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Susceptibility of *Schistocerca gregaria* (Forskål) and *Euprepocnemis plorans* (Charpentier) to *Metarhizium anisopliae* var. *acridum* (Metchnikoff) Soroken, *Beauveria bassiana* (Bals.) Vuill. and *Nosema locustae* Canning

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ABSTRACT

Effects of the fungi; *Metarhizium anisopliae* var. *acridum* (Metchnikoff) Soroken (=*Metarhizium flavoviride*) *Beauveria bassiana* (Bals.) Vuill. and the microsporidian *Nosema locustae* Canning on the acridiids *Schistocerca gregaria* (Forskål) and *Euprepocnemis plorans* (Charpentier) were studied in the laboratory. The three pathogens were tested at doses of 10^3, 10^4, 10^5 and 10^6 spores/nymph. Mortalities were subjected to probit analysis to study the dose and time mortality response. The results showed that *M. anisopliae* var. *acridum* was the most virulent pathogen to the 3rd nymphal instar of *S. gregaria* and *E. plorans* followed by *B. bassiana*, then *N. locustae*, where the LD_{50}s for desert locust at the 7th day were $3.5X10^7$, $7.4X10^8$ and $3.0X10^{10}$ spores/nymph, on the 14th day were $4.5X10^4$, $7.0X10^5$ and $1.2X10^8$ spores/nymph and on the 21st day were $3.6X10^3$, $3.5X10^4$ and $1.1X10^5$ spores/nymph, after infection with *M. anisopliae*, *B. bassiana* and *N. locustae*, respectively. In case of *E. plorans*, the LD_{50}s on the 7th day were $3.5X10^7$, $1.6X10^8$ and $3.7X10^9$ spores/nymph, on the 14th day were $2.3X10^4$, $7.0X10^5$ and $3.3X10^6$ spores/nymph on the day 21st were $1.3X10^3$, $8.7X10^4$ and $1.3X10^5$ spores/nymph, after infection with the same respective pathogens. Concerning the speed of kill, *M. anisopliae* was the fastest pathogen in its action followed by *B. bassiana*, then *N. locustae* at LT_{50} values bases. The LT_{50} values of *M. anisopliae*, *B. bassiana* and *N. locustae* when applied to *S. gregaria* at the dose 10^3 were 24.66, 36.39 and 51.26 days, at the dose 10^4 were 16.37, 21.84 and 27.05 days, at the dose 10^5 were 11.76, 17.85 and 20.13 days and at the dose 10^6 were 10.76, 13.59 and 17.40 days, respectively. When applied to *E. plorans*, the LT_{50} values of *M. anisopliae*, *B. bassiana* and *N. locustae* at the dose 10^3 were 19.72, 36.61 and 40.47 days, at the dose 10^4 were 14.90, 23.24 and 23.54 days, at the dose 10^5 were 11.76, 17.85, and 20.12 days, and at the dose 10^6 were 10.43, 13.47 and 16.08 days, respectively. The fungi *M. anisopliae* var. *acridum* and *B. bassiana* showed significant prolongation to infected 3rd nymphal insatr of both insect species. The entomopathogens lead to significant prolongation in duration of infected 4th and 5th nymphal instars. *M. anisopliae* var. *acridum* caused the longest prolongation to all nymphal instar durations followed by *B. bassiana* then *N. locustae*. The pathogens also caused significant reduction in number of egg pods and number of eggs per pods, in addition they caused significant prolongation in the prepovipositional period and the period between each egg pod. They also induced significant reduction in the adult longevity. The most effective pathogen on the fecundity of *S. gregaria* and *E. plorans* was *M. anisopliae* var. *acridum* followed by *B. bassiana* then *N. locustae*.

Key words: Entomopathogenic fungi, protozoa, desert locust, grasshopper, bioassay, mortality, duration, fecundity.

INTRODUCTION

Locusts and grasshoppers are major economic pests of crops and grasslands throughout the world's dry zones, their attacks attract much public attention, few other pests make headline news (Lomer *et al.*, 1999). Desert locust *Schistocerca gregaria* consumes approximately their own weight (ca. 2g) of fresh vegetation each day. Swarms often contain 50 million individuals per Km^2, so that even a moderate swarm measuring 10 Km^2 could consume about 1000 tons of fresh vegetation daily during migration (Coper, 1982). Since long time, grasshopper *Euprepocnemis plorans* (Charp.) is considered the most economic grasshopper in Egypt, causing a serious damage to cultivated crops specially in the newly reclaimed lands (Nakhla, 1957). Recently, *E. plorans* caused serious damage to maize cultivation in Sharkia and Dakahlia Governorates (unpublished data, El-Maghraby 2003). Millions of ha. of land in many countries received aerial or ground insecticide treatments to control locusts and grasshoppers, using millions of liters of insecticides, the large scale repeated application of insecticides, raised concern about the possible impact on the environment as well as on human health (Anonymous , 1990 and 2006). All fungi isolates whish are highly virulent to *S. gregaria* belonged to genus *Metarhizium*. The isolate that was adopted as a standard and used in all assays for comparison was *M. anisopliae* var. *acridum* IMI330189 which originated from the grasshoppers *Ornithacris cavroisi* (Finot) collected in Niger. No highly virulent isolates to locust have yet been found originating in non orthopteran hosts (Prior, 1992). About 58 species of Orthoptera were known to be susceptible to infection by *N. locustae*, while there is no species out order Orthoptera susceptible to *N. locustae* (Henry, 1969). Also, most economic grasshopper species in Egypt are susceptible to *N. locustae* infection (Abdelatef, 1998).

The aim of the present study is to evaluate the susceptibility of *S. gregaria* and *E. plorans* to the three entomopathogens: *Metarhizium anisopliae* var. *acridum*, *Beauveria bassiana* and *Nosema locustae*.

MATERIALS AND METHODS

Test insects

Insects used were the 3rd nymphal instar of the locust, *S. gregaria* and the grasshopper, *E. plorans*. The insects were obtained from stock cultures maintained for several generations at the Locust and Grasshopper Research Department, Plant Protection Research Institute, Agricultural Research Center (A.R.C.), Dokki, Giza, Egypt. The cultures are usually fortified with some fresh insects brought from the field every year.

Metarhizium anisopliae var. *acridum* IMI 330189

Spores of *M. anisopliae* var. *acridum* used were from isolate IMI330189, kindly provided by (Biological Control Products), South Africa. The spores were first used to contaminate desert locust, *S. gregaria* and the grasshopper, *E. plorans* nymphs, then the nymphs were kept under 31 °C and observed for mortality. Cadavers were removed and sterilized according to Lacey and Brooks (1997) and kept in sterilized Petri dishes to dry for 24 h. Then sterilized moistened pieces of cotton were placed in the Petri dishes. Every cadaver was kept alone in a Petri dish, incubated at 27 °C and observed for sporulation. Spores grown from 10 cadavers were suspended in 1ml. of sterile sunflower oil then 100 micro liter were used to contaminate 1 Petri dish (ten Petri dishes were used), each dish contain: 0.36 g. KH_2PO_4, 1.42 g. Na_2HPO_4 $12H_2O$, 0.62 g. $MgSO_4$ $7H_2O$, 1 g. KCL, 0.70 g. NH_4NO_3, 10 g. Maltose, 5 g. yeast extract, 18 g. agar-agar and 0.5 g. chloramphenicol per 1000 ml of distilled water. Then *M. anisopliae* var. *acridum* spores were collected by using small brush.

Beauveria bassiana

The spores of *B. bassiana* used were originally isolated from mycosed red palm weevil adult. The spores were used to contaminate nymphs of the desert locust, *S. gregaria* and the grasshopper, *E. plorans*. The infected nymphs were treated as previously described in case of *M. anisopliae*.

Nosema locustae

The spores of *N. locustae* used were originally obtained from Rangeland Insect Laboratory, Montana State University, A.R.S, USDA, U.S.A. in 1990. Spores were in many types of formulations: contaminated wheat bran, aqueous solution of spores and polyethylene glycol solution of the spores, dried spores and infected grasshopper cadavers. All formulations were used for the mass rearing by infecting the grasshopper, *E. plorans*. Then after 2 generations of *E. plorans* the harvested spores were used in this study.

Inoculation methods

The three pathogens, *M. anisopliae* var. *acridum*, *B. bassiana* and *N. locustae* were used at doses of 10^3, 10^4, 10^5 and 10^6 spores/nymph to contaminate *S. gregaria* and *E. plorans* nymphs of 3rd instar. Sixty nymphs were used for each treatment, divided into 4 groups each of 15 nymphs placed in a cylinder plastic cage covered with a piece of white light cloth. The appropriate doses of both fungi were suspended in sunflower oil and the appropriate volumes were placed using micro pipette under the pronotum of the insects according to Prior *et al.* (1995). The spores of *N. locustae* were suspended in water then placed into 7mm diameter lettuce disc, then allowed to dry for two hrs at room temperature, and then introduced to the nymphs individually. Nymphs which did not consume the entire lettuce disc were discarded (Henry and Oma, 1974). Each treatment received 60 nymphs divided into 4 groups; each of 15 nymphs placed in cylinder plastic cage. The infected and non infected nymphs were daily cleaned, fed and observed for mortality. The mortality data were subjected to probit analyses according to Finney (1971) to calculate dose mortality responses and its regression lines; also time mortality responses and its regression lines were calculated using Ldp line software (http://www. ehabsoft.com/ldpline/).

Effect of the entomopathogens on the development and fecundity

To study the effect of the entomopathogens on the development and fecundity, newly molted 3rd instar nymphs (100 nymphs/treatment) of *S. gregaria* and *E. plorans* were infected with dose 10^3 spores/nymph. Twenty infected nymphs with each pathogen and control were individually kept in white plastic cups, the nymphs were daily fed and observed for molting till reaching adult stage and duration of every nymphal stadium was recorded. When 5th nymphal instar molted to adult stage, 20 males and females paired of each treatment, then each couple was kept in a glass cylinder. The glass cylinders were placed in a box, which had appropriate holes leading to plastic cups full of sterilized sand for the oviposition. These cups were screened carefully to detect any new egg pods. Fresh food, especially Egyptian clover was added daily.

The pre-ovipositional period was recorded for each female. When any egg pod was found, it was isolated and recorded. The number of eggs/pod was counted in randomly selected 10 pods of each treatment. Days between egg pods/female were also recorded as well as longevity of the adult. All infected and non infected insects were kept in an incubator at 31 ± 0.5 °C, 65±5% R.H. and 12:12 hrs light: dark photo period.

RESULTS AND DISCUSSION

Dose mortality response

Table (1) shows calculated lethal doses of the tested pathogens to kill 25, 50 and 90 % of *S. gregaria* and *E. plorans* individuals at the mentioned days and slopes of dose-mortality regression lines. The results revealed that *Metarhizium* is the most virulent pathogen to *S. gregaria* and *E. plorans* followed by *Beauveria* then *Nosema*, where LD_{50}s of *Metarhizium* at all the experimental durations were lower than those of *Beauveria* and *Nosema*, while LD_{50}s of *Nosema* were the highest at all experimental periods. The LD_{25}s show the same trend as LD_{50}s, while LD_{90}s show the same trend except at day 21, in case of *S. gregaria* where LD_{90}s of *Nosema* were lower than those of *Beauveria*, the same in case of *E. plorans* in day 14.

Regarding the differences among LD_{50}s of the tested pathogens, on the 7[th] day after infection, in case of *S. gregaria*, there was no significant difference between *Metarhizium* and *Beauveria* while upper and lower limits (fiducial probability) for *Nosema* couldn't be calculated due to low mortality observed (8.33, 10, 15and 20 for doses 10^3, 10^4,10^5 and 10^6 spores/nymph, respectively). In case of *E. plorans,* there was no significant difference between *Metarhizium* and *Nosema*, upper and lower limits for *Beauveria* couldn't be calculated due to that doses 10^5 and 10^6 caused the same mortality percentage (23.33 %). On the 14[th] day, there was no significant difference between *Metarhizium* and *Beauveria*, while upper and lower limits for Nosema in case of *S. gregaria* couldn't be calculated due to heterogenic response of locust individuals to *Nosema* (slope = 0.168). In case of *E. plorans,* there was significant difference between *Nosema* and both *Metarhizium* and *Beauveria*. On the 21[st] day, there was significant difference between *Metarhizium* and both *Beauveria* and *Nosema*, while there was no significant difference between the last two pathogens.

Time Mortality responses

Data presented in table (2), illustrate LT_{25}, LT_{50}, LT_{90} and slope of time mortality responses and their regression lines after infection of *S. gregaria* and grasshopper *E. plorans* with the tested entemopathogens at the doses of 10^3, 10^4, 10^5 and 10^6 spores/nymph. The fungus, *Metarhizium* was significantly faster in action against *S. gregaria* and *E. plorans* individuals where the corresponding LT_{50} values were significantly lower than the other two pathogens at all tested doses, as well LT_{25}, LT_{90}, values were lower than those of *Beauveria* and Nosema. Mean while *Beauveria* was faster than *Nosema*, there was no significant difference between *Beauveria* and *Nosema* in their speed of kill at all used doses, except in case of dose 10^6 spores/nymph. Also, LT_{25}, LT_{90}, values for *Beauveria* were lower than those of *Nosema* except in case of LT_{25} and LT_{90} of *Nosema* at dose 10^5 spores/nymph against *S. gregaria* and in case of LT_{90} against *E. plorans*.

Bidochka & Khachatourians (1990) identified the *B. bassiana* extracellular protease as a virulence factor in pathogenicity toward the migratory grasshopper, *Melanoplus sanguinipes*. So, the virulence of the three tested pathogens against the two acridiied species could be arranged ascendingly as follow:*M. anisopliae* var. acridium > *B. bassiana* > *N. locustae*. In the present study *Nosema* was the

Table (1): Virulence of *M. anisopliae* var. *acridum*, *B. bassiana* and *N. locustae* against *Schistocerca gregaria* and *Euprepocnemis plorans* after 7, 14 and 21 days post treatment.

Days after treatment	Pathogens	*Schistocerca gregaria*				*Euprepocnemis plorans*			
		LD_{50}[ab]	LD_{25}[a]	LD_{90}[a]	Slope	LD_{50}[ab]	LD_{25}[a]	LD_{90}[a]	Slope
7	*M. anisopliae*	$3.5X10^7$a	$4.5X10^4$	$1.1X10^{13}$	0.233	$3.5X10^7$a	$4.5X10^4$	$1.1X10^{13}$	0.233
	B. bassiana	$7.4X10^8$a	$2.1X10^5$	$4.2X10^{15}$	0.190	$1.6X10^8$*	$2.5X10^5$	$3.5X10^{13}$	0.240
	N. locustae	$3.0X10^{10}$*	$8.4X10^6$	$1.7X10^{17}$	0.190	$3.7X10^9$a	$1.4X10^7$	$1.4X10^{14}$	0.281
14	*M. anisopliae*	$4.5X10^4$ a	55.73	$1.5X10^{10}$	0.232	$2.3X10^4$b	1.926	$1.3X10^{12}$	0.165
	B. bassiana	$7.0X10^5$ a	531.58	$5.8X10^{11}$	0.216	$7X10^5$ ab	$4.4X10^2$	$8.2X10^{11}$	0.211
	N. locustae	$1.2X10^8$*	$1.1X10^4$	$5.1X10^{15}$	0.168	$3.3X10^6$ a	$1.2X10^4$	$1.3X10^{11}$	0.279
21	*M. anisopliae*	$3.6X10^3$b	27.76	$3.6X10^7$	0.320	$1.3X10^3$b	12.29	$9.4X10^6$	0.332
	B. bassiana	$3.5X10^4$a	164.99	$9.5X10^8$	0.289	$8.7X10^4$ a	645.8	$9.8X10^8$	0.316
	N. locustae	$1.1X10^5$ a	$1.3X10^3$	$5.7X10^8$	0.347	$1.3X10^5$a	$1.0X10^3$	$1.3X10^9$	0.320

[a]Spores/nymph. [b]Values with same letter did not differ significantly. *Significance could not be calculated.

Table (2): Time mortality response of *Schistocerca gregaria* and *Euprepocnemis plorans* to *M. anisopliae* var. *acridum*, *B. bassiana* and *N. locustae* after treatment with 10^3, 10^4, 10^5 and 10^6 spores/nymph.

Dose Spores/nymph	Pathogens	*Schistocerca gregaria*				*Euprepocnemis plorans*			
		LT_{50}^{ab}	LT_{25}^{a}	LT_{90}^{a}	Slope	LT_{50}^{ab}	LT_{25}^{a}	LT_{90}^{a}	Slope
10^3	*M. anisopliae*	24.66 b	11.34	107.97	1.99	19.72 b	9.97	72.12	2.28
	B. bassiana	36.39 a	14.11	220.36	1.64	36.61 a	13.48	232.22	1.60
	N. locustae	51.26 a	20.73	286.39	1.72	40.47 a	19.68	159.22	2.15
10^4	*M. anisopliae*	16.37 b	9.06	50.42	2.62	14.90 b	8.76	40.87	2.92
	B. bassiana	21.84 a	10.88	82.03	2.23	23.24 a	10.98	96.62	2.07
	N. locustae	27.05 b	14.04	94.03	2.37	23.54 a	14.02	63.02	3.00
10^5	*M. anisopliae*	11.76 b	6.81	33.19	2.84	11.76 b	6.81	33.19	2.84
	B. bassiana	17.85 a	8.35	75.54	2.05	17.85 a	8.35	75.54	2.04
	N. locustae	20.13 a	7.21	56.20	2.87	20.12 a	11.72	56.20	2.87
10^6	*M. anisopliae*	10.76 c	6.18	30.89	2.80	10.43 c	6.14	28.53	2.93
	B. bassiana	13.59 b	7.27	44.57	2.48	13.47 b	7.29	43.16	2.53
	N. locustae	17.40 a	9.46	55.44	2.55	16.08 a	10.11	38.84	3.35

[a]values in days. [b]Values with same letter did not differ significantly. *Significance could not be calculated.

Table (3): Effect of *Metarhizium anisopliae* var. *acridum*, *Beauveria bassiana* and *Nosema locustae* on durations of the 3rd, 4th and 5th nymphal instars of *Schistocerca gregaria* and *Euprepocnemis plorans*.

Treatment	*Schistocerca gregaria*[a]				*Euprepocnemis plorans*[a]			
	3rd	4th	5th	Total	3rd	4th	5th	Total
Metarhizium anisopliae	6.22a	9.13a	9.50a	24.85a	6.00a	7.27a	9.38a	22.65a
Beauveria bassiana	6.17a	9.07a	9.42a	24.66a	5.94a	7.20a	9.23a	22.37a
Nosema locustae	5.78ab	8.93a	9.25a	23.96a	5.52ab	7.06a	8.92a	21.50a
Untreated control	5.55b	6.71b	6.75b	19.01b	5.21b	5.71b	6.63b	17.55b

[a]Means in same columns with same small letters didn't differ significantly.

slowest killing pathogen to *S. gregaria* and *E. plorans*, many other investigators reported that *N. locustae* is slow acting pathogen so it's suitable to be used as long term control agent (Tanada & kaya 1993 and Olson *et al.,* 2002). Fungal species have numerous strains that differ in their virulence and pathogenicity Tanada & Kaya (1993). The pathogenicity of fungus may be associated with the production of enzymes or and mycotoxins during the course of infection in an insect (McCoy *et al.,* 1988).

The present data confirm those of Sieglaff *et al.,* (1997), they found that *M. flavoviride* was much more virulent than *B. bassiana* to *Schistocerca americana* and *Melanoplus sanguinipes*.

Effect on the development and fecundity

Data in table (3) demonstrate the effect of the tested pathogens on the duration of 3rd, 4th and 5th nymphal instars of *S. gregaria* and *E. plorans*. The infection with *Metarhizium* and *Beauveria* caused significant prolongation to 3rd nymphal instar of both insects compared with untreated nymphs, while prolongation of infected nymphs with *Nosema* did not differ significantly from untreated ones, also did not differ significantly from those infected with the two fungi. It was obvious that the tested pathogens also caused significant prolongation to the 4th and 5th nymphal instars of *S. gregaria* and *E. plorans* when compared with untreated nymphs.

Figure (1) shows percentages of reduction in number of egg pods/female and eggs/pod of *S. gregaria* and *E. plorans* after infection with each of the three pathogens. It was clear that the infection caused significant reduction in number of egg pods/female and eggs/pod. The highest reduction was induced by *Metarhizium*, followed by *Beauveria* and *Nosema*.

Figure (2) shows percentages of prolongation of the pre-oviposition period and period between egg pods of *S. gregaria* and *E. plorans*, after infection with the entomopathogens. The infections caused significant prolongation in the pre-ovipositional period and the period between egg pods of infected insects, except the pre-ovipositional period of infected *E. plorans* insects with *Nosema*. Highest prolongation was achieved in case of *Metarhizium* infection, followed by *Beauveria* then *Nosema*.

Figure (3) shows percentages of reduction in the adult longevities of *S. gregaria* and *E. plorans*, after infection. In case of *S. gregaria*, *Metarhizium* infection caused the highest reduction in the adult longevity of males and females as well as males plus

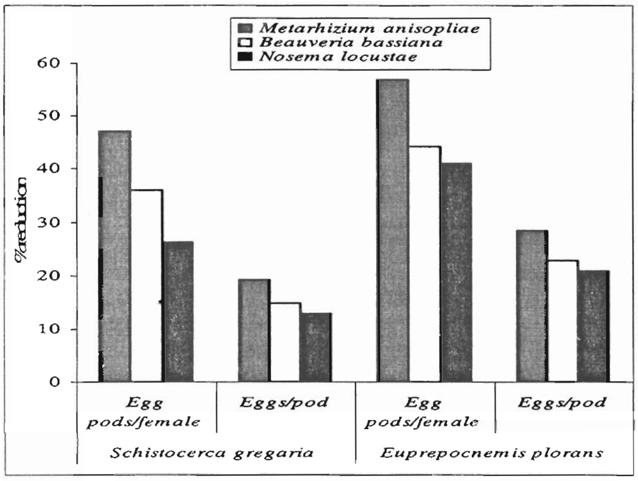

Fig. (1): Effect of infection of the three entomopathogens on *Schistocerca gregaria* and *Euprepocnemis plorans* fecundity.

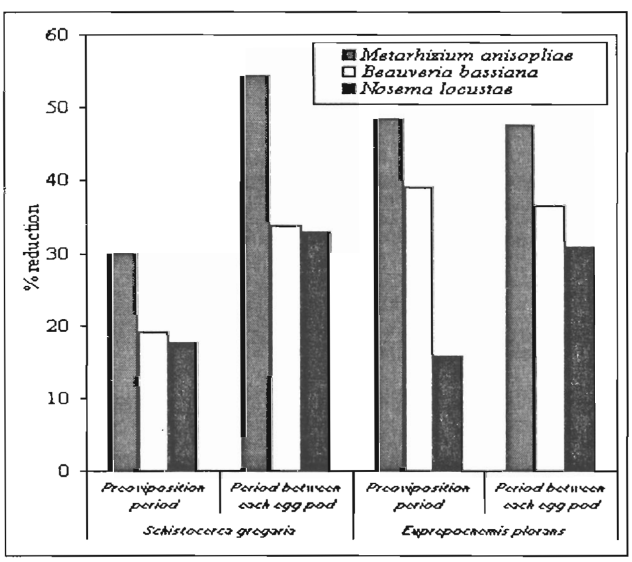

Fig. (2): Effect of infection of the three entomopathogens on the pre-ovipositional period and period between each egg pod of *Schistocerca gregaria* and *Euprepocnemis plorans*.

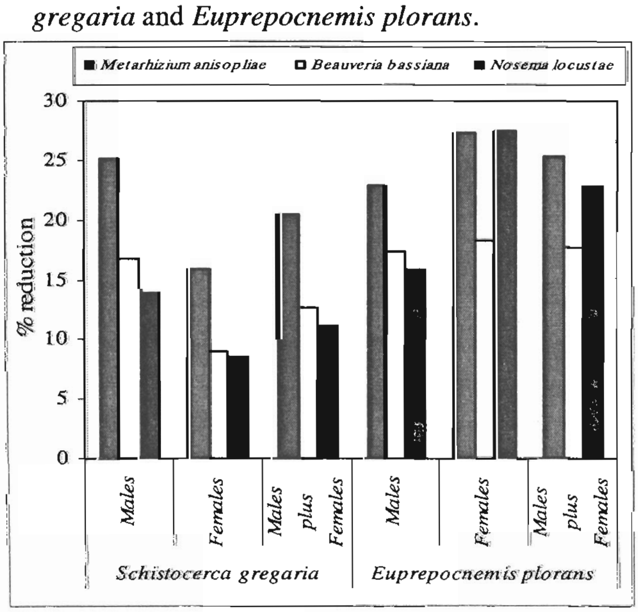

Fig. (3): Percentages of reduction in the adult longevities of *S. gregaria* and *E. plorans*, after infection.

females, followed by *Beauveria* then *Nosema*. While in case of *E. plorans*, the infections caused significant reduction in the males' longevity. The highest reduction was found in infected males with *Metarhizium*, followed by *Beauveria* then *Nosema*. In case of females, the infection also caused significant reduction in infected females' longevity. The highest reduction caused by infection with *Nosema* followed by *Metarhizium* and *Beauveria*. The longevity of males and females were affected by the infections with the three pathogens. The greatest effect was achieved after infection with *Metarhizium* followed by *Nosema* and *Beauveria*.

In general the most effective pathogen on adult fecundity of both *S. gregaria* and *E. plorans* was *Metarhizium* followed by *Beauveria* then *Nosema*. Except in case of *E. plorans* female's longevity, *Nosema* was the most effective pathogen.

Prolongation in the nymphal duration may be attributed to one or all of the flowing reasons: 1- Secretion of juvenile hormone by the fungi and *Nosema*, (Schneiderman *et al.*, 1960 and Fisher & Sanborn, 1964), 2- Hormonal imbalance as suggested by Gaugler & Brooks (1975) and Sloman & Reynolds (1993), 3- Hormone substance disturbance or other factors during the host-pathogens interaction as mentioned by Beckage & Riddiford (1982), El-Maghraby (1984), and El-Maghraby *et al.*, (1988). 4- Starvation of the host; several authors reported the reduction of feeding as a result of infection by many pathogens, *e. g.* Abdelatef (1998), Arthurs & Thomas (2000) and Tefera & Pringle (2003).

In contrast to the present results, the infection with *M. anisopliae* var. *acridum* did not affect the egg production of the brown locust *Locustana pardalina* (Walker), as reported by Arthurs and Thomas (2000) and *S. gregaria* Blanford and Thomas (2001). In these studies, the reduction in pre-oviposition period in infected females was associated with more egg pods. Such reduction of pre-ovipositional period was hypothesized to be due to that the synthesis of juvenile hormone was affected (Blanford and Thomas, 2001).

Henry (1971) observed that grasshoppers, exhibiting light or higher levels of infection with *N. locustae,* rarely possessed detectable quantities of ovarian tissue or egg debris. Also, Sajap & Lewis (1992) reported that the ovarian tissue of *Ostrinia nubilalis* was infected with *Nosema pyrausta*. Abdelatef (1998) studied the effect of *N. locustae* on grasshopper *E. plorans* fecundity and confirmed this result. Malone (1987) suggested that the infection of ovaries is a possible explanation for such reduction.

Such explanation may be accepted in case of *N. locustae* infection, but it was not true for *M. anisopliae* var. *acridum* and *B. bassiana* infections, because the fungal propagation occurs mainly in heamolymph of its host. Moreover, infection may diminish the uptake of nutrients by the host, where the depletion of the nutritive resources was suggested as possible factor for reduction of egg production by other investigators, *e.g.* Gaugler & Brooks (1975) for the infection of *Heliothis zea* by *Nosema heliothidis*, and Ewen & Mukerji (1980) for the infection of *Melanoplus sangunipes* and *M. packardii* by *N. locustae.* The prolongation of the period between eggs pods may be due to that *N. locustae* competes with developing organs for nutrient resources. Gaugler & Brooks (1975) suggested that the adult longevity of *H. zea* was reduced after infection with *N. heliothidis* because of the extensively infected adult fat body. Also, Arthurs and Thomas (2000) found that treatment with *M. anisopliae* var. acridum showed a significant reduction in body fat accumulation at sexual maturity compared with controls.

REFERENCES

Abdelatef G. M. 1998. Studies on most economic grasshoppers (Effect of *Nosema locustae* on *Euprepocnemis plorans* and *Hetracris annuloza*). M. Sc. Theses. Faculty of Agricultural, Zagazig University 105 pp.

Anonymous 1990. A Plague of Locusts Special Report OTA-F-450. OTA. (Office of Technology Assessment). US Government Printing Office, Washington DC. 129 pp.

Anonymous 2006. Hunger in their wake "Inside the battle against desert locust". Food and Agriculture Organization of the United Nations.

Arthurs, S. and M.B., Thomas. 2000. Behavioural changes in *Schistocerca gregaria* following infection with a fungal pathogen: implications for susceptibility to predation. Ecological entomology. V. 26: 227-234.

Beckage, N.E. and L.E. Riddiford. 1982. Effects of parasitism by *Apanteles congregatus* on the endocrine physiology of the tobacco hornworm *Manduca sexta.* Genral and Comparative Endocrinology. 47 (3): 308-322.

Bidochka, M. J., and Khachatourians, G. G. 1990. Identification of *Beauveria bassiana* extracellular protease as a virulence factor in pathogenicity toward the migratory grasshopper, *Melanoplus sanguinipes* J. Invertebr. Pathol. 56, 362-370.

Blanford, S. and Thomas, M.B. 2001. Adult survival, maturation, and reproduction of the desert locust *Schistocerca gregaria* infected with the fungus *Metarhizium anisopliae* var *acridum.* J. Invertebr. Pathol., 78: 1-8.

Coper, A. 1982. The locust and grasshopper agricultural manual for Overseas Pest Research, London. Pp 690. In Van Huis, A., (Ed). Desert locust control with exiting techniques. Proceeding of the seminar held in Wageningen. Pp. 11 – 17.

El-Maghraby, M. M. A. 1984. Der einfluB von *Bacillus thuringeiensis* Berl. auf zwei Wirt Parasit- Systeme : *Spodoptera littoralis* (Boisd.) - *Microplitis rufiventris* Kok.; *Pieris brassicae*-*Apanteles glomeratus* L. Diss. Landw. Fak., Univ. Goettingen.

El-Maghraby, M. M. A., A. Hegab and S. I., Yosif - Khalil 1988. Interactions between *Bacillus thuringeiensis* Berl., *Beauvaeria bassiana* (Bals.) Vuill. and the host parasitoid system: *Spodoptera littoralis* (Boisd.)-*Microplitis rufiventris* Kok. J. Appl. Ent. 106: 417-421.

Ewen, Al. B. and M. K., Mukerji. 1980. Evaluation of *Nosema locustae* (Microsporida) as control agent of grasshopper populations in Saskatc-hewan. J. Invertebr. Pathol. 35: 295-303.

Finney, D. J. 1971. Probit analysis. Cambridge University Press, Cambridge, United Kingdom.

Fisher, F. M. and R. C., Sanborn, 1964. *Nosema* as a source of juvenile hormone in parasitized insects.Biol. Bull., 126: 235-252.

Gaugler, R. R. and W. M., Brooks 1975. Sublethal effects of infection by *Nosema heliothidis* in the corn earworm *Heliothis zea.* J. Invertebr. Pathol. 26: 57-63.

Henry, J. E. 1969. Extension of the host range of *Nosema locustae* in Orthoptera. Annal. Entomol. Soc. America 62: 452-453.

Henry, J. E. and E. A., Oma. 1974. Effects of infection by *Nosema locustae* Canning, *Nosema acridophagus* Henry, and *Nosema cuneatum* Henry (Microsporida: Nosematidae) in *Melanoplus bivittatus* (Say) (Orthoptera: Acrididae). Acrida. 3: 223-231.

Lomer, C. J., R. P. Bateman, D. Dent, H. De Groote, O.K. Douro Kpindou, C. Kooyman, J. Langewald, Z. Ouambama, R. Peveling and M. Thomas. 1999. Development of strategies for the incorporation of biological pesticides into the integrated management of locusts and grasshoppers. Agricultural and Forest Entomology 1:71-88.

Lacey, L. A., and W. M. Brooks. 1997. Initial handling and diagnosis of diseased insects. In Lacey, L. A. [Ed] Manual of techniques in insect pathology. Academic Press p: 1-15.

Malone, L. A. 1987. Longevity and fecundity of argentine stem weevils *Listronotus bonariensis* (Coleoptera: Curculionidae) infected with *Microsporidium itiiti* (Protozoa: Microspra). J. Invertebr. Pathol. 50: 113-117.

McCoy, C. W., R. A. Samson, and D. G.,

`Boucias.1988. Entomogenous fungi. In: IgnoVo, C.M. and N.B. Mandava, (Eds.), Handbook of Natural Pesticides, Vol. V. Microbial Pesticides Part A. Entomogenous Protozoa and Fungi. CRC Press, Boca Raton, FL, pp. 151–236.

Nakhla, N. B. 1957. The life- history, habits and control of the bersim grasshopper,*Euprepocnemis plorans* Charp., in Egypt. Bull. Soc. Entomol. Egypt. 41: 411- 428.

Olson, D., P. Glogoza, and L. Charlet. 2002. Biological Control in the Urban Environment: Part II Parasites and Pathogens. North Dakota State University. (http www.ag.ndsu. edu/ pubs/ plantsci/pests/e1229.pdf)

Prior, C. 1992. Discovery and characterization of fungal pathogens for locust and grasshopper control. In Lomer, C. J. and Prior, C. (eds.) Biological control of locust and grasshoppers. C.A.B. International/University of Arizona Press; Wallingford (UK)/Tucson, AZ (USA), 159-180.

Prior, C., M., Carey, Y.J., Abraham, D., Moore, and R.P., Bateman. 1995. Development of a bioassay method for the selection of entomopathogenic fungi virulent to the desert locust, *Schistocerca gregaria* (Forskaal), J. Appl. Entomol. 119 (8): 567-573.

Sajap, A. S. and L.C., Lewis. 1992. Chronology of infection of aculate corn borer (Lepidoptera: Pyralidae) with microsporidium *Nosema pyrausta*: effect on development and vertical transmission. Environ. Entomol. 21: 178-182.

Schneiderman, H. A., L. I. Gilbert and M. J. Weinstein. 1960. Juvenile hormone activity in microrganism and plants. Nature 188:1041-1042.

Sieglaff, D. H; Pereira, R. M; Capinera. J. L. (1997). Pathogenicity of *Beauveria bassiana* and *Metarhizium flavoviride* (Deuteromycotina) to *Schistocerca americana* (Orthoptera: Acrididae)., J.-Econ. Entomol., 90: 6, 1539-1545;

Sloman, I.S. and S.E., Reynolds. 1993. Inhibition of ecdysteroid secretion from *Manduca prothoracic* glands in vitro by destruxins-cyclic depsipeptide toxins from the insect pathogenic fungus *Metarhizium anisopliae*. Insect biochemistry and molecular biology. V. 23 (1): 43-46.

Tanada, Y. and H. K., Kaya, 1993. Protozoa and fungal. In: Tanada, Y. and H.K., Kaya. (eds.) Insect Pathology. Academic Press, NewYork, pp. 318–366.

Tefera, T. and K. L., Pringle. 2003. Food consumption by *Chilo partellus* (Lepidoptera: Pyralidae) larvae infected with *Beauveria bassiana* and *Metarhizium anisopliae* and effects of feeding natural versus artificial diets on mortality and mycosis J. Invertebr. Pathol. 84(3): 220-225.