

FUNGUS BIO EFFICACY *BEAUVERIA BASSIANA* IN CONTROLLING SUNNA INSECT *EURYGASTER MAURA* (GEOFROY) (HEMIPTERA : SCUTELLERIDAE) ON THE WHEAT CROP IN THE PROVINCE OF BABYLON.

Asaad Mudhehe Al-Sultani ^{1a *} , A.M. Dr. Najeha – M. Bari ^{2b}

^{1*} Biological control technical Department , Al-Mussaib Technical College, AL-Furat Al-Awsat Technical University, (ATU). Iraq

² Biological control technical Department, Al-Mussaib Technical College, AL-Furat Al-Awsat Technical University, (ATU).
Iraq

^aE-mail assaad.modhey.tcm.2@student.atu.edu.iq

^bE-mail com.naj@atu.edu.iq

Corresponding author E-mail: assaad.modhey.tcm.2@student.atu.edu.iq

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Abstract This study was conducted at the AL- Musayyib Technical College / Department of Biological-Control Techniques for the period from 1/8 /2021. to 1/5/2022 The study aimed to test the effect of fungus filtrate *Beauveria bassiana* On adults and nymphs of the Sunna insect *E.maura* (Geoffroy) .The results of the study showed the effect of the fungus filtrate on the adults and nymphs of the Sunna insect, where the concentration of 75% gave a death rate of 60.0% for the adults during a period of 7 days, and it was significantly superior to the rest of the concentrations during the studied periods. The results also showed the effect of the filtrate on the second and fourth instar nymphs, where the concentration of 75% gave the highest mortality rate of 73.3 and 66.7%, respectively, during a period of 7 days. The results of the study also showed that the second nymph was more affected by the fungal filtrate compared to the adult and the fourth nymph.

Keywords: Sunn Pest , wheat, *Beauveria bassiana*.

1. INTRODUCTION

The wheat crop occupies *Triticum aestivum* L. Belonging to the Grass family (Gramineae) The first place among the cultivated cereal crops in the world in terms of nutritional value and demand. The nutritional value is due to the fact that it contains carbohydrates as well as proteins, gluten and some nutrients such as calcium, phosphorus and magnesium [19]. It provides the world with 55% of the total carbohydrates and 20% of the food calories, and contributes to providing 20% of the human need for food [12]. The crop is considered the first productive crop of other cereal crops globally, including rice and maize, becoming undisputed as it is cultivated within a wide range of environmental differences [15]. The crop is planted in large areas in the Iraqi country, especially in the northern region, as it occupies a large area of the total areas planted with winter crops, so the amount of wheat production in Iraq in the winter season 2019 (4343) thousand tons for a cultivated area of 6331 thousand acres. (Central Statistical Organization, 2019). During the growing season, the wheat crop is exposed to great economic losses due to being infected with a number of pests, which differ in the percentages of damage they cause, and one of the most important of these economically important pests is the Sunna insect *Eurygaster maura*

(Linnaeus) Belonging to the family Scutelleridae, order Hemiptera. Its damage is represented by feeding nymphs and adults on leaves, stems and grains, and losses in yield as a result of direct feeding range between 50-90% [14]. The severity of the Sunna insect infestation affects the ratio of starches to proteins in the components of the bean, and consequently on the total components of the paste and the activity of the amylase enzyme [13]. Sunnaa injury also affects the formation of gluten proteins in wheat and its effect on the quality of flour [27]. The activity of yeast and the fermentation time in the dough produced from wheat flour infested with the *Sunna insect*, which contains protease enzymes, Decreased production of essential m *Sunna insect* etabolites (carbon dioxide and ethanol). As well as secondary metabolites (glycerol, acetic acid, and succinic acid)

[22]. Sunna insect infestation affects the structure of the protein network surrounding the starch granules. This network is destroyed due to the activity of protease enzymes resulting from the insect infestation And it gives a viscous dough with a weak and heterogeneous gluten network and therefore bad technological specifications [9];[24]. The study was conducted to mitigate the effect of *Sunn Pest* infestation on the technological, rheological properties and specific gravity of wheat grains by mixing them with healthy grains [8]. For this reason, attention has been drawn in recent research to find alternative methods for using chemical pesticides, and the most important method is biological control[20]. Fungi and plant extracts are good means of control in this field [4]. More than 700 species of entomopathogenic fungi have been diagnosed, and these fungi infect nearly 200 species of insects, causing them to have Muscardian disease [10]. Among the most important of these fungi are *Metarhizium anisopliae* and *Beauveria bassiana*. The use of **Fungi** is one of the alternative ways to control harmful insects, including store house insects [11] Because it does not pollute the environment and is safe for humans, as the International Environmental Protection Agency (E.P.A.) in 1999 excluded **Fungus** from the list of **Fungus** prohibited for use in free form, as **Fungus** were produced as a biological preparation, Among the most important biological preparations used globally against insects are Boverin and Mycotrol [28].

Aims of the study: Study of the effect of the biological fungus *B. bassiana* on the biological performance of the *Sunna insect*. And use of the fungus as an alternative to chemical pesticides.

2. MATERIALS AND METHODS

2.1- Insect collection and breeding

The adults of the wintering Sunnaa were collected from their places of dispersal inside the sedge plant (*Imperata cylandrice*) It is located on the shoulders of the water fountains in the palm groves in the province of Babylon, where the Lemon graas plant is the secondary host for the wintering of the *Sunna insect* in the middle Euphrates region, Insects were collected at different stages during the months of August, September and October of 2021 And the month of March 2022. The insects were then transferred to the laboratory, where they were raised on wheat plants by planting seeds of Barcelona variety on sandy soil inside plastic containers with dimensions of 35 x 25 x 18 cm, according to mentioned [7]. The process of watering the seeds continued every three days using a hand sprinkler, and when the plant reached a length of 3-10 cm, 30 insects were placed inside each plastic container. With 15 males and 15 females, males were distinguished from females by observing the last ventral ring, which is divided in the female and not divided in the male, according to [3]. The open end of the containers was covered with two layers of boring cloth, openings of 2 mm in diameter, and secured with a rubber band to prevent insects from escaping. Use a 100-watt electric lamp for the purpose of obtaining a period of 16 hours of illumination. The insect was diagnosed by Assistant Professor Dr. Hana Hani Abdul-Hussein/Natural History Museum and Research Center/University of Baghdad.

2.2- Isolate the roles of the insect

The insects raised in the laboratory were monitored, and after the adult females started laying egg blocks on the boring cloth, on the leaves of the wheat plant and the walls of the plastic containers in the laboratory, with 14 eggs per block, some egg blocks were gently lifted and each block was placed in a petri dish with a diameter of 9 cm. It contains filter paper. As for the rest of the eggs, they were left until hatching. The first nymph phase took place immediately after the eggs hatched. As for the rest of the other phases, they were isolated according to the shape, size, growth of wing buds and shields, and the number of molting skins in the nymph phases that preceded them.

2.3- Culture media used for isolating and diagnosing fungi

2.3.1- medium preparation Potato Dextrose Agar (P.D.A)

Prepare the medium by taking 200 gm of potato tubers, peeled and cut into small pieces, and add 500 ml of distilled water to it. It was boiled in a glass beaker for 20 minutes, and after the end of the boiling period, the mixture was filtered with layers of medical gauze to obtain the filtrate, Dissolve 20 gm of dextrose and 17 gm of agar in another 500 ml of distilled water, then add the potato filter to it and complete the volume to 1 liter. Distribute the medium into 500 ml glass beakers and close the nozzles with cotton stoppers It was sterilized in a steam sterilizer (autoclave) at a temperature of 121°C and a pressure of 15 pounds/in for 20 minutes. After the sterilization period ended, the flasks were left to cool, then the antibiotic Chloramphenicol was added to it at a rate of 250 mg/L to reduce bacterial growth, The flasks were left to cool before solidification, then poured the medium into petri dishes with a diameter of 9 cm and left the medium to solidify and kept the dishes in the refrigerator until use[26].

2.3.2- medium preparation Potato Dextrose Broth (P.D.B).

200 g of peeled and cut into small pieces were boiled with (500) ml of distilled water for 20 minutes in a (1) liter glass beaker, the cooked potatoes were filtered through a clean gauze cloth, the filtrate was taken and added to it (20) g of dextrose, fill the volume to (1) liter by adding distilled water, distribute the filtrate into (250) ml glass beakers at a rate of (150) ml/beaker, close the nozzles with a cotton plug and sterilize with a purifier at 121 °C and a pressure of 15 psi/in for 20 One minute, the bottles were left to cool and then the antibiotic Chloramphenicol was added to them. It is used for the purpose of preparing the mushroom filtrate.

2-3-3 Preparation of mushroom filtrate *B. bassiana*

The liquid nutrient medium PDB was prepared as in the paragraph and distributed in 250 ml beakers with an amount of 150 ml / beaker. Chloramphenicol antibiotic was added at an amount of (250) mg/L, inoculated with three discs with a diameter of (5) mm that were perforated from the edge of the purified fungal colonies in PDA culture medium and diagnosed at the age of seven days. Every 3-4 days for the distribution of fungal growth, and after 28 days, the inoculum was filtered using Whatman No.1 filter paper and re-filtered using a Millpore 45 mm fine filter. After obtaining the crude filter, concentrations (25, 50 and 75%) were prepared from it for use in experiments suffix. The laboratory work was carried out in the postgraduate laboratory of the Department of Biological Resistance / Technical College / Al-Musayyib / Middle Euphrates University.

Insect role:

1-Studying the effect of 25, 50 and 75% concentrations and periods of *B. bassiana* filtrate on the destruction of sunn adults in the laboratory.

Five adult insects were taken to a petri dish with a diameter of 9 cm for the purpose of treating them with fungal infiltrates. Three replicates were used for each concentration of 25, 50 and 75% concentrations. These concentrations were prepared by withdrawing a quantity of the fungal infiltrate using a sterile medical syringe and completing by adding sterile distilled water according to each concentration, and the replicates were treated by Small manual sprayer with a capacity of 100 ml. The sprayer was calibrated before use by fixing the nozzle position and pressing the hand of the sprayer, which is 10 cm away from an empty Petri dish.

Three capsules were used for each replicator. The comparison treatment was sprayed with sterile distilled water. Insects treated with mushroom concentrations were transferred to small plastic containers of dimensions (6 x 12 x 18) tight from the top. Their covers were punctured with a sterile needle for the purpose of ventilation and put for feeding the wheat branches, to which water was added to prevent drying, and the branches were changed every three days to ensure continued feeding Insect The experiment was carried out in laboratory conditions at a temperature of 5 ± 25 °C and a humidity of 5 ± 75 °C. The mortality rate for the treated adults was calculated after 1, 3, 5 and 7) days of treatment.

2-Studying the effect of 25, 50 and 75% concentrations and periods of *B. bassiana* filtrate on the destruction of second-phase nymphs of Sunn pest in the laboratory.

The same method was followed in the previous paragraph by taking five nymphs from the second instar into a petri dish with a diameter of 9 cm for the purpose of treating them with mushroom filters. Three replicates were used for each concentration of 25, 50 and 75% concentrations. The lids of the dishes were perforated with a sterile needle for the purpose of ventilation and placed for feeding. Wheat branches were added to water to prevent drying, and the branches were changed every three days to ensure continued feeding of the insect The experiment was conducted in laboratory conditions at a temperature of 5 ± 25 °C and humidity of 5 ± 75 The mortality rate of treated nymphs was calculated after (1, 3, 5 and 7) days of treatment.

3-Studying the effect of 25, 50 and 75% concentrations and periods of *B. bassiana* filtrate on the destruction of the fourth instar nymphs of Sunn pest in the laboratory.

The same method was followed in the previous paragraph by taking five nymphs from the fourth instar into a petri dish with a diameter of 9 cm for the purpose of treating them with mushroom filtrate. Three replicates were used for each concentration of 25, 50 and 75% concentrations. The lids of the dishes were perforated with a sterile needle (needle) for the purpose of ventilation and placed for feeding. Wheat branches were added to water to prevent drying, and the branches were changed every three days to ensure continued feeding of the insect. The experiment was conducted in laboratory conditions at a temperature of 5 ± 25 °C and humidity of 5 ± 75 . The mortality rate for treated nymphs was calculated after (1, 3, 5 and 7) days of treatment.

statistical analysis:

The data experiments were analyzed according to the factorial experiment model and a completely randomized Facepperiments with completely randomized Design (C.R.D) and by using the Leas torial t significant difference test (L.S.D under the probability level (0.05) to show the significance of the existing differences, the percentage of loss was corrected according to the Abbott equation [2].

$$\text{Corrected Fatality Percentage} = \frac{(\text{The comparison is a treatment in perishing \%} - \text{treatment in perishing \%})}{(\text{The comparison in perishing \%} - 100)} \times 100$$

The corrected values were converted to angular values that are not included in the statistical analysis [25]. The statistical program (SAS) Statistical Analysis System (2012) was used in the statistical analysis.

3. RESULTS AND DISCUSSION

3.1 Studying the effect of 25, 50, 75 concentrations and time periods of *B. bassiana* filtrate on the mortality of sunn adults in the laboratory.

The results of Table (1) showed that there were significant differences in the average effect of concentrations and the rate of effect of periods, that the effect of mushrooms increased with the increase in concentration and time period, where the average effect of the concentrations gave the concentration 75%, the highest rate of death was 35.0% and the lowest rate of destruction was in the concentration 25% with an average 15.0% and 50% concentration gave a rate of 28.3%. As for the effect of the periods, the 7-day period gave the highest mortality rate of 33.3, as it differed significantly with all the studied time periods of 1, 3 and 5 days, which gave a death rate of 0.0, 18.3 and 26.7 As for the results of the interaction between concentrations and periods, they show that there are significant effects between concentrations and time periods, where the concentration of 75% gave the highest death rate of 60.00% during a time period of 7 days and the lowest death rate was in the concentration 25% during a time period of 3 days, while the All concentrations did not give any death rate during the time period 1 day. As for the comparison treatment, it did not give any death rate during the four studied periods.

The results of the study indicated that the fungal filter *B. bassiana* has a clear effect in killing the adults of the insect, and that the percentage of killing increases with the increase in the concentration of the fungal filtrate. This is due to the effects caused by the toxins present in the filtrate. Among [23] that the fungus *B. bassiana* works to produce many toxins in the liquid medium, and the most important secretion is the poison Beauvericin, which is attributed to it the main reason for killing insects treated with the fungus in addition to the ability of the fungus to produce cyclodepsipeptides toxin that accumulates in the mitochondria of cells And thus leads to its explosion and destroys the active cells in the body of the host, where these toxins reach inside the body of the host through the respiratory stomata or through feeding or by surface contact with the body of the pest, in addition to activating the enzyme Prophenoloxidase, which is important in This study agrees with the findings of [5], which showed that the .biological construction processes filtrate of the fungus *B. bassiana* had an effect on the whole of black bean *Aphis Fabae* Scopolli, as the concentration of 100% gave the highest death rate of 54.10% and that this concentration differed significantly from the concentrations of 75, 50 and 25% In which the death rates were 33.94, 30.50 and 21.50%, respectively.

Table (1) Effect of concentrations 75, 50, 25% and periods of *B. bassiana* filtrate on the mortality of sunn adults in the laboratory.

concentration %	Percentage of loss during the time period (day)				Concentration Effect Rate
	1	3	5	7	
0	0.0	0.0	0.0	0.0	0.0
25	0.0	13.3	20.0	26.7	15.0
50	0.0	26.7	40.0	46.7	28.3
75	0.0	33.3	46.7	60.0	35.0
Interval effect rate	0.0	18.3	26.7	33.3	
L.S.D 0.05	For Concentrations = 5.88, For Periods = 5.88, Overlap = 11.76				

1- Study of the effect of concentrations 75, 50, 25% and periods of *B. bassiana* filtrate on the destruction of second-phase nymphs of Sunn pest in the laboratory.

Table (2) shows that there are significant differences between the average effect of concentrations and the rate of effect of periods, where the concentration 75% gave the highest mortality rate was 47.1% and the lowest death rate was 5.0% in the comparison treatment, while concentration 25 and 50% gave a mortality rate of 6.7 and 15.8%, respectively. . As for the average effect of periods, the 7-day period gave the highest rate of 33.8%, as it differed significantly with all studied time periods of 1, 3 and 5 days, which gave a mortality rate of 8.3, 11.7 and 20.8, respectively.

As for the interaction between concentrations and periods, the results showed significant differences in the mortality rate. The concentration 75% gave the highest mortality rate of 73.3 at the 7-day period, while the lowest mortality rate in the comparison treatment was 0.0 in the comparison treatment and the concentration 25% after a period of time 1 and 3 days in a row.

[21] explained the reason for the difference in the toxicity of fungi types, perhaps due to the extent of their secretion of enzymes and mycotoxin, as these substances destroy or disrupt some tissues or may affect the growth and development of the insect through the process of parasitism. While [16] mentioned that the difference in the rates of mortality of fungi is due to their ability to secrete decomposing enzymes and mycotoxins that affect the vital activities of living organisms and thus lead to their death.

[18] tested several isolates of *B. bassiana* against adults of maize weevil *Sitophilus zeamais* (Motsch), and high significant differences appeared in the pathogenicity of the isolates, which caused a killing rate of 37-100% within the average lethal period ranging from 3 to 9 days. Between . [1], The results of previous studies proved that the percentage of killing of *B. bassiana*, was similar against other insect pests.

Table (2) shows the effect of concentrations 75, 50, 25% and periods of *B. bassiana* filtrate on the destruction of second-phase nymphs of Sunn pest in the laboratory

concentration %	Percentage of loss during the time period (day)				Concentration Effect Rate
	1	3	5	7	
0	0.0	6.7	6.7	6.7	5.0
25	6.7	0.0	6.7	13.3	6.7
50	6.7	6.7	8.3	41.7	15.8
75	20.0	33.3	61.7	73.3	47.1
Interval effect rate	8.3	11.7	20.8	33.8	
L.S.D 0.05	For Concentrations = 9.32, For Periods = 9.32, Overlap = 18.63				

2-Studying the effect of 25, 50, 75 concentrations and periods of *B. bassiana* filtrate on the destruction of the fourth instar nymphs of Sunn pest in the laboratory.

Table (3) shows that there are significant differences between the average effect of concentrations and the rate of effect of periods, where the concentration of 75% gave the highest mortality rate of 39.2%, while the lowest destruction rate was in the comparison treatment of 5.0%, while the concentration of 25 and 50% gave the death rate of 25.0 and 27.1 As for the impact rate of the time periods, the 7-day period gave the highest death rate of 43.3% and differed with all studied periods of 1, 3 and 5 days and gave a death rate of 10.0, 11.7 and 31.2%, respectively. As for the results of the interaction, it showed that there were significant differences between the concentrations and periods, where the concentration 75% gave the highest mortality rate of 66.7% during a period of 7 days, while the comparison treatment gave the lowest mortality rate of 0.0% during a period of 1 and 3 days. The results show that the effect of the fungus increases Increasing focus and time. We conclude from this that the rate of killing is directly proportional to the increase in concentration. This study agreed with [6] that the death rate was decreasing with each decrease in the concentration of *B. bassiana* mushrooms.

The results of this study confirm the ability of *B. bassiana* to achieve the best result in killing the early stages of cutworm insect under laboratory and field conditions. The use of biological control agents together or in conjunction with chemical pesticides is the future basis for integrated control to reduce the damage caused by agricultural pests, through the ability provided by biological control agents to continue to reduce pest density below the level of the critical economic limit. Explained by [17]. The fungus *B. bassiana* produces the compound Beauvericins and has the same toxic effects as dystroxin (DTX), and the production of Beauvericin increases with the higher concentration of conidia. Biological control is an alternative way to control stored grain insects. Biological control must be advanced to improve its efficiency, effectiveness and reliability.

Table (3) shows the effect of concentrations 75, 50, 25% and periods of *B. bassiana* filtrate on the destruction of the fourth instar nymphs of Sunn pest in the laboratory.

concentration %	Percentage of loss during the time period (day)				Concentration Effect Rate
	1	3	5	7	
0	0.0	0.0	6.7	13.3	5.0
25	13.3	13.3	26.7	46.6	25.0
50	6.7	13.3	41.7	46.7	27.1
75	20.0	20.0	50.0	66.7	39.2
Interval effect rate	10.0	11.7	31.2	43.3	
L.S.D 0.05	For Concentrations = 8.61, For Periods 8.61, Overlap = 17.23				

4. CONCLUSIONS

1-The biological fungus *B. bassiana* had a good effect on the percentage of killing of insect roles at different concentrations, and the percentage of death increased with increasing concentration and time period.

2- The second nymphal phase was the most affected by the fungus compared with the fourth and adult instars, and it showed weak resistance against the biological fungi.

5. Recommendations:

1-Using the biological fungus *B.bassiana* in I.P.M integrated pest management programs. It is preferable to treat with the fungus when insects descend into the field.

2-Using mushrooms on other pests, knowing their effect on them, the percentage of destruction they cause, and knowing the family range of the fungus and its parasitic ability.

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