Evaluation of the efficiency of hot aqueous extract of *Bougainvillea glabra* and spore suspension *Beauveria bassiana* (Bals) to control *Myzus persicae* (Sulzer) (Aphididae: Homoptera)

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Abstract

The purpose of this study was to determine the efficiency of the hot aqueous extract of *Bougainvillaea glabra* and fungal extract *Beauveria bassiana* in the evaluation of its efficiency in controlling the green peach aphid *Myzus persicae*Many different active compounds were identified by the GC-Mass, depending on retention time and peak area, among these were caprolactam, arginine, methyl ester, phytol, oleic acid, 9,12-octadecadienoic acid, trifluoroacetate, and hydrazine carboxamide.

We prepared three concentrations of the hot aqueous extract of *Bougainvillaea glabra* leaves (0.25,0. 50, and 0.75%) to assess its impact on the mortality of the second nymph instar. Similarly, we prepared three concentrations of the *Beauveria bassiana* fungus (0.25, 0.50, and 0.75%) to assess its effectiveness in the mortality of of the second nymph instar of the green peach aphid.

The results showed that second-instar mortality occurred more often when the concentrations of the hot aqueous extract and the fungal extract were raised compared to the control treatment. At a concentration of 0.75%, the aqueous extract had the highest mortality rate, at 28.6%.

However, at a concentration of 0.25, the lowest percentage of mortality was 0.26%. The concentration of the fungal extract *B. bassiana* fungus had a significant effect on the mortality rate. At a concentration of 0.75%, the mortality rate was 62.8%, which was higher than at a concentration of 0.75%.

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The mortality rate was the lowest at 32.3% at a concentration of 0.25%. As for the effect of the period factor, the mortality irate increased with an increase in the time period, reaching 32.3, 44.8, and 59.2%, respectively, after 1 and 3 days.
As for the interaction between concentrations and time duration, the highest percentage of mortality was 86.3 at the 0.75% concentration on the third day, while the lowest rate was 24.0 at the 0.25% concentration on the first day.
The hot aqueous extract and fungus extract have been found to influence the death rate of the green peach aphid. These extracts are considered safe alternatives to chemical pesticides, as they are cost-effective and environmentally benign.

Key words : GC-Mass, agricultural crop, *Beauveria bassiana* , *Myzus persicae* and hot aqueous extract.

Introduction

Aphids are among the most important pests in agricultural crop production due to their high reproductive ability and unique nutritional ability (Nalam *et al*., 2019).
It causes significant damage to it, either directly or indirectly, by absorbing the plant's juices and secreting honeydew, on which mould fungi grow, and dust collects, leading to deformities, yellowing, and stunting in the plant. It is also considered an indirect transmitter of many plant viruses to plants (Sadeghi *et al*., 2008).
Bougainvillea glabra, which belongs to the Nyctaginaceae family, is one of the most important ornamental plants in India. It is a source of betacyanin, which has excellent anti-diabetic, anti-microbial, and antioxidant properties (Shaheena *et al*., 2019).
The main active substances in *Bougainvillea glabra* are flavonoids, alkaloids, proteins, tannins, and phenolic compounds (Liu *et al*., 2019).
Manufactured chemical pesticides effectively controlled conventional pests, but repeated and incorrect use led to significant damage, including poisoning of non-target organisms such as bees and fish and environmental damage (Paoletti & Pimentel,2000).
Because they contained compounds similar to those in manufactured pesticides, known for their high toxicity to pests and rapid decomposition in the environment, the available alternatives to botanical insecticides had sufficient ability to combat insect pests (Rahman, et al; 2016).
Plant extracts are safe and do not harm the environment due to the extraction and concentration of the active substances using water and water-soluble organic solvents (Leong et al., 2018).
B. glabra extract gave 100% mortality at concentrations of 10%, 5%, 96.4%, and 3% against the termite insect *Heterotermes indicola* (Aihetasham et al.;2017).
The effectiveness of *Citrullus colocynthis* and Nicotiana extract in controlling aphids, and the highest percentage of mortality reached (100% and 70%, respectively) (AL-juboory, et al; 2015).
Many plant extracts possess toxic properties against various insect pests and affect the behaviour of target pests (Abbas et al., 2013).
Plants are environmentally friendly because they are biodegradable and have no negative

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impact on our health or the environment. Plant products regulate the growth and development of insects and possess insecticidal and antifeedant properties. However, they also negatively affect small organisms (Edori & Ekpeti, 2015).

The study's goal was to find the compounds in the *Bougainvillea glabra* using GC-Mass analysis and to find the best concentration of the *Beauveria bassiana* fungus's aqueous extract and filtrate to control the green peach aphids.

Materials and methods

Prepare samples

M. persicae nymph and adults were collected from the *Malva parviflora* and *Beta Vulgaris* in the Al-Mahawil area by observing their presence on the plants. They were raised in a laboratory in an incubator with a temperature of 25 ± 1 C and a relative humidity of 65 ± 5% to obtain on the colony (AL—Aradhi and Al-Rubaie,2009).

We obtained the *B. glabra* leaves from public gardens, washed the distilled leaves to remove dust, and dried them completely to prevent rotting. We prepared the plant extracts according to Adedire and Akinneye (2003).

Grind the *B. glabra* leaves into a fine powder using an electric grinder. Weigh 10 grams of the powder and dissolve it in 100 ml of boiling distilled water. After leaving the solution for 24 hours, please place it in a centrifuge for half an hour, filter it using filter paper, and then concentrate using the rotary to evaporate and dry it.

The substance was dried in an electric oven at 45°C. Then, the dried residue was taken, placed in glass containers with a known weight, and stored in the refrigerator until needed. The process was repeated several times to obtain the required quantity.

To determine the biological effectiveness of the aqueous extract of *B. glabra* leaves, 4 g of the dried extract was taken, dissolved in 5 ml of distilled water and 3 ml of Tween 20. The volume was completed to 100 ml with distilled water, and the concentration of the stock solution became 4%, or the equivalent of 40 mg/ml. Concentrations were prepared (0.25%,0.50%, and 0.75% mg/ml). The control treatment was distilled water only.(AL—Aradhi and Al-Rubaie,2009).

GC-MS analysis of bioactive compounds in hot aqueous extract of *Bougainvillea glabra* leaves

Identified the active compounds in the essential oil of the *B. glabra* plant using a chromatographic Gc- MS QP2010 Ultra, JAPAN, SHIMADU

The Gc- MS has a silica column that measures (30 m x 0.25 mm,i.d.0.25 μm).

The operating conditions of the Gc—MS were as follows: helium was used as a carrier gas at a flow rate of 0.89 ml/min, the injector temperature was 250 °C, and the detector temperature was 280 °C.held the column temperature was held at 50°C for 1 minute, then raised to 130°C at a rate of 20°/min.

We then held the temperature for 1 minute, raised it from 130°C to 250°C at a rate of 9°/min, and fixed it at 250°C for 8.33 minutes, resulting in a total duration of 27.66 minutes

for the thermal programme. The sample injected into the device contained one microliter of oil (Alia & Akrama,2019).

Effect of a hot aqueous extract of *Bougainvillea glabra* on the mortality of the second aphid nymphal instar

Use Petri dishes and place sterile filter paper in each. Place *Beta Vulgaris* leaves in the dishes after wrapping their stems with sterile cotton moistened with water to feed the insect. Ten nymphs, three for each concentration, were placed in each replicate. Using a sterile medical syringe, we sprayed the replicates with extract and distilled water concentrations as a control treatment at a 1 ml/replicate rate.

The Petri dishes were surrounded with adhesive tape to prevent the treatment of aphids from leaving, and they were then transferred to the incubator at a temperature of (25 ± 1) C and a relative humidity of (65 ± 5)%. We recorded the mortality rates in the second nymphal instar after 24, 48, and 72 hours.

Effect of Fungus *Beauveria bassiana* on the mortality of the second nymph

We prepared three concentrations of the filtrate (0.25,0. 50, and 0.75%) by withdrawing a portion using a sterile medical syringe and then adding sterile distilled water to 100 ml. The control treatment consisted only of distilled water.

We treated the second nymphal instar using the same method as we treated the aqueous extract, but we replaced the aqueous extract concentrations with those of the fungal extract. We calculated the percentage of mortality using the Abbot equation.

$$\text{Mortality Rate}=\frac{\textit{Death rate per treatment}\,\% - \textit{death rate in control treatments}\%}{100 - \textit{Death rate in control treatments}}\times 100$$

Number of dead after test

Total number of used in the test

Result and discussion

GC-MS analysis of bioactive compounds in hot aqueous extract of *Bougainvillea glabra* leaves

The results of the GC-MS analysis revealed the presence of many active compounds in the Bougainvillea leaf extract, with up to 30 identified compounds belonging to different chemical groups (Figure 1). The eight most important compounds are determined by their retention time and area, including hydrazine carboxamide, caprolactam, and tri isobutyl. (3-phenylpropoxy), Arginine, methyl ester, Phytol, Oleic Acid, 9,12-Octadecadienoic Acid, and Trifluoroacetate.

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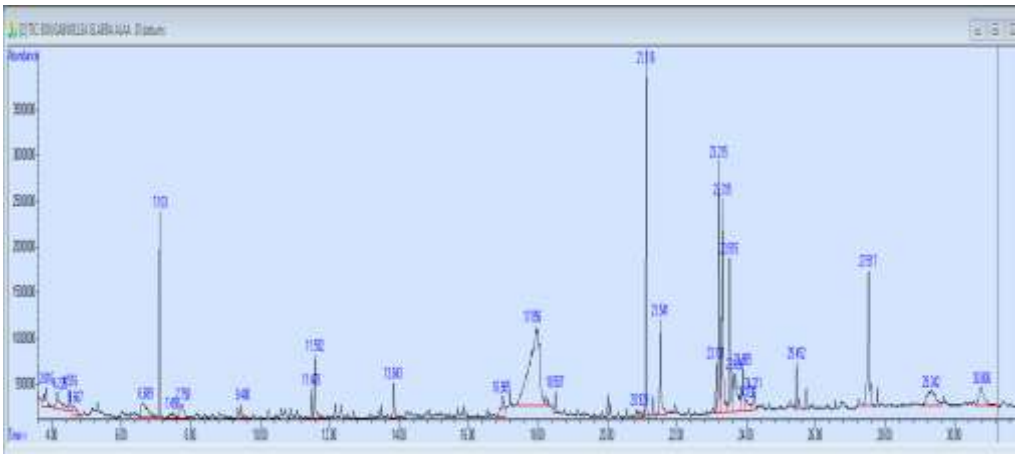

Figure 1 shows the chromatogram of the chemical compounds isolated from Bougainvillea leaf extract.

It also agrees with Liu et al. (2019), who indicated that the Bougainvillea plant contains main chemicals such as flavonoids, alkaloids, proteins, tannins, and phenolic compounds.

Effect of a hot aqueous extract of Bougainvillea on the mortality of the second aphid nymphal instars

The results in Table 1 showed the effect of the concentrations of hot aqueous extract on the leaves of the Bougainvillea plant on the mortality of the second instar of the green peach aphid. The statistical analysis results indicated the significance of the differences found due to the effect of the extract concentrations on the percentage of mortality. The increased concentrations of the extract led to a rise in mortality rates when compared to the control treatment. The highest mortality rate was 28.6% at a concentration of 0.75%; the highest mortality rate was 28.6%.

Naidu et al. (2016) screened the Bougainvillea extract for its components, which included flavonoids, tannins, phenolic compounds, carbohydrates, proteins, and alkaloids.

The lowest percentage of the mortality rate was 0.26 at a concentration of 0.25%. As for the effect of the period factor, the mortality rate increased with an increase in the period factor, reaching 14.3, 30.3, and 41.3%, respectively, after 72, 48, and 24 hours. Regarding the interference's effect, Between concentrations and periods, the highest percentage of mortality was 41.3% at a concentration of 0.75% after 72 hours of treatment, while the lowest percentage was 0.0 at 0.25% after 24 hours of treatment.

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Table 1: Effect of a hot aqueous extract of Bougainvillea on the mortality of the second Myzus persicae nymphal instars

Time peroid(day) Concentrations(%)	24	48	72	Mean
0	0.0	0.0	0.0	0.0
0.25	0.0	0.10	0.16	0.26
0.50	0.15	2.3	4.3	2.25
0.75	14.3	30.3	41.3	28.6
Mean	4.8	10.9	15.2	---
L.S.D of concentrations (0.05)				4.2
L.S.D of time peroid (0.05)				2.2
L.S.D of Interference (0.05)				6.0

This study aligned with the findings of Alhatab's 2019 study; the hexane extract of the *Carissa* was superior in controlling green peach aphid nymphs in the laboratory, as the highest mortality rate reached 46.95 at a concentration of 20 mg/ml after 72 hours of treatment.

The ethyl alcohol extract of *Lautropia mirabilis* leaves on the corn aphid extract demonstrated its effectiveness on nymphs and adults, resulting in a mortality rate of 54.8% and 46.9% at a concentration of 20 mg/ml, respectively, after 48 hours of treatment, compared to 6.1 and 12.3% in the control treatment (Hatem, 2019).

The high mortality rate of nymphs compared to adults is due to their incomplete defence systems and other morphological and physiological characteristics (El-Kady, 2010).

Effect of *Beauveria bassiana* fungus on the mortality of the second aphid nymphal instars

The results of Table (3) demonstrated the impact of the interaction between the concentrations of the fungus *B.bassiana* filtrate and the period on the percentage mortality rate of the second larval instar of the green peach *M. persicae*. The statistical analysis revealed the significance of the differences observed due to the concentrations of the fungus *B.bassiana* filtrate. At a concentration of 0.75%, mortality was 62.8%, while at a concentration of 0.25%, the lowest percentage of deaths was 32.3%.

As for the effect of the period factor, the mortality rates increased with increasing period, reaching 32.3, 44.8, and 59.2%, respectively, after 1 and 3 days. As for the effect of the interaction between concentrations and period, the highest percentage of mortality was 86.3 at a concentration of 0.75% on the third day. While the lowest percentage reached 24.0 at a concentration of 0.25% on the first day, no percentage of mortality occurred for the control treatment.

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Table 3: Effect of *Beauveria bassiana* fungus on the mortality of the second *Myzus persicae* nymphal instars

Time peroid(day) Concentrations(%)	24 hr	48 hr	72 hr	Mean
0	0.0	0.0	0.0	0.0
0.25	24.0	33.0	40.0	32.3
0.50	30.7	41.3	51.3	41.1
0.75	42.3	60.3	86.3	62.8
Mean	32.3	44.8	59.2	---
L.S.D of concentrations (0.05)				4.2
L.S.D of time peroid (0.05)				4.2
L.S.D of Interference (0.05)				8.1

Al-Shuwaili (2010) used the fungal filtrate *B. bassiana* against the black bean aphid Aphis fapa, and this study supported his findings. A 100% concentration of the fungal filtrate resulted in a 54.10 and 52.17 per cent mortality rate for nymphs and adults, respectively.

The reason for the mortality is that the higher concentrations of the fungal filtrate lead to the accumulation of toxic substances in the insect cells, causing them to explode.

These results are consistent with Ghailan and Al-Masoudi's (2013) study, in which the filtrate of the fungus *B.bassiana* caused a percentage death to the larvae of the insect *Spodoptera litura*, which reached 73.33% at a concentration of 100%.

The fungus's ability to secrete enzymes like chitinase, protease, and lipase, which break down the body wall, is crucial in destroying the insect's body.

The secondary metabolites of the fungus, which can interfere with the immune system and cause changes in the host's behaviour, such as a reduction in nutrition and activity, paralysis of the insect, and changes in tissue structures, maybe the cause of the high mortality rate (Charnley, 2003).

Increased concentrations of mycotoxins lead to an increase in the mortality rate after the entry of these toxins. This occurs through contact with the body wall, the mouth, the respiratory stomata, or a fungus penetrating the body wall; the areas where the rings articulate are the most susceptible to penetration, which weakens the insect's defences and increases its susceptibility to disease infection (Hatem, 2020).

Conclusions

We conclude that hot aqueous and fungus extract affect the mortality of the green peach aphid, as they are safe alternatives to chemical pesticides, have an economical cost and are environmentally friendly pesticides

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