

## **IMPACT OF ENTOMOPATHOGENIC FUNGUS *Beauveria bassiana* (BALSAMO) ON ADULT OF SUGAR-BEET TORTOISE BEETLE, *Cassida vittata* (VILL.).**

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### **ABSTRACT**

Adults of *Cassida vittata* were treated in the laboratory with different concentrations of the fungal spores of *Beauveria bassiana*. The insects were dipped in the spore suspensions for five seconds. The concentrations used in the treatments were as follows,  $10^8$ ,  $5 \times 10^7$ ,  $10^7$ ,  $5 \times 10^6$  and  $10^6$  spore/ml water. The results show that the concentration of  $10^8$  caused insect mortality of 5, 10, 20, 20, 35, 50, 70, and 90 % in the period from the second to the ninth day. A complete mortality was reached in the tenth day. The concentration of  $5 \times 10^7$  caused 95 % mortality on the twelfth day. On the same day there were no differences between the two concentrations of  $10^7$  and  $5 \times 10^6$  in which the mortality was recorded as 85 %. The percent mortality was recorded as 80% when the insect were treated with the concentration of  $10^6$  spore/ml. Insect treatment with the concentration of  $10^8$  caused insect mortality of 5, 10, 20, 20, 35, 50, 70, and 90 % in the period from the second to the ninth day respectively. A complete mortality was reached in the tenth day.

According to the field application with the concentration of  $10^8$  spore/ml, the total numbers of adult at the beginning of the experiments were counted and recorded as 114 /25 and 107 / 25 plant in treated and control respectively. After two weeks it became 65/25 plant and 121 / 25 plants in treatment and control respectively. In the fourth week the number of insects was noticed to be decreased in both treatment and control and recorded 40 and 73 adult insect per 25 plants in treatment and control respectively. In the sixth week the insect population was gradually increased in both treatment and control in which the total number of adult was recorded as 69 and 198 / 25 plants in treatment and control respectively.

### **INTRODUCTION**

The entomopathogenic fungi such *Beauveria bassiana* and *Metarhizium anisopliae* are very promising in the bio-control of agriculture pests. Increasing pathogenicity of the fungal strains was studied and reported by Rabie, (1994, 2002). The effect of chemical pesticides on the efficiency of *Metarhizium anisopliae* and *Beauveria bassiana* was also studied and reported by Rabie *et al.*, (1995). The relationship between fungal pathogenicity and the production of fungal toxin in the target insects and fungal liquid cultures was also reported by Rabie, (2002).

*Beauveria bassiana* is a common soil borne fungus that occurs worldwide. It attacks a wide range of both immature and adult insects. Besides silkworm, the extensive list of hosts includes such important pests as whiteflies, aphids, grasshoppers, termites, Colorado potato beetle, Mexican

bean beetle, Japanese beetle, boll weevil, cereal leaf beetle, bark beetles, lygus bugs, chinch bugs, fire ants, European corn borer codling moth, and Douglas fir tussock moth. Natural enemies, such as lady beetles, are susceptible too, and it has even been found infecting the lungs of wild rodents.

Sugar-beet plants are liable to infestation by a variety of insects. Some of these insects are key pests of regular occurrence and cause serious damage to their host plants that leads to reduction of crop yield, in both quantity and quality.

Isolation, production and use of entomopathogenic fungi for controlling the sugar - beet insect pests in Egypt was reported by El-Husseini, *et. al.*, (2004). Utilization of biological control agents for controlling some Sugar-beet insect pests at Kafr El-Sheikh region was studied and reported by Mesbah *et. al.*, (2004). Abedel-Rahman, *et. al.*, (2004) studied also the natural occurrence of entomopathogenic fungi on cereal aphids at Assiut and they made a comparison study between field and laboratory observations.

The present work was planned to study the possibility of using the entomopathogenic fungus *Beauveria bassiana* as effective biocide against tortoise beetle, *Cassida vittata* (Vill.).

The over all purpose of this work is to through some lights on the possibility using of *Beauveria bassiana* as an effective field applicant biocide against the adult of *Cassida vittata*.

## **MATERIALS AND METHODS**

### **1- Laboratory application of the adult insects with the fungal spores**

Adults of *Cassida vittata* were gathered from the field and reared for two weeks in the laboratory. The insects were treated with the conidial spores of *Beauveria bassiana* by the concentrations of  $10^8$ ,  $5 \times 10^7$ ,  $10^7$ ,  $5 \times 10^6$  and  $10^6$  spores / ml. Each treatment contained four replicates and each replicate contained twenty insects. Five individuals each in Petri – dish, placed on a wetted filter paper. After dipping the insects in the suspension, the insects were transmitted on the parts of sugar-beet leaves to feed. Each treatment was incubated at  $24 \pm 1^\circ\text{C}$  and  $65 \pm 5\%$  R.H. and observed daily, and the control leaves were treated with distilled water.

### **2- Field Application with *Beauveria bassiana*:**

The application of *Beauveria bassiana* Blast spore in sugar-beet fields was carried out in Kafr El-Sheikh (Abu Kalab) during season 2002 – 2003. Sugar – beet plants were sprayed with the fungal suspensions to control *Cassida vittata*. Blast spore were grown on agitated bio malt liquid medium. Blast spores of *B. bassiana* were applied to sugar- beet plants by using 10 lit. Knapsac sprayer with a concentration of ( $10^8$  spores / ml.).

The treated plots were (10 x 10 m) and other untreated were used as control. Numbers of adults were counted on the treated and control plots before and after fungal treatment. Twenty five plants were investigated in

every plot. The spraying was repeated each two weeks for three times .Data were submitted to the statistical analysis using the analyses of variance.

## RESULTS AND DISCUSSION

### 1- Laboratory application of the adult insects with the fungal spores

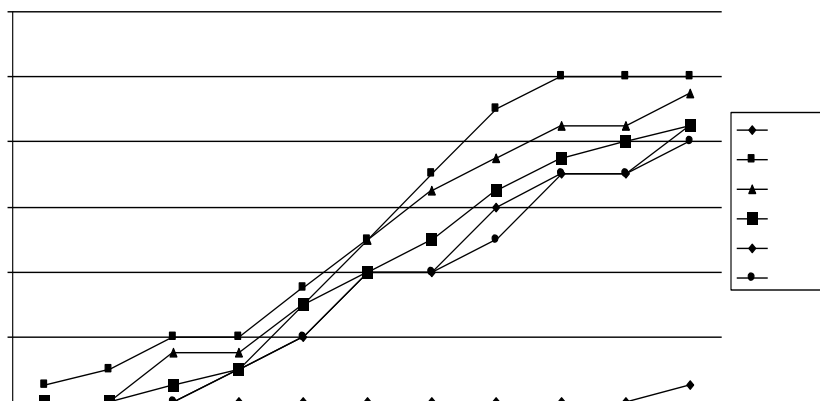
Adults of *Cassida vittata* which gathered from the field and reared for two weeks in the laboratory were treated with different concentrations of the fungal spores. The Insect deeding technique in the spore suspensions for five seconds was used to infect the insects. The concentrations used in the treatments were as follows,  $10^8$ ,  $5 \times 10^7$ ,  $10^7$ ,  $5 \times 10^6$  and  $10^6$  spore/ml water. The percent mortality caused by every concentration was recorded daily and reported in Table (1) and showed clearly in fig (1). Table (1) shows that the concentration of  $10^8$  caused insect mortality of 5, 10, 20, 20, 35, 50, 70, and 90% in the period from the second to the ninth day. A complete mortality was reached in the tenth day. The concentration of  $5 \times 10^7$  was caused 95 % mortality on the twelfth day. On the same day there were no differences between the two concentrations of  $10^7$  and  $5 \times 10^6$ . Both concentrations caused 85% insect mortality. The same table indicates that only 80 % mortality was recorded as a result of treated the insects with the concentration of  $10^6$ .

**Table (1): The percent mortality of adults of *C. vittata* as results of treatment with five of spore concentrations of *B. bassiana* at  $24 \pm 1^\circ\text{C}$  and  $65 \pm 5\%$  R.H.**

Days	Control	$10^8$	$5 \times 10^7$	$10^7$	$5 \times 10^6$	$10^6$
2	0	5	0	0	0	0
3	0	10	0	0	0	0
4	0	20	15	5	0	0
5	0	20	15	10	10	10
6	0	35	30	30	20	20
7	0	50	50	40	40	40
8	0	70	65	50	40	40
9	0	90	75	65	60	50
10	0	100	85	75	70	70
11	0	100	85	80	70	70
12	5	100	95	85	85	80

As shown also in fig (1) it could be proved that the first concentration of  $10^8$  was the most effective one among all concentrations. It was the only one which gave complete mortality in the insect population after only ten days.

Fig. ( ) % Mortality of Adult of *C. vittata* infected with *B. bassiana*  
( Laboratory treatments )



## 2 - Field Application of the adult insects with the fungal spores

The field experiments were carried out to investigate the suitability of the spores of *B. bassiana* as a biocide against the adult insects. All applications were planned to be done in the sugar-beet field in Kafr El – sheikh Governorate. March and April 2003 was the beginning of the experimental work. Spores fungal concentrations were prepared in the lab. Just before the beginning of the experiments. The suspensions were sprayed every two weeks for three times. As mentioned above, number of insects was counted before and after the applications in both treatments and control applications.

According to the laboratory experiments, the concentration of  $10^8$  was the most effective one among all the others. Therefore the field application was carried out using this concentration. Percent mortality was recorded every two weeks and reported in table (2) and illustrated in fig (2). The data in Table (2) indicates that at the beginning of experiments (zero time), the total number of adult in was 114 /25 and 107 per 25 plant in treated and control respectively. After two weeks from application with *B. bassiana* the total number of adult was recorded to be 65 per and 121 per 25 plants in treatment and control respectively.

In the fourth week which seems to have a difficult weather, number of insect was noticed to be decreased in both treatment and control. Table (2) recorded 40 and 73 adult insect per 25 plants in treatment and control respectively.

In the sixth week in which the weather seems to be better, the insect population was gradually increased in both treatment and control. As recorded in table (2), the total number of adult was 69 and 198 per 25 plants in treatment and control respectively.

Table (2): Field application of  $10^8$  concentration of *Beauveria bassiana* against adult of *Cassida vittata*.

After weeks	Treatment	Control
0	114	107
2	65	121
4	40	73
6	69	198
Total	288	499
Mean	72	124.75
F- Value	*	*

### CONCLUSION

The present work is an attempt to through heavy lights on the possibility of using the entomopathogenic fungi as very effective alternatives of chemical insecticides. It is known that, the preparation of effective biocide such as *Beauveria bassiana* enough to control the pests in one feddan is very cheaper than chemical insecticides. It is aimed in this work to prove also that the fungal biocides are not only cheaper but also very effective against insect pests. Laboratory treatments and field application using the entomopathogenic fungi proved the high affectivity of the spore fungal preparations against the agriculture pests either in lab or in field. The concentration of  $10^8$  spore/ml water caused complete mortality in insect in the lab after only 10 days. The same concentration decreased the insect population in the field from 114 to 69 insect per 25 plants in six weeks. Therefore it could be proved that *Beauveria bassiana* could be used practically in the field. As given by Rabie (1994) and Rabie *et.al.*, (1995) fungal pathogenicity could be gradually improved in the lab by the continuous infection and fungal reisolation through the target insect. Thus a modification in fungal pathogenicity to increase fungal virulence could be very benefit for introducing promising methods for agricultural pest control.

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**تأثير الفطر الممرض ( *Beauveria bassiana* ) على الطور الكامل لحشرة خنفساء بنجر السكر السلحفائية *Cassida vittata* Vill. على بنجر السكر في مصر .**

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تم معاملة الحشرات الكاملة لخنفساء بنجر السكر السلحفائية في المعمل بتركيزات مختلفة من جراثيم فطر *Beauveria bassiana* . تم وضع الحشرات في محلول جراثيم الفطر لمدة ٥ ثواني . التركيزات التي استخدمت كانت ١٠<sup>١</sup> و ١٠ x ٥<sup>٢</sup> و ١٠<sup>٣</sup> و ١٠<sup>٤</sup> و ١٠<sup>٥</sup> و ١٠<sup>٦</sup> جرثومة / مل ماء . ومن النتائج اتضح أن التركيز ١٠<sup>٤</sup> سبب نسبة موت ٥ و ١٠ و ٢٠ و ٢٠ و ٣٥ و ٥٠ و ٧٠ و ٩٠ % في الفترة من اليوم الثاني إلى اليوم التاسع . وقد تم موت جميع الحشرات في اليوم العاشر . التركيز ١٠ x ٥<sup>٢</sup> تسبب في موت ٩٥ % في اليوم الثاني عشر . وفي نفس اليوم لا يوجد اختلاف بين التركيزان ١٠<sup>٦</sup> و ١٠ x ٥<sup>٢</sup> حيث وصلت نسبة الموت إلي ٨٥ % . ونسبة الموت وصلت إلي ٨٠ % عندما عوملت الحشرات بتركيز ١٠<sup>٦</sup> . وقد تم التطبيق الحقل باستخدام التركيز ١٠<sup>٤</sup> وتم عد الحشرات عند بداية التجربة وكانت ١١٤ حشرة / ٢٥ نبات و ١٠٧ حشرة / ٢٥ نبات في المعاملة والكنترول على الترتيب . بعد أسبوعين أصبحت ٦٥ حشرة / ٢٥ نبات و ١٢١ حشرة / ٢٥ نبات في المعاملة والكنترول على الترتيب . في الأسبوع الرابع لوحظ انخفاض في تعداد الحشرات في كل من المعاملة والكنترول وسجلت ٤٠ و ٧٣ حشرة / ٢٥ نبات في المعاملة والكنترول على الترتيب . في الأسبوع السادس ازداد تعداد الحشرات في كلا من المعاملة والكنترول وكان التعداد الكلي للحشرات ٦٩ و ١٩٨ حشرة / ٢٥ نبات في المعاملة والكنترول على الترتيب .