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Effects of entomopathogenic fungi *Metarhizium anisopliae* (Met.) and *Verticillium lecanii* (Zimm.) on the predation efficiency of *Chrysoperla carnea* (Stephens)

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Abstract:

This research was conducted to evaluate the effects of entomopathogenic fungi M. anisopliae and V. lecanii at various concentrations (1 x 10^5, 1×10^7, 1× 10^9 spore/ml) on the predation efficiency of Chrysoperla carnea (Stephens) on the Ephestia cautella eggs under laboratory conditions (25 ± 2°C and 60 - 70 % RH and photoperiod (16 L : 8 D h). The results had been revealed that the treated of larval stages with the tested fungi affect the predator capacity in general and the largest decline predator capacity has been recorded when the first and second larval instar treated with the concentration ***10^9*** *spore/ml of V.lecanii which were 16, 35.3 egg respectively , while the predator capacity were* ***27.7*** *,* ***65*** *egg respectively when treated with M.anisopliae at the same concentration and the predator capacity of third larval stage were 596, 88****5.3*** *egg when treated with M.anisopliaea and V.lecanii at the same concentration respectively compare with the control treatment 1154.3 egg.*

Key words: *C. carnea, V. lecanii, M. anisopliae*, *E. cautella*, Predation efficiency.

INTRODUCTION

Biological control or the use of parasitoids, predatory or beneficial bacteria and fungi (Natural Enemies) is an

alternative pest management strategy for dealing with arthropods (Van Driesche and Heinz 2004).

The green lacewings, *C. carnea* (Stephens) (Neuroptera: Chrysopidae) is a worldwide polyphagous predator, generally found in a wide range of agricultural system (McEwen *et al.*2001).

The effective predator, is a ravenous feeder of aphids, and white flies and gaining importance in (IPM) (Sattar and Abroo, 2011; Hashami, 2001). It can be reared by the laboratory (Hamed *et al.* , 2009 ; Nadeem, 2010).

It is normally present in larval and adult forms on vegetables, leaves of crops, ,fruit plants in high humidity areas like in greenhouses and irrigated crops adults of *C.carnea* feed plant nectars (Atlihan *et al*., 2004).

Larvae of *C. carnea* are voracious predator of thrips, mites., aphids, and eggs of lepidopterous (Soft bodied arthropods) insects larvae of *C. carnea* are voracious and feed on insect pests (Carrillo *et al*., 2004).

Entomo. pathogenic fungi are known to infect a broad range of insects including Lepidoptera, Homoptera, Hymenoptera, Coleoptera, Diptera, Hemiptera, Orthoptera and Thysanoptera (Lecuona *et al.*, 2001; Lezama-Gutierrez *et al.*,2001).

The fungi *M. anisopliae* (Met.) is natural enemies of a broad range of insects and arachnids and both fungi have a worldwide distribution (Rehner, 2005).

The fungus *V. lecanii* (Deuteromycetes) can be used for crop protection for the biocontrol of insects of agricultural importance (Thiery, 2015).

MATERIALS AND METHODS

Insect and Entomopathogenic Fungi Source

Laboratory experiments were carried out in the insectary belonging Entomology, University of Wasit, College of science, Department of Biology. constant temperature of 25 ± 2° C and relative humidity of 60 -70 % and (16 L : 8 D h). The predators *C. carnea* (Steph.) , prey *E. cautella* (Walk.) eggs were obtained from a maintained culture in the Insectary, and entomopathogenic fungi of *V. lecanii* (V5) and *M. anisopliae* (M87) were obtained from Laboratories of Agricultural research directorate , integrated pest control center, Zuafraniya - Baghdad- Iraq.

Preparation for all experiments

Insect Colony Rearing

C. carnea (Steph.) adults were reared in a rectangular cage, made of 6 cm thick, transparent plastic sheet. The cage is(35 h, 35 l, and 20 w) cm. Two circular windows, each of 13 cm diameters, enclosed with lids of the same material, situated diagonally near opposite corners of a front wall of the cage,are made for handling adults, and provision of water in petri dish etc. Synthetic foods include honey , yeast, sugar and water were provided in petri dish. A sieve of circular holes 2 mm diameter is drilled into the sidewalls to ventilation in the cage, for better survival of adults (Ahmad-Ur-Rahman *et al.*, 2015).

Ephestia cautella

The colony of *Ephestia cautella* (Lepidoptera : Pyralidae) was established using growth chamber set up at 25 ± 2°C, 60 -70 % RH and a photoperiod L16:D 8 h . Artificial diet as described earlier was used for *E. cautella* continuance (Hameed 2002). From side to side this method enough numbers of eggs were

obtained to rear *C. carnea* continuously under laboratory condition and to carry predator capacity of larval instar (1st instar , 2nd instar and3rd instar) test.

Entomopathogenic Fungi

Maintenance of culture and Preparation of fungus inoculums

A metal loop of inocula from subculture plates of isolates were shifted to PDA slants and sustained as pure cultures. For laboratory studies, the fungus was grown on PDA medium. After complete sporulation, conidia from the medium were harvested by washing them carefully with sterilized water containing Tween-80 (0.2%) for direct use. Spores were also harvested with the sterile Lab spoon. Harvested conidia was dried in the air under laminar flow and stored in the refrigerator at 25 C." Suspension of spores was made using (D.W) with Tween-80 (0.2%) and filtered through a double layered muslin cloth to obtain a spore suspension". Three concentrations of all fungal isolates 1 x 10^5, 1 x 10^7 and 1x 10^9 were determined by the aid of hemocytometer. The viability of spore was determined as in Masoud and Bahar (2012).

Bioassay

Effect of entomopathogenic fungi on the predator capacity of Predator larval stage.

Newly hatched predator larva (1st instar, 2nd instar and 3rd instar) treatment direct exposure these larvae to fungal suspensions of *V. lecanii* and *M. anisopliae* by 2 ml of concentration of (1 x 10^5, 1 x 10^7 and 1x 10^9 spore / ml) each put single in a petri dish (10 cm) with filter paper on its bottom was prepared and repeated five times for each prey. Known surplus numbers from each prey (*Ephestia cautella*) eggs was

offered and the devoured were replaced daily each repeater in the comparison group were treated by spraying 2 ml of ((D.W) . Attacked prey individuals were counted daily during lavae instar of predator. The duration period, feeding capacity of larval stage (1st instar, 2nd instar and 3rd instar) were recorded (Mesbah, 2001).

Statistical analysis

Statistical analysis was carried out using "completely randomized design(CRD) and Duncan's multiple range test (DMRT) " .

RESULTS AND DISCUSSION

Data in table (1) show increase predator capacity daily larval stages consistent with the requirements of growth and development of *C. carnea* and the effects of two entomopathogenic fungi, *M. anisopliae* (Met.) and *V. lecanii* (Zimm.) were conducted to determine the predator capacity of *C. carnea* larval instar treated with different concentrations of fungal isolates.

The results had been revealed that that treated of larval stages with the tested fungi affect the predator capacity in general and the largest decline predator capacity has been recorded when the first and second larval instar treated with the concentration $\mathbf{10^9}$ spore/ml of *V. lecanii* which were 16, 35.3 egg respectively , while the predator capacity were **27.7** , **65** egg respectively when treated with *M.anisopliae* at the same concentration and the predator capacity of third larval stage were 596, 88**5.3** egg when treated with *M.anisopliae* a*nd V. lecanii* at the same concentration respectively as compare with the control treatment 1154.3 egg .

The life predator research effectiveness and efficiency predatory which are effective influenced by environmental

factors which include the virulence of Entomopathogenic fungi. The predator *C. carnea* was decrease in food consumption of insect one of the possible effects of the doses under deadly caused by entomopathogenic fungi (Hajek and Goettel, 2000).

Table (1): The effect of different concentrations of entomopathogenic Fungi *lecanii Verticillium* and *Metarhizium anisopliae* in predator capacity of the larval stage of the predator *Chrysoperla carnea* feeding on *Ephestia*

Treatment	Con.	Mean consumption		
		1st instar	2nd instar	3rd instar
Control	0	38.0a	84.3a	1154.3a
V. lecanii	10^5	24.7b	75.7b	975.3b
	10^7	19.3c	45.7c	817c
	10^9	16c	35.3d	769.3d
Control	0	38.0a	84.3a	1154.3a
M. anisopliae	10^5	34.7ab	ab81	782.3b
	10^7	29.7b	b78	672c
	10^9	.7b27	65c	576d

Different letters for columns indicate to the significant difference between each fungal isolate concentrations and control.(L.S.D = 0.05)

On the other hand, Similar results have been reported by Roditakis *et al.,*(2008) who refer that the predator consumption decrease to those who *Myzus persicae* and are significantly when the fungus infection as *longisporum Lecanicillium*.These results were in confirmation with the findings of Alma(2005) decrease who in predator capacity of predator *Dicyphus hesperus* infected when the fungus *Paecilomyces fumosoroseus*. Many of the predator species avoid by feeding on infected prey fungal and moved away from predation, recorded avoid predation white flies infected with fungi from predator *D.hesperus* (Labbe et al ,. 2006).The obtained results are in accordance with that (Meyling and Pell, 2006) who proof that predator *Anthocoris nemorum was* not feed to prey (aphids) infected with entomopathognic fungi

B.bassiana. A positive correlation has been found between germination speed and the virulence of some *L. lecanii* isolates (Diaz *et al*. 2009). In another study about the three tested treatments of the *V. lecanii* (Zimm.) at the concentrations 10^8 spores/ml., the predator *C. carnea* (Steph.) and *V. lecanii* followed by release of *C. carnea* second instar in faba bean field for controlling cowpea aphid, *Aphis craccivora* (Koch), These treatments indicated the potential use of these treatments to control *A. craccivora* on faba bean. Reduction in *A. craccivora* population and subsequent yields were significant between treatments, highest reduction and yield gain was observed when fungus *V.lecanii* was applied (Abd El-Gawad and Atef,2008). In other study of laboratory indicated (Leyva *et al*. 2011) is not affected by any age predator Chrysopa exterior when exposed to the fungal pathogen *Beauveria bassiana (*Bals.*)* and *Vuillemin Strain LBB-1,* which promotes the use of workers in the application of (IPM) of insect pests.

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