

ARTICLE

Chemical analyses of two plant essential oils and their effects on functional response of *Habrobracon hebetor* Say to *Sitotroga cerealella* Olivier larvae

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ABSTRACT *Salvia officinalis* L. and *Glycyrrhiza glabra* L. are two valuable medicinal plants from Kermanshah province in Iran. In this study, chemical analyses of their essential oils were performed by gas chromatography-mass spectrometry and the effects investigated on functional response of *Habrobracon hebetor* Say on larval stage of *Sitotroga cerealella* Olivier. Accordingly, emerged females of *H. hebetor* were treated by LC₃₀ of the isolated essential oils for 24 h. Then, six wasps were accidentally selected and introduced to densities of host larvae for 24 h. The results showed that naphthalene, decahydro-4a-methyl and alpha-thujone were dominant compounds in both essential oils, respectively. Holling model (1959) by using regression analyses confirmed functional response type III in the control wasps and type II in both essential oils treatments. The highest and lowest attack rates were observed in the control wasps ($0.0443 \pm 0.00278 \text{ h}^{-1}$) and *S. officinalis* treatment ($0.0349 \pm 0.00257 \text{ h}^{-1}$), respectively. Moreover, the treated wasps by *G. glabra* essential oil showed shorter handling time than *S. officinalis* treatment ($0.4497 \pm 0.0373 \text{ h}$ versus $0.5196 \pm 0.0589 \text{ h}$). Accordingly, *G. glabra* due to lower negative effects on the functional response of *H. hebetor* was more compatible than *S. officinalis* for their combination in integrated pest management schedules.

Acta Biol Szeged 65(2):211-220 (2021)

KEY WORDSanalysis
ectoparasitoid
functional response
stored pest
volatile compound**ARTICLE INFORMATION**

Submitted

01 September 2021

Accepted

18 October 2021

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Introduction

Salvia officinalis L. (sage, also called as garden sage, common sage, or culinary sage) (Fig. 1A) is an important medicinal plant belonging to family Lamiaceae (Labiatae). This species is a perennial and shrubby plant with evergreen stems and large, elongated, and entire leaves. The height of the plant is about 50 to 80 cm and its young stems are dark green that covered with dense gray hairs (Asadi et al. 2018). This plant is native to the Mediterranean regions; but, grows in most parts of the world and is also cultivated as an ornamental plant. *S. officinalis* needs heat and dry conditions during growing season and it is cultivated in some provinces of Iran that have mentioned conditions. *Glycyrrhiza glabra* L. (Liquorice or Licorice) (Fig. 1B) is an important medicinal plant with many secondary metabolites which many features of them is remained unknown (Duan et al. 2016; Esmaeili et al. 2019). This species is a perennial plant that has adapted with mesophyte and xerophyte habitats during its evolutionary period. It is native for southern Europe,

northern Africa, and temperate regions of Asia; but is found in abundance in many parts of Iran, especially in western half (Ghahreman and Attar 1999). According to Bao and Larsen (2010), *G. glabra* is considered as the most important species in genus *Glycyrrhiza* and family Fabaceae (Leguminosae) which has been widely used as an important medicinal plant.

Habrobracon hebetor Say (Fig. 2) is a minute wasp from family Braconidae that is an ectoparasitoid agent of moth caterpillars. This wasp feed quickly, aided by their gut enzymes which quickly destroy blood proteins in hosts larvae; this increases value of the species as an effective biological control agent (Salvador and Consoli 2008). Mass rearing projects of *H. hebetor* are frequently performed on the larval stage of *Sitotroga cerealella* Olivier and the other laboratory hosts in different commercial insectariums from Iran (Abedi et al. 2012; Mahdavi and Saber 2013). Angoumois grain moth (*S. cerealella*) is an important species of family Gelechiidae, commonly referred as the rice grain moth. It is most commonly associated as a pest of field and stored cereal grains. They burrow within the kernel grains of crop plants and rendering

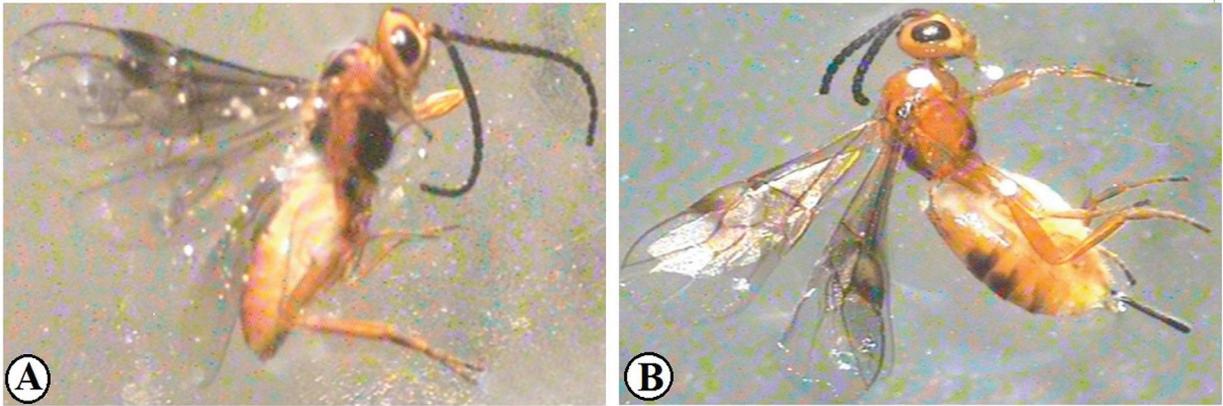


Figure 2. Adult male (A) and female (B) wasps of *H. hebetor*.

them unusable for human consumption. By laying eggs between the grains and hatching in later time during the processing, transportation, or storage stages; this moth can be transported to households or countries presently free of infestations. Thus, constant protection against this important pest is required for grain up until consumption time (Throne and Weaver 2013). To date, *H. hebetor* parasitoid wasp has been released for effective control of *Helicoverpa armigera* (Hübner), *Sesamia cretica* Lederer, and *Ostrinia nubilalis* (Hübner) in agricultural crops of Iran (Baker and Fabrick 2000).

Essential oils are volatile materials in different plant species that contain terpenes, terpenoids, aromatic, and non-aromatic compounds. Identifying these compounds and understanding their roles are very important issues in plant science (Isman 2000; Isman et al. 2008). Essential oils, also known as the plant secondary metabolites, are mainly abundant in Myrtaceae, Lauraceae, Lamiaceae, and Asteraceae families due to their contact, fumigation, repellent, and anti-feeding effects. These compounds are one of the main components in defense mechanisms of various plants against herbivores during many centuries ago (Bakkali et al. 2008; Rafiee-Dastjerdi et al. 2013; Asadi et al. 2018, 2019). In terms of isolation method, the aromatic products from distillation process that are separated by volatile mechanisms are called the essential oil (Asadi et al. 2019). Research on these compounds has shown that the essential oils are used in different aspects due to their fumigant and medicinal effects. These compounds are different in each plant species and also in each geographical areas; therefore, it is not possible to expect same compounds in one plant species from different regions; although, there may be similarities between their compounds.

Biological control or biocontrol is one of main methods in integrated pest management (IPM). Functional response

is the intake rate of a consumer as a function of food density or the amount of food available in a given ecotope (Baker and Fabrick 2000; Moezipour et al. 2008; Salvador and Consoli 2008). Following Holling (1959), functional responses are generally classified into three types, which are called Holling's type I, II, and III (Carneiro et al. 2010; Asadi et al. 2018). Totally, functional response type I has linear shape. In functional response type II, number of hosts attacked by each natural enemy reach to fix rate and most of the biocontrol agents show this type. Also, the functional response type III has sigmoid shape that is a preferred type or response about the natural enemies (Holling 1959; Hassell 1978).

Predators and parasitoids as important biocontrol agents commonly could be combined with compatible pesticides or botanical compounds (plant extracts and essential oils). To date, there are few information about the effects of these compounds on the functional response of *H. hebetor* to its hosts (Rafiee-Dastjerdi et al. 2009; Abedi et al. 2012; Mahdavi and Saber 2013; Mahdavi et al. 2013; Faal Mohammad-Ali et al. 2015; Jarrahi and Safavi 2015; Rashidi et al. 2018; Asadi et al. 2018, 2021). Due to great importance of functional response in population dynamics of plant pests by their natural enemies, the main idea for performing of this research was to determining safe botanical compounds on this important biocontrol agent for their efficient combination in IPM schedules.

Material and Methods

Identification of plant species

Identification of medicinal plants species collected from their natural habitats in Kermanshah province from Iran was done by sending complete samples to Razi University Herbarium (RUHK), Herbarium code: 2905 and 2906,

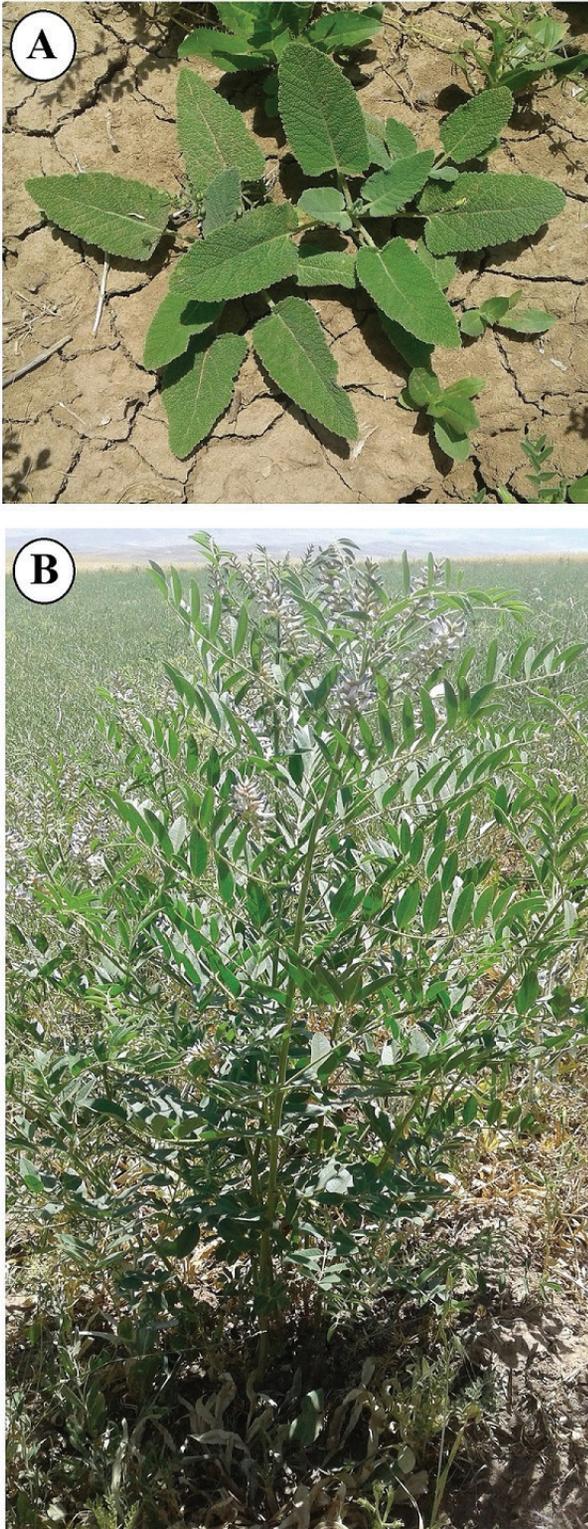


Figure 1. Mature plants of *S. officinalis* and *G. glabra* that collected from their natural habitats.

respectively; Kermanshah, Iran.

Isolation process of essential oil

After drying the plant specimens in shade (temperature about 25 °C), they were transferred to laboratory and their essential oils were gradually isolated. For this, aerial parts of the plants that have the highest content of volatile compounds were pulverized. Then, 50 g of the plant powder was mixed with 500 ml of distilled water in 1 liter balloon of Clevenger apparatus (Asadi et al. 2018). When the balloon heats up, volatile compounds are transferred to top of the tube and then cooled by condenser. After three hours, the essential oil was separated as a pale green layer on the water. The essential oil content of two medicinal plants was sufficient for these experiments and in each mentioned process approximately 0.5 ml was isolated. In order to remove water and purify the isolated essential oils, sodium sulfate (Na_2SO_4) was used (Asadi et al. 2019). Finally, the purified essential oils were stored in special glasses (5 ml) covered with aluminum tape in refrigerator (temperature about 4 °C) until GC-MS analyses and their usage in the experiments (Negahban et al. 2007; Shiva Parsia and Valizadegan 2015; Asadi et al. 2018).

Gas chromatography-mass spectrometry (GC-MS)

Chemical compounds in the purified essential oils of *S. officinalis* and *G. glabra* were identified by using a gas chromatographic device equipped with a split-splitless inlet (SSI) connected to mass spectrometer (GC-MS, Agilent 7980, USA). This device was available in the central laboratory, Islamic Azad University, Science and Research Branch, Tehran, Iran. The MS was equipped with EI ionization system and a four-coupled single analyzer (SQA) (Agilent, USA). To achieve the highest sensitivity in the detector, there was a triple detector (EMP) type that had very little noise and drift. After injecting of the essential oils by Hamilton syringe into the device, different compounds were gradually detected based on their molar mass at different times (run time). Based of the analyses by GC-MS, different compounds were explored according to their peak number, run time, and area.

Rearing of the host (*S. cerealella*)

Initial population of *S. cerealella* was gotten from a private insectarium (registered name: Jalilian) in Eslamabad-e Gharb city located in Kermanshah province (Iran), during 2019. The population was reared on wheat seed and maintained under the laboratory conditions including 25 ± 1 °C, $65 \pm 5\%$ relative humidity, and a photoperiod of 16:8 (L:D) h (Naseri et al. 2017).

Rearing of the parasitoid wasp (*H. hebetor*)

Females of *H. hebetor* were gotten from a private insecta-

Table 1. Fifteen dominant compounds in *S. officinalis* essential oil.

Compound number	Compound name	Run time (min)	Area%
1	Alpha-thujone	8.904	24.22
2	Bicyclo [2.2.1] heptan-2-one	9.865	15.51
3	1, 8-cineole 2-oxabicyclo	7.250	10.00
4	Thujone bicyclo [3.1.0] hexan-3	9.110	6.20
5	Veridiflorol	24.330	4.47
6	1-naphthalene propanol	31.786	4.10
7	1s-alpha-pinene	5.408	3.52
8	Camphene bicyclo [2.2.1] heptan	5.688	3.17
9	Alpha-humulene	20.416	2.65
10	Iron, monocarbonyl-(1, 3-butadien)	37.828	1.67
11	Borneol	10.369	1.65
12	Caryophyllene	19.341	1.60
13	12-oxabicyclo [9.1.0] dodeca-3	24.765	1.30
14	3, 6-dioxa-2, 4, 5, 7-tetrasilaoctan	38.034	1.13
15	1-naphthaleneethanol	37.565	1.09

rium (registered name: Jalilian) in Eslamabad-e Gharb city (Kermanshah province located in west of Iran), during 2019. The obtained wasps were urgently transferred to the growth chamber and reared on the last larval stage of *S. cerealella* as its laboratory host in 25 ± 1 °C, 65 ± 5 % relative humidity, and a photoperiod of 16:8 (L:D) h. Honey solution (produced in Mahram factory, Iran) was used as food for the adult parasitoid during the mentioned processes (Rafiee-Dastjerdi et al. 2009; Abedi et al. 2012).

Bioassays experiments

For study the fumigant toxicity of two isolated essential oils on the young female wasps of *H. hebetor*; different concentrations of them (8.5, 17.8, 38, 79.45, and 166.5 $\mu\text{L/L}$ air for *S. officinalis* and 8.9, 21.9, 54.95, 134.9, and 333.5 $\mu\text{L/L}$ air for *G. glabra*) that showed the mortality rate between 20% and 80% were put on filter papers (1 \times 1 cm) in 80 ml glass Petri dishes by using of sampler. The distilled water was used on the control wasps. Then, 20 female wasps of *H. hebetor* were introduced to each Petri dish and then the Petri dishes were immediately covered with parafilm tape to prevent of essential oil exit. For feeding of the parasitoids, the honey solution was used on a piece of white paper in each unit. Each concentration was assayed in four replications and after 24 h; numbers of dead wasps were carefully recorded (Shiva Parsia and Valizadegan 2015).

Functional response experiments

LC₃₀ of each essential oil (6.657 and 6.754 $\mu\text{L/L}$ air, respectively) was used as the lowest lethal concentration for the functional response experiments. For this, 100 mated females of *H. hebetor* were firstly treated by LC₃₀

of selected essential oils on filter papers (1 \times 1 cm) by using of sampler in 10 cm Petri dishes for 24 h. All stages were done for on control wasps with the distilled water. After 24 h, six treated wasps were randomly selected and separately transferred to the Petri dishes contained the different densities including 2, 4, 8, 16, 32, and 64 of *S. cerealella* larvae in a growth chamber that was set at 25 ± 1 °C, 65 ± 5 % relative humidity, and a photoperiod of 16:8 (L:D)h for 24 h. The honey solution was supplied as food source for the parasitoids. The experiments were designed in eight replication for the control wasps and each essential oil treatment and the numbers of paralyzed larvae were recorded after 24 h.

Functional response model

Model of Holling (1959) was used about the functional response of *H. hebetor* on variable densities of *S. cerealella* larvae which given as follows:

$$N_a = aT_t N_0 / (1 + aT_h N_0)$$

The components of above equation are:

- N_a = Number of attacked larvae
- N_0 = Different densities of *S. cerealella* larvae
- T_t = Total time of experiments (24h)
- a = Attack rate
- T_h = Handling time

Simpler form of the above equation is:

$$a = (d + bN_0) / (1 + CN_0)$$

Table 2. Twenty dominant compounds in *G. glabra* essential oil.

Compound number	Compound name	Run time (min)	Area%
1	Naphthalene, decahydro-4a-methyl	25.578	15.62
2	2, 6-octadiene-1-ol, 3, 7-dimethyl	24.702	6.96
3	Butanoic acid, 3, 7-dimethyl-2	23.587	5.79
4	Lavandulyl acetate	18.294	4.93
5	3-hexene-1-ol, benzoate	23.890	3.45
6	Nerolidol 1, 6, 10-dodecatrien	23.735	3.22
7	Geranyl tiglate	27.037	3.04
8	Geranyl propionate 2, 6-octadiene	21.137	2.59
9	Geranyl benzoate	31.151	2.55
10	(e)-2-formyl-6-methyl-3-(1-propylen)	24.805	2.24
11	Benzyl benzoate benzoic acid	28.473	2.14
12	1, 6, 10-dodecatrien-3-ol	25.091	2.12
13	Naphthalene, 1, 2, 3, 4, 4a, 5, 6, 8a	25.337	2.09
14	2-naphthalenemethanol	25.967	1.98
15	Propanoic acid, 2-methyl	22.242	1.82
16	Hinesol	25.664	1.57
17	(-)-endo-2, 6-dimethyl-6-(4-methyl)	19.890	1.46
18	Neryl propionate 2, 6-octadiene	28.193	1.34
19	Beta-eudesmol 2-naphthalene	25.898	1.28
20	1h-cycloprop [e] azulene, decahydro	20.926	1.02

In this status; “b”, “c” and “d” are model constants (Hassell 1978; Juliano 1993).

Statistical analysis

Logistic and non-linear regression models by using of SAS ver. 9.1 software were used to determine the functional response type and estimation of its parameters (attack rate and handling time), under essential oils treatments and in the control wasps, respectively (SAS Institute 2002).

Results

Chemical analysis of the essential oils

The GC-MS results showed that *S. officinalis* essential oils contained 93 chemical compounds with a total run time of 38.230 min. Among the identified compounds based on the exit analysis by detector device, fifteen compounds were occupied more than 1% of area and make up 82.28 % of total sum of areas in the essential oil (Table 1). Also, alpha-thujone (8.904 min - 24.22%) was

the highest compound in this essential oil. Moreover, *G. glabra* essential oil contain 239 different chemical compounds with total run time of 38.469 min. As shown in Table 2, among the identified compounds, 20 compounds accounted more than 1% of sum of areas and mentioned as dominant compounds. By comparing of area in each compound to sum of areas, it was found that naphthalene, decahydro-4a-methyl (25.578 min -15.62 %) had the highest area in this essential oil. These data were explored from the file produced by the chromatographic device.

Bioassay

The obtained LC₃₀, LC₅₀, and LC₉₀ values for *S. officinalis* and *G. glabra* essential oils against the females of *H. hebetor* are shown in Table 3. The adult bioassays indicated that acute toxicity of *S. officinalis* essential oil on *H. hebetor* was higher than *G. glabra* essential oil.

Functional response type

Models of logistic regressions by linear (P₁) and non-linear parameters indicated functional response type in

Table 3. Acute toxicity of two plant essential oils on the female wasps of *H. hebetor*.

Essential oil	n	Slope ± E	LC30 µL/L air (95% CL)	LC50 µL/L air (95% CL)	LC90 µL/L air (95% CL)	Chi-Square
<i>S. officinalis</i>	480	1.564 ± 0.177	6.657 (4.181 - 9.159)	14.405 (10.744 - 18.083)	95.025 (70.572 - 145.715)	10.07
<i>G. glabra</i>	480	1.123 ± 0.134	6.754 (3.564 - 10.370)	19.792 (13.475 - 26.667)	273.871 (176.772 - 527.283)	8.99

Table 4. Functional response parameters in *H. hebetor* wasps were exposed to LC₃₀ of two essential oils.

Treatments	Coefficient	Estimate	Stansard Error	Chi-Square	P-value
Control	P ₀ (Constant)	1.2962	0.5316	5.95	0.0148
	P ₁ (Linear)	0.0554	0.0813	0.46	0.4958
	P ₂ (Quadratic)	-0.00292	0.00306	0.91	0.3406
	P ₃ (Cubic)	0.00002	0.00003	0.81	0.3668
<i>S. officinalis</i>	P ₀ (Constant)	1.5690	0.4790	10.73	0.0011
	P ₁ (Linear)	-0.1267	0.0701	3.27	0.0707
	P ₂ (Quadratic)	0.00410	0.00260	2.48	0.1151
	P ₃ (Cubic)	-0.00004	0.00002	2.58	0.1082
<i>G. glabra</i>	P ₀ (Constant)	1.6326	0.5021	9.85	0.0017
	P ₁ (Linear)	-0.0248	0.0767	0.10	0.7469
	P ₂ (Quadratic)	-0.00078	0.00285	0.07	0.7846
	P ₃ (Cubic)	0.00001	0.00002	0.17	0.6828

the control and each essential oil treatment. If P₁ value be equal or more than 1, the response is type III and if be lower than 1 is type II (Table 4). According to the results, the functional response type III (P₁ ≥ 0) in the control and type II (P₁ < 0) in *S. officinalis* and *G. glabra* essential oils were determined, respectively (Fig. 3).

Functional response parameters

Estimation results of the attack rate, handling time, and theoretical maximum attack rate values for the treated wasps of *H. hebetor* by two isolated essential oils and the control are given in Table 5. Accordingly, the highest and lowest attack rates were obtained in the control wasps (0.0443 ± 0.00278 h⁻¹) and *S. officinalis* (0.0349 ± 0.00257 h⁻¹) treatment, respectively. The descending order of attack rate values was observed in the control, *G. glabra*, and *S. officinalis* treatments, respectively. In addition, the treated wasps by *G. glabra* essential oil showed shorter handling time than *S. officinalis* (0.4497 ± 0.0373 h versus 0.5196 ± 0.0589 h). Moreover, the respective ascending order of handling time values was observed in the control, *G. glabra*, and *S. officinalis* treatments. The highest and lowest values for theoretical maximum attack rate based on the obtained T/T_h (total time / handling time) were observed

in the control and *S. officinalis* essential oil (62.19 versus 46.19), respectively. In addition, the respective descending order for this parameter was determined in the control, *G. glabra*, and *S. officinalis* treatments.

Discussion

Medicinal plants are god-given natural resources in each region and study of their chemical properties is very important. Due to high importance of *S. officinalis* and *G. glabra*, various researches have been done on these valuable plants over the world. There are good studies on the extract of these plants; but, we will abandon them. In addition, there are some studies on their essential oils that we will review them and explain differences or similarities with our results. Totally, the effects of essential oils on the functional response of *H. hebetor* and the other natural enemies could be considered as a useful tool for predicting the success of this useful agents in IPM schedules, especially on stored pest, *S. cerealella*.

Studies about the acute toxicity of different essential oils on *H. hebetor* is very limit (Seyyedi et al.

Table 5. Logistic regression analysis from *S. cerealella* larvae that parasitized by *H. hebetor* wasp.

Treatment	Functional response type	Attack rate (h ⁻¹) a ± SE (Lower - Upper)	Handling time (h) T _h ± SE (Lower - Upper)	Theoretical maximum attack rate (T _{total} / T _{handling})	Correlation coefficient (R ²)
Control	III	0.0443 ± 0.00278 (0.0387 - 0.0449)	0.3859 ± 0.0317 (0.3221 - 0.4498)	62.19	0.96
<i>S. officinalis</i>	II	0.0349 ± 0.00257 (0.0343 - 0.0446)	0.5196 ± 0.0589 (0.4012 - 0.6381)	46.19	0.95
<i>G. glabra</i>	II	0.0364 ± 0.00340 (0.0296 - 0.0433)	0.4497 ± 0.0373 (0.3747 - 0.5247)	53.36	0.95

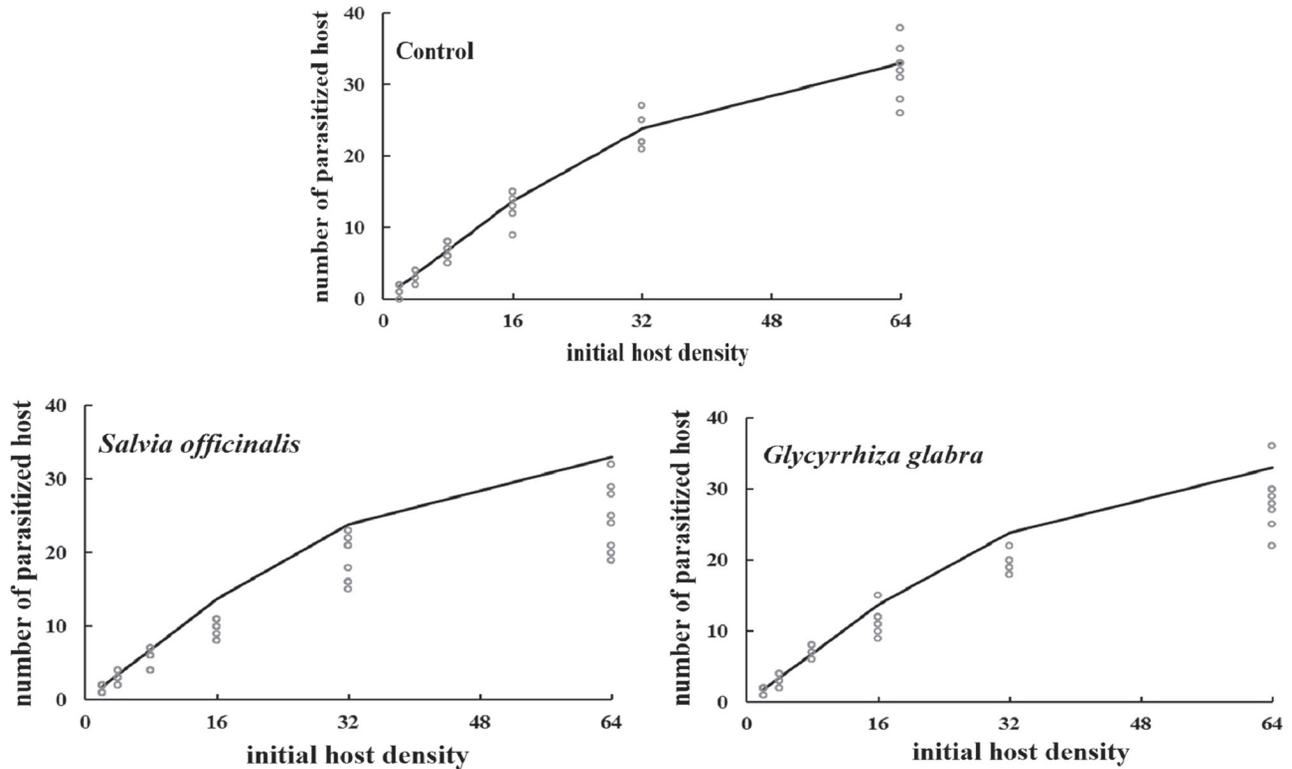


Figure 3. Curves of functional response from females of *H. hebetor* that treated by LC_{30} of selected essential oils and the control wasps to different densities of *S. cerealella* larvae.

2011; Hashemi et al. 2014; Ahmadpour 2017; Asadi et al. 2018, 2019). In our study, the tested essential oils showed different acute toxicity on the females of *H. hebetor* that extremely are similar to the results of Seyyedi et al. (2011), who studied the effects of *Ferula gummosa* L. essential oil on *H. hebetor* and concluded that the mortality was increased during 24h. Moreover, Hashemi et al. (2014) concluded that *Ferula assafoetida* L. essential oil had high toxicity on *H. hebetor*. Furthermore, Ahmadpour (2017) concluded that among *Ocimum basilicum* L., *Achillea millefolium* L., *Foeniculum vulgare* Mill., and *Zataria multiflora* Boiss, the essential oil of *F. vulgare* showed higher mortality compared with the others. Asadi et al. (2018, 2019) concluded that acute toxicity of *Rosmarinus officinalis* L. essential oil on the female wasps of *H. hebetor* was higher than the other essential oils including *Allium sativum* L., *Piper nigrum* L., *Salvia officinalis* L., and *Glycyrrhiza glabra* L. Also, *G. glabra* essential oil had the lowest acute toxicity in their research that is completely similar to our results.

In our research, alpha-thujone and naphthalene decahydro-4a-methyl were main components in *S. officinalis* and *G. glabra* essential oils, respectively.

These compounds are volatile and aromatic with high toxicity on the female wasps of *H. hebetor*. These compounds have active molecules that make fumigant and contact effects and could be considered as natural insecticides against the insect pests especially in enclosed environments (Yazdgerdian et al. 2015). Accordingly, these essential oils and the other botanical compounds also can have negative effects on the natural enemies especially on behavioural features such as functional response that must be seriously considered (Croft 1990; Mahdavi and Saber 2013; Jarrahi and Safavi 2015). The results also showed that the attack rate values in treated wasps of *H. hebetor* by LC_{30} of the essential oils was lower than the control; but, the handling time values were higher. This subject indicated that these essential oils negatively have affected these important parameters in *H. hebetor*; because, when handling time increase the attack rate naturally decrease and this is a negative effect of each compound on a biocontrol agent.

There are limited research about the effects of essential oils on the functional response of *H. hebetor*; but, research on the insecticides effects are to some extent available. Asadi et al. (2018) concluded

that among *A. sativum*, *R. officinalis*, *P. nigrum*, *S. officinalis*, essential oil of *G. glabra* showed minimum negative effects on the functional response type and its parameters in *H. hebetor*. Their results indicated that *G. glabra* essential oil can be recommended with *H. hebetor* in IPM that is similar with our results. Although, the studied host in their research was *Ephestia kuehniella* Zeller that is different with our host. Features of host directly could be effective on the natural enemies that this subject must be considered in related research on any natural enemy.

About the effects of pesticides on the functional response of this biocontrol agent, Rafiee-Dastjerdi et al. (2009) obtained the functional type II in *H. hebetor* on its laboratory host in the control and all insecticides treatments. Abedi et al. (2012) indicated that cypermethrin showed more adverse effects on this ectoparasitoid wasp compared with azadirachtin, methoxyfenozide, and pyridalil. In comparison, our results indicated that *S. officinalis* essential oil made higher negative effects on this biocontrol agent. Mahdavi and Saber (2013) concluded that malathion was compatible insecticide on the functional response of *H. hebetor* to *E. kuehniella* larvae compared with diazinon. But, our results showed that *G. glabra* were compatible. Mahdavi et al. (2013) studied the effects of abamectin, carbaryl, chlorpyrifos, and spinosad on the functional response of *H. hebetor* on adults treatment and reported functional response type III in all treatments; but, spinosad and abamectin were relatively compatible on the functional response parameters of this ectoparasitoid wasp. The dissimilarity of the results can belong to the type of host, laboratory conditions, quality of investigated wasps, type and mode of action of selected treatments, and their compatibility on *H. hebetor*.

Faal Mohammad-Ali et al. (2015) investigated the effects of chlorpyrifos and fenprothrin on the functional response of *H. hebetor* in larval and pupal treatments and stated that the functional response of this parasitoid wasp in the control and all treatment were type III and the treatments have not any significant effects on the attack rate compared to the control; but, their effects on the handling time were significant. Different stages of *H. hebetor* have special features and their treatments could be made variable results. Jarrahi and Safavi (2015) concluded that Proteus® in pupal stage treatment made the highest handling time and lowest attack rate in *H. hebetor* compared to entomopathogenic fungus *Metarhizium anisopliae* sensu-lato and the control that their results were in agreement with our results about *S. officinalis* treatment. The observed differences in results can belong to treated growth stage of *H. hebetor*, experimental con-

ditions, colony quality of the parasitoid wasp, and type of examined treatments. Moreover, Rashidi et al. (2018) studied the effects of diazinon, phosalone, fipronil, and pyriproxifen on the functional response of *H. hebetor* on two lepidopteran host and concluded that the type of functional response in the treatments and control were similar; but, the attack rate and handling time negatively were affected by LC₃₀ of mentioned insecticides that their results were in agreement with our results; although, in our research the type of functional response in all treatments were changed compared with the control. Asadi et al. (2021) studied the sublethal effects of chemical and botanical insecticides on the functional response of *H. hebetor* to larvae of *E. kuehniella* and stated that palizin and dayabon due to the lowest negative effects on *H. hebetor* were compatible insecticides for combination with this parasitoid wasp in IPM. In this study, we investigated the effects of two selected essential oils on the functional response in the ectoparasitoid wasp *H. hebetor* for second time. So far, the isolated essential oils from these medicinal plants have not been formulated as a commercial insecticide and we hope to find a way to formulate this natural compounds in future.

Acknowledgment

We wish to thank Mrs. Nashmieh Fathi the curator of Razi University Herbarium (RUHK) for her co-operation during this research.

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