EFFECT OF TWO NEEM COMMERCIAL PRODUCTS AND MINT OIL ON SOME BIOLOGICAL ASPECTS OF THE EGG PARASITOID, *TRICHOGRAMMA EVANESCENS* WEST. (HYMENOPTERA: TRICHOGRAMMATIDAE) WHEN REARED ON *PECTINOPHORA GOSSYPIELLA* (SAUNDERS) EGGS.

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INTRODUCTION

The use of natural biochemical pesticides in commercial agricultural and horticultural industries has increased in recent years. These bio-pesticides offer desirable alternatives to synthetic chemical pesticides in agricultural systems where protection of the environment and preservation of beneficial organisms are important. Azadirachtin (AZA) is a botanical compound of interest that can be effective, biodegradable, and rabidly metabolized in the environment (Isman 1999). This compound is a liminoid that accumulates in the seeds of the neem tree (*Azadirachta indica* A. Juss.), from which it can be efficiently extracted (Schroeder and Nakanishi 1987). Recent work from various countries on the use of neem extracts as botanical insecticides is reviewed, and tables are given showing the formulation and results applicable to each of over 50 species of insect pests of agricultural, medical or veterinary importance from the orders Orthoptera, Hemiptera, Lepidoptera, Coleoptera and Diptera. These included both direct toxic effects and inhibition or disruption of feeding, growth, development, reproduction or egg maturation. (Ksmal and Madanlar, 1988). In addition, these natural compounds apparently have minimal toxicity to non-target organisms such as parasitoids, predators, pollinators and earthworm *Lampito mauritii* (Ramakishnan 2004). Thacker *et al.* (2003) indicated that neem extracts are regarded as environmentally friendly chemicals for pest management. In other words, the use of these natural compounds can be substantial contribution toward the preservation of biodiversity in ecosystems despite the fact that they are not completely safe to all stages of beneficial nematodes, mites, and insects. Hence, the present study was conducted on the behavioral and physiological effects of three natural products on the important lepidopterous egg parasitoid, *Trichogramma evanescens*. The use of *Trichogramma*

in biological control has gained widespread interest in several countries. Recently about 18 species of *Trichogramma* are being mass-reared to control pests on corn, sugarcane, tomato, rice, cotton, sugar beet, apple, plum, vineyard, pasture, cabbage, chestnut, sweet pepper, and forest pests in at least 23 countries (Hassan, 1992). During the last few years, *Trichogramma evanescens* (native parasitoid in Egypt) is mass reared (in the Biological Control Research Department, Plant Protection Research Institute, ARC, Cairo) and released in large numbers in sugarcane fields in Upper Egypt for controlling the lesser sugarcane borer, *Chilo agamemnon*. Other trials are carried out in the Bollworm Department at the same Institute for mass production and release of *Trichogramma* spp. on cotton cultivations for controlling the pink and spiny bollworms (Abd El- Hafez, Alia and Nada, 2000, Shalaby *et al.*, 2002). From the previous studies, neem extracts protect crops against insects by causing a series of behavioural and physiological effects. The present study was conducted to determine whether these effects apply to the eggs of pink bollworm *Pectinophora gossypiella* and to the egg parasitoid *T. evanescens*, and what are the precautions that should be taken to minimize those adverse effects.

MATERIAL AND METHODS

The experiments were carried out in the Integrated Pest Management Laboratory, Bollworms Department, Plant protection Research Institute, ARC, Dokki, Giza, Egypt.

Natural products:

The Products are NeemAzal T/s 1% Azadirachtin (German company product), Neemix 4.5% Azadirachtin (Indian company product) and Mint oil. NeemAzal T/s and Neemix are two natural commercial products from *A. indica*, while Mint oil is also a commercial product from spearment plant, *Mintha sativa*. The effect of the three products was evaluated on the eggs of the pink bollworm *P. gossypiella* and the parasitoid at a concentration of 1% (water extract).

Rearing of insects:

Host rearing:

Pink bollworm was reared for several generations on modified artificial diet as described by Abd El-Hafez, Alia *et al.* (1982). Ten pairs of freshly emerged moths were confined in a glass chimney cage, inside which a piece of cotton wool previously soaked in 10 % sugar solution was suspended to be renewed every 48

hours for moths' nutrition. The top and bottom of each cage were covered with screening mesh kept in position by rubber bands for stimulating egg-laying response in the females. Eggs were deposited through the screening mesh on a piece of paper placed upper and under the cage in open Petri-dishes. Cages were maintained at 27±1 °C & 80 ±5 % R. H. and were examined daily for collecting the pieces of papers carrying eggs.

Parasitoid rearing:

Trichogramma used in this study were taken from the culture maintained in laboratory at 27±1 °C & 80 ±5 % R. H, using eggs of the pink bollworm as host. For rearing the parasitoid, fresh host egg sheets (2000-2500 eggs) were exposed to 100-150 adult females into 0.4-1 liter glass jars provided with 10% sucrose solution for nutrition, and covered with cloth-wrapped cotton kept in position by rubber band.

Experimental techniques:

1-Effects on hatchability of *P. gossypiella* eggs:

The same products at the same concentrations were tested against *P. gossypiella* eggs < 24 hr old. Ten tested cards (100-150 eggs/card) were sprayed with each natural product, while ten cards were sprayed with water as control. After drying, the treated cards were kept individually in glass vials and incubated until egg hatch. The numbers of hatched and dead eggs were calculated.

2-Effect on the parasitoid:

a. The sterility effect:

Five pairs of freshly emerged *T. evanescens* were transferred into a glass vial containing treated sugar solution as food, and an egg card containing about 400 pink bollworm eggs for parasitism. Each tube served as a replicate. Five replicates were done for each treatment. Untreated sugar solution was introduced to females as control with similar numbers of replicates. Adults were removed 24 h after exposure to the host eggs. Data were collected on mortality percentage of adults, number of parasitized eggs, percent of adult emergence, sex- ratio, longevity of both treated males and females, and longevity of both emerged male and female parasitoids were calculated.

b. The effect on oviposition (parasitisation).

The effect of the three natural products on oviposition was evaluated under choice and no-choice conditions. Ten cards of pink bollworm eggs (approximately 400/card) were sprayed with a concentration of 1% of each natural product (ten replicates for each treatment). Another ten cards were sprayed with water served as control. Each

card was placed in a glass vial; five pairs of freshly emerged *T. evanescens* adults were introduced into each vial with the help of a fine camel-hair brush and provided with 10% sucrose solution for adult nutrition. In the choice test, the five pairs were provided with cards bearing ≈400 treated and ≈400 untreated eggs in a tube. Tubes were plugged with cloth-wrapped cotton kept in position by rubber band, incubated at the same rearing conditions. Adults were removed after 24 h of exposure to the host eggs. Data on the level of parasitism both in the choice and in the no-choice testes were recorded 5 days after parasitism (oviposition). Data on mortality of the adult parasitoid after 24 h were also recorded, and were subjected to correction by Abbott's formula (1925).

c. The effect on settlement/ feeding Behavior:

Effects of the three natural products on feeding were examined both under the choice and no-choice conditions. Each natural product was mixed with sugar solution (10%) in a ratio of 1:1. In the choice tests, treated sugar solution and untreated one were provided as 2 cm long area on the walls of a glass vial. Around ten freshly emerged *T. evanescens* adults were introduced into each vial. Five replicates were prepared for each treatment. The duration of settlement of adults on the treated and untreated sugar solution was taken as criteria for feeding deterrent effect based on visual observation. In the no-choice tests, ten adult parasitoids were transferred to glass vial containing treated sugar solution (there were five glass vials, each vial serve as a replicate). A comparison in this case was made with control kept in separate five tubes. The method of evaluation was the same as in the choice test.

d. The effect of pretreatment of the host eggs.

Ten cards of pink bollworm eggs ≈400 eggs/card, each card formed as a replicate. There were 10 such replicates for each treatment. Egg cards were sprayed with the three natural products. Another ten egg cards were sprayed with water as control. After drying, each card was kept individually in glass vials. Five freshly emerged *T. evanescens* females were introduced into every vial for parasitism for 24 h. The data on the percentage of adult emergence and their longevity were recorded. Females which succeeded to complete their development in the treated host eggs, were offered new egg cards to determine their efficacy in parasitizing new host eggs. Ten newly emerged mated females were confined individually in glass vials each with new un-treated egg card. Observations were done to determine female fecundity.

Statistical analysis:

Analysis of variance was done on all data (ANOVA) and Duncan's multiple range tests was used to separate the means (Snedecor & Cochran 1980).

RESULTS AND DISCUSSION

1-Effects on hatchability of *P. gossypiella* eggs:

Table (1) showed that, NeemAzalT/S, Neemix and mint oil exhibited ovicidal effect against pink bollworm eggs, as survival of pink bollworm eggs was significantly affected by treatment with the tested products. Mint oil was significantly the most effective compound against pink bollworm egg, as it caused mortality of eggs estimated by 83.14% compared with control (11.76%). NeemAzal was the least effective one, as the percentage of egg mortality was 65.91%; where as treatment with Neemix caused a moderate percentage of mortality (72.32%). No such information is available in the literature for the effect of these products on eggs of the pests. However, Das (1987) studied the toxicity of neem kernel oil to eggs and larvae of the stored products pest *Callosobruchus chinensis,* and he found that treatment of seeds with 4, 6, 8 and 10 ml neem oil/kg increased egg mortality significantly, mortality being 87.2% at the highest concentration. Rovesti and Das (1991) tested aqueous neem seed kernel extracts against eggs of the apple pest *Leucoptera malifoliella* in the laboratory and found that an extract from a batch of kernels at 20g/litter gave a 60% reduction of egg hatching rate. Weathersbee and Tang (2002) observed reproductive effects of Neemix against the root weevil *Diaprepes abbreviatus* (L), and the numbers of larvae hatching per egg mass were reduced by 27% and 68% at 30 and 90mg/Liter.

TABLE (I)

Effect of the three products on eggs of *Pectinophora gossypiella* eggs.

Treatment	% Mortality mean± SD	Rang
NeemAzal T/S	65.91^{b}±9.99	50.45-76.5
Neemix	72.32^{b}±12.04	57.95-85.37
Mint oil	83.14^{a}±11.06	64.93-95.09
Control	11.76^{c}±1.76	9.77-14.63

Means followed by different letters are significantly different (P=0.05), F value= 111.545, LSD=7.485.

2-Effect on the parasitoid:

a. The sterility effect:

Table (2) presents data collected on the effect of feeding on treated sugar solution with the three natural products on parasitism, adult emergence, sex-ratio of emerged progeny, and longevity of both treated and emerged adults. The number of parasitized eggs decreased significantly from 226.8 eggs/5females when females fed on untreated sugar solution (control) to 118, 126.2 and 143 eggs/ 5 females when

females fed on sugar solution treated with NeemAzal T/s, Neemix and mint oil, respectively. The percentage of adult emergence decreased significantly from 95.34% in control to 82.47 and 84.8 when females fed on sugar solution treated with NeemAzal T/s, Neemix, while this percentage increased insignificantly to 96.48% when females fed on sugar solution treated with mint oil. In control, the percentage of females was 64.23%, this percentage decreased insignificantly to 55.57% when females fed on sugar solution treated with mint oil, and significantly to 42.77 and 49.02% when females fed on sugar solution treated with NeemAzal T/s, Neemix, respectively. No detrimental effect was noticed on longevity of both sexes in either treated or emerged adults except for the treatment with NeemAzal. No such information is available in the literature for the effect of these products on the parasitoids. Randen and Roitberg (1998) observed suppression of egg development when the flies, *Rhagoletis indifferens* ingested the neem–based insecticide formulation, containing azadrachtin, and the total suppression of egg development was obtained in adults exposed continuously to concentrations >0.5%.

b. Effect on oviposition.

The three natural products, in the choice test, drastically reduced egg laying of *T .evanescens.* All the three treatments were significantly different from control (Table 3). The reduction in parasitism ranged from 71.08 to 93.13%. In the no-choice test, the difference was significantly, the number of parasitized eggs reduced drastically from 165.8 in control to 52.8, 75.4 and 75.6 in case of treatment with NeemAzal, Neemix and mint oil, respectively. The reduction in parasitism ranged from 68.15 to 54.4%. Naumann and Isman (1995) found that a 1% crude oil emulsion of neem seed oil significantly reduced the proportion of eggs laid by *Spodoptera litura* on treated plants. Kalita *et al.* (2002) indicated that neem act as strong ovipositional deterrent and grain protectant against pulse beetle in storage. Little information exists on the effects of natural extracts on ovipositional behavior of parasitoids. Joshi *et al.* (1982) reported that 2% aqueous neem seed kernl did not deter egg laying on *S. litura* by the egg parasitoid *Telenomus remus* Nixon. But in the present study we found that, the two neem extracts exhibit significant oviposition deterrent effect, while mint oil exhibit insignificant oviposition deterrent effect to *T. evanescens.* The effect of NeemAzal and neemix in this study is similar to results obtained by Klemm and Schmutterer (1993) who showed that spraying of eggs of *Plutella xylustella* with 0.2% neem oil reduced the number of eggs parasitized by *T. principicum* in the laboratory and by *T. pretiosum* in the field. But Raguraman (1994) reported that an aqueous suspension of neem seed kernel at or above 1.2% caused >50% reduction in parasitization to *T. chilonis.*

TABLE (II)

Effect of feeding on sugar solution treated with the three products on parasitism, adult emergence, and longevity of *T. evanescens*

Treatment	Number of eggs parasitized[a]/ 5 females in ≈400 host eggs in 24h.	Adult emergence%	Sex-ratio	Average longevity of treated adults, days		Average longevity of emerged adults, days	
				female	Male	female	Male
NeemAzal T/S	$118^{b\pm}$23.76	82.47^{b}±13.71	42.77^{c}±6.51	1.68^{b}±0.42	1.56^{b}±0.41	2.6^{b}±0.89	1.4^{b}±0.55
Neemix	126.2^{b}±28.78	84.87^{b}±3.87	49.02^{bc}±9.77	2.24^{a}±0.22	$1.76^{ab\pm}$0.30	3.6^{ab}±0.89	2.6^{ab}±0.89
Mint oil	143^{b}±39.36	96.48^{a}±1.55	55.57^{ab}±7.78	2.2^{a}±028	1.92^{ab}±0.18	3.8^{ab}±0.84	$2.5^{ab\pm}$1.29
Control	226.8^{a}±55.04	95.34^{a}±1.37	64.23^{a}±10	2.68^{a}±0.52	2.04^{a}±0.09	4.4^{a}±0.89	3.2^{a}±0.84
LSD	**51.803**	**9.650**	**11.581**	**0.507**	**0.365**	**1.180**	**1.216**

In a column, means followed by a common litter are not significantly different (P=0.05) by ANOVA.
a mean of 5 replications, 5 females/replicate

TABLE (III)

Effect of the three products on oviposition of *T. evanescens* in choice and no choice tests.

Treatments	Choice test				No-choice test		
	No. of eggs parasitized[a]/5females in approximately400 host eggs in 24h.		Difference from un-treated host eggs	% increase (+) or decrease (-) of parasitism from un-treated control	Number of eggs parasitized[a]/ 5 females in ≈400 host eggs in 24h.	Different from un-treated host eggs	% increase (+) or decrease (-) of parasitism from un-treated control
	Treated	Un treated					
NeemAzal	12.8	186.2	173.4**	-93.13	52.8^{b}	113**	-68.15
Neemix	33.4	152.0	118.6**	-78.03	75.4^{b}	90.4**	-54.5
Mint oil	31.33	108.33	77^{ns}	-71.08	75.6^{b}	90.2**	-54.4
Control	-	-	-	-	165.8^{a}		
LSD 0.05					24.758		

In a column, means followed by a common litter are not significantly different (P=0.05) by ANOVA. **, significant; ns, not significant; comparison by LSD. a mean of 5 replications, 5 females/replicate.

c. The effect on settlement/ feeding Behavior:

In the choice test, the percent of adults settled on treated sucrose solution were much lower than the untreated one (Table: 4). The difference from untreated control was highly significant in case of treatment with NeemAzal, and significant in treatment with neemix, while it was insignificant in treatment with mint oil, the reduction in settlement ranged from 37.5 to 71.09% . In no-choice test, the percent of adults settled on treated sucrose solution were reduced insignificantly than control. The reduction in settlement ranged from 48.15 to 55.6%. Schmutterer (1990) reported that neem seed kernel extracts have been reported to deter the feeding of almost all insects tested thus far, albeit the degree of sensitivity varies greatly.

TABLE (IV)

Settling/feeding response of *T. evanescens* on sucrose solution mixed with three products in choce and no-choce tests.

Treatments	Choice test				No-choice test		
	% adults[a] settled on sucrose solution		Different from untreated control	% increase (+) or decrease (-) from untreated control	% adults[a] settled on sucrose solution	Different from untreated control	% increase (+) or decrease (-) from Untreated control
	treated	Un-treated					
NeemAzal	23.3	76.67	53.37***	-69.61	24[a]	30*	-55.6
Neemix	16.67	57.67	41**	-71.09	28[a]	26*	-48.15
Mint oil	27.5	44.0	16.5[ns]	-37.5	26.67[a]	28[ns]	-51.19
Control	-	-	-	-	54.32[a]	-	-
LSD					29.297		

In a column, means followed by a common letter are not significantly differed (p=0.05) by ANOVA. ***Highly significant,**Significant, ns, not significant; comparison by LSD. a mean of 5 replications, 10 adults/replicate. Observations were taken for 1-2 min.

d. The effect of pretreatment of the host eggs.

Table (5) showed that, pretreatment of host eggs with the three natural products reduced the adult emergence insignificantly from 91.58% in control to 86.73% when the eggs were treated with mint oil and significantly to 80.25 and 78.87% when treated with Neemix and NeemAzal, respectively. Longevity of both sexes was unaffected except for longevity of males in case of treatment with NeemAzal, as it decreased to 1.4 days compared with 3.4 days in control. Fecundity of emerged females, which succeeded to complete its development, was decreased insignificantly from 78.4 individuals/female in control to 73.25 progeny/female when the host eggs were treated with mint oil, and significantly to 41 and 43.2 individual/female when the host was treated with NeemAzal and Neemix,

respectively. Raguraman and Singh (1999), found that neem seed oil reduced parasitism by *T. chilonis* at 2.5 and 5.0% concentrations, and stated that this effect was probably resulted from the mortality (36-40%) of the adult parasitoids.

TABLE (V)

Effect of pretreatment of host eggs with the three products on adult emergence, longevity and fecundity of emerged adults of *T. evanescens*.

Treatments	Adult emergence% [a]	Average longevity of adults, days [a]		Fecundity of emerged females [b]
		female	Male	
NeemAzal	$78.87^{b}\pm8.03$	$2.6^{a}\pm0.89$	$1.4^{b}\pm0.55$	$41.0^{b}\pm15.33$
Neemix	$80.25^{b}\pm7.25$	$3.6^{ab}\pm0.89$	$2.6^{a}\pm0.89$	$43.2^{b}\pm11.78$
Mint oil	$86.73^{ab}\pm7.94$	$2.4^{ab}\pm0.83$	$3.8^{ab}\pm1.14$	$73.25^{a}\pm73.25$
Control	$91.58^{a}\pm1.99$	$3.4^{a}\pm0.89$	$3.4^{a}\pm0.55$	$78.4^{a}\pm19.97$
LSD	**9.029**	**1.180**	**1.115**	**20.397**

In a column, means followed by a common letter are not significantly differed (p=0.05) by ANOVA. a mean of 5 replications, 5 females/replicate b Mean of 10 replicates, 1 female/replicate.

e. Toxicity effect.

Mortality of both females and males after 24h post settling on treated sucrose solution (feeding toxicity) and on treated host eggs (contact toxicity) are presented in Table (6). In the feeding toxicity test, there were insignificant differences among the three treatments, but mortality of females was higher than males, and it is perhaps due to that females need nutrition more than males in order to mature and laying eggs. In the contact toxicity test, a maximum of 48% mortality among *T. evanescens* females was recorded at NeemAzal treatment, this percentage of mortality decreased insignificantly to 30% at the treatment of Neemix and decreased significantly to 11.25% in case of mint oil treatment. Males also were affected with the three treatments, as their mortality percentages averaged 40-46.03%, and there were insignificant differences between the three treatments. These result is in contrast with findings of Raguraman and Singh (1999) who stated that males were unaffected by contact with the different neem seed oil concentrations. However, no specific reference is available on the toxic effects of treated substrates or Neem contaminated food against egg parasitoids, except for Raguraman and Singh (1998b) who found toxic effects of aqueous and ethanolic extracts of neem seed kernel on *T. chilonis*. On contrast, nearly the same concentration of neem seed oil and seed kernel extracts did not influence adults of the larval parasitoid, *Bracon hebetor* (Raguraman and Singh, 1998a).

TABLE (VI)

Toxic effect of three products on *T. evanescens*.

Treatments	Corrected mortality (%) [A] of adults in 24 h			
	Feeding toxicity		Contact toxicity	
	Female	Male	Female	Male
NeemAzal	31.19[a]	14.66[a]	48[a]	43.75[a]
Neemix	40.44[a]	17.63[a]	30[ab]	40[a]
Mint oil	31.18[a]	23.07[a]	11.25[b]	68.75[a]
LSD(0.05)	37.98	21.71	33.528	48.176

In a column, means followed by a common letter are not significantly differed (p=0.05) by ANOVA. A mean of 5 replications, 5 females/replicate.

SUMMARY

Two natural products named NeemAzal T/S 1% and Neemix 4.5% (commercial products of Neem plant, *Azadirachta indica*), and Mint oil (a commercial product from Spearmint plant, *Mintha sativa*) were evaluated in the laboratory for their effects on the hatchability percentage of *Pectinophora gossypiella* eggs, and were tested for oviposition deterrence, feeding deterrence, toxicity, sterility effects against the egg parasitoid, *Trichogramma evanescens*. Mint oil was more effective against pink bollworm eggs, as it caused mortality of eggs estimated by 83.14%, followed by Neemix (72.32%), where as, NeemAzal was the less effective one (65.91%). Where as, the effect on the parasitoid was significantly different, as the less effective one on the pest was the harmful one on the parasitoid. No sterility effect was observed when the parasitoid was fed on sucrose solution mixed with the tested products, where as the number of parasitized eggs decreased from 226.8 eggs/5females to 118, 126 and 143 eggs/5females when the parasitoid fed on sucrose solution mixed with NeemAzal, Neemix and mint oil, respectively. The sensitivity of the parasitoid to these products varied considerably under both choice and no-choice conditions. The pretreatment of host eggs showed no adverse effects on the development of the parasitoid, however, pretreatment of the host egg with NeemAzal and Neemix decreased fecundity of emerged females to 41 and 43.2 individuals/female, respectively. In feeding toxicity, females were affected more than males, but in contact toxicity, males were affected more than females except for the treatment with NeemAzal, as it caused 48% mortality to females.

REFERENCES

ABBOTT, W. S. (1925): A method of computing the effectiveness of an insecticide. *(J. Econ. Entomol., 18: 265-267).*

ABD EL-HAFEZ, A. and M. A. NADA (2000): Augmentation of *Trichogrammatoidea bactrae* Nagaraja in the IPM programs for control of pink bollworm, *Pectinophora gossypiella* (Saund.) in Egypt. *(2000 Beltwide cotton conferences, cotton Insect research and Control Conference, 1009-1014).*

ABD EL-HAFEZ, A.; A. G. METWALLY and M. R. A. SALEH (1982): Rearing pink bollworm *Pectinophora gossypiella* (Saund.) on kidney bean diet in Egypt (Lepidoptera: Gelechiidae). *(Res. Bull., Fac. Agric., Zagazig Univ., April, No.576, 10pp).*

BAKTHAVATHSALAM, R., and G. RAMAKRISHNAN (2004): Acute toxicity of neem products to the earthworm, *Lampito mauritii* (Templeton) kept in different substrates of agricultural importance. *(Environ. and Ecol. 22(4): 929-933).*

DAS, G. P (1987): Efficacy of neem oil on the egg and grub mortality of *Callosobruchus chinensis* Linn. (Bruchidae: Coleoptera). *(Tropical Grain Legume Bulletin. 1987; (34): 14-15).*

HASSAN, S. A. (1992): Guide lines for the evaluation of side affects of plant protection products on *Trichogramma cacoeciae, (IOBC/WPRS Bulletin 1992/ XV13:18-39).*

JOSHI, B., G. RAMAPRASAD and S. SITARAMAIAH (1982): Effect of neem seed kernel suspension on *Telenomus remus,* an egg parasite of *Spodoptera litura. (Phytoparasitica 10: 61-63).*

KALITA, J.; P. GOGOI; P. R. BHATTACHARYYA; and R HANDIQUE (2002): A study on effect of traditional storage protectants on oviposition of *Callosobruchus chinensis* and their damage to stored pulses. *(J. Appl. Zool. Res. 13(2/3): 251-254).*

KLEMM, U. and H. SCHMUTTERER (1993): Wirkungen von Niem- praparaten auf die Kohlmotte Plutella xylostella L. und ihre naturlichen Feinde Gattung *Trichogramma. (Z. Pflanzenkr. Pflatzenschutz. 100: 113-128).*

KSMAL, S. and N. MADANLAR (1988): An examination of the effects of *Azadirachta indica* A. Juss (Meliaceae) on insects. *(Turkiye Entomoloji Dergisi. 12(4): 239-249)*

NAUMANN K. and M. B. ISMAN (1995): Evaluation of neem Azadirachta indica seed extracts and oils as oviposition deterrents to noctuid moths. *(Entomol. Exp. Appl. 76(2): 115-120).*

RAGURAMAN, S. (1994): Behavioural and physiological effects of neem (*Azadirachta indica* A. Juss) seed kernel extracts on insect egg parasitoid, *Trichogramma chilonis* Ishii and larval parasitoid, *Bracon hebitor* Say. *(Ph. D. dissertation, post-graduate school, Indian Agricultural Research Institute, New Delhi).*

RAGURAMAN, S., and R. P. SINGH (1998a): Behavioral and physiological effects of neem (*Azadrachta indica*) seed kernel extracts on larval parasitoid, *Bracon hebtor. (J. Chem. Ecol. 24: 1241-1250).*

RAGURAMAN, S., and R. P. SINGH (1998b): Biological effects of neem (*Azadirachta indica*) seed kernel extracts on *Trichogramma chilonis* Ishii, an egg parasitoid. *(Intl. J. Pest Manage. (in press)).*

RAGURAMAN, S., and R. P. SINGH (1999): Biological effects of neem (*Azadirachta indica*) seed oil on an egg parasitoid, *Trichogramma chilonis. (J. Econ. Entomol. 92 (6): 1274-1280).*

RANDEN, E.J. VAN and B.D. ROITBERG (1998): Effect of a neem (*Azadirachta indica*)-based insecticide on oviposition deterrence, survival, behavior and reproduction of adult western cherry fruit fly (Diptera: Tephritidae). *(J. Econ. Entomol.1998; 91(1): 123-131).*

SABBOUR, M. M., and S.E. ABD EL-AZIZ (2002): Efficacy of some botanical oils formulated with microbial agents against the cotton leafworm and Greasy cutworm attacing cotton plants. *(Bull. ent. Soc. Egypt, Econ. Ser., 28:135).*

SCHMUTTERER, H. (1990): Properties and potential of natural pesticides from the neem tree. *Azidarachata indica. (Ann. Rev. Entomol.35: 271-297).*

SHALABY, F.F.; A. ABD EL-HAFEZ; E. F. EL-KHAYAT and M.A.A. EL-SHARKAWY (2002): Applied control of *Pectinophora gossypiella* (Saund.) and *Earias insulana* (Boisd.) in cotton fields by releasing *Trichogrammatoidae bactrae* Nagaraja. *(2nd International Conference, PPRI, ARC, Cairo, Egypt, 21-24 Dec., 1: 402-411).*

SCHROEDER, D. R., and K. NAKANISHI (1987): Simplified isolation procedure for azadirachtin. *(J. Nat. Prod. 50: 241-244).*

SNEDECOR, G. W. and W. G. COCHRAN (1980): Statistical Methods. *(2nd Ed. The lowa State University Press, Ames, lowa, pp. 318).*

THACKER, J. R. M; W. J. BRYAN; C. MCGINLEY; S. HERITAGE; and R. H. C. STRANG (2003): Field and laboratory studies on the effects of neem (*Azadirachta indica*) oil on the feeding activity of the large pine weevil (*Hylobius abietis* L.) and implications for pest control in commercial conifer plantations. *(Crop-Protection. 2003; 22(5): 753-760).*

WEATHERSBEE III, A. A., and Y. Q. TANG (2002): Effects of neem seed extract on feeding, growth, survival, and reproduction of *Diaprepes abbreviatus* (Coleoptera: Curculionidae). *(J. Econ. Entomol. 95 (4): 661-667).*