PHYTOTOXIC EFFECTS OF *Sonchus oleraceus* ON EMERGENCE AND SEEDLING GROWTH OF *Echinocloa cruss-galli*

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ABSTRACT

The study was aimed to explore the phytotoxic potential of Sonchus oleraceus L. on the emergence and seedling growth of *Echinocloa cruss-galli* L. and quantification of allelochemicals present in leaves, stem and fruit of S. oleraceus weed. The experiment comprised of aqueous extracts of leaf, roots and fruits applied at 0.25, 0.5, 1, 2, 4 and 8% (w/v) along with a control (0%). Data regarding seed emergence, emergence index, emergence percentage, root length, shoot length and seedling dry weight were recorded. All the tested concentrations of all the parts of S. oleraceus significantly inhibited the seed emergence, emergence index, emergence percentage (%), growth and delayed mean emergence time and time taken to 50% emergence of *E. cruss-galli*. However, maximum mean emergence time (5.20 days) and time taken to 50% emergence (3.49 days), were noted at 8% concentration of fruit and leaf aqueous extracts, respectively. Fruit aqueous extract at 8% concentration caused the lowest germination index (1.70), germination percentage (40%), root length (0.09 cm), shoot length (0.45 cm) and dry weight (0.16 g) of *E. cruss-galli*. Results suggested that the extract of S. oleraceus weed at concentration (8%) can be used as potential bio-herbicide for the control of *E. cruss-galli* weed.

Keywords: Allelopathy, concentration, germination, phytotoxic, *Sonchus oleraceus*

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INTRODUCTION

Weeds are unwanted plants in crops that compete with crops for resources such as carbon dioxide, water, light, nutrients and space and also create biotic hazards by releasing certain chemicals which result in low crop production (Gallandt and Weiner, 2015). The use of herbicides in controlling weeds is one of the major weed control strategies and is considered as the time saving, most effective and economical method to control the targeted weeds (Kraehmer*et al.,* 2014). However, the excessive use of herbicides in weed management causes serious threat to both environment and public health (Asghari and Tewari*,* 2007). Furthermore, increasing herbicide resistance in weeds have made chemical weed control an unsuitable method to sustain crop production (Abbas *et al.,* 2016a), allelopathy is quite effective to control resistant weeds (Abbas *et al.,* 2016b). Therefore, there is necessity to develop alternate weed control methods which are less or no dependent on chemical herbicides and less toxic to environment (Singh *et al.,*2011). Weed crop competition is one of the major biotic constraints in the production of rice. In rice 12% reduction in the yield is due to the weeds (Singh *et al.,* 2011). Weed crop competition is more in early growth stages and un-control of weeds during early period of crop result in poor growth of crops and decrease in grain yield (Fahad *et al.,* 2015). In Pakistan, an average of 20-30% decrease in yield of different crops due to weed crop competition. In cereal crops like wheat, rice, maize and gram, the reduction in the yield due to the weed infestation is 30, 40, 4 and 5 billion rupees respectively (Razzaq *et al.,* 2010). Therefore, it is important to control weeds to decrease the weed crop competition and improve the yield (Macias *et al.,* 2002). Biological control of weeds is an important method to control weeds and is also environment friendly and cheap (Abbas*et al.,* 2016b). Grassy and broad-leaf plant shave the potential to suppress weeds (Javaid*et al.,*2005; Nadeem *et al.,* 2020b). Annual sow thistle (*S. oleraceus* L.) is an annual widespread deleterious winter weed of Pakistan. It is one of the sophisticated weed species found in diverse places all over the world (Hutchinson and Lewin 1984). Sow thistle plants are upright and fleshy with slender and smooth stems oozes milky latex when broken. Plant can grow from 20 cm to 150 cm while the growth pattern may vary from straight to rosette. Numerous weed species have been reported to be allelopathic (Qasem and Foy, 2001). The leaves of *Sonchus oleraceus* contain several bioactive compounds including flavonoids, tannins, phenols, alkaloids and steroids (Yin *et al.,* 2007; Singh, 2010; Banaras *et al.,* 2020). The aqueous extracts of sow thistle showed an inhibitory effect on the germination and seedling growth of radish (Alaib *et al.,* 2017).

Rice is an important cereal crop in the Pakistan, and barnyard grass (*E.cruss-galli* L.) is the principal weed of rice in this region. The competitiveness, photoperiodic insensitivity, C_4 pathway, and higher seed production capacity make the barn yard grass a problematic weed (Bagavathiannan *et al.,* 2014). Barnyardgrass typically causes 30% reduction in yield of rice crop and may lead to complete crop failure under severe infestations (Bagavathiannan*et al.,*2012). The chemical weed control is common practice for control of barnyard grass but is criticized due to environmental issues and development of herbicide resistance (Pieterse, 2010). Allelopathic potential of lot of weeds had been reported by earlier workers. It is therefore, extremely encouraging to investigate the phytotoxic potential of weed on obnoxious weeds like *E. cruss-galli*so that plants having ability to serve as source of natural herbicides may be exploited to control the growth of weeds. The present study has therefore been planned to investigate the phytocidal, phytostatic or phytoregulatory potential of *S. oleraceus* against *E. cruss-galli (*barnyard grass*)*, the one of the worst crops and environmental weeds of rice and quantification of allelochemicals in different parts of *S. oleraceus*.

MATERIALS AND METHODS

The study was carried out in summer 2017 at Weed Science

Laboratory, Department of Agronomy, University of Agriculture Faisalabad (UAF), Pakistan to investigate the phytotoxic potential of aqueous extracts of different parts of *Sonchus oleraceus* L. (sow thistle) on *Echinochloacruss-galli* L. weed. The study was arranged in in a Completely Randomized Design with factorial arrangement having four replications.

Collection of *S. oleraceus* plant parts

To prepare aqueous extract of *S. oleraceus*, its plants were collected from weed Agronomic Farm, University of Agriculture Faisalabad. The plants of *S. oleraceus* were harvested above the ground surface at maturity and dried for two weeks at ambient temperature. After drying properly, different parts of plants were separated and chopped into pieces of about 2 cm.

Preparation of aqueous extracts of *S. oleraceus* weed parts

S. oleraceus aqueous extracts of various parts were made by adding 10 g of chopped dried plant material into 100 ml of distilled water in bottles separately at ratio of 1:10 w/v using procedure as described by Qasem (2013). At room temperature plant material were soaked in the water for at least 24 hours. These aqueous extracts were made from each desired part of *S. oleraceus* such as leaves, stem, fruit, flower etc. Then the material was passed through a cheese cotton cloth to attain the 10% water extracts of different parts of *S. oleraceus*which was used as stock solution. The stock solution diluted to prepare the required concentration of 0.25%, 0.5%, 1%, 2%, 4% and 8%.

Laboratory experiment

In laboratory experiment, the aqueous extracts of *S. oleraceus* weed parts (leaves, stem and fruits) in different concentrations viz.0.25%, 0.5%, 1%, 2%, 4%, and 8% were used to explore their phytotoxic effects on emergence and growth of *E.cruss-galli (*barnyard grass*)*. A distilled water control was included in the treatment. The study was carried out in petri plates of 9 cm and filter paper of double layer was used as sowing medium. Twenty-one treatment combinations of *S. oleraceus* weed parts *viz* leaves, stem and fruits in different concentrations viz. 0% (control), 0.25%, 0.5%, 1%, 2%, 4%, and 8% were tested against *E. cruss-galli*. Ten seeds of *E. cruss-galli* were placed in every Petri plates. Then 7 ml of all *S. oleraceus* extracts dilutions were added in all the Petri plates. Each treatment had 4 replications. Distilled water was used as control treatment. To reduce evaporation, petri plates were covered and rapped with scotch tape. The petri plates were kept at 30°C temperature and were moistened after one week.

Data collection

The data regarding emergence of seeds were recorded every day for 14 days. After 14 days, emerged seedlings of *E. cruss-galli* were taken and used to record different seedling growth parameters like shoot and root length, fresh and dry weight. Fresh weight was recorded instantly after harvesting while the seedlings dry weight was observed after oven drying for two days at 60°C.

Mean emergence time (MET) (days)

MET was determined by using the formula proposed by Ellis and Reborts, (1981).

$$MET = \sum \frac{(Dn)}{\Sigma n}$$

Emergence index (EI)

The emergence index was observed as per Association of Official Seed Analysts (1990) by using formula

$$GI = \frac{Number\ of\ seeds\ emerged}{Days\ of\ first\ count} + ----+ \frac{Number\ of\ seeds\ emerged}{Days\ of\ final\ count}$$

Emergence percentage (EP) (%)

No of emerged seeds were counted daily according to the method of the association of Official Seed Analysts (1990) and converted into emergence percentage by the following formula.

$$Emergence\ percentage = \frac{Number\ of\ seeds\ emerged}{Total\ Number\ of\ seeds} \times 100$$

Time to 50% emergence

The time taken to obtain 50% emergence (E_{50} or T_{50}) was calculated by the formula proposed by (Coolbear*et al.,* 1984).

$$T50 = ti + \left[\frac{\frac{N}{2} - ni}{nj - ni}\right](tj - ti)$$

Shoot length (ShL)(cm)

All seedlings from each Petri plate were separated 14 days after emergence. Shoot length was calculated by using scale from the junction of root and shoot to tip of the seedling and average was taken.

Root length (RL) (cm)

Root length was measured from junction of root and shoot to the tip of root and then average was work out.

Dry weight (DW) (g)

The seedlings dry weight was noted by oven drying the seedlings for 48 hours at 60°C. Then weighed by using digital balance to get average dry weight of seedling.

Statistical analysis

Data analyses were performed by using Statistix software (version 8.1 Statistix, Tallahassee, FL, USA). The least significant difference (LSD) test was used to compare the means at probability level of 5% (Steel *et al.,* 1997).

RESULTS

Mean Emergence Time (MET)

The application of aqueous extracts of *S. oleraceus*significantly affected the mean emergence time of barnyard seeds (Fig. 1). Leaf, stem and fruit aqueous extract of *S. oleraceus* influenced significantly the mean emergence time of *E. cruss-galli*. The leaf aqueous extracts resulted in highest (4.22 days) while fruit aqueous extracts the lowest (4.15 days) mean emergence time of *E. cruss-galli*. The interaction effect of plant parts and concentrations of extracts was found significant. Maximum MET (5.20 days) was examined under leaf extract at 8% concentration while minimum (3.36 days) under control (0%) *S. oleraceus*. Different concentrations of aqueous extract of *S. oleraceus* produced non-significant effect on mean emergence time of *E.cruss-galli.*

This showed that leaf aqueous extracts resulted in more inhibition of emergence as compared to fruit and stem aqueous extracts. The higher concentrations of aqueous extracts resulted in more delayed emergence. The maximum increase was recorded at 8% concentration with the leaf aqueous extracts while the minimum mean value was recorded at lower concentrations of aqueous extracts.

Emergence Index

Influence of aqueous extracts of *S. oleraceus* on the emergence index of *E. cruss-galli* seeds was significantly different (Fig. 2). The aqueous extracts of *S. oleraceus* posed inhibitory effect towards emergence index regarding concentration of aqueous extracts, plant parts as well as their interactive possessions. Concerning aqueous extract concentration, the highest value of emergence index (4.39) was noted at control (0%) while significantly the lowest value of emergence index (2.15) was noted at 8% concentration. At high concentrations, maximum reduction in emergence index was observed. Among the plant parts, fruit and stem extracts were responsible for lower emergence index values while leaf aqueous extract gave maximum emergence index (4.03). Interactive effect of plant part and concentration directed that highest emergence index (4.89) was observed at 0.5% concentrated leaf aqueous extracts while lowest (1.70) was recorded at 8% concentrated fruit aqueous extracts. Higher concentrations of weed aqueous extracts showed inhibitory effects towards emergence index.

Emergence percentage (%)

Emergence percentage of barnyard grass seeds was significantly affected by the water extracts of leaf, stem and fruit of *S. oleraceus* (Fig. 3). Maximum emergence percentage (82.80%) of *E. cruss-galli* was examined under leaf extract while minimum (69.58%) by fruit aqueous extract. Various concentrations of *S. oleraceus* aqueous extracts produced significant effect. Among concentrations, maximum emergence percentage (100%) was achieved under control (0%) whereas minimum (46.67%) by 8% aqueous extract. However, interactive effect of plant parts and their concentration was non-significantly on emergence percentage of *E. cruss-galli*. Increase in concentration of extracts resulted in decreased emergence of barnyard grass and the maximum reduction was observed with highest concentration (8%) of *S. oleraceus* aqueous extract.

Time to complete 50% emergence of *E. cruss-galli*

Time taken to complete 50% emergence of *E. cruss-galli* seed was significantly affected by aqueous extracts of leaf, stem and fruit of *S. oleraceus* and their various concentrations (Fig. 4). Seed of *E. cruss-galli* treated with stem extract of *S. oleraceus* took more time (3.01 days) to complete 50% emergence while minimum (2.75 days) time was taken by fruit extract. Among different concentrations *E. cruss-galli* control (0%) took the maximum (3.47 days) while 8% concentration the minimum (1.75 days) time to complete 50% emergence. Plant parts× concentration produce non-significant effect on time taken to complete 50% emergence of *E. cruss-galli* seed.

Root length of *E. cruss-galli*

Root length of *E. cruss-galli* seedlings was significantly affected by the leaf, stem and fruit aqueous extracts of *S. oleraceus* (Fig. 5). Leaf aqueous extract produced seedlings with the longest root length (1.47cm) whereas fruit extract the shortest root length (1.03 cm). Among various concentrations, maximum (1.63 cm) and minimum (0.44 cm) root length was examined at 1% and 8% concentrations, respectively. Different plant parts and their various concentration produced significant effect on root length of *E. cruss-galli*. The significantly highest mean value (2.38 cm) was recorded with 0% concentration while the significantly the lowest mean value (0.09 cm) was observed with fruit aqueous extracts of 8% concentration.

All the extracts showed significant reduction in barnyard grass root length at 8% concentration. The lower concentrations of the extracts showed promontory effect except stem extract whereas inhibitory effect was observed by higher concentrations.

Shoot length of *E. cruss-galli*

The shoot length of barnyard grass seedlings was not affected by the aqueous extract of *S. oleraceus* parts such as leaf, stem and fruit. Interaction of plant parts and their various concentrations produced significant effect on shoot length of *E. cruss-galli* (Fig. 6).The highest shoot length (6.74 cm) was recorded at 0.5% while the lowest shoot length (2.37cm) was recorded at 8% concentration. This showed the inhibitory actions of extracts at higher concentrations. Various concentrations of aqueous extract of *S. oleraceus* caused **s**tatistically significant effect on the shoot length of *E. cruss-galli* seedlings. The highest mean value (8.38 cm) of shoot length was observed at 0.5% concentration while the lowest mean value (0.45 cm) was recorded at 8% concentrated fruit extract. Although the application of extracts at lower concentrations up to 0.5% promoted the shoot length but these differences were not statistically different from each other.

Dry weight of *E. cruss-galli* seedling

Aqueous extracts of *S. oleraceus* plant parts caused significant effect on *E. cruss-galli* seedling dry weight (Fig. 7). The lowest seedling dry weight (1.35 g) was produced by the application of fruit aqueous extracts while the highest (3.13 g) by the application of leaf aqueous extracts. The results regarding concentrations of weed aqueous extracts were found non-significant. However, the interactive effect of different plant parts and various concentrations significantly influenced the seedling dry weight. The highest dry weight (6.24 g) was recorded at 0.5% concentrated leaf aqueous extract while the lowest mean value (0.17 g) was observed at 8% fruit aqueous extract.

Total phenolics (mg/g) and flavonoids (mg/g) in aqueous extract of different parts of *S. oleraceus* weed

The total phenolics and flavonoids showed differences among plant parts. The highest amount of phenolics and flavonoid was observed in fruit extracts. The minimum amount of phenolics and flavonoid was observed in leaf extract (Fig. 8).

DISCUSSION

The water extracts of all plant parts showed inhibitory effects on the emergence of seed of *E. cruss-galli*. However, maximum suppression was recorded with application of leaf extract. The lower inhibitory effect of leaf extract on the germination can be attributed to lower concentration of phenolics and flavonoids in leaf extract compared to extracts from other plant parts. The significant inhibitory effect of weed aqueous extracts on the germination of plants has also been reported by Dongre and Yadav (2005). Tanveer *et al*. (2008 and 2010) evinced the inhibitory effects imparted by aqueous extracts of different parts of *Alternanthera* species. They reported that the leaves of *Alternanthera* species contain more allelochemicals than other plant part therefore resulted in greater inhibitory effect.

The germination index was decreased with increasing concentrations of the extracts and minimum germination index was recorded with fruit extract. These findings are similar to those of Kadioglue *et al*. (2005) and Tanveer *et al*. (2008 and 2010) who reported decrease in germination index of rice seeds with the application of aqueous extracts of two weeds namely *Alternanthera sessilis*and *Alternanthera philoxeroides.* Nadeem *et al.*(2020b) reported that Water extracts of leaf of *C. tinctorius* at 8% concentration result in lowest *E. cruss-galli* emergence index. The germination percentage and time to complete 50% germination were influenced significant by plant parts and concentrations of aqueous extracts. The germination percentage and time to germination was decreased with increase in concentration. The results of germination percentage were analogous to the findings of Dessalegne *et al.* (2013) who studied allelopathic effects of leaf, stem and root parts of *Datura stramonium*, *Argemone mexicana*, *Parthenium hysterophorus* and *Amaranthus hybridus* on seed germination, seedling growth and biomass production of wheat cultivars. They reported that aqueous extracts of weeds caused maximum reduction in germination of wheat seeds. Decreased mean value with increasing extract concentrations showed significant inhibitory actions of extracts. According to Nadeem *et al.*(2020a) who reported that all the concentrations of *C. tinctorius* enhance the time to complete 50% emergence of *O. punctata* with 8% concentration Similar inhibitory effects of aqueous extracts were also demonstrated by Chandra *et al.*(2011) who reported that aqueous extracts of *Achyranthes aspera* imparted strong inhibitory effects on the germination and seedling growth of *Vicia faba* L. at its higher concentrations. Reduced seedling growth may also be the result of reduced shoot and root length of seedlings. The differences among the plant parts can be attributed to differences in total phenolics and flavonoids. The stimulatory effect at lower concentration of leaf and flower extract can be attributed to the hormeotic effect of phytotoxic compound and has also been reported by Nadeem *et al.* (2017).

The decrease in root length of *E. cruss-galli* at higher concentrations might be due to greater amount of the allelochemicals at these concentrations. The increase in root length with leaf and flower extracts at lower concentrations compared with control might be due to hormeotic effect of these extracts. These results showed that overall, more inhibitory effects was shown by fruit extracts while leaf and stem extracts showed minimum inhibitory effects on root length. This might be possible due to the presence of more phytotoxic substances in fruit extracts as compared to leaf and stem extracts. Nadeem *et al.* (2020b) studied the effect of aqueous extracts of various parts of *C. tinctorius* on the root length of barnyard grass results revealed that minimum root length of barnyard grass was produced by produced by safflower leaves aqueous extract whereas, seedlings with lengthiest roots were noted by application of stem aqueous extracts of *C. tinctorius*.

The results regarding the allelopathic behaviour of aqueous extracts of different plant parts of sow thistle on the root length were analogously obtained by Della *et al*. (2009) and Aljubory *et al*. (2010). On the other hand, root extracts were found to be more stimulatory while leaf extracts had more inhibitory effects compared to other parts extracts and control (Aljubory *et al,* 2010). The differences among different concentrations were non-significant. Safdar *et al.* (2016) reported drastic reduction in germination percentage, germination index, and seedling growth; and delay in germination of parthenium weed by aqueous extracts derived from *A. aspera* and*A. philoxeroides.* However, these results are contradictory to those of Park *et al.* (2011) who reported the negative effects of higher concentrations of *S. oleraceus* stem extracts on root length. This contradiction in results can be attributed to the differences in the test plant species and site conditions from where samples were collected for preparation of extracts.

The differences among plant parts were non-significant. The lower seedling dry weight was observed with fruit extract. The results are different from those of Prasad and Priyadar-shani (2006) who reported that fruit extracts showed more inhibitory actions. They demonstrated that *Parthenium hysterophorus* and *Adhatodavasica* leaf and fruit extracts reduced the seedling growth of *Brassica rapa* L. The differences in the results can be attributed to difference in plant material used for extract preparation and test plants under study. The inhibitory effect at higher concentration can be attributed to presence of allelochemicals that inhibited the growth of the seedlings. The inhibitory effect of plant water extracts on the seedling growth has also been advocated by Mitra and Prasad (2010). The maximum shoot length with leaf extract was might be possible due to hormeotic effect of leaf extract. Nadeem *et al.*(2020b) studied the effect of aqueous extracts of various parts of *C. tinctorius* on the root length of barnyard grass. Results revealed that minimum root length of barnyard grass was produced by produced by safflower leaves aqueous extract whereas, seedlings with lengthiest roots were noted by application of stem aqueous extracts of *C. tinctorius*.

These results showed that fruit aqueous extracts contained more phenolics and flavonoids that causes the reduction in dry weight of barnyard seedlings at higher concentrations. This reduction might be possible due to the inhibition of seedling growth due to the actions of allelochemicals and phenolics. By the application of different annual weeds aqueous extracts on the soybean plants, inhibition in seedling growth has also been reported by Bhowmik and Doll (1984).

CONCLUSION:

This study was aimed to explore the phytotoxic potential of *Sonchus oleraceus* L. on the emergence and seedling growth of *Echinocloa cruss-galli* L. and quantification of allelochemicals present in leaves, stem and fruit of *S. oleraceus* weed. Results suggested that the extract of *S. oleraceus* weed at concentration (8%) can be used as potential bio-herbicide for the control of *E. cruss-galli* weed.

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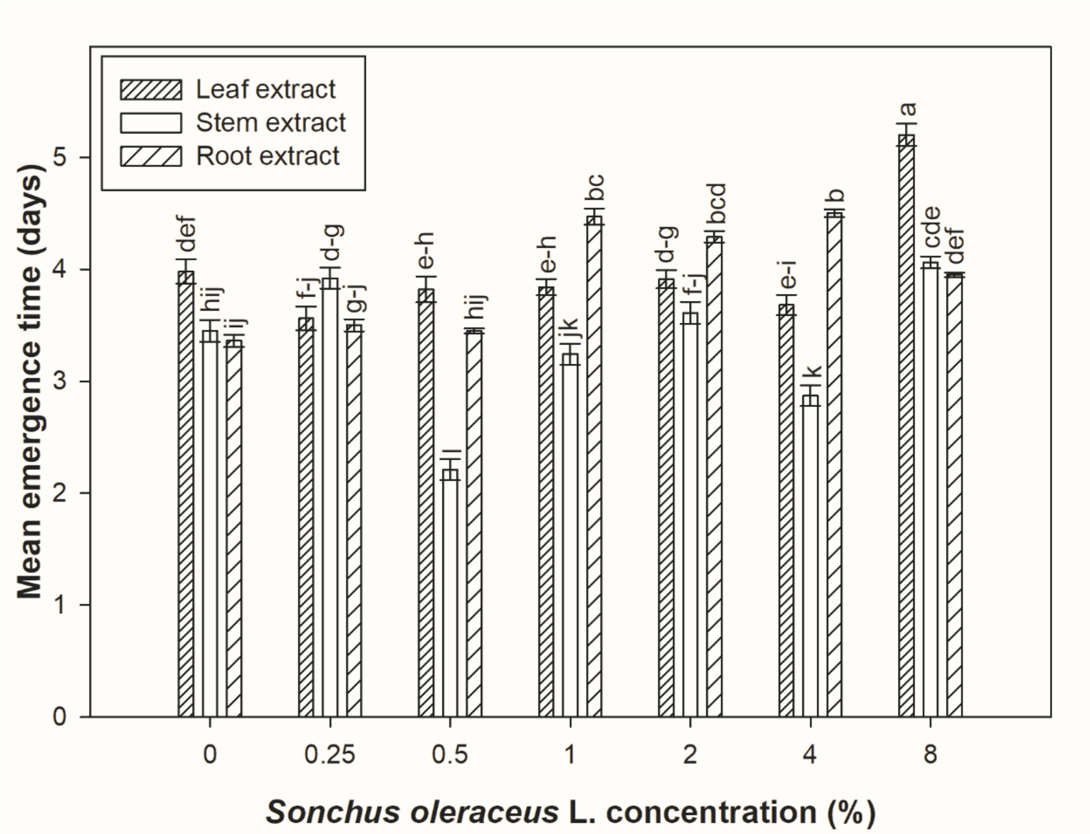

Fig.1: Phytotoxic effects of *sonchus oleraceus* on emergence time (days) of *echinocloa cruss-galli*

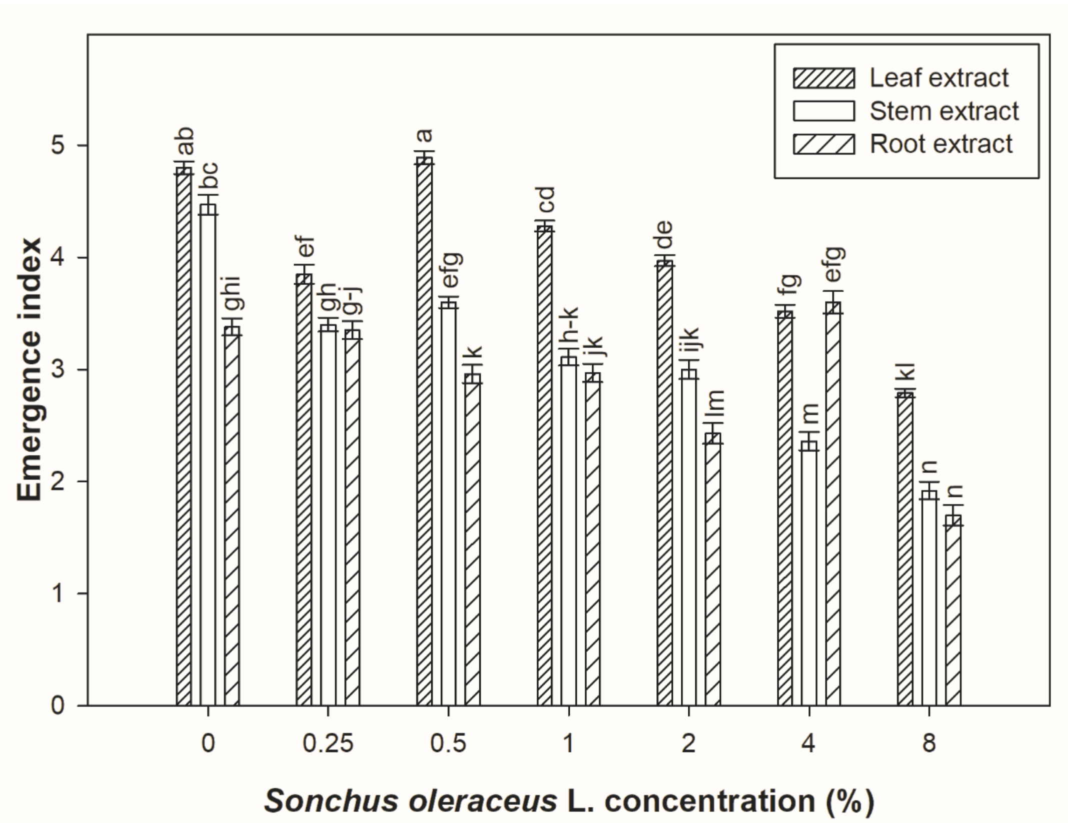

Fig.2: Phytotoxic effects of *sonchus oleraceus* on emergence index of *echinocloa cruss-galli*

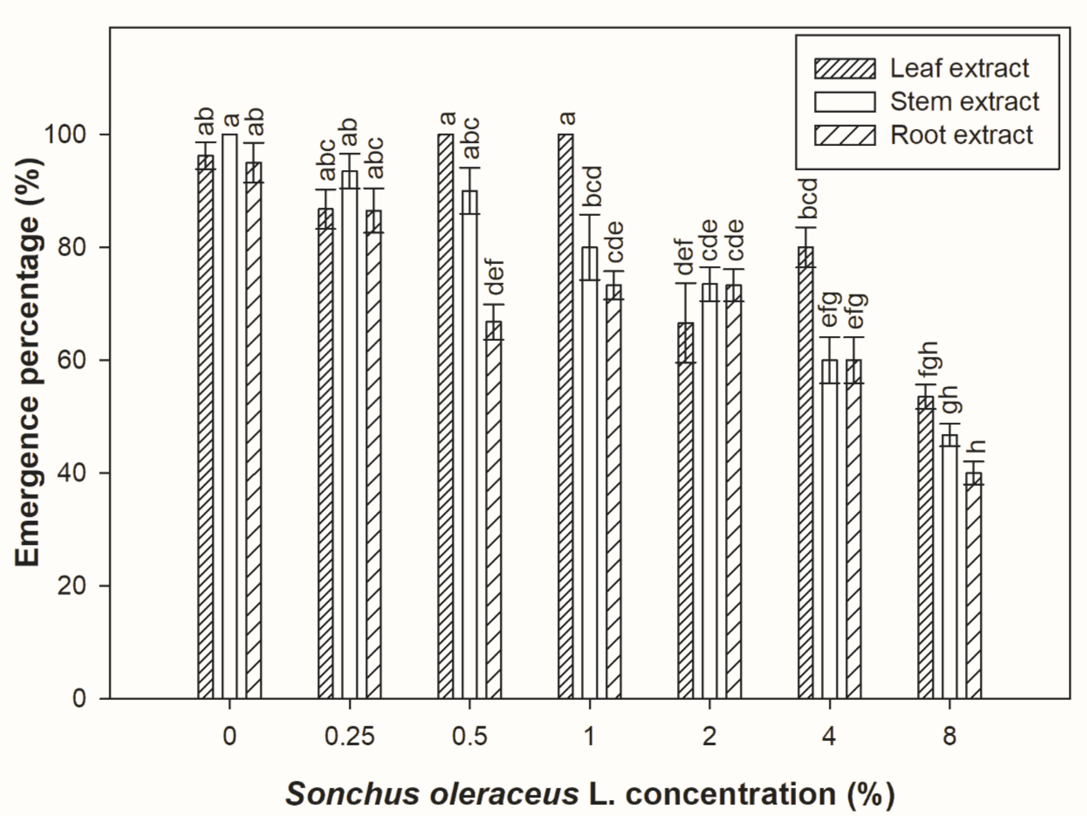

Fig.3: Phytotoxic effects of *sonchus oleraceus* on emergence percentage (%) of *echinocloa cruss-galli*

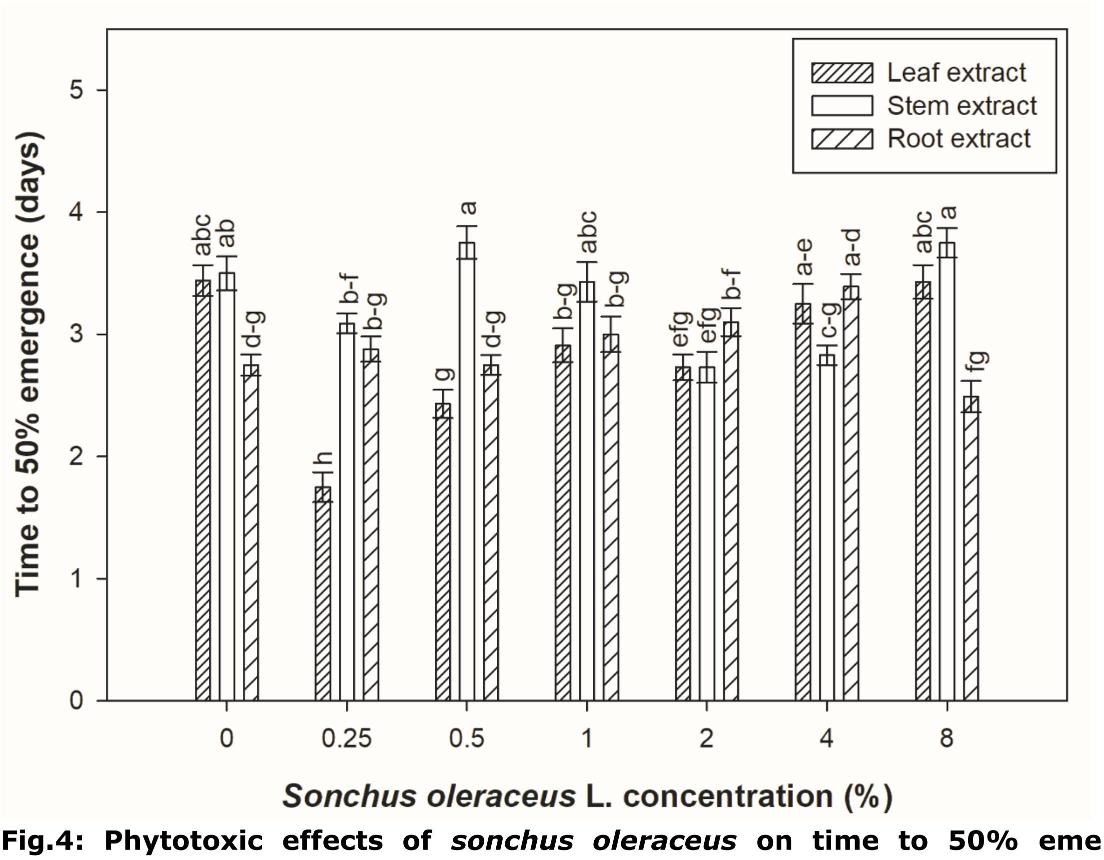

Fig.4: Phytotoxic effects of *sonchus oleraceus* on time to 50% emergence (days) of *echinocloa cruss-galli*

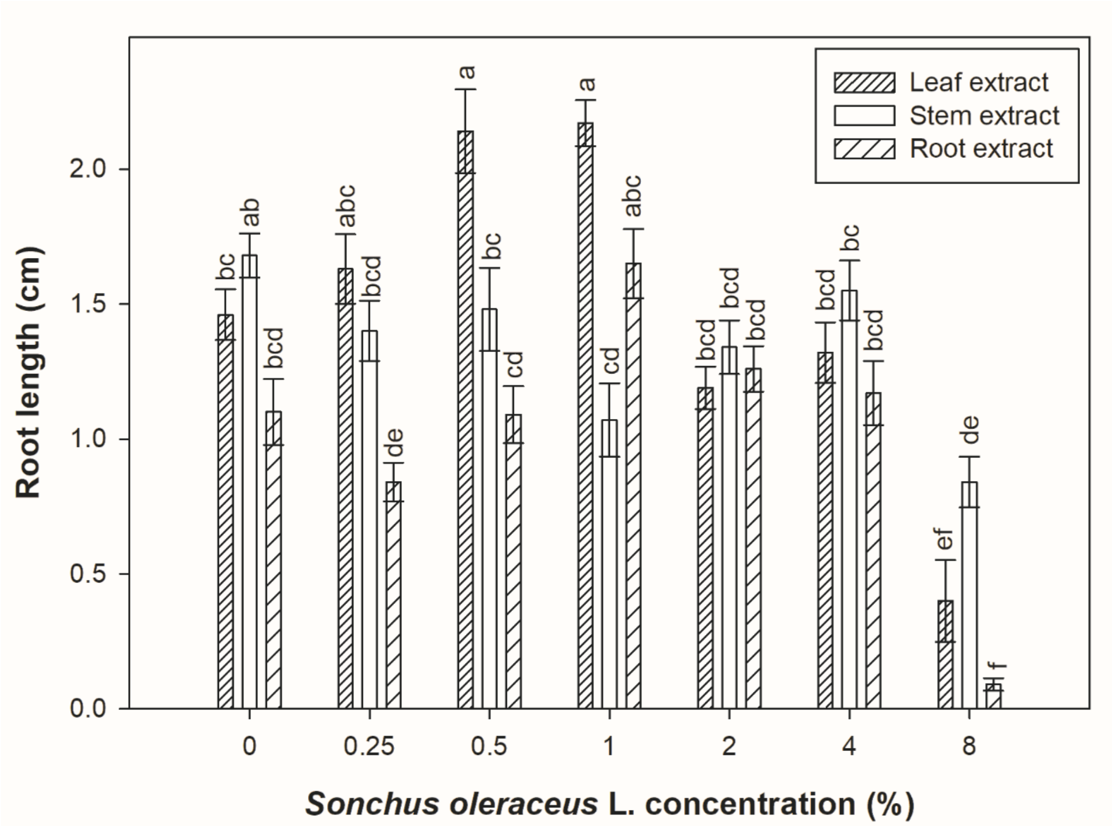

Fig.5: Phytotoxic effects of *sonchus oleraceus* on root length (cm) of *echinocloa cruss-galli*

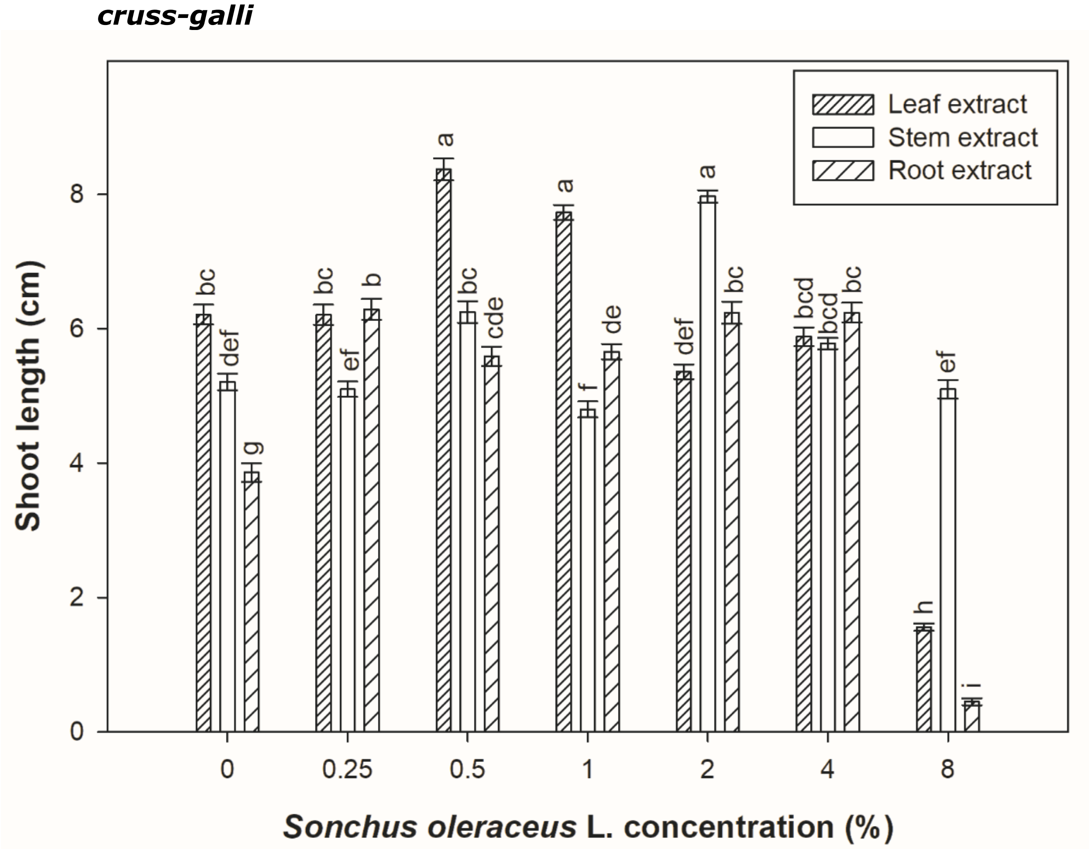

Fig.6: Phytotoxic effects of *sonchus oleraceus* on shoot length (cm) of *echinocloa cruss-galli*

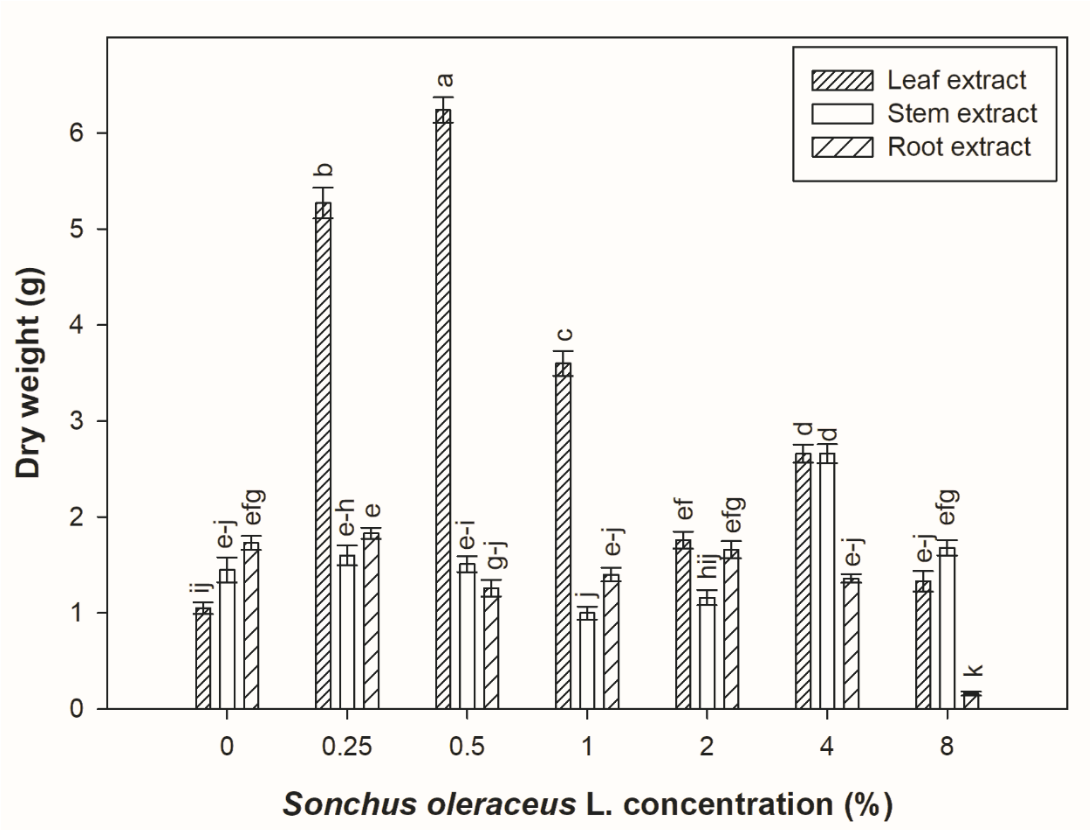

Fig.7: Phytotoxic effects of *sonchus oleraceus* on dry weight (g) of *echinocloa cruss-galli*

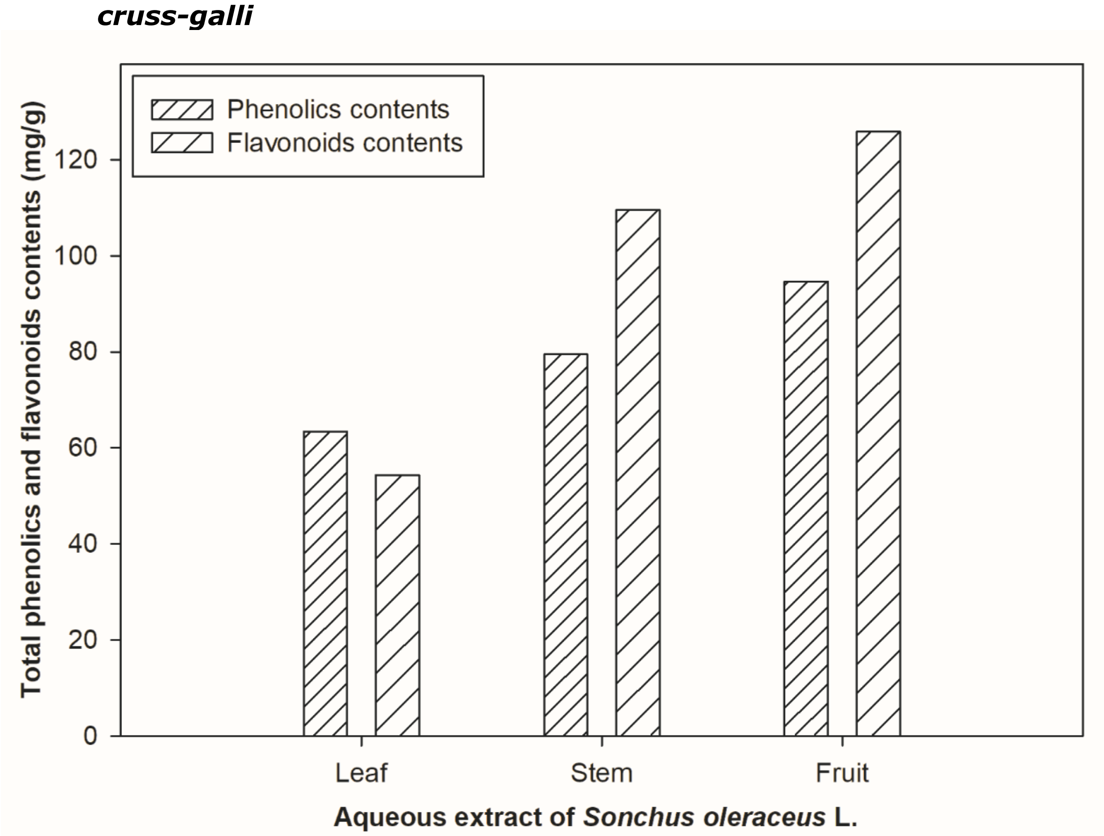

Figure:8 Total phenolics (mg/g) and flavonoids (mg/g) in aqueous extract of different parts of *S. oleraceus* weed