J. Plant Prot. and Path., Mansoura Univ., Vol. 5 (12): 1065 - 1071, 2014

NOVELPESTECIDESFORDESERT LOCUST *Schistocerca gregaria* (Forskal) CONTROL

Gehan A. Mohamed. ; Hala. M. Ibrahim[2] and, G. M. M. Abd-Elatef[1]
Plant Protection Research Institute,Locust and Grasshoppers Research Dep.
Central Pesticides Laboratory, Pesticides AnalsysResearch Dep.
Nadi El-Seid St, Dokki, Egypt.

ABSTRACT

The effect of Abamectin and D-limonene was evaluated against desert locust nymphs under laboratory and field conditions, as well as the combined effect of both insecticides with the fungus *Metarhizium anisopliae* var. *acridum*, also the stability of Abamectin and D-limonene was assessed under different enveronmental condition. The obtained results revealed that both Abamectin and D-limonene could be used to control desert locust, where the reduction in 5th nymphal instar under the laboratory condition reached to 67.5 % after three days post treatment with 40% Abamectine. Whereas under field condition the reduction among young nymphs three days post treatment were 83.33 and 66.66 % in case of 40 % Abamectin and D- limonene, respectively. Moreover the combined effect of 10% Abamectin or D- limonene with *M. anisopliae* var. *acridum* caused significant increase in the toxic effect of the fungi, reached to 58.33 and 51.66 % , respectively.

Keywords *Schistocerca gregaria, Metarhizium anisopliae* var. *acridum,* Abamectin, d-limonene, field, laboratory, nympal instar.

INTRODUCTION

By 1987 the use of Dieldrin -the main tool for locust preventive control- was prohibited for environmental reasons, Dieldrin because of its high efficacy and persistence was successful control agent. since then no alternative insecticides (organophosphates, pyrethroids and carbamates) showed the required qualities to efficiently control desert locust (Balanca and de Visscher 1997**)**. Wide use of chemical pesticides to control locust invasions is a central concern and alternatives a growing necessity. But it is not so easy to find alternatives to these chemicals and to integrate them into operational campaigns **(**Lecoq 2010). *Metarhizium anisopliae* Var. *acridum* is a mycopesticide recommended by FAO as succefull biocontrol agent for the preventive strategies of desert locust control (Anonymous 2009)**.** Abamectin is a broad spectrum insecticide and acaricide with high pesticidal activity, it is a natural product of the soil microorganism *Streptomyces avermitilis* (Qiao et al. 2012).Many plant essential oils show a broad spectrum of activity against pest insects, most essential oil chemicals are relatively non-toxic to mammals and fish in toxicological tests, and meet the criteria for “reduced risk” pesticides (Koul et al. 2008), Limonene is a naturally occurring monoterpene found in citrus, other fruits, The toxicity and neurotoxic effects of d-limonene are discussed by Coats et al. 1991, and the suitability of limonene for control of insect pests has been reviewed by Ibrahim et al.(2001). Several reports mention using limonene for control of plant pests (Tiberi et al. 1999 and

Hollingsworth 2005)**.** The aim of present work is to study the possibility of using Abamectin or D-limonene as novel pesticides for desert locust *S. gregaria*, also the effect of their mixture with *Metarhizium anisopliae* var. *acridum* on desert locust in the laboratory and field, in addition the effect of some environmental conditions on the stability of Abamectin and d-limonene.

MATRIALS AND METHODS

Pesticides used:-

Metarhizium anisopliae var. *Acridum* isolate IMI 330189 (Green muscle 5× 10^{10} Aerial conidia /g dry powder) kindly provided by Biological Pest Control BPC company South Africa.

Abamectin (Vertemic 1.8 % EC) 5-*O*-demethylavermectin A_{1a} (i) mixture with 5-*O*-demethyl-25-de(1-methylpropyl)-25-(1-methylethyl) avermectin A_{1a} (ii).

D- Limonene (Prev AM 6% EC) R-1 methyl- 4-(1-methyl ethaenyl cyclohexane).

Laboratory studies

Both Abamectin and d-limonene were used at three concentration (10%, 20% and 40%) against 5th nymphal instar of desert locust, each treatment consists of four replicates ten nymph each, each nymph were dropped for ten seconds in 100 ml contain the suitable concentration of the used pesticides, then every ten nymphs were placed in cylinder plastic cage covered with a piece of white light cloth. Then feed on clover for three days and the mortality were recorded after 24, 48 and 72 hrs post treatment.

To study the effect of the mixture of each pesticide and *Metarhizium anisopliae* Var. *acridum* three concentrations were prepared as follow:1-100ml 10 % of Abamectin + 0.001 g of *Metarhizium anisopliae* var. *acridum* spores + emulsifiable agent (0.1% Tween 80), 2-100 ml 10 % of d-limonene + 0.001 g of *Metarhizium anisopliae* var. *acridum* spores + Tween 80, and 3-0.001 g of *Metarhizium anisopliae* var. *acridum* spores + Tween 80 alone. Each treatment was applied as previous to 5th nymphal instar. The combined action of the mixtures was calculated in term of co-toxicity factor according to the equation given by Mansour *et al.,*(1966) :

$$\text{Co-toxicity factor} = \frac{\text{Observed \% mortality} - \text{Expected \% mortality}}{\text{Expected \% mortality}} \times 100$$

Where: Observed % mortality: the mortality percentage among treated insects with pesticide and fungi combination and Expected % mortality: the sum of mortality percentage among treated insects with pesticide and fungus in sole.

To study the effect of certain environmental condition on the fate of the two used pesticides, the previous Abamectin and d-limonene formulation were stored in dark glass package according to CIPACF 1995, then stored in oven at 25, 35, 45 and 54 ° C ± 2 for 14 days, in sunny place for 14 days and in room temperature (in door) for 14 days (FAO/WHO 1992**)**, for determination of the active ingredient of both Abamectin and d-limonene before and after storage, HPLC instrument was used under the following

condition: the chromatographic system consisted of Jusco HPLC, diodoarry detector model PU_2089 Ac_{18} stainless column (4.6 nm id X 25 Cm) and the column temperature was 40 ° C. Abamectin and d-limonene were eluted isocratic with acetonitril: methanol: water (60:35:5 v/v), at rate of 0.9 ml/min, under these conditions. The retention time (Rt) for Abamectin and d-limonene were 7.57 and 3.64 minutes, respectively. The results of tested pesticides were quantitatively determined by comparison with the standard under the identical HPLC conditions.

Field experiment

The field trials against desert locust, *S. gregaria* were carried out at Garf Hussein at western shore of Nasser lake south of Egypt, in area infested with desert locust hoppers most of them were 2nd nymphal instar. An area of about 100 X 50 m were treated with each treatment of the following: 1- Abamectin at concentration of 40% 2- d-limonene at concentration of 40% 3- *M. anisopliae* var. *acridum* spores at a rate of 2.5 X 10^8 spores/m^2 4- Abamectin at concentration of 10% + *M. anisopliae* var. *acridum* spores at a rate of 2.5 X 10^8 spores/m^2 5- d-limonene at concentration of 10% + *M. anisopliae* var. *acridum* spores at a rate of 2.5 X 10^8 spores/m^2. Sixteen nymphs were collected and placed in three cages each contain 20 nymph before application, then after application another 60 nymphs were collected from each plot area and kept in three cages.

RESULTS AND DISSECTIONS

Laboratory Bioassay

The obtained results presented in table (1) showed that the mortality percentages of 5th nymphal instar of *S. gregaria* after treatment with Abamectin and d-limonene, it's clear that both pesticieds showed slow action, where the mortality increased by time increase, also d-limonene showed lower toxicity effects than Abamectine. By the 3rd day post treatment the mortality reached 22.5, 37.5 and 67.5 and 15, 25 and 30 % after treatment with Abamectine and d-limonene at the used concentrations.

Table (1) Mortality percentages of 5th nymphal instar of *Schistocerca gregaria* after Abamectin and d-limonene treatment in laboratory.

Periods / Treatments	After 24 hrs			After 48 hrs			After 72 hrs		
	10%	20%	40%	10%	20%	40%	10%	20%	40%
Abamectin	0	2.5	12.5	10	17.5	35	22.5	37.5	67. 5
d-limonene	0	0	0	5	12.5	15	15	25	30

The appilty of *M. anisopliae* var. *acridum* to used in mixture with Abamectin and d-limonene is clearly obvious as data in table 2 shows that mixtures of the fungi *M. anisopliae* var. *acridum* with both used insecticides caused potential effect as described by Mansour *et al.,*(1966). Such potentiating effects as represented by Co toxicity factor were higher in case of *M. anisopliae* var. *acridum* with Abamectine after 24 hr, while it was higher

when *M. anisopliae* var. *acridum* mixed with d-limonene after 48 and 72 hrs. post treatment.

Table (2) The effect of *Metarhizium anisopilae* mixture with Abamectin or d-limonene on the mortality of 5th nymphal instar of *Schistocerca gregaria* in laboratory

Days post treatment	***Metarhizium anisopilae* (Ma) + Abamectin**					***Metarhizium anisopilae* (Ma) + d-limonene**				
	Ma alone	Abamectin alone	Expected (Ma+ Abamectin)	Observed (mixture)	Co toxicity factor	Ma alone	d-limonene alone	Expected (Ma+ d-limonene)	Observed (mixture)	Co toxicity factor
24 hr	1	0	1	5	80.0	1	0	1	2.5	60.0
48 hr	5	10	15	20	25.0	5	5	10	15	33.3
72hr	10	22.5	32.5	45	27.8	10	15	25	35	28.6

Field trails

Data Illustrated in Figure (1) showed that the effect of the used pesticides and the fungi *M. anisopliae* var. *acridum* (Ma) alone as well as the combination of the two pesticides at concentration of 10% each with the fungi. The field data indicated that Abamectine caused reduction in desert locust population reached to 83.33% by the 3rd day post treatment while D-limonene reached to 66.66% and *M. anisopliae* var. *Acridum* were 25 after 3 days post treatment. It's clear that the combination of Abamectine and D-limonene at concentaration of 10 % enhanced the effect of *M. anisopliae* var. *Acridum* against desert locust nymphs under field condition where the mortality reached to 58.33 and 51.66, respectively.

Results obtained from laboratory boiassay and field trails, indicate that abamectine showed slow act against desert locust, this may be du to it's mode of action where Jansson et al 1996 and Wolstenholme and Rogers 2005 indicated that, it is likely that abamectin bind to multiple sites (including glutamate and GABA) in insect chloride channels. In general, the chloride ion flux produced by the opening of the channel into neuronal cells results in loss of cell function and disruption of nerve impluses. Consequently, invertebrates are paralyzed irreversibly and stop feeding. Maximum mortality of arthropods is achieved within 4 days.

The effect of abamectin against desert locust was expectied, since Abamectin was very potent against mites and certain insect species (Dybas *et al.,* 1989). While d-limonene mode of action in insects is not fully understod, it may cause an increase in the spontaneous activity of sensory nerves, this hightened activity sends spurious information to motor nerves and results in twitching, lake of coordination and convulsions, the central nervous system may also be affected, resulting in additional stimulation of motor nerves, which may led to rapid knockdown paralysis (Weinzierl and Henn 1994). *M. anisopliae* Var. *acridum* proved to be succefull biocontrol agent against desert locust and grasshoppers under Egyptian condetions but it's relatevely slow of action may be disadvantage of this fungi when making control desigen (Abdelatef 2005)**.**

Figure (1) Effect of Abamentin, D-limonene, *M. anisopliae* var. *acridum*, *M. anisopliae* var. *acridum*+ Abamectin and *M. anisopliae* var. *acridum*+ D-limonene under on the mortality of Schistocerca gregaria nymphs under field condetion.

Effect of storage conditions

Data presented in table (3) showed that the percentage of active ingredient in Abamectin 1.8% and D-limonene 6% were 1.79 and 5.96 % before storage, recording dissipation and loss equal to 0.223 and 0.671 respectively. The percentages of loss of Abamectin and D-limonene were increased as temperature increase, it reached to 9.96 and 71.43% after storage at 54 ° C for 14 days, it's clear also that d-limonene showed great loss when stored at 45 it was 35.44. While the storage in sunny place for 14 days caused loss in Abamectin and d-limonene reached to 6.13 and 10.91. It's clear that storage in door caused slight loss in the percentage of active ingredient of both insecticides which were 0.897 and 1.18 %, respectively. It could be concluded that the residue of all the tested samples greatly deteriorated when exposed to direct sun light for long periods. The great reduction of D-limonene may be due to oil essential. The reduction in the used pesticides residues may be due to thermal, evaporation and light intensity consideration as stated by several investigators (Halley et al. 1993 and Mrozik *et al.* 1988).

Table (3) Effect of storage at different degree of temperature, in sunny place and room temperature (in door) on active ingredient of abamectine and d-limonene

		Abamectine concentration	% of loss	d-limonene concentration	% of loss
Before storage		1.796	0.223	5.96	0.671
After storage at	25° C	1.768	1.81	5.92	1.35
	35° C	1.759	2.33	5.66	6.01
	45° C	1.68	7.14	4.43	35.44
	54° C	1.637	9.96	3.5	71.43
After storage in sunny place		1.696	6.13	5.41	10.91
After storage in door		1.784	0.897	5.93	1.18

In the present work it could be concluded that mixture of Abamectin or d-limonene with *M. anisopliae* Var. *acridum* could be succeful in locust control under field conditin specially against young desert locust nymphs.

REFERENCES

Abdelatef G. M., (2005). Effect of green muscle on locust and grasshoppers. Final report of project (FAO) CRC/EMPRES (PR27209). pp: 53.

Anonymous 2009. Second international workshop on the future of biopesticides for desert locust management (Rome, 10-12 February 2009). Food and Agriculture Organization of the United Nations, Rome.

Balanca G., and M. N., de Visscher. 1997. Impacts on non target insects of a new insecticides compound used against the desert locust Schistocerca gregaria (Forskal). Arch. Environ. Contam. Toxicol. Vol. 32, 58-62.

Coats, J. R., L. L. Karr, and C. D. Drewes. 1991. Toxicity and neurotoxic effects of monoterpenoids: in insects and earthworms, pp. 305-316. In P. A. Hedin [ed.], Naturally occurring pest bioregulators. ACS Symposium Series, American Chemical Society, Washington, DC.

Dybas, R. A. Hilton, N. J., Babu, J. R., Preiser, F. A. and Dolce, G. J. (1989). Novel second-generation avermectin insecticides and miticides for crop protection. In: Novel Microbial Products for Medicine and Agriculture (ed. A.L. Demain et al.) pp. 203–212. Society Industrial Microbiology, Annandale, VA.

CIPAC F. 1995. Collaborative international pesticides analytical council.

FAO/WHO 1992. Meeting on pesticide has discussed and evaluated.

Halley B. A.; W. A. Vanden Heuvel and P.G. Wisloki 1993. Environmental effect of the usage of vermectin in live stock. Vet. Parasitol.48:109-125.

Hollingsworth R. G. 2005. Limonene, a Citrus Extract, for Control of Mealybugs and Scale Insects. J. Econ. Entomol. 98(3): 772-779.

Holopainen. 2001. Insecticidal, repellent, antimicrobial activity and phytotoxicity of essential oils: with special reference to limonene and its suitability for control of insect pests (Review). Agric. Food Sci. Fin. 10: 243-259.

Ibrahim, M. A., P. Kainulainen, A. Aflatuni, K. Tiilikkala, and J. K. Tiberi, R., A. Niccoli, M. Curini, F. Epifano, M. C. Marcotullio, and O. Rosati. 1999. The role of the monoterpene composition in *Pinus* spp needles, in host selection by the pine processionary caterpillar, *Thaumetopea pityocampa*. Phytoparasitica 27: 263-272.

Jansson R.K., R. Brown, B. Cartwright, D. Cox, D.M. Dunbar, R.A. Dybas, C. Eckel, J.A. Lasota, P.K. Mookerjee, J.A. Norton, R.F. Peterson, V.R. Starner and S. White. 1996. Emamectin benzoate: a novel avermectin derivative for control of lepidopterous pests. The management of diamondback moth and other crucifer pests. Proceedings of the Third International Workshop, 29 October - 1 November, 1996, Kuala Lumpur, Malaysia. 171-177.

Koul O., S. Walia and G. S. Dhaliwal. 2008. Essential Oils as Green Pesticides: Potential and Constraints. Biopesticides International. 4(1): 63–84

Lecoq M. 2010. Integrated Pest Management for locusts and grasshoppers: are alternatives to chemical pesticides credible?. J. Orthoptera Research 19 (1): 131-132.

Mansour, N.A; M.E., El-Defrawi; A., Toppozada, and M., Zeied, 1966. Toxicological studies on the Egyptian cotton leaf-worm, *Prodenia litura*. VI potentiation and antagonism of organophosphorus and carbamate insecticides. J. Econ. Entomol., 59 (2):307-311.

Mrozik H., P. Eskola, G. F. Reynolds, B. H. Arison G. M. Smith, and M. M. Fisher.1988.Photoisomers of avermectins. J. Org.Chem.53:1820-1823.

Qiao K., X. Liu, H. Wang, X. Xia, X. Ji and K. Wang. 2012. Effect of Abamectin on root-knot nematodes and tomato yield. Pest Management Science. 68 (6): 853-857.

Weinzierl, R. and T. Henn. 1994. Chapter 48: Botanical insecticides and insecticidal soaps, pp. 541-556. In: A. R. Leslie [ed], Handbook of Integrated Pest Management for Turf and Ornamentals. Lewis Publishers, Boca Raton, FL.

Wolstenholme A.J. and A. T. Rogers. 2005. Glutamate-gated chloride channels and the mode of action of the avermectin/milbemycin anthelmintics. Parasitology 131, S85–S95.

مبيدات حديثة لمكافحة الجراد الصحراوي

جيهان على محمد[1] و هالة محمد ابراهيم[2] و جمال محمد محمود عبد اللطيف[1]

1- قسم بحوث الجراد و النطاط, معهد بحوث وقاية النباتات.

2- قسم بحوث تحليل المبيدات, المعمل المركزى للمبيدات.شارع نادى الصيد الدقى

تم دراسة تأثير مبيد الأبامكتين و الدليمونين على حوريات الجراد الصحراوى تحت ظروف المعمل و الحقل كذلك مخلوط هذان المبيدان مع الفطر ميتاريزيوم انيسوبلاى فار اكريديوم. ايضا تم تقييم ثبات هذان المبيدان تحت ظروف تخزين مختلفه. النتائج المتحصل عليها اظهرت ان كل من الأبامكتين و الدى ليمونين يمكن استخدمهما فى مكافحة الجراد الصحراوى. وجد ان نسبة الخفض فى حوريات العمر الخامس للجراد فى المعمل وصلت الى 67,5 % بعد 3 ايام من المعاملة. بينما فى الحقل وصلت نسبة الموت بين الحوريات المعامله الى 83.33 و 66.66 % عند المعاملة بالأبامكتين و الدى ليمونين بتركيز 40 % لكلاهما. بينما ادى اضافة كل من المبيدين بنسبة 10% الى الفطر ادى الى زيادة الفعل السام للفطر و التى وصلت الى 58.33 و 51.66% على التوالى.