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ALLELOPATHIC EFFECT OF OREGANO (*Origanum onites* L.) ON GERMINATION AND SEEDLING DEVELOPMENT OF SOME WEED AND CULTIVARS

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Abstract

The present study carried out at Suleyman Demirel University, Faculty of Agriculture of the Department of Field Crops in 2010. The aim of the study was to examine allelopathic effects of oregano oil (Origanum onites L.) on germination and seedling development of amaranth (Amaranthus retroflexus L.), dock (Rumex crispus L.) and mustard (Sinapis arvensis L.) and crop plants [wheat (Triticum aestivum cv Gün 91), sunflower (Helianthus annuus cv. Sirena) and chickpea (Cicer arietinum)]. Seeds of these plants were germinated in petri and pot. Oregano oil doses for petri study was 0 (control), 3, 6, 10 and 20 µl, for pot study was 0 (control), 0.5, 1.0, 2.0 and 4.0 mg kg^{-1}. The germination rate, root length, seedling length and dry matter in each germination condition were investigated. All parameters were significant ($p<0.01$). The germination rate of seeds in petri and pot at control application was determined as 16.0 and 46.7% for amaranth, 53.3 and 74.0% for dock, 56.0 and 72.0% for mustard, 89.3 and 92.0% for sunflower, 100.0 and 100.0% chickpea, 100.0 and 100.0% for wheat, respectively. The lowest oregano oil doses (3 µl and 0.5 mg kg^{-1}) in petri and pot study reduced, germination rate of dock, amaranth and mustard dramatically, germination rate of other plants did not change. The increased doses of oregano oil in petri and pot study negatively affected the root length of dock, amaranth and mustard. The seedling length of amaranth, mustard and dock at 0.5 mg kg^{-1} dose was decreased compared to control. As a result, it was determined that oregano oil (3 µl and 0.5 mg kg^{-1}) at low doses significantly inhibited weed seed germination and was less damaging to crop plants seed germination.

Key words: *allelopathy, oregano oil, germination, seedling growth.*

INTRODUCTION

There are large numbers of weeds in agricultural production areas. 7000 species in the world and 1800 species in Turkey have been identified as harmful weeds in agricultural areas and their damage is around 32% in our country (Aydın and Tursun, 2010). Herbicides compose 50% of all pesticides used in the world. Herbicide use has increased due to easier and quicker application to large areas and expensive labour force in agriculture. This situation can cause to an irreversible problem (Rice, 1984).

One of the most important strategies against weeds is phytotoxic damage in the agricultural areas, which depends on preventing the enzyme activity, inhibiting germination and growth of plant compounds. So far, studies on extracts with herbicidal effects derived from secondary metabolites in plants have been demonstrated. Monoterpenes are the major compounds responsible for phytotoxic effect. Lavandula (*Lavandula hybrida* L.), oregano (*Origanum onites*), sage (*Salvia officinalis* L.), mint (*Mentha piperita*), rosemary (*Rosmarinus officinalis*) and fennel (*Foeniculum vulgare*) essential oil as well as many from medicinal and aromatic plants are obtained, and the main components of these oils are monoterpenes.

One of the alternative methods is to use the allelopathic substances (secondary metabolites, allelochemicals) against weeds, pests and plant diseases. Awareness of the availability of these substances in biological control against weeds increased the importance of allelopathic effects in crop production practices (Rice, 1984). Allelopathy is defined as a limiting or enhancing effect as the development of plants of each other as a result of various chemical mechanisms (Türkmen and Turhan, 2006).

The aim of this research was to determine allelopathic effects of oregano oil (*O. onites* L) on germination and seedling development of amaranth (*A. retroflexus* L.), dock (*R. crispus* L.) and mustard (*S. arvensis* L.) and crop plants

[wheat (*T. aestivum* cv Gün 91), sunflower (*H. annuus* cv. Sirena) and chickpea (*C. arietinum*)].

MATERIALS AND METHODS

This research was carried out Suleyman Demirel University, Faculty of Agriculture at the Department of Field Crops. In this study, three weeds species and three cultivated species were used as plant material. Oregano essential oil was obtained from hydrodistillation.

Weed seeds were collected during the months of July to September 2009 from the crop cultivation areas.

In petri experiments; 0 (control), 3, 6, 10 and 20 µl doses, in pot experiment; 0, 0.5, 1.0, 2.0, and 4.0 mg kg^{-1} doses of oregano oil was applied (Azirak and Karaman, 2008; Gülsoy et al., 2008). Twenty five seeds were put into perti dishes and 10.0 ml distilled water was added to each petri dishes. Field soil with the texture clayed-calcareus, with pH 8.1, Cation Exchange Capacity 36.0% and total salt composition 0.025%, rich in lime (75.4 K_2O da^{-1}) and with poor organic material (1.34%). Only distilled water was used for control doses. Seeds within petri dishes and pots were allowed to germinate during 15 days at room temperature (25°C). At the end of this period, germination rate, root and stem length and dry matter rate were investigated.

Gas Chromatography/Mass Spectrometry (GC-MS) analysis of the oregano oil was performed on Shimadzu 2010 Plus GC-MS equipped with a Quadrapole (QP-5050) detector. The analysis was performed under the following conditions: capillary column, CP-Wax 52 CB (50 m x 0.32 mm, film thickness 0.25 µm); injector and detector temperature, 240°C; stove heat program, from 60°C (10 min. hold) to 90°C rising at 4°C/min., and increasing to 240°C (11.5 min. hold) rising at 15°C/min.; flow speed, 1 psi; detector: 70 eV; ionization type, EI; carrier gas, helium (20 ml/min.); sample injected 1 µl. Identification of constituents was carried out with the help of retention times of standard substances by composition of mass spectra with the data given in the Wiley, Nist, Tutor library (Stein, 1990).

This experiment was designed in completely randomized plot design with 3 replications. Germination time in experiments was determined according to ISTA (2009) rules.

All characters means were subjected to analysis of variance (ANOVA) using SAS (1998) program and differences among treatments were separated with Duncan Multiple Range Test. Before the analyses, data was normalized with arcsine transformation.

RESULTS AND DISCUSSIONS

GC-MS analysis, a total of 6 components was identified in oregano oil. The major component in oregano oil was carvacrol (91.39%). The other components were cymene, gamma terpinen, linalool, isoborneol and myrecene, 4.01, 1.55, 1.51 1.44 and 0.10%, respectively (Table 1). Species, oil doses and their interaction was significant ($P<0.01$ level) for all the investigated characters in petri and pot experiments (Table 2).

Table 1. The essential oil composition of oregano oil

Components	RT	Rate (%)
Gamma terpinene	17.6	1.55
Cymene	19.2	4.01
Linalool	39.9	1.51
İsoborneol	46.1	1.44
Myrecene	50.1	0.10
Carvacrol	74.2	91.39

RT: Retention time

Essential oils have different effects on plant growth one of which is the inhibition of germination (Foe et al., 2002; Barney et al., 2005). As seen in Table 3, germination rates were decreased with the increased doses of oregano oil in both petri and pot experiments. In petri experiment, dock and sunflower seeds at doses above 3 µl and chickpea seed at doses above 10 µl were did not germinate. While amaranth and mustard seeds germinated only in the control application, oregano oil completely inhibited germination of amaranth and mustard seeds. Wheat seeds in control application completely germinated, but germination decreased with higher doses of oregano oil at the lowest germinate was observed in 20 µl. In pot experiment, while germination of wheat and chickpea seeds were not affected by 1.0 mg kg^{-1} dose compare to control application, germination of sunflower seeds were decreased by 67.4% at the same dose. Germination of

dock, amaranth and mustard seeds decreased by 39.2, 89.3 and 76.8%, respectively, at 1.0 mg kg^{-1} oregano oil application.

Dudai et al. (2000) indicated that monoterpenes, comprising essential oils, inhibit germination of seeds at low levels and the plants exposed to monoterpene steam were damage seriously.

Azirak and Karaman (2008), reported that while 3 and 6 µl application to *Coriandrum sativum, Foeniculum vulgare, Lavandula stoechas, Pimpinella anisum, Rosmarinus officinalis* and *Salvia officinalis* essential oils did not affected germination, 10 and 20 µl application reduced germination of 7 weed seeds significantly; namely *Alcea pallida, Amaranthus retroflexus, Centaurea salsotitialis, Raphanus raphanistrum, Rumex nepalensis, Sinapis arvensis* and *Sonchus oleraceus*.

Allelochemicals affect physiological interactions such as photosynthesis and respiration, cell division, cell development, membrane permeability, and ion exchange (Ağar et al., 2006). Scrivanti et al. (2003) emphasized that essential oils destroyed cell organelles in root apical meristem and damaged cell membranes causing slow root growth.

The application of oregano oil has been adversely affected seedling root length in petri and pot experiments. Root length of dock was decreased by 93.8% compared to control application at 3 µl dose in petri experiment. While root length of sunflower, chickpea and wheat at control application were 10.31, 5.57 and 6.22 cm, their root lengths at 3 µl dose were 3.94, 3.85 and 3.58 cm, respectively.

Root lengths of dock, amaranth and mustard at 0.5 mg kg^{-1} dose in pot experiments were decreased 26.4, 8.5 and 25.5%, respectively. The roots of sunflower seedlings among crop plants have much less damaged compare to the roots of chickpea and wheat at 0.5 mg kg^{-1} (Table 4).

Monoterpenes limited oxygen intake thus preventing germination and growth. Penuelans et al., (1996) reported that α-pinene reduced oxygen consumption in soybean cotyledons, which prevented seed germination and plant growth. Averages of stem length of all species generally decreased with increasing essential oil doses in both of experiment. The stem length of dock, sunflower, chickpea and wheat at 3 µl dose compared to control application in petri experiment were decreased by 74.1, 6.2, 14.6, 26.4%, respectively.

In pot experiment, while stem length of sunflower, chickpea and wheat at control application were 20.82, 21.65 and 24.93 cm, their stem length at 0.5 mg kg^{-1} dose decreased to 18.39, 15.69 and 19.98 cm, respectively. The stem length of sunflower seedlings at the lowest doses at the both experiments (3 µl and 5 mg kg^{-1}) had shortened.

Topal and Kocaçalışkan (2006) reported juglon which is important component of walnut, decreased seedling length of *Sinapis arvensis* (48.1%), *Cirsium arvense* (79.0%) and *Lamium amplexicaule* (74.9%). Same researchers at the same time chlorophyll content decreased at higher doses. The same study, stem length of wheat and amount of chlorophyll decreased 11.1% and 20.2%, respectively.

Dry matter content of seedlings of all species was raised with increasing dose of oregano oil in both experiments (Table 6). The highest dry matter was determined in chickpea seedlings in petri experiment and in wheat seedling in pot experiments.

CONCLUSIONS

As a result, allelochemicals are important to avoid herbicide residues in crops, environmental damage. Essential oils, one of most important of these chemicals, can be used directly instead of herbicides, are the basis of new synthetic herbicides. In this study, germination of weed seeds significantly inhibited reduced at lower dose applications, while germination rates of cultivated species were not adversely affected at same doses

Table 2. The variance analysis for germination rate, root and stem length and dry matter of rate in petri and pot experiments

VS	DF	Petri				Pot			
		Germination rate MS	Root length MS	Stem length MS	Dry matter rate MS	Germination rate MS	Root length MS	Stem length MS	Dry matter rate MS
Species	5	7100.9**	24.5**	49.1**	1619.3**	7499.6**	88.9**	626.2**	1033.9**
Dose	4	97.7**	70.6**	64.9**	635.4**	12046.8**	140.4**	381.1**	1152.7**
Species x Dose	20	627.7**	6.7**	15.1**	503.9**	589.6**	18.8**	50.6**	661.9**
Error	60	11. 9	0.2	0.1	3.5	20.0	0.3	0.6	5.4
CV (%)		13.3	23.1	12.9	15.4	10.9	14.8	10.4	13.1

** Significant at P ≤ 0.01, CV: Coefficient of Variation, MS: Mean square

Table 3. Average germination rates (%) in petri and pot experiments

	Petri						Pot					
Species	**0 µl**	**3 µl**	**6 µl**	**10 µl**	**20 µl**	**Avr.**	**0 mg kg^{-1}**	**0.5 mg kg^{-1}**	**1.0 mg kg^{-1}**	**2.0 mg kg^{-1}**	**4.0 mg kg^{-1}**	**Avr.**
Dock	53.3 aC	45.3 bB	0.0 cC	0.0 cC	0.0 cB	**19.7**	74.0 aB	50.0 bD	45. 0 cB	5.0 dC	0.0 eB	34.8
Amaranth	16.0 aD	0.0 bD	0.0 bC	0.0 bC	0.0 bB	**3.2**	46.7 aC	25.0 bE	5.0 cD	0.0 cC	0.0 cB	15.3
Mustard	56.0 aC	0.0 bD	0.0 bC	0.0 bC	0.0 bB	**11.2**	72.0 aC	54.0 bB	16.7 cD	0.0 dC	0.0 dB	28.5
Sunflower	89.3 aB	14.0 bC	0.0 cC	0.0 cC	0.0 cB	**20.7**	92.0 aA	85.0 aB	30.0 bC	0.0 cC	0.0 cB	41.4
Chickpea	100.0 aA	95.3 aA	50.0 bB	7.0 cB	0.0 cB	**50.5**	100.0 aA	100.0 aA	96.7 aA	66.7 bB	0.0 cB	72.7
Wheat	100.0 aA	90.0 aA	66.7 bA	66.7 bA	15.0 cA	**67.7**	100.0 aA	100.0 aA	96.7 abA	90.0 bA	30.0 cA	83.3
Average	**69.1**	**40.8**	**19.5**	**12.3**	**2.5**		**80.8**	**69.0**	**48.4**	**26.9**	**5.0**	

*Differences between doses and species were indicated with lower and capital letters at the 1% level. Values within a column followed by the same letter are not significantly different.

Table 4. Average root lengths (cm) in petri and pot experiments

	Petri						Pot					
Species	**0 µl**	**3 µl**	**6 µl**	**10 µl**	**20 µl**	**Avr.**	**0 mg kg^{-1}**	**0.5 mg kg^{-1}**	**1.0 mg kg^{-1}**	**2.0 mg kg^{-1}**	**4.0 mg kg^{-1}**	**Avr.**
Dock	2.10 aD	0.13 bB	0.00 bB	0.00 bC	0.00 bB	**0.45**	3.11 aC	2.29 bC	1.90 bcC	1.40 cC	0.00 dB	**1.74**
Amaranth	3.49 aC	0.00 bB	0.00 bB	0.00 bC	0.00 bB	**0.70**	1.42 aD	1.30 aC	1.13 aC	0.00 bD	0.00 bB	**0.77**
Mustard	2.39 aCD	0.00 bB	0.00 bB	0.00 bC	0.00 bB	**0.48**	2.16 aD	1.61 bC	1.13 cC	0.00 dD	0.00 dB	**0.98**
Sunflower	10.31 aA	3.94 bA	0.00 cB	0.00 cC	0.00 cB	**2.85**	13.24 aA	10.76 aA	1.77 bC	0.00 bD	0.00 bB	**5.15**
Chickpea	5.57 aB	3.85 bA	3.25 bA	0.20 cB	0.00 cB	**2.57**	12.39 aA	9.13 bA	3.45 cB	2.06 cB	0.00 dB	**5.41**
Wheat	6.22 aB	3.58 bA	3.00 bA	2.04 cA	0.75 dA	**3.12**	10.13 aB	7.31 bB	6.12 cA	3.91 dA	2.80 eA	**6.05**
Average	**5.01**	**1.92**	**1.04**	**0.37**	**0.13**		**7.08**	**5.40**	**2.58**	**1.23**	**0.47**	

*Differences between doses and species were indicated with lower and capital letters at the 1% level. Values within a column followed by the same letter are not significantly different.

Table 5. Averages of stem length in petri and pot experiments (cm)

	Petri						Pot					
Species	**0 µl**	**3 µl**	**6 µl**	**10 µl**	**20 µl**	**Avr.**	**0 mg kg^{-1}**	**0.5 mg kg^{-1}**	**1.0 mg kg^{-1}**	**2.0 mg kg^{-1}**	**4.0 mg kg^{-1}**	**Avr.**
Dock	1.93 aD	0.50 bD	0.00 cC	0.00 cC	0.00 cB	**0.49**	4.29 aC	4.16 aC	3.20 bCD	2.40 cC	0.00 dB	**2.81**
Amaranth	2.90 aC	0.00 bD	0.00 bC	0.00 bC	0.00 bB	**0.58**	2.77 aD	2.75 aC	2.45 aDE	0.00 bD	0.00 bB	**1.59**
Mustard	1.94 aD	0.00 bD	0.00 bC	0.00 bC	0.00 bB	**0.39**	2.52 aD	2.49 aC	1.61 bE	0.00 cD	0.00 cB	**1.32**
Sunflower	11.16 aA	10.47 aA	0.00 bC	0.00 bC	0.00 bB	**4.33**	20.82 aB	18.39 bA	4.64 cC	0.00 dD	0.00 dB	**8.77**
Chickpea	5.63 abB	4.81 bB	5.81 aA	1.90 cB	0.00 dB	**3.63**	21.65 aB	15.69 bB	10.26 cB	4.59 dB	0.00 eB	**10.44**
Wheat	5.31 aB	3.91 bC	2.68 cB	2.25 dA	1.85 dA	**3.20**	24.93 aA	19.98 bA	18.58 bA	13.59 cA	11.50 cA	**17.72**
Average	**4.81**	**3.28**	**1.42**	**0.69**	**0.31**		**12.77**	**10.58**	**6.84**	**3.43**	**1.92**	

*Differences between doses and species were indicated with lower and capital letters at the 1% level. Values within a column followed by the same letter are not significantly different.

Table 6. Averages of dry matter rates in petri and pot experiments (%)

	Petri						Pot					
Species	**0 µl**	**3 µl**	**6 µl**	**10 µl**	**20 µl**	**Avr.**	**0 mg kg^{-1}**	**0.5 mg kg^{-1}**	**1.0 mg kg^{-1}**	**2.0 mg kg^{-1}**	**4.0 mg kg^{-1}**	**Avr.**
Dock	6.26 bE	21.00 aB	0.00 cC	0.00 cC	0.00 cB	**5.45**	12.47 cC	19.70 bB	27.60 aBC	28.50 aA	0.00 dB	**17.65**
Amaranth	8.37 aDE	0.00 bD	0.00 bC	0.00 bC	0.00 bB	**1.67**	15.60 bC	24.47 aB	2.50 cE	0.00 aC	0.00 aB	**8.51**
Mustard	53.10 aA	0.00 bD	0.00 bC	0.00 bC	0.00 bB	**10.62**	47.80 bA	63.60 aA	33.67 cAB	0.00 dC	0.00 dB	**29.01**
Sunflower	16.83 bC	21.94 aB	0.00 cC	0.00 cC	0.00 cB	**7.75**	12.70 cC	21.47 bB	39.37 aA	0.00 dC	0.00 dB	**14.71**
Chickpea	11.53 bD	12.34 bC	16.79 bB	44.00 aA	0.00 aB	**16.93**	7.30 cD	8.50 cC	13.90 bD	23.90 aB	0.00 dB	**10.72**
Wheat	29.93 bcB	27.53 cA	29.54 bcA	31.23 bB	34.80 aA	**30.61**	19.50 eB	21.40 dB	23.47 cC	28.30 bA	39.03 aA	**26.34**
Average	**21.00**	**13.80**	**7.72**	**12.54**	**5.80**		**19.23**	**26.52**	**23.42**	**13.45**	**6.51**	

*Differences between doses and species were indicated with lower and capital letters at the 1% level. Values within a column followed by the same letter are not significantly different.

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