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Bio-control of velvetleaf (*Abutilon theophrastii*) and field bindweed (*Convolvulus arvensis*) by essential oil of sweet bay (*Laurus nobilis*) as an inhibitor of seed germination

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Abstract

In order to study bio-controlling posibility of velvetleaf (*Abutilon theophrastii*) and field bindweed (*Convolvulus arvensis*) by essential oil of sweet bay (*Laurus nobilis*) two laboratory and greenhouse experiments were conducted in Tabriz, Iran during 2013. Studied treatments in this part of our experiment were essence concentrations of (*Laurus nobilis*) as 0, 100, 200, 300 and 400 ppm. The stepwise regression analysis carried out for the data obtained to test the significance of the independent variables affecting the seedling vigor index. Mean comparisons indicated that 10 days after beginning of experiment 69% of field bindweed seeds germinated, but only 66.8% from velvetleaf seeds. With increasing of essential oil concentration more than 200 ppm as well as control one leaves number per plant reduced 17.5%. Leaf area per plant ranged from 13.5 cm^2 in 200 ppm essential oil concentration to 9.7 cm^2 in means of 300 ppm and 400 ppm. With increasing of essence concentration from 200 ppm seedling biomass reduced significantly. We concluded that higher studied concentrations of *Laurus nobilis* essential oil had high potential in controlling of velvetleaf (*Abutilon theophrastii*) and field bindweed (*Convolvulus arvensis*). The stepwise regression analysis verified that the final germination percentage, seedling biomass and leaf area per plant of both weeds had a marked increasing effect on their seedling vigor index.

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Introduction

In agriculture there is a world-wide effort to reduce the amount of chemicals used in production by introducing modern biological and ecological methods. One of the possible solutions is allelopathy, the utilization of the chemical interaction between plants. Evidence for allelopathic interactions in nature caused by aromatic plants containing volatile allelochemicals have been described frequently (Muller *et al.* 1964, Rice 1984). Essential oils were reported as inhibitors of seed germination and plant growth (Asplund, 1968).

Sweet bay (*Laurus nobilis*), from *Lauraceae* family, is an aromatic evergreen tree or large shrub with green, glossy leaves, native to the Mediterranean region. It is one of the plants used for bay leaf seasoning in cooking. The most abundant essential oil found in laurel is cineole, also called eucalyptol. The leaves contain about 1.3% essential oils, consisting of 45% eucalyptol, 12% other terpenes, 3–4% sesquiterpenes, 3% methyleugenol, and other α- and β-pinenes, phellandrene and linalool (Panza *et al.*, 2011).

Velvetleaf (*Abutilon theophrastii*) is a large genus of *Malvaceae* family. It is distributed throughout Asia (Chisholm, 2011). Field bindweed (*Convolvulus arvensis*) is a species of *Convolvulaceae* family, native to Europe and Asia. Flowering occurs in the mid-summer, when white to pale pink, funnel-shaped flowers develop (Rauciu, 2009). The both above mentioned plants are as troublesome weeds in crop plant fields.

Study of Hemada and El. Darier (2011) revealed that GP attended a value of about 100% at control level for both the two target thyme species and the two test species. GP of *Lepidium sativum* values of about 33 and 60% for *T. capitatus* and *T. vulgaris*, respectively at the maximum oil concentration. The relative records for *Raphanus sativus* were about 65 and 80%, respectively. It is important to mention that *R. sativus* seeds were highly resistance for oil application as compared with *L. sativum*, which is the most sensitive. In Arouiee *et al.* (2006) experiment, leaf chlorophyll content in the control was highest in compare to other treatments. Challa and Ravindra (1988) reported that *Portulaca* leaf leachates reduced the root growth of onion. Also, leaf leachates of *Portulaca oleracea* and *Cyperus* caused 63% and 50 % reduction of root growth in radish.

In several reports, stem length and root and shoot dry weights of cabbage (*Brassica oleracea*), carrot (*Daucus carota*), cucumber (*Cucumis sativus*), squash (*Cucurbita pepo*), onion (*Allium cepa*), pepper (*Capsicum annum*), or tomato (*Lycopersicon esculentum*) were reduced by root exudates of medicinal plants (Qasem, 2001). The essential oils of *Salvia hierosolymitana* Boiss. and *Salvia multicaulis* Vahl. var. *simplicifolia* Boiss. Have shown to have phytotoxic activity against seed germination and initial radical elongation of radish and garden cress, two species frequently used in biological assays (Mancini *et al.*, 2009). In *S. multicaulis* var. *simplicifolia* oil, which contain the major amount of oxygenated terpenoids, has been reported to have inhibitory effects on seed germination and seedling growth inhibitors (Kordali *et al.*, 2007).

In an experiment *Eugenia uniflora, Piper guineense*, *Chromolaena odorata* and *C. odorata* appears to be allelopathically inactive as measured by lettuce seedling bioassay, whereas *E. uniflora* appears to be growth stimulatory, and *P. guineense* is growth inhibitory at concentrations above 400 ppm according to this same bioassay (Oguntimein and Elakovich, 1991)

Results of Azizi and Fuji (2012) showed that an undiluted extract of St. John's wort (*Hypericum perforatum*) and sage (*Salvia officinalis*) had a significant inhibitory effect on seed germination percent and germination rate for *Amaranthus retroflexus*, but not for *Portulaca oleraceae*. Eucalyptus essential oils had a strong inhibitory effect on the germination of both weed species. Dudai *et al.* (1999) have previously reported on the effect of monoterpenes from a number of aromatic plants on the germination of wheat and have also shown that wheat seeds exposed to defined monoterpenes, such as citral, citronellal, pulegone and carvacrol are able to metabolize it (Dudai *et al.* 2000). The effect of exogenous eucalyptol applied to germinating seeds has not been studied previously. In this study, we describe the fate of essential oil of sweet bay, when they are applied to velvetleaf and field bindweed seeds as an inhibitor of germination.

Materials and methods

In order to study bio-control of velvetleaf (*Abutilon theophrastii*) and field bindweed (*Convolvulus arvensis*) with application of essential oil of sweet bay as a inhibitor of seed germination two laboratory and greenhouse experiments were conducted at the Agricultural Research Station of Islamic Azad University of Tabriz, Iran during 2013 with three replications.

Germination bioassay

Studied treatments in this part of our experiment were essence concentrations of (*Laurus nobilis*) as 0, 100, 200, 300 and 400 ppm. Twenty five seeds for each replicate were placed in germinator at 25±1 °C for a germination test in a Petri dish containing Whatman filter paper No. 1 that had been thoroughly moistened with essence solution and germination was checked once a day for 10 d. The recorded data were final germination percentage, seedling biomass, seedling length, seedling vigor index, leaves number and leaf area index.

Final germination percentage (GP) was calculated as the cumulative number of germinated seeds with normal radicles by using below equation, as described by Larsen and Andreasen (2004).

$GP = \Sigma n / N \times 100$

Where, n is number of germinated seeds at each counting and N is total seeds in each treatment.

Seedling vigor index (SVI) was calculated according to Abdul-Baki and Anderson (1973) by using below equation.

$SVI = SDW \times GP$

Where, GP, SDW and SVI are final germination percentage, seedling dry weight and seedling vigor index, respectively.

In this study, to formulate the relationship among five independent growth variables measured in our experiment for weed plants with a dependent variable, multiple regression analysis was carried out for the final germination percentage (X_1), seedling biomass (X_2), seedling length (X_3), leaves number (X_4) and leaf area per plant (X_5); and seedling vigor index as a dependent variable. The multiple regression equation for seedling vigor index is as follows:

Furthermore, the stepwise regression analysis was also carried out for the data obtained to test the significance of the independent variables affecting the seedling vigor index. The stepwise regression equations are as follows:

Velvetleaf:

$Y_{SVI} = -111.101 + 1.222\ (X_1) + 1.412\ (X_2) + 5.012\ (X_5)$

Field bindweed:

$Y_{SVI} = -108.444 + 1.510\ (X_1) + 1.007\ (X_2) + 3.435\ (X_5)$

Results and discussion

Analysis of variance indicated that there is significant difference between weed species with a view to studied variables. Also, effect of studied treatments on all studied variables was significant (Table 1). Kocacaliskan and Terzi (2001) indicated that jouglone and leaf extracts of walnut influenced seed germination and seedling growth of tomato, cucumber, garden cress and alfalfa.

Mean comparisons

Mean comparisons indicated that 10 days after beginning of experiment 69% of field bindweed seeds germinated, but only 66.8% from velvetleaf seeds, with 12.6 cm and 11.2 cm seedlings length, respectively. Rietveld (1983) shown that seed germination and seedling growth of several species were sensitive to juglone. Similar results were indicated that juglone inhibited significantly from the germination of cucumber (Tekintas *et al.*, 1988), tomato and bean (Neave and Dawson, 1989) and alfalfa (Dornbos and Spencer, 1990). In Arouiee *et al.* (2006) experiment, plant length decreased in Lavender and thyme extract treatment. Increasing in the levels of extract accompanied with plant length declined. The highest plant length in rosemary and eucalyptus extract belong to 20 and 10% of treatments, respectively.

Table 1. Mean squares for studied variables in weed plants under essential oil treatment.

SOV	df	Final germination percentage	Seedling length	Leaves number per plant	Leaf area per plant	Seedling biomass	Seedling vigor index
Weed species (A)	1	151.21*	100.00*	2.21*	40.52*	0.23*	5.45*
Essence concentrations (B)	4	0.25**	110.22**	8.81**	65.55**	0.25*	15.11**
A×B	4	10.25	4.45	0.41	7.54	0.11	1.24
Error	20	42.21	20.52	0.49	10.65	0.07	1.30
CV (5%)	-	22.22	26.00	29.49	29.29	24.21	26.25

*, ** mean significant difference at 5% and 1% probability levels, respectively.

On the contrary, seedlings from velvetleaf had greater leaves number per plant and higher leaf area per plant than field bindweed. Similarly, velvetleaf plants had greater biomass and seedling vigor index than field bindweed (Table 2). Study of Hemada and El. Darier (2011) revealed that the effects of essential oils extracted from the dry shoots of Thymus capitatus and T. vulgaris was evaluated upon germination percentages (GP) of Lepidium sativum and Raphanus sativus. Commonly, GP decreased with increasing essential oil concentration.

Table 2. Mean comparisons for studied variables in weed plants.

Weed species	Final germination percentage	Seedling length (cm)	Leaves number per plant	Leaf area per plant (cm^2)	Seedling biomass (g $plant^{-1}$)	Seedling vigor index
Velvetleaf	66.8	11.2	17	12.5	0.50	33.4
Field bindweed	69.0	12.6	14.7	10	0.36	24.8

Higher concentration of the leaf extracts of *Thymus vulgaris, Lavadula* sp., *Rosmarinus officinalis* and *Eucalyptus citriodora* on seed germination and some growth characteristics of *Solanum nigrum* and *Amaranthus retroflexus* had more effective than others. Increasing in the levels of extract accompanied with declined in the germination and growth of both weeds. Germination percent, germination rate and MDG influenced by plant extracts in comparison of control, so that the all germination parameters decreased (Arouiee *et al.*, 2006).

It seems that treatment with 200 ppm essential oil of *Laurus nobilis* cause to improvement of 73.3% in final germination percentage of weed seeds, but only 64.7% in average of other treatments (Table 3). The inhibitory effect of essential oil was concentration dependent. Among treatments, higher seedling height measured in 100 ppm and 200 ppm essential oil concentrations, and the lowest one in control. Concentrations of 300 ppm and 400 ppm were better than control (Table 3).

Leaves number per plant ranged from 17.5 in 200 ppm essential oil concentration to 14 leaves in control. With increasing of essential oil concentration more than 200 ppm as well as control one leaves number per plant reduced 17.5% (Table 3).

Similarly, leaf area per plant ranged from 13.5 cm^2 in 200 ppm essential oil concentration to 9.7 cm^2 in means of 300 ppm and 400 ppm. There is no significant difference in leaf area per plant between 100 ppm and control. With decreasing of essential oil concentration from 200 ppm to 100 ppm leaf area restricted significantly (Table 3). With increasing in concentration of extract, LAI and chlorophyll content increased. There were many researches that the allelopathy is a kind of stress. It may be increase the amount of leaf chlorophyll content for increasing efficiency of photosynthesis of the plants (Salisbury and Ross, 1991).

Table 3. Mean comparisons for studied variables in weed plants under essential oil treatment.

Essential oil concentrations (ppm)	Final germination percentage	Seedling length (cm)	Leaves number per plant	Leaf area per plant (cm^2)	Seedling biomass (g $plant^{-1}$)	Seedling vigor index
Control (0)	64.8 b	24.0 c	14.0 c	11.0 b	0.44 b	28.6 b
100	65.3 b	38.5 a	15.8 bc	11.0 b	0.43 b	27.4 b
200	73.3 a	38.0 a	17.5 a	13.5 a	0.69 a	50.6 a
300	63.8 b	33.0 b	16.2 b	10.0 bc	0.30 c	19.6 c
400	65.0 b	33.9 b	15.8 bc	9.5 c	0.30 c	19.4 c

In Rowshan and Karimi (2013) experiment seedling growth of *Zea mays* in control were significantly higher than in *Salvia macrosiphon* extract treatments. Concentrations of 5% to 100% decreased germination of corn seeds significantly, compared to control. Increasing of leaf and root extracts concentration from 5% to 100% inhibited germination from 27% to 96% and 20% to 75% in comparison with control. Highest significant inhibition effects on germination were obtained by 100% leaf *S. macrosiphon* extract. *S. macrosiphon* extract treatments not only decreased germination but also decreased growth and dry matter in corn seedlings. Treatments with 5%, 10%, 25%, 50% and 100% extracts decreased dry weight of corn significantly. Decreased seedling dry weight was resulted by decreased seedling growth. The plumule length, plumule fresh weight and dry weight of seedlings were reduced significantly in response to the *S. macrosiphon* extracts. Among two parts, leaves were the most allelopathic. Compared to control treatment, *S. macrosiphon* extracts at concentrations of 100% decreased corn radicle length, plumule length, radicle fresh weight, plumule fresh weight, radicle dry weight, plumule dry weight 76, 52, 65, 45, 68 and 40% with root extract and 99, 100, 100, 100, 100 and 100% with leaf extract, respectively.

When plants treated with 200 ppm essential oil, dry weight per plant increased nearly 100% compared to 300 ppm and 400 ppm concentrations. With increasing of essence concentration from 200 ppm seedling biomass reduced significantly (Table 3). We understood that treatment of 200 ppm essence produced 1.8, 2.6, 2.6 and 1.8 times vigorous seedlings compared to 100, 300, 400 ppm and control, respectively (Table 3).

Conclusion

We concluded that higher studied concentrations of *Laurus nobilis* essential oil had high potential in controlling of velvetleaf (*Abutilon theophrastii*) and field bindweed (*Convolvulus arvensis*). The stepwise regression analysis verified that the final germination percentage, seedling biomass and leaf area per plant of both weeds had a marked increasing effect on theie seedling vigor index.

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