# INHIBITORY EFFECTS OF WILD BARLEY (*HORDEUM SPONTANEUM* Koch.) RESIDUES ON GERMINATION AND SEEDLING GROWTH OF WHEAT (*TRITICUM AESTIVUM* L.) AND ITS OWN PLANT

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## ABSTRACT

*Laboratory studies were conducted to examine the effects of different concentrations of wild barley (Hordeum spontaneum Koch.) shoot and seed extracts on germination and seedling growth of wheat (Triticum aestivum L.) and wild barley. In this study, all wild barley shoot extract concentrations (with exception of lowest concentration) significantly reduced wheat seed germination after 7 days. Shoot extract concentrations of 60 and 120 g / L significantly reduced shoot and root lengths, shoot fresh and dry weights, and, root fresh and dry weights of wheat. Seed germination of wheat was not affected by intermediate wild barley seed extracts. Considerably, some extracts of wild barley seeds stimulated the germination and growth of wheat. Wild barley shoot and seed extracts at low level stimulated the growth of its own plant, however, its germination and seedling growth were inhibited at higher shoot extract concentrations. The results of this investigation show that wild barley shoot extracts exert more allelopathic effects on germination and growth of wheat and its own plant than those of seed extracts.*

***Key words:*** *allelopathy, HORDEUM SPONTANEUM, wheat, germination, seedling growth.*

## Introduction

Crop growth and development are influenced by a wide range of abiotic and biotic factors that usually create less than an optimal crop production environment (Einhelling, 1996). Weeds, as one of the major biotic factors, are known to be plants of negative value, which interfere with main crop through competition for space, water, nutrients and carbon dioxide for photosynthesis (Klingsman et al., 1982) and allelopathy (Rice, 1984) or both (Weidenhamer et al., 1989).

The phenomenon of allelopathy, where a plant species chemically interferes with germination, growth or development of

neighbor plant species has been known for over 2000 years (Rice, 1984). Chemicals that impose allelopathic influences are called allelochemicals and are present in virtually all plant tissues, including leaves, flowers, fruits, stems, roots, rhizomes, seeds and pollen. They may be water-soluble and released from plant parts into the environment by means of volatilization, leaching, root exudation, and decomposition of plant residues (Whittaker et al., 1977).

Biological activities of receiver plants to allelochemicals are known to be concentration dependent with a response threshold. Responses are, characteristically, stimulation at low concentrations of allelochemicals, and inhibition as the concentration increases (Bais et al., 2003, Lovet, 1989, Weidenhamer, 1996). Most researches on allelopathy have focused on the effect of interactions among weed species (Narwal, 1994), weeds and crops (Rice, 1984) and crop species (Hedge et al., 1990).

Laboratory bioassays are an important part of allelopathic research. In the laboratory, plant extracts and leachates are commonly screened for their effects on seed germination with further isolation and identification of allelochemicals from greenhouse tests and field soils, confirming laboratory results. Many laboratory bioassays, however, show little or no correspondence to field interactions because of their dissimilarity to field conditions (Whittaker et al., 1977).

Investigation of allelopathic effects of weeds on wheat germination, growth and yield is not new. In a comparison of numerous species of genus Brassica, Mason-Sedun et.al. (Mason-Sedun et al., 1986) found that water extracts from residues of the genus Brassica significantly reduced root and coleoptile length of wheat with little effect on germination. In the bioassay experiments, Bialy et al. (Bialy et al., 1990) showed that wheat seed germination inhibition was occured at 500 ppm of 2-phenetyl ITC, a known allelochemical derived from species of genus Sinapis. Ghadiri and Hamidi (Ghadiri et al., 1991) studied the effect of different concentrations of seed extract of bur parsley [Turgenia latifolia (L.) Hoff.] on germination and seedling growth of three dryland wheat cultivars. They reported that bur parsley seed extract concentrations of 60, 80, and 100% significantly reduced the germination of three wheat cultivars after 3 days and germination of weed seeds were not sensitive to the lower concentrations of their own extracts. Inderjit and Dakshini (Inderjit et al., 1998) investigated the allelopathic effects of chickweed [Stellaria media (L.) Vill.] plant parts on seedling growth of wheat. Results indicated that both young and mature growth stages of chickweed contribute water-soluble phenolics to the soil and inhibit seedling growth of wheat.

Two-rowed wild barley, Hordeum spontaneum Koch, is a dominant troublesome weed in most wheat fields in Iran and its natural population has been reported

in many other parts of the world (Harlan et al., 1966). It is a winter annual plant from Poacea, reproducing by seed. Seed dispersal is usually limited to within several meters of the mother plant; although seeds can be carried in the fur of animals over longer distances (Zohary, 1969).

H. spontaneum,first found in 1848, is the most original species of cultivated forms. The known distribution areas of this plant is Afghanistan, Turkey, Jordan, Syria, Iraq, Iran, Pakistan, individual parts of North Africa, Outer Caucasus, and the Southern part of Middle Asia. Normally it grows from 350-1500 m above sea level (Shao et al., 1983).

Over the past 20 years, an extensive amount of information has been obtained on the extent and structure of genetic variation in H. spontaneum (Brown et al., 1978, Volis et al., 2001, Volis et al., 2002), but little or no information is available on allelopathic effects of its plant parts on other plants. The present study was conducted to evaluate the effects of various aqueous extract concentrations of H. spontaneum seed and shoot on wheat and its own seed germination and seedling growth.

## Materials and Methods

**Plant sampling and preparation of extracts**

Mature wild barley plants and seeds were collected from the experiment station farm, College of Agriculture, Shiraz University, located in Bajgah valley,18 km north of Shiraz, Iran.

All above-ground plant materials except seeds, were chopped by hand into small 1-cm long pieces and then oven-dried at 48 C for 48 hours (Inderjit et al., 1995). Extracts were prepared by soaking appropriate amounts of seed (1, 2, 4, 8, and 16 g) and chopped plant materials (7.5, 15, 30, 60, and 120 g) in 1000 ml distilled water for 24 h at room temperature. The containers were shaken at intervals and after 24 h, the extracts were collected and filtered through 3 layers of Whatman # 2 filter paper and stored in cool temperature (5 C) until experiments were conducted.

**Seed bioassay**

Germination tests were conducted for each of the extracts. Twenty five surface-sterilized(with 75% ethanol for 2 minutes) wheat (Triticum aestivum cv. Pishtaz ) and naked wild barley seeds were germinated in sterilized 9-cm Petri dishes contained two Whatman # 2 filter papers moistened with 5 ml of the appropriate extract or with distilled water (control treatment) at constant temperature of 25 C in germinator. 3 ml of each appropriate extract or distilled water (control) were added to each Petri dish after 3 days to prevent drying. After 7 days, wheat and wiled barley seed germination (SG) were counted and shoot (SL) and root lengths (RL), No. of seminal roots (NSR), shoot fresh (SFW) and dry weights(SDW), root fresh (RFW) and dry weights (RDW) of 5 randomly chosen seedlings of both plants were measured and averaged for each replicate

within each treatment. Germination was considered to occur when radicle length was 1mm or longer. Polyethylene glycole (PEG) was not used in this study because the extract solution concentrations did not exceed 50 milliosmoles (about –0.11 Mpa) (Bell, 1974).

**Experimental design and statistical analysis**

Germination and seedling growth bioassays were conducted in a completely randomized design (CRD) with three replications. Homogenity of variances was tested and those data not normally distributed were log10 transformed and retransformed data are presented in the results. Data were analyzed by analysis of variance procedure and differences between means were subjected to Duncan's new multiple range test at the p=0.05 level.

## Results

**Effects of wild barley shoot and seed extracts on wheat germination and seedling growth**

All wild barley shoot extract concentrations (with exception of 7.5 g/L) significantly reduced wheat SG after 7 days (Figure 1A). The degree of reduction was different. Highest extract concentration (i.e. 120 g/L) significantly caused the highest reduction in wheat SG compared with the distilled water. Shoot extract concentration of 7.5 g/L increased wheat SG after 7 days, but as compared with control treatment, this increase was not significant.

The most severe reductions were observed in wheat SL and RL treated with 60 and 120 g/L wild barley shoot extract concentrations (Figures 1B and 1C). While NSR was not affected by all wild barley shoot extract concentrations (Figure 1D), wheat SFW was significantly inhibited by extract concentrations of 60 and 120 g/L to the extent of 68.9 and 76%, respectively (Figure 1E). On the other hand, the reduction in wheat SDW as affected by these concentrations was 57.12 and 67.8%, respectively (Figure 1F).

The RFW and RDW of wheat were significantly decreased at higher shoot extract concentrations (Figure 1G and 1H).

In general, wheat SG was not decreased by intermediate wild barley seed extracts and only the highest concentration significantly inhibited wheat SG (Figure 2A). The effects of wild barley seed extract treatments on wheat SL and RL are showed in Figures 2B and 2C. Considerably, all treatments significantly stimulated SL as compared with control treatment (Figure 2B). The RL responses to aqueous extracts were nearly the same of SL (Figure 2C). Wheat NSR and SFW were not affected by wild barley seed extracts (Figures 2D and 2E), whereas, lower and higher extract concentrations significantly stimulated SDW (Figure 2F). Wheat RFW and RDW showed the same responses, in these cases, lower extract concentration (i.e. 1 g/L) significantly

increased RFW and RDW (Figures 2G and 2H).

**Effects wild barley shoot and seed xtracts on its own seed germination and seedling growth**

Wild barley SG responses to various extract concentrations of its own shoot extracts is shown in Fig. 3A. With exception of the first shoot extract concentration (i.e. 7.5 g/L), the rest of treatments significantly reduced SG percentage after 7 days. At extract concentration of 60 g/L only 8.7% of seeds were germinated but failed to develop into normal seedlings. Wild barley SG responses to own extract concentrations largely depends upon the extract concentration, so that, the highest concentrations produced highest germination inhibition (Fig. 3A). Wild barley seed extracts at concentrations of 2, 4, 8 and 16 g/L significantly reduced its own SG, whereas the lower extract concentration (i.e. 1g/L) showed stimulation effects on SG (Fig. 4A).

Of the six shoot extract treatments that were tested, the concentrations of, 60 and 120 g/L were most likely to exhibit reductive effects (Fig. 3). In compare to control treatment, the concentration of 7.5 g/L increased SL and RL of its own seedlings by 8 and 6%, respectively, however, no significant effects were observed. The extract concentrations of 60 and 120 g/L severely suppressed its own SL and RL (Fig. 3B and 3C). All seed extract concentrations increased its own SL, but the highest of the five extract concentrations included in this study, i.e. 16 g/L was more effective for stimulation of its own SL (14%). Similar trends were observed in RL, but in this case the major effective concentration was 8 g/L(Fig. 4B and 4C).

Among all applied treatments, the highest shoot extract concentrations (i.e. 60 and 120 g/L) severely suppressed seminal roots growth (Fig. 3D).

Differences in shoot FW and root FW, due to the highest levels of extract concentrations, were found significant as compared to the rest of treatments (Fig. 3E and 3G). Among all shoot treatments, concentrations of 7.5, 15 and 30 g/L increased the own shoot FW, however, these observed effects were not significant. Surprisingly, aboveground wild barley extract treatments (i.e. 7.5, 15 and 30 g/L) significantly increased the shoot and root fresh weights of its own seedling (Fig. 3E and 3G). Both shoot FW and DW showed positive responses to all seed extract concentrations. As compared with distilled water, the extract concentration of 8 g/L significantly increased its own shoot FW and DW by 27.3 and 32%, respectively (Fig. 4E and 4F).

The effects of various concentrations of wild barley shoot extract on its own shoot and root DW are shown in Fig. 3E and 3G. In compare to distilled water, extract concentrations showed stimulated effects on shoot DW at 7.5, 15 and 30 g/L by 7.17, 14.34 and 1.02%, respectively (Fig. 3F). The data in Fig. 4G and 4H demonstrates the effects of wild barley seed extract

concentrations on its own root FW and DW. In general, root FW and DW responses were similar to those of shoot. The greatest stimulative effects of seed extract concentrations occurred with 8 g/L (59%) and 1 g/L (54.3%) for root FW, 47 and 51% for root DW, respectively.

## Discussion

There are many literature reviews that suggest various members of the Poaceae family have allelopathic potential (Abdul-Wahab et al., 1967, Bokhari, 1978, Einhelling et al., 1992, Goslee et al., 2001, Hanson et al., 1981, Hanson et al., 1983, Liu et al., 1993, Liu et al., 1998). The results presented above clearly show differential phytotoxicity and autotoxicity of aqueous extracts of wild barley aboveground body plant. Putnam and Duke (Putnam et al., 1974) have suggested that “wild types” of existing crops may have possessed high allelopathic potential and that this characteristic may have been reduced or lost during agronomic selection.

The results of this study also demonstrated that the wild barley plant components produce compound(s) that could be inhibitor(s) for other plants. In this experiment no attempt was made to isolate and identify any specific phytotoxic substances in wild barley, however, some literatures have indicated the presence of effective allelochemicals including gramine (N,N-dimethyl-3-aminomethylindole) (Hanson et al., 1981, Hanson et al., 1983), DIBOA (2,4-dihydroxy-1, 4-benzoxazin-3-one) (Baria et al., 1991, Gianoli et al., 1998) and hordenine (N,N-dimethyltyramine) (Lovett et al., 1995, Overland, 1966) that are phytotoxic to seed germination and seedling growth.

In general, both shoot and seed extracts reduced germination and seedling growth of wheat especially at higher concentrations. Shoot extracts inhibited wheat root FW more than shoot FW. These results are in agreement with earlier studies reporting that water extracts of allelopathic plants have more effects on root growth than shoot growth (Ben-Hammouda et al., 1995, Bowmick et al., 1982).

Compare to wheat, the large depression in wild barley seed germination due to its own shoot extract levels may be attributed to lower resistance of this species to allelochemicals. This finding generally agree with that of Overland (Overland, 1966) who reported that barley is more sensitive to allelochemicals than wheat.

To distinguish competition and allelopathy, Rice (Rice, 1984) and Liu and Lovet (Liu et al., 1998) reported that the response to competition is usually reduction in plant growth, while the response to allelopathy is characteristically stimulation at low concentrations and inhibition as the concentration of allelochemical increases. The results of this investigation indicated that wheat and wild barley growth parameters were stimulated when low rates of both shoot and seed extracts are used (Fig. 3 and 4)

Although, involvement of wild barley competition with wheat has not been reported

earlier, it could be concluded that wild barley vegetative residues can release water soluble compound(s) into the soil and reduce germination and seedling growth of wheat through allelopathy phenomenon.

Extracts from the seeds of wild barley almost stimulated its own seedling growth. This valuable effect shows an ecological adaptation involvement, because in natural conditions and in competition with other competitors, a phenomenon must be suitable and reasonable to enhance plant seedling establishment. In this relation, allelopathy, as an ecological significant mechanism, plays an important role.

Visual observations show that wild barley population densities have been increased in many parts of Iran during the last 20 years. The allelopathic potential of this weed, but not as an unify theory, may be involved (Goslee et al., 2001, Inderjit et al., 1998), on the other hand, resource availability determined amount of allelochemicals in plant tissues (Einhelling, 1996), it could be expected that wild barley population increasing occur in the places where resources especially soil moisture are in the shortage.

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