

Journal of Herbal Drug

journal homepage: www.jhd.iaushk.ac.ir



Effects of Jasmonic acid on essential oil yield and chemical compositions of two Iranian landraces of basil (*Ocimum basilicum*) under reduced irrigation

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ARTICLE INFO

Type: Original Research **Topic:** Physiology of Medicinal Plants **Received** September 19th 2014 **Accepted** February 18th 2015

Key words:

- ✓ Ocimum basilicum L.
- ✓ Jasmonic acid
- ✓ Secondary metabolites
- ✓ Reduced irrigation

ABSTRACT

Background & Aim: Sweet basil (*Ocimum basilicum*), a plant that is extensively cultivated in some countries, is used to enhance the flavour of salads, sauces, pasta, confectioneries and other products as both a fresh and dried herb. The effect of foliar application of Jasmonic acid and reduced irrigation on essential oil yield and chemical components of two Iranian landrace of basil (*Ocimum basilicum*) were investigated.

Experimental: Treatments comprised control, 0.0, 200, and 400 μ L Jasmonic acid applied to plants under normal irrigation and stressed conditions (30 and 60% F.C) based on a completely randomized design with three replications. The essential oils from the aerial parts of basil were analyzed by GC-FID and GC/MS.

Results & Discussion: Results indicated that the different levels of Jasmonic acid and irrigation had significant effects on oil yield and some main components of the essential oil. The highest value of oil content was obtained from application of 400 μ L JA. Percentage of some chemical constituents in the essential oil extracted from the plants under stress was higher than non-stressed plants. Foliar application of Jasmonic acid significantly improved methyl chavicol in the oils, but reduced Germacrene- D, α -Cadinol and δ -Cadinene amounts.

Industrial and practical recommendations: Since, essential oil of basil, particularly methyl chavicol component, have many application in pharmaceutical and perfumery industry and has health benefits such as antiviral, antibacterial and antispasmodic activity, we can use Jasmonic acid elicitor to enhance the therapeutic properties of basil and also essential oil content and methyl chavicol of plant.

1. Introduction

Ocimum basilicum L. belongs to the family Lamiaceae is an herb that is extensively cultivated in some countries. Arial parts, especially leaves of sweet basil

are widely used to enhance the flavour of foods such as salads, pasta, tomato products, vegetables, pizza, meat, soups, marine foods, confectioneries and other products (Ghasemi Pirbalouti, 2014). Traditionally basil has

been used as a medicinal plant in the treatment of headaches, coughs, diarrhea, constipation, warts, worms, and kidney malfunctions (Ghasemi Pirbalouti, 2014; Simon et al., 1984). In Iranian traditional medicine, the aerial parts of the plant are perceived as a galactogogue, stomachic, carminative, and antispasmodic (Sajjadi, 2006). Moreover, basil possesses a range of biological properties, including action as an insect repellent and nematocide (Deshpande and Tipnis, 1997), having antibacterial (Hussain et al., 2008), antiviral (Chiang et al., 2005), antifungal activity (Hussain et al., 2008), and antioxidant properties (Kwee and Niemeyer, 2011). Methyl chavicol, methyl cinnamate, methyl eugenol, citral, and linalool are generally the main chemotypes in sweet basil. Investigations (Sajjadi, 2006) on the chemical composition of the essential oil of basil, have demonstrated considerable variability.

In recent years, scientists have been looking for new alternatives to conventional methods for plant protection, and simultaneously for improvements in health properties and bioactive compound content. One of these methods is elicitation, which is induction of natural plant resistance mechanisms using biotic or abiotic factors. Elicitation can be an important strategy towards obtaining improved production of plant secondary metabolites *in vivo* (Hussain *et al.*, 2012). There are several commercially available chemical compounds that could be used as elicitors to modify plant secondary metabolites and subsequently the bioactivity of medicinal plants (Ghasemi Pirbalouti, *et al.*, 2014).

It has been reported that plant hormonal chemicals such as jasmonic acid (JA) and methyl jasmonate (MeJA), can used as elicitors (Rabea et al., 2003). Endogenous JA is a signaling compound that modulates various physiological processes in plants (Wasternack and Parthier, 1997). Therefore, exogenous JA has been used to induce useful secondary metabolites such as alkaloids, terpenoids, and phenolics in some plants such as Nicotina species, Hyoscyamus muticus, Norway spruce stems, and some plant cell cultures (Halitschke et al., 2000; Keina et al., 2001; Martin et al., 2002). The increased amounts of secondary metabolites increase not only plants' disease resistance but also their market values regarding the increased health benefits. Moreover, environmental factors such as moisture deficit are known to play an

important role in the synthesis of secondary metabolites in plants (Ghasemi Pirbalouti *et al.*, 2015). A lot of researches (Khalid, 2006; Bettaieb, *et al.*, 2009; Ghasemi Pirbalouti *et al.*, 2013) showed the effect of water stress on secondary metabolites such as essential oils in different plant species. The present study was performed to evaluate the effect of various concentrations of Jasmonic acid on essential oil yield and essential oil composition two landraces of basil (*Ocimum basilicum* L.) under water deficit.

2. Materials and Methods

2.1. Plant material and experimental site description

Plastic pots were filled with clay loam at a pH of 7.23, containing 0.8% organic matter comprised of 0.01% total N, 11.20 mg/kg available phosphorus, 694 mg/kg available potassium, and a saline value measured at E.C.: 1.35 dS/m.

Two Iranian landraces of sweet basil (*Ocimum basilicum* L.) seeds were obtained the Pakan Seed Company, Isfahan, Iran. In May 2014, pots transferred to the filed in Shahrekord (latitude $32^{\circ} 20^{\prime}$ N, longitude $50^{\circ} 51^{\prime}$ E, altitude 2061 m above sea level), southwestern Iran.

Type of study area climate by Emberger's climatology method is cold and semiarid and semi humid with temperate summer and very cold winter by Karimi's climatology method (IRIMO, 2012). In this study, no inorganic fertilizer and systemic pesticide was used during the entire experiment, and weed control was done manually.

2.2. Experimental design and treatments

Treatments included three irrigation regimes, viz., I_1 (unstressed or control), I_2 (irrigation in 70% field capacity when 30% of maximum total available soil water was depleted in the upper 30 cm of the soil profile), I_3 (irrigation in 40% field capacity when 60% of maximum total available soil water was depleted in the upper 30 cm of the soil profile) and three Jasmonic acid treatments (0.0, 200 and 400 µl) sprayed thrice at 10-12 leaves, before flowering, and two weeks later. Jasmonic acid was dissolved in ethanol 80%, diluted in distilled water with various concentrations. These solutions were sprayed at dew point (approximately 100 ml per plant) with a hand sprayer (untreated control plants were sprayed with an equivalent volume

of distilled water). The aerial parts of basil were collected after third spray of Jasmonic acid in July 2014.

2.3. Essential oil isolation

Dried plant material (100 g) was powdered and subjected to hydro–distillation for three hours using a Clevenger–type apparatus (British Pharmacopoeia, 1988). The essential oils were dried with anhydrous sodium sulphate and kept in amber vials at 4 °C prior to use.

2.4. Identification of the oil constituents

Composition of the essential oils was determined by gas chromatography (GC) and mass spectrophotometry (GC/MS). The GC analysis was done on an Agilent Technologies 7890 GC (Agilent Technologies, Santa Clara, CA) equipped with a single injector and a flame ionization detector (FID). A polar HP Innowax column and an apolar HP-5 capillary column (30 m x 0.25 mm, 0.25 μ m film thicknesses) coated with 5% phenyl, 95% methyl polysiloxane were used. The flow of the carrier gas (N2) was 0.8 ml/min. Initial column temperature was 60°C and programmed to increase at 4°C/min to 280°C. The injector temperature was set at 280 and 300°C. Split injection was conducted with a ratio split of 1:40. Essential oil samples of 0.1 μ L were injected neat (directly).

GC-MS analyses of aromatic oil samples were performed on an Agilent Technologies 7890 gas chromatograph coupled to Agilent 5975 C mass selective detector (MSD) and quadrupole EI mass analyzer (Agilent Technologies, Palo Alto, CA, USA). A HP-5MS 5% column (coated with methyl silicone) (30 m x 0.25 mm, 0.25 µm film thicknesses) was used as the stationary phase. Helium was used as the carrier gas at 0.8 mL/min flow rate. The temperature was programmed from 60 to 280°C at 4°C/min ramp rate. The injector and the GC-MS interface temperatures were maintained at 290°C and 300°C, respectively. Mass spectra were recorded at 70 eV. Mass range was from m/z 50-550. The ion source and the detector temperatures were maintained at 250 and 150°C, respectively.

Oil constituents were identified based on their retention indices (determined with reference to homologous series of C5-C24 *n*-alkanes), by comparison of their mass spectra with those reported in the literature (Adams, 2007) and stored in NIST 08

(National Institute of Standards and Technology) and Willey (ChemStation data system) libraries. The peak area percentages were computed from HP-5 column without the use of FID response factors.

2.5. Statistical analysis

Simple and interaction effects of experimental factors were derived from two-way analysis of variance (ANOVA) based on the GLM procedure of the SAS statistical package (SAS/STAT® v.9.2. SAS Institute Inc., Cary, NC).

The assumptions of variance analysis were tested by ensuring that the residuals were random and homogenous, with a normal distribution about a zero mean. The significance of differences among treatment means was tested using Duncan's multiple range test (DMRT) at $p \le 0.05$.

3. Results and discussion

3.1. Essential oil yield

All essential oils extracted from the aerial parts of two landraces of basil under different treatments were clear and yellow liquid. An analysis of variance indicated that different irrigation regimes had a significant effect on oil yield of basil ($p \leq 0.01$) (Table 1). The highest oil yield was obtained from mild drought stress treatment (I_3) with 0.82 ml/100 g dry material (Table 1). Our results indicated that Jasmonic acid at 400 µl produced maximum oil content (0.53 ml/100 g) (Table 1). It has been demonstrated that signal molecules such as Jasmonic acid are very potential elicitors for induction of plant secondary metabolites (Zhao et al., 2005). Recent years, the applications of signal components as elicitors have evolved an effective strategy for the production of target secondary metabolites in plant. Foliar application of Jasmonic acid on oil content of basil was influenced $(p \le 0.01)$ by irrigation regime (Table 2). The highest oil yield (0.89 ml/100 g) was achieved by foliar spray of 400 µL Jasmonic acid under reduced irrigation or mild stress drought (I3 \times J4) (Table 2). The lowest oil content was obtained when Jasmonic acid was applied under normal irrigation (Table 2).

There was significant jasmonic acid × landrace interaction effect on oil content (Table 3). In this study, landrace × irrigation had significant effects on oil content ($p \le 0.01$) (Table 4).

Treatments	Oil yield (ml/100 g)	1,8-Cineole	Linalool	Methyl chavicol	Nerol	Neral	Geranial	β- Caryophyllene	α-Humulene	Germacrene- D	δ-Cadinene	Caryophyllene oxide	α- Cadinol
Irrigation													
I1 (control)	0.183c	1.120b	4.39	33.24a	1.44b	6.30	7.10	4.82	3.12ab	2.98a	1.79a	1.854b	5.92a
I2 (60% FC)	0.488b	1.349a	4.41	34.71ab	1.77a	5.93	6.93	4.55	2.88b	2.66b	1.59b	1.79b	4.79b
I3 (30% FC)	0.822a	1.478a	4.62	35.67a	1.51b	5.74	6.82	4.99	3.16a	2.43b	1.41c	2.24a	5.42a
ANOVA	$p \le 0.01$	$p \le 0.01$	ns	$p \le 0.05$	$p \le 0.01$	ns	ns	ns	$p \le 0.05$	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	<i>p</i> ≤ 0.01
Jasmonic acid													
J1 (control)	0.471c	1.317a	4.44a	34.55ab	1.63ab	6.39a	7.36a	0.49ab	0.99a	4.98a	3.35a	2.70ab	1.68ab
J2 (ethanol)	0.467d	1.316a	4.23a	32.91b	1.51b	5.81ab	6.65a	0.50a	1.09a	5.15a	3.18ab	2.89a	1.78a
J3 (200)	0.518b	1.251a	4.48a	34.41ab	1.79a	6.26ab	7.27a	0.44b	0.99a	4.88a	2.98bc	2.66ab	1.54bc
J4 (400)	0.535a	1.378a	4.74a	36.29a	1.28c	5.50b	6.51a	0.45ab	0.98a	4.14b	2.71c	2.51b	1.39c
ANOVA	$P \le 0.01$	ns	ns	$P \le 0.05$	$P \le 0.01$	ns	ns	ns	ns	$P \leq 0.01$	$P \le 0.01$	ns	<i>P</i> ≤ 0.01
Landraces													
S1 (Green)	0.71a	0.321b	1.50b	28.10b	2.84a	10.83a	12.64a	6.86a	3.79a	2.65	0.32b	2.85a	2.12b
S2 (Purple)	0.285b	2.31a	7.45a	40.98a	0.27b	1.14b	1.25b	2.71b	2.33b	2.73	2.87a	1.06b	8.64a
ANOVA	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	ns	$p \le 0.01$	$p \le 0.01$	$p \leq 0.01$

Table 1. The effects of irrigation regime, jasmonic acid and landrace on the essential oil from basil

ns: non-significant

Table 2. The interaction effects of irrigation regime × jasmonic acid on the essential oil from basil

Interaction effect	Oil yield	1,8- Cineole	Linalool	Methyl chavicol	Nerol	Neral	Geranial	β- Caryophyllene	α- Humulene	Germacrene- D	δ- Cadinene	Caryophyllene oxide	α- Cadinol
$I1 \times J1$	0.175i	1.00cd	3.84c	29.34e	1.46cd	6.28ab	6.88ab	5.56a	3.52a	3.47a	2.31a	2.15ab	7.28a
$I1 \times J2$	0.175i	1.22bc	4.72abc	34.63bcd	1.42cde	6.33ab	7.11ab	5.03ab	3.30abc	2.86bc	1.76bc	1.94a-d	6.02b
$I1 \times J3$	0.179i	0.80d	3.94bc	35.21bcd	1.44cde	6.35ab	7.24ab	4.77abc	3.05abcde	2.96b	1.52cdef	1.84bcde	4.94bcd
$I1 \times J4$	0.204h	1.44ab	5.06a	33.79bcd	1.44cd	6.23ab	7.16ab	3.94cd	2.61ef	2.63bcd	1.58cde	1.43e	5.44bc
$I2 \times J1$	0.450g	1.42ab	4.36abc	32.09de	1.70abc	5.64ab	6.59ab	4.62bcd	2.80-f	2.70bcd	1.60cd	1.56cde	4.81cd
$I2 \times J2$	0.462f	1.19bc	3.88bc	32.12de	1.87ab	6.76a	7.90a	4.82abc	3.35abc	2.88bc	1.76bc	2.04abc	4.95bcd
$I2 \times J3$	0.525d	1.33abc	4.69abc	34.85bcd	1.95ab	6.09ab	7.24ab	4.91ab	2.89c-f	2.55bcd	1.53cdef	2.08ab	5.31bc
$I2 \times J4$	0.513e	1.44ab	4.70abc	39.80a	1.32de	5.21b	5.99b	3.85d	2.51f	2.52bcd	1.32ef	1.50de	4.08d
$I3 \times J1$	0.775c	1.51ab	4.49abc	37.30ab	1.36cde	5.50ab	6.47ab	5.28ab	3.22a-d	2.51bcd	1.43def	2.40a	5.56bc
$I3 \times J2$	0.775c	1.53ab	4.73abc	36.91abc	1.61bcd	6.07ab	7.07ab	5.08ab	3.42ab	2.37d	1.38def	2.26a	5.37bc
$I3 \times J3$	0.850b	1.60a	4.81ab	33.15cde	1.99a	6.32ab	7.32ab	4.96ab	3.01b-e	2.47cd	1.57cde	1.96a-d	5.46bc
$I3 \times J4$	0.888a	1.25bc	4.45abc	35.29bcd	1.08e	5.05b	6.39ab	4.65bcd	3.01b-e	2.38d	1.26f	2.36a	5.31bc
ANOVA	$p \le 0.01$	$p \le 0.01$	ns	$p \le 0.01$	$p \le 0.05$	ns	ns	ns	ns	ns	$p \le 0.05$	$p \le 0.05$	$p \le 0.05$

Interaction effect	Oil yield	1,8- Cineole	Linalool	Methyl chavicol	Nerol	Neral	Geranial	β- Caryophyllene	α- Humulene	Germacrene - D	δ- Cadinene	Caryophyllene oxide	α- Cadinol
$J1 \times S1$	0.683c	0.25b	1.20c	28.92d	2.74b	10.46a	12.07a	7.33a	3.96a	2.81ab	0.33d	2.94a	2.22c
$J1 \times S2$	0.250g	2.38a	7.26a	36.90c	0.27d	1.15b	1.23b	2.98c	2.40d	2.98a	3.24a	1.14bc	9.54a
$J2 \times S1$	0.683c	0.24b	1.24c	28.37d	2.94b	11.46a	13.26a	7.08a	3.82ab	2.79ab	0.31d	2.80a	2.12c
$J2 \times S2$	0.258f	2.39a	7.65a	40.73b	0.32d	1.31b	1.46b	2.87cd	2.89c	2.62ab	3.05a	1.36b	8.77ab
J3 × S1	0.733b	0.31b	1.55bc	28.95d	3.26a	10.99a	12.87a	6.93a	3.80ab	2.50b	0.32d	2.93a	2.11c
$J3 \times S2$	0.303e	2.19a	7.40a	39.86bc	0.33d	1.52b	1.67b	2.83cd	2.17de	2.81ab	2.75b	0.99bc	8.36b
J4× S1	0.742a	0.47b	2.00b	26.15d	2.42c	10.41a	12.37a	6.11b	3.56b	2.51b	0.31d	2.74a	2.01c
$J4 \times S2$	0.328d	2.27a	7.47a	46.43a	0.14d	0.59b	0.65b	2.17d	1.85e	2.50b	2.46c	0.78c	7.87b
ANOVA	$p \le 0.01$	ns	ns	$p \le 0.01$	$p \le 0.05$	ns	ns	ns	$p \le 0.05$	ns	$p \le 0.01$	ns	ns

Table 3. The interaction effects of jasmonic acid × landrace on the essential oil from basil.

Table 4. The interaction effects of irrigation regime × landrace on the essential oil from basil.

Interaction effect	Oil yield	1,8- Cineole	Linalool	Methyl chavicol	Nerol	Neral	Geranial	β- Caryophyllene	α- Humulene	Germacrene- D	δ- Cadinene	Caryophyllene oxide	α-Cadinol
$I1 \times S1$	0.225d	0.37d	1.72c	27.71cd	2.72b	11.35a	12.98a	6.65b	3.71b	2.75b	0.31d	2.67b	2.22cd
$I1 \times S2$	0.142e	1.86c	7.05b	38.77b	0.16c	1.25b	1.22b	3.00c	2.54c	3.21a	3.28a	1.01c	9.62a
$I2 \times S1$	0.750b	0.29d	1.52c	30.37c	3.06a	10.49a	12.22a	6.33b	3.45b	2.73b	0.35d	2.37b	1.54d
$I2 \times S2$	0.225d	2.40b	7.29b	39.05b	0.36c	1.36e	1.63b	2.77c	2.32cd	2.60bc	2.83b	1.22c	8.04b
$I3 \times S1$	1.15 a	0.29d	1.25c	26.21d	2.74b	10.65a	12.72a	7.61a	4.20a	2.48bc	0.30d	3.52a	2.59c
$I3 \times S2$	0.488c	2.66a	1.99a	45.12a	0.28c	0.82b	0.91b	2.37c	2.13d	2.38c	2.52c	0.96c	8.26b
ANOVA	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	$p \le 0.01$	ns	ns	ns	$p \le 0.01$	$p \le 0.01$	$p \le 0.05$	$p \le 0.01$	$p \le 0.01$	$p \le 0.05$

 Table 5. The interaction effects of irrigation regime × .jasmonic acid × landrace on the essential oil from basil.

Interaction effect	Oil yield	1,8-Cineole	Linalool	Methyl chavicol	Nerol	Neral	Geranial	β- Caryophyllene	α- Humulene	Germacrene- D	δ- Cadinene	Caryophyllene oxide	α- Cadinol
$I1 \times J1 \times S1$	0.225k	0.34h	1.32fg	23.70jk	2.74cde	11.28abc	12.65a-d	7.71ab	4.13ab	3.19abc	0.33g	2.98bcd	1.92fgh
$I1 \times J1 \times S2$	0.125n	1.67fg	6.35de	34.99de	0.17gh	1.28de	1.12e	3.40gh	2.91efg	3.75a	4.30a	1.33gh	12.63a
$I1 \times J2 \times S1$	0.225k	0.40h	1.63fg	27.23g-j	2.64cde	11.48ab	13.07a-d	7.07a-d	3.85a-d	2.89b-f	0.31g	2.79cde	2.12gh
$I1 \times J2 \times S2$	0.125n	2.04ef	7.80abc	42.03bc	0.20gh	1.18de	1.15e	3.00g-j	2.77fgh	2.83b-f	3.21bc	1.09hi	9.93b
$I1 \times J3 \times S1$	0.02001	0.21h	1.95fg	33.15def	2.60de	10.34abc	12.04a-d	5.78ef	3.42c-f	2.61c-f	0.32g	2.57c-f	2.87g
$I1 \times J3 \times S2$	0.158m	1.39g	5.92e	37.28cd	0.27gh	2.37d	2.44e	3.76g	2.69ghi	3.31ab	2.72de	1.11hi	7.01e
$I1 \times J4 \times S1$	0.250j	0.52h	1.98fg	26.78h-k	2.88bcd	12.32a	14.16ab	6.05def	3.44c-f	2.32def	0.29g	2.33def	1.98fgh
$I1 \times J4 \times S2$	0.158m	2.35cde	8.13ab	40.79bc	0.01h	0.15e	0.16e	1.82j	1.77k	2.95b-e	2.88cd	0.53i	8.89bcd
$I2 \times J1 \times S1$	0.700f	0.15h	1.11fg	32.80de	3.13bc	10.26abc	11.91a-d	6.44c-f	3.42c-f	2.81b-f	0.33g	2.08f	1.73fgh
$I2 \times J1 \times S2$	0.2001	2.68a-d	7.62a-d	31.38e-h	0.28gh	10.03de	1.26e	2.79g-j	2.17h-k	2.60c-f	2.88cd	1.04hi	7.89cde
$I2 \times J2 \times S1$	0.700f	0.12h	0.99g	29.51e-i	3.31ab	11.73ab	13.64abc	6.52b-f	3.46b-e	2.96bcd	0.31g	2.10ef	1.56gh
$I2 \times J2 \times S2$	0.225g	2.26de	6.77cde	34.72de	0.43gh	1.78de	2.15e	3.11ghi	3.23d-g	2.80b-f	3.49b	1.98fg	8.33b-e
$I2 \times J3 \times S1$	0.850d	0.42h	1.58ab	38.83f-j	3.34ab	10.25abc	12.21a-d	7.04a-e	3.71a-d	2.39def	0.37g	3.12abc	2.09fgh
$I2 \times J3 \times S2$	0.2001	2.25de	7.79abc	40.88bc	0.56g	1.93de	2.27e	2.77g-j	2.07ijk	2.71b-f	2.69de	1.03hi	8.53b-e
$I2 \times J4 \times S1$	0.750e	0.47h	2.41f	30.36e-i	2.47de	9.71bc	11.13d	5.30f	3.22d-g	2.76b	0.38g	2.18ef	0.77h
$I2 \times J4 \times S2$	0.275i	2.41b-e	6.99b-e	49.23a	0.16gh	0.71de	0.85e	2.40hij	1.80k	2.28ef	2.25f	0.81hi	7.39de
$I3 \times J1 \times S1$	1.125c	0.24h	1.16fg	30.27e-i	2.36ef	9.85bc	11.64cd	7.83a	4.32a	2.42def	0.33g	3.76a	3.02fg
$I3 \times J1 \times S2$	0.425h	2.79abc	7.82abc	44.34ab	0.36gh	1.15de	1.31e	2.73g-j	2.12h-k	2.59c-f	2.53def	1.04hi	8.09cde
$I3 \times J2 \times S1$	1.125c	0.20h	1.10g	28.38f-j	2.87bcd	11.17abc	13.06a-d	7.65abc	4.17a	2.51c-f	0.31g	3.52ab	2.67fg
$I3 \times J2 \times S2$	0.425h	2.86ab	8.36a	45.44ab	0.35gh	0.97de	1.09e	2.50g-j	2.66g-j	2.23f	2.45ef	0.99hi	8.06cde
$I3 \times J3 \times S1$	1.150b	0.29h	1.12fg	24.87ijk	3.82a	12.38a	14.35a	7.96a	4.28a	2.52cdef	0.28g	3.09abc	1.37gh
$I3 \times J3 \times S2$	0.550g	2.92a	8.49a	41.44bc	0.17gh	0.26de	0.30e	1.95ij	1.74k	2.42def	2.86cd	0.82hi	9.54bc
$I3 \times J4 \times S1$	1.225a	0.43h	1.61fg	21.32k	1.92f	9.19c	11.84bcd	6.99а-е	4.03abc	2.46def	0.27g	3.72a	3.28f
$I3 \times J4 \times S2$	0.550g	2.06ef	7.29a-d	49.27 a	0.24gh	0.91de	0.94e	2.30hij	1.99jk	2.29def	2.24f	0.99hi	7.34de
ANOVA	$p \le 0.01$	$p \le 0.05$	$p \le 0.05$	$p \le 0.01$	$p \le 0.01$	$p \le 0.05$	$p \le 0.01$	ns	ns	ns	$p \le 0.01$	$p \le 0.05$	$p \le 0.01$

The highest oil yield was observed in the green landrace of basil under reduced irrigation or mild stress drought (Table 4). In this study, jasmonic acid × irrigation × landrace had significant effects ($p \le 0.01$) on oil yield (Table 5). The highest oil yield (1.22 ml / 100 g dry weight) was achieved by foliar spray of jasmonic at 400 µl under reduced irrigation in the green landrace of basil (Table 5).

Similarly, Simon et al. (1992) reported that water stress increased essential oil accumulation via higher density of oil glands due to the reduction in leaf area. Moreover, Shokrani et al. (2012) reported an increase in essential oil content and yield in basil with increasing water stress severity and higher yield of essential oil obtained from mild water stress. In general, moisture deficit increases the essential oil content of more medicinal and aromatic plants, because in case of stress, more metabolites are produced in the plants and substances prevent from oxidization in the cells. Essential oil helps to plant that easier adopt to the environmental stress conditions. In addition, stress induced alterations in oil accumulation are considered to be mainly due to its effect on plant growth and differentiation, due to the fact that plants produce high terpene concentrations under water deficit conditions due to a low allocation of carbon to the growth, suggesting a trade-off between growth and defense (Turtola et al., 2003). Probably, improvement in oil yield by exogenous application of Jasmonic acid might be due to the increase in cycle growth, nutrients uptake or changes in leaf oil gland population and monoterpenes biosynthesis (Ghasemi Pirbalouti et al., 2014). Boonlertnirun et al. (2008) found that foliar application of elicitor had a significant effect on the growth, and yield of drought-stressed rice plants compared to control plants.

3.2. Chemical compositions of oil

The chemical constituents of the essential oil identified by GC-FID and GC/MS. Tables 1-5 show the effect of different treatments on the chemical composition of essential oil extracted from two landraces of basil. The major components in the essential oil were methyl chavicol, gernial, neral, and β -caryophyllene (Table 1). Our results demonstrated that the essential oils obtained from the landraces of basil contained phenylpropanoids (methyl chavicol), oxygenated monoterpenes (linalool, gernial, and neral),

sesquiterpenes hydrocarbons (β -caryophyllene), and oxygenated sesquiterpenes (α -cadinol), confirming earlier reports that major chemical groups obtained from the aerial parts of basil were phenylpropanoids and oxygenated monoterpenes (Sajjadi, 2006; Ghasemi Pirbalouti *et al.*, 2013; Ghasemi Pirbalouti, 2014).

An analysis of variance indicated that the different factors had a significant effect on some major constituents of the essential oil from basil. Results also indicated that there were significant differences between Jasmonic acid, and landraces treatments in term methyl chavicol (Table 1). Results of mean comparison revealed that amount of methyl chavicol, as a main compound in the basil oil, not considerably affected by drought levels (Table 1). Different levels of the foliar application of Jasmonic acid had significant effects on the major constitutes of the essential oils (Table 1). This demonstrated that Jasmonic acid at 400 µL produced the highest levels of methyl chavicol (36.29%), linalool (4.74%), and 1,8-Cineole (1.37%), but at 200µL produced the highest levels of nerol (1.79%), geraniol (0.94%) (Table 1).

Results of interaction effects of irrigation × Jasmonic acid indicated that the highest contents of some main components were obtained from the application of Jasmonic acid under reduced irrigation (Table 2). Our results further indicated that foliar application of Jasmonic acid reduced the effect of water deficit stress on amounts of linalool, methyl chavicol, and geranial, the major compounds in the essential oil from the landraces of basil (Table 2). Jasmonic acid may play an important signaling role in the activation of various plant defense responses, such as the biosynthesis of special secondary metabolites.

In present research, the ANOVA showed that interaction effects of landrace \times jasmonic acid had significant effects on the major constitutes of basil essential oils (Table 3). In addition, interaction effects of irrigation \times landrace had significant effects on many volatile constituents (Table 4). Morevere, interaction effects of jasmonic acid \times irrigation \times landrace had significant effects on many volatile constituents (Table 5). The maximum amount of methyl chavicol (49.27%) was obtained by 400 µl jasmonic acid in the purple landrace under mild drought stress.

4. Conclusions

In this research, effects of foliar application of jasmonic acid and drought stress on yield and chemical composition of essential oils of two landraces of basil have evaluated. The investigation showed two landraces of basil are sensitive to the application of elicitors such as jasmonic acid and water deficit. The results revealed that essential oil yield two landraces of basil increased with application of elicitors under drought stress. Not only oil ratio but also oil composition was affected since some components were increased and other components were decreased. Finally, it could be concluded that foliar application of jasmonic acid and deficit irrigation, as a possible technique, can be used to increase volatile oil ratio in basil cultivation. This study provides the basis for further research on improving the quality and prohealth functional value of two landraces of basil using elicitors.

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