Allelopathic effects of *Heliotropium bacciferum* leaf and roots on *Oryza sativa* and *Teucrium polium*

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Abstract: In the present study an attempt has been made to investigate the potential allelopathic effects at different concentrations (0, 10, 25, 50 and 75%) of water extract of roots and leaves of *Heliotropium bacciferum* Forssk. on seed germination, seedling growth and biochemical constituents changes *Oryza sativa* L and medical plants *Teucrium polium* L. The results showed that the roots and leaves extracts of *H. bacciferum* caused a general phytotoxic effect on seed germination and seedling growth of *O. sativa* and *T. polium* at all concentrations. It was noticed that the germination in both tested plants was delayed for one and two days at the highest levels of extract (50 and 75%). The degree of seed germination inhibition was higher in leaf extract more than root extract of heliotrope plants. Also, *O. sativa* seeds were more affected at different extract concentration than *T. polium*. With respect to the internal metabolites, a gradual increase in the proline content, decrease in the chlorophyll content and consequently the soluble sugar content is proportional to the increase the concentrations of water extract of roots and leaves of heliotrope. Aqueous extract of heliotrope leaves induced more inhibitory effects than aqueous extract of heliotrope roots. A greater inhibitory effect was observed on *T. polium* as compared with *O. sativa*. Consequently, the aqueous extracts from *H. bacciferum* have allelopathic potential and should be evaluated as an allelopathic species, presenting arisk to seed germination and seedling growth of crops or weeds.
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1. Introduction

Heliotropium bacciferum Forssk. (Common heliotrope) widespread in Eastern province of Saudi Arabia (Chaudhary, 2001). This species has been known as medicinal plant (Diana et al., 2002). Allelopathy is a phenomenon observed in many plants that release chemicals into the near environment either from their aerial or underground parts in the form of root exudation leaching by dews and rains, and volatilization or decaying plant tissue (Rice, 2000). Any direct or indirect and harmful or beneficial effect by one plant (including micro-organisms) on another through production of chemical compounds that escape into the environment is called allelopathy (Rice, 1984).

Allelopathy in agricultural practices has become more important in biological control of weeds (Piyatida and Noguchi, 2010). The inhibition of the plant depends on the concentrations of the allelochemicals (Ashrafi et al., 2009; Batlang and Shushu, 2007). It is defined as the direct or indirect harmful or beneficial effects of one plant on another through the release of chemical compounds into the environment. It plays a significant role in agro ecosystems and affects the growth, quality and quantity of the produce (Kohli et al., 1998; Singh et al., 2001). Allelopathy exists in almost all plants, and it has been proposed as a mechanism for influencing plant populations and communities by organize the germination and establishment of species (Khanh et al., 2007; Gleissman, 1983). Also, it is generally accepted as a significant ecological factor in determining the structure and composition of plant communities (Scrivanti et al., 2003). Allelochemicals are present in almost all plants and their tissues such as leaves, stems, roots, flowers, seeds, bark, and buds (Weston and Duke, 2003). Generally leaves are the most effective source of allelochemicals, the allelopathy is accepted as an important ecological factor in determining the chemistry of plant communities (Lorber and Muller, 1976; Mizutani, 1989; Seigler, 1996).

The weed have allelopathic superiority over crops besides their competition superiority (Zzet and Yusuf, 2004). In allelopathy, relations between weeds and crops, between weeds and weeds and between crops and crops (Rice, 1984) because the modern agriculture relays on synthetic chemicals to get rid of these unwanted plants. Also, many researchers have found that inhibitory substances involved in allelopathy are terpenoids and phenolic substances (Khanh et al., 2007). A number of weeds grow in rice field, which compete with crop and reduce the crop yield (Ampong-Nyarko and de Datta, 1991). If the allelopathic effect of specific rice varieties on specific weeds can be known, weed management becomes economically more effective for the crop grower (Azania et al., 2003).

In recent decades, many researchers such as (Einhellig, 2002; Yang et al., 2002; Setia et al., 2007;

Jabeen et al., 2011; Raoof and Siddiqui, 2012 and Mangal et al., 2013, 2014) have reported the effect of various allelochemicals of different plants on physiological and biochemical processes but reports regarding effects of invasive plant allelochemicals on germination, seedling growth and protein expression of some native species are still scanty.

There are several reports that accumulation of proline (Cattivelli et al., 2008) content and soluble sugars (Izanloo et al., 2008) of leaves decreased under stressful. Production and accumulation of free amino acids, especially proline by plant tissue. The present work aimed to determine the allopathic effect of water extracts of heliotrope roots and leaves on *Oryza sativa* L. and *Teucrium polium* L. germination, seedling growth, total chlorophyll content, total soluble sugar content and proline content.

Salient features of the plants used

Teucrium polium L. locally called ('Jaada') is a dwarf shrub plant which grows wild in Mediterranean and Middle East countries. The genus *Teucrium* (Germander) belongs to the family Lamiacae (El-Shazly and Hussein, 2004). Its flowers are small and range from pink to white, is a medicinal plant that has been used for more than two thousand years for treating many diseases such as abdominal pain, indigestion and diabetes in the Middle East (Kandouza et al., 2010).

Oryza sativa L. var. Hassawy is cultivated in the Eastern Province of Saudi Arabia, trobpical and subtrobical region, commonly known as Asian rice, is a genus which belongs to Poaceae (Graminae) family. (Chaudhary, 1989).

2. Materials and Methods

Collection of material

Heliotropium bacciferum plants were collected during the flowering stage from naturally growing stand in the salty, gravelly soil (26, 41503°N, 50.09906°E, 200 m altitude) in Eastern province, Dammam city, Saudi Arabia. Plant seeds of *O. sativa*, were obtained from the Agriculture Faculty of Alhassa, Saudi Arabia, while *T. polium* were obtained from the Camel and Range Research Centre, Saudi Arabia. The experiment was carried in the laboratory at the Department of Biology, Faculty of Science, University of Dammam, Saudi Arabia, during the period of April to May, 2013.

Preparation of extracts

The plants were gently washed with distilled water, dried between two paper towels. The parts were brought into the laboratory and each part of the fresh plant was cut into small pieces, shade dried and then ground separately with help of electronic grinder and made fine powder. The leachates from the leaves were prepared by immersing 100 g leaves in 1.0 L double distilled water in a flask for 24 h at room temperature (20±2°C) (Djanaguiraman et al., 2002). The precipitates so formed were recovered through centrifugation (2000 rpm) for 30 min .The extracts were then filtered with double layer of muslin cloth followed by Whatman filter paper No.1. and stored at 5°C. Stock extracts (leaf and root) were diluted with sterile distilled water to give final concentrations of 0, 10, 25, 50 and 75%.

Seed germination and seedling growth

Before germination tests, the plant seeds were surface-sterilized with 1% NaOCl for 5 min and then washed six times with distilled water. Distilled water was used as a control treatment.

Germination tests were performed for each of the root and leaf extracts from *H. bacciferum* as follows: According to seed size, 25 or 50 the sterilized seeds were evenly placed on two layers of filter paper in sterile Petri dishes (9 cm). Due to differences in the size of *O. sativa* and *T. polium* seed, 10 and 5 ml of extract solution were added to Petri dish containing *O. sativa* and *T. polium*, respectively. Distilled water was used as a control treatment. All Petri dishes were placed in a dark room at 25°C. Treatments were arranged in a completely randomized design with four replications. Germination was determined by counting the number of germinated seeds at 24-h intervals over a 7-days. The rate of germination was calculated by dividing the number of germinating seed each day by the number of days and summing the values (Maguire, 1962). Germination was deemed to occur only after the radicle had protruded beyond the seed coat by at least 1 mm. After7-days. Radicle and plumule lengths of *O. sativa* and *T. polium* seedlings were measured. After measuring the radicle and plumule lengths, the dry weights of seedlings were determined by drying the plant material in an oven at 60 °C for 24 h prior to weighing. The inhibitory or stimulatory percent was calculated using the following equation given by Chung et al., (2001).

$$\text{Inhibition \%} = \frac{[(\text{Control} - \text{extracts})] \times 100}{\text{control}}$$

At the end of the experiments 14-dayes of emergence the plumules tissue were used to determine their contents of chlorophyll, soluble sugars and proline.

Measurement of chlorophyll content

The total chlorophyll content from leaves of treated or control plants were extracted from a known fresh weight of leaves in 85% (v/v) aqueous acetone. The extract was taken and diluted by 85% aqueous acetone to the suitable concentration for spectrophotometric measurements using spectrophotometer (UV-1800) according to the method used by Todd and Basler (1965). Contents of total chlorophyll were expressed as mg/100g fresh weight.

Measurement of soluble sugars content

Soluble sugars were determined based on the method of phenolsulfuric acid by (Dubois et al., 1956). 0.5 g fresh weight of roots and shoots was homogenized with deionized water, extract was filtered and the extract treated with 5% phenol and 98% sulfuric acid, mixture remained for 1h and then absorbance at 485 nm was determined by spectrophotometer (Biochrom S 2100). Contents of soluble sugar were expressed as mg/100g fresh weight.

Measurement of proline content

Free proline accumulation was determined using the method of Bates et al., 1973. 0.5 gm fresh weight of roots and Leaves was homogenized with 3% sulfosalicylic acid and 2 ml of filtrate was reacted with 2.0 ml acid ninhydrin and 2.0 ml of glacial acetic acid in a test tube. The mixture was boiled for 1 hour at 100°C in water bath the reaction terminated in an ice bath .The reaction mixture was then extracted with 4.0 ml toluene and vortexed in a mixer for 10-15 seconds. Warmed to room temperature 25 °c and the absorbance red at 520 nm was determined by Uv-visible spectrophotometer (Biochrom S 2100) using toluene for a blank .Contents of proline were expressed as µmol g-1 FW.

Experimental design and statistical analysis

Germination and seedling growth bioassays were calculated in a complete randomized design with four replications. The data were subjected to one-way analysis of variance, and treatment means separated from the control at P<0.05 applying post hoc Dunnett's test. Statistical analysis was done with SPSS 15.0 for Windows statistical software package.

3. Results

Germination percent

The data of germination percentage of *O. sativa* grains and *T. polium* are presented in Fig. 1 and 2, respectively. The % germination of rice grains significantly reduced by aqueous extract of heliotrope roots which produced 25, 38, 64 and 80% seed germination at 10, 25, 50 and 75%, respectively (Fig. 1). Whereas, the inhibition of *T. polium* seeds recorded 25, 35, 57, and 74% at 10, 25, 50 and 75%, respectively (Fig. 2). It was observed that the treatments with aqueous leaf extract of heliotrope plants induced inhibition of % germination in both tested plants (Fig. 1 and 2).

The degree of inhibition in rice grains reached about 17, 29, 50 and 66% at 10, 25, 50 and 75% from aqueous leaf extract percentages, respectively (Fig. 1). Whereas, the inhibition of *T. polium* seed germination was recorded at aqueous leaf extract percentages 10, 25, 50 and 75%, respectively (Fig. 1). Also, it noticed that as the extract level increased the inhibitory effect on % germination increased.

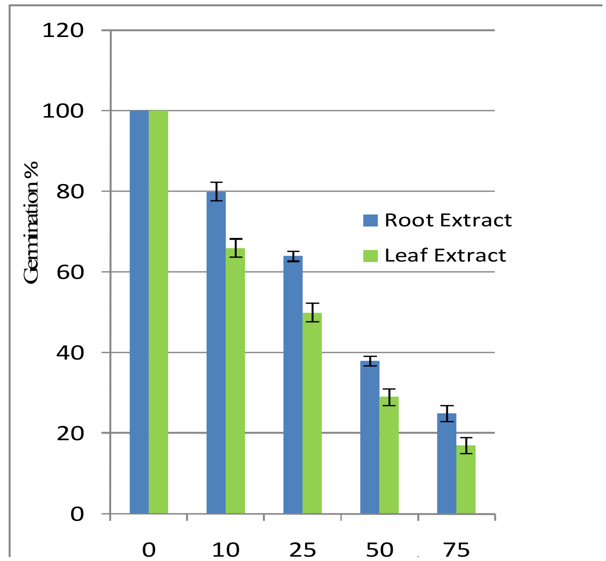

Figure 1. Percentage germination of *O. sativa* seeds germinated under different levels of root and leaves extract of heliotrope plants.

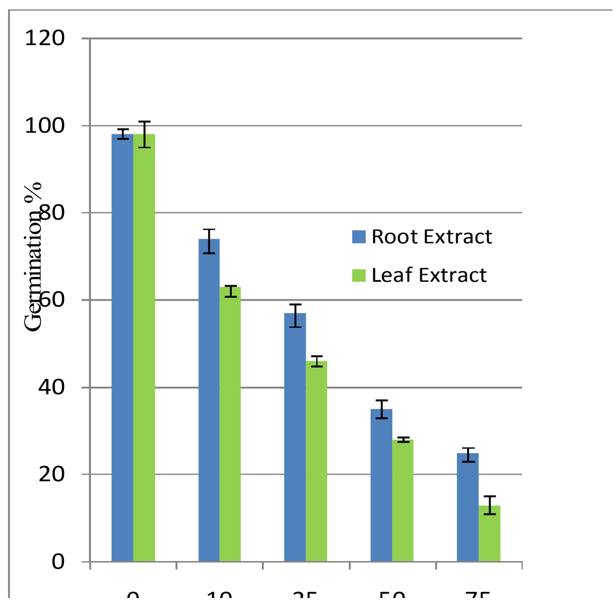

Figure 2. Percentage germination of *T. polium* seeds germinated under different levels of root and leaves extract of heliotrope plants.

The maximum seed germination percentage was reported at the low levels 5% of extract in *T. polium* and *O. sativa* as compared to the water extract (control) which was 100 and 98%, respectively. The degree of inhibition gradually increase with increasing extract concentration. It was observed that the germination in both tested plants was delayed for one and two days at the highest levels of extract (50 and 75%). Also, it was noticed that the degree of seed germination inhibition was higher in leaf extract more than root extract of Heliotrope plants. Also, *O. sativa* seeds were more affected at different extract concentration than *T. polium*.

Effect of allopathic extract on radical and plumule lengths

Data in Table (1) showed that the plumule and radicle length significantly ($P<0.05$) influenced by various aqueous extract concentrations as compared with the control treatment and the degree of inhibition increased with increasing extract concentration.

Table 1. Effects of heliotrope leaf and root aqueous extracts on the radicle length of *O. sativa* and *T. polium.*

Plant	*H. bacciferum* parts	Extract conc. (%)	Radicle length (cm) (Mean ± SD)	%stimulation (+) or inhibition (–) over control
O. sativa	Root	0	2.58±0.30**	--
		10	1.93±0.10**	-25.19
		25	1.33±0.13*	-48.45
		50	1.03±0.05*	-60.08
		75	0.80±0.08	-68.99
	Leaf	0	3.38±0.15*	--
		10	1.83±0.10**	-45.51
		25	1.25±0.06	-63.95
		50	0.63±0.05**	-75.58
		75	0.45±0.06**	-81.39
T. polium	Root	0	3.38±0.15**	--
		10	2.43±010**	-28.11
		25	1.35±0.13*	-39.94
		50	1.13±0.10**	-66.56
		75	0.63±0.13**	-81.36
	leaf	0	3.38±0.15**	--
		10	1.83±0.10*	-45.86
		25	1.25±0.06*	-63.08
		50	0.63±0.05**	-81.36
		75	0.45±0.06**	-86.69

* Indicate significant differences between treatments at $P<0.05$.

The percentage of rice radical inhibition reached to -68.99% at 75 % water extract of roots and -81.39% in case of water leaf extract, respectively (Table 1). However, at the high concentrations the emergence of plumule was delayed to in the 5th and 7th day, respectively and continued to grow very slowly and reached at 50% of leaf concentration only 0.5 and 0.3 cm in the *O. sativa* and *T. polium*, respectively. In case of *T. polium* seeds, the plumule appeared at the sixth day after sowing at the low and medium concentrations of the leaf extraction (10 and 25%) while the plumule did not emerged at the high leaf extract concentration 75% (Table 2).

Table 2. Effects of heliotrope leaf and root aqueous extracts on the plumule emergence and length of *O. sativa* and *T. Polium.*

Plant	*H. bacciferum* parts	Extract Conc. (%)	Plumule length (cm) (Means ± SD)	stimulation (+) or inhibition (–) over control	Days of plumule emergence
O. sativa	Root	0	1.43±0.10*	--	3
		10	1.05±0.06*	26,57	3
		25	0.90±0.08**	37.06	4
		50	0.80±0.08**	44.06	4
		75	0.00±0.00	--	4
	Leaf	0	1.43±0.10*	--	3
		10	0.90±0.08*	37.06	3
		25	0.73±0.08*	48.95	4
		50	0.50±0.08*	65.03	5
		75	0.00±0.00	--	5
T. polium	Root	0	1.30±0.08**	--	4
		10	1.00±0.08	23.07	5
		25	0.83±0.10**	36.15	5
		50	0.70±0.08**	46.15	5
		75	0.00±0.00	---	6
	leaf	0	1.30±0.08**	--	4
		10	0.85±0.06**	34.62	6
		25	0.63±0.10**	51.54	6
		50	0.30±0.08**	76.92	7
		75	0.00±0.00	--	-

* Indicate significant differences between treatments at $P<0.05$.

Also, the radicle of *T. polium* seedlings was changed brown at the highest concentration (75%) immediately after emergence due to the high toxicity .The root length was more sensitive to all types of extract used in comparison to the other plant parameters measured.

Between the two test species, a greater inhibitory effect was observed on *T. polium* (-86.69%) at 75% of leaf aqueous extract compared with *O. sativa* (Table 2). Maximum radicle dry weight were recorded in *O.*

sativa at the lowest concentration as compared to control of root aqueous extract.

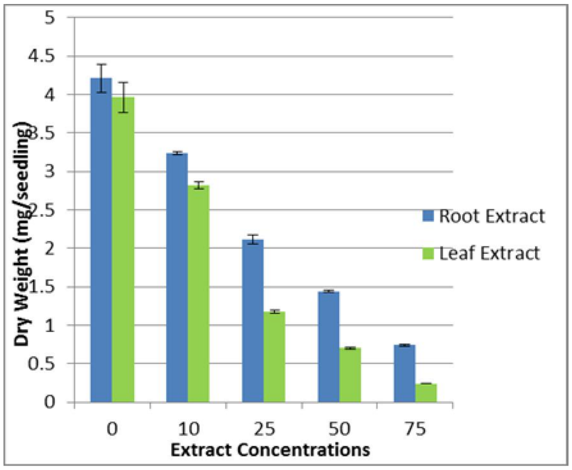

Figure 3. Dry weight (mg/ seedlings) of *O. sativa* germinated in different levels of Heliotrope leaf and root water extract.

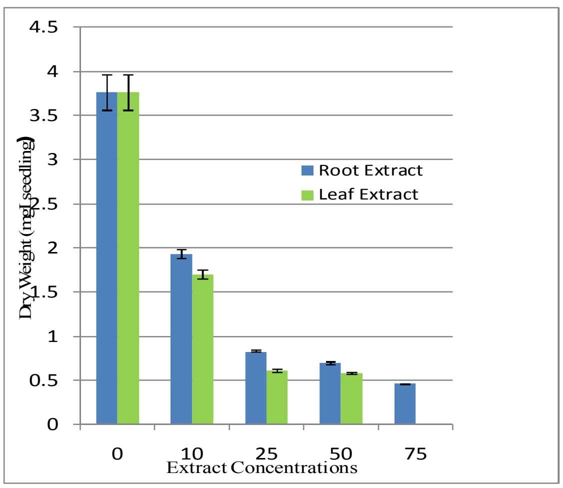

Figure 4. Dry weight (mg/seedlings) of *T. polium* germinated in different levels of heliotrope leaf and root water extract.

Effect of allopathic extract on total chlorophyll

Data in Table (3 and 4) showed the result of the effect of different concentrations of root leaf extract of heliotrope and the leaf extract on total chlorophyll content.

The results of total chlorophyll of *O. sativa* and *T. polium* germinated under different levels of both roots and leaf extract of heliotrope plants showed a significant decrease as the level of % water extract of both roots and leaves of heliotrope increased in the medium of germination. The total chlorophyll contents of *O. sativa* and *T. polium* seedling plumules showed an increase especially at the low levels (10 and 25%) of root or leaf heliotrope extract (Table 3,4) whereas, the high levels (50 and 75 %) of both extract roots and leaves showed a significant decrease in total chlorophyll contents. Also, the data showed the inhibitory effect of leaf extract more the inhibitory effect of root extract.

Effect of allopathic extract on soluble sugars content

Results in Table (3, 4) revealed that the total soluble sugars content in both roots and leaves of *O. sativa* and *T. polium* significantly decrease with increasing extract concentration, however soluble sugar levels in *O. sativa* was more than *T. polium* under control plants. The data showed that the highest content of these osmotic adjustments was found in *O. sativa* (P<0.05). Moreover, soluble sugar content in leaves was more than roots (P<0.05). The higher concentrations (75%) reduced the total soluble sugar contents by 33 and 20 mg/100g.F.wt in of *O. sativa* and *T. polium* , respectively as compared to the control (Table 3, 4).Whereas, the low levels (10 and 25%) of both extract roots and leaves showed an increase in total soluble sugars contents increased with increasing extract concentration.

Effect of allopathic extract on proline content

Data analysis of variance (Table 3, 4) showed that the means for effects of both extract roots and leaves heliotrope extract were significant at all concentrations and their interaction was also significant. Comparison of means (Table 3, 4) showed that there was a significant increase in proline contents with increasing extract concentration. This indicating that, the chemical stress is induced by leaf extract .The minimum amount of proline was recorded in controls (24 μmol/g FW) *T. polium*. While, the highest amount at high levels (50 and 75 %) of both extract roots and leaves heliotrope extract. Moreover, the leaves extract was more effective than the root extract in increasing the proline content of *O. sativa* and *T. polium* seedlings (Table 3, 4).

4. Discussion

In the present study, the results indicated that the aqueous extracts from different parts of heliotrope show a phytotoxic influence on *T. polium* and *O. sativa* (Tables 1 and 2). Extracts from heliotrope root and leaf solutions showed inhibitory effects on seed germination of two tested plant species. This result is consistent with Soltani et al., (2006) where they reported that the aqueous extracts of *Thymus kotschyanus* had a considerable inhibitory effect on germination of *Bromus tomentellus* and *Trifolium repens*. The degree of inhibition increased with increasing extract concentration. Also, similar finding were also observed by many researchers (Oudhia, 2001; Oueslati, 2003; Siddiqui et al., 2009).

Table 3: Effects of heliotrope leaf and root aqueous extracts on total chlorophylls, total soluble sugars and proline of *O. sativa*.

H. bacciferum	Extract Conc. (%)	Total chlorophylls (mg/100g.F.wt)	Total soluble sugars (mg/100g.F.wt)	Proline (µmol g-1 FW)
Root	0	7. 35±1.75*	258±.29	48±0.92*
	10	5.42±0.16*	120±0.31**	60±1.53*
	25	4.11±0.73*	95 ±0.52*	73±0.20**
	50	2.50±1.75*	70 ±0.43**	100±0.39
	75	1.89±0.16	33±1.14**	133±0.81*
Leaf	0	8.01±1.75**	300±.29	48±0.92*
	10	4.83±.0.37*	180 ±1.53*	83±0.19*
	25	2.22±0.09*	115 ±0.39*	110±0.20**
	50	1. 00±0.10*	83± 0.49**	180±0.73*
	75	0.65±0.65*	60±2.32	192±1.14*

* Indicate significant differences between treatments at P<0.05.

Table 4: Effects of heliotrope leaf and root aqueous extracts on total chlorophylls total soluble sugars and proline of *T. polium*.

H. bacciferum	Extract Conc. (%)	Total chlorophylls (mg/100g.F.wt)	Total soluble sugars (mg/100g.F.wt)	Proline (µmol g-1 FW)
Root	0	4.50±0.42*	198±0.20	24±0.30*
	10	3.11±0.03*	80±.0.61*	33±0.73**
	25	2.48±±0.33*	66±1.14*	47±0.06
	50	1.99±0.73*	32±0.29	56±0.10
	75	0.66±0.03**	14±0.92	80±0.20**
Leaf	0	5.52±0.42*	210±0.15*	24±0.30*
	10	2. 33±0.10*	104±0.09**	49±0.08
	25	1.60±0.09*	75±0.12*	61±0.03*
	50	0.84±0.03	52±0.38	92±0.20
	75	0.23±0.42**	20±0.81*	140±0.13**

* Indicate significant differences between treatments at P<0.05.

Mahmoodzadeh and Mahmoodzadeh, 2013). Increase the extract concentration significantly ($p<0.05$) increased the inhibitory effect on germination of two tested species compared with the control (Tables 1 and 2). This finding is identical with the results of Chung and Miller (1995) who found that the degree of inhibition increased with increasing extract concentration. The data revealed that leaf extracts exhibited the greatest inhibition at all concentrations, while root extracts was the least inhibitory (Figure 1 and 2). The results found in this study are consistent with those of Turk and Tawaha (2002) who reported that leaf extracts of black mustard is the most inhibitory, while root extract is the least inhibitory the test species is lentil (*Polygala tatarinowii* Regel.). Furthermore, Hossain et al., (2012) reported that the rate of germination of *Vigna radiata* decreased with the increasing concentration of aqueous extract. Tanveer et al., (2008) reported that germination rate in maize, barley, rice, wheat and sunflower was reduced by leaf leaches of *Xanthium strumarium*. The germination was delayed for one and two days in the last three treatments and 75% respectively. These results are in agreement with those of Jalal et al., (2013) who stated that seed germination and plant growth delayed at the higher concentrations, and the final germination percentage was decreased by increasing leaf extract concentration of *X. strumarium*.

This study demonstrated that all heliotrope aqueous extracts, especially root and leaf extracts inhibited seedling growth of two tested plant species (Tables 1 and 2). Oudhia (1999) found that extracts of some weeds as *Calotropis gigantea* has caused allelopathic effects inhibited germination and growth of *Lathyrus sativus*. The radicle length was found to be more sensitive to all types of heliotrope aqueous extract in comparison to the other parameters measured. These results are in agreement with those of Turk and Tawaha (2002) who reported that leaf extracts of black mustard is the most inhibitory, while root extract is the least inhibitory the test species is lentil (*Polygala tatarinowii* Regel.). Moreover, Randhawa et al., (2002) reported that root length of *Trianthema portulacastrum* was affected by sorghum water extract and significantly reduced by high concentration of 75 and 100% sorghum water extract. Similarly, Gulzar et al., (2014) reported that the root length and shoot length of weed species decreased significantly when plants were exposed to increasing aqueous concentration of *Cassia sophera*. Between the two tested plant species, a greater inhibitory effect was observed on *T. polium* than on *O. sativa* which suggests species specificity.

Radicle and plumule are the first organs emerged from the seed and exposed to allelopatic matter since it can be predictable that their growth decrease when they expose to allelopathic compounds. Increase in extract concentration significantly decreased the length of plumule and put a stop on it at the highest level of extract (75%). Dongre and Yadav (2005) found inhibition in the length of plumule and radicle, a reduction in their dry weights and total seedling weights in wheat, pea and lentil with water extracts of various weeds.

Oueslati (2003) and Turk et al., (2003) showed similar observations on other plant species. Nasrine et al., (2011) stated that the germination efficiency, plumule and radicle length of Bromus was completely inhibited at the highest concentration of aqueous extracts of the donor species level (10%).

Decrease in plumule length can cause by inhibiting in cell division and elongation and or decreasing in hormones such as acetic acid and gibberellin .Consequently these may cause short and weakness seedlings and then decrease the establishment of them. Anaya (1999) stated that a decrease in plumule and radicle length might be because of decreasing in cell division. Several enzymes like proteases, lipases and a-amylases play an important role during seed germination. Many enzymatic functions are inhibited by the presence of allelochemicals (Turk and Tawaha, 2002; Rice, 1984). Seeds proteases play an important role in the hydrolysis of proteins during germination. To a large extent the activity of these enzymes is primarily related to water imbibition by the seeds. Although enzyme activity was not investigated in this study, an indirect association between lower seed germination and allelopathic inhibition may be the consequence of the inhibition of water uptake and enzyme activity (Turk and Tawaha, 2003).

The biochemical constituents, total chlorophyll and total sugar contents of both *O. sativa* and *T. polium* were significantly reduced in the all treatment declined at higher concentrations of extract. Reduction in chlorophylls may decrease the photosynthesis and thereby substantially decrease all the metabolites viz., total sugars, proteins and soluble amino acids (Singh and Rao, 2003). The reduction in chlorophyll contents observed in all the concentrations might be due to degradation of chlorophyll contents or reduction in their synthesis (a and, b) the action of flavanoids trepenoids or other phytochemicals present in leaf leachates (Tripathi et al., 1999, 2000). This finding is supported by many investigators (Kavitha et al, 2012; Gonzalez et al. 1997 and Einhellig et al., 1993) who stated that a decrease in that a decrease in photosynthesis efficiency is a common effect of allelopathic phenolics.

Einhellig et al., (1993) and Gonzalez et al., (1997) confirmed that the root exudates of Sorghum bicolor inhibited the oxygen evolution of soybean leaf disk and isolated pea chloroplast, which interfere in photosystem II electron transfer reaction and consequently growth reduction. Rice (1984) has suggested that the synthesis of porphyrin precursors of chlorophyll biosynthesis may be impeded by some allelopathic compounds. Allelochemicals may enhance the activities of enzymes such as chlorophyllase and Mg-dechelatase, responsible for the Chl derivative pathway (Yang et al., 2004).

The reduction of soluble sugar content in both *O. sativa* and *T. polium* in agreement with Mohamadi and rajaie (2009) who found that the chlorophyll and total sugar contents of eucalyptus were significantly reduced to the increase in concentration of leaf leachate in both *Phaseolus vulgaris* and *S. bicolor*. Previous studies conducted by El-Khawas and Shehata (2005) also established that phenolics present in the leaf extracts of *Acacia nilotica* inhibited the leaf sugar content of maize leaves in pot experiments.

The free proline content in both *O. sativa* and *T. polium* was increased, this indicating that, the chemical stress is induced by leaf and root of heliotrope. Pawar and Chavan (2004) also observed the similar type of result in sorghum. Proline protects proteins from denaturation by maintaining the hydration level. Abdulghadar and Nabat (2008) reported that with application of heliotrope leaves extracts, level of proline significantly increased in leaves of Dodder. The high level of proline may be protecting plants from stress conditions (Kumar et al., 2003).

This experiment was conducted in laboratory condition therefore it suggests that more research could be carried out in greenhouse condition because in natural condition the results may change as a result of differences in growth conditions. It also suggested that more investigation about the allelopathic effect of this species should be carried out on the other species.

5. Conclusion

Heliotrope (mainly leaves and root) an allelopathic plant. It is potentially an inhibitor of seed germination and seedling growth of crop (*O. sativa*) and weed (*T. polium*) that are commonly intercropped with heliotrope. Therefore, heliotrope must be considered as an allelopathic species posing risk in a rotation or an intercropping or mixed cropping system. Also, we propose that, the active compound that has allelopathic potential and herbicidal activity to be isolated and characterized in the future.

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