Toxicity of Some Commonly Used Synthetic Insecticides Against Spodoptera exigua (Fab) (Lepidoptera: Noctuidae)

Qamar Saeed¹, Mushtaq A. Saleem^{1*} and Munir Ahmad²

¹Department of Crop Protection, University College of Agriculture, B. Z. University Multan, Pakistan ²Department of Entomology, Pir Mehr Ali Shah Arid Agriculture University, Murree Road, Rawalpindi, Pakistan

Abstract. Spodoptera exigua (Lepidoptera: Noctuidae) is one of the major pests of many crops in Pakistan. Effectiveness of different insecticides was evaluated against field populations of 2^{nd} instar larvae of *S. exigua* under laboratory conditions because in lepidopterous pests resistance is developed in 3^{rd} instars. Bioassays were performed through leaf dip method to evaluate the dose- and time-mortality response for emamectin benzoate, lufenuron, chlorpyrifos and cypermethrin. Significant variation was revealed in lethal concentration and lethal time values. Among all the tested insecticides emamectin benzoate gave the lowest LC₅₀ value *i.e.*, 0.005 mg/l (95% FL: 0.004–0.007 mg/l) followed by lufenuron *i.e.*, LC₅₀ value 0.65 mg/l (95% FL: 0.38–0.93 mg/l). While there was a non significant difference in the LC₅₀ values of cypermethrin and chlorpyrifos *i.e.*, 146 mg/l (95% FL: 108–188 mg/l) and 175 mg/l (95% FL: 113–256 mg/l), respectively. LT₅₀ values showed that cypermethrin was more effective compared to chlorpyrifos; however, both required less than half the time required by emamectin to kill 50% population of 2^{nd} instar larvae of *S. exigua*. Lufenuron, however, required more time as compared to all tested insecticides. The order of effectiveness in terms of LT₅₀ values was; cypermethrin > chlorpyriphos > emamectin benzoate > lufenuron. Responsiveness of *S. exigua* larvae provides important information with dose- and time-mortality for selection of insecticides in field for better pest management.

Key words: Spodoptera exigua, insecticides, dose-mortality, time-mortality.

INTRODUCTION

Xenobiotics like insecticides are important constituent of our daily life from preservation of food materials to insect pest management. These xenobiotics have beneficial as well as deleterious effects on our food, air, water and surrounding environment. Hazardous effects like decreased egg hatching have been observed not only in insect pests like *Spodoptera exigua* and *Spodoptera frugiperda* but also the predatory beetle like *Poecilus cupreus* L. and *Hippodamia convergens* (Adamski *et al.*, 2009; Alvarez *et al.*, 2009a,b; Antwi and Peterson, 2009). These chemicals are also the constituents of plants on which these insects feed and are ultimately affected in terms of development and reproduction etc (Adamski *et al.*, 2005).

Insecticides are important tool for insect pest management. Determination of their effective dose and time mortality factors are important for spray systems. Selection of *S. exigua* with conventional insecticides has been reported in different parts of the world (Meinke and Ware, 1978; Chaufaux and Ferron, 1986; Yoshida and Parrella, 1987; Brewer and Trumble, 1989). Exposure to these insecticides increase the larval survival and delimits their effective use (Kim *et al.*, 1998). Similarly, resistance to conventional and some new chemistry insecticides have also been reported, hindering the proper pest management (Perez *et al.*, 2002; Ahmad *et al.*, 2009).

S. exigua is a serious insect pest many crops in Southeast Asian region. Being polyphagous, strong migratory capabilities and wide distribution (Metcalf and Flint, 1962; Aarvik, 1981; Stewart *et al.*, 2002), it can infest large areas of field crops and horticultural cultivations. (Mitchell, 1979; Han *et al.*, 2008). It has been recorded from more than 50 genera of over ten plant families (Smits *et al.*, 1987). Although its outbreaks are sporadic yet its population develops rapidly because of inefficient management at late larval stages. Its short life span and multivoltine nature makes its control rather difficult if not monitored regularly. Effective doses and time related factor are important for selection of

^{*} Corresponding author: mushtaqasaleem@hotmail.com 0030-9923/2012/0005-1197 \$ 8.00/0 Copyright 2012 Zoological Society of Pakistan

insecticides depending on severity of infestation. So the present study was conducted to determine the lethal/ effective dose and lethal time of commonly used insecticides. Some commonly used insecticides were used in this experiment to evaluate susceptibility and time trends in mortality against S. *exigua* under laboratory conditions.

MATERIALS AND METHODS

Insect collection and rearing

S. exigua egg batches were collected from cauliflowers in Multan during 2010 and transported in well-ventilated plastic box at the Post Graduate Laboratory in the Department of Agricultural Entomology, University College of Agriculture, Bahauddin Zakariya University Multan, Pakistan. The hatched larvae were further reared on castor (Ricinus communis) leaves to complete their life cycle. Castor leaves were collected at morning time, washed thoroughly with tap water and air-dried. Castor leaves were changed daily till the pupation started. Pupae were kept individually in separate Petri dishes lined with filter papers. Newly emerged adults were placed in cylindrical transparent plastic jars (4 kg capacity) having cotton balls soaked in 10% honey solution as adult diet. Nappy liner strips of about 1-2 inch wide and 6-7 inch long were suspended with the help of rubber bands in the jars as egg substrate and were collected every 24 h and kept in batches separately in plastic jars. Second instar larvae of F₁ laboratory generations were used for bioassays throughout the present experimentations.

Insecticides

Commercial formulations different of insecticides used for bioassays comprised emamectin benzoate (Proclaim® 1.9EC, Syngenta, Pakistan), lufenuron (Match® 05EC, Syngenta, Pakistan), chlorpyrifos (Lorsban® 40EC; Dow AgroSciences, Pakistan), cypermethrin (Arrivo® 10EC, FMC, Pakistan) A non-ionic surfactant (Stapple®Dupont, Pakistan) was added @ 5 mg ml⁻¹ to each insecticide to enhance their adhesiveness to leaf surface.

Bioassays for LC_{50} and LT_{50}

Bioassays were conducted on 2nd instar larvae of S. exigua (3-6 h after moulting) using a standard leaf disc bioassay method (Sayyed et al., 2000; Ahmad et al., 2009). Castor leaves were collected from unsprayed plants, washed and air-dried and made 5 cm diameter leaf discs with the help of a leaf cutter. Stock solution of each tested insecticide was made from the formulation available with different concentrations (emamectin benzoate (Proclaim® 1.9EC, Syngenta, Pakistan), lufenuron (Match® 05EC, Syngenta, Pakistan), chlorpyrifos (Lorsban® 40EC; Dow AgroSciences, Pakistan), cypermethrin (Arrivo® 10EC, FMC, Pakistan)) and then 6-8 serial concentrations (chlorpyriphos: 1024–32ppm, cypermethrin: 1024-32 ppm, lufenuron: 0.13-2.0ppm, emmemectin benzoate: 0.03-0.001875ppm) were prepared by calculation the field doses from the available insecticides in the market. Leaf discs were dipped for 10 s in a test solution and allowed to dry at ambient temperature for about 20-30 min in a fume hood. Test solutions were prepared fresh in distilled water and then Stapple® (5 mg ml⁻¹) was added as surfactant. Leaf discs immersed in distilled water were labeled as control. Air-dried leaf discs were then placed in individual plastic Petri dishes (5 cm diameter) containing moistened filter paper. Each treatment (concentration) including controls were replicated eight times. Five 2nd instar S. exigua larvae were placed on each leaf disc and thus total numbers of tested larvae per concentration were 40. The bioassays were kept at a temperature of 28±2°C, 50-60±5% relative humidity and 14:10 (light-dark) photoperiod. Larval mortality at the end point was assessed after 48 and 72 h of insecticidal exposure.

Time-mortality data for the tested insecticides were observed at 12 h interval at their respective LC_{50} levels. Time taken for 50% mortality was used to measure lethal time (LC_{50}) values.

Data analysis

Mortality as endpoint was corrected for analysis by Abbott's formula using POLO-PC software (LeOra Software, 1987). Fiducial limits at 95% interval were used for differentiation of significance levels (Litchfield and Wilcoxon, 1949).

RESULTS

 LC_{50}

All the insecticides tested in the present studies caused concentration–dependent mortality in 2^{nd} instar larvae of field population of *S. exigua*. The slopes of regression lines and the lethal concentration values at 50% kill were significantly different from each other. Slopes of cypermethrin and chlorpyrifos were non-significant to each other due to overlapping of their regression lines (Table I). The LC₅₀ values of lufenuron and emamectin were, however, significant not only from each other but also from cypermethrin and chlorpyrifos (Table I). Their slope values were also steeper than cypermethrin but less than that of chlorpyrifos.

LT_{50}

The time-mortality studies for the four tested insecticides at their respective LC_{50} values were performed. Cypermethrin required the least time to kill the 50% population followed by chlorpyrifos and emamectin, respectively. Lufenuron, however, required the maximum time of 32 h to kill 50% exposed insects (Table II). Fiducial values of all the insecticides were non-significant to each other; however, it was least in cypermethrin making it the most effective. There existed steeper slopes in cypermethrin and chlorpyrifos making them both as effective as other insecticides (Table II).

DISCUSSION

Similar mode of action as nerve toxicity can be attributed for similar results with respect to the lethal time factor. Cypermethrin, chlorpyrifos and emamectin benzoate being nerve poisons have similar modes of action as effectors of nerve impulses with rapid action against insect pests. These insecticides have been found effective in terms of dose and time factor against leaf worm, *S. litura* (Ahmad *et al.*, 2006) like that of *S. exigua*. No cross resistance was observed between methomyl and chlorpyrifos against *S. exigua* suggesting the effectiveness of organophosphates as well as carbamates (Argentine *et al.*, 2002). Byrne and Toscano (2001) suggested target site insensitivity as major mechanism of resistance to methomyl. However, they observed insensitive acetylcholine esterase activity against a number of organophosphates like profenofos, chlorpyrifos and sulprofos (Byrne and Toscano, 2002). Increase in mortality rate with prolonged larval duration, delay in pupal formation and adult emerge were significant when S. frugiperda larvae were provided diet incorporated with insect growth inhibitors as flavinoids. Variation may exist due to stage, feeding pattern and requirement and behaviour of the insect with respect to particular insecticide. However, these factors were not considered as endpoint during this study.

Table I	Probit analysis of dose-mortality data for
	different insecticides against 2 nd instar larvae
	of Spodoptera exigua field population

Insecticide	LC ₅₀ FL at 95% (mg/l) level ^a		Slope <u>+</u> SE ^b	χ^2	
Cypermethrin	146	108-188	2.49±0.39	2.01	
Chlorpyrifos	175	113-256	1.50 <u>±</u> 0.17	6.15	
Lufenuron	0.65	0.38-0.93	1.84 <u>±</u> 0.40	2.90	
Emamectin benzoate	0.005	0.004-0.007	1.74 <u>+</u> 0.24	1.69	

^a FL, fiducial	l limits at 95%	confidence	level; SE,	Standard error.
---------------------------	-----------------	------------	------------	-----------------

Table II.-Probit analysis of time-mortality data for
different insecticides against 2nd instar larvae
of Spodoptera exigua field population at their
respective LC₅₀

Insecticide	LC ₅₀ (mg/l)	FL at 95% level ^a	Slope <u>±</u> SE ^b	χ^2
cypermethrin	7.20	2.43-12.9	1.44 <u>+</u> 0.16	13.4
chlorpyrifos	14.7	9.54-22.3	1.75 <u>+</u> 0.16	8.77
lufenuron	32.3	20.8-56.5	2.54 <u>+</u> 0.23	16.1
emamectin benzoate	26.0	23.2-8.9	4.23 <u>+</u> 0.38	1.49

^aFL, fiducial limits at 95% confidence level; ^bSE, Standard error.

Emamectin benzoate, a derivative of abamectin, is quite effective against a number of lepidopteran insect pests including *S. exigua* and *S. litura* (Ahmad *et al.*, 2006). This insecticide is not only photostable but also has translaminar efficacy with lack of cross-resistance with many commercial

insecticides (Mrozik, 1994; White *et al.*, 1997). Its foliar application as insecticide was observed many folds toxic as compared to diet incorporation against important lepidopteron insect pests including *S. exigua* (Argentine *et al.*, 2002). However, the results of LC₅₀ values and LT₅₀ values are species specific and need to be performed for *S. exigua* (Thomsen and Eilenberg, 2000). Lack of resistance has been observed for this insecticide against different insect pests worldwide which stresses its incorporation into resistance management programs (Shad *et al.*, 2010).

The discovery of insect growth regulators in the last two centuries has created a new class of insecticides for their effective control (Wing, 1988; Wing et al., 1988). Factors like lack of larval molting to next instar, cessation of feeding and slow movements followed by ultimate death has been observed due to their acute dose application in certain insect pests (Wing et al., 1988; Smagghe and Degheele, 1994a). Lufenuron required maximum time of 32 h to kill 50% population which was due to its mode of action through ingestion and affecting the physiological processes. However, disorders in oogenesis and spermatogenesis have also been main features at their chronic dose rates (Smagghe and Degheele, 1994b). These results are very similar to those of the xenobionts which affect the hatching success of S. exigua eggs (Adamski et al., 2009).

Integrated pest management approach combining insecticides with regular field monitoring of their respective resistance levels are important not only to keep these insecticides for their long term usage but also for regulation of insecticide registration with respect to a particular insect pest species (Thomsen and Eilberg, 2000; Perez et al., 2002). Presence of cross-resistance among different insecticide groups with different mode of action also requires regular monitoring programs (Ahmad et al., 2006; Ahmad et al., 2009). Incorporation of new chemistry insecticides especially for the pests like S. exigua of vegetables need safer insecticides like emamectin benzoate with least phytotoxic effects with efficient control of the inset pests (Clarke and Fleischer, 2003).

This study provides basic information regarding the concentration and time for *S. exigua* mortality under laboratory conditions. Field studies

can foster the effectiveness of these and other insecticides for long term and effective management of *S. exigua*.

ACKNOWLEDGEMENTS

The first author is thankful to the Higher Education Commission for financing fellowship for his PhD studies under the supervision of Prof. Mushtaq A. Saleem and Bahauddin Zakariya University Multan for providing laboratory facilities to conduct the experiments.

REFERENCES

- AARVIK, L., 1981. The migrant moth, *Spodoptera exigua* Hubner (Lepidoptera: Noctuidae) recorded in Norway. *Fauna Norvegicab*, **28**: 90-92.
- ADAMSKI, Z., ZIEMNICKI, K., MARCINIAK, P., HALAMUNDA, J., NAWROCKA, M. F., LELARIO, L.S. AND BUFO, S.A., 2009. Effect of various xenobiotics on hatching success of *Spodoptera exigua* eggs as compared to a natural plant extract. *J. Toxicol. environ. Hlth.*, A. **72**: 1132-1134.
- ADAMSKI, Z., NIEWADZI, M. AND ZIEMNICKI, K., 2005. Inheritance of chorionic malformations and insecticide resistance by *Spodoptera exigua*. J. appl. Ent., 129:526–533.
- AHMAD, M., SALEEM, M. A., AHMAD, M. AND SAYYED, A. H., 2006. Time trend in mortality for conventional and new chemistry insecticides against leafworm, *Spodoptera* litura (Lepidoptera: Noctuidae). *Pakistan J. biol. Sci.*, **9**: 360-364.
- AHMAD, M., SALEEM, M.A. AND SAYYED, A.H., 2009. Efficacy of insecticide mixtures against pyrethroid and organophosphate resistant population of *Spodoptera litura* (Lepidoptera: Noctuidae). *Pest Manage. Sci.*, 65: 266-274.
- ALVAREZ, A. F., ORTEGO, F. AND CASTANERA, P., 2009b. Bt maize fed-prey mediated effect on fitness and digestive physiology of the ground predator Poecilus cupreus L. (Coleoptera: Carabidae). J. Insect Physiol., 55: 143-149.
- ALVAREZ, A., PERA, L. M. LOTO, F., VIRLA, E. G. AND BAIGORI, M.D., 2009a. Insecticidal crystal proteins from native *Bacillus thuringiensis*: numerical analysis and biological activity against *Spodoptera* frugiperda. *Biotechnol. Lett.*, **31**: 77-82.
- ANTWI, F.B. AND PETERSON, R.K., 2009. Toxicity of deltaphenothrin and resmethrin to non-target insects. *Pest Manag. Sci.*, 65: 300-305.
- ARGENTINE, J. A., JANSSON, R. K., STARNER, V. R. AND HALLIDAY, W. R., 2002. Toxicities of emamectin

benzoate homologues and photodegradates to Lepidoptera. *J. econ. Ent.*, **95**: 1185-1189.

- BREWER, M. J. AND TRUMBLE, J. T., 1989. Field monitoring of resistance in beet armyworm (Ledpidoptera: Noctuidae). J. econ. Ent., 82: 1520-1526.
- BYRNE, F. J. AND TOSCANO, N.C., 2001. An insensitive acetyl cholinesterase confers resistance to methomyl in the beet armyworm *Spodoptera exigua* (Lepidoptera: Noctuidae). *J. econ. Ent.*, **94**: 524-528.
- CHAUFAUX, J. AND FERRON, P., 1986. SensibiliLe differenle de deux populations de *Spodoptera exigua* Hubner. (Lepidoptera: Noctuidae) aux baculovirus ct aux pyrelhrinoides de synthesc. *Agronomie*, **6**: 99-104.
- CLARKE, H. D. AND FLEISCHER, S. J., 2003. Sequential sampling and biorational chemistries for management of lepidopteran pests of vegetable amaranth in the Caribbean. *J. econ. Ent.*, **96**: 798-804.
- HAN, R.D., PARAJULEE, M., HE, Z. AND GE, F., 2008. Effects of environmental humidity on the survival and development of pine caterpillars, *Dendrolimus tabulaeformis* (Lepidoptera: Lasiocampidae). *Insect Sci.*, **15**:147–152.
- KIM, Y., POWELL, E. N., WADE, T. L., PRESLEY AND SERICANO, J., 1998. Parasites of sentinel bivalves in the NOAA Status and Trends Program: distribution and relationship to contaminant body burden. *Mar. Poll. Bull.*, 37:45-55.
- LEORA SOFTWARE. 1987. POLO-PC. A user's guide to Probit Or LOgit analysis. LeOra Software Inc., Berkeley, California, pp. 22.
- LITCHFIELD, J.T. AND WILCOXON, F., 1949. A simplified method of evaluating dose-effect experiments. J. Pharmacol. exp. Ther., **99**: 99-103.
- MEINKE, L. AND WARE, G.W., 1978. Tolerance of three beet armyworm strains in Arizona to methomyl. *J. econ. Ent.*, **71**: 645-646
- METCALF, C. L. AND FLINT, W.P., 1962. *Destructive and useful insects.* 4th edition (revised). McGraw Hill, New York, USA
- MITCHELL, E. R., 1979. Monitoring adult populations of the fall armyworm (Lepidoptera: Noctuidae). *Flor. Entomol.*, **62**: 91–98.
- MROZIK, H., 1994. Advances in research and development of avermectins. *Am. chem. Soc. Symp.*, **551**: 54-73.
- PEREZ, C. J., ALVARADO, P., NARVAEZ, C., MIRANDA, F., HERNANDEZ, L., VANEGAS, H., HRUSKA, A. AND SHELTON, A.M., 2002. Assessment of insecticide resistance in five insect pests attacking field and vegetable crops in Nicaragua. J. econ. Ent., 93: 1779-1787.

- SAYYED, A. H., HOWARD, R., HERRERO, S., FERRE, J. AND WRIGHT, D. J., 2000. Genetic and biochemical approach for characterization of resistance to *Bacillus thuringiensis* toxin Cry1Ac in a field population of the diamondback moth. *Appl. environ. Microb.*, **66**: 1509– 1516.
- SHAD, S. A., SAYYED, A. H. AND SALEEM, M. A., 2010. Cross-resistance, mode of inheritance and stability of resistance to emamectin in *Spodoptera litura* (Lepidoptera: Noctuidae). *Pest Manag. Sci.*, **66**: 839– 846.
- SMAGGHE, G. AND DEGHEELE, D., 1994a. Action of the nonsteroidal ecydysteroid mimic RH-5849 on larval development and adult reproduction of insects of different orders. *Invert. Reprod. Develop.*, 25: 227-236.
- SMAGGHE, G. AND DEGHEELE, D., 1994b. Action of a novel nonsteroidal ecdysteroid mimic, tebufenozide (RH-5992), on insects of different orders. *Pestic. Sci.*, 42: 85-92.
- SMITS, P. H., VAN VELDEN, M. C., VAN DEVRIE, M. AND VLAK, J. M., 1987. Feeding and dispersion of *Spodoptera exigua* larvae and its relevance for control with a nuclear polyhedrosis virus. *Ent. Ex. Appl.*, 43: 67–72.
- STEWART, R., FRANCIS, C.M. AND MASSEY, C., 2002. Age-related differential timing of spring migration within sexes in Passerines. *Wilson Bull.*, **114**:264–271.
- THOMSEN, L. AND EILENBERG, J., 2000. Timeconcentration mortality of *Pieris brassicae* (Lepidoptera: Noctuidae) and *Agrotis segetum* (Lepidoptera: Noctuidae) larvae from different destruxins. *Environ. Ent.*, **29**: 1041-1047.
- WHITE, S.M., DUNBAR, D.M., BROWN, R., CARTWRIGHT, B., COX, D., ECKEL, C., JANSSON, R.K., MOOKERJEE, P.K., NORTON, J.A., PETERSON, R.F. AND STARNER, V.R., 1997. Emamectin benzoate: a novel derivative for control of lepidopterous pests in cotton. In: *Beltwide Cotton Conf.*, 2: 1078-1082. New Orleans,
- WING, K.D., 1988. RH-5849, a nonsteroidal ecdysone agonist: Effects on a *Drosophila* cell line. *Science*, 241: 467-469.
- WING, K. D., SLAWECKI, R. A. AND CARLSON, G. R., 1988. RH-5849, a nonsteroidal ecdysone agonist: Effects on larval Lepidoptera. *Science*, 241: 470-472.
- YOSHIDA, H. A. AND PARRELLA, M.P., 1987. The beet armyworm in ßoricultural crops. *Calif. Agric.*, **41**: 13-15.

(Received 23 January 2011, revised 3 July 2012)