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N.Z. DIMETRY, S.A.A. AMER, F.M. MOMEN

Laboratory trials of two neem seed extracts on the predatory mites *Amblyseius barkeri* (Hughes) and *Typhlodromus richteri* Karg.

Abstract - The effect of commercial products of two neem seed extracts (Neem Azal-S and Margosan-O) on the predacious mites *Amblyseius barkeri* (Hughes) and *Typhlodromus richteri* Karg was studied. In spite of the fact that both Margosan-O and Neem Azal-S decreased eggs laying as well as the food consumption rate, both extracts at the two tested concentrations were considered to be safe for *A. barkeri*. Margosan-O can be considered harmless for *T. richteri* as no or only slight adverse effects were observed on the predator. Neem Azal-S seems to be harmful to *T. richteri* at 0.2% but slightly harmful at 0.05% concentration. The sex ratio of the progeny in both species was recorded.

Riassunto - *Prove di laboratorio sull'attività di due estratti di semi di neem su* Amblyseius barkeri *(Hughes) e* Typhlodromus richteri *Karg*.

È stata studiata l'efficacia di due prodotti commerciali contenenti estratti di semi di Neem su due acari predatori, *Amblyseius barkeri* (Hughes) e *Typhlodromus richteri* Karg. I due formulati causano una diminuzione dell'ovideposizione e una riduzione nel consumo di cibo ma possono essere considerati selettivi nei riguardi di *A. barkeri*. Margosan-O può essere considerato non nocivo nei riguardi di *T. richteri* in quanto sono stati osservati su questo predatore solo leggeri effetti negativi. Neem Azal-S risulta nocivo nei riguardi di *T. richteri* allo 0,2% ma lo è solo in modo leggero alla concentrazione dello 0,05%. Viene riportata inoltre la sex ratio della discendenza in ambedue le specie.

Key words: *Amblyseius barkeri, Typhlodromus richteri*, neem.

INTRODUCTION

Extracts from seeds and leaves of the neem tree (*Azadirachta indica* A. Juss) are known to contain several insecticidal, nematicidal and acaricidal

compounds (Gill, 1972; Jacobson et al., 1978; Ketkar, 1976 and Warthen, 1979). Up to now, little research has been done on the effect of neem and its products on mites and their natural enemies. Some authors (Jacobson et al., 1978; Schauer & Schmutterer, 1981; Mansour & Ascher, 1984) saiw deterrent effects of neem kernel extracts on the citrus red mite, *Panonychus citri* (McGregor), the two-spotted spider mite, *T. urticae* and the carmine spider mite *T. cinnabarinus*. Dimetry et al. (1993) investigated the efficacy of Margosan-O and Neem Azal-S on the two-spotted spider mite *T. urticae*.

The effect of neem extracts on the predacious mites has not been studied extensively with the exception of the work of Mansour et al. (1987). However a few studies on other types of plant extracts have been conducted on phytoseiid mites such as *Typhlodromus fallacis* (Garmain), (Hamstead, 1970, *Phytoseiulus persimilis* Athias Henriot (Sopp et al. 1990), *Amblyseius gossipi* El-Badry (Dimetry & Amer, 1992). Therefore, it is important to investigate the effect of two neem formulations, namely Margosan-O and Neem Azal-S(*), on the predacious mite *A. barkeri*, which preys exclusively on *Thrips palmi* (Kajita, 1986) and on tetranychid mite *T. urticae*, (Reda & Momen, 1993).

Studies were also extended to another predacious mite, *T. richteri* Karg found on *Malus domestica* at Alnarp, which has been introduced recently by Momen into Egypt from South Sweden.

MATERIALS AND METHODS

Rearing of the prey

The prey was obtained from a laboratory breeding of the two spotted spider mite *T. urticae* Koch on Lima bean, *Phaseolus vulgaris* L. at 28 ± 2 C and 55 ± 5% R.H.

Build up of predators colony

The first breeding of phytoseiid strain was started from 20 females of *A. barkeri*, previously collected from infested grape leaves and twigs of vineyards located at Katta Village in Giza Governorate.

The second phytoseiid colony was started from 20 females of *T. richteri* introduced from Eneskogen (South Sweden) into Egypt. They were collected from *Malus domestica* at Alnarp and on *Picea abies* at Eneskogen (South Sweden). In Egypt this predator was reared successfully under laboratory

(*) Trademark of commercial preparations of neem seed extract.

conditions as mentioned before and fed on *T. urticae* for several generations before being used in the present investigations.

Both phytoseiids were kept separately on cut raspberry leaves which were heavily infested with nymphs of the prey mites T. urticae at 28±2 °C and 55±5% R.H.

Commercial products

Margosan-O and Neem Azal-S are commercial neem seed extracts with 0.3 and 0.35% azadirachtin, respectively, obtained by courtesy of Professor Dr. H. Schmutterer (Giessen, F.R.G.). Two concentrations of each product were selected and prepared (0.2 and 0.05%), which had been proved by Dimetry et al. (1993) to be effective against *T. urticae*. Each concentration gave > 50% mortality.

Treatments

Direct effect on the adult females.

Newly mated females of both predator species (*A. barkeri* and *T. richteri*) were treated with different concentrations and transferred singly to the under surface of raspberry leaf discs (3 cm in diameter) previously treated with the prepared concentrations of either Margosan-O or Neem Azal-S. The upper surface was placed on cotton saturated with water. Tangle foot([1]) was applied on the edge of the discs to prevent the predator from escaping. Untreated nymphs of *T. urticae* were offered as food to both predators. After five days, the adult predators were transferred to clean discs for another 5 days. Four replicates for each concentration were done (20 females/replicate). In every test, control experiments were carried out. During a ten day period, mortality of the females, food consumption, number of eggs laid and the sex ratio of progeny were recorded.

Statistical analysis were carried out using the «F» test. Calculation of the adverse effect of the different extracts on the predator was done according to the formula of Overmeer and van Zon (1982) as follows:

$$E = 100 - (100-M)\ R,$$

where E = Adverse effect;

M = Corrected mortality of treated series with that of untreated (Abbott, 1925);

R = Quotient of the total number of eggs produced in treated and untreated series.

([1]) Tangle foot consists of Canada balsam and citronella oil.

The results obtained from the previous equation will throw light on the total adverse effect of each treatment of the predator.

According to Hassan (1985) the adverse effect of the treatment was classified into the following categories:

1. E = 0-25% Harmless
2. E = 26-50% Slightly harmful
3. E = 51-75% Moderately harmful
4. E = 76-100% Harmful

The percentage of reduction in food consumption was calculated according to Samsoe-Petersen (1983).

$$\% \text{ Reduction} = \frac{C - T}{C} \times 100$$

where C = total number of prey consumed/female in the control;
T = Total number of prey consumed/female in the test.

RESULTS AND DISCUSSION

Effect of Neem Azal-S and Margosan-O treatments:

a) On mortality and eggs production:

The data obtained in Table 1 show that both Neem Azal-S and Margosan-O appeared to be harmless for *Amblyseius barkeri* as no mortalities occurred at the tested concentrations.

On *Typhlodromus richteri*, Neem Azal-S caused 50% and 30% mortalities for individuals tested at 0.2 and 0.05% concentrations respectively, but Margosan-O seems to be harmless for this predator as only 10% mortalities happened at the high concentration tested.

As to the effect on the eggs production, the data obtained in Table 1 show that at high concentration (0.2%) of Neem Azal-S and Margosan-O, a significant reduction in the total number of eggs laid per female during the ten day period in comparison to the control for both species occurred. At low concentration, i.e. 0.05%, Neem Azal-S is harmless for *A. barkeri* but not for *T. richteri* as a significant reduction in the total number of laid eggs during the ten day period occurred.

Using Margosan-O at 0.05%, the number of eggs laid per female is not significant in comparison to the control.

The adverse effect of the two formulations, was calculated according to Overmeer and van Zon (1982); no or only slight harmful effects on *A. barkeri* at the two concentrations ($E < 50$ %) were observed.

Table 1 - Effect of Neem Azal-S and Margosan-O on the reproduction and percentage of mortality of females of A. barkeri *and* T. richteri.

Treatments	*A. barkeri*				*T. richteri*			
	Total No. eggs/♀ /10 days	No. eggs/♀ /day	% M after 10 days	% adverse effect	Total No. eggs/♀ /10 days	No. eggs /♀ /day	% M after 10 days	% adverse effect
Neem Azal-S								
0.2%	22.1	2.21	0	29.51	13.98	1.4	50	68.08
0.05%	27.9	2.79	0	11.00	16.99	1.7	30	45.69
Margosan-O								
0.2%	21.7	2.17	0	30.78	17.02	1.7	10	30.05
0.05%	26.7	2.67	0	14.83	20.00	2.0	0	8.68
Control	31.35	3.14	0	-	21.90	2.19	0	-
L.S.D. at								
0.05	-	0.419			-	0.369		
0.01	-	0.559			-	0.493		

Neem Azal-S can be considered moderately harmful for *T. richteri* at 0.2%, where E = 68.08%, but slightly harmful at the lower concentration (0.05%), where E = 45.69.

Margosan-O, on the other hand, can be considered harmless as no or only slight adverse effects were observed on to the predator.

Therefore, it can be concluded that all the tested extracts were considerably more toxic to the pest *T. urticae* (Dimetry et al., 1993), than to the predator *T. richteri* and at the same time they can be considered safe for the other predator *A. barkeri.*

The present findings are in agreement with that one of Chu et al. (1975) and Saxena et al. (1984), who stated that *Lycosa pseudoannulata*, a spider which is an important predator of rice pests, was not affected by neem oil even at the higher dose of 50 ug/spider, applied topically.

Chiu (1985), showed that a dosage of 250 ug/spider of the seed oil of *Melia azedarach* and of 100 ug/spider of neem oil applied topically had no effect on *L. pseudoannulata.*

Mansour et al. (1987), pointed out that the low concentrations (that is to say 0.2-0.5% of the different neem extracts) which were active against spider mites (Mansour & Ascher, 1984) were not toxic for an important predator, the spider *Chiracanthium mildei.* Kleeberg (1992) stated that recent studies indicate that the effect of neem ingredients on useful insects are negligible. Momen and

Amer (1994), recorded that *A. barkeri* suffered a drop in reproduction when fed on prey formerly kept on leaves treated with lupin, turnip and fenugreek extracts.

b) On sex ratio:

The eggs laid by treated females were observed until the larvae hatched and completed development to protonymphs, deutonymphs and adults. The resulting adults were sexed. The results obtained show that the sex ratio of *A. barkeri* was nearly unaffected in comparison to the control for all the treatments with Margosan-O (table 2).

After treatments with Neem Azal-S, the sex-ratio of the resulting adults is 51% females: 49% males at 0.2%; 66% females: 34% males at 0.05%; 73% females: 27% males in the control.

This means that treatments of *A. barkeri* with either Margosan-O or Neem Azal-S did not affect the sex ratio of the progeny. On the contrary, Neem Azal-S increased the incidence of males in comparison to the control and this

Table 2 - Sex ratio of the progeny of treated females of A. barkeri *and* T. richteri *with Neem Azal-S and Margosan-O.*

Treatments	No. eggs for 20 ♀ ♀ /10 days	Progeny		Sex ratio	
				No.	%
		♀	♂	♀ : ♂	♀ : ♂
		A. barkeri			
Neem Azal-S					
0.2%	442	225	217	1.04:1	51:49
0.05%	558	368	190	1.94:1	66:34
Margosan-O					
0.2*	434	308	126	2.44:1	71:29
0.05%	534	379	155	2.45:1	71:29
Control	627	455	172	2.65:1	73:27
		T. richteri			
Neem Azal-S					
0.2%	262	66	196	1:2.97	25:75
0.05%	312	60	252	1:4.20	19:81
Margosan-O					
0.2%	316	100	216	1:2.16	32:68
0.05%	400	76	324	1:4.26	19:81
Control	438	224	214	1.05:1	51:49

confirms the probability of increased oviposition. These data confirm those of Bonde (1989), who stated that females without access to males did not oviposit and that oviposition is induced by mating.

In *T. richteri*, the sex-ratio was greatly changed in all the treatments. The percentage of males increased greatly in comparison to the females for all treatments, while that for the control ones was 51% females: 49% males (table 2).

c) On food consumption:

The results obtained show that during a 10 day period, a treated female of *A. barkeri* consumed an average 11.53 and 15.85 nymphs of *T. urticae* per day using 0.2 and 0.05% of Neem Azal-S compared to 19.3 nymphs of the prey for the control individuals (table 3).

Treatments with Margosan-O decreased the food consumption to 10.32 and 14.23 nymphs of prey/female/day at 0.2 and 0.05%, respectively. The food consumption of the adults in the control increased significantly to 19.30 nymphs/female/day.

Calculating the percentage reduction in food consumption, the data obtained show that both formulation of neem extract at 0.2% caused a significant reduction in the consumed food (more than 40%). At low concentration (0.05%), the percentage reduction in the food consumption

Table 3 - Effect of Neem Azal-S and Margosan-O on the food consumption of A. barkeri *and* T. richteri.

Treatments	*A. barkeri*			*T. richteri*		
	Total prey /♀/ 10 days	Total prey /♀/ day	% Reduction in food consumption	Total prey /♀/ day	Total prey /♀/ days	% Reduction in food consumption
Neem Azal-S						
0.2%	115.25	11.53	40.26	56.64	5.66	47.43
0.05%	158.50	15.85	17.88	71.56	7.16	33.59
Margosan-O						
0.2%	103.15	10.32	46.53	54.95	5.50	49.00
0.05%	142.30	14.23	26.27	85.40	8.54	20.74
Control	192.95	19.30	-	107.75	10.78	-
L.S.D. at						
0.05	-	2.035		-	1.206	
0.01	-	2.718		-	1.611	

decreased to 17.88 and 26.27% for Neem Azal-S and Margosan-O respectively (table 3).

These data are in accordance with those of Momen and Amer (1994), who found that the number of food consumed by the female predator *A. barkeri* was significantly lower with lupin extract.

A significant lower consumption and an inhibitory effect on reproduction were recorded on *A. gossipi* when exposed to treated prey with fenugreek and Canna extracts (Dimetry and Amer, 1992).

For *T. richteri*, the food consumption decreased significantly for both extracts and for all the concentrations tested. The percentage reduction of the consumed food amounts to 47.43 and 33.59% for Neem Azal-S at 0.2 and 0.05%, respectively. However, the corresponding figures for Margosan-O were 49 and 20.74% at 0.2 and 0.05%, respectively.

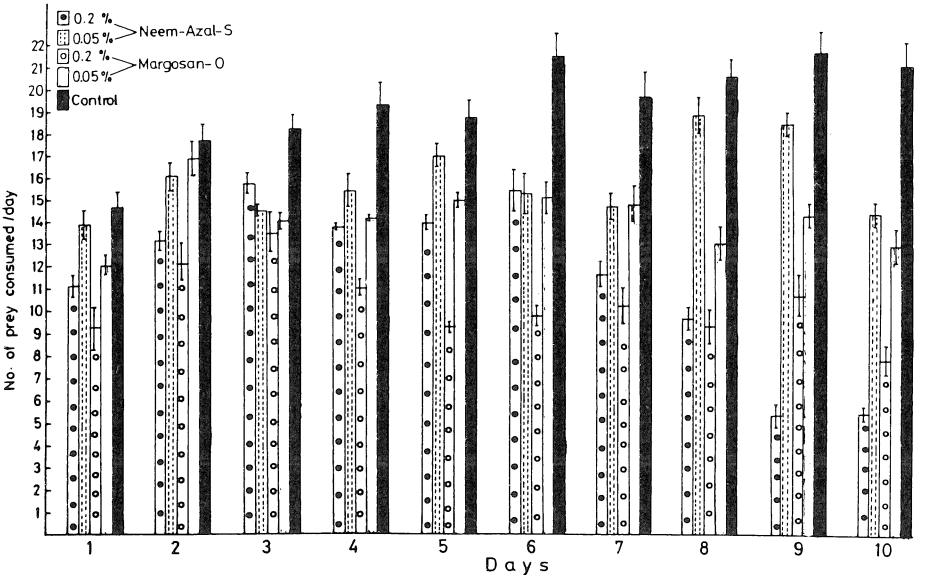

Fig. 1 - The food consumed/female *Amblyseius barkeri* during 10 days period.

Concerning daily food consumption of the female *A. barkeri*, the data obtained in fig. 1 show that females of *A. barkeri* treated with 0.05% Neem Azal-S or Margosan-O behaved nearly in the same way as in the control. However, by increasing the concentration of both formulations to 0.2% the activity of the predator in food consumption is nearly similar to that of the control one during the first five days after which the food consumption in the female decreased greatly.

In the case of *T. richteri*, the female shows non-consistent behaviour in daily food consumption (fig. 2) and generally speaking, the total number of prey consumed per female per day decreased greatly during the whole period of the experiment and for both formulations, especially at 0.2%. In addition, the rates at which nymphs of mites are consumed daily are different.

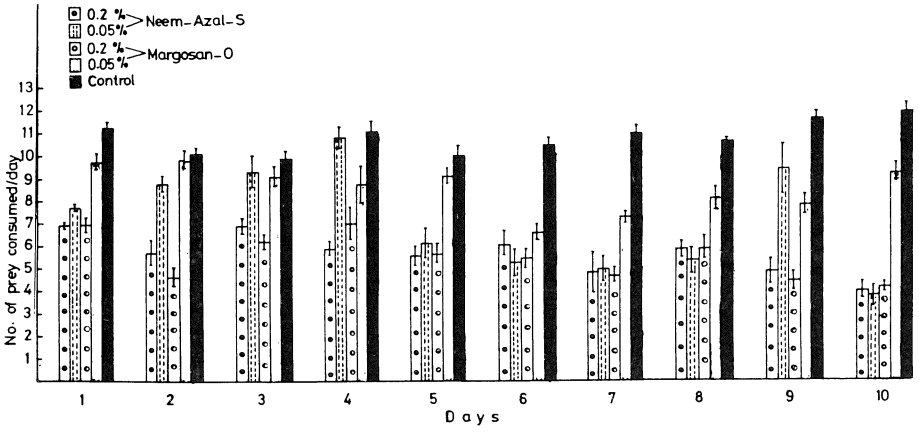

Fig. 2 - The food consumed/female *Typhlodromus richteri* during 10 days period.

CONCLUSION

On the basis of the foregoing results, it can be concluded that both neem extract exert no or only slight harmful effect on A. barkeri at the two tested concentrations. In the case of *T. richteri*, Margosan-O can be considered harmless as no or only slight adverse effect occurred to the predator, while Neem Azal-S exerts 50% mortality, a significant lower consumption rate and an inhibitory effect on reproduction at high concentrations were recorded. Moreover, a positive correlation between rates of feeding and oviposition has been observed for both species.

Thus, it is important to say that the lack of adverse effects of neem extracts on the predator must be considered of great importance when their field application is contemplated.

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PROF. DR. NADIA Z. DIMETRY, SANAA A.A. AMER, FATEN M. MOMEN - Plant Protection Department, National Research Centre, Dokki, Cairo, Egypt.

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