Pakistan J. Zool., vol. 39(3), pp. 179-184, 2007.

Efficacy of Mixtures of an Organophosphate Malathion and a Synthetic Pyrethroid Deltamethrin against Lesser Grain Borer, *Rhyzopertha dominica**

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Abstract.- Adults of six strains of stored grain pest, *Rhyzopertha dominica viz.* Chichawatni (C), Karachi (K), Wazirabad (W), Sialkot (S), Lahore (L), and Multan (M) were treated with an organophosphate malathion, and a synthetic pyrethroid, deltamethrin (Decis), alone and in two different combinations. C strain was found to be most resistant to malathion (LC_{50}, 115.50 ppm), whereas M strain was least resistant (LC_{50} 12.40 ppm). On the other hand, M strain was most resistant to deltamethrin (LC_{50} 10.55 ppm), whereas L strain was most susceptible (LC_{50} 2.83 ppm). A mixture of malathion : deltamethrin (10:1) was most effective against K, S and M strain with LC_{50} of 4.71, 5.33 and 8.38 ppm, respectively, whereas mixture 10:1.5 and 9:2 was most effective against W strain (LC_{50} 4.52 and 3.13 ppm) and M strain (LC_{50} 8.25 and 5.27 ppm), respectively. *R. dominica* did not show any definite pattern of susceptibility to both types of mixtures. It is concluded that mixture 10:1 with LC_{50} ranging between 4.53 to 24.94 ppm, mixture 10:1.5 with LC_{50} ranging between 4.52 to 8.74 ppm and mixture 9:2 with LC_{50} ranging between 3.13 to 61.53 ppm were found to be most effective for almost all the strains.

Key words: Deltamethrin (Decis), malathion, pyrethroid, organophosphate, stored grain pests, *Rhyzopertha dominica*, pest control, LC_{50}.

INTRODUCTION

Rhyzopertha dominica, the Lesser Grain Borer is a pest of economic significance in the United States of America, Southern Canada, Argentina, New South Wales, South East Australia and Indo-Pakistan sub-continent (Cuperus *et al*.,1986 Shakoori *et al*., 2000; Toews and Subramanyam, 2003; Flinn *et al*., 2004; Toews *et al*., 2005; Fields, 2006). *R. dominica* is a secondary grain pest, in which the first instar larvae have been observed to enter grain through the intact kernel (Arthur, 1999) and cannot be removed from the grain through normal cleaning procedures (Flinn, 1998).

Number of organophosphorous (OP) insecticides have been and are being used against stored grains pests which have resulted in the development of severe resistance in many insect species including lesser grain borer (Champ and Dyte, 1977; Haliscak and Beeman, 1983; Zettler and Cuperus, 1990; Khan *et al*., 1995; Kotze and Wallbank, 1996; Ma *et al*., 2004; Syed *et al*., 2005).

* Part of Ph.D. thesis of first author.

0030-9923/2007/0003-0179 $ 8.00/0

Stored grain pests including lesser grain borer has also developed resistance against number of other insecticides *e.g.*, chlorinated hydrocarbons *viz*., BHC/lindane (Bhatia and Pradhan, 1972), DDT (Lloyd, 1969), and certain fumigants *e.g.,* methyl bromide and phosphine (FAO, 1975; Emery *et al.*, 2003; Daglish, 2004), but cyanogen and allyl acetate are still very toxic fumigants to many stored grain insect pests including *R. dominica* (Hooper *et al*., 2003; Rajendran and Muralidharan, 2005). *R. dominica* in Brazil was also found to be resistant to deltamethrin (Lorini and Galley, 1999).

Due to development of resistance against OP and pyrethroids, a search for a new class of insecticides has been initiated to achieve successful control over stored grain insect pests including lesser grain borer (Cordova *et al.,* 2005; Ahmet and Gulay, 2005; Ferizli and Beris, 2005). On the other hand, efforts have been initiated in some labs to use mixtures of insecticides against resistant insects. Laboratory and field experiments in Australia have established the effectiveness of OP and pyrethroid combinations against beetle species that have developed resistance (Bengston *et al.*, 1980, 1983, 1987). Deltamethrin along with piperonyl butoxide (PBO) resulted in 89% reversal in deltamethrin resistance in just one generation in larvae of yellow fever mosquito, *Aedes aegypti*, which after 15 generations returned to the original level (Kumar *et al.*, 2002). Nayak *et al.* (2003) also reported similar effects of mixture of OP with carbaryl on three psocid species.

The objective of this study was to determine the toxicity of mixtures of malathion and deltamethrin in different ratios against *R. dominica* to identify the most effective dosage for the control of this pest.

MATERIALS AND METHODS

Beetle culture and its maintenance

Six strains of *R. dominica* were collected from godowns of different regions of Pakistan like Chichawatni (C), Karachi (K), Wazirabad (W), Sialkot (S), Lahore (L), and Multan (M) in the form of original cultures. These strains were maintained in sterilized jam bottles covered with muslin cloth in a culture room at 30±2°C with relative humidity of 65±5%. Beetles were fed wheat after fumigation with phosphine for 24 hours. Newly hatched adult beetles collected 43±2 days after egg laying were used in the present study (Anonymous, 1974).

Insecticides

Technical grades of malathion [diethyl (dimethoxyphosphinothioylthio) succinate] 57EC and deltamethrin [(S)-α-cyano-3-phenoxybenzyl ester; decis; K-othrine], 25mg/kg were obtained from the Agricultural Chemical Group of FMC Corporation, Lahore, Pakistan.

Estimation of LC_{50}

Using serial dilutions of malathion, deltamethrin and their mixtures in acetone, LC_{50} was determined. Different concentrations of insecticides alone and their combinations in triplicate were applied to glass Petri plates (dia. mm, 130 cm^2) by residual film method. These concentrations were prepared by dissolving calculated amount of insecticides in 10 ml of acetone.

Two sets of mixtures of malathion and deltamethrin were prepared (Table I). In mixtures of set I (1-5) concentration of malathion was kept constant, whereas, concentration of deltamethrin was changed. In set II (6-8) combinations, concentration of malathion was varied, whereas, concentration of deltamethrin was kept constant.

Table I.- Composition of mixtures of malathion and deltamethrin used in the study for determination of LC_{50} against *Rhyzopertha dominica.*

Mixture No.	Ratio of mixtures	Quantities mixed malathion+deltamethrin (g)
1	10:0.5	40 + 2
2	10:1.0	40 + 4
3	10:1.5	40 + 6
4	10:2.0	40 + 8
5	10:2.5	40 + 10
6	9.5:2.0	38 + 8
7	9.0:2.0	36 + 8
8	8.5:2.0	34 + 8

About 9 serial dilutions *viz*., 256 ppm, 128 ppm, 64 ppm, 32 ppm, 16 ppm, 8 ppm, 4 ppm, 2 ppm and 1 ppm were made to estimate LC_{50} of each insecticide and their mixtures. 1.3 ml of these dilutions were applied in the centre of glass Petri plates (in triplicate) with the help of glass pipette. To spread the insecticides uniformly the Petri plates were rotated manually. The acetone was allowed to evaporate after which ten healthy beetles were placed in each Petri plate and covered. Three control Petri plates were also prepared for each set of mixtures and individual insecticides. The control plates contained acetone only with 10 healthy adult beetles. Beetles were checked by using camel hair brush for mortality after 48 hours. They were considered dead if on touching with brush they did not show any movement. Lloyd method (1969) was used for counting the mortality. Lc_{50} was calculated by computerized probit analysis (Finney, 1971).

RESULTS AND DISCUSSION

Toxicity of malathion

Table II shows the toxicity of malathion and deltamethrin alone and in combination, on six different strains of *R. dominica*. C strain was found to be the most resistant (LC_{50} 115.5 ppm) and M strains the least resistant (LC_{50} 12.40 ppm) against malathion. The gradation followed by different strains C > W > K > L > S > M. Researches in the past shows that number of OP including malathion resulted in the development of severe resistance in many insect species including lesser grain borer). (Badmin, 1990; Rossiter *et al*., 2001; Li *et al*., 2005). Only freshly prepared malathion 2ppm-8ppm produced 100% mortality in adults of hairy fungus beetle, *Typhae stercorea* (Tigar and Pinniger, 1996). Guedes *et al*., 1996, reported the failure of many other OP protectants *e.g.*, chlorpyrifos-methyl and primiphos-methyl to control the Brazilian and U.S. populations of *R. dominica*.

Table II.- Comparison of the LC_{50} (in ppm) of malathion and deltamethrin administered alone and in mixtures in different ratios against different strains of *Rhyzopertha dominica*.

Insecticides (Dose)	Strains of *Rhyzopertha dominica*					
	Chichawatni (C)	Karachi (K)	Wazirabad (W)	Sialkot (S)	Lahore (L)	Multan (M)
Malathion	115.50	71.91	90.39	28.32	47.18	12.40
Deltamethrin	5.00	5.17	6.25	8.35	2.83	10.55
Malathion:Deltamethrin Mix. (10:0.5)	12.08	18.80	17.38	12.29	15.35	16.31
Malathion:Deltamethrin Mix. (10:1)	24.94	4.71	8.95	5.33	4.53	8.38
Malathion:Deltamethrin Mix. (10:1.5)	6.32	8.74	4.52	7.29	5.08	8.25
Malathion:Deltamethrin Mix. (10:2)	5.40	11.81	10.61	8.98	19.05	15.83
Malathion:Deltamethrin Mix. (10:2.5)	14.52	15.98	8.24	16.50	9.64	19.61
Malathion:Deltamethrin Mix. (9.5:2)	9.77	52.10	9.89	13.24	28.88	11.39
Malathion:Deltamethrin Mix. (9:2)	6.09	57.42	3.13	61.53	3.41	5.27
Malathion:Deltamethrin Mix. (8.5:2)	23.27	31.76	9.99	43.75	27.03	7.80

Toxicity of deltamethrin

Strain M of *R. dominica* was found to be most resistant (LC_{50} 10.55 ppm) and L strain was most sensitive (LC_{50} 2.83 ppm) to deltamethrin. The gradation followed by different strains was M > S > W > K > C > L. Comparatively deltamethrin was more toxic when its LC_{50} values are compared with those of malathion (115.5 and 12.4ppm). Arthur (1997) has also reported high toxicity of deltamethrin dust (0.05%) to *R. dominica* and *T. castaneum*. It is found more efficient with 0.25ppm against *S. oryzae* than other pyrethroids (Athanassiou *et al*., 2004). High sensitivity to pyrethroids, deltamethrin and esfenvalerate, has also been determined in aquatic insects larvae (Beketov, 2004). Deltamethrin and cypermethrin have been shown to be inefficient in mosquito control (Enayati and Hemingway, 2006). Development of severe resistance to pyrethroids including deltamethrin has been reported in granary weevil *S. granarius* (L.) (Curculionidae) adults (KIjajic and Peric, 2006).

Toxicity of mixtures of malathion and deltamethrin

Table II also shows the effect of different mixtures of two insecticides (malathion and deltamethrin) on lesser grain borer. Mixture 10:0.5 was found to be quite ineffective to all tested strains as its LC_{50} values are quite high (12.08-18.80 ppm) when compared with those of deltamethrin alone (2.83-10.55 ppm). The order of gradation of resistance shown by all the strains for this mixture was K > W > M > L > S > C.

For mixture 10:1 the different strains followed the gradation C>W>M>S>K>L. This mixture is effective for K, S and M strain with LC_{50} values 4.71, 5.33 and 8.38 ppm, respectively. For mixture 10:1.5 it was K>M>S>C>L>W. It is effective for W and M strain with LC_{50} values 4.52 and 8.25 ppm, respectively. For mixture 10:2 it was L>M>K>W>S>C with LC_{50} values ranging from 5.40-19.05 ppm. It is ineffective for each of the six strains. For mixture 10:2.5 it was M>S>K>C>L>W with LC_{50} values ranging from 8.24 -19.61 ppm. It is ineffective, too. Therefore, the order of effectiveness of the set I combinations mixtures is

10:1 > mixture 10:1.5 > mixture 10:0.5 = mixture 10:2 = mixture 10:2.5.

Treating the six strains of lesser grain borer with set II combinations the mixture 9.5:2 was quite ineffective with LC_{50} values ranging between 9.77-52.10 ppm. Mixture 9:2 was very effective for W and M strain with LC_{50} values of 3.13 and 5.27 ppm, respectively. Mixture 8.5:2 was effective for only M strain (LC_{50} 7.80 ppm). The order of effectiveness of the set II combinations is mixture 9:2 > mixture 8.5:2 > mixture 9.5:2.

Of the first set of combinations, mixture 10:1 is effective for K, S and M strains. Mixture 10:1.5 and mixture 9:2 from set II combinations with comparatively low LC_{50} values proved to be the most effective for the W and M strains of the lesser grain borer.

The results of the present study is in accordance with the findings of Shakoori and Saleem (1989) on *Tribolium castaneum* and by Ali *et al*. (2003) on *R. dominica* adults, who concluded that OP in combination with pyrethroid is much more effective than when administered alone. In some other studies, pyrethroid (resmethrin and bioresmethrin) combined with chlorpyrifos-methyl (OP) and cyfluthrin (pyrethroid) plus piperonyl butoxide plus chlorpyrifos-methyl proved very effective against lesser grain borer (Arthur, 1992, 1994).

Considering all the effective combinations together, the efficacy of the mixtures in descending order for K and S strains is: mixture 10:1 > mixture 10:1.5 > mixture 9:2 for W and M strain, mixture 9:2 > mixture 10:1.5 > mixture 10:1.

Previously the effectiveness of organophosphate-pyrethroid combinations against *R. dominica and Sitophilus oryzae* has also been reported by Pereira *et al*. (1997) and Pinto *et al*. (1997) where a mixture of fenitrothion, deltamethrin and piperonyl butoxide caused 100% mortality in these beetles. The mixture of bifenthrin, PBO and chlorpyrifos prevented *R. dominica* and other beetles from producing live progeny (Daglish *et al.*, 2003). Ahmad (2004) also established the effectiveness of OP-pyrethroid combinations against *Helicoverpa armigera*. According to Haung and Subramanyam (2005) a mixture of primiphos-methyl (OP) and synergized pyrethrins was not superior to primiphos-methyl alone against the five insect pests including *R. dominica.*

Overall results of this study conclude that OP-pyrethroid mixtures have more powerful and beneficial control over this beetle than when these insecticides are used alone. Furthermore, mixture 10:1, mixture 10:1.5 and mixture 9:2 with comparatively low LC_{50} values were found to be the most effective against *R. dominica.* Therefore, controlling resistant insects with insecticide mixtures is strongly recommended.

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(Received 19 December 2006, revised 10 February 2007)