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The biological effect of *Lecanicillium lecanii* on the fertility and Oviposition of grain weevil *Sitophilus granaries*

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ABSTRACT

The pestilence of commercially significant crops by insects threatens global food security. Over the past few decades, chemical pesticides have been widely utilized to control insect pests all over the world. These substances, however, are harmful to both the ecology and human health. Several problems have also developed resistance to a variety of pesticides. The search for environmentally suitable substitutes prompted researchers to produce biocontrol agents such as entomopathogenic fungi, this fungus belonging to a number of taxa that effectively infect and kill insects. Additionally, they are highly host-specific, have a minimal impact on organisms other than the target, and are easily and swiftly manufactured in large quantities. The entomopathogenic fungus, *Lecanicillium lecanii*, was used to probe whether it has an impact on the mating behavior, fertility, and egg production of adults *Sitophilus granaries*. showed the efficiency of the fungal concentrations on the adults at ages 24 h that effect, reflecting that effect in the mating capacity and the rate of egg production, and the lowest rate of productivity eggs when mated male's treatment with female treatment was 18.3 at 2×10^8 spores/ ml, also the lowest percentage of hatching eggs was 38 % at 2×10^8 spores/ ml and the highest rate of hatching period was 6.8 at 2×10^8 spores/ ml. when mated males untreated with females treated with fungal the rate of productivity of eggs was 18.8, 24.2, and 24.8 at concentrations (2×10^8 , 2×10^6 , 2×10^4 spores/ ml) respectively while the lowest percentage of hatching eggs was 56 % at 2×10^8 spores/ ml. This results indicate that *Lecanicillium lecanii* may have the potential for usefulness in the integrated control program of *Sitophilus granaries*.

Keywords: *Sitophilus granarius*, entomopathogenic fungus, biological control, Oviposition

1. Introduction:

The grain weevil *Sitophilus granarius* (Coleoptera: Curculionidae) is considered one of the most important primary stored grain pests, the adults and larvae attack an assortment of stored cereals, including grains of wheat, maize, oats, and rice, the infestation not only affects direct weight loss and infection but also causes stored cereals exposed to secondary storage insects and toxicogenic fungi (Nietupski, et al., 2021) In addition, the weevils can enormously reduce the germination and viability of cereals and finally reduces the nutritious and economical value of the cereal, also can actually pose health hazards to people liable to allergies (Magan, et al., 2003) Also the degree of harm is directly associated with the infestation average which, in turn, is specified by aspects such as the eggs laying, and the survival and fertility (Nawrot, et al., 2010). Chemical pesticides are applied as cereal protectants from insect infection and offer adequate protection of cereals for sundry months (Tsaganou et al., 2021). But the use of chemical pesticides poses a large threat to the ecosystem also insects can develop resistance to pesticides at the long term, So the scientists focused on finding safer and more environmentally friendly control technologies. One of the most effective methods for managing insects is biological control, which makes use of predators, parasitoids, or diseases. Entomopathogens, sometimes known as insect pathogens, are bacteria, fungi, viruses, protozoa, or nematodes that infect and eventually kill the pest (Silva, et al., 2020). *Lecanicillium lecanii* is one of the essential entomopathogenic fungi species previously known as *Verticillium* the spores of this fungi grow, produce hyphae on pests (insects), and penetrate inside the body cavity where they rapidly occur tissue devastation. (Altinok, et al., 2019). Given the importance of the insect as one of the most destructive pests in the world, and the great economic damage it causes to grain, as well as the lack of studies on controlling this insect by managing fertility and egg production. The aim of the current research is to find out the efficacy of *L. lecanii* on the fecundity of *S. granaries*

2. Materials and Methods

2.1. Culture of *Sitophilus granaries*

Sitophilus granaries was obtained from the University of Baghdad/College of Agriculture/ plant protection / Entomology Lab, and culture was formed by putting 20 pairs of adults on wheat grains in plastic containers (15X 7). 5 grams of dried bread yeast was added and fine cloth was used to cover the culture, provisioned with a rubber band to prohibit the adults from escaping. This culture of insects was incubated at $28 \pm 2^\circ\text{C}$ and $60 \pm 2\%$ relative humidity until the experimentation time.

2.2. Fungal Isolate

Lecanicillium lecanii isolate used in this experiment was obtained from the Department of Plant Protection / College of Agriculture / University of Baghdad, Insect Diseases Laboratory. The isolate was maintained in Petri dishes containing the culture medium (SDA/sabouraud dextrose agar) at a temperature ($25 \pm 2^\circ\text{C}$) and remained for a period ranging between 7-10 days before starting the experiments. (Lord,2007) Conidia of *L. lecanii* were harvested by carried out using the technique described by (Lacey,1998) by adding 5 ml of distilled water to the Petri dishes containing the fungal culture and scraping with a Loop, the contents of the dish were filtered with

a filter paper, which was fixed on a funnel, and adding another 5 ml of distilled water to ensure complete removal of fungal spores. The sporophyte was collected in a flask, and thus the stock of the fungal suspension was obtained. Prepared three concentrations of *L. lecanii* conidial suspension (2×10^8 , 2×10^6 and 2×10^4 spores/ ml) by counting spores harvested from stock of the fungal suspension by using a hemocytometer noubarchamber and making dilution adjustments as necessary.as reported previously (Zhao et al. 2021).

2.3.Laboratory bioassays

In this experiment, collected adults 24 h old, by flowing up the pupa to grow into adults, were divided into four groups, each group has taken 20 adults (10 males,10 female), was treated by direct spraying with three concentrations (2×10^8 , 2×10^6 , 2×10^4 spores/ ml) in addition to the controlling test, the mortality rate of adults, the average egg production, the hatching rate, and the incubation period of eggs, were recorded. after exposure for matings, were carried out as follows:

A: Males treated with the fungal concentrations X females treated with the fungal concentrations

B: Males untreated with the fungal concentrations X females treated with the fungal concentrations

C: Males treated with the fungal concentrations X females untreated with the fungal concentrations

D: Males untreated with the fungal concentrations X females untreated, which are insects of the control group

3. Results and Discussion

The results in Table 1 showed the efficiency of the fungal concentrations on the adults at ages 24 h that effect, reflecting that effect in the mating capacity and the rate of egg production, and the lowest rate of productivity eggs when mated male's treatment with female treatment was 18.3 at 2×10^8 spores/ ml, also the lowest percentage of hatching eggs was 38 % at 2×10^8 spores/ ml and the highest rate of hatching period was 6.8 at 2×10^8 spores/ ml. when mated males untreated with females treated with fungal the rate of productivity of eggs was 18.8 , 24.2, and 24.8 at concentrations (2×10^8 , 2×10^6 , 2×10^4 spores/ ml) respectively while the lowest percentage of hatching eggs was 56 % at 2×10^8 spores/ ml . The highest percentage mortality of adults after mating males treated with females treated was 64% at concentration 2×10^8 spores/ ml, and, the most increased percentage mortality of adults after breeding males untreated with females treated was 60 % at concentration 2×10^8 spores/ ml while the highest percentage mortality of adults after mating males treated with females untreated was 62% at concentration 2×10^8 spores/ ml

We noted as stated in Table (1) that when females are treated and mated with treated males, it is found greatly affected the average number of eggs. This case was explained by: Bajan, et al., (1977); Gottwald and Tedders (1984) showed that treating female insects with concentrations of entomopathogenic fungi causes partial damage to the tissues of their ovaries, which leads to disturbances in the fertilization of live adults, and inhibition of the egg-laying process. Sookar, et al. (2014) evaluated efficiently the *Metarhizium anisopliae* on the fertility of fruit fly *Bactrocera*

zonata and *Bactrocera cucurbitae* the results showed that treated females laid fewer eggs than untreated flies. Similar result was recorded by Dimbi, et al. 2013 when that study the effect of *Metarhizium anisopliae* on Fertility and eggs Laying in three species of fruit fly, there was an impact of entomopathogenic fungal on female egg production in the flies as untreated flies laid more eggs than entiompathogenic fungus treated females, also percentage lowering in fecundity in fruit flies treated with *M. anisopliae*. In conclusion, this research has displayed that. The results of ZHOU, et al. 2023 In a previous study, it was reported that entomopathogenic fungi *Isaria javanica* could efficiently infect *Sogatella furcifera* and that lead to high adult mortality, as well as the high concentrations of fungal 50% decline in the total egg laying was observed in adults derived from infected nymph as compared with uninfected. Our results indicated that *L. lecanii* shows efficacy to control pests under laboratory conditions, revealing the strong potential for their applications in biological control.

Table (1)

Matings		Number of eggs	Hatching eggs %	Hatching period (days)	Mortality rates of adults %	
					Male	Female
A	2x 10 ⁸	18.3	38	6.8	64	61
	2 x 10 ⁶	21.2	46	5.2	54	56
	2x 10 ⁴	22	48	5.2	42	39
B	2x 10 ⁸	18.8	56	5.5	10	60
	2 x 10 ⁶	24.2	54	5.5	6	57
	2x 10 ⁴	24.8	54	5.2	6	41
C	2x 10 ⁸	39.4	77	4.8	62	12
	2 x 10 ⁶	42.2	74	4.6	55	8
	2x 10 ⁴	41.7	92	4.6	42	4
D	Control	77	100	4.5	0	0
LSD value		11.07 *	17.63 *	2.04 *	9.41 *	8.66 *
* (P≤0.05).						

A: Males treated X females treated

B: Males untreated X females treated

C: Males treated X females untreated

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Sarah Ibrahim Mahmood / Afr.J.Bio.Sc. 6(8) (2024)

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Sarah Ibrahim Mahmood / Afr.J.Bio.Sc. 6(8) (2024)

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