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Allelopathic Effects of Water Extract of Artemisia Princeps Var. Orientalis on Wheat under Two Type of Soils

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Abstract: Study was conducted in the greenhouse of Biology Department at the College of Science, Taif University. The allelopathic effect of *Artemisia princeps var. orientalis* was measured in terms of germination rate and radical length of a bread wheat variety 'Ariana' (*Triticum aestivum* L.) in sandy and silty soil. Diluted extracts of *Artemisia princeps* shoot system was applied to seeds of the test plant. The allelopathy of *Artemisia* species varied with the concentration of the extracts. The concentrated extracts (75 and 100%) of *A. princeps* were phytotoxic to germination of wheat in the two soils. The reduction in the germination percentage reached to 20-35% by treatment with *A. princeps* at conc. of 75 and 100% at sandy soil, while at silty soil reduction reached to 6-15% respectively after 8 days. The aqueous extract of *A. princeps* at 25% caused an increase in the root and shoot growth of the wheat seedlings at sandy soil comparison with control. However the growth of root and shoot gradually decreased with the increase extract concentration (50-100%). The biomass production paraled with that of root and shoot lengths as activated at low concentration (in sandy soil) and inhabited at high ones. The inhibitive effect of *A. princeps* extracts were proportional to their concentration increase. Allelopathy might be reduced by prompt tillage and other practices that promote rapid decomposition of *A. princeps* residue in silty soil. Allelopathy as a mechanism and future strategy for agricultural pest control and farm management and the potential use and development of some allelochemicals as natural pesticides or plant growth regulators are also considered and discussed.

Key words: *Triticum aestivum* · *Artemisia princeps var. orientalis* · allelopathy · growth creature · chlorophyll · sand and silt soil

INTRODUCTION

Allelopathy is an important mechanism of plant interference mediated by the addition of plant-produced phytotoxins to the plant environment and competitive strategy of plants [1]. There is no denying that allelopathy plays a prominent role in ecology and evolution of plant communities. However, its pervasive interacting nature intrigues us as well as challenges us as scientists to dig deeper into the understanding of its mechanism of action. Working on this challenge will lead to new discovery that will keep us excited to learn more and gain a better understanding of the phenomenon. Equipped with this new knowledge and understanding, we should be able to solve many difficult environmental problems of our time.

Allelochemicals are released from plant tissue in a variety of ways including emission of volatile substances from living plant parts, exudation from roots, or leaching from above ground parts by rain, dew, fog, etc. [2]. The allelopathic effect may be so striking that competition for resources does not explain why, in plant communities, many species appear to regulate one another through the production and release of chemical attractants, stimulators or inhibitors [2]. Several crops are known to have allelopathic effects on other crops, e.g. Beet (*Beta vulgaris* L.), Lupin (*Lupinus lutens* L.), maize (*Zea mays* L.), wheat (*Triticum aestivum* L.), oats (*Avena sativa* L.) and barley (*Hordeum vulgare* L.) [3]. Moreover allelopathic effect of *Avena fatua* L., *Cyprus rotundus* L., *Polygonum hydropiper* L. and *Solanum nigrum* L. were examined on seedling growth of certain commonly used varieties of *Triticum aestivum* L. [4]. Roth, *et al.* [5] showed that water extracts from sorghum residues inhibited germination and decreased root and shoot growth of corn (*Zea mays* L.) and wheat.

Khanh *et al.* [6] found that inhibitory substances involved in allelopathy are mainly terpenoids and phenolic substances. A wide array of biologically active

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constituents is produced by plants in the genus *Artemisia* [7]. Bertholdesson, [8] showed that volatile oil of *Artemisia afra* have several biological activities, notably antibacterial, antifungal and anti-oxidative properties. Monoterpene vapors may cause anatomical and physiological changes in plant seedlings and exposure to volatile terpens can lead to accumulation of lipid globules in the cytoplasm, reduction in organelles including mitochondria and disruption of membranes surrounding mitochondria and nuclei [9]. The root tip cells subjected to the alkaloids gramine and hordenine caused damages to the cell walls, disorganization of organelles, increase cell vacuoles and the appearance of lipid and globules, showing food reserves [10]. Large amounts of monoterpene hydrocarbons and/or sesquiterpenes are found to lower the antimicrobial activity of essential oils [11].

Management practices can reduce residues on the soil surface and decrease the impact of phytotoxin concentrations from the residues in the soil and depending on soil type [7]. Five new monoterpenes and seven new sesquiterpene lactones, have allopathic effects, were isolated from the aerial parts of *Artemisia suksdorfii* [12].

The goal of the present research to determine the influence of *Artemisia princeps var. orientalis* water extract on wheat (*Triticum aestivum* L) germination through, growth criteria and chlorophyll in two type of soils.

MATERIALS AND METHODS

Artemisia princeps was collected from the desert plains at Taif area and sampled in March 2008. Whole plants were pulled out of the field at the stage of flowering development. The plants were gently washed with distilled water, dried between two paper towels. The plant materials were chopped into 1 cm long pieces and dried at 50°C for 24 h. The components were soaked in distilled water for 24 h at the rate of 5 g fresh weight per 100 ml distilled water. Extract was filtered through four layers of cheese cloth, Whitman filter paper under vacuum and stored at <5°C. Dilutions were made of the original extracts as, 100, 75, 50 and 25% of the origin extracts. Two types of soils were used in pots, sandy soil (85% sand, 10% Silt and 5% clay) and silty soil (35% sand, 55% Silt and 10% clay). Twenty seeds were placed in each pot at 25°C in the greenhouse at Biology Department Taif University, S.A. The experimental design was a complete randomized design with four replications pots as:

- C = Pots irrigated with dist H_2O
- SA_1 = Pots irrigated with *Artemisia* extract, 25% in sandy soil
- SA_2 = Pots irrigated with *Artemisia* extract, 50% in sandy soil
- SA_3 = Pots irrigated with *Artemisia* extract, 75% in sandy soil
- SA_4 = Pots irrigated with *Artemisia* conc., extract 100% in sandy soil
- SiA_1 = Pots irrigated with *Artemisia* extract, 25% in silty soil
- SiA_2 = Pots irrigated with *Artemisia* extract, 50% in silty soil
- SiA_3 = Pots irrigated with *Artemisia* extract, 75% in silty soil
- SiA_4 = Pots irrigated with *Artemisia* conc., extract 100% in silty soil

After 2, 4, 6 and 8 days germination counts, root and shoot lengths were measured and recorded. Seeds were considered germinated when the radical extended through the seed coat. Relative water content was also measured at day 8 and expressed as a percentage according to the following equation:

$$RWC (\%) = (FW - DW)/FW \times 100.$$

Chlorophyll content was determined, at 8 days after germination. It was extracted by homogenizing 0.5 g fresh weight of green tissues (leaf plus stem) of the seedlings in 10 ml of 95% ethanol. After centrifugation for 10 min. at 5000 rpm, the chlorophyll content was analyzed spectrophotometrically on the ethanolic supernatant at 654 nm as described Einhellig and Rasmussen [13]. Absorbance was measured with a Hitachi U2000 UV-visible spectrophotometer.

Statistical analysis: Results indicated as mean values±SEM, differences between control and treated seeds were analyzed by one–way ANOVA, taking $P<0.05$ a significant, according to Tukey s Multiple Range Test (MRT).

RESULTS

Artemisia princeps var. orientalis shoot extracts significantly affected germination of wheat seeds (Table 1) in the two type of soil. Results obtained revealed that *A. princeps* allelopathy takes the form of heterotoxicity, depressive to wheat. The germination

Table 1: Effect of diluted extracts of *Artemisia princeps* shoots on germination (%) of bread wheat at sand and silt soils

	Time (days)			
Treatments	2	4	6	8
C	58±5	96±6	100±6	100±6
SA1	53±5	88±5	90±6	92±6
SA2	44±4**	65±5**	80±5*	88±6*
SA3	34±3***	51±4***	74±5**	80±6**
SA4	22±2***	34±3***	46±4***	65±6***
LSD, SA (5%)	1.2	0.65	0.55	0.43
SiA1	54±5	90±6	95±6	100±6
SiA2	50±4	84±5*	90±6	98±6
SiA3	43±3**	79±5*	85±5	94±6
SiA4	36±3**	65±5**	78±5**	85±6*
LSD, SiA (5%)	1.56	0.95	0.75	0.75

Means within a column followed by different number of stars are significantly different according to Fisher's:
*=Significant at P< 0.5 **=Significant at P< 0.1 ***=Significant at P< 0.05

Table 2: Effect of diluted extracts of *Artemisia princeps* shoots on root and shoot lengths (cm) of bread wheat at sand and silt soils, day 8

Treatments	Root	Shoot
C	4.9±0.4	14.5±0.9
SA1	5.1±0.5	15.2±1.0
SA2	3.5±0.3*	12.5±0.4*
SA3	2.3±0.2**	8.5±0.6***
SA4	1.6±0.2***	6.7±0.4***
LSD, SA (5%)	0.26	0.92
SiA1	5.0±0.5	14.2±1.0
SiA2	4.5±0.4	13.5±0.7
SiA3	4.0±0.3*	12.6±0.7*
SiA4	3.4±0.3*	10.7±0.6*
LSD, SiA (5%)	0.36	1.2

Means within a column followed by different number of stars are significantly different according to Fisher's: *=Significant at P< 0.5 **=Significant at P< 0.1 ***=Significant at P< 0.05

percentage of wheat grain decreased more than 50 and 35% due to extract treatment at concentration of A_4 and A_3 respectively after 2 days. The reduction decreased to 35 and 20% after 8 days in sandy soil. While at silt soil the reduction reduced to 15 and 6% in day 8 at A_4 and A_3 respectively. Treatment with A_1 extract had un-significant effect on the germination of bread wheat compared with control in the two soil. In general the reduction effects of *A. princeps* extract on the germination percentage of wheat grains increase with the increasing in concentrations of extracts.

Table 3: Effect of diluted extracts of *Artemisia princeps* shoots on water content (W.C) and degree of succulence (D.Su.) of bread wheat seedlings root and shoot at sandy and silty soils after 8 days

	Root		Shoot	
Treatments	W.C (%)	D. Su.	W.C (%)	D. Su.
C	66.03	2.94	70.93	3.44
SA1	55.17*	2.23*	70.83	3.43
SA2	75.00	4.00	85.90	3.72
SA3	69.70	3.30	88.89*	3.74*
SA4	63.64	2.75	88.24*	3.09*
LSD, SA (5%)	1.020	0.08	1.67	0.07
SiA1	60.78	2.55	74.44	3.91
SiA2	62.50	2.67	74.12	3.86
SiA3	65.12	2.87	72.97	3.70
SiA4	65.63	2.91	70.37	3.38
LSD, SiA (5%)	1.030	0.07	1.82	0.08

Means within a column followed by different number of stars are significantly different according to Fisher's: *=Significant at P< 0.5 **=Significant at P< 0.1 ***=Significant at P< 0.05

Table 4: Effect of diluted extracts of *Artemisia princeps* shoots on Fresh weight (F.W) and dry weight (D.W) of bread wheat seedlings at sandy and silty soils after 8 days

	Root (g/plant)		Shoot (g/plant)	
Treatments	F.W	D. W	F.W	D. W
C	0.053	0.018	0.086	0.025
SA1	0.058	0.026	0.096	0.028
SA2	0.044	0.011*	0.078	0.021
SA3	0.033**	0.01*	0.063	0.017*
SA4	0.022***	0.008**	0.034**	0.011**
LSD, SA (5%)	0.005	0.004	0.008	0.002
SiA1	0.051	0.02	0.09	0.023
SiA2	0.048	0.018	0.085	0.022
SiA3	0.043	0.015	0.074	0.02
SiA4	0.032**	0.011*	0.054	0.016*
LSD, SiA (5%)	0.006	0.005	0.01	0.002

Means within a column followed by different number of stars are significantly different according to Fisher's:
*=Significant at P< 0.5 **=Significant at P< 0.1 ***=Significant at P< 0.05

Root and shoot lengths, were measure under both normal and treated conditions (Table 2). The maximum shoot and root lengths as compared to control was estimated at 25% (SA_1) treatment at sandy soil, day 8. The reduction in the lengths reached to 60-75 % by treatment with *A. princeps* at SA_3 and SA_4 respectively. The root and shoot length of wheat seedlings were gradually

Table 5: Effects of diluted extract of *Artemisia princeps* shoots on chlorophyll content (ug/g F.W) of bread wheat seedling at sandy and silty soils

Treatments	Time (days) 2	4	6	8
C	88±5	212±11	326±16	564±21
SA1	93±6	238±15	490±18*	686±26
SA2	74±4*	176±7**	256±14**	420±16*
SA3	54±3**	151±4***	174±5***	280±6***
SA4	32±2***	94±4***	116±6***	165±8***
LSD, SA (5%)	1.2	0.65	0.55	0.43
SiA1	93±6	230±16	460±16	656±23
SiA2	84±4	210±11	324±14	512±19
SiA3	74±4*	191±9**	300±11*	470±13**
SiA4	62±3**	114±4***	216±10**	315±9***
LSD, SiA (5%)	1.56	0.95	0.75	0.75

Means within a column followed by different number of stars are significantly different according to Fisher's:

*=Significant at P< 0.5 **=Significant at P< 0.1 ***=Significant at P<0.05

decreased with the increase extract concentration from SA_2 to SA_4 at sandy soil, while at silt soil the lengths gradually decreased with the increases of concentration of extract.

Water content (W.C) and degree of succulence were lower in the SA_1 group compared with C group, Table 3. The treatment with SA_2 and SA_3 increase the water content as well as degree of succulence of the wheat root and shoot at sandy soil. Meanwhile at SA_4 extract, W.C increase in the shoot and decrease in the root samples. At silt soil water content and degree of succulence gradually increase in seedling roots with increasing of extract concentration, meanwhile gradually decreased in seedling shoots.

Priming treatments significantly affected fresh and dry weight of seedlings (Table 4). Fresh weights of seedlings was drastically decreased due to high concentration of *A. princeps* water extract, meanwhile low concentration (SA_1) improved fresh and dry weight of seedlings as compared with control in sandy soil. The reduction of fresh and dry weight reached to 65% at seedlings treated with SA_4. Meanwhile the reduction reached to about 44% at silty soil in the same concentration.

Total chlorophyll was determined in green tissues in *T. aestivum* at two type of soil under the effect of irrigation with *A. princeps* water extract. In SA_1 and SiA_1 seedlings were higher than those of C seedlings, Table 5. On the eight day it was observed that the C seedlings had more than three fold of chl. that of SA_4, while about two fold that of SiA_4. So *T. aestivum* effective in the sandy soil than at silty ones. The chlorophyll content gradually decrease with the increasing in extract concentration at the two soils.

DISCUSSION

Allelopathic compounds in *Artemisia princeps* that was tilled into the soil probably were solubilized rapidly by soil moisture from ample rainfall [5]. The high concentration treatments showed a significant effect on the germination percentage of wheat as well as root and shoot lengths in sand and silt soils. The maximum shoot and root lengths as compared to control was estimated at (A_1) 25% treatment, the result was in agreement with Kil and Yun, [14]. They shows that germination percentage and dry weight of wheat was slightly increased at lower concentrations of the *Artemisia princeps* var. '*orientalis*' extracts, whereas it was proportionally inhibited at higher concentrations at the two type of soil.

The mode of action of a allelochemicals can broadly be divided into a direct and an indirect action [15]. Effects through the alternation of soil properties, nutritional status and an altered population or activity of micro-organisms and nematodes represent the indirect action. The direct action involves the biochemical/physiological effects of allelochemicals on various important processes of plant growth and metabolism. Processes influenced by allelochemicals involve: Mineral uptake allelochemicals can alter the rate at which ions are absorbed by plants. A reduction in both macro- and micronutrients are encountered in the presence of phenolic acids [16], Cytology and ultra structure a variety of allelochemicals have been shown to inhibit mitosis in plant roots [16]. Phytohormones and balance the plant growth hormones indole acetic acid (IAA) and gibberellins (GA) regulate cell enlargement in plants. IAA is present in both active and inactive forms and is inactivated by IAA- oxidase. IAA- oxidase is inhibited by various allelochemicals [16] Other inhibitors block GA-induced extension growth. Membranes and membrane permeability many biological compounds exert their action through changes in permeability of membranes. Exudation of compounds from roots on root slices have been used as an index of permeability because plant membranes are difficult to study [17].

Water content and degree of succulence were lower in the 25 and 100% treatment groups as compared with the control group in sandy soil. The treatment with 50 and

75% extracts increase the water content and degree of succulence of wheat at sandy soil. At silty soil the water content and degree of succulence decrease in all treated sample compared with control.

Fresh and dry weights of seedlings was drastically decreased due to the increase in the concentration of extracts from 25 to 100% at silty soil, but seedlings treated with 25% of sandy soil (SA_1) improved fresh and dry weights as compared to control. Increase in the concentration of extract from 50 to 100% significantly decrease root and shoot biomass of *T. aestivum* by 50 % and 58% respectively.

There are hundreds of secondary metabolites in the plant kingdom and many are known to be phytotoxic [18]. Allelopathic effects of these compounds are often observed to occur early in the life cycle, causing inhibition of seed germination and/or seedling growth [19]. The compounds exhibit a wide range of action mechanisms, from affects on DNA (alkaloids), photosynthetic and mitochondrial function (quinones), phytohormone activity, ion uptake and water balance (*phenolics*). Interpretations of action mechanisms are complicated by the fact that individual compounds can have multiple phytotoxic effects [13].

Chlorophyll molecules are the core component of pigment-protein complexes embedded in the photosynthetic membranes and play a major role in photosynthesis. A Chl reduction must result in a decrease of photosynthesis efficiency. *A. princeps* water extracts reduce Chl accumulation in all treated samples except at low concentration (SA_1 and SiA_1). Yang *et al.* [20] showed that allelochemical may reduce Chl accumulation in three ways: the inhibition of Chl biosynthesis, the stimulation of Chl degradation, or both. The present study strongly indicates that Chl biosynthesis of wheat seedlings is affected by the exogenously applied allelopathic water extracts. The extract exhibited apparently different degrees of inhibition according to soil type.

Solubilization and leaching of allelopathic compounds from *A. princeps* depended on precipitation, as was the case with *Artemisia* incorporated into soil [21]. The amount, intensity and duration of precipitation and the air temperature probably influenced the leaching pattern of allelopathic compounds, just as they affected loss of other soluble constituents from senescing tissue [22]. The delayed effect of no-till *A. princeps* also suggested that allelopathic compounds degraded slowly, if at all, in non-incorporated compared with their rapid degradation in soil [23].

Our results suggest that *A. princeps* residue effect on the following wheat crop and this depends in large part on the degree of decomposition of the stover before the wheat is planted. Prompt tillage of the stover could alleviate allelopathy by extending the duration for decomposition and, in many cases, enabling it to occur at a more favorable soil temperature before wheat is planted. Although it was noted here, that chopping the *Artemisia* stover finely also might accelerate decomposition. The obtained results as a suggested by Purvis, [21] that silt soil could promote decomposition and lessen allelopathy from *A. princeps* on wheat.

The merits of practices that reduce allelopathy from *A. princeps* must be treated sandy soil with more irrigations well as used silt soil for more decomposition of plant residue due more microbial activity.

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