

Congress grass possess herbicidal potential against weeds in wheat fields

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Abstract

An understanding about occurrence of weed species and patterns of change in incidence is dynamic in emerging weed managing approaches and give directions to future study endeavors. To appraise this threat, we studied the allelopathic effect of aqueous extracts of different parts of invasive weed congress grass, *Parthenium hysterophorus* L.) to biologically minimized weed risk in Wheat in 2012-13. Aqueous extracts of different plant parts (root, shoot, leaf, fruit and entire plant) of *Parthenium* were applied for three times as pre-emergence, post emergence and pre + post emergence. No significant grain yield differences were noticed between leaf extract and whole plant extract. Maximum weed density reduction (85.50%), weed dry biomass reduction (77.21%), weed control efficiency (85.67%), plant height (91.44 cm), biological yield (13426 kg ha⁻¹) and grain yield (4437 kg ha⁻¹) were found where leaf extract was applied as pre emergence spray. Whole plant extract sprayed as twice i.e. once as pre emergence and secondly as post emergence also gave better results showing weed density reduction (79.93%), weed dry biomass reduction (73.77%), weed control efficiency (80.09%), biological yield (12253 kg ha⁻¹) and grain yield (4414 kg ha⁻¹). On other hand 11.08% decrease in grain yield occurred where fruit extract was applied as post emergence spray. Therefore for better wheat grain yield and reduced weed risk farmers can use spray of leaf extract of parthenium before weed emergence.

Keywords: Aqueous extract, Bioherbicide, Weeds, Wheat, *Parthenium hysterophorus* L.

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Introduction

The increased production and land diversification

would meet future cereals demand. National strategies and market factors will determine food diversification (Gupta and Seth, 2007). Wheat though possess genetic



yield potential of 6-8 t ha⁻¹ but the actual average yield in Pakistan is about 2.7 t ha⁻¹ (Qamar et al., 2014). Wheat production (25.48 million tonnes against the last year's production of 25.98 million tonnes) during 2014-15 was 1.9% less than previous year (Anonymous, 2015). Delayed harvesting of kharif crops was the main reason for low production. Among the other factors were delayed wheat sowing, reduced fertilizer use, non-availability of certified seed, and weed loss (Ibrahim et al., 2013). Weeds are among serious plant pests (Zimdahl, 2013).

Weeds cycle can have three stages affected by climatic variables: germination-emergence, plant development and seed production (Fernandez-Quintanilla, 1988). Weeds negatively affect crop growth (Rajcan and Swanton, 2001). Yield loss by weeds vary among crops (Oerke, 2006). Plants have defense mechanisms and hormones which help in managing weeds (Pickett et al., 2014). Weed control diversification is necessitated by the limitations affiliated with the conventional weed control (Jabran and Farooq, 2013). During recent times, farmers use toxic chemicals for quick weed control (Khan et al., 2009; Tang et al., 2010). Agricultural systems use three million tons of herbicides annually indicating weed infestation severity (Stephenson, 2000). Herbicide application can lead to herbicide resistance. As the microbial activity and organic matter decay is more concentrated in upper soil layer hence the nitrate is the only inorganic ion found in higher concentration in soil solution affecting germination of many weed species (Espeby, 1989; Adkins et al., 1984).

Limiting weeds through allelopathic interaction is a novel biological way (Zeng, 2014). For keeping the environment healthy, natural methods for weed control are strongly suggested (Mubeen et al., 2012). Allelochemicals with plants origin can potentially be used to formulate new bio-herbicides (Duke et al., 2002). The allelopathic interaction may be between two crops, a crop and weed/s or between two weeds or vice-versa (Rice, 1984). There are studies on exploring the new allelochemicals and formulating natural herbicides. Parthenium (congress grass) is an annual herbaceous harmful weed belonging to Asteraceae family (Jalata, 2009). *High conceptive potential, quick development rate, obstruction by allelopathy and versatile nature causing wild spread of this weed (Singh et al., 2005). It impedes nodulation of legumes as the inhibitory effect of allelochemicals on nitrifying and nitrogen fixing bacteria (Dayama, 1986). Since*

Parthenium is widespread in the world, *exploring the herbicidal uses if any could be important.*

Researchers have studied the synergistic impacts of different mixtures of allelopathic water extracts alongside weeds in various crops (Elahi et al., 2011; Awan et al., 2012). The objective of the research was to investigate suitable plant part extract of Parthenium to be used as pre or post emergence to reduce weeds infestation in wheat fields.

Material and Methods

Field investigations into the allelopathic efficacy of water extracts of Parthenium on other weeds and yield of wheat were carried out at the University of Poonch, Rawalakot AJ&K during 2013-14. Mature plants of *Parthenium hysterophorus* were harvested from different places including roadsides, barren lands and cropped lands around the Rawalakot city. Roots, shoots, leaves and fruits were distinctly separated by using scissors and dried under shade for twenty five days. The fully dried roots, shoots, leaves, fruits and whole plants were soaked in distilled water at 150 g L⁻¹ (15%) at room temperature (21 ± 3°C) for 48 hours. These soaked plant parts were filtered by using two layers of muslin cloth to obtain their aqueous extracts separately. Water extracts were separately bottled and tagging was done for future use in the experiments. Experiment was laid out using Randomize Complete Block Design (RCBD) under factorial arrangement with net plot size of 1m×3m replicated three times. Wheat cultivar Lasani-2008 was used as a test crop with seed rate of 100 Kg ha⁻¹. Wheat sowing was done in October, 2012 using single row hand drill on raised beds for provision of drainage to melted snow water and to provide sufficient aeration to wheat tillers to avoid growth of fungal spores. All other agronomic practices were applied as per standard recommendations. The first pre-emergence spray of extracts was applied after 3 days of sowing while the second spray was done after 30 days of sowing using hand held knapsack sprayer. Weed samples were taken after 60 days of sowing from each treatment to record their density and dry weight (g) by using electric balance. Taken 5g from each weed sample and placed in oven at 60°C till constant weight. Data on density and dry weight of weeds were used to calculate efficiency indices by formulae as under (Misra and Misra, 1997)



$$\text{Weed Control Efficiency} = \frac{W_c - W_t}{W_c} \times 100$$

$$\text{Weed Persistence Index} = \left(\frac{W_c}{W_t}\right) \times \left(\frac{DM_c}{DM_t}\right)$$

Here (Wc) is weed density of un-treated plot, (DMc) is treated plot weed dry matter, (Wt) is treated plot weed density and (DMt) is weed dry matter of control. At wheat crop maturity (during the 3rd week of June) recorded the data on plant height (cm), spike length (cm), number of tillers per plant, productive tillers per plant, number of spikelets per spike, 1000 grain weight, grain yield and harvest index. The data means were then accordingly subjected to analysis of variance (ANOVA) individually and means were separated using Tukey’s test to identify significant differences.

Results and Discussion

Spray of Parthenium aqueous extracts pre emergence sole bring significant differences in weeds density of wheat but the post emergence application revealed significant reduction in weeds density. Generally, aqueous extracts of leaf applied as pre emergence treatments provided satisfactory control of broad leaved weeds present in the field as compared with other treatments (Table 1). Highest reduction (85.50%) in weed density was achieved where leaf extract was applied as pre emergence followed by 79.93% reduction where whole plant extract was applied twice i.e. pre + post emergence. More the weeds, more is the allelochemicals depletion from the soil and the more is their competition with crop plants. However, the minimum reduction (9.96%) in weed density was observed in plots where fruit extract was sprayed at the time of post emergence. Cheema et al., 2002 and Iqbal et al., 2010 supported the idea of application of allelopathic plant water extract, which reduce 64-85% weed density of *Convolvulus arvensis* L. as compared to control. These results showed that leaf extract applied as pre emergence is most effective for weed control whereas fruit extract applied as post emergence is less effective.

Mean value (77.21%) of dry biomass reduction in tested variety showed that the highest value noted for leaf extract applied as pre emergence, followed by 73.77% reduction where whole plant extract applied as pre + post emergence. Biomass reduction (66.36%) noted for leaf extract applied as combined pre + post

emergence in treated plot (Table 1).

Table-1: Effect of foliar spray of water extracts of congress grass at different stages on percent weed population and biomass reduction 60 DAS and increase in wheat yield.

Treatments	Weed density reduction (%)	Weed biomass reduction (%)	Grain yield Increase Over Control (%)
T1: Root extract (Pre-emergence)	67.95 bc	62.42 ab	57.96
T2: Root extract (Post-emergence)	45.34 e	33.07 d	49.85
T3: Root extract (Pre + Post emergence)	53.80 cde	40.31 cd	51.67
T4: Shoot extract (Pre-emergence)	64.54 bcd	56.87 bc	56.71
T5: Shoot extract (Post-emergence)	44.92 e	31.80 d	54.59
T6: Shoot extract (Pre + Post emergence)	45.07 e	29.21 d	50.12
T7: Leaf extract (Pre-emergence)	85.50 a	77.22 a	60.77
T8: Leaf extract (Post-emergence)	77.04 ab	66.37 ab	58.89
T9: Leaf extract (Pre + Post-emergence)	15.65 f	3.97 e	24.62
T10: Fruit extract (Pre-emergence)	53.76 cde	37.82 d	54.99
T11: Fruit extract (Post-emergence)	9.96 fg	1.74 e	11.09
T12: Fruit extract (Pre + Post emergence)	52.10 de	29.60 d	49.34
T13: Whole plant extract (Pre-emergence)	52.57 cde	37.01 d	53.3
T14: Whole plant extract (Post-emergence)	21.25 f	7.96 e	34.06
T15: Whole plant extract (Pre + Post emergence)	79.94 ab	73.78 ab	60.57
T16: Control	0 g	0 e	0

The lowest reduction (1.74%) in dry biomass was recorded in plots where fruit extract was applied as post emergence followed by 3.96% reduction in the treatment where leaf extract was applied combine i.e. pre + post emergence. The results are quite in consonance with other scientists who testified that allelopathic plants water extracts suppressed total dry weight of weeds in wheat crop (Sharif et al., 2005; Bhatti et al., 2006).



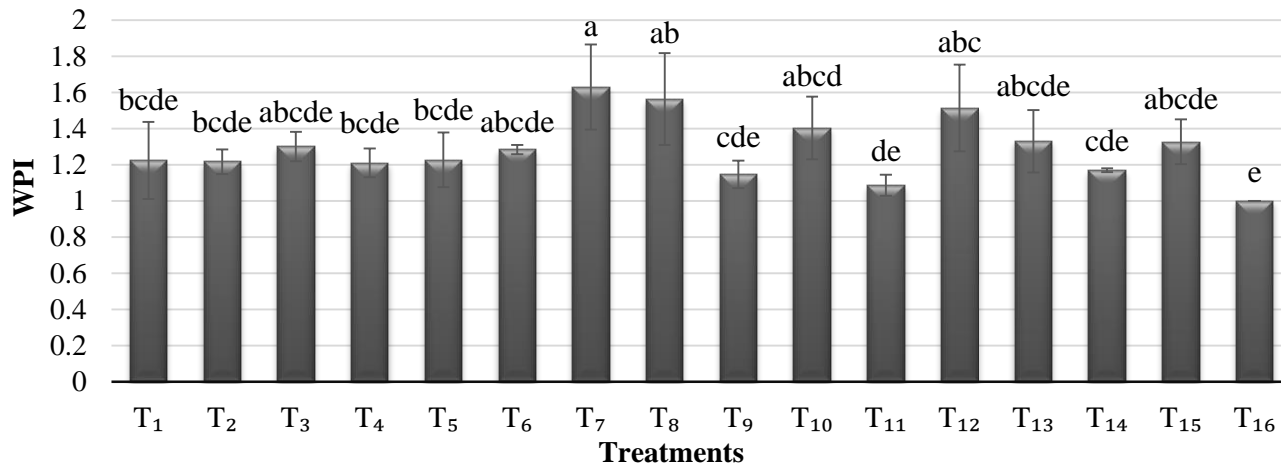


Figure-1: Effect of Parthenium aqueous extracts and time of application on weed persistence index.

(T₁: Root extract (Pre-emergence); T₂: Root extract (Post-emergence); T₃: Root extract (Pre + Post emergence); T₄: Shoot extract (Pre-emergence); T₅: Shoot extract (Post-emergence); T₆: Shoot extract (Pre + Post emergence); T₇: Leaf extract (Pre-emergence); T₈: Leaf extract (Post-emergence); T₉: Leaf extract (Pre + Post-emergence); T₁₀: Fruit extract (Pre-emergence); T₁₁: Fruit extract (Post-emergence); T₁₂: Fruit extract (Pre + Post emergence); T₁₃: Whole plant extract (Pre-emergence); T₁₄: Whole plant extract (Post-emergence); T₁₅: Whole plant extract (Pre + Post emergence) and T₁₆: Control)

Weed persistence determines the tolerance of weeds to aqueous extracts of Parthenium treatments as well as their effectiveness to eliminate the weeds (Figure 1). Cooperative effect of aqueous extracts of Parthenium with wheat plants was important for Weed Persistence Index. Maximum value (1.63) of WPI was recorded in leaf extract applied as pre emergence followed by leaf extract (1.56) applied as post emergence and fruit extract (1.51) sprayed combine i.e. Pre + Post emergence. On other hand minimum value of WPI was found in weedy check plot (1). These findings are similar to those obtained by Khaliq et al., 2014 who stated that application of various plant extracts resulted in relatively higher WPI than rest of the wheat cultivars.

Weed control efficiency (WCE) was suggestively affected by different aqueous extracts of Parthenium treatments used in this experiment (Figure 2). It reflects the effectiveness of applied Parthenium extracts in securing yield loss against weed competition and a lower value of weed index mean high efficiency. Pre emergence application of fruit extract recorded maximum (85.67%) WCE than rest of the treatments. Whole plant (80.19%) applied as pre + post emergence and leaf extract (77.19%) used as post emergence also showed highest values respectively. On other hand lowest WCE value was found in control

plot where no extract was applied. It is evident in the study that the fruit extract (pre emergence) was becoming more operative for controlling weed at inferior time of that extract.

Allelochemicals of *P. hysterophorus* have stronger effects on growth as compared to seed germination (Batish et al., 2002). Allelochemicals released by *P. hysterophorus* through aqueous extracts demonstrated to be adequate to provide significant phytotoxicity and the comparative role of allelopathic plants. Therefore, present results offer indications that the allelochemicals released has the capability to play a primary role. If such interference of *P. hysterophorus* with plants in the vicinity exists, it will be directed by numerous manipulating aspects. These findings also relate with Dawar et al., 2010 and Regina et al., 2007, who indicated that the perceived variability of allelochemicals content in *D. alba* and *P. hysterophorus* extracts recommends a main role limited to conditions where allelochemicals leading the range of inhibitors released from these plants. Ahn et al., 2005 exhibited that the releasing rate of allelochemicals from allelopathic plants is relative to the total quantity extant within the alleged plants and one could assume that allelochemicals will be the influential aspect for allelopathy only if high stages are accrued in the plants.

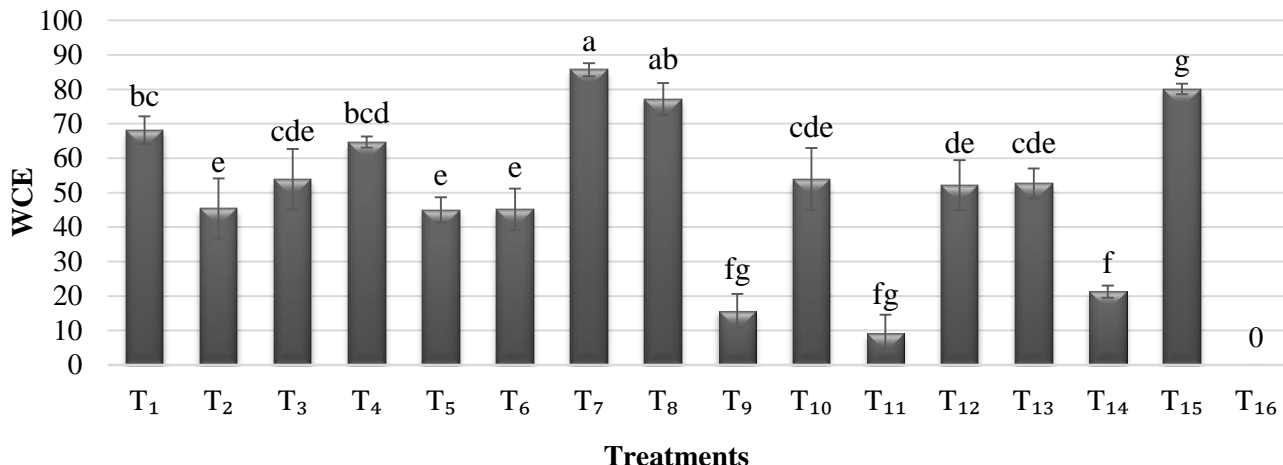


Figure-2: Effect of Parthenium aqueous extracts and time of application on weed control efficiency. (T₁: Root extract (Pre-emergence); T₂: Root extract (Post-emergence); T₃: Root extract (Pre + Post emergence); T₄: Shoot extract (Pre-emergence); T₅: Shoot extract (Post-emergence); T₆: Shoot extract (Pre + Post emergence); T₇: Leaf extract (Pre-emergence); T₈: Leaf extract (Post-emergence); T₉: Leaf extract (Pre + Post-emergence); T₁₀: Fruit extract (Pre-emergence); T₁₁: Fruit extract (Post-emergence); T₁₂: Fruit extract (Pre + Post emergence); T₁₃: Whole plant extract (Pre-emergence); T₁₄: Whole plant extract (Post-emergence); T₁₅: Whole plant extract (Pre + Post emergence) and T₁₆: Control)

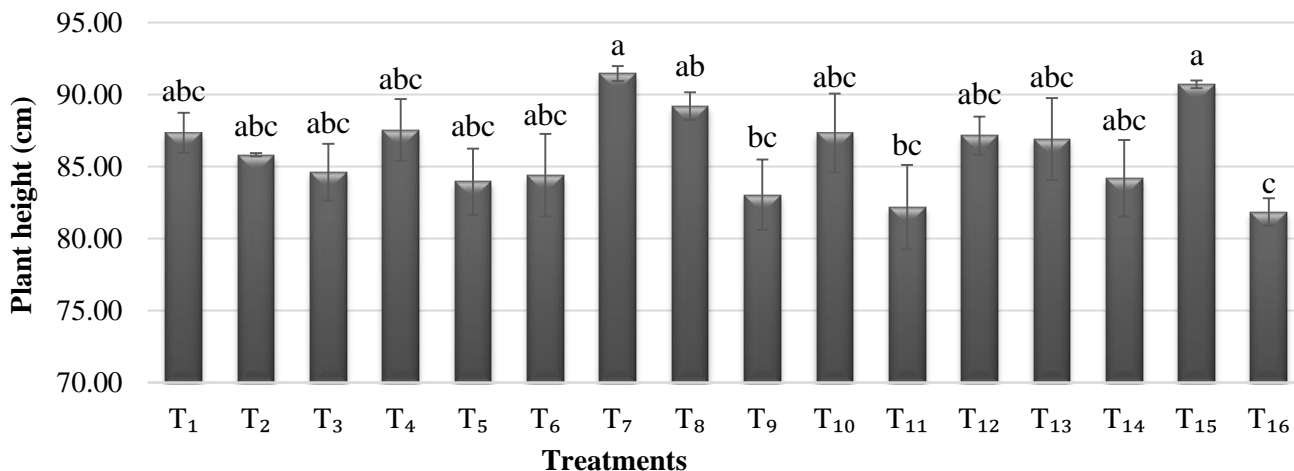


Figure-3: Effect of Parthenium aqueous extracts and time of application on plant height of wheat. (T₁: Root extract (Pre-emergence); T₂: Root extract (Post-emergence); T₃: Root extract (Pre + Post emergence); T₄: Shoot extract (Pre-emergence); T₅: Shoot extract (Post-emergence); T₆: Shoot extract (Pre + Post emergence); T₇: Leaf extract (Pre-emergence); T₈: Leaf extract (Post-emergence); T₉: Leaf extract (Pre + Post-emergence); T₁₀: Fruit extract (Pre-emergence); T₁₁: Fruit extract (Post-emergence); T₁₂: Fruit extract (Pre + Post emergence); T₁₃: Whole plant extract (Pre-emergence); T₁₄: Whole plant extract (Post-emergence); T₁₅: Whole plant extract (Pre + Post emergence) and T₁₆: Control)

Statistical data for aqueous extracts of Parthenium exhibited that different plant parts extracts had significant effect on the plant height of the tested specie (Figure 3). Mean values of the tested wheat cultivar showed that the maximum plant height (91.44 cm) was measured for fruit extract applied as pre emergence followed (90.71 cm) by whole plant extract applied two times (pre + post) and leaf extract (89.2 cm) applied as

post emergence. The minimum mean plant height 81.83 cm was recorded in the control treatments followed by fruit extract (82.16 cm) applied as post emergence spray and leaf extract (82.97 cm) applied as (pre + post) respectively.

All the plant extracts significantly reduced both spike length and awn length as compared with the control treatment (Figure 4). Spike length with the highest mean

values (10.78 cm) was recorded in plots where leaf extract was applied as pre emergence spray, whole plant extracts (9.81 cm) applied as combined (pre + post emergence) and leaf extracts (9.07 cm) applied as post emergence spray, respectively. While the minimum value (5.79 cm) was noted for control plot, (7.18 cm) for fruit extracts applied as post emergence and (7.46 cm) for fruit extracts applied as pre emergence. Similarly maximum awn length (4.98 cm) found in plots where leaf extract was applied at the time of pre emergence followed by (4.24 cm) for whole plant extract applied as combined (pre + post emergence) and 4.03 cm for leaf extracts sprayed as post emergence (Figure 4 and 5). The increase in spike length may be due to suppression of vegetative growth of weed (Majeed et al., 2012) due to spray of parthenium leaf extracts.

Comparison of the treatment means (Figure 6) depicted that the highest number of fertile tillers per plant (12.62) were in the plots where leaf extract was sprayed as pre emergence followed by (12.26) whole plant extracts applied as combined (pre & post emergence) and (11.40) for leaf extracts applied as post emergence spray. Treatments showing escalation in the number of fertile tillers may be due to relatively better weed control which eventually enabled relatively further translocation of photosynthates concerning reproductive growth because of less competition of weeds with wheat crop. Data existing in Figure 7 showed that significantly higher thousand grain weight (45.38 g) was found in treatment where leaf extract was applied as pre emergence followed by whole plant extract (43.61 g) applied as both

pre + post emergence. On the other hand, minimum thousand grain weight (27.83g) was found in weedy check (control) plot.

Data revealed that biological and grain yields of wheat did not contrast significantly among numerous treatments (Figure 8 and 9). Though, both were significantly affected by spray of Parthenium aqueous extracts. Maximum biological yield (13426 kg ha⁻¹) and grain yield (4437 kg ha⁻¹) were recorded in leaf extract sprayed as pre emergence followed by whole plant extract (4414.8 kg ha⁻¹) and (4253 kg ha⁻¹) treatment applied as both pre + post emergence. The higher yield may be due to favorable temperature, higher rainfall and effective weed control. While control treatment showed the lowest grain yield (1740.7 kg ha⁻¹) and biological yield (7266 kg ha⁻¹). Fruit extract showed grain yield (1957.8 kg ha⁻¹) and biological yield (7953 kg ha⁻¹) applied as post emergence spray.

This study indicated that increased grain yield with aqueous extract of Parthenium may be attributed to more fertile tillers, number of spikelets because of the suppressive allelopathic effect of leaf extract of Parthenium on weed density, which ultimately favors the higher grain yield (Cheema et al., 2002).

Our results showed resemblance with the results reported by Fujii et al., 2003 and Dawar et al., 2010 that water extract of *D. alba* and *W. somnifera* influenced some bioactive compounds which considerably reserved the growth of shoot and root of *R. crispus*, highly competitive weed commonly found in wheat fields.

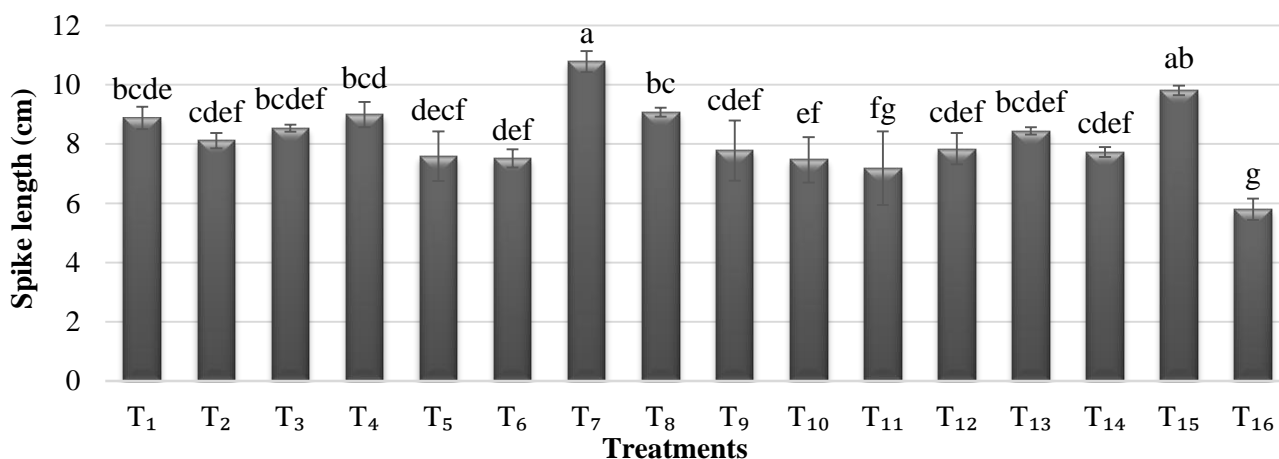


Figure-4: Effect of Parthenium aqueous extracts and time of application on spike length of wheat

(T₁: Root extract (Pre-emergence); T₂: Root extract (Post-emergence); T₃: Root extract (Pre + Post emergence); T₄: Shoot extract (Pre-emergence); T₅: Shoot extract (Post-emergence); T₆: Shoot extract (Pre + Post emergence); T₇: Leaf extract (Pre-emergence); T₈: Leaf extract (Post-emergence); T₉: Leaf extract (Pre + Post-emergence); T₁₀: Fruit extract (Pre-emergence); T₁₁: Fruit extract (Post-emergence); T₁₂: Fruit extract (Pre + Post emergence); T₁₃: Whole plant extract (Pre-emergence); T₁₄: Whole plant extract (Post-emergence); T₁₅: Whole plant extract (Pre + Post emergence) and T₁₆: Control)



Meihua et al., 2006 observed that allelochemicals condensed the chlorophyll content in plants. The reduced differences between various treatments could be due to the genetic information contained in the cultivar with no marked effect of extracts and the time of extract application on cell division thereby no significant increase or decrease in height of wheat plants. The existence of allelochemicals has been discovered and many researchers give opinion that allelopathins are extant in numerous parts of the plants and can

significantly affect the plants which receive them, in many ways. Sesquiterpene lactone parthenin were the most common inhibitors in *P. hysterophorus* found at high level in Capitulate Sessile Trichomes on stem, leaves and the achene complex (Reinhardt et al., 2004). Results showed that presence of allelopathic weeds in the agricultural fields and allelochemicals may provide opportunity for use in weed management (Cheema and Khaliq, 2000).

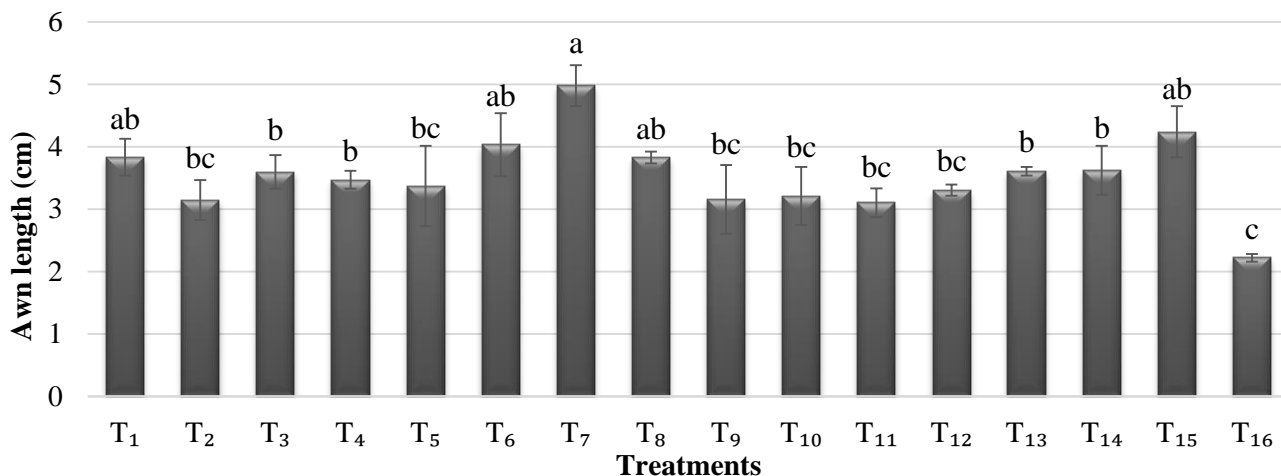


Figure-5: Effect of Parthenium aqueous extracts and time of application on awn length of wheat.

(T1: Root extract (Pre-emergence); T2: Root extract (Post-emergence); T3: Root extract (Pre + Post emergence); T4: Shoot extract (Pre-emergence); T5: Shoot extract (Post-emergence); T6: Shoot extract (Pre + Post emergence); T7: Leaf extract (Pre-emergence); T8: Leaf extract (Post-emergence); T9: Leaf extract (Pre + Post-emergence); T10: Fruit extract (Pre-emergence); T11: Fruit extract (Post-emergence); T12: Fruit extract (Pre + Post emergence); T13: Whole plant extract (Pre-emergence); T14: Whole plant extract (Post-emergence); T15: Whole plant extract (Pre + Post emergence) and T16: Control)

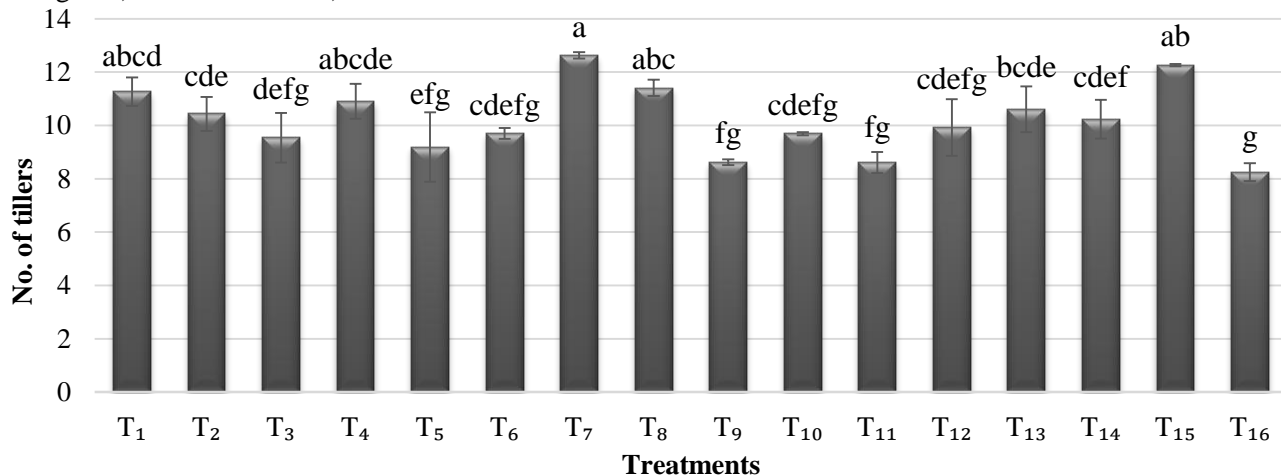


Figure-6: Effect of Parthenium aqueous extracts and time of application on number of tillers of wheat.

(T1: Root extract (Pre-emergence); T2: Root extract (Post-emergence); T3: Root extract (Pre + Post emergence); T4: Shoot extract (Pre-emergence); T5: Shoot extract (Post-emergence); T6: Shoot extract (Pre + Post emergence); T7: Leaf extract (Pre-emergence); T8: Leaf extract (Post-emergence); T9: Leaf extract (Pre + Post-emergence); T10: Fruit extract (Pre-emergence); T11: Fruit extract (Post-emergence); T12: Fruit extract (Pre + Post emergence);



T₁₃: Whole plant extract (Pre-emergence); T₁₄: Whole plant extract (Post-emergence); T₁₅: Whole plant extract (Pre + Post emergence) and T₁₆: Control)

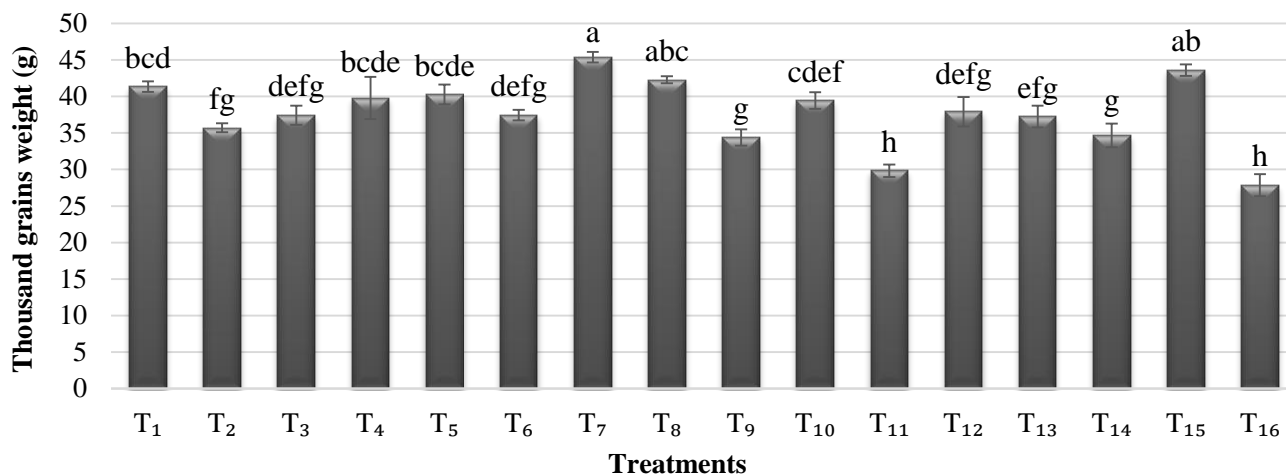


Figure-7: Effect of Parthenium aqueous extracts and time of application on 1000-grain weight of wheat.

(T₁: Root extract (Pre-emergence); T₂: Root extract (Post-emergence); T₃: Root extract (Pre + Post emergence); T₄: Shoot extract (Pre-emergence); T₅: Shoot extract (Post-emergence); T₆: Shoot extract (Pre + Post emergence); T₇: Leaf extract (Pre-emergence); T₈: Leaf extract (Post-emergence); T₉: Leaf extract (Pre + Post-emergence); T₁₀: Fruit extract (Pre-emergence); T₁₁: Fruit extract (Post-emergence); T₁₂: Fruit extract (Pre + Post emergence); T₁₃: Whole plant extract (Pre-emergence); T₁₄: Whole plant extract (Post-emergence); T₁₅: Whole plant extract (Pre + Post emergence) and T₁₆: Control)

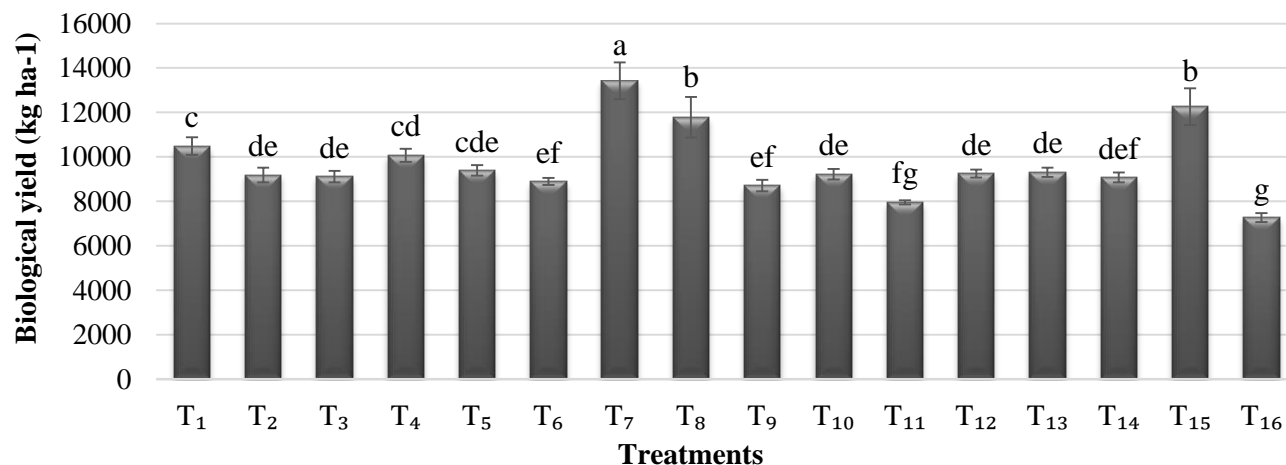


Figure-8: Effect of Parthenium aqueous extracts and time of application on biological yield of wheat.

(T₁: Root extract (Pre-emergence); T₂: Root extract (Post-emergence); T₃: Root extract (Pre + Post emergence); T₄: Shoot extract (Pre-emergence); T₅: Shoot extract (Post-emergence); T₆: Shoot extract (Pre + Post emergence); T₇: Leaf extract (Pre-emergence); T₈: Leaf extract (Post-emergence); T₉: Leaf extract (Pre + Post-emergence); T₁₀: Fruit extract (Pre-emergence); T₁₁: Fruit extract (Post-emergence); T₁₂: Fruit extract (Pre + Post emergence); T₁₃: Whole plant extract (Pre-emergence); T₁₄: Whole plant extract (Post-emergence); T₁₅: Whole plant extract (Pre + Post emergence) and T₁₆: Control)

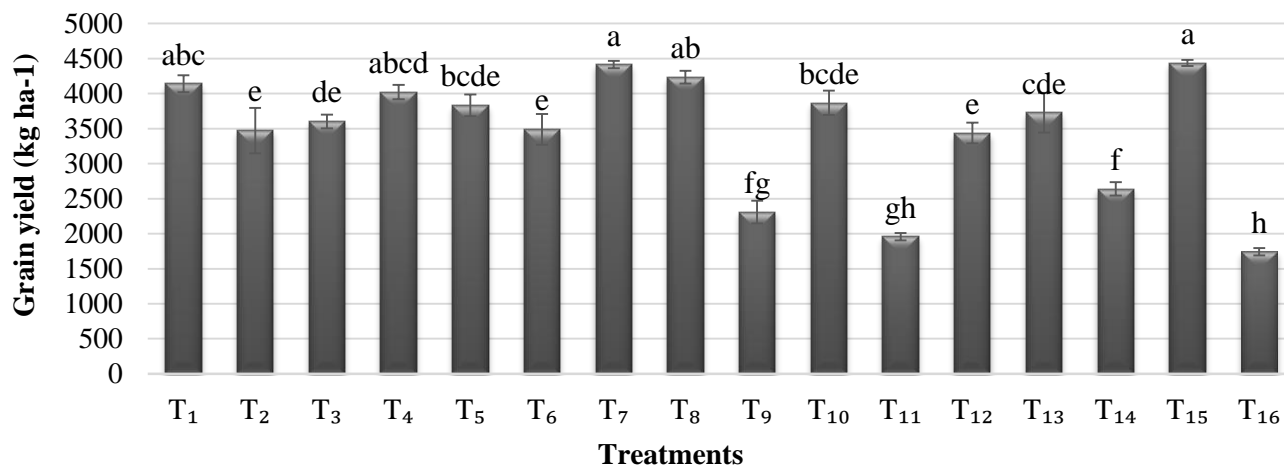


Figure-9: Effect of Parthenium aqueous extracts and time of application on grain yield of wheat.

(T₁: Root extract (Pre-emergence); T₂: Root extract (Post-emergence); T₃: Root extract (Pre + Post emergence); T₄: Shoot extract (Pre-emergence); T₅: Shoot extract (Post-emergence); T₆: Shoot extract (Pre + Post emergence); T₇: Leaf extract (Pre-emergence); T₈: Leaf extract (Post-emergence); T₉: Leaf extract (Pre + Post-emergence); T₁₀: Fruit extract (Pre-emergence); T₁₁: Fruit extract (Post-emergence); T₁₂: Fruit extract (Pre + Post emergence); T₁₃: Whole plant extract (Pre-emergence); T₁₄: Whole plant extract (Post-emergence); T₁₅: Whole plant extract (Pre + Post emergence) and T₁₆: Control)

Conclusion

The results of this study showed that the allelochemicals in leaf extracts applied as pre emergence significantly inhibited growth of weeds by decreasing their density, dry biomass, efficiency percentage and improved wheat yield and yield components. Therefore, it is suggested that leaf extracts of *P. hysterophorus* can be explored further as pre emergence spray. The whole plant extract can also be sprayed as pre and post emergence to downsizing weeds population, weed biomass and to boost grain yield of wheat. However, these water extracts needs to be tested further in different climatic and edaphic situations for adoption by the farmers for effective and safe weed control. We believe this research will lay down the foundation for promotion of biological weed control in wheat.

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Aziz M: Helped in statistical analysis
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