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Chemical composition and yield of essential oil from lemon balm (*Melissa officinalis* L.) under foliar applications of jasmonic and salicylic acids

Abdollah Ghasemi Pirbalouti[a,*], Mahdieh Nekoei[b], Mehdi Rahimmalek[c], Fatemeh Malekpoor[b]

[a] *Research Center for Medicinal Plants, Shahr-e-Qods Branch, Islamic Azad University, Tehran, Iran*
[b] *Department of Medicinal Plants, Shahrekord Branch, Islamic Azad University, P O Box: 166, Shahrekord, Iran*
[c] *Department of Agronomy and Plant Breeding, College of Agriculture, Isfahan University of Technology, Isfahan, 84156 83111, Iran*

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A B S T R A C T

Jasmonic acid (JA) is regarded as endogenous regulator that plays important roles in regulating stress responses, plant growth, and development. Salicylic acid (SA) has been identified as an important signaling element involved in establishing the local and systemic disease resistance response of plants after pathogen attack. A field experiment was conducted to assess the foliar applications effect of JA and SA on quantity and quality yields of essential oil of lemon balm (*Melissa officinalis* L.). Experimental treatments were: *I)* water foliar application; *II)* water + 1% ethanol foliar application (as a solvent); *III-V)* JA at 0.05–0.40 mg L^{-1}; *VI-IX)* SA at 0.14–14.00 g L^{-1}. The essential oil contents of the treatments ranged between 0.55 to 0.61% (v/w). The results of analysis of the essential oils by GC-FID and GC/MS indicated that monoterpene oxygenated and sesquiterpenes were the major components. There were no significant differences among treatments for essential oil content, dry matter weight, and some of volatile components (neral and geranial). The foliar spray JA significantly improved the amounts of two phenolic monoterpenes compounds such as thymol from 0.42% in control to 4.37% in JA at 0.40 mg L^{-1} and carvacrol from 0.77% in control to 14.76% in JA at 0.40 mg L^{-1}. In addition, the highest percentages of β-caryophyllene and caryophyllene oxide were obtained from the foliar application of SA and JA. In conclusion, the exogenous application of JA and SA can considerably improve some secondary metabolites especially monoterpene oxygenated and sesquiterpenes in lemon balm plants under filed conditions.

1. Introduction

Lemon balm (*Melissa officinalis* L.), belonging to the family Lamiaceae, which it grows in various areas from the eastern Mediterranean regions to western Asia. In addition, *M. officinalis* is widely cultivated throughout Europe (Koytchev et al., 1999). It is a well-known herb used to give fragrance to different food and beverage products. The aerial parts of lemon balm are used for treatment of headaches, gastrointestinal disorders, nervousness, and rheumatism (Bisset and Wichtl, 2001). *M. officinalis* (Persian name: "Badrnajboyeh" or "Varangboo") is a plant growing and cultivated in some parts of Iran (Mozaffarian, 2008). In Iranian folk medicine, the aerial parts of lemon balm are used for treatment of indigestion, headaches, flatulence, colic, nausea, nervousness, anaemia, vertigo, syncope, malaise, asthma, bronchitis, amenorrhea, cardiac failure, arrhythmias, insomnia, epilepsy, depression, psychosis, hysteria, ulcers, and wounds and also as carminative, antispasmodic, sedative, analgesic, tonic, and anti-diarrheal (Ghasemi-Dehkordi, 2002).

The extracts and essential oil extracted from *M. officinalis* have been shown to have biological and pharmacological activities, such as an anti-bacterial (Mimica-Dukic et al., 2004; Canadanovic-Brunet et al., 2008), anti-fungal (Mimica-Dukic et al., 2004), anti-oxidant (http://www.sciencedirect.com/science/article/pii/S0023643807001247,Ivanova et al., 2005; Mimica-Dukic et al., 2004; Venkutonis et al., 2005; Dastmalchi et al., 2008), anti-viral (Allahverdiyev et al., 2004), insecticidal (Koliopoulos et al., 2010), antispasmodic (Masakova et al., 1979), anti-tumor (De Sousa et al., 2004), anti-glycative (Miroliaei et al., 2011), amoebicidal (Malatyali et al., 2012), anti-diabetic (Chung et al., 2010), and use for Alzheimer's disease (Akhondzadeh et al., 2003; Dastmalchi et al., 2009), neuro-

Abbreviations: JA, Jasmonic Acid; MJ, Methyl Jasmonate; SA, Salicylic acid; GC-MS, Gas Chromatography-Mass Spectrometry; CRD, Completely Randomized Design

* Corresponding author. Research Center for Medicinal Plants, Shahr-e-Qods Branch, Islamic Azad University, Tehran, Fath Highway, P.O. Box 37541-374, Iran. Tel. +98 21 4689 6512; fax: +98 21 4689 6513.
E-mail addresses: ghasemi955@gmail.com, ghasemi@qodsiau.ac.ir (A. Ghasemi Pirbalouti).

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protective (Bayat et al., 2012), and gastrointestinal spasms (Sadraei et al., 2003).

SA (2-hydroxybenzoic acid, $C_7H_6O_3$) naturally occurs in plants in very low concentration. SA is a common plant–produced signal molecule that is responsible for inducing tolerance to a number of biotic (infection caused by fungi, bacteria and viruses) and abiotic stresses (drought, high soil salinity, chilling, heat, UV light, ozone, and heavy metal) (Wang et al., 2004; Gharib, 2006; Idrees et al., 2010; Ghasemi Pirbalouti et al., 2014a,b; Dianat et al., 2016). (3 R, 7 R, 9Z)-JA ($C_{12}H_{18}O_3$) and its methyl ester (MJ) were first identified as odorant compounds in essential oil of jasmine (Ghasemi Pirbalouti et al., 2014c). Jasmonates are involved in many critical functions, including defense against insects and pathogens by inducing phytoalexin production, protection from abiotic stresses, immunity, and plant growth and development, suggesting that they have critical roles in plant physiology (Avanci et al., 2010; Ghasemi Pirbalouti et al., 2014c; Alavi-Samani et al., 2015; Talebi et al., 2018).

SA and JA play roles in the stimulation and production of volatile compounds, antioxidants, anthocyanin, phenolic and other main compounds in medicinal and aromatic plants. The hypothesis of this study was that elicitors JA and SA would have bioactivity or influence plant growth, the stimulation and production of secondary metabolites in lemon balm. Few studies have been carried out to investigate the effects of foliar application of chemical elicitors such as JA and SA on the growth, accumulation of essential oil, and chemical components profile of essential oil from lemon balm cultivated in agricultural systems. Thus, the main goals of this research was to evaluate the exogenous foliar of SA and JA on quantity and quality of yields of the essential oil from lemon balm for first time.

2. Materials and methods

2.1. Chemicals

Alkane standard solution C_5–C_{24}, JA and SA were purchased from Sigma–Aldrich Co. (Steineheim, Germany). Ethanol and anhydrous sodium sulphate were bought from Merck Co. (Darmstadt, Germany).

2.2. Plant material and field site description

M. officinalis seeds were obtained from the Pakan Seed Company, Isfahan, Iran. In the first week of March 2016, five seeds were sown in each plastic pot (20 cm diameter and 35 cm height), and after four weeks were thinned to two healthy seedlings per pot. The seedlings were transplanted to the experimental field in Saman (latitude, 32° 29′ N; longitude, 50° 54′ E; altitude, 1890 m above sea level), Southwestern Iran. The climate of study area is cold and semiarid by Emberger's climatology standard and semi humid with temperate summer and very cold winter by Karimi's climatology standard (annual average of temperature = 13.2 °C and annual average rainfall = 342 mm) (Chaharmahal Va Bakhtiari Meteorological, 2012). The soil characteristics were the following: loam sandy soil, pH 8.09, electrical conductivity, 0.49 dS m^{-1} and organic matter, 1.1%. Each experimental plot size was 2 m × 2 m with three replications and in each plot; plants were grown in six equidistant rows, 0.4 m apart. In this study, no inorganic fertilizer and systemic pesticide was used during the entire experiment, and weed control was done manually. For the first month (establishment phase), a watering level equivalent to 60–65% of soil field capacity was applied once a day. For the subsequent months, the plots were irrigated when 50% of soil available water was depleted (irrigation intervals varied from 2 to 3 days) by drip system irrigation. The aerial parts of *M. officinalis* were collected from each plot before flowering stage (85 days after planting), all of which were dried at 40 °C in the dark until it reached a constant weight.

2.3. Experimental design and treatments

The experiment was conducted in a complete randomized design (CRD) with three replications.

Experimental treatments, included:

(I) Water foliar application (control),
(II) Water + 1% ethanol foliar application (as a solvent),
(IIIsbndV) JA at 0.05, 0.10, 0.20, and 0.40 mg L^{-1},
(VI-IX) SA at 0.14, 1.40, 5.60, and 14.00 g L-1.

The experimental concentrations of JA and SA were used according earlier investigations (Ghasemi Pirbalouti et al., 2014c; Alavi-Samani et al., 2015; Talebi et al., 2018). The leaves were treated by foliar application twice on 15 April at 4 to 6 leaves stage and 4 May at 8 to12 leaves stage. Ten mL JA and SA solutions (adjusted to pH 7.0 with NaOH) were sprayed using an atomizer onto the leaves until it completely ran off. The control plants were sprayed with distilled water, also adjusted to pH 7.0 with NaOH.

2.4. Essential oil isolation

The aerial parts of *M. officinalis* were dried inside for 1 week at room temperature. The dried samples were ground to fine powder by using a blender (Moulinex food processor, Spain). Then, the aerial parts of *M. officinalis* were subjected to hydro-distillation for 3 h using a Clevenger-type apparatus according to the method recommended in British pharmacopoeia (The British Pharmacopoeia Commission, 1988). Samples were dried with anhydrous sodium sulphate and kept in amber vials at 4 ± 1 °C prior to use.

2.5. Gas chromatography/mass spectrometry (GC/MS) analysis

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) using a HP–5MS capillary column (30 m × 0.25 mm, 0.25 μm film thicknesses) coated with 5% phenyl, 95% methyl polysiloxane. The carrier gas was helium (99.999% pure) at a flow rate of 0.8 mL min^{-1}. Initial column temperature was 60 °C and programmed to increase at 4 °C min^{-1} to 280 °C. The injector temperature was set at 300 °C. Split injection was conducted with a ratio split of 1:40. Essential oil samples of 1 μL 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 × 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^{-1} flow rate. The temperature was programmed from 60 to 280 °C at 4 °C min^{-1} 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. The samples (1 μL) were injected neat with 1:40 split ratio.

2.6. Identification of components

Essential 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 percentage composition was computed from the GC peak areas without using any correction factors.

Table 1
The chemical compositions and contents of the essential oil and dry matter yield of *Melissa officinalis* L. under sprayed with various concentrations of JA and SA.

Characteristics	†RI	*ANOVA*	Water	Ethanol	Concentrations							
					Jasmonic acid (mg L^{-1})				Salicylic acid (g L^{-1})			
					0.05	0.10	0.20	0.40	0.14	1.40	5.60	14.00
Dry matter yield (g m^{-2})	–	$p > 0.05$	189.9 ± 27.1	191.1 ± 21.9	193.5 ± 19.5	186.4 ± 11.9	184.5 ± 9.8	201.3 ± 27.2	188.4 ± 20.9	195.2 ± 6.8	189.8 ± 14.4	179.8 ± 24.6
Oil yield (v/w %)	–	$p > 0.05$	0.59 ± 0.22	0.61 ± 0.11	0.53 ± 0.29	0.57 ± 0.09	0.59 ± 0.06	0.54 ± 0.19	0.54 ± 0.03	0.59 ± 0.17	0.60 ± 0.03	0.58 ± 0.10
Essential oil components (%)‡												
α-Pinene	940	–	0.06	–	–	–	–	-	tr¶	–	0.05	tr
Octen-3-ol	982	–	0.58	0.81	0.48	0.75	0.71	tr	0.8	0.82	0.51	0.86
β-Myrcene	995	–	0.27	0.25	tr	0.3	0.33	tr	0.21	0.32	0.24	0.25
α-Phellandrene	1011	–	0.05	–	–	tr	–	–	–	tr	0.04	tr
p-Cymene	1026	–	0.06	0.06	tr	tr	0.11	0.11	tr	0.13	0.05	0.05
β-Phellandrene	1030	–	0.11	0.17	–	–	–	–	–	tr	0.09	–
1,8-Cineole	1033	–	0.12	tr	–	–	–	–	–	–	0.11	–
(*Z*)- β-Ocimene	1038	–	0.31	tr	tr	–	0.05	–	tr	0.06	0.27	0.09
Benzene-acetaldehyde	1044	–	0.19	0.22	–	0.12	0.19	–	0.12	0.34	0.17	0.14
(*E*)-β-Ocimene	1048	–	0.19	1.27	tr	0.4	0.18	–	0.25	0.33	0.15	0.62
γ-Terpinene	1057	–	tr	–	–	–	–	1.48	–	0.07	–	tr
Linalool	1101	$p \le 0.05$	1.71 ± 0.11a§	2.04 ± 0.42 a	1.19 ± 0.33a	1.73 ± 0.1a	1.66 ± 0.22a	0.05 ± 0.05b	1.95 ± 0.27a	1.79 ± 0.18a	1.51 ± 0.22a	2.37 ± 0.39a
(*E*)-Rose oxide	1111	–	0.16	tr	tr	0.15	0.12	tr	0.11	0.11	0.14	0.15
Citronellal	1153	$p \le 0.05$	5.38 ± 0.88a	3.06 ± 0.67b	4.38 ± 0.77ab	5.46 ± 1.11a	3.89 ± 0.89ab	2.16 ± 0.61c	4.39 ± 0.87ab	5.21 ± 0.91a	4.02 ± 0.55ab	3.81 ± 0.76ab
α-Terpineol	1189	–	0.05	tr	–	–	–	–	–	–	0.16	tr
Nerol	1228	–	tr	0.37	tr	–	0.38	–	0.5	0.42	tr	0.48
Neral	1248	$p > 0.05$	31.93 ± 3.55	30.49 ± 2.77	31.74 ± 2.22	33.65 ± 2.01	31.17 ± 3.21	27.34 ± 4.11	32.87 ± 3.11	33.02 ± 2.65	28.32 ± 2.48	31.79 ± 3.07
Geraniol	1255	–	tr	tr	tr	–	tr	-	tr	0.18	0.24	0.24
Geranial	1281	$p > 0.05$	38.34 ± 1.91	35.63 ± 3.11	41.33 ± 3.61	40.12 ± 1.58	36.96 ± 2.17	34.26 ± 2.52	39.58 ± 1.91	37.8 ± 1.01	37.8 ± 2.78	38.02 ± 2.91
Essential oil components (%)‡												
Bornyl acetate	1286	–	0.09	tr¶	–	–	tr	–	–	0.06	0.08	tr
Thymol	1290	$p \le 0.01$	0.42 ± 0.04b	0.05 ± 0.00b	0.26 ± 0.11b	0.45 ± 0.03b	0.24 ± 0.15b	4.37 ± 0.61a	0.06 ± 0.11b	0.41 ± 0.19b	0.38 ± 0.11b	0.13 ± 0.27b
Carvacrol	1298	$p \le 0.01$	0.77 ± 0.22b	0.11 ± 0.03b	1.38 ± 0.52b	0.85 ± 0.63b	0.81 ± 1.01b	14.76 ± 3.26a	0.21 ± 0.16b	0.93 ± 0.36b	0.68 ± 0.38b	0.34 ± 0.12b
Methyl geranate	1321	–	1.01	tr	0.78	0.67	tr	–	0.78	0.68	0.9	0.72
Geranyl acetate	1381	$p \le 0.05$	0.05 ± 0.00c	0.48 ± 0.09bc	1.60 ± 0.28a	1.42 ± 0.06ab	1.38 ± 0.11ab	2.05 ± 0.16a	2.05 ± 0.53a	0.75 ± 0.24b	1.61 ± 0.49ab	1.73 ± 0.32ab
(*E*)-Caryophyllene	1415	$p \le 0.05$	4.31 ± 1.32b	3.61 ± 1.53b	3.55 ± 0.99b	3.12 ± 1.17b	4.84 ± 0.69ab	2.99 ± 0.11b	3.64 ± 0.29b	3.02 ± 0.76b	3.29 ± 1.01b	5.03 ± 0.91a
α-Humulene	1447	–	tr	0.56	0.25	–	0.37	–	0.24	0.18	0.28	0.34
β-Farnesene	1452	–	tr	0.06	–	tr	0.11	–	–	–	0.12	0.05
Germacrene-D	1475	–	0.11	1.1	0.52	0.47	0.38	–	0.24	0.13	0.36	0.49
Ionone	1480	–	–	–	1.4	–	0.13	tr	–	–	0.17	–
α-Farnesene	1503	–	–	0.09	–	–	0.14	–	0.15	tr	0.17	0.18
Cadinene	1517	–	–	0.34	–	–	0.06	–	0.12	–	0.28	0.16
Spathulenol	1570	–	–	–	–	–	–	–	0.55	0.16	0.19	tr
Caryophyllene oxide	1576		0.11 ± 0.77c	0.63 ± 0.59bc	0.05 ± 0.03c	0.38 ± 0.21c	1.74 ± 0.47a	0.05 ± 0.11c	0.05 ± 0.0c	0.48 ± 0.17bc	2.09 ± 0.41a	1.37 ± 0.48ab
Cadin-4-en-10-ol	1623	–	–	–	–	–	0.16	tr	–	–	–	0.13
Muurolol	1645	–	–	–	–	–	–	–	0.11	–	0.18	–

Values with a common letter are not significantly different using LSD at $p \le 0.05$.
† RI: Retention indices determined on HP-5MS capillary column.
‡ Calculated from total ion chromatogram (TIC) data.
¶ Trtrace (< 0.01%).
§ Values of major compounds (> 1% in total oil) are given as means ± SD.

2.7. Statistical analysis

The data was statistically analyzed using one way analysis of variance by the program SPSS. Treatment means were compared by least significant differences (LSD) at $P < 0.05$ level.

3. Results

3.1. Essential oil content

The color of the essential oil of *M. officinalis* extracted was light yellow. The essential oil contents of studied treatments ranged between 0.53 to 0.61% (v/w), based on dry weight. Statistical analysis indicated that there were no significant differences among treatments for essential oil contents of lemon balm (Table 1).

3.2. Chemical compositions of the essential oil

According to results of GC-FID and GC/MS analysis 35 components were identified representing more than 85–97% of the essential oil composition (Table 1). The analysis of essential oils indicated some major constituents, viz. geranial, neral, carvacrol, β-caryophyllene, citronellal, thymol, and geranyl acetate. The major chemical components in the essential oils from *M. officinalis* contained oxygenated monoterpenes and sesquiterpenes (Table 1). The result of analysis of variance of this investigation indicated that different levels of the foliar application of JA and SA have significant impacts on some main constitutes in the essential oil from the *M. officinalis* leaves (Table 1), however, JA and SA did not influence on the concentrations of geranial and neral in the essential oil of the herb. The foliar applications of SA and JA significantly improved sesquiterpenes hydrocarbons and oxygenated sesquiterpenes such as β-caryophyllene and caryophyllene oxide (Table 1). On the other hand, foliar application of JA increased oxygenated monoterpenes such as carvacrol and thymol (phenolic monoterpenes compounds), and gerenyl acctate (Table 1).

3.3. Herbage dry weight

In this study, the foliar applications of JA and SA and JA had no significant effects on herbage yield of lemon balm (Table 1).

4. Discussion

Growth and quantity and quality yields of medicinal and aromatic plants, especially essential oil content and chemical compositions are known to be affected by genetic (ecotype, chemotype, cultivar etc.), agronomic managements, ecological and environmental factors, and their interaction effects. In this study, the foliar spray of SA and JA had not significantly impacts on dry herbage weight and essential oil content of *M. officinalis*. Similarly, Ashrafi et al. (2012) reported that the foliar application of JA (50–400 $\mu g\,L^{-1}$) did not effect on dry herbage and essential oil yields of Iranian thyme (*Thymus daenensis* Celak.). Results a study (Rahimmalek et al., 2012) stated that the foliar spray of SA (1–40 $mol\,L^{-1}$) and JA (50–400 $\mu g\,L^{-1}$) had not significantly effects on leaf biomass and essential oil yield of sage (*Salvia officinalis* L.). Opposing results, however, indicated that the exogenous application of MeJA (0.5 mM) resulted in higher essential oil of Basil (*Ocimum basilicum* L.) (Talebi et al., 2018). In addition, results of an investigation by Gharib (2006) indicated that the foliar application of SA at various concentrations (10^{-5}-10^{-3} M) increased biomass and essential oil yields of sweet basil (*Ocimum basilicum*) and marjoram (*Majorana hortensis*). Different results in these phytohormones effects on essential oil content may be related to concentration level, species as well as environmental conditions.

Plant secondary metabolites are unique sources for pharmaceuticals, food additives, flavors and other industrial materials. 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. Various physiological and biochemical effects of elicitors, including SA and JA on plant systems have been documented. In this investigation, the major compounds in the essential oils from the leaves of cultivated lemon balm were geranial, neral, carvacrol, β-caryophyllene, citronellal, thymol, geranyl acetate, and caryophyllene oxide. Results of other investigations on GC/MS analysis of the essential oil from cultivated lemon balm in Iran also indicated that the main constituents in the essential oil are geranial, neral, citronellal, geranyl acetate, *β*-caryophyllene and caryophyllene oxide (Sadraei et al., 2003; Asgari and Sefidkon, 2004; Sharafzadeh et al., 2011; Taherpour et al., 2012). Mimica-Dukic et al. (2004) reported that the main constituents in *M. officinalis* essential oil were monoterpene aldehydes, ketones (neral/geranial, citronellal, isomenthone, and menthone), mono- and sesquiterpenes hydrocarbons (*β*-caryophyllene). A comparison of our results with the previous reports suggests few differences in the volatile composition of the plant could be attributed to the geographic origin of the plant sample, methods of extraction, and agronomic practices (Ghasemi Pirbalouti et al., 2017; Torki-Harchegani et al., 2018; Moghaddam et al., 2018).

Accumulation of metabolites often occurs in plants subjected to stresses including various elicitors or signal molecules. Indeed, SA and JA as natural compounds are important components in the signal transduction pathway metabolic. Jasmonic and salicylic acids compounds have long been observed to be transducers of elicitor signals for the production of plant secondary metabolites. In this study, the exogenous applications of JA and SA improved concentrations of oxygenated monoterpenes such as carvacrol and thymol (phenolic monoterpenes compounds), gerenyl acctate and also sesquiterpenes (β-caryophyllene and caryophyllene oxide) in the essential oils. Similarly, Ashrafi et al. (2012) reported the foliar application of JA increased thymol and carvacrol contents in essential oil of *T. daenensis* aerial parts. In addition, Talebi et al. (2018) reported that the foliar application of MeJA raised the percentages of linalool and 1,8-cineole, whereas reduced the percentages of α-cadinol, α-bergamotene, β-maaliene, and eugenol in the extracted essential oils from both basil cultivars. The JA signaling pathway is generally regarded as an integral signal for biosynthesis of many plant secondary products (Sanz et al., 2000; Zhao et al., 2005), anthocyanins (http://www.sciencedirect.com/science/article/pii/S0014579306004005,Zhao et al., 2005; Uppalapati et al., 2005), and defense-related volatiles (Thaler et al., 2002; Ament et al., 2004). Increasing evidence indicates that JA-induced changes in secondary metabolism constitute a ubiquitous plant defense response (Goossens et al., 2003; Zhao et al., 2005). JAs are produced and accumulated in plants, but exogenous application of JA and MJ can elicit secondary metabolite accumulation in defense response induction (Thiem and Krawczyk, 2010). In this investigation, the lowest percentages of citronellal, linalool, and geranyl acetate were observed from JA at 0.40 $mg\,L^{-1}$. Decreased amount of these constitutes in the essential oil of the lemon balm plants sprayed with JA might be attributed to stress condition such as abiotic or biotic stresses, which would activate the synthesis of secondary metabolites. In addition, the foliar application of SA and JA resulted not in disappearance or appearance of the compounds in the essential oil of *M. officinalis*.

Generally, elicitors such as SA and JA as the newest plant growth regulators play lots of roles in plants. These elicitors have been shown extended range of varying effects on all plants, ranging from inhibition to promotion of plant processes. In this investigation, the foliar application of SA and JA did not effect on amount of essential oil content and dry herbage weight. Whereas, SA and JA applications changed the percentage of some major and minor constituents of lemon balm and we can select better elicitor and concentration due to our purpose. In final, using the exogenous SA and JA on plants could raise natural components such as oxygenated monoterpenes and sesquiterpenes in the essential oils.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bcab.2019.101144.

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