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Seed germination and seedling growth of wheat (*Triticum aestivum*) as influenced by safed behman (*Centaurea behen*) water extract

Yasin NOROUZI, Gholamreza MOHAMMADI* and Iraj NOSRATTI

Department of Crop Production and Breeding, Faculty of Agriculture and Natural Resources,
Razi University, Kermanshah, Iran. Postal code: 6715685438
*Corresponding author, G. Mohammadi,: E-mail: mohammadi114@yahoo.com
Fax: 098-831-8321083, Telephone: 098-831-8326919.

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Abstract. Plant species belonging to the family *Asteraceae* produce wide varieties of biologically active components. Safed behman (*Centaurea behen* L.) a member of *Asteraceae* family, is a perennial weed species which is native to south Asia and during recent years has become a dominant weed of wheat fields in west Iran. In order to investigate the phytotoxic potential of safed behman water extract on wheat, an experiment was conducted at the Plant Physiology Laboratory of Faculty of Agriculture and Natural Resources, Razi University, Kermanshah, Iran in 2014. The experiment was a factorial with two factors arranged in a completely randomized design with three replications. The first factor was safed behman water extract concentrations [0 (control), 25, 50, 75 and 100%) and the second was wheat cultivars (Sardari and Ryzhav).Results showed that for both wheat cultivars safed behman water extract significantly decreased all of the traits under study including germination percentage, mean germination rate and the length and dry weight of plumule and radicle. Decreasing effects were intensified in response to increasing water extract concentration, as the highest concentration (100%) of safed behman water extract reduced wheat germination percentage and mean germination rate by 84.6 and 31%, respectively as compared with control. In control and the lowest concentration of water extract (25%) treatments Ryzhav showed significant higher radicle length than Sardari but at the higher concentrations (more than 50%) there were no significant differences between two cultivars in terms of this trait. Safed behman water extract had also a more reducing effect on Sardari plumule length so that at the highest concentration the reductions for this trait were 67.2 and 56.1% for Sardari and Ryzhav, respectively compared to control.

Key words: germination, phytotoxicity, safed behman, seedling growth, wheat.

Introduction

Safed behman (*Centaurea behen* L.) a member of *Asteraceae* family, is a perennial weed species which isnative to south Asia (Flamini et al. 2004). In dry farmlands of west Iran,it recently has become an important weed for both summer and winter season crops such as wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.) and chickpea (*Cicer arietinum* L.). According to the author' spersonal observations and local reports, wheat crop cultivated in the farms infested with safed behman has lower growth and yield which could be attributed to the weed allelopathic activity. Allelopathy is defined as the ability of certain plant species to produce secondary chemical compounds that exert some sort of biological effects on other plants (Rice 1984).Some of the main effects of allelochemicals on crops are as prolonged emergence or delayed germination and emergence, reduced biomass of the roots and inhibited seedling growth (Kobayashi 2004, Liebman & Sundberg 2006, Hiradate et al. 2010). These effects further increase the competition ability of weeds for resources such as light, water and nutrients. Allelochemicals may leach out from various parts of plants to the surrounding rhizosphere either as exudates or rain-residues or from dead tissues (Macias et al. 2004, Haramoto & Gallandt 2005, Duke 2015).

Centaurea species exude chemicals from their organs that can have allelochemical activity (Callaway et al. 1999, Bais et al. 2002, 2003). For example, *Acroptilon repens* (L.) DC (Russian knapweed; formerly *Centaurea repens*), a perennial herbaceous plant belonging to the family Asteraceae. Ni et al. (2010) reported that competitive ability, perhaps through an allelopathic mechanism, may contribute to the competitive success of *Acroptilon*. Several natural products from this plant have negative effects on other organisms. Phytochemicals from Russian knapweed are harmful to other plants (Stermitz et al. 2003). It secretes a phytotoxic flavonoid, 7.8-benzoflavone. This chemical was identified under soil-free conditions and caused toxicity in other species at 100 mg ml^{-1} (Stermitz et al. 2003). In another study, Bais et al. (2002) isolated (±)-catechin from the root exudates of *C. maculosa* and showed that (–)-catechin can have strong allelopathic effects in strile culture.

Some chemical investigations on *Centaurea* species have also shown the presence of flavonoids, sesquiterpene lactones, especially guaianolides, germacranolide-typesesquiterpene lactones (Rustaiyan et al. 1981a, 1981b, 1982, 1984). However, we didn't find any report on the phytotoxicity effects of safed behman on crops. Since, safed behman has recently become a major weed in wheat fields of west Iran and wheat usually shows a weaker establishment and growth in the presence of this weed species, therefore, the main objective of the present study was to assess the possible phytotoxic effects of safed behman water extract on seed germination and initial seedling growth of two wheat cultivars (Sardari and Ryzhav) which commonly cultivated in west Iran.

Materials and Methods

This experiment was carried out at the Plant Physiology Laboratory of Faculty of Agriculture and Natural Resources, Razi University, Kermanshah, Iran in 2014. Two non-irrigated wheat cultivars used were Sardari and Ryzhav which are widely grown in the region. The experiment was a factorial with two factors arranged in a completely randomized design with four replications. The first factor was safed behman water extract concentrations [0 (control), 25, 50, 75 and 100%) and the second was wheat cultivars (Sardari and Ryzhav). In order to prepare the safed behman water extract, weed plants were collected from the non-irrigated wheat fields located at Javanroud (34° 48' 24"N, 46° 29' 29"E; elevation 1380 m), Kermanshah province, west Iran, just before flowering stage. The collected weed tissues were dried at room temperature until a constant weight, then ground with a mill equipped with a 1-mm-mesh sieve. 100 g of ground plant tissue was soaked added to 1 liter of distilled water. The solution was placed on an orbital shaker with 260rpm for 24 h at room temperature (25 °C) then filtered several times through a four-layer cheese cloth and subsequently through a two-layer filter paper (Whatman # 2).To prepare the different concentrations of safed behman water extract, the filtered solution was diluted by distilled water. Wheat seeds were sterilized by NaOCl 5% for 5 minutes then washed by distilled water. Twenty seeds of each wheat cultivar were placed in glass petri dishes of 9 cm diameter lined with a two-layer filter paper(Whatman # 2).The petri dishes were moistened with 10 ml of safed behman water extracts or distilled water (as control). The petri dishes were sealed with parafilm and placed in a growth chamber at 25°C for 8d.

Seed germination was recorded daily up to 8 d after the start of the experiment. A seed was considered germinated when radicle emerged by about 2 mm in length. The mean germination rate (MGR) was calculated according to the following equation (Ellis & Roberts 1980):

$$MGR= \sum n / \sum Dn$$

Where: n is the number of seeds germinated on day D and D is the number of days from the start of test.

Moreover, seed germination percentage (SGP) was determined at the end of test. To determine the radicle length (RL) and plumule length (PL) after the 8^{th}d, normal plumules and radicles produced in each petri dish were separated from the seeds, their lengths were measured, then dried at 70°C for 48 h and their dry weights were recorded. Data analyses were carried out using SAS software (SAS Institute 2003). The Means were separated using LSD test at the 5% level of probability.

Results

According to the analysis of variance (data not shown) all of the wheat traits under study including germination percentage, mean germination rate and the dry weight of plumule and radicle were significantly influenced by safed behman water extract concentration. Moreover, there was a significant two-way interaction (water extract concentration × cultivar) for the length of plumule and radicle.

Safed behman water extract applied in all concentrations significantly reduced the wheat traits under study and the reductions were intensified in response to increasing water extract concentration (Table 1). The weed water extract at the lowest level (25%) decreased wheat germination percentage by 27.5% whereas; the reduction was 84.6% by the highest level (100%), as compared with control. However, mean germination rate showed a milder decreasing trend in response to increasing safed behman water extract concentration, asit was reduced by 24.1 and 31% at the lowest (25%) and the highest (100%) level of the weed water extract, respectively compared to control (Table 1).

Significant reductions were also observed for both plumule and radicle dry weights when wheat seeds were treated by safed behman water extract. Compared to control, at the lowest concentration of the weed water extract the reductions were 22.2 and 40% and at the highest concentration were 77.8 and 80% for plumule and radicle dry weights, respectively (Table 1) indicating higher sensitivity of radicle dry matter accumulation in the presence of safed behman water extract even at the lowest concentration. However, radicle dry weight showed a significant milder reducing response to increasing the weed water extract concentration when compared with plumule dry weight (Table 1).

There was a significant two-way interaction (water extract concentration × cultivar) for the length of plumule and radicle. In control and the lowest concentration of water extract (25%) treatments, Ryzhav showed significant higher radicle length than Sardari but at the higher concentrations (more than 50%) there were no significant differences between two cultivars in terms of this trait (Fig. 1). Safed behman water extract had also a more reducing effect on Sardari plumule length so that at the highest concentration the reductions for this trait were 67.2 and 56.1% for Sardari and Ryzhav, respectively compared to control (Fig. 2). It seems that the radicle and plumule elongation in Ryzhav is more resistant against the phytotoxicity of safed behman when compared with Sardari.

Discussion

In general, wheat seeds treated by safed behman water extract showed significant lower germination and seedling growth parameters and for most traits, the lowering effect was intensified in response to increasing the weed water extract concentration. The allelopathic effect of *Centaurea* species has been reported by other workers. Bais et al. (2002) isolated (±)-catechin from the root exudates of *C. maculosa* and showed that (–)-catechin can have strong allelopathic effects in sterile culture. Pirzad et al. (2013) also found that *Centaurea repens* extract in all concentrations showed phytotoxicity effect on wheat seedling growth. However, based on our literature review, there are no reports on the allelopathic effect of safed behman, especially on wheat.

Table 1. Wheat germination and seedling growth parameters as influenced by different concentrations of safed behman water extract.

Water extract concentration (%)	Germination percentage	Mean germination rate (no/day)	Plumule dry weight (g/plant)	Radicle dry weight (g/plant)
0 (control)	93.125^{a}	0.293^{a}	0.098^{a}	0.052^{a}
25	67.500^{b}	0.221^{b}	0.077^{b}	0.035^{b}
50	41.875^{c}	0.218^{b}	0.061^{c}	0.020^{c}
75	38.125^{c}	0.216^{b}	0.040^{d}	0.018^{c}
100	14.375^{d}	0.200^{b}	0.021^{e}	0.010^{d}
$LSD_{(0.05)}$	10.018	0.023	0.011	0.005

Means followed by the same letters in each column are not significantly different according to LSD test at the 0.05 level of probability.

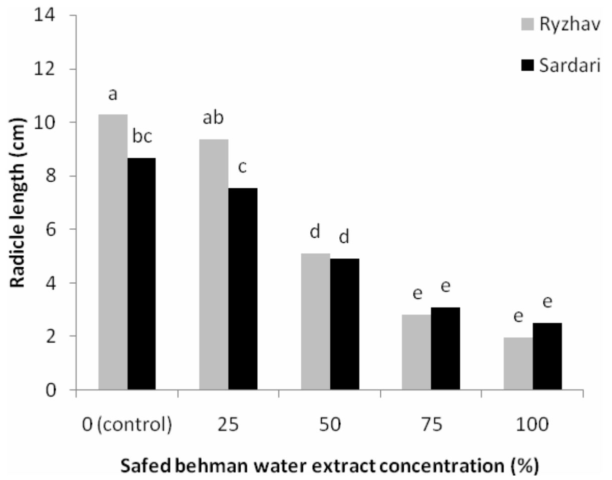

Figure 1. Radicle length of two wheat cultivars in response to safed behman water extract applied in different concentrations.

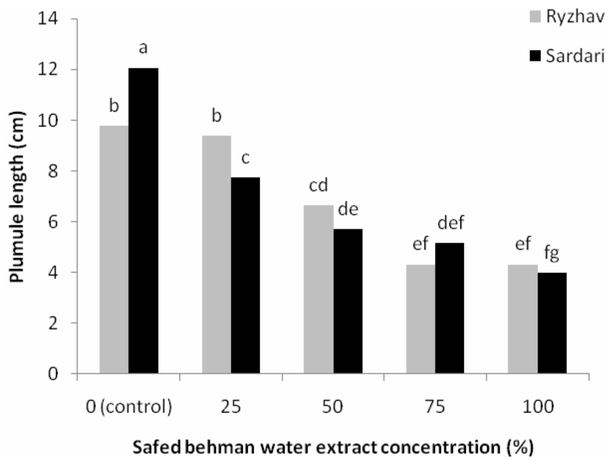

Figure 2. Plumule length of two wheat cultivars in response to safed behman water extract applied in different concentrations.

The inhibition of seed germination induced by *Centaurea* was attributed to disturbance in the activities of peroxidase, alpha-amylase and acid phosphates (Alam & Islam 2002). In general, allelochemicals can suppress seed germination and seedling growth in different ways. Some known sites of action for allelochemicals have been identified to include cell division, pollen germination, nutrient uptake, photosynthesis and specific enzyme function, with commonly cited effects being reduced seed germination and seedling growth (Ferguson & Rathinasabapath 2009). Inderjit & Duke (2003) also suggested several action modes for allelochemicals, including direct inhibition of PSII components and ion uptake, interruption of dark respiration, and ATP synthesis and r*eactive oxygen species*-mediated allelopathic mechanisms.

In our study, the suppressing effect of safed behman water extract was intensified when its concentration was increased. According to Einhellig (1986) the biological activity of allelochemicals in allelopathic plants is highly concentration dependent. For some traits, the weed cultivars under study responded differently to safed behman water extract, as cv. Sardari showed more sensitivity to the weed water extract especially in low concentrations.

Generally, our findings confirm the hypothesis that allelopathy is one of the mechanisms by which safed behman can suppress wheat establishment and growth in field. However, further studies are needed in order to a better understand of the phytotoxic interactions between wheat and safed behman especially in field condition and also to identify the allelopathic active components responsible for the inhibitory effect of this important weed species.

It is **c**oncluded that safed behman water extract even at low concentrations can reduce wheat germination and seedling growth parameters. However, the reductions were more obvious at high concentrations of the weed extract confirming other findings based on the fact that phytotoxicity ability is highly concentration-dependent. For some traits, wheat cultivars responded differently to safed behman water extract as at low concentrations Ryzhav showed higher radicle length than Sardari. Moreover, compared to control the weed water extract applied at different concentrations had more reducing effect on Sardari plumule length than Ryzhav.

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