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Fumigant Toxicity of New Formulations Prepared from *Artemisia sieberi* (Asteraceae) Essential Oil against *Sitophilus oryzae* (Col.: Curculionidae)

E. Shahinfar[1], A. Heidari[2*], S. Imani[1], A. Ahadiyat[1], and M. Negahban[2]

ABSTRACT

Nowadays, the increased pest resistance to chemical pesticides and the pesticide residues in food, together with the toxic effects of pesticides on mammals and the environment, necessitate the use of newer and safer alternatives for controlling insect pests. The use of encapsulation technology for pesticides formulations has led to increased efficiency and controlled release of these substances. In this study, four formulations containing *Artemisia sieberi* Besser essential oil based on biodegradable polymers such as polyethylene glycol, starch, glycerol mono stearate, and maltodextrin were prepared as powder and pellet to control the rice weevil, *Sitophilus oryzae*. Chemical composition of *A. sieberi* essential oil was determined and 16 components were identified. Cis-thujone, Santolinyl acetate, Trans-thujone, and Camphor were the major components of the oil. These formulations were different in the amount and type of polymers and surfactants. Also, these formulations were combined with a different rotation speed of the homogenizer. In this study, the fumigant toxicity of these formulations was compared with the *A. sieberi* oil. The experiments were carried out on the adult stage of the rice weevil at 27±1°C, 70±5% RH and in total darkness condition. The results showed that the LC_{50} value of the essential oil after 72 hours was 11.33 ppm, while the LC_{50} of formulations 1 to 4 were 47.61, 57.55, 23.94, and 44.66 ppm, respectively. Therefore, encapsulation of plants essential oils as a safe pesticide with slow release and fumigant toxicity can be used in integrated pest managements.

Keywords: Encapsulation, Lethality, Pesticides, Rice weevil.

INTRODUCTION

The rice weevil, *Sitophilus oryzae* Linnaeus (Coleoptera: Curculionidae), is one of the most important pests of stored products, distributed throughout the world by international commerce. This insect attacks many cereals, but rice is the main target. Depending on the product, the damage will be up to 50 % by weight (Hosseini *et al.*, 2008; Singh, 2017).

Increasing resistance of storage pests to conventional chemical pesticides such as methyl bromide and phosphine in the storage room and the problems of pesticide residues in food, water, and environmental pollution, plant contamination, and toxic effects on mammals and the environment have necessitated the need for newer and safer compositions for pesticides (Boyer *et al.*, 2012; Nobari *et al.*, 2014). The tendency to use natural compounds with environmental compatibility and desired pesticide characterization is expanding, so, nowadays, a large amount of research has focused on bio-rational pesticides (Isman *et al.*, 2011). Plants are a rich source of defensive chemicals (Wink *et al.*, 1998). These materials may exhibit insecticidal properties, repellency, attractiveness, anti-nutritional, and the effect of growth regulators on insects (Champagne *et al.*,

[1] Department of Plant Protection, Science and Research Branch, Islamic Azad University, Tehran, Islamic Republic of Iran.
[2] Department of Pesticide Research, Iranian Research Institute of Plant Protection, Agricultural Research, Education and Extension Organization (AREEO), Tehran, Islamic Republic of Iran.
* Corresponding author; e-mail: ah.heidari@areeo.ac.ir

1992). *Artemisia sieberi* Besser, belonging to the family Asteraceae, is indigenous to Iran. It is a green, grayish bushy plant, highly branched, dense, colony shaped, with thick woody vertical roots, branched at the bottom (Mir Heidar, 1994; Bagheri *et al*., 2007). There are 400 species of *Artemisia* in the world, with about 34 species growing in Iran (Rabie *et al*., 2006). This plant is the most valuable species in terms of livestock nutrition and resistant to desert and semi-desert areas in Iran. It is important in protecting the environment, especially for soil erosion (Kargar *et al*., 2010). The genus *Artemisia* has numerous medicinal qualities for various treatments, including anti-coughing properties, antispasmodic properties, anesthetic, anti-parasite, analgesic and headache (Mir Heidar, 1994; Shafizadeh, 2002), antioxidant (anti-cancer), antidiabetic, anti-fungal as well as insecticidal properties (Shahbazi and Arshadi, 2018).

Zandi-Sohani *et al.* (2018) studied the fumigant toxicity of *A. sieberi* essential oil on *Bemisia tabaci* Gennadius and its predator *Orius albidipennis* Reuter then measured the LC_{50} for both species as 0.059 and 0.621 µL L^{-1}, respectively. The result representing the difference in toxicity of essential oil for this insect pest and its predator is promising for the use of essential oil for pest control as fumigants without seriously affecting its predator. Naseri *et al*. (2017) showed that the LC_{50} of *Artemisia khorassanica* Podl essential oil had higher toxicity than that of *A. sieberi*. The results of this study suggest that tested essential oils have a good potential to apply in integrated pest management of *Sitotroga cerealella* Olivier. Nouri-Ganbalani and Borzoui (2017) showed that the lethal and sub-lethal effects of *A. sieberi* essential oil was evaluated against 4^{th} instar larvae of *Trogoderma granarium* Everts. Bioassays showed that the larvae were susceptible to both contact and fumigant toxicity.

The presence of volatile compounds with high evaporation rate and degradation are the constraints for using essential oils (Majeed *et al.,* 2015). However, terpenes mixture of *Melaleuca alternifolia* Cheel was quantified using GC-MS after processing through high shear and high-pressure homogenizer. They observed degradation of active compounds (Donsi *et al.,* 2011a). In another study, Gaysinsky *et al*. (2005) reported temperature stability of micellar encapsulated eugenol & carvacrol. They observed that with increased eugenol concentration in micelle, the temperature stability decreased i.e, at 0.1% eugenol the micelles were stable at 90˚C, but at 0.9% eugenol the micelles were stable at 60˚C (Gaysinsky *et al*. 2005). Therefore, one way to increase the stability and performance improvement of these plant compounds is to formulate essential oil (Majeed *et al.,* 2015). The choice of formulation depends on a variety of factors, including bioavailability, physical and chemical factors of the essential oil, the mode of action and the type of product (Negahban *et al.,* 2013). Plant essential oils require a transition system and their protection for greater efficacy by using the controlled release formulation (Donsi *et al.,* 2011b). Encapsulation technology reduces the amount of active ingredient loss and, by a coating of active ingredients, protects them from environmental factors such as temperature, light, and humidity. In their formulation, the release of the active ingredient is gradual. Their action seems attributable by progressive paralysis of *Limantria dispar* Linnaeus larvae, which become motionless and, consequently, stop feeding until death occurs (Moretti *et al.,* 2002).

Production of encapsulated powder formulation contains plant essential oils as the safe and healthy biorational pesticide with high performance. Due to its significant toxic effects on pests, new and safe technologies can be obtained that have the least harmful effect on human and environments instead of chemical pesticides. This is a completely natural formulation based on industrial materials at an affordable price.

In this research, we aimed to prepare a new herbal source and environmentally friendly pesticide as powders and pellet from *A. sieberi* oil to control the rice weevil.

MATERIALS AND METHODS

Insect Cultures

Adult rice weevils were collected from rice cultivar 'Shiroudi' storage depots and grown in pest control laboratory at the Iranian Research Institute of Plant Protection, Tehran, Iran, at 27±1°C, 70±5% RH in darkness condition in the germinator system. To obtain identical insects sample after four generations (growth period of insects, 28–30 days), the adult insects of 1 to 3 days (in both sexes) were used for initial tests and final experiments.

Preparation of Essential Oil

Artemisia sieberi essential oil used in this research was purchased from Barij Essence Pharmaceutical Co. (Kashan, Iran).

GC-MS Analysis

The essential oil of *A. sieberi* was analyzed by GC-MS (Agilent 7890A) coupled with a mass spectrometer (Agilent 5975C) with a strong library for identification of isolated substances, located at the Research Institute of Forests and Rangelands, Tehran, Iran. The GC-MS system equipped with a DB-5 fused silica column (30 m×0.25 mm id, film thickness 0.25 μm) with the oven temperature ehich programmed as follows: the initial temperature of 60°C was immediately increased to 220°C at a rate of 3°C min^{-1}, subsequently, the temperature was increased to 240°C at 20°C min^{-1} and held at this temperature for 3 minutes. The injector and transfer line temperature were 260 and 280°C, respectively; carrier gas was helium with a linear velocity of 30.6 cm s^{-1}; split ratio 1:100, ionization energy 70 eV, scan time 1 second, mass range 40–340 amu.

Formulations

Formulation 1.

To prepare 5% (w/w) formulation, 5 g of *A. sieberi* essential oil and 5 g of Castor oil (India), *Ricinus communis* Linnaeus, were mixed using magnet stirrer [Model: MR HEI-TEC ø145(EU)] at the rotation speed of 100-200 rpm with manual regulation (about 50 rmp per 3 minutes). Polyethylene glycol (85 g) were heated at 120°C in a separate container using magnet stirrer at the rotation speed 100-200 rpm. After melting, 5 g of Ethoxylated Nonylphenol surfactant were added. The polymeric temperature was slowly reduced to a point where the physical state of the solution was not changed (70°C), and then the mixture of essential oil and castor oil was added dropwise into the polymer container. In order to prevent evaporation of the essential oil at this stage, the cap of the polymer container was sealed by parafilm. The resulting mixture was poured into a plastic container and kept inside the freezer. After freezing (-20°C), the powder was prepared using a mill [Model: 8 BL28 (HGB600), USA].

Formulation 2.

For 10 % (w/w) formulation, 10 g of *A. sieberi* essential oil was mixed with 5 g of Glycerin, 5 g of Polyethylene glycol and 5 g of Tween 80 [Polyoxyethlene (20) sorbitan monooleate], and with the help of a mechanical mixer at the speed of 400–1,200 rpm (Model: Hei-TORQUE Value 400 Basis). Then, 75 g of starch were added. Finally, to prevent adhesion and absorption of moisture, 4 g of sodium sulfate (Na_2SO_4) were added to the powder.

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Formulation 3.

For 10% (w/w) formulation, 10 g of *A. sieberi* essential oil, 10 g of Polyethylene glycol and 1 g of coconut oil (coconut fatty acid diethanolamide) as a surfactant were added and mixed for 15 minutes with a mechanical mixer at the rate of 500–1,000 rpm. In another container, 55 g of starch, 10 g of Maltodextrin, 5 g of Na_2SO_4 and a specific amount of Xanthan gum were mixed. The essential oil solution was then added to the powder containing starch drop by drop. The final formulation was powdered.

Formulation 4.

For 10 % (w/w) formulation, a mixture of 10 g of essential oil with 3 g of tween 80 and 7 g of Polyethylene glycol were mixed up using a mechanical mixer at the speed of 400–1,200 rpm. In another container, 80 g of Glycerol Mono Stearate (GMS) were heated on a magnet-stirrer at the speed of 100–200 rpm to 250°C. After complete melting, the solution temperature was reduced to 60°C, and the essential oil solution was added slowly to the dish, containing melted GMS. Finally, the solution was poured into small molds and the final formulation was prepared as a pellet.

Bioassay Tests.

Experiments were carried out at the Bioassay Laboratory of the Pesticide Department at the Institute of Plant Protection Research. All experiments were conducted in a germinator at 28±1°C and 70±5% RH in continuous darkness.

Fumigant Toxicity.

For this purpose, preliminary tests were carried out to determine the necessary concentrations for mortality between 20–80% by *A. sieberi* essential oil on adult rice weevils. Then, logarithmic distance was used to obtain the main concentrations (Robertson *et al*., 1991). After determining the concentrations, according to Negahban and Moharramipour, (2007) filter (Whatman No. 1, cut into 2 cm diameter pieces) was placed inside the cap of 280 mL glass jar to deposit essential oil on them. Then, 10 adult insects with 1 to 3 days old were transferred to the jars and 1 g of rice as insect food was added. With the aid of the sampler, different concentrations of essential oil were deposited on the filter paper of 1.7, 9.3, 12.5, 16.4 and 21.4 µL L^{-1} air.

The most appropriate concentrations for each formulation were as follows:

Formulation 1 were 35.7, 42.9, 50, 57.1, 67.9 and 78.6 µL L^{-1} air of essential oil that were equal to 0.17, 0.20, 0.23, 0.27, 0.32 and 0.37 g of powder, respectively.

Formulation 2 were 46.4, 50, 57.1, 64.3, 71.4 and 78.6 µL L^{-1} air of essential oil that were equal to 0.11, 0.12, 0.13, 0.15, 0.17 and 0.18 g of powder, respectively.

Formulation 3 were 17.9, 18.4, 25, 28.6, and 32.1 µL L^{-1} air of essential oil that were equal to 0.04, 0.05, 0.06, 0.07 and 0.08 g of powder, respectively.

Formulation 4 the concentrations were considered as 35.7, 42.9, 53.6, 57.1, and 71.4 µL L^{-1} air of essential oil that were equal to 0.08, 0.10, 0.12, 0.13 and 0.17 g of pellet, respectively.

In terms of the active ingredient, the formulations were weighted and poured in tea bag papers, then placed inside the jar caps. In order to avoid the contact of the insect with the filter papers and tea bags, the crates of the jars were covered with a mesh cloth fabric. Then, the lid was closed tightly and covered with parafilm. This experiment was performed in four replicates (each with 10 adult weevils) with control. Distilled water was used as water control of ordinary essential oil and non-essential oil formulation was used as control of powders and pellet. When no leg or antennal movements were observed, insects were considered dead. The number of alive and dead insects of each jar was counted 72 hours after initial exposure to the essential oil and formulations.

Remained Weight Percent of Essential Oil and Formulation of Essential Oil.

For this purpose, according to the method of Passino *et al*. (2004), 2 g of powder formulations 2, 3 and pellet formulation and 4 g of powder formulation 1, containing 0.2 g of essential oil of *A. sieberi*, were weighed in petri dishes. The petri dishes were covered with lace and after 1, 3, 6, 9, 12, 15, 18, 21, 27 and 31 days, were weighed. The experiments were carried out in a temperature range of 27±1°C, the relative humidity of 65±5%, with three replications.

Durability of Fumigant Toxicity

According to method of Negahban *et al.* (2011), d**urability of fumigant toxicity** for each compound was performed at LC_{90} concentration. With the help of a micropipette, different concentrations of ordinary essential oil were poured on filter paper to a diameter of 2 cm, and after placing it inside the container with the insect food, the lid of the container was tightly closed. Also, the determined concentrations from the formulations were weighed and raped in a tea bag and then placed in cap of 280 mL jar with insect food inside. Three days after the treatment, 10 adult insects of rice weevil were released into the glass containers, and after 72 hours, the number of dead insects was counted. The same process was repeated for 6, 9, 12, 15, 18, 21, 24, 27, 30, 33 and 36 days every three days until no mortality was observed for the treatment. The lid of the container was tightly closed before the pest transfer, and after the samples were poured into the container, the lid of the container was closed again and remained blocked throughout this period. The experiments were performed in 3 replications with water controls.

Data Analysis

The Probit analysis was employed to estimate 50% Lethal Concentration (LC_{50}) and 50% Lethal Time (LT_{50}) for each formulation and normal essential oil (Finney, 1971), and comparisons of these values using Relative Median Potency (RMP), as showed in Equation (1) (Finney, 1964) were performed using SPSS 22 software, and the graphs were plotted using the Excel 2016.

$$RMP = \frac{LC_{50} \text{ of Essential oil}}{LC_{50} \text{ of Formulation}} \qquad (1)$$

RESULTS

Chemical Composition of *A. sieberi* Essential Oil

The GC-MS results of essential oil analysis for *A. sieberi* showed that the four major components of this essential oil were Cis-thujone (38.20%), Santolinyl acetate (23.77%), Trans-thujone (11.31%) and Camphor (6.30%) (Table 1). The GC/ MS profile of *A. sieberi* essential oil is presented in Figure 1.

Fumigant Toxicity

The results of the fumigant toxicity of *A. sieberi* essential oil and formulations containing this essential oil on the rice weevils is represented in Table 2. The LC_{50} and LC_{90} values after 72 hours on the rice weevil for pure essential oils were 11.33 and 25.80 μL L^{-1} air; for powder1: 47.16 and 82.52 μL L^{-1} air; for powder 2, 55.57 and 82.16 μL L^{-1} air; and for powder 3 23.94 and 33.78 μL L^{-1} air, respectively, and for the pellet formulation was equal to 66.64 and 73.25 μL L^{-1} air, respectively (Table 2). Additionally, no mortality was observed in all the controls during the tests.

The RMP between LC_{50} values obtained from ordinary essential oil and formulations prepared after 72 hours indicated a

Table 1. GC-MS analysis of *Artemisia sieberi* essential oil.

NO	Compound	Retention time (Min)	Percentage	Kovats Index (KI)
1	α-Pinene	1.94	0.70	94
2	Camphene	2.04	4.27	950
3	Yomogi alcohol	2.13	2.17	1003
4	p-Cymene	2.34	1.04	1026
5	1,8-Cineol	2.39	3.16	1037
6	Artemisia alcohol	2.52	1.55	1085
7	Cis-thujone	2.72	38.20	1105
8	Trans-thujone	2.77	11.31	1117
9	Comphor	2.81	6.30	1141
10	Borneol	2.89	2.62	1165
11	Santolinyl acetate	2.95	23.77	1171
12	Terpinen-4-ol	3.03	0.88	1180
13	p-Cymen-8-ol	3.05	0.57	1182
14	Verbenone	3.18	1.14	1202
15	Cis-chrysanthenyl acetate	3.27	1.14	1268
16	Bornyl acetate	3.4	0.69	1281
17	Other compounds	-	0.49	-
	Total		100	

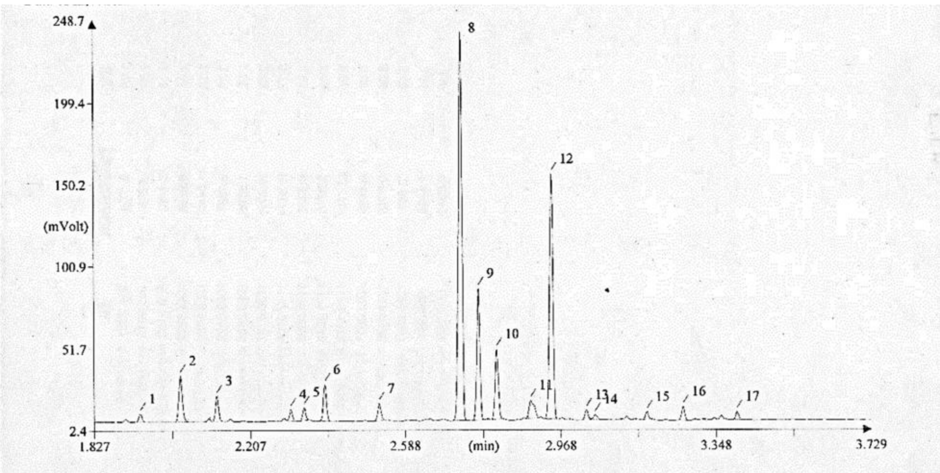

Figure 1. GC/MS profile of *Artemisia sieberi* essential oil

significant difference between them, so that the relative toxicity of the essential oil compared to powders 1, 2, 3 and pellet formation were 0.25, 0.20, 0.46 and 0.27, respectively.

The remaining weight of the essential oil and formulations after 1, 3, 6, 9, 12, 15, 18, 21, 27 and 31 days are shown in Table 3. The formulations had the highest weight percentage of residual essential oil in comparison with the normal essential oil. For formulations 1, 2, 3, and pellet formulation 4, the weight percentage of residual essential oil after 31 days were 96.77, 92.48, 90, and 93.10%, respectively. For the essential oil, the remaining weight percentage in 31 days was 29.30%. Therefore, it can be concluded that the formulation of essential oil can significantly prevent the rapid weight loss of the essential oil. The essential oils release from the

Table 2. LC_{50} and LC_{90} values of fumigant toxicity of essential oil and formulations of *Artemisia sieberi* essential oil on *Sitophilus oryzae.*

Compound	Time (h)	N	P-value	χ^2 (df)	Intercept ±SE	Slope ±SE	LC_{50} (μL L^{-1} air) Confidence Limit 95%	LC_{90} (μL L^{-1} air) Confidence Limit 95%
Essential Oil	72	200	0.76	1.18 (3)	(-3.79±0.64)	3.59±0.59	11.33 9.90 -12.78	25.80 20.85- 36.88
Powder 1	72	240	0.99	0.24 (4)	(-9.20±1.39)	5.49±0.81	47.61 43.59 - 51.18	81.52 72.32 - 99.71
Powder 2	72	240	0.95	0.70 (4)	(-14.59±2.03)	8.29±1.14	57.55 54.56 - 60.38	82.16 75.78 - 93.71
Powder3	72	200	0.93	0.43 (3)	(-11.82±1.65)	8.57±1.19	23.94 22.64 - 25.23	33.78 31.09 - 38.65
Pellet	72	200	0.90	0.59 (3)	(-9.84±1.67)	5.97±0.98	44.66 40.56 - 48.06	73.25 65.21 - 90.22

capsules was slower and remained for a longer time (Table 3).

Durability of Fumigant Toxicity

The results show that pure essential oil in the concentration of 25.12 ppm (LC_{90}) did not have a significant durability, such that 3 days after insecticide application, the mortality of insects was only 30%, and this trend continued until the 9th day when it decreased and reached 10%. As a result, the durability of ordinary essential oil was not significant.

In the powder formulation 1, the mortality rate was 83.33% 3 days after the treatment. After 15 days, the mortality rate reached 16.67%. However, in the powder formulation 2, after 3 days, the mortality rate was 100%, and this rate was maintained until the 6th day, and then gradually decreased and reached 13% on the 36th day. Three days after essential oil was added, the lethality rate of powder formulation 3 was 100% and gradually decreased to null on day 33rd. In pellet formulation, after 3 days, the mortality rate was 90% and 27 days after the treatment, the mortality rate reached 13.33% (Table 4).

Finally, the LT_{50} values for powder formulations 1, 2, 3 and pellet formulation were calculated to be 6.95, 21.36, 16.1 and 14.4 days, respectively (Table 5).

DISCUSSION

Insecticidal compounds of many essential oils are monoterpenoids. Camphor, camphene, 1.8-cineol, α-pinene α, linalool, methyl acetate, limonene, menthone, graniol, citral, citronellal, thymol, carvacrol, eugenol, and trans-anethol are examples of pesticide compounds of plant essential oils (Isman and Machial, 2006; Phillips *et al*., 2010). In addition, monoterpenoids are highly regarded as control agents and are actually toxic to insects (Moharramipour and Negahban, 2014). In this study, *Artemisia* essential oil had different monoterpenoids with high evaporation rate. Due to their high volatility, they had fumigant activity that might be of importance for controlling stored-product insects. Bayramzadeh *et al*. (2019) showed that two essential oils including *Cuminum cyminum* Linnaeus and *Lavandula angustifolia* Miller, were nanocapsulated by solvent evaporation emulsion method and their fumigant toxicity was investigated against *Tribolium castaneum* Herbst and *Sitophilus granarius* Linnaeus. Their results suggested that the content of essential oils could influence encapsulation efficiencies.

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Table 3. The remaining weight percent (Mean±SE) of *Artemisia sieberi* essential oil before and after formulation at 27±1°C and 65±5% relative humidity.

Compound	Time (Day)										
	0	1	3	6	9	12	15	18	21	27	31
Essential oil	100 ±0.0	44.00 ±2.08	37.50 ±1.15	35.50 ±0.82	33.33 ±0.93	32.50 ±0.76	31.80 ±0.67	30.50 ±0.58	30.30 ±0.44	29.67 ±0.60	29.30 ±0.44
Powder 1	100 ±0.0	98.50 ±0.04	97.58 ±0.03	97.40 ±0.01	97.12 ±0.02	97.01 ±0.01	96.86 ±0.02	96.84 ±0.01	96.80 ±0.01	96.79 ±0.0	96.77 ±0.01
Powder 2	100 ±0.0	94.92 ±0.06	94.88 ±0.03	93.63 ±0.02	93.50 ±0.05	93.18 ±0.04	92.72 ±0.07	92.60 ±0.06	92.55 ±0.05	92.50 ±0.04	92.48 ±0.04
Powder 3	100 ±0.0	94.05 ±0.08	92.62 ±0.46	92.20 ±0.05	91.72 ±0.16	91.45 ±0.18	91.10 ±0.10	90.80 ±0.06	90.55 ±0.17	90.10 ±0.14	90.00 ±0.11
Pellet	100 ±0.0	99.82 ±0.12	99.53 ±0.23	99.08 ±0.41	98.32 ±0.65	97.78 ±0.83	97.20 ±0.96	96.57 ±1.14	95.55 ±1.30	94.22 ±1.23	93.10 ±1.26

Table 4. Morality percentage of *Sitophilus oryzae* at LC_{90} for 36 days.

Compounds	Concentration (ppm) (LC_{90})	Time mortality±SE												
		1	3	6	9	12	15	18	21	24	27	30	33	36
Essential oil	25.12	100.0 ±0.0	33.33 ±3.33	20.0 ±5.77	10.0 ±5.77	0.0 ±0.0	-	-	-	-	-	-	-	-
Powder 1	81.52	87.50 ±4.79	83.33 ±3.33	56.67 ±3.33	40.0 ±5.77	26.67 ±3.33	16.67 ±3.33	0.0 ±0.0	-	-	-	-	-	-
Powder 2	82.16	100.0 ±0.0	100.0 ±0.0	100.0 ±0.0	90.0 ±0.0	76.67 ±3.33	66.67 ±3.33	60.0 ±5.77	53.33 ±3.33	46.67 ±3.33	40.0 ±5.77	33.33 ±3.33	26.67 ±3.33	13.33 ±3.33
Powder 3	33.78	100.0 ±0.0	100 ±0.0	93.33 ±3.33	83.33 ±3.33	73.33 ±3.33	60.0 ±5.77	43.33 ±3.33	33.33 ±3.33	23.33 ±3.33	16.67 ±3.33	10.0 ±0.0	0.0 ±0.0	-
Pellet	73.25	97.50 ±2.50	90.0 ±5.77	83.33 ±3.33	70.0 ±0.0	56.67 ±3.33	46.67 ±3.33	40.0 ±0.0	33.33 ±3.33	16.67 ±3.33	13.33 ±3.33	0.0 ±0.0	-	-

Table 5. LT_{50} values for formulations of *Artemisia sieberi* against *Sitophilus oryzae*.

Formulations	Concentration (ppm) (LC_{90})	N	P-Value	χ^2 (df)	Intercept±SE	Slope±SE	LT_{50} Confidence Limit 95%
Powder 1	81.52	150	0.99	0.11(3)	2.28±0.45	(-2.71±0.48)	6.95 5.57-8.37
Powder 2	82.16	360	0.97	3.27(10)	4.76±0.54	(-3.58±0.41)	21.36 19.36-23.63
Powder 3	33.78	300	0.99	1.53(8)	4.94±0.57	(-4.10±0.46)	16.1 14.6-17.75
Pellet	73.25	210	0.97	0.97(5)	2.52±0.42	(-2.18±0.39)	14.4 11.87-18.62

Different studies have shown that the fumigant toxicity of some essential oils is almost higher than their contact toxicity (Kim *et al*., 2003; Abdelgaleil *et al*., 2009). According to the results of fumigant toxicity of essential oils and formulations containing *A. sieberi* essential oil, this study showed that the essential oil and formulations had fumigant toxicity on *S. oryzae*. It was also found that the essential oil was trapped after being formulated in the cover of polymers and, therefore, had no insecticidal properties during the first 24 hours, but after 72 hours with gradual and controlled release, led to significant mortality and retained its insecticide property. This issue is in accordance with the findings of Khademi (2012). The nanocapsulated essential oil of

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Cuminum cyminum and *Carum copitum* C.B. Clark did not show fumigant toxicity on and *S. oryzae* in the first 24 hours of release, but over time, the toxicities of these compounds increased (Khademi, 2012).

This study showed that the LC_{50} value of normal essential oil of *A. sieberi* on the adults of *S. oryzae* was 11.33 ppm after 72 hours, while after formulation, this index increased as a result of trapping essential oil in polymer cover, which led to slow and gradual release by maintaining its insecticidal properties.

Negahban and Moharramipour, (2007) reported that the LC_{50} rate for the fumigant toxicity of *A. sieberi* essential oil on the flour beetle, *T. castaneum*, was 16.76 ppm, while in the later studies, the LC_{50} rate obtained for fumigant toxicity of this essential oil after nanoencapsulation was 41.32 ppm. Also, higher fumigant toxicity (LC_{50}) was reported for *A. sieberi* essential oil (3.78 ppm) over 24 hours, compared to the nanocapsuleted essential oil (11 ppm) against third-instar larvae of the diamondback moth, *Plutella xylostella* Linnaeus (Negahban, 2012).

Researches performed by Passino *et al.* (2004) on *Plodia interpunctella* Hübner with the micro-capsule formulation of *Rosmarinus officinalis* Linnaeus and *Thymus vulgaris* Linnaeus essential oil showed that by microcapsulation of essential oils, mortality of the pest increased significantly, and after 25 days, the microcapsule contained 75% essential oil. Ziaee *et al.* (2014) reported greater toxicity of a nanogel formulation of the *Carum copitum* essential oil compared to the unformulated essential oil.

Louni *et al*. (2018) showed that nanoemulsion, containing the essential oil of a mint species, *Mentha longifolia* Linnaeus, with its gradual release, kept fumigant toxicity on larvae and eggs of the Mediterranean flour moth, *Ephestia kuehniella* Zeller. The estimated LC_{50} values on the larval stage using normal essential oil and nanoemulsion essential oil were very close to each other, indicating that the essential oil after the formulation was able to maintain its insecticidal properties (Louni *et al*., 2018).

The results suggest that the process of encapsulating plant essential oils releases the active ingredient in a controlled manner and maintains its toxicity for a longer period of time. Several researchers have reported the durability of formulations prepared with plant essential oils against storage pests. According to the present findings, Negahban (2012) determined a longer fumigant and contact durability of the nanocapsulated essential oil of *A. sieberi* in comparison with ordinary essential oil. The researcher stated that the formulation of nano-capsules was able to increase the fumigant durability of essential oil against *Plutella xylostella* on larvae and adult insect of *Tribolium castaneum* by 7.3 and 6.73 times, respectively. Also, encapsulation increased the persistence of contact toxicity of essential oil on the *P. xylostella* (13.92 times) and the adult insect of *T. castaneum* (9.38 times). In a similar study, nanoemulsion formulation of *Zataria multiflora* Boiss essential oil increased the durability of contact toxicity against *Ephestia kuehniella* Zelle on larvae (Emamjomeh *et al*., 2017). Choupanian *et al*. (2017) also showed that nanoemulsion prepared from neem essential oil at 25 °C had a high physical stability and remained unchanged for 90 days. *Rosmarinus officinalis* nano-encapsulated essential oil also caused gradual release and maintenance of the fumigant toxicity of the essential oil against *Tribolium castaneum* in the long-run (Khoobdel *et al*. 2017). The high durability of *Carum copitum* essential oil on *Sitophilus granarius* L. and *Tribolium confusum* has also been confirmed (Ziaee *et al.*, 2014). Also, Passino and Moretti (2004) showed that the highest (75%) release of *R. officinalis* microscopic essential oil occurred within 25 days.

CONCLUSIONS

At the present, the introduction of low risk, effective, and low potentials resistance insecticide to control the pests is felt more than ever. Essential oils and plant extracts of natural origin can be effective in controlling pests in the warehouses by solving commercialization problems and their formulation issues. Also, considering the recent trend of society to use safe food with minimal synthetic pesticide residues reveals the need for bio-based pesticides production, while also being effective and low-risk for the environment. The use of encapsulating technology by creating polymer coating around the particles of essential oil is a novel and suitable method for preserving the essential oils from fast oxidation, which maintains its insecticide effect and longer durability.

The type of polymer selected can vary in the amount of essential oil released from the capsules. Therefore, with the proper choice of formulation in the pest management programs, rapid release formulation can be used to have impacts on severe contamination of stored products. The formulation with gradual release can also be used to prevent pest attack.

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سمیت تنفسی فرمولاسیون جدید حاوی اسانس درمنه *Artemisia sieberi* (Asteraceae) علیه شپش برنج Sitophilus oryzae (Col.: Curculionidae)

ا. شاهین فر، ا. حیدری، س. ایمانی، ع. احدیت، و م. نگهبان

چکیده

امروزه افزایش مقاومت آفات انباری به سموم شیمیایی و همچنین مشکلات باقی مانده سموم در مواد غذایی، اثرات سمی آن ها روی پستانداران و محیط زیست، نیاز به استفاده از ترکیب های جدیدتر و ایمن تر برای محیط زیست و انسان را ضروری می نماید. استفاده از تکنولوژی کپسوله کردن اسانس ها منجر به افزایش کارآیی و رهایش تدریجی آنها می شود. در این مطالعه، چهار فرمولاسیون حاوی اسانس درمنه *Artemisia sieberi* Besser بر اساس پلیمرهای زیست تخریب پذیر مانند پلی اتیلن گلیکول، نشاسته، گلیسرول مونو استئارات و مالتودکسترین به صورت پودر و پلت برای کنترل شپشه برنج *Sitophilus oryzae* تهیه شد. ترکیب شیمیایی اسانس *A. sieberi* اندازه گیری و ۱۶ ترکیب شناسایی شد. ترکیبات عمده شامل Cis-thujone، Santolinyl acetate، Trans-thujone و Camphor بود. فرمولاسیون های تهیه شده از نظر میزان و نوع پلیمر، سورفاکتانت و همچنین سرعت چرخش هموژنایزر متفاوت بودند. در این مطالعه، سمیت تنفسی فرمولاسیون های تهیه شده با اسانس خالص درمنه مورد مقایسه قرار گرفت. آزمایش ها بر روی حشرات کامل شپشه برنج در دما ۱±۲۷درجه سلسیوس، رطوبت نسبی ۵ ± ۷۰ ٪ و در شرایط تاریکی انجام شد. نتایج نشان داد، مقدار LC_{50} تنفسی اسانس خالص درمنه پس از ۷۲ ساعت، ۱۱/۳۳ پی پی ام بود، در حالی که LC_{50} فرمولاسیون ۱ تا ۴ به ترتیب ۴۷/۶۱، ۵۷/۵۵، ۲۳/۹۴ و ۴۴/۶۶ پی پی ام بود. براساس نتایج به دست آمده کپسوله کردن اسانس های گیاهی به عنوان آفتکش ایمن با رهایش تدریجی و سمیت تنفسی می تواند در مدیریت تلفیقی آفات در نظر گرفته شود.