American-Eurasian J. Agric. & Environ. Sci., 7 (3): 298-302, 2010
ISSN 1818-6769

Allelopathic Effects of Spurge (*Euphorbia hierosolymitana*) on Wheat (*Triticum durum*)

[1]*Saeid Abu-Romman,* [1]*Mohammad Shatnawi and* [2]*Rida Shibli*

[1]Department of Biotechnology, Al-Balqa' Applied University, Al-Salt 19117, Jordan
[2]Department of Horticulture and Crop Science, University of Jordan, Amman, Jordan

Abstract: This study was conducted to investigate the allelopathic potential effect of *Euphorbia hierosolymitana* in wheat (*Triticum durum* local var. Hourani 27), in both laboratory and glasshouse. Since *E. hierosolymitana* is a common weed in filed crops and orchards in Jordan. The effects aqueous leachate of *E. hierosolymitana* on germination, seedling growth, total chlorophyll and protein contents of wheat were examined. Higher c oncentration of aqueous leachate of *E. hierosolymitana* reduced the germination rate. On the other hand, the radicle and coleoptile length of the germinated seeds of wheat were significantly inhibited by the leachate. Also, the aqueous leaf leachate of *E. hierosolymitana* was found to inhibit significantly the growth of wheat seedlings. Allelochemicals caused significant reduction in decreased root and shoot length, fresh, dry weights and decreased the amount of total chlorophyll and protein contents.

Key words: Allelopathy · Aaqueous leachate · *Euphorbia hierosolymitana* · Wheat

INTRODUCTION

Wheat (*Triticum durum*) is the most grown crop in the world and has an economical significance for the humankind [1]. Residues and leachate of several crops and weeds have been shown to possess negative impacts on wheat growth and yield. Ben- Hammouda *et al.,* [2] reported that some barley cultivars are phytotoxic to durum wheat (*T. durum*) and bread wheat (*T. aestivum*). In comparison of numerous species of genus *Brassica*, Mason-Sedum *et al*., [3] reported that water extracts from residues of the genus *Brassica* significantly reduced root and coleoptile length of wheat with little effect on germination. In bioassay experiments, Bialy *et al*., [4] showed that inhibition of wheat seed germination occurred upon treatment with 2-phenetyl ITC, a known allelochemical derived from species of the genus *Sinapsis*. Roth *et al.,* [5] found that prompt tillage of sorghum stalks delayed development and reduced grain yields of wheat.

In allelopathy, the chemical inhibition of one plant species by another, represents a form of chemical warfare between neighbouring plants competing for limited light, water and nutrient resources. Allelopathy is believed to be involved in many natural and manipulated ecosystems and plays a role in the evolution of plant communities, exotic plant invasion and replant failure [6, 7]. Most plant species, including crops, are capable of producing and releasing biologically active compounds (allelochemicals) into the environment to suppress the growth of other plants.

Allelochemicals are toxic (e.g. phenolics, terpenoids and alkaloids and their derivatives) and may inhibit shoot/root growth, nutrient uptake, or may attack a naturally occurring symbiotic relationship thereby destroying the plant's usable source of a nutrient. The consequent effects may inhibited or retarded germination rate, reduced root or radicle and shoot or coleoptile extension, lack of root hairs, swelling or necrosis of root tips, curling of the root axis, increased number of seminal roots, discolouration, reduced dry weight accumulation and lowered reproductive capacity [8]. Allelochemicals are considered to be secondary metabolites or waste products of the main metabolic pathways in plants and they do not appear to play a role in the primary metabolism essential for plant survival [9, 10].

Allelopathy effects have been noted for several Euphorbiacae. Aqueous extracts of stems and leaves leafy spurge (*Euphorbia esula*) inhibited radicle elongation and germination of several test species [11]. On the other hand, Hussain [12] reported the allelopathic effect of *Euphorbia granulata* on different plant species (both weeds and cultivated plants). Therefore, the present study was conducted to investigate the allelopathic effects of *E. hierosolymitana* on wheat in terms of germination, seedling growth, chlorophyll and protein content.

Corresponding Author: Dr. Saeid Abu-Romman, P.O. Box 317, Amman 11910, Jordan. E-mail. ssadroman@yahoo.com

MATERIALS AND METHODS

Allelopathic Plant and Preparation of Leachate: *E. hierosolymitana* plants were planted in the greenhouse obtained from seed, 100 g leaves of *E. hierosolymitana* plants, harvested at the vegetative stage, were soaked in 100 ml of distilled water with stirring for 24 h. The collected leachate was filtered through cheese cloth to remove debris and finally filtered using Whatman No. 1 filter paper to have 100% concentration. The leachates of 25, 50 and 75% concentrations were made by diluting the parent leachate with distilled water.

Seed Germination and Seedling Growth: Ten uniform seeds (2% sodium hypochlorite for 15 min and then washing in doubled distilled water thoroughly) of wheat (*T. durum* local var. Hourani 27) were germinated in sterilized petri-dishes lined with two layers of Whatman No. 1 filter paper and moistened with 5 mL of respective leaf leachate concentration in treatment and distilled water in control. Each treatment had eight replicates and each experiment was repeated twice. The petri-dishes were incubated at 25±2°C in growth chamber. After three days of culture, germination percentages were recorded and, the radicle and coleoptile length were measured.

Soil Culture: In another set of experiment, seeds were surface sterilized as mentioned above. Five seeds were sown at equal distance in experimental pots (15 cm in diameter, 25 cm in height) filled with homogenous soil. The experiment was conducted in the glasshouse. Eight replicates were assigned for each treatment and the experiment was repeated twice. The pots were irrigated every two days with 100 ml of graded concentration of leachate according to the treatments. The control pots were irrigated with distilled water. After three weeks, data were recorded for root and shoot length, fresh and dry weights. For dry weight determination, tissues were dried at 70°C for 24 h. Chlorophyll from leaves were extracted with 80% acetone and quantified following the procedure of Danilov and Ekelund [13]. Protein content was determined following the method of Bradford [14]. The amount of protein was calculated with reference to standard curve obtained from bovine serum albumin.

Statistical Analysis: Data were subjected to one-way ANOVA; differences between individual means were determined by the least significant difference (LSD) test at the 0.05 level of probability. Experimental design was performed as a randomized complete block design; each experiment was consisting of five treatments. Data were analyzed using SAS program [15].

RESULTS

Seed Germination and Seedling Growth: Germination, length of radicle and coleoptile were recorded after three days of culture. Leachte of *E. hierosolymitana* leaves showed inhibitory effects on seed germination and seedling growth of wheat grown in petri dishes. Leachate of *E. hierosolymitana* inhibited wheat seed germination compared with the control (Table 1). The control treatment produces the highest germination rate (96.3%). Using 100% leachate, germination reduced to 30%. Higher concentration of leachate exhibited inhibitory effect on radicle and coleoptile length. At 25% leachate, the radicle and coleoptile length were reduced to nearly (31%) and (34%), respectively (Table 1). Length of radicle and coleoptile length was inhibiting at 100% concentration (Table 1), where radicle and coleoptile length dropped to reach 1.05 cm and 0.47 cm, respectively.

Soil Culture: Leachates of *E. hierosolymitana* caused significant alteration in growth of wheat seedlings. At higher concentrations of leachate, allelochemicals gradually decreased in root length as well as root fresh and dry weights. The shortest roots and the lowest root fresh and dry weights were recorded at the 100% concentration (Table 2). Wheat shoot growth were significantly affected by the allelochemicals.

Table 1: Effect of aqueous leaf leachate of *E. hierosolymitana* on germination and seedling growth of *T. durum*

Treatments	Germination %	Radicle Length (cm)	Coleoptile Length (cm)
0	96.3 a	2.55 a	1.22 a
25	85.5 b	1.75 b	0.81 b
50	82.4 bc	1.54 bc	0.73 bc
75	74.5 cd	1.12 cd	0.51 c
100	67.5 d	1.05 d	0.47 c

Mean values followed by the same letters within each column are not significantly different according to LSD (P =0.05) n = 8. C, control; 25, 50, 75 and 100 denote the concentration of *Euphorbia* leachate

Table 2: Effect of aqueous leaf leachate of *E. hierosolymitana* on root growth of *T. durum* seedlings grown under glasshouse conditions for three weeks

Treatments	Root length (cm)	Root fresh weight (g)	Root dry weight (g)
0	9.34 a	0.302 a	0.0237 a
25	8.79 a	0.280 b	0.0222 b
50	8.56 b	0.216 c	0.0178 c
75	7.74 c	0.206 d	0.0168 c
100	4.90 d	0.170 e	0.0145 d

Mean values followed by the same letters within each column are not significantly different according to LSD (P =0.05) n = 8. C, control; 25, 50, 75 and 100 denote the concentration of *Euphorbia* leachate

Table 3: Effect of aqueous leaf leachate of *E. hierosolymitana* on shoot growth of *T. durum* seedlings grown under glasshouse conditions for three weeks

Treatments	Shoot length (cm)	Shoot fresh weight (g)	Shoot dry weight (g)
0	16.52 a	3.31 a	0.290 a
25	15.24 a	3.24 a	0.278 b
50	11.16 b	3.05 b	0.260 c
75	7.35 c	2.87 c	0.242 d
100	5.15 c	2.65 d	0.215 e

Mean values followed by the same letters within each column are not significantly different according to LSD (P =0.05) n = 8. C, control; 25, 50, 75 and 100 denote the concentration of *Euphorbia* leachate

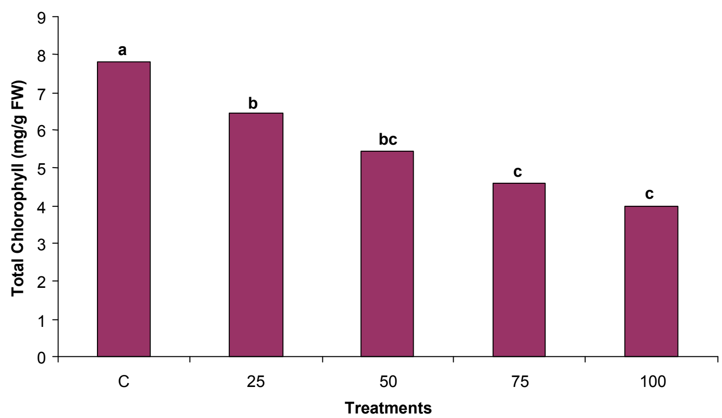

Fig. 1: Effect of aqueous leaf leachate of *E. hierosolymitana* on total chlorophyll content of *T. durum* seedlings grown under glasshouse conditions for three weeks. Values with the same letters are not significantly different according to LSD (P =0.05) n = 8 C, control; 25, 50, 75 and 100 denote the concentration of *Euphorbia* leachate

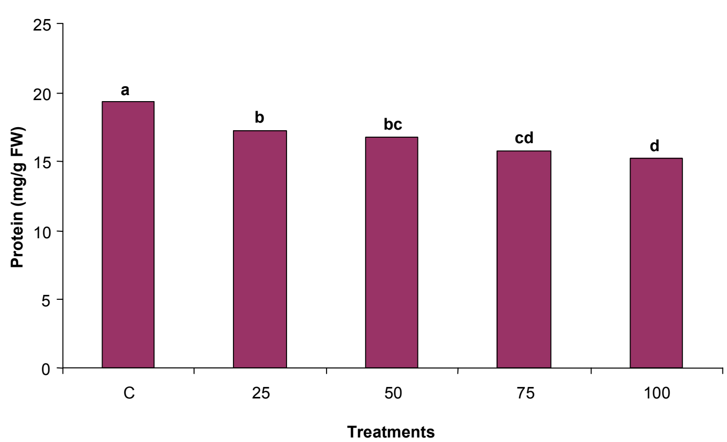

Fig. 2: Effect of aqueous leaf leachate of *E. hierosolymitana* on protein content of *T. durum* seedlings. grown under glasshouse conditions for three weeks. Values with the same letters are not significantly different according to LSD (P =0.05) n = 8. C, control; 25, 50, 75 and 100 denote the concentration of *Euphorbia* leachate

The shortest shoots were noticed at 75 or 100%. Leachate concentration above 25%, resulted in significant reduction in shoot fresh weight. Treated wheat with 100% leachate concentration resulted in a significant reduction in shoot fresh weight. Shoot dry weight decreased significantly when leachest concentration increased, the lowest shot dry weight (0.215 g) was recorded at 100% leacheat concentration (Table 3). A decrease in total chlorophyll content of wheat seedlings under allelochemical stress was occurred. The control treatment gave significantly the highest total chlorophyll content (7.8 mg/g FW). The chlorophyll content was reduced by 41% and 48%, when wheat seedlings were treated with 75 and 100% leachate, respectively (Fig. 1). Leachate decreased protein content of wheat seedlings. The control gave significantly the highest protein content (19.4 mg/g FW) as compared to other treatments. Protein content of wheat seedling dropped to reach 15.3 mg/g FW at 100% leachate (Fig. 2).

DISCUSSION

The results of the present study indicated that the allelopathic of *E. hierosolymitana* reduced germination percentages, radicle and coleoptile growth of wheat seedlings. This result is similar to previous finding on wheat by Bhatt *et al.,* [16] were the bark, leaf and leaf extract of *Quercus glauca* and *Q. leucotricophora* significantly reduced germination, plumule and radicle length of wheat (*Triticum* sp.).

An indirect relation between lower germination rate and allelopathic inhibition may be the consequence of inhibition of water uptake [17] and alteration in the synthesis or activity of gibberellic acid (GA_3) [18]. This may be due to the presence of phenolic compounds inhibit the activity of GA_3 [19] or inhibit the synthesis of GA_3 which regulate de novo amylase production during seed germination [20]. Impaired metabolic activities caused by allelochemicals decreased root and shoot length.

Allelochemicals decreased elongation, expansion and division of cells which are growth prerequisite [19]. Also, allelochemicals inhibit absorption of ions [21] and therefore, resulted in arrested growth [22]. Thus the reduction in total chlorophyll content under allelopathins was recorded. This is similar to previous findings obtained by Venkateshwarlu *et al.,* [23] in radish seedlings under mango leaf leachate treatment. Bagavathy and Xavier [24] also reported the reduction in chlorophyll a, chlorophyll b and total chlorophyll in sorghum plants when treated with *Eucalyptus* leaf extract. However, the reduction of chlorophyll under leachate treatments could be attributed to the inhibition of chlorophyll biosynthesis and/or the stimulation of chlorophyll degradation [25]. Rice [26] has suggested that allelopathic compounds impede the synthesis of porphyrin precursors of chlorophyll biosynthesis. Romagni *et al.,* [27] reported that lichen metabolite, usnic acid; irreversibly inhibit the activity of protoporphyrinogen IX, the enzyme involved in the chlorophyll biosynthesis pathway. Moreover, Latakowska *et al.,* [28] found that usnic acid can accelerate the degradation of chlorophyll. Higher degradation rate of chlorophyll b or controlled conversion of chlorophyll a to chlorophyll b in leaves of maize seedlings treated with higher concentration of *Nicotiana* leachate resulted in increase of chlorophyll a/b ratio [29]. This can be explained by the fact that the first step in chlorophyll b degradation involves its conversion to chlorophyll a [30].

Allelochemicals decreased the amount of protein in wheat seedlings. Impairment of various metabolic activities under the influence of leachate inhibited the protein synthesis and/or stimulated the degradation [31]. Normal ways of protein synthesis is inhibited in lettuce seedlings (*Lactua sativa*) when treated with cinnamic acid [19]. In conclusion, the present study showed that aqueous leachate of *E. hierosolymitana* delayed the germination, reduced the seedling growth, total chlorophyll and protein content.

REFERENCES

1. Löve, A., 1984. Conspectus of the Triticaceae. Feddes Repertorium, 95: 425-521.
2. Ben-Hammouda, M., H. Ghorbal, R.J. Kremer and O. Oueslati, 2001. Allelopathic effects of barley extracts on germination and seedlings growth of bread and durum wheat. Agronomie., 21: 65-71.
3. Mason-Sedun, W., R.S. Jessop and J.V. Lovett, 1986. Different phytotoxicity among species and cultivars of the genus *Brassica* to wheat. I. Laboratory and field screening of species. Plant Soil, 93: 3-16.
4. Bialy, Z., W. Oleszek, J. Lewis and G.R. Fenwick, 1990. Allelopathic potential of glucosinolates (mustard oil glycosides) and their degradation products against wheat. Plant Soil, 129: 277-281.
5. Roth, C.M., J.P. Shroyer and G.M. Paulsen, 2000. Allelopathy of sorghum on wheat under several tillage systems. Agron. J., 92: 855-860.

6. Ridenour, W.M. and R.M. Callaway, 2001. The relative importance of allelopathy in interference: the effects of an invasive weed on a native bunchgrass. Oecologia, 126: 444-450.
7. Inderjit, D.S.O., 2003. Ecophysiological aspect of allelopathy. Planta, 217: 529-539.
8. Ayeni, A.O., D.T. Lordbanjou and B.A. Majek, 1997. *Tithonia diversifolia* (Mexican sunflower) in South Western Nigeria; occurrence and growth habit. Weed Res., 37: 443-449.
9. Swain, T., 1977. Secondary compounds as protective agents. Ann. Rev. Pl. Physiol., 28: 479-501.
10. Chon, S.U. and J.D. Kim, 2002. Biological activity and quantification of suspected allelochemicals from alfalfa plant parts. J. Agron. Crop Sci., 188: 281-285.
11. Steenhagen, D.A. and R.L. Zimdahl, 1979. Allelopathy of leafy spurge (*Euphorbia esula*) Weed Sci., 27: 1-3.
12. Hussain, F., 1980. Allelopathic effects of Pakistani weeds: *Euphorbia granulate* Forssk. Oecologia, 45: 267-269.
13. Danilov, R.A. and N.G.A. Ekelund, 2001. Effects of Cu^{2+}, Ni^{2+}, Pb^{2+}, Zn^{2+} and pentachlorophenol on photosynthesis and motility in *Chlamydomonas reinhardtii* in short term exposure experiments. BMC Ecology. 1: 1.
14. Bradford, M.M., 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal. Biochem., 72: 248-254.
15. SAS Institute Inc. 1988. The SAS System for Windows Release 6.12. SAS Institute Inc, Cary, NC.
16. Bhatt, B.P., D.S. Chauhan and P. Todaria, 1994. Effect of weed leachate on germination and radicle extension of some food crops. India. J. Plant Physiol., 36: 170-177.
17. Tawaha, A.M. and M.A Turk, 2003. Allelopathic effects of black musard (*Brassica nigra*) on germination and growth of wild barley (*Hordeum spontaneum*). J. Agron. Crop. Sci., 189: 298-303.
18. Olofsdotter, M., 2001. Rice-A Step Toward Use of Allelopathy. Agron. J., 93: 3-8.
19. Einhellig, F.A., 1996. Mechanism of action of allelochemicals in allelopathy. Agron. J., 88: 886-893.
20. Chandler, P.M., J.A. Zucar, J.V. Jacobson, T.J.V. Higgins and A.S. Inglis, 1984. The effect of gibberellic acid and abscisic acid on a-amylase mRNA levels in barley aleurone layers studies using an α-amylase cDNA clone. Plant Mol. Biol., 3: 407-408.
21. Qasem, J.R. and T.R. Hill, 1989. Possible role of allelopathy in competition between tomato, *Senecio vulgaris* L. *Chenopodium album* L. Weed Res., 29: 349-356.
22. dos Santosh, W.D., M.L.L. Ferrarese, A. Finger, A.C.N. Teixeira and O. Ferrarese-Filho 2004. Lignification and related enzymes in Glycine max root growth-inhibition by ferulic acid. J. Chem. Ecol., 30: 1199-1208.
23. Venkateshwarlu, G., V. Ravindra and C. Prabha, 2001. Mangiferin: an allelopathin from mango (*Mangifera indica* L.) leaves. Allelopathy J., 8: 221-224.
24. Bagavathy, S. and G.S.A. Xavier, 2007. Effects of aqueous extract of *Eucalyptus globules* on germination and seedling growth of sorghum. Allelopathy J., 20: 395-402.
25. Yang, C.M., I.F. Chang, S.J. Lin and C.H. Chou, 2004. Effects of three allelopathic phenolics on chlorophyll accumulation of rice (*Oryza sativa*) seedlings: II. Stimulation of consumption-orientation. Bot. Bull. Acad. Sin., 45: 119-125.
26. Rice, E.L., 1984. Allelopathy, 2nd edn. Academic Press, Orlando.
27. Romagni, J.G., G. Meazza, N.P.D. Nanayakkara and F.E. Dayan, 2000. The phytotoxic lichen metabolite, usnic acid, is a potent inhibitor of plant p-hydroxyl phenyl pyruvate dioxygenase. FEBS Lett., 480: 301-305.
28. Latakowska, E., Z. Lechowski, J. Bialczyk and L. Pilarski, 2006. Photosynthesis and water relations in tomato plants cultivated long term in media containing (+)-usnic acid. J. Chem. Ecol., 32: 2053-2066.
29. Singh, A., D. Singh and N.B. Singh, 2009. Allelochemical stress produced by aqueous leachate of *Nicotiana plumbaginifolia* Viv. Plant Growth Regul., 58: 163-171.
30. Fang, Z., J.C. Bouwkamp and T. Solomas, 1998. Chlorophyllase activities and chlorophyll degradation during leaf senescence in non yellowing mutant and wild type of *Phaseolus vulgaris* L. J. Exp. Bot., 49: 503-510.
31. Mersie, W. and M. Singh, 1993. Phenolic acids affect photosynthesis and protein synthesis by isolated leaf cells of velvet-leaf. J. Chem. Ecol., 19: 1293-1301.