Tikrit Journal of Pharmaceutical Sciences 2007, 3(1) :63 - 68

Efficacy of Enriched *Melia azedarach* L. Extract on Immature Stages of the Pest *Spodoptera cilium latebrosa*(Guerine) (Lepidoptera: Noctuidae)

Atallah F. Mekhlif
Dept. of Biology, College of Education, Mosyl University, Mosyl, Iraq

Received 24/12/2006 :accepted 21/1/2007

Abstract

The enriched methanolic extract from unripe fruits of *Melia zedarach* was used for diet treatment of *Spodoptera cilium latebrosa*. Artificial diets were treated with concentrations of 0.25, 0.5, 1.0, 5.0, 10.0 and 15.0 ppm. Starting with 3rd instar, mortality percentage were 33.3, 45.8, 75.0 and 100% at the concentrations 0.5, 1.0, 5.0 and 10.0 ppm respectively. Residual activity was high toxic during one-day-old at the higher concentrations 10.0 and 15.0 ppm. Duration of the life cycle was affected by diet treatment; larval and pupal periods were prolonged, while that of prepupa shortend. The extract lost its residual toxicity by the 8th day after treatment.

تاثير مستخلص ثمار السبحبح المركز في الاطوار غير الكاملة للافة Spodoptera cilium latebrosa (Lepidoptera: Noctuidae)

عطا الله فهد مخلف

المستخلص

تمت معاملة غذاء الافة *Spodoptera cilium latebrosa* بالمستخلص الميثانولي المركز للثمار غير الناضجة للسبحبح، كانت تراكيز المستخلص الى غذاء يرقات 0.25، 0.5، 1.0، 5.0، 10.0 و 15.0 جزء في المليون (ج ف م). وجدت نسبة الموت 33.3، 45.8، 75.0 و 100% عند اضافة التراكيز 0.5، 1.0، 5.0 و 10.0 (ج ف م) على التوالي. كان الاثر المتبقي للمستخلص باضافة التركيزين 10.0 و 5.0 (ج ف م) ذو سمية عالية بعد يوم من المعاملة. وتاثرت دورة حياة الافة، اذ طالت فترة طوري اليرقة والعذراء وقصرت فترة طور قبل العذراء. وفقد المستخلص السمية المتبقية بعد اليوم الثامن من المعاملة.

Introduction

The species number of the genus *Spodoptera* are second only to of *Heliothis* (*Helicoverpa*) among pestiferous Lepidoptera worldwild(1).

S. cilium latebrosa (Guerine) is one of the harmful pests in Iraq(2). In Saudi Arabia were found host plants of *S. cilium latebrosa* belongs to the families. *Boragenaceae*, *Asteraceae*, *Leguminosae*, *Solobnaceae*(3).Also, it was reported that *S. cilium* is one of the rice pests in Senigal(4).*Melia azedarach L.* is a tree of the family Meliaceae native to India and introduced to Iraq many years ago. The extracted liminoids from fruits of chinaberry (*M. azedarach*) had significant antifeedant and insecticidal properties(5). De *et al.* (6) found that aqueous extracts of *M. azedarach* applied on bean plants leaves interfere with longevity and development of immature stages of *Bemisia tabaci*, the vector of Golden Mosaic virus. The powdered fruits of *M. azedarach* were extended larval period of *Helcoverpa armigera*(7). The observation of strong biological activity of methanolic extract of seeds of *M. azedarach* makes this plant a potential tool in the control of chagas disease by lowering ecdysis percent of fourth instar numphs of *Rhodnius prolixus* to about 25%(8). Extracts of leaves and fruits of *M. azedarach* were effective in reducing the number of larvae of Maize stalk borer *Busseola fusca* (Fuller)(9). *M. azedarach* leaves and seeds extracts in benzene were found significantly effective in the repellency of red pumkin beetle *Aulacophora faveicolla*(10).Toosendanin is one of the commercial bioinsecticides, it was isolated from *M. azedarach.* Cespedes *et al.* (12) used toosendanin as positive control, caused Lc_{50} 8.0 ppm at 7 days for fall armyworm *S. frugiperda.*The present investigation reports attempts, under laboratory conditions, to evaluate the efficacy of *M. azedarach* fruits extract as one of the sources of bioinsecticides.

Materials and Methods

Insect rearing

The fertilized flying females of *Spodoptera cilium latebrosa* were collected by handle net near outdoor lights. The captured female was egg oviposited in one liter jar.

Preparation of the extract

The green unripe fruits were collected in October 2005 from *Melia zedarach* L. trees. After removing the pedicels, the fruits were washed and stored in a freezer.In order to prepare an enriched extract, 50 gm of frozen fruits was ground and mixed with 100 ml 80% methanol. The suspension was preserved in a refrigerator for 24 h for maceration, then stirred for 4 h and filtered under low pressure. The solvent was evaporated at room temperature. The dray crude extract was redisolved in 80% methanol and washed with equal volume of petroleum ether (b.p 30-50 °C) by shaking for half an hour. After separating the layers in the funnel, the methanol extract layer was separated, the solvent was evaporated under room temperature. Then dray methanol extract was dissolved in two solvents, distilled water and the same volume of ethyl acetate in a separator funnel, and mixture was shacked for half an hour and was left for 24. The ethyl acetate extract layer was dried by evaporization and redissolved in 80% methanol to prepare 1000 ppm stock solution. Five larvae were used for each concentration the statistical concentrations were based on ten readings.Larval mortality if any, was recorded daily as a percentage of died larvae, twenty five larvae were used for each concentration mortality within prepupal period was not seen. Otherwise, malformed prepupal stage was added to mortality of pupae.

All developmental stages of a continuous culture of *S. cilium latebrosa* were maintained at standard conditions of 27 ± 1 °C, 60 ± 5% R.H and 16:8 (L:8) photoperiod in the incubator.Larvae were fed in artificial diet as prepared for *S. exigna* (Hubner)(12,13). The adults were fed on a 15% honey water solution.

Experimental treatment

Stock solution of the extract was mixed with the artificial diet before gelling, at initial concentration of 0.25 ppm to 250 gm of the weight of the duet. The carrier was evaporated and controlled diet were prepared with carrier alone.Single larva was used to avoid cannibalism that is prevalent in the larvae of S. ciliumTo determine the effect of *M. azedarach* extract on *S. cilium* life cycle, newly moulted third-instar larvae, were starved for 5 h and transferred singly to 30 cm^3 plastic containers. In order to avoid mortality at immature stages at used applied concentration, twenty.

Results and Discussion

Figure (1) shows high levels of larval mortality of Spodoptera cilium latebrosa. When 3rd instar larvae diet were treated with enriched methanolic fruits extract of Melia zedarach, the concentration 0.25 ppm did not cause mortality, but mortality begin at 0.5 ppm with a relatively high percentage. The mortality was proportionally increased with diet application at the concentrations 1.0, 5.0 and 10.0 ppm. The value of Lc_{50} was calculated 1.7 ppm. As compared with other pests whose diet treated with M. azedarach extract, S. cilium lalebrosa larvae were very susceptible to M. azedarach fruits, for instance, Lc_{50} was 8.8 ppm for S. exigua (Mekhlif, 2004).The relative sensitivity of S. cilium latebrosa was too high in relation to other pests, it was equal to 10 folds more than both S. exigua and S. littoralis and 20 folds for Agrotis ipsilon (13,14).Also, Figure (1) shows that mortality percent of pupae decreased in comparison with larval mortality, the effect of limonoids of the extract on pupal stage only through pathways of metabolism. While, as well as that effect on larval stage, the alimentary tract of feeding larva distorted by these limonoids.Data presented in Table (1) show that the daily residual toxicity of enriched methanolic extract of *M. zedarach* on developed 3rd instar larvae. The toxic effect started since 4-day-old residue activity for the concentrations 0.5 and 1.0 ppm. The toxicity at concentrations 5.0, 10.0 and 15.0 ppm started at one-day-old residue activity, but the residual effect was very high at last two concentrations (92.0 and 96% respectively).Permanent larvae at concentrations 10.0 and 15.0 ppm (4.0%) were only seen. The moulting failure of permanent larvae may be attributed either to erosion of epidermal layer and/or to disruption of neurosecretion organs by extract azadirachtin constituents (15).The methanolic fruits extract lost nearly its residual toxicity by the eight day after the diet treatment.Table (2) shows that the sublethal doses of enriched fruits extract caused a significant disruption of immature stages periods, this disruption ranged between extending and shortening for those periods. The significant extension of the larval period (3rd-prepupa) began at concentration 0.5 ppm, maximum extension was 4.4 days at the highest sublethal extract concentration 5.0 ppm.In spite of short control prepupal period (1.9 days), but it was affected by treated diets, and decreased for about a half day at extract concentrations 1.0 and 5.0 ppm. Previous studies did not focus on the effect of plant extracts on prepupal stage, but, on the basis of the weight, dramatic deficiency had been occurred in the larvae weight through metamorphosing to prepupa. In the

biochemistry language, drastic tissues catabolism and cell patches rearrangement occurred for prepupa as preparation step to pupal period.The average time to pupation and emergence of adults was prolonged as compared with untreated control. The concentration 0.25 ppm was disrupted pupation period, but had not influenced the larval and prepupal periods, this sensitivity may be attributed to interference of the extract active ingredients with histolysis and histogenesis processes within pupal stage.

Table (1): Residual toxicity of *Melia azedarach* extract on larvae of *Spodoptera cilium latebrosa* (Guerine)*

Date of examination	Concentration (ppm)						
	control	0.25	0.5	1.0	5.0	10.0	15.0
19.9	-	-	-	-	4	23	24
20.9	-	-	-	-	3	1	-
21.9	-	-	-	-	4	-	-
22.9	-	-	1	1	2	-	-
23.9	-	-	6	2	5	-	-
24.9	-	-	2	7	1	-	-
25.9	-	-	-	5	-	1**	1**
26.9	-	-	-	-	-	-	-

* 25 larvae for each treatment.
** permanent larvae

Table (2): Effect of fruits extract of *Melia azedarach* on the development of *Spodoptera cilium latebrosa* immature stages (days)*

Concentration (ppm)	III-Prepupa	Prepupa	Pupa
Control	11.5 ± 0.8 c	1.9 ± 0.3 a	8.1 ± 0.8 d
0.25	11.5 ± 1.4 c	1.9 ± 0.8 a	8.5 ± 0.7 cd
0.5	13.2 ± 0.4 b	1.7 ± 0.5 ab	9.1 ± 0.7 bc
1.0	15.5 ± 3.2a	1.3 ± 0.5 b	9.6 ± 0.9 ab
5.0	15.9 ± 1.1 a	1.3 ± 0.5 b	9.9 ± 0.9 a

* means within in column followed by the same letters are not significantly different (P = 0.085; Duncan's multiple range test).

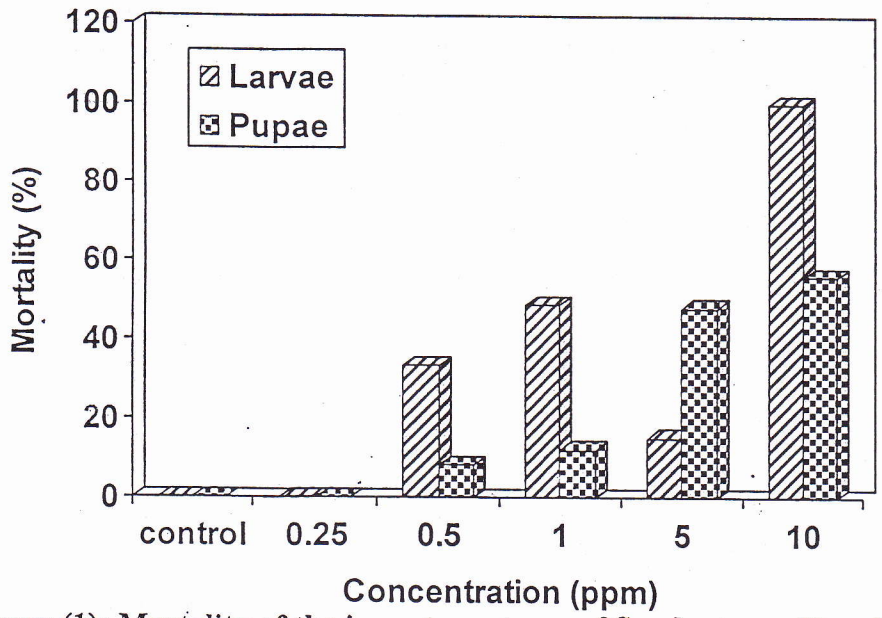

Figure (1): Mortality of the immature stages of Spodoptera cilium latebrosa, diet of the developed larvae was treated with fruits extract of Melia azedarach

References

1- Schmutterer H. (ed.) (1995). The neem tree, source of unique natural products for integrated pest management, medicine, industry and other purposes/ VCH/ Weinheim, 696.

2- Wiltshire E.P. (1957). The Lepidoptera of Iraq. Adlard and Son, Bartholomew Press, Dorking, p. 160.

3- Al-Ahmadi A.Z. and Salem M.M. (1995). Entomofuna of Saudi Arbaia. Part 2. King Saudi Univ. Academic Pub. and Press, p. 187.

4- Helnrichs E.A. and Barrlon A.T. (2004). Rice-feeding insects and selected natural enemies in West Africa. In International Rice Research Institute, p. 247.

5- Carpinella M.C., Defago H.T., Valladares and Palacios S.M. (2002). Antifeedant and insecticide properties of limonoid from Melia azedarach (Meliaceae) with potential use for pest management. J. Agric. Food Chem., 51(2): 369-374.

6- De N.E.A.B., Costa A.C. and Lourencao A.L. (1987). *Melia azedarach* extract as an antifeedant to *Bemisia tabasi* (Homoptera: Aleryodidae). Florida Entomologist, 80(1): 92-94.

7- Anwar T., Jabbar A., Khalique F., Tahir S. and Shakel M.A. (1992). Plants with insecticidal activities against four major insect pests in Pakistan. Trop. Post Manage., 38: 431-437.

8- Cabral M.M., Garcia E.S., Rembold H.De Simone, S.G. and Kelecom A. (1996). Anti-moulting activity in Brazilian *Melia azedarach*. Mem. Inst. Oswaldo Gruz, Rio de Haneiro, 91(1): 117-118.

9- Amlak G. (1999). Insecticidal activity of chinaberry, endodand pepper tree against the maize stalk borer (Lepidoptera: Nocluidae) in Southern Ethiopia. International J. Pest Management, 45(1): 9-13.

10- Khan S.M. and Wasum M. (2001). Assessment of different plant extracts for their repellency against red pumpkin bettele (*Aulacophora faveicollis Lucas*) attacking muskmelon (*Cucummis melo L.*) crop/Online J. Biological Sciences, 1(4): 198-200.

11- Cespedes C.L., Calderon J.S., Lina L. and Aranda E. (2000). Growth inhibitory effect on fall armyworm *Spodoptera frugiperda* of some limonoids isolated from Cedrela spp. (Meliaceae). J. Agric. Food Chem., 48(5): 1903-1908.

12- Smagghe G. and Degheele D. (1994). Action of a novel nonsteröidal ecdysteroid mimic, tubufenozide (RH-5992), on insects of different orders. Pestic. Sci., 42: 85-92.

13- Mekhlof A.F. (2004). Effect of chinaberry tree, *Melia azedarach* L. extract on the reproduction and development of beet armyworm Spodoptera exigua (Hubner) Noctuidae: Lepidoptera). Ph.D. Thesis, College of Science, Mosul University.

14- Schmidt G.H., Adel A.I. and Breur M. (1997). Effect of *Melia azedarach* extract on larval development and reproduction parameters of *Spodoptera littoralis* (Boisd.) and Agratis ipsilon (Hufn.) (Lep-Noctuidae). Anz. Schadlingskde., Pflanzenschutz, 70: 4-12.

15- Rembold H., Subrahmanyam B. and Muller T. (1989). Corpus cardiacum-a target for azadirachtin. Experientia., 45: 361-363.