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Article

Efficiency of the fungal filtrate Beauveria bassiana and the alcoholic extract of Artemisia herba-alba against the greater wax worm Galleria mellonella L. (Lepidoptera: Pyralidae) in Babylon Province

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ABSTRACT

A laboratory study was conducted to determine the effect of the filtrate of the fungus Beauveria bassiana and the alcoholic extract of the wormwood plant Artemisia herba-alba on the destruction of the larval stages of the waxworm major; the results of the study showed that the filter of the fungus B. bassiana affected the destruction of the larval stages of the waxworm. The highest mortality rate was 76.7% for the second larval stage after 24 hours of treatment with a 30% concentration, while it reached 86.7% after 168 hours of treatment with the same concentration. The lowest mortality rate was 13.3% for the last larval stage after 24 hours of treatment, with a concentration of 10%. In comparison, the death rate was 30.0% after 168 hours of treatment, with a concentration of 10%. The results also showed that the alcoholic extract of the wormwood plant affected the destruction of the larval stages of the insect.

Keywords: Fungal; Alcoholic Extract; Wormwood.

INTRODUCTION

The wax moth Galleria mellonella of the Lepidoptera family Pyralidae is one of the most important pests of bees that cause great damage to beekeeping and the main destroyer of wax tablets to feed on them and the tunnels that kill them. Its larvae feed on tire wax, honey, pollen grains, leftover tissue that destroys honey, and its residues of molting skins that destroy honeybee cells and expel the remaining bees. Honey bee colonies are exposed to many pests and diseases that cause serious economic damage to both agriculture and beekeepers and have significantly reduced the number of healthy beekeepers. In addition to the losses in bee hive products [1, 2, 3, 4]. Pest control was carried out mainly through chemical insecticides, and chemical control is one of the most used methods in controlling the great wax worm in the store. It may require a special arrangement for the possibility that pesticides will reach honey, such as methyl bromide, sulfur dioxide, aluminum phosphate, Paradichlorobenzene, Calcium cyanide, and others [1, 2, 3, 4]. Excessive and indiscriminate use of traditional, non-specialized, and highly toxic chemical pesticides in controlling insect pests has led to the development of resistance by them against a wide range of chemical pesticides, as well as the emergence of many

health and environmental problems and their negative side effects in non-targeted organisms, including parasites, predators and bees. In addition to the risks of their residuals on human and animal health. For this reason, the world has recently turned to finding alternative methods for modern and safe chemical pesticides that are more effective and compatible with the environment, or what is called today the environmentally friendly alternative methods included in the integrated management program [5]. The tendency was to use natural enemies, plant extracts, and physical methods in pest control [6, 7, 8,] and 9. When researchers seek to find natural substances of plant origin that do not have side effects, the role of plant extracts is very important in combating the great wax worm G. mellonella. In this context, the importance of this research comes to finding important compounds in Artemisia herba alba. Coinciding with the recent development in the field of biological technologies and modern biological preparations, which are commonly used in many countries of the developed world as an alternative to chemical pesticides [10], and one of the species that has the leading position in the field of biocontrol is the mushroom Beauveria bassiana because of its distinct characteristics and mechanisms that made it advanced elements in the manufacture of bio-fertilizers and pesticides. The use of mushrooms is one of the alternative methods for controlling harmful insects, including the great wax worm, as it does not constitute any environmental pollution and is safe for humans, as the International Environmental Protection Agency (EPA) in 1999 excluded B. bassiana from the list of fungi prohibited to be used freely, as it was produced Mushrooms as a biological preparation, and the most important biological preparations are Mycotrol and Boverin.

MATERIALS AND METHODS

Insect Collection and Breeding

This experiment was conducted in the insect lab for postgraduate studies in the Department of Bio-control Techniques / Al-Musayyib Technical College. Tires infected with the waxworm G.mellonella L. were obtained from different apiaries in Babylon Governorate. Wax infested with all roles of the major waxworm G.mellonella L. They were placed in plastic bottles with a diameter of 20 cm and a height of 30 cm, and their nozzles were closed with a muslin cloth and tied with a rubber band. On the top of the bottles, pieces saturated with a sugar solution 100% were placed to feed the emerging adults and stimulate them to lay eggs to obtain the first larval phase. Three replicates were used for each treatment, with ten larvae for each replicate. Dishes with a diameter of 10 cm were used. Then, samples of adults and both sexes were sent to the Natural History Museum and Research Center / University of Baghdad. Diagnosed by AMD. Hana Hani Abdel Hussein Al-Saffar.

Fungi Beauveria Bassiana

An isolate was obtained from the Biotechnology Laboratory / Department of Biological Resistance, Al-Musayyib Technical College. It was diagnosed by A. Dr . Aahd Abd Ali Hadi. The isolate was grown on sterile potato agar and dextrose culture medium prepared in Petri dishes. After the incubation period, the dishes were kept in the refrigerator for use in subsequent experiments.

Culture Medium

Medium Potato Dextrose Agar (PDA)

39 gm was taken from the ready-made PDA culture medium produced by (Himedia, India). Dissolve in 1L of distilled water in a 1L glass beaker in a water bath and add the antibiotic Chloramphenicol at 250 mg/L. The center is divided into 250 ml glass beakers at a rate of 150 ml for each beaker, sealed its nozzle with cotton plugs, and then sterilized with an osmosis device at a temperature of 121

°C. And pressure 15 pounds / ang for 20 minutes and then leave to cool and keep in the refrigerator until use. Used to isolate and purify fungi.

Medium Potato Dextrose Broth

200 g of peeled and cut potatoes were boiled with 500 ml of distilled water for 20 minutes in a 1L glass beaker. The cooked potatoes were filtered through a clean gauze cloth. The filtrate was taken, and 20 g of dextrose was added. Fill the volume to 1L by adding distilled water. Distribute the filtrate into 250 ml glass beakers at a 150 ml/beaker rate. Sterilized by a purifier at a temperature of 121°C and a pressure of 15 lbs. Ang for 20 minutes. Used to prepare filter fungi.

Preserving Innate Isolation

Discs of 5 mm in diameter were transferred from the edge of the purified one-week-old fungal colonies by cork piercing with a sterile needle into 15 ml test tubes containing sterile/diluted PDA culture medium supplemented with the antibiotic Chloramphenicol in an amount of 250 mg/L. The tubes were incubated at 25 ± 2 °C for seven days and then kept in the refrigerator until use in subsequent experiments, considering the renewal of the isolates as needed.

Preparation of Mushroom Filtrate

PDB liquid nutrient medium was prepared as in paragraph 3. 5 . 1 . 2 Distribute into 250 ml and 150 ml/beakers. Chloramphenical antibiotic in 250 mg/L was added, inoculated with three 5 mm diameter discs with a cork piercing from the edge of the purified fungal colonies in PDA culture medium and diagnosed at seven days of age. The flasks were incubated at 25 ± 2 °C, taking into account the shaking of the flasks every 3-4 days to distribute the fungal growth. After 28 days, the inoculum was filtered using a Whatman No. 1 filter paper with a Buechner funnel with the help of a vacuum pump and re-filtered using a Millipore microfilter. The filter was used in subsequent experiments. The laboratory work was carried out in the postgraduate laboratory of the Department of Biological Resistance / Al-Musayyib Technical College / Al-Furat Al-Awsat Technical University.

Preparation of Plant Parts for Study

The aerial parts of the wormwood plant were obtained from the local markets in the province of Babylon. The plant parts were cleaned well, and a quantity of them dried. Then, the plant parts were ground with an electric grinder to obtain the powder and kept in a glass bottle in the refrigerator until use. The plant was classified by A. M. Dr . Ibrahim Mardi Radi from the Musayyib Technical College / Department of Plant Production.

Preparation of Extracts

Preparation of Alcoholic Extracts of Wormwood Alba A. Herb

Method [11] was followed in preparing alcoholic extracts by taking 10 g of dry matter powder from the wormwood plant and placing it in the Soxhlet extractor. 200ml of ethyl alcohol was added and extracted for 24 hours at a temperature of 45°C. Then, the dry residual was taken and placed in a tightly sealed glass container with a known weight and kept in the refrigerator until use. To determine the biological activity of the crude alcoholic extract of wormwood plants, 2 g of the dry residue of the extract was taken. If 2 ml of ethyl alcohol was dissolved and the volume was completed to 100 ml with distilled water, the concentration of the basic solution became 2%, equivalent to 20 ml\mg, and concentrations (0.5 - 1.0 - 1.5) ml\mg were prepared from it. The control treatment was 2 ml of ethyl alcohol, 2 ml of ethyl alcohol and 2 ml of ethyl alcohol. Diffuser material, then complete the volume to 100 ml of distilled water.

Statistical Analysis

The data experiments were analyzed according to the factorial experiment model and a completely randomized Facepperiments with a completely randomized Design (CRD) and using the least significant difference test (LSD) under the probability level (50.0) to show the significance of the existing differences. The percentage of loss was corrected according to the Abbott equation.

$$\text{Corrected mortality rate\%}=\frac{\text{mortality } in\ treatment\%-mortality\ in\ control\ treatment\%}{mortality\ in\ control\ treatment\%-100}\times 100 \quad (1)$$

The corrected values were converted to angular values not included in the statistical analysis. The statistical program (2011) (GenStat) General Statistics was used in the analysis.

RESULTS

Effect of using the alcoholic extract of wormwood herba-alba A. on the killing of the second-stage larvae of the great waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours)

The results of the statistical analysis of the data (Table 1) showed that there were significant differences, where the concentration exceeded 1.5 mg/ml, which gave a fatality rate for the second phase amounted to (43.3, 53.3, 66.7%) for the periods 24 hours, 72 hours and 168 hours, respectively, which showed significant differences compared to With the control treatment, which gave a depreciation rate of 0.0% for all previous periods. The concentration of 0.5 mg/ml gave a mortality rate for the second phase amounted to (23.3, 26.7, 33.3%) for 24 hours, 72 hours and 168 hours, respectively, with a significant difference from the comparison treatment, which gave 0.0%. We did not find significant differences between concentrations (1.5 and 1.0) mg/ml and (1.0 and 0.5) mg/ml. In contrast, significant differences were found between (1.5 and 0.5) mg/ml, and the 168 hours had the highest mortality rate in all concentrations than 72 hours and 24 hours. These results are in agreement with the study conducted by [12], where the alcoholic extract of the wormwood plant surpassed the highest effect of destroying the second-stage larvae of the cotton leafworm, which reached (0.025 and 0.016 mg L-1) and (0.140 and 0.065 mg L). -1) and (0.127 and 0.012 mg L-1) after 72 and 96 hours. We conclude from Table (1) that the increase in concentration led to an increase in the percentage of loss and an increase in time. The cause of the death of the second phase larvae may be attributed to the treatment with wormwood extract. Studies have shown that wormwood extract is acidic because it contains many glycosidic compounds, phenolics, saponins, resins, flavonoids, alkaloids and terpenes. The inhibitory activity of wormwood extract is due to the glycoside compounds, which represent one of the active compounds in this plant, such as santhonine glycoside and thujone glycoside [5, 13, 14].

concentration mg/ml				**Concentration rate**
	24hours	72hours	168hours	
0.5	23.3	26.7	33.3	27.8
1.0	36.7	40.0	50.0	42.2
1.5	43.3	53.3	66.7	54.4
control	0.0	0.0	0.0	0.0
pesticide 50mg/dish	100.0	100.0	100.0	100.0
time rate	40.7	44.0	50.0	
L.S.D Concentration = 9.73		**Time Period= 7.5**		**Interaction =16.86**

Table 1. The effect of using the alcoholic extract of wormwood herba-alba A. on the destruction of the second-stage larvae of the Great Waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours)

The effect of using the alcoholic extract of wormwood herba - alba A. on the killing of the fourth phase larvae of the Great Waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours)

The results of the statistical analysis of the data (Table 2) showed significant differences where the concentration exceeded 1.5 mg/ml. The mortality rate for the fourth phase amounted to (30.0, 43.3, 56.7%) for 24 hours, 72 hours and 168 hours, respectively, which showed significant differences compared to the control treatment, which gave a death rate of 0.0% for all previous periods. The concentration of 0.5 mg/ml gave a fatality rate for the fourth phase that amounted to (3.3, 16.7, and 23.3%) for 24 hours, 72 hours and 168 hours, respectively. And with a significant difference from the control treatment, which gave 0.0%. Significant differences were found between concentrations (1.5 and 1.0) mg/ml for 24 hours, 72 hours and 168 hours, respectively. While there were no significant differences between the two concentrations (1.0 and 0.5) mg/ml for 24 and 72 hours. However, significant differences were found for the two concentrations (1.0 and 0.5) mg/ml in 168 hours. Also, significant differences were found between the average periods. The 168 hours were characterized by giving the highest percentage of mortality in all concentrations, with significant differences from the period of 72 hours and 24 hours. This study agrees with [15], where the alcoholic extract of the wormwood plant had a role in eliminating the first and third larval stages of the sugar beet worm Spodoptera exigua. The percentage of death in the first and third larval phases after 72 hours of treatment was (58.89, 52.06%), respectively. Increasing the concentration of the alcoholic extract leads to an increase in the percentage of larval mortality after 72 hours of treatment, which amounted to (46.89, 53.73%, and 67.219%) due to the concentration of 0.5, 1.0, and 1.5, respectively. We conclude from Table (2) that the increase in concentration led to an increase in the percentage of loss and an increase in time. The cause of the death of the fourth phase larvae may be due to the treatment with wormwood extract. Studies have shown that the extract of the wormwood plant has an acidic nature because it contains many glycosidic compounds, phenolics, saponins, resins, flavonoids, alkaloids and terpenes. The inhibitory activity of wormwood extract is due to the glycoside compounds, which represent one of the active compounds in this plant, such as santhonine glycoside and thujone glycoside [5, 13, 14].

concentration mg/ml				**Concentration rate**
	24hours	72hours	168hours	
0.5	3.3	16.7	23.3	14.4
1.0	6.7	20.0	36.7	21.1
1.5	30.0	43.3	56.7	43.3
control	0.0	0.0	0.0	0.0
pesticide 50mg/dish	100.0	100.0	100.0	100.0
time rate	28.0	36.0	43.3	
L.S.D Concentration = 7.99		**Time Period= 6.19**		**Interaction =13.84**

Table 2. The effect of using an alcoholic extract of the herba-alba A. wormwood plant on destroying the fourth phase larvae of the greater wax worm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours).

The effect of using the alcoholic extract of wormwood herba-alba A. on the destruction of the last phase larvae of the waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours)

The results of the statistical analysis of the data (Table 3) showed that the concentration 1.5 mg/ml excelled where it gave a mortality rate for the last phase of (6.67, 13.33, 20.00%) for the periods 24 hours, 72 hours and 168 hours, respectively, which showed significant differences compared to the control treatment, which gave The depreciation rate is 0.0% for all previous periods. The concentration of 0.5 mg/ml gave a mortality rate for the last phase that reached (0.00, 3.33, 13.33%) for 24 hours, 72 hours and 168 hours, respectively, with a significant difference from the control treatment, which gave 0.0%. No significant difference exists between the concentrations (1.5 and 1.0) mg/ml. Also, there was no significant difference between the two concentrations (1.0 and 0.5 mg/ml) for 24 hours, 72 hours and 168 hours. Significant differences were found between the average concentration (1.5 and 0.5 mg/ml). The period of 168 hours was characterized by giving the highest mortality rate in all concentrations than the period of 72 hours and 24 hours. This study agrees with [15, 12]. Table (3) The effect of using the alcoholic extract of wormwood herba-alba A. on the destruction of the last phase larvae of the Great Waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours). We conclude from Table (3) that the increase in concentration led to an increase in the percentage of loss and an increase in time. The cause of the fourth phase larvae's death may be due to the treatment with wormwood extract, as studies have shown that wormwood extract has an acidic nature because it contains many glycosidic compounds, phenolics, saponins, resins, flavonoids, alkaloids and terpenes. The inhibitory activity of wormwood extract is due to the glycoside compounds, which represent one of the active compounds in this plant, such as the santhonine glycoside and thujone glycoside [5, 13, 14].

concentration mg/ml				**Concentration rate**
	24hours	72hours	168hours	
0.5	0.00	3.33	13.33	5.56
1.0	3.33	10.00	16.67	10.00
1.5	6.67	13.33	20.00	13.33
control	0.0	0.0	0.0	0.0
pesticide 50mg/dish	100.0	100.0	100.0	100.0
time rate	22.00	25.33	30.00	
L.S.D Concentration = 6.089		**Time Period = 4.716**		**Interaction = 10.546**

Table 3. The effect of using an alcoholic extract of wormwood (Herba - alba A.) on the mortality of the late phase larvae of the greater wax worm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours).

The efficiency of the fungus B. bassiana in the mortality rate of the larvae of the second stage of the great waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours)

The results of the statistical analysis of the data (Table 4) showed significant differences where the concentration exceeded 30%. The mortality rate for the second phase amounted to (76.7, 83.3, and 86.7) for the periods 24 hours, 72 hours and 168 hours, respectively, which showed significant differences compared to the control treatment, which gave a death rate of 0.0% for all previous periods. The 10% concentration gave a fatality rate for the second phase (36.7, 50.0, 56.7%) for 24 hours, 72 hours and 168 hours, respectively, with a significant difference from the control treatment, which gave 0.0%. There is a significant difference between the two concentrations (30% and 20%) in the 24 hours. There is no significant difference between the two concentrations (20% and 10%) for 24 hours, 72 hours and 168 hours. The period of 168 hours was characterized by giving the highest

percentage of mortality in all concentrations, with significant differences from the period of 72 hours and 24 hours. This study agrees with 16 that the pathogenicity of B. bassiana on the larvae of the waxworm Galleria mellonella reached 100% as a percentage of death after 3-5 days of treatment. We conclude from Table (4) that the reason for the death of the second phase larvae is that the fungus B. bassiana contains toxins, which have several functions, including poisoning events inside the host's body, which leads to its death in the end and the immune inhibition of the host's immune defenses and overcoming them as a result of the growth of the fungus inside the host and secretion of toxins Which works to reduce its effectiveness and cause paralysis in addition to lack of nutrition.

concentration mg/ml	24hours	72hours	168hours	**Concentration rate**
0.5	36.7	50.0	56.7	47.8
1.0	53.3	63.3	73.3	63.3
1.5	76.7	83.3	86.7	82.2
control	0.0	0.0	0.0	0.0
pesticide 50mg/dish	100.0	100.0	100.0	100.0
time rate	53.3	59.3	63.3	
L.S.D Concentration = 12.35 Time Period= 9.56 Interaction =21.38				

Table 4. The efficiency of the fungus B. bassiana in the mortality rate of the second phase Galleria mellonella larvae at the following times (24 hours, 72 hours, 168 hours).

The efficiency of the fungus B. bassiana in the mortality rate of the fourth phase larvae of the Great Waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours)

The results of the statistical analysis of the data (Table 5) showed that the concentration exceeded 30%, as it gave a mortality rate for the fourth phase amounted to (43.3, 56.7, 63.3%) for the period 24 hours, 72 hours and 168 hours, respectively, which showed significant differences compared to the comparison treatment, which gave a death rate of 0.0 % for all previous periods. The 10% concentration gave a fourth phase mortality rate of (26.7, 33.3, and 43.3%) for 24 hours, 72 hours and 168 hours, respectively, with a significant difference from the control treatment, which gave 0.0%. The two concentrations have no significant difference (30, 20%). Also, the two concentrations have no significant difference (20, 10%). For all periods, 24 hours, 72 hours and 168 hours. The period of 168 hours was characterized by giving the highest percentage of mortality in all concentrations, with significant differences from the period of 72 hours and 24 hours. This study agrees that the fungus B. bassiana gave the fourth larval phase mortality rate of 100% at the concentration (10 6 x 5.5, 5.86 x 10 5 and 4.8 x 10 6) spores/ml, respectively. We conclude from Table (5) that the cause of the fourth phase larvae's death is the secretion of the protease enzyme B. bassiana, which separates and weakens the association and formation of the cytoskeleton, as well as inhibits the phagocytosis of plasma cells in the great waxworm [17].

concentration mg/ml	24hours	72hours	168hours	Concentration rate
0.5	26.7	33.3	43.3	34.4
1.0	36.7	46.7	56.7	46.7
1.5	43.3	56.7	63.3	54.4
control	0.0	0.0	0.0	0.0
pesticide 50mg/dish	100.0	100.0	100.0	100.0
period rate	41.3	47.3	52.7	
L.S.D Concentration = 13.62 Time Period= 10.55 Interaction =23.58				

Table 5. The efficiency of the fungus B. bassiana in the mortality rate of the fourth phase larvae of the Great Waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours).

The efficiency of the fungus B. bassiana in the mortality rate of the last phase larvae of the great waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours)

The results of the statistical analysis of the data (Table 6) showed that there were significant differences, where the concentration exceeded 30%, as it gave a mortality rate for the last phase of (36.7, 46.7, 56.7%) for the periods 24 hours, 72 hours and 168 hours, respectively, which showed significant differences compared to the comparison treatment, which It gave a depreciation rate of 0.0% for all previous periods. The 10% concentration gave a fatality rate for the last phase amounting to (13.3, 20.0, and 30.0%) for 24 hours, 72 hours and 168 hours, respectively, with a significant difference from the comparison treatment, which gave 0.0%. There is no significant difference between the two concentrations (30% and 20%). Also, there is no significant difference between the two concentrations (20% and 10%) for 24 hours, 72 hours and 168 hours. The period of 168 hours was characterized by giving the highest percentage of mortality in all concentrations, with significant differences from the period of 72 hours and 24 hours.

concentration mg/ml	24hours	72hours	168hours	Concentration rate
0.5	13.3	20.0	30.0	21.1
1.0	26.7	33.3	43.3	34.4
1.5	36.7	46.7	56.7	46.7
control	0.0	0.0	0.0	0.0
pesticide 50mg/dish	100.0	100.0	100.0	100.0
period rate	35.3	40.0	46.0	
L.S.D Concentration = 9.41 Time Period= 7.29 Interaction =16.30				

Table 6. The efficiency of the fungus B. bassiana in the death rate of the last phase larvae of the Great Waxworm Galleria mellonella at the following times (24 hours, 72 hours, 168 hours).

DISCUSSION

Eighteen used the fungus B. bassiana to treat the eggs of the major wax moth. The percentage of unhatched eggs was (45.4%, 24.9% and 45.2%), and it was shown that the mortality rates in the different larval stages of the major wax worm increased with the increase in the concentrations of the fungus B.bassiana. Also, [15] showed that the fungus B.bassiana can kill the corn stalk borer insect, as the death rate reached 87.5-100%. The results of the study showed that the reason for the increase in the percentage of mortality by increasing the concentrations, which

leads to a weak immune system of the insect, is attributed to the cause of the death of the second and fourth larval stages, where the body wall and tissues are thin, which allows easy penetration by the fungal infiltrate, as the main method for the entry of fungi is direct penetration into the body wall Also, when appropriate conditions are available, suitable temperature and humidity are available [5]. The reason for the decrease in the sensitivity of the larvae towards the fungus with the progression of their larval age is that the older the larvae increase, the number of blood cells increases, through which the number of phagocytes floating in the blood increases, such as plasma cells and granular hemocytes, which have the greatest advantage in devouring The foreign bodies that enter the body of the larvae, and this was confirmed by [6] when they mentioned that the cellular defense factors of the squamous wing order reside in these types of cells, while the larvae of the primary stages have not completed their defense systems, especially the blood cells formed that Most of them are of the primitive type Prohaematocytes, which usually do not participate in devouring the foreign bodies entering their body cavity [7].

CONCLUSIONS

The highest mortality rate was 43.3% for the second larval stage when treated with a concentration of 1.5 mg/ml after 24 hours of treatment, and it reached 66.7% for the same larval stage and concentration after 168 hours of treatment. The mortality rate for the last larval stage was 0.00% after 24 hours of treatment with a concentration of 0.5 mg/ml and 13.33% after 168 hours of treatment with the same concentration.

References

1. Hristov, P.; Shumkova, R.; Palova, N.; Neov, B. Honey bee colony losses: Why are honey bees disappearing?. *Sociobiology*, **2021**, 68.1: e5851-e5851.
2. Olynyk, M.; Westwood, A. R.; Koper, N. Effects of natural habitat loss and edge effects on wild bees and pollination services in remnant prairies. *Environmental Entomology*, **2021**, 50.3: 732-743.
3. Wood TJ; Michez , D.; Paxton , R.J.; Drossart, M.; Neumann , P.; Gerard, M.; Vanderplanck, M.; Barraud, A.; Martinet, B.; Leclercq, N.; Vereecken, N. J. Managed honey bees as a radar for wild bee decline?. *Apidologie,* **2020**, 51.6: 1100-1116.
4. Graystock, P.; Blane, E. J.; McFrederick, Q. S.; Goulson, D.; Hughes, W. O. Do managed bees drive parasite spread and emergence in wild bees?. *International Journal for Parasitology: Parasites and Wildlife*, **2016**, 5.1: 64-75.
5. Al-Taif, I. M.; Hamoudi, M. S.; Mahmoud, A. R. R.; Shaker, M. A. Study of the active compounds and mineral elements of wormwood fruit extracts and their effect on GOT liver enzymes. *Journal of the College of Basic Education*, **2016**, 22.95: 137-150.
6. Prasad, A.; Syed, N. Evaluating prospects of fungal biopesticide Beauveria bassiana (Balsamo) against Helicoverpa armigera (Hubner): An ecosafe strategy for pesticidal pollution. *Asian J Exp Biol Sci*, **2010**, 1.3: 596-601.
7. Ferro, T. A.; Araújo, J. M.; dos Santos Pinto, B. L.; Dos Santos, J. S.; Souza, E. B.; Da Silva, B. L.; Colares, V. L.; Novais, T. M.; Filho, C. M.; Struve, C.; Calixto, J. B. Cinnamaldehyde inhibits Staphylococcus aureus virulence factors and protects against infection in a Galleria mellonella model. *Frontiers in microbiology*, **2016**, 7: 2052.
8. Silva, L. N.; Da Hora, G. C. A.; Soares, T. A. Bojer, M. S.; Ingmer, H.; Macedo, A. J. Myvicetin protects Galleria mellonella As Against Staphylococcus Aureus Infection and Inhibits Multiple Virulence Factors. Sci. Rep, **2017**, 7:2823 .
9. Bezerra Filho, C. M.; da Silva, L. C. N.; da Silva, M.V.; Løbner-Olesen, A.; Struve, C.; Krogfelt, K. A.; Correia, M. T. D. S.; Vilela Oliva, M. L. Antimicrobial and Antivirulence Action of Eugenia brejoensis Essential Oil in vitro and in vivo Invertebrate Models. *Frontiers in Microbiology*, **2020**, 11: 42.
10. Musa, A. A. M. Biological comparison of plant diseases, Academic Library, Arab Republic of Egypt, **2000**: 684.

11. Harbonne J. B. *Phytochemical methods a guide to modern techniques of plant analysis*. Springer science & business media,**1998**.
12. Khamis, W. M.; Farag, D. A.; Selim, S. Efficacy of Artemisia judaica extract and certain insecticides against cotton leafworm, Spodoptera littoralis (Lepidoptera: Noctuidae). *Egyptian Academic Journal of Biological Sciences, F. Toxicology & Pest Control*, **2018**, 10.2: 35-52.
13. Al-Ghanimi, D. A. H. *Effect of hot aqueous extract of wormwood on some functional and histological parameters of some male organs of rats induced by diabetes*. **2012**. Master Thesis. Department of Life Sciences. College of Education for Pure Sciences, Karbala University. 124.
14. Lubna, O. *Study of some biochemical properties of wormwood, Artemisia herba alba Asso*. **2010**. Master Thesis. Department of Biology. Faculty of Science, Farhat Abbas University. 90.
15. Al-Bahili, H. M. R. M.; Abdul-Hassan, H. A. J.; Mohammed, F. A. R. The effect of some biological fungi and alcoholic extracts of wormwood and bitter melon on the life of the larval stages of the sugar beet worm Hubn (Noctuidae: Lepidoptera) Spodoptera exigua on tomato. University of Maysan College of Education. *Maysan Research Journal*, **2005**, 1.2: 171-189
16. Ahmed, M.; Ghazal, I.; Rajab, L. Isolation of the fungus Beauveria bassiana and evaluation of its activity on the larvae and pupae of the cotton leafworm Spodoptera littoralis (Boisd) (Lepidoptera: Noctuidae). *Tishreen University Journal of Research and Scientific Studies*, **2016**, 38.4: 177-188.
17. Griesch, J.; Wedde, M.; Vilcinskas, A. Recognition and regulation of metalloproteinase activity in the haemolymph of Galleria mellonella: a new pathway mediating induction of humoral immune responses. *Insect Biochemistry and Molecular Biology*, **2000**, 30.6: 461-472.
18. Taher, K. H. *Laboratory study of the pathogenicity of some pathogens in the waxworm Galleria mellonella*. **2011**. Master Thesis. College of Agriculture, University of Baghdad. 107.

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