parthenium weed.

Glyphosate is one of the most toxic herbicides, with many species of wild plants being damaged or killed by applications of less than ten micrograms plant^{-1}.

Glyphosate can be more damaging to wild flora than many other herbicides. Glyphosate kills plants by inhibiting the enzyme 5-enolpyruvoyl-shikimate-3-phosphate synthase, which forms the aromatic amino acids phenylalanine and tyrosine, and tryptophan. Paraquat is used to kill both broadleaf and grassy weeds. It is the second most widely used herbicide in the United States, after glyphosate. However, it is highly persistent in soil and has been classified as a Restricted Use Pesticide (RUP) due to its potential for groundwater contamination (Ware, 1986). Therefore, another effective herbicide, i.e., carfentrazone ethyl (5%) + glyphosate (36%), was done in the field study, along with another three herbicides, which were effective pre and post-application.

Parthenium weed control at field condition

Wilting and necrosis of leaves started after a few days of herbicides application, but mortality was only seen after 14 DAS irrespective of herbicides (Fig. 5). Carfentrazone Ethyl (5%) + Glyphosate (36%) resulted in 40 and 91% mortality at 7 and 14 DAS, respectively and it was increased to 100% at 21 DAS. Quizalofop-p-Ethyl resulted in 19 and 60 % mortality at 7 and 14 DAS, respectively however, it was increased to 90% at 21 DAS. No mortality was seen in another two herbicides. These findings indicate that Carfentrazone Ethyl (5%) + Glyphosate (36%) could provide effective control of parthenium in the crops for which they are registered. Shabbir (2014) concluded that glyphosate and isoproturon are effective in killing *P. hysterophorus* but glyphosate is comparatively more effective than isoproturon in field conditions.



Vertical bars show standard errors of means of three replication.

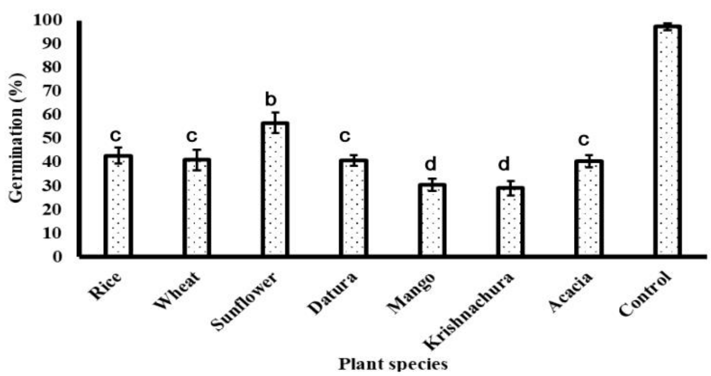
Figure 5. Effect of herbicides on mortality rate (%) of *P. hysterophorus* at 7, 14 and 21 days after spray (DAS) in the field.

In the present study, since the test herbicides were found effective even at a lower recommended dosage, their usage has comparatively fewer environmental risks.

Since the plant did not regrow when the top died, this also makes these herbicides' choice against parthenium more crucial. Thus in light of the results of the present study, it is recommended that parthenium should be managed by using the lowest doses of these herbicides, especially those which take comparatively less time to kill this alien weed species. Moreover, these results indicate that several herbicides registered for PRE and POST control of weeds in maize, cotton, peanut, rice, and soybean could provide effective control of parthenium. Under such circumstances, the herbicidal approach seems viable for controlling parthenium in Bangladesh.

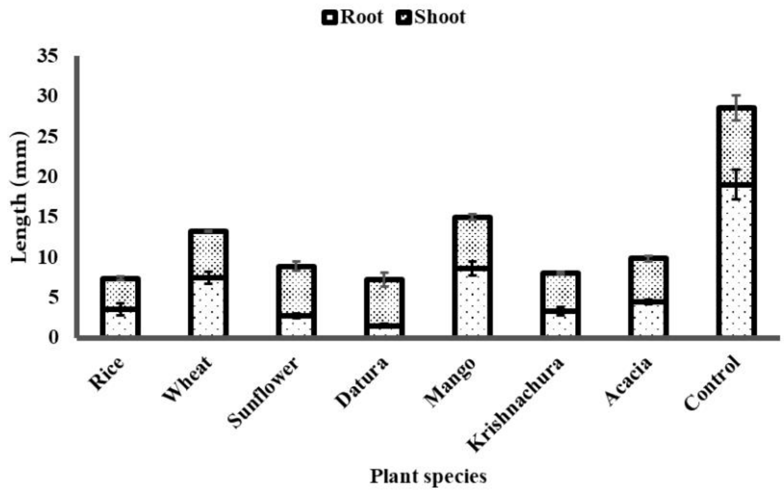
Effect of plants extracts on germination and seedling growth of parthenium weed

Aqueous extracts of all the test species showed markedly herbicidal activity against the germination of parthenium seeds (Fig. 6), where *Delonix regia* was found to be the most effective for the inhibition of germination (29%). On the other hand *Datura metel* extracts and *Oryza sativa* var. Boteswar straw extracts significantly reduced root (1.5 mm) and shoot (3.8 mm) length, respectively (Fig. 7). However, the highest reduction in fresh biomass (1.93 mg plant^{-1}) was observed due to the application *Magnifera indica* extracts (Fig.8). These results support the earlier findings that parthenium's germination and seedling growth can be checked by aqueous leaf extracts of the allelopathic tree and grass species (Safdar et al., 2013; Safdar et al., 2016; Ojija et al., 2019). Likewise, Shafique et al. (2005) found that aqueous leaf extracts of allelopathic trees, namely *Azadirachta indica* (L.) A. Juss., *Ficus bengalensis* L., *Melia azadarach* L., *Mangifera indica* L. and *Syzygium cumini* (L.) Skeels exhibit the herbicidal potential in controlling parthenium weed. Besides, (Javaid et al., 2011) reported that aqueous extracts of allelopathic crops such as rice, wheat, and dicotyledonous plants such as *Datura metel*, leaf, and bark extracts of *Alstonia scholaris* (L.) R. Br.significantly suppressed the germination and growth of parthenium weed.



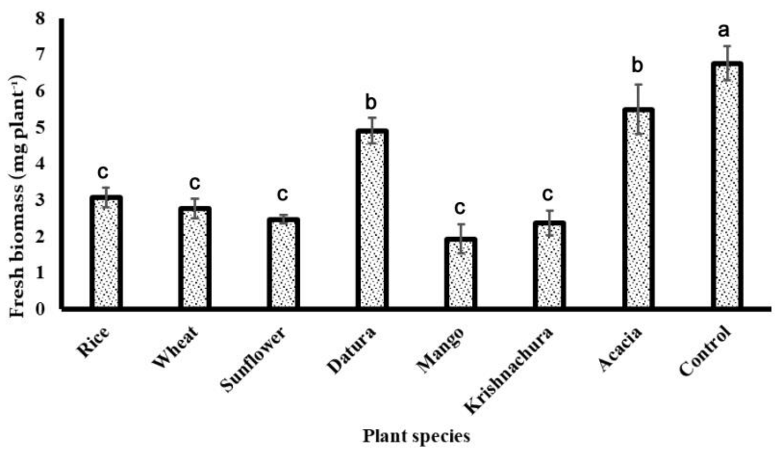
Vertical bars show standard errors of means of three replication. Values with different letters at their top show significant difference ($p \leq 0.05$) as determined by LSD test

Figure 6. Effect of aqueous extracts of different plant species on germination of *P. hysterophorus.*



Vertical bars show standard errors of means of three replication.

Figure 7. Effect of aqueous extracts of different plant species on seedling growth of *P. hysterophorus.*



Vertical bars show standard errors of means of three replications. Values with different letters at their top show significant difference ($p \leq 0.05$) as determined by LSD test

Figure 8. Effect of aqueous extracts of different plant species on fresh biomass of *P. hysterophorus.*

CONCLUSION

The results indicated that well-grown plants of parthenium weed could effectively be controlled with pretilachlor, pendimethalin, bensulfuron methyl + pretilachlor, bensulfuron methyl + acetachlor, oxadiazon, and pyrazosulfuron-ethyl + pretilachlor before emergence, and glyphosate, paraquat and carfentrazone ethyl (5%) + glyphosate (36%) at both rosette and bolted stage in non-cropped areas. Carfentrazone ethyl (5%) + glyphosate (36%) could be applied in the cropped area to overwhelm crop injury. We also recommend that for effective control of *P. hysterophorus* integrated control approaches, for example, *D. regia* and *D. metel* leaf bio-herbicides and *O. sativa* var. Boteswar are imperative. However, further field studies must be conducted to verify the safety of these leaves crude extracts for native plant and animal species before implementation. In addition to deploying control measures via a community approach, awareness of the effects and dispersal mechanisms of *P. hysterophorus* should be raised in local communities to prevent new invasions in other habitats.

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