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THE INSECTICIDAL ACTIVITY OF ESSENTIAL OILS ON EGGPLANT CULTIVARS INFESTED WITH THE COTTON APHID (APHIS GOSSYPII GLOVER (HEMIPTERA: APHIDIDAE)) UNDER GREENHOUSE CONDITIONS

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Abstract

The cotton aphid, *Aphis gossypii* Glover (Hemiptera: Aphididae), is a major pest that causes considerable economic damage worldwide, infesting many vegetable and fruit crops. Due to its developed resistance to chemical pesticides and harm to humans and the environment, it has become necessary to move towards finding alternatives. This study evaluated new nanoemulsion formulations of three essential oils, i.e., thyme (*Thymus vulgaris*), clove (*Eugenia caryophyllus*), and lavender (Lavandula angustifolia), against three local eggplant cultivars (Naser, Jawaher, and Barcelona) infested with the aphid. A nanoemulsion pesticide formulation was prepared at a concentration of 20% of the above oils. Then, three concentrations of 1, 5, and 10 ml/L of each essential oil were tested separately against *A. gossypii* and compared to the Sivanto and Levo-registered pesticides under greenhouse conditions. After 14 days of treatment, the 10 ml/L thyme oil concentration produced significant differences in reducing the population density levels of this pest, followed by cloves and then lavender. This study recommends using thyme, clove, and lavender oils as one of the safe alternatives for pest management and to achieve one of the goals of sustainable development.

Keywords: Thyme, Cloves, lavender, Essential oils, Aphids, Insecticides.

فعالية الزيوت العطرية كمبيدات حشرية على أصناف الباذنجان المصابة بحشرة من القطن APHIS GOSSYPII GLOVER (HEMIPTERA: Aphididae) تحت ظروف البيت البلاستيكي

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الخلاصة

تعتبر حشرة من القطن، *Aphis gossypii* Glover (Hemiptera: Aphididae)، واحدة من أهم الآفات التي تسبب أضرارا اقتصادية كبيرة في جميع أنحاء العالم. تصيب هذه الآفة العديد من محاصيل الخضر وشجار الفاكهة. نظرا لتطور مقاومة حشرة من القطن للمبيدات الكيماوية وبسبب أضرار المبيدات المعروفة على الإنسان والبيئة، أصبح من الضروري التوجه نحو إيجاد البدائل. في هذه الدراسة، اختيرت مستحضرات نانوية مستحلبة من ثلاثة زيوت عطرية، وهي زيت الزعتر *Thymus vulgaris*، وزيت القرنفل *Eugenia caryophyllus*، وزيت اللافندر *Lavandula angustifolia*، ضد ثلاثة أصناف محلية من الباذنجان (نصر، جواهر، وبرشلونة) مصابة بحشرة من القطن *A. gossypii*. حضر مستحضر نانوي مستحلب للمبيد بتركيز 20% من الزيوت المذكورة أعلاه. بعد ذلك، اختبرت ثلاثة تراكيز من مستحضر الزيتي المستحلب 1 و5 و10 مل/لتر من كل زيت عطري بشكل منفصل ومقارنتها بالمبيدات الحشرية المسجلة Sivanto و Levo ضد حشرة *A. gossypii* تحت ظروف البيت البلاستيكي. أشارت النتائج وبعد 14 يوما من المعاملة إلى أن زيت الزعتر المستخدم بتركيز 10 مل/لتر قد حقق فروقاً معنوية في خفض مستويات الكثافة السكانية لحشرة من القطن، يليه القرنفل ثم الخزامى. تقترح هذه الدراسة استخدام زيوت الزعتر والقرنفل واللافندر كمبيدات نباتية الاصل بديلة آمنة للمبيدات الكيميائية لتحقيق أحد أهداف التنمية المستدامة.

كلمات مفتاحية: زعتر، قرنفل، لافندر، زيت عطري، حشرة المن، مبيد حشري.

Introduction

The eggplant (*Solanum melongena*) is a fruit of much economic importance worldwide because of its large number of diverse cultivars in terms of color, shape, and size, and growth in different seasons of the year (6). The plants are infested in open and greenhouse conditions by many arthropods, mainly the cotton aphid *A. gossypii*, a multi-host pest around the world (7). Its adult and nymphal stages feed on the underside of leaves, sucking nutrients from the plant (6, 12 and 20) that can lead to poor quality plants and low productivity (24).

The widespread use of synthetic pesticides has produced significant and harmful effects not only on pollinators and beneficial insects but also on human health and the environment (26, 30 and 39). In addition, it has led to the emergence of insect resistance to many pesticide groups, with more than 600 species of arthropods that are resistant to one or more pesticides (22 and 34). The most important of these insects are *A. gossypii* and *Myzus persicae*, which have the ability to develop such resistance quickly (22 and 32). Hence, moving towards effective alternatives compatible with integrated pest management programs is crucial. In this context, botanical pesticides based on essential oils have become an important alternative to chemical pesticides (8, 17 and 31). Essential oils are easy to extract and have low toxicity to natural enemies (4 and 31). They have become common in organic farms as green pesticides (11 and 16).

Several studies have shown the effectiveness of essential oils against pests, such as contact pesticides, fumigation, deterrents, or attractants (15), as well as the effectiveness and potential use of essential oil products to control insect pests in both open and protected agriculture (16 and 37). More than fifty essential oils have significant insecticidal activity against *Trialeurodes vaporariorum* (Westwood), which is one of the most important pests that attack various vegetable crops grown in greenhouses in Korea (9).

Other studies indicated that over twenty essential oils have toxic effects on the third larva instar of *Spodoptera littoralis* (Boisduval) (28). Furthermore, other essential oils have shown insecticidal activity and inhibit the egg-laying ability of the cowpea weevil, *Callosobruchus maculatus* (Fabricius) (29). In addition, plant-based essential oils and their constituents are known to have both repellent and toxic effects on the fruit fly *Ceratitis capitata* (Wiedemann) and the Asian citrus psyllid, *Diaphorina citri* Kuwayama (21 and 40).

Aphids are among the main economic pests that attack a wide range of crops (33). A recent study found seven plant essential oils, such as *Ocimum sanctum* L., *O. basilicum* L., *O. gratissimum* L., *Mentha piperita* L., *M. arvensis* L., *Tagetes erecta* L., and *Lavandula angustifolia* Mill., as eco-friendly pesticides to control *A. gossypii* (38). Also, essential oils obtained from *Tagetes minuta* L. that were tested against the aphids *Acyrthosiphon pisum* (Harris), *M. persicae*, and *Aulacorthum solani* (Kaltenbach) showed a significant reduction in their reproduction (37). In other studies, rapeseed oil also significantly reduced damage caused by the black cherry aphid *M. cerasi* (Fabricius). The black bean aphids reduced the production of beans and caused economic loss on the infested crops (3 and 18). In addition, thyme oils significantly decreased the fecundity and survival of the cabbage aphid *Brevicoryne brassicae* (13).

By employing nanotechnology to develop novel nanoformulations based on EO in aqueous solutions, bioinsecticides can be made more efficient and environmentally friendly (19).

This study evaluated the insecticidal activity of emulsifiable concentrate formulations (EC) prepared from thyme oil (*T. vulgaris*), clove oil (*E. caryophyllus*), and lavender oil (*L. angustifolia*) on three *A. gossypii*-infested local eggplant cultivars (Naser, Jawaher, and Barcelona) under greenhouse conditions.

Materials and Methods

Thyme (*T. vulgaris*), clove (*E. caryophyllus*), and lavender (*L. angustifolia*) oils were purchased from the Green Sahel Factory Company for Oil Production, 113 Princess Raya, 11910 Amman, Jordan) and Tween 80 (polyoxyethylene sorbitan monooleate), used as surfactant or emulsifier, was purchased from Alpha Chemika company, India.

Nano emulation formulation: The formulation (EC) was prepared following (19). Briefly, the three essential oils (thyme, clove, and lavender) were mixed with Tween 80 and distilled water in a 1: 2: 2 ratio to obtain a 20 % concentration for each oil. Then, the emulsion was stirred by a magnetic stirrer at 600 rpm for 90 min and moved into a Falcon plastic container (12 x 75 mm). They were then sonicated ultrasonically at 65 W nominal power at 20 kHz frequency to obtain the nano-emulsion. The stability of the emulsion over time was also observed. The samples were stored at 25° C for use in the experiments.

Preparing the greenhouse: A 180-square-meter greenhouse (9 meters wide and 20 meters long) at the Department of Plant Protection, College of Agricultural Engineering Sciences, University of Baghdad, Baghdad, Al-jadryia, was prepared for the 2023-2024 agricultural season. Agricultural operations were conducted by sterilizing the soil with fungicides and organic and chemical fertilizers.

The soil was flooded with water after the pesticides (Nematin 10 G and Extra 25 WDG) and fertilizers were added. The entire area of the greenhouse was covered with agricultural nylon for 45 days. Then, the soil was plowed, and fertilizers were applied according to the recommended doses. Before planting the three eggplant cultivar seedlings (Naser, Jawaher, and Barcelona), 10 agricultural lines were prepared and distributed over 5 rows. The rows were covered with black nylon netting to reduce the growth of harmful weeds and preserve soil moisture. The dimensions of each row were 0.8 × 13 meters, and the rows were laid a meter apart. The eggplant seedlings were planted in the first week of October 2023. They were spaced 40 cm apart, and a drip irrigation system was used to water the plants.

After calculating the numerous densities of the aphid (*A. gossypii*), three concentrations of each of the essential oils at 1 mL/L, 5 mL/L, and 10 mL/L were prepared separately and sprayed at a concentration of 20% that was previously formulated. The pesticides Levo 2.4 SL (Oxymatine) and Sivanto SC 200 g/L (Flupyradifurone) were used as a positive control. Water with Tween 80 was also used as a negative control. The seedlings were sprayed with four replicates in the early morning. The experiment was carried out in a randomized complete block design (RCBD). After applying the treatments, the number of dead adults was counted after

1, 2, 3, 7, and 14 days of spraying the twelfth treatment (Table 1), with four replicates for each. The efficiency of treatments used in the experiment was calculated according to the equation of Henderson and Tilton (14).

Table 1: Treatments on the three eggplant cultivars infested by *A. gossypii*.

No.	Treatments	Rate of use
1	Thyme oil 20%	1 mL/L
2	Thyme oil 20%	5 mL/L
3	Thyme oil 20%	10 mL/L
4	Clove oil 20%	1 mL/L
5	Clove oil 20%	5 mL/L
6	Clove oil 20%	10 mL/L
7	Lavender 20%	1 mL/L
8	Lavender 20%	5 mL/L
9	Lavender 20%	10 mL/L
10	Sivanto SC 200 g/l	0.6 mL/L
11	Levo 2.4 SL.	0.25 mL/L
12	Control	10 mL/L

Assessment of the population density of A. gossypii: The first date of appearance of the aphid was determined by periodically examining the eggplant cultivars used in the study. Random samples of the eggplant leaves were taken weekly in 3 replicates for each cultivar and from three levels of the plant (upper, middle, and lower).

Each sample was placed in transparent, sealed polyethylene bags, and the date, name, and replicate numbers were recorded. The samples were taken to the Biological Control Laboratory, Plant Protection Department, College of Agricultural Engineering Sciences, University of Baghdad, where they were examined using an optical microscope (German company Boeco) at magnification (x4). Temperatures and relative humidity were recorded using a Hobo device (American Onset Company, Model MX1101).

Statistical Analysis: All data obtained from the greenhouse experiments were analyzed using a one-way ANOVA followed by an LSD test. The statistical analyses were performed using SPSS V.26.

Results and Discussion

The effectiveness of different essential oil treatments on the three cultivars infested by *A. gossypii* showed statistically significant differences compared to the control (Table 2). Specifically, the Naser cultivars showed significant differences ($p < 0.001$) on the first day (Figure 1), with the 10 mL/L thyme concentration being most effective in reducing the aphid population to 4% compared to the positive control (Sivanto 3% and Levo 6%). In contrast, the negative control was 23%. Moreover, the thyme 10 mL/L was still superior among the treatments in reducing the aphid population for all tested periods.

The results for the Jawaher cultivar (Figure 2) were significant among the tested treatments ($p < 0.002$). Thyme 10 mL/L and lavender 10 mL/L were the most effective in reducing the aphid population during all tested periods. Specifically, on the first day, it was 3% each for the 10 mL/L thyme and lavender, while in the positive control, it was 4% with Sivanto and 8 % using Levo; however, for the control treatment, it was 28%.

For the Barcelona cultivar, the results also indicated significant differences among the tested treatments ($p < 0.008$) (Figure 3). Thyme 10 mL/L and clove 5 mL/L and 10 mL/L were the most effective in reducing aphid populations during the tested periods. Specifically, on the first day, the aphid population was reduced to 2% with thyme 10 mL/L, while for the 5 and 10 mL/L clove, it was 2% and 3%, respectively. However, for positive control with Sivanto and Levo, it was 4% and 5%, respectively, while with negative control, it was 22%.

Table 2: Statistical differences for the three eggplant cultivars infested by *A. gossypii* following different treatments after 14 days ($p < 0.05$).

Treatments	Day	Naser		Jawaher		Barcelona	
		df	P - value	df	P - value	df	P - value
CONTROL							
THYME 1 mL/L	1	15.3	0.001	3.6	0.002	3.6	0.008
THYME 5 mL/L	2	9.7	0.001	3.8	0.001	2.8	0.007
THYME 10 mL/L							
CLOVE 1 mL/L	3	7.3	0.001	2.9	0..007	1.15	0.35
CLOVE 5 mL/L							
CLOVE 10 mL/L	7	7.4	0.001	4.7	0.001	1.5	0.16
LAVENDER 1 mL/L							
LAVENDER 5 mL/L	14	6.6	0.001	6.9	0.001	2.38	0.025
LAVENDER 10 mL/L							
SIVANTO							
LEVO							

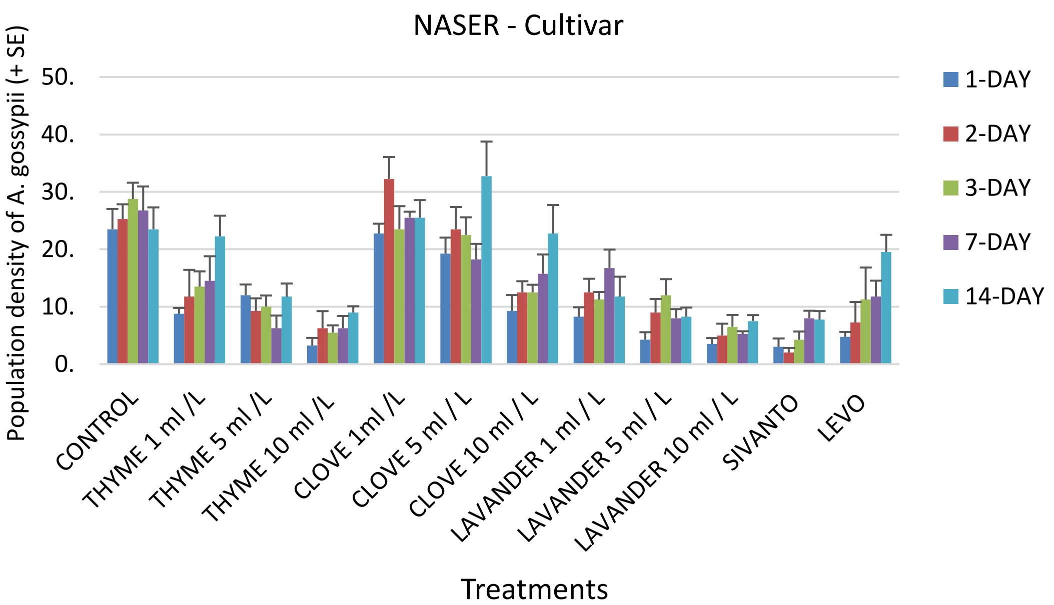

Figure 1: Population dynamics of the *A. gossypii* infested Naser eggplant cultivar following different treatments ($p < 0.05$).

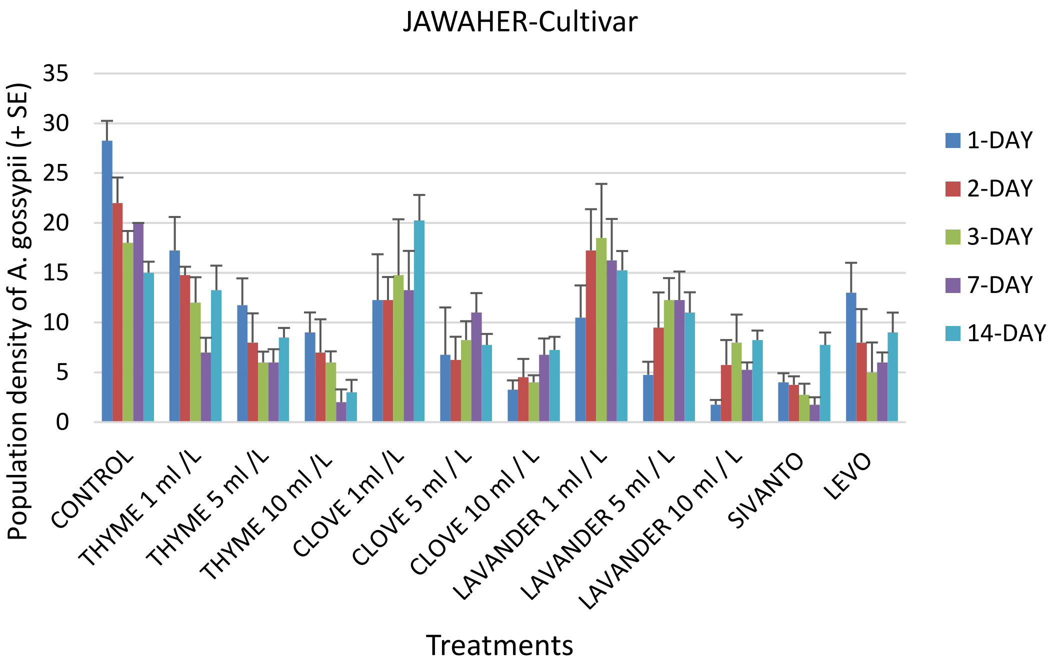

Figure 2: Population dynamics of the *A. gossypii* infested Jawaher eggplant cultivar following different treatments ($p < 0.05$).

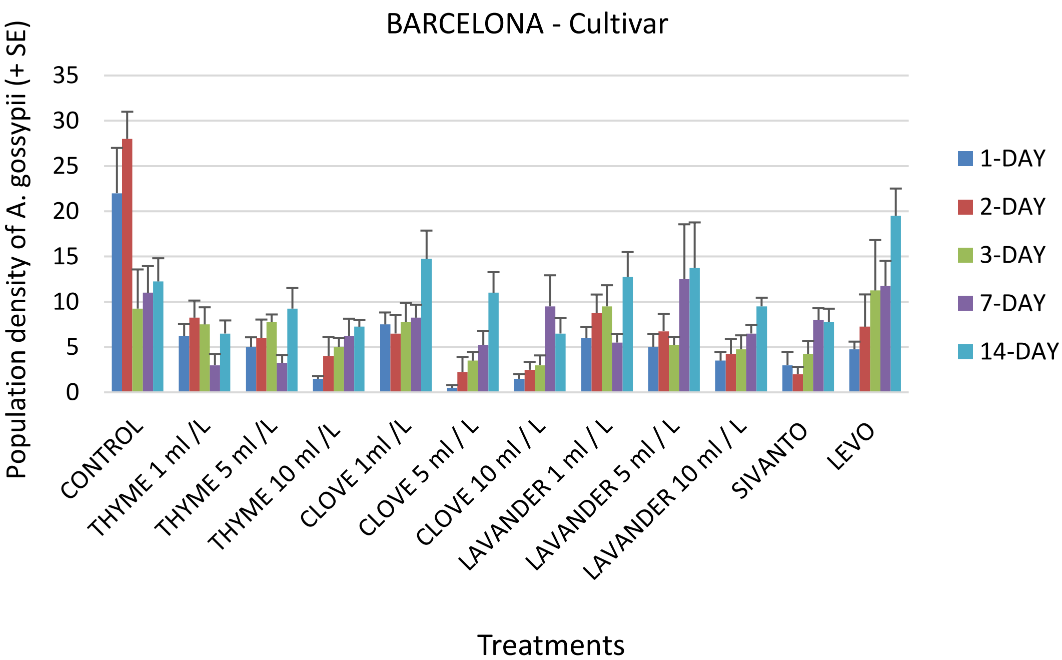

Figure 3: Population dynamics of the *A. gossypii* infested Barcelona eggplant cultivar following different treatments ($p < 0.05$).

The results showed significant effects on the adult stage of *A. gossypii*, with thyme being the most effective, followed by cloves and lavender, especially at the high dose of 10 mL/L. Studies show that exposure to essential oils can directly or indirectly affect the growth and development of the insect through the toxic effects of contact or fumigation exposure. It may also have antifeedants, repellents, or even be attractive to natural enemies. For example, *Tagetes minuta* L. essential oil significantly reduced the life cycle development of *Aulacorthum solani* Kaltenbach, *M. persicae*, and

Acyrthosiphon pisum Harris (37). Moreover, (2) indicated that essential oils can be used as biopesticides commercially because of their high toxicity against springtails *Hypogastrura vernalis* (Collembola: Hypogastruridae). The emulation formulation of essential oils caused high mortality in the *A. gossypii* population that infested sweet peppers under greenhouse conditions (1)**.**

Essential oils affect the sensory receptors and nervous system of insect pests, leading to inhibition of functions and eventual death of the insect; for example, exposure to *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae), *Trichoplusia ni* Hübner (Lepidoptera: Noctuidae) and *Callosobruchus maculatus* Fabricius (Coleoptera: Chrysomelidae) to lemongrass essential oil through contact or vaporization affects their nervous systems and negatively affects their growth and development (22, 23, 25, and 36). Studies show that the toxicity of essential oils is due to the presence of various chemical groups, such as monoterpenes, diterpenes, and sesquiterpenes (1 and 2). The advantage of these compounds is that they are lipophilic and highly volatile, which helps them easily penetrate the bodies of insects, affecting their physiological functions and causing death, or they may indirectly affect aphids by attracting natural enemies (16). Also, the quantity and quality of compounds may determine the level of toxicity, where small concentrations may directly or indirectly affect one of the vital functions, such as antagonism or synergy effects (36). The essential oils used on celery bunches infested with *Hypogastrura vernalis* in the study by (2) were found useful in reducing the pest population**.**

The results of this study show that thyme oil was the most effective against the adult stage of aphids. This toxic activity is due to one of its components being thymol and carvacrol (27 and 35). This study also proved that clove oil has a toxic effect on aphid adults because it contains compounds having toxic activity, such as eugenol (88.61%), eugenol acetate (8.89%), and β-caryophyllene, which form 99.89% of its components (10).

Finally, lavender (*L. coronopifolia*) also showed high toxicity toward *A. gossypii* adults. Lavender species have been widely investigated against insect pests. For example, *L. angustifolia* Mill showed insecticidal activity against the pea aphid *A. pisum*, and this activity was due to the chemical structure in which linalool was the most abundant component (38.57%), followed by linalyl acetate (29.95%), 1.8-cineole (13.66%), camphor (13.13%), alpha-pinene (3.14%), and terpene-4-ol (1.54%) (5).

Conclusions

This study demonstrated the importance of using essential oils, especially thyme oil, as a safe and effective pesticide against adults of the A. gossypii aphid. It reduces toxicity towards natural enemies, is low-cost, and has minimal impact on the environment, thus contributing to the Sustainable Development Goal of having clean agriculture.

Supplementary Materials:

No Supplementary Materials.

Author Contributions:

Conceptualization - Qasim and Alaa; Data curation - Qasim; Investigation - Qasim; Methodology - Qasim and Alaa; Project administration - Qasim; Resources - Qasim and Alaa; Supervision - Qasim; Writing original draft - Alaa; Writing, reviewing and editing - Qasim and Alaa. All authors have read and agreed to the published version of the manuscript.

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All data availability statement.

Conflicts of Interest:

The authors declare no conflict of interest.

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