INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY
1560–8530/2005/07–5–768–772
http://www.ijab.org

Toxicity of Some Insecticides with Novel Modes of Action Against Malathion–Resistant and Organophosphate–Susceptible Strains of *Tribolium castaneum* Larvae

RIAZ HUSSAIN[1], MUHAMMAD ASHFAQ, MUSHTAQ A. SALEEM† AND SOHAIL AHMED
Ecotoxicology Lab., Department of Agricultural Entomology, University of Agriculture, Faisalabad–38040, Pakistan
†University College of Agriculture, Bahauddin Zakariyya University, Multan, Pakistan
[1]Corresponding author's e–mail: riahuss@excite.com

ABSTRACT

The LC_{50} values for malathion–resistant (PAK) and organophosphate–susceptible (FSS–II) strains of red flour beetle, *Tribolium castaneum* larvae were determined through residual film/filter paper impregnated method against six insecticides with novel modes of action such as abamectin (Sure 1.8 EC), spinosad (Tracer 240 SC), indoxacarb (Steward 150 SC), azadirachtin (Nimbokil 60 EC), buprofezin, and polychlorinated petroleum hydrocarbon (Tenekil 100 EC). The LC_{50} values of these insecticides were worked out as 178 and 261 ppm for abamectin, 6020 and 7455 ppm for spinosad, 4071 and 2154 ppm for indoxacarb, 20025 and 16405 ppm for azadirachtin, 9351 and 12450 ppm for buprofezin 25 WP, and 9933 and 6286 ppm for Tenekil 100 EC against PAK and FSS-II strains, respectively. The results revealed that abamectin was the most toxic of all insecticides tested in this study followed by indoxacarb, spinosad, buprofezin, Tenekil 100 EC and azadirachtin. Furthermore, abamectin, spinosad, and buprofezin proved more toxic to larvae of PAK strain, while indoxacarb, azadirachtin, and Tenekil 100EC to larvae of FSS-II strain.

Key Words: Insecticides; Toxicity; *Tribolium;* Abamectin; Spinosad; Indoxacarb; Azadirachtin

INTRODUCTION

To prevent losses from insect feeding in grain bins, a sound integrated pest management program involves: (i) grain bin and harvest equipment sanitation, (ii) proper harvest equipment adjustment, (iii) grain cleaning, (iv) uniform distribution of the grain during uploading into the bin, (v) moisture management, (vi) temperature management, (vii) periodic grain monitoring, (viii) residual insecticide treatment for long–term storage, and (ix) fumigation as a last resort. If the first eight tactics are used effectively, fumigation will be needed only rarely.

Currently, the measures taken to control the pest infestation in grains rely heavily upon the use of insecticides, which pose possible health hazards (to warm–blooded animals) and a risk of environmental contamination. Economic entomologists throughout the world spend a great deal of time and effort in attempt to determine the presence of beetles in stored grains, check their infestations and design better and safer methods to bring them under control. Control measures of different nature are being adopted at farm, market, and public sector storage that consist of use of native or natural methods of control by plant materials, and / or contact insecticides and fumigants. Chemicals currently used around the world to treat stored grains include chlorpyrifos methyl, lindane, malathion, pyperonyl butoxide plus pyrethrins (White & Leesch, 1995), pirimiphos methyl, methyl parathion, dichlorvos, diazinon (Lessard *et al*., 1998), some pyrethroids (Hutson, 1979; Hargreaves *et al*., 2000).

The development of insecticide resistance is widespread in stored grain insect pests including *Tribolium castaneum*. Various strains of *T. castaneum* had been reported to develop resistance against gamma–HCH (Dyte & Blackman, 1970; Kumar & Bhatia, 1982); malathion (Saleem & Shakoori, 1989; Zettler, 1991). There are other reports showing development of resistance in the said insect against fumigants, e.g., methyl bromide and phosphine (FAO, 1975; Nakakita & Winks, 1981; Mills, 1983; Tyler *et al.,* 1983), synthetic pyrethroids, e.g., cypermethrin, deltamethrin, cyfluthrin, fenvalerate and permethrin (Collins, 1990; Hasan *et al.,* 1996; Hussain *et al.,* 1996 a & b; Saleem *et al.,* 1998; Shakoori *et al.,* 1998) and some juvenile hormone analogues (Dyte, 1972).

Although *T. castaneum* is being controlled successfully with fumigants like phosphine in Pakistan (Rehman, 2000) yet keeping in view the threat of development of resistance against conventional insecticides as reviewed above, it was thought important to test some insecticides bearing novel modes of action, e.g., abamectin, spinosad, indoxacarb, azadirachtin and buprofezin and also polychlorinated petroleum hydrocarbon as it is dicovered by Pakistani scientists (STEDEC, 2004), against *T. castaneum* larvae.

MATERIALS AND METHODS

Insect strains. The malathion–resistant (PAK) and the organophosphate–susceptible (FSS–II) strains of *T. castaneum* were obtained from the Department of Zoology, University of the Punjab, Lahore–Pakistan. The sixth instar larvae of PAK strain have developed 40–fold resistance against malathion on the basis of comparison of LC_{50} of

FSS–II strain with that of PAK strain (Saleem & Shakoori, 1989).

The larvae of these two strains were reared in empty glass jam jars of 300 mL capacity. Whole meal wheat flour was used as the culture medium. The culture medium was also placed in an oven at 60°C for 3 h to provide maximum reduction of contamination. Both the strains were bred at 30±1°C and 60±5% relative humidity (Saleem & Shakoori, 1984). The new cultures were prepared every third day to obtain sufficient numbers of larvae for experimentation. Each bottle was filled 1/4th with sterilized wheat flour and about 50 adult beetles were introduced into each. The sixth instar larvae collected were used in insecticidal bioassays.

Insecticides used. Commercially available formulations of the following insecticides in Pakistan were used in the present study:

Group of insecticides	Brand name	Generic name	Name and address of manufacturer/distributor
Insecticides with natural origin	Sure 1.8 EC	Abamectin	Pan Pacific (Pvt.) Ltd., Vehari
	Tracer 240SC	Spinosad	SCL AgroSciences Pakistan (Pvt.) Ltd., Karachi
	Nimbokil 60EC	Azadirachtin	Scientific and Technological Development Corporation (STEDEC) of Pakistan, STEDEC House, Lahore
Oxadiazine	Steward 150 SC	Indoxacarb	DuPont Pakistan Operations (Pvt.) Ltd., Pakistan
Chitin synthesis inhibitor	Buprofezin 25 WP	Buprofezin	PakChina Chemicals, Lahore
Chlorinated hydrocarbon	Tenekil 100EC	Polychlorinated petroleum hydrocarbon	Scientific and Technological Development Corporation (STEDEC) of Pakistan, STEDEC House, Lahore

Determination of toxicities. Initial tests were conducted to check the dose response to five insecticides used in the present study. Based on these results, dose ranges were selected, which would be expected to yield LC_{50} values of the insecticides. For all experiments, calculated quantities of insecticides were dissolved in acetone.

The insecticide solutions were then diluted to give a geometric series of at least 3–5 different concentrations. The insecticide dilutions (1 ml/dish) of abamectin, spinosad, indoxacarb, and buprofezin were applied in the centre of glass Petri dishes (130 cm²) and then spread uniformly by gently rotating the dishes, while of azadirachtin, and polychlorinated petroleum hydrocarbon on filter paper akin to Petri dish. The control dishes had no insecticide, but only the acetone. Three replicates of each dose were used simultaneously for each insecticide. After evaporation of acetone at room temperature, 20 healthy sixth instar larvae were released in each dish. The larvae were kept without food, and mortality counts were made after 48 h exposure to insecticides and the percentage kill was corrected by Abbott's formula for any control mortality (Abbott, 1925). The criterion for death was the one described by Lloyd (1969) according to which "the insects were judged to be dead when the pressure from a brush failed to produce a response".

The mortality data so obtained were subjected to Probit Analysis. The LC_{50} values were calculated as outlined by Busvine (1971). Where there Chi–square (χ^2) values were found significant, the variances were adjusted for heterogeneity to have the good fit of regression lines.

RESULTS AND DISCUSSION

The LC_{50}, 95% fiducial limits, regression equations, regression lines and Chi–square (χ^2) of toxicities of some novel insecticides against larvae of malathion–resistant (PAK) and organophosphate–susceptible (FSS–II) strains of *Tribolium castaneum* after 48 h of treatment are presented in Table I and Figs. 1–2. The LC_{50} values of these insecticides for PAK and FSS-II strains were worked out as 178 and 261 ppm for abamectin, 6020 and 7455 ppm for spinosad, 4071 and 2154 ppm for indoxacarb, 20025 and 16405 ppm for azadirachtin, 9351 and 12450 ppm for buprofezin 25 WP, and 9933 and 6286 ppm for Tenekil 100 EC, respectively. The results revealed that abamectin was the most toxic of all insecticides tested in this study followed by indoxacarb, spinosad, buprofezin, Tenekil 100EC, and azadirachtin. Furthermore, abamectin, spinosad, and buprofezin proved more toxic to larvae of PAK strain and indoxacarb, azadirachtin, and Tenekil 100EC to larvae of FSS-II strain.

Some workers recorded resistance to abamectin in field populations of diamondback moth, *Plutella xylostella* (L) (Iqbal *et al.*, 1996; Iqbal & Wright, 1997; Sayyed *et al.*, 2004), when compared with a laboratory insecticide–susceptible population. Liang *et al.* (2003) reported little

Table I. The relative toxicities of some novel insecticides against malathion-resistant (PAK) and organophosphate-susceptible (FSS-II) strains of *Tribolium castaneum* (Herbst.) larvae

Insecticides	Strain	LC_{50}s (ppm)	95 % Fiducial limits	Slope ± SE	Regression equation	Chi–square (χ^2)
Abamectin (Sure 1.8EC)	PAK	178	146–216	2.02 ± 0.047	Y = 0.45 + 2.02 x	1.82
	FSS-II	261	223–323	2.05 ± 0.083	Y = 0.043 + 2.05 x	2.7
Spinosad (Tracer 240SC)	PAK	6020	4477–8109	1.77 ± 0.067	Y = –1.69 + 1.77 x	1.23
	FSS-II	7455	5445–10092	1.96 ± 0.070	Y = –2.59 + 1.96x	2.31
Indoxacarb (Steward 150SC)	PAK	4071	3265–5081	1.64 ± 0.055	Y = –0.92 + 1.64 x	1.56
	FSS-II	2154	1738–2629	1.80 ± 0.067	Y = –1.00 + 1.8 x	0.83
Azadirachtin (Nimbokil 60EC)	PAK	20025	16218–24774	1.89 ± 0.056	Y = –3.13 + 1.89 x	2.68
	FSS-II	16405	13365–20137	2.00 ± 0.059	Y = –3.43 + 2.00x	3.27
Polychlorinated Petroleum Hydrocarbon (Tenekil 100EC)	PAK	9933	6776–14554	3.088 ± 0.153	Y = –7.343 + 3.088 x	7.248
	FSS-II	6286	4808–8203	5.393 ± 0.223	Y =–15.485 + 5.393 x	6.84
Buprofezin 25WP	PAK	9351	6839–12793	1.27 ± 0.042	Y = 1.4421 + 0.9879 x	4.12
	FSS-II	12450	9418–16443	1.66 ± 0.081	Y = –0.40 + 1.32 x	2.93

Fig. 1. Regression lines of toxicities of some insecticides with novel mode of action against malathion-resistant (PAK) strain of *Tribolium castaneum* larvae

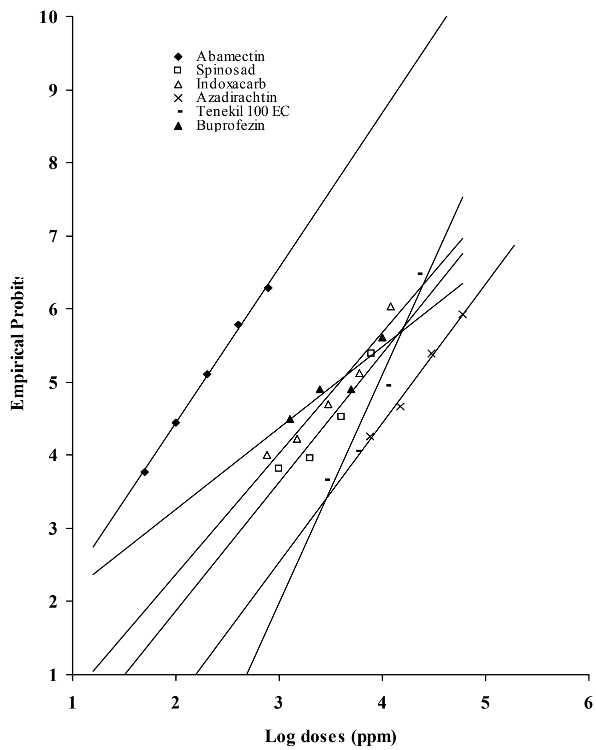
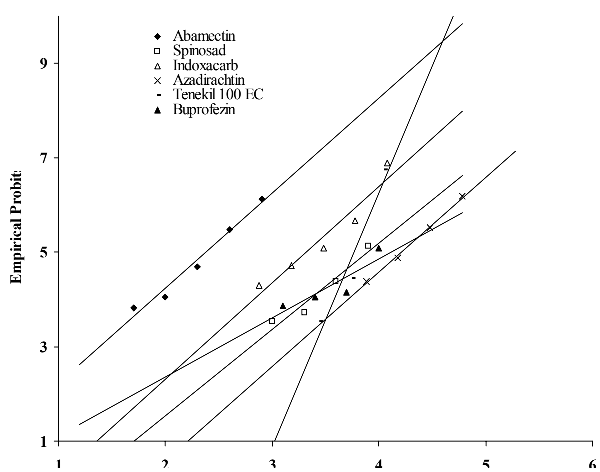


Fig. 2. Regression lines of toxicities of some insecticides with novel mode of action against organophosphate-susceptible (FSS-II) strain of *Tribolium castaneum* larvae



cross–resistance between abamectin and four pyrethroid insecticides (deltamethrin, beta–cypermethrin, fenvalerate and bifenthrin) in abamectin–resistant and abamectin–susceptible strains of the diamondback moth, *Plutella xylostella* (L). But in our experiments the larvae of malathion–resistant (PAK) strain remained more susceptible to abamectin.

The results of spinosad against larvae of *T. castaneum* are consistent to the findings of other workers against various insect pests. Spinosad remained effective against house flies as well as against its cyclodiene–resistant strain (Scott, 1998; Scott *et al.,* 2000); German cockroach, *Blatella germanica* (L.) (Wei *et al.,* 2001); most of the field populations of diamondback moth, *Plutella xylostella* (L.) (Zhao *et al.,* 2001); eggplant flea beetle, *Epitrix fuscula* Crotch (McLeod *et al*., 2002); *Sitophilus oryzae* (Toews & Subramanyam, 2003); *Oryzaephilus surinamensis* and *T. castaneum* on durum wheat (Fang *et al.,* 2002); *Tribolium* spp. on concrete surface (Toews *et al.,* 2003); and rice moth, *Corcyra cephalonica* (Huang & Subramanyam, 2004).

Spinosad proved more toxic to larvae of malathion-resistant strain (PAK) of *T. castaneum* in our studies, while in case of field populations of beet armyworm, *Spodoptera exigua*, it was found less toxic (Moulton *et al.,* 1999; 2000).

Indoxacarb proved effective against larvae of *T. castaneum*. Pasquier and Charmillot (2003) reported that the LC_{50} value was over 2800 mg kg^{-1} for indoxacarb administered by topical application to diapausing larvae from a susceptible codling moth strain. Studebaker and Kring (2003) stated that indoxacarb induced significantly higher mortality of laboratory–reared males, females and third instar nymphs of *Orius insidiosus* (Say), an important predator of several economic pests in cotton, with treated Petri dishes than on treated cotton plants in the field or greenhouse. Bostanian *et al.* (2004) state that indoxacarb is a novel oxadiazine pro–insecticide that has been found toxic to *Hyaliodes vitripennis* (Say), a predacious mirid (LC_{50}: 0.054 g AI L^{-1}) in several Quebec orchards where IPM programs are used. Tillman *et al.* (2001) studied the toxicity of a 145 g L^{-1} indoxacarb SC formulation (StewardTM) on the tarnished plant bug, *Lygus lineolaris,* and the big–eyed bug, *Geocori spunctipes*. Both insect species responded very similarly to indoxacarb in topical, tarsal contact and plant feeding toxicity studies. Prolonged tarsal contact with dry indoxacarb residues did not result in mortality for either insect species. Ahmad *et al.* (2002) found that the organophosphate–resistant Berrien strain of obliquebanded leafroller, *Choristoneura rosaceana*, exhibited a very high level of resistance (>700–fold) to indoxacarb, which has never been used in Michigan to control this insect pest. Our results showed that indoxacarb was less toxic to *T. castaneum* larvae of malathion–resistant (PAK) strain as compared to larvae of organophosphate–susceptible (FSS–II) strain on the basis of LC_{50} values.

According to various reports azadirachtin exhibited good efficacy against *Musca nobulo* L., *Culex fatigans*, *T. castaneum* and *Anthrenus flavipes* (Paul *et al.,* 1963); *Rhyzopertha dominica* (Muda & Cribb, 1999); pulse beetle (Jairaj, 1976); *S. oryzae, R. dominica, Sitotroga cerealella* (Savitri & Rao, 1976); *T. castaneum* (Tabassum *et al.,* 1994); *Corcyra cephalonica* Staint and a stored product insect pest, *Epilachna vigintioctopunctata* F. (Mathur &

Nigam, 1996); *Tribolium* spp infesting sunflower stored seed in the laboratory (Zahid *et al.,* 2000); larvae of *Thecodiplosis japonensis* and intermediate nymphs of *Matsucoccus thunbergiannae* (Lee *et al.,* 2000); and larvae of balsam fir sawfly, *Neodiprion abietis* (Harris) (Li *et al.,* 2003). In our experiments, azadirachtin remained effective against larvae of *T. castaneum* but at high doses (LC_{50} = 20025 ppm).

On the other hand a few workers namely Jagdale and Grewal (2002), Yadav (1993), and Schuster and Stansly (2000) found azadirachtin no more effective against some pests or insects.

Polychlorinated petroleum hydrocarbon showed efficacy against *T. castaneum* but on the basis of LC_{50}, it proved more effective against larvae of organophosphate–susceptible (FSS–II) strain as compared to malathion–resistant (PAK) strain of *T. castaneum*. Development of resistance in *T. castaneum* against HCH has been reported by Dyte and Blackman (1970), Kumar and Bhatia (1982), Brun and Attia (1983), Barwal and Kalra (1982), and Saleem and Shakoori (1990). In our experiment, Tenekil 100 EC also proved less toxic to resistant strain of *T. castaneum* larvae.

Our data suggested that buprofezin was more effective against larvae of PAK strain of *T. castaneum* as compared to larvae of FSS-II strain. Many workers found chitin synthesis inhibitors quite effective against resistant strains of various insect pests as against multi–resistant *Musca domestica* (Pospischil *et al*., 1996), against whiteflies (Dennehy & Williams, 1997; Denholm & Jespersen, 1998), and against field strains of *Spodoptera littoralis* (Ishaaya *et al*., 2003). On contrary the chitin synthesis inhibitors were found less effective against resistant strains of *T. castaneum* (Ishaaya & Yablonski, 1987), *Bombyx mori* (L.) (Vassarmidaki *et al.,* 1999), and *Trialeurodes vaporariorum* (Gorman *et al*., 2002).

In conclusion, abamectin (Sure 1.8 EC), indoxacarb (Steward 150 SC), and spinosad (Tracer 240 SC) proved more toxic to *T. castaneum* larvae than buprofezin, azadirachtin, and Tenekil 100 EC.

REFERENCES

Abbott, W.S., 1925. A method for computing the effectiveness of insecticides. *J. Econ. Entomol*., 18: 265–7

Ahmad, M., R.M. Hollingworth and J.C. Wise, 2002. Broad–spectrum insecticide resistance in obliquebanded leafroller, *Choristoneura rosaceana* (Lepidoptera: Tortricidae) from Michigan. *Pest Manag. Sci*., 58: 834–8

Barwal, B.N. and R.L. Kalra, 1982. Cross resistance characteristics of lindane resistant and susceptible strains of *Tribolium castaneum* (Coleoptera: Tenebrionidae). *Entomon*., 7: 91–6

Bostanian, N.J., C. Vincent, J.M. Hardman and N. Larocque, 2004. Toxicity of indoxacarb to two species of predacious mites and a predacious Mirid. *Pest Manag, Sci*., 60: 483–6

Brun, L.O. and F.I. Attia, 1983. Resistance to lindane, malathion and fenitrothion in Coleopterous pests of stored products in new Caledonia. *In*: *Proc. Hawaiian Entomol. Soc.,* 24: 211–6

Busvine, J.R., 1971. *A Critical Review of the Techniques for Testing Insecticides*. CAB, London

Collins, P.J., 1990. A new resistance to pyrethroids in *Tribolium castaneum* (Herbst.). *Pestic. Sci*., 28: 101–15

Denholm, I. and J.B. Jespersen, 1998. ENMARIA–A new initiative in European insecticide and acaricide resistance management. http://www.rothamsted.bbsrc.ac.uk/enmaria/workshops/pesticideoulookintro.html. (5 May 2004)

Dennehy, T.J. and L. Williams, 1997. Management of resistance in *Bemisia* in Arizona cotton. *Pestic. Sci*., 51: 398–406

Dyte, C.E., 1972. Resistance to synthetic juvenile hormone in a strain of the red flour beetle, *Tribolium castaneum*. *Nature*, London, 238: 48–9

Dyte, C.E. and D.G. Blackman, 1970. The spread of insecticide resistance in *Tribolium castaneum* (Herbst.). *J. Stored Prod. Res*., 6: 255–61

Fang, L., B. Subramanyam and F.H. Arthur, 2002. Effectiveness of spinosad on four classes of wheat against five stored–product insects. *J. Econ. Entomol*., 95: 640–50

FAO, 1975. Recommended methods for the detection and measurement of resistance of agricultural pests to pesticides: Tentative method for adults of some major pest species of stored cereals with methyl bromide and phosphine. FAO method No. 16. *FAO Pl. Prot. Bull.,* 23: 12–35

Gorman, K., F. Hewitt, I. Denholm and G.J. Devine, 2002. New developments in insecticide resistance in the glasshouse whitefly (*Trialeurodes vaporariorum*) and the two–spotted spider mite (*Tetranychus urticae*) in the UK. *Pest Manag. Sci*., 58: 123–30

Hargreaves, K., L.L. Koekemoer and B.P. Bruke, 2000. *Anopheles funestos* resistant to pyrethroid and insecticides in South Africa. *J. Med. Vet. Entomol.,* 14: 181–9

Hasan, M., F. Ahmed, K. Ashraf and M. Ahmad, 1996. Survey of resistance against insecticides indifferent strains of *Tribolium castaneum* (Herbst.) collected from Bahawalpur Division. *Pakistan Entomol*., 18: 41–2

Huang, F. and B. Subramanyam, 2004. Responses of *Corcyra cephalonica* (Stainton) to pirimiphos–methyl, spinosad and combinations of pirimiphos–methyl and synergized pyrethrins. *Pest Manag. Sci*., 60: 191–8

Hussain, A., W. Akram and F.S. Khan, 1996a. Determination of insecticide resistance in red flour beetle, *Tribolium castaneum* (Herbst.) collected from Rawalpindi. *Pakistan Entomol*., 18: 59–61

Hussain, A., Z. Hussain and M.A. Javed, 1996b. Field detection of resistance to various insecticides in *Tribolium castaneum* (Herbst.) collected from Multan and Bahawalpur. *Pakistan Entomol*., 18: 76–7

Hutson, D. M., 1979. The metabolic fate of synthetic pyrethroid insecticides in mammals. *In:* Chasseaud, L.F. and J.W. Bridges (Eds.), Progress in Drug Metabolism, 3: 215-252. Wiley, Chichester

Iqbal, M. and D.J. Wright, 1997. Evaluation of resistance, cross–resistance and synergism of abamectin and teflubenzuron in a multi–resistant field population of *Plutella xylostella* (Lepidoptera: Plutellidae). *Bull. Entomol. Res*., 87: 481–6

Iqbal, M., R.H.J. Verkerk, M.J. Furlong, P.C. Ong, S.A. Rahman and D.J. Wright, 1996. Evidence of resistance to *Bacillus thuringiensis* (*Bt*) subsp. *kurstaki* JD–1, *Bt* subsp. *aizawai* and abamectin in field populations of *Plutella xylostella* from Malaysia. *Pestic. Sci*., 48: 89–97

Ishaaya, I. and S. Yablonski, 1987. Toxicity of two benzoylphenyl ureas against insecticide resistant mealworms, pp. 131–40. *In*: Wright, J.E. and A. Retnakaran (eds.), *Chitin and Benzoylphenyl Ureas*. Dr. Junk Publishers, Dordrecht, The Netherlands

Ishaaya, I., S. Kontsedalov and A.R. Horowitz, 2003. Novaluron (Rimon), a novel IGR: Potency and cross–resistance. *Arch. Insect Biochem. Physiol*., 54: 157–164.

Jagdale, G.B. and P.S. Grewal, 2002. Identification of alternatives for the management of foliar nematodes in floriculture. *Pest Manag. Sci*., 58: 451–8

Jairaj, 1976. *Neem Products–entomological Trials*. Utilization report. Department of Entomology, T.N.A.U., Coimbatore

Kumar, J. and S.K. Bhatia, 1982. Inheritance of resistance to lindane in a laboratory – selected strain of *Tribolium castaneum* (Herbst.). *Pestic. Sci*., 13: 513–6

Lee, S.G., J.D. Park and Y.J. Ahn, 2000. Effectiveness of neem extracts and carvacrol against *Thecodiplosis japonensis* and *Matsucoccus thunbergianae* under field conditions. *Pest Manag. Sci*., 56: 706–10.

Lessard, F.F., M.M. Vidal and H. Budzinski, 1998. Modeling biological efficacy decrease and rate of degradation of chlorpyrifos methyl on wheat stored under controlled conditions. *J. Stored Prod. Res.,* 34: 341–54

Li, A.Y., T.J. Dennehy and R.L. Nichols, 2003. Baseline susceptibility and development of resistance to pyriproxifen in *Bemisia argentifolii* (Homoptera: Aleyrodidae) in Arizona. *J. Econ. Entomol*., 96: 1307–14

Liang, P., X.W. Gao and B.Z. Zheng, 2003. Genetic basis of resistance and studies on cross–resistance in a population of diamondback moth, *Plutella xylostella* (Lepidoptera: Plutellidae). *Pest Manag. Sci*., 59: 1232–6

Lloyd, C.J., 1969. Studies on the cross tolerance to DDT related compounds of a pyrethrum resistant strain of *Sitophilus granarius* L. (Coleoptera: Curculionidae). *J. Stored Prod. Res*., 5: 337–7

Mathur, Y.K. and S. Nigam, 1996. Insecticide, antifeeding and juvenilizing effects of neem (*Azadirachta indica*) oil against *Corcyra cephalonica* Staint. and *Epilachna vigintioctopunctata* F. *In*: Singh, R.P., M.S. Chari, A.K. Raheja and W. Kraus (eds.). *Neem and Environment*, Vol. I, pp. 335–42. Oxford and 1BH Publ. Co. Pvt. Ltd., New Delhi

McLeod, P., F.J. Diaz and D.T. Johnson, 2002. Toxicity, persistence, and efficacy of spinosad, chlorfenapyr, and thiomethoxam on eggplant when applied against the eggplant flea beetle (Coleoptera: Chrysomelidae). *J. Econ. Entomol*., 95: 331–5.

Mills, K.A., 1983. Resistance to the fumigant hydrogen phosphide in some stored–product species associated with repeated inadequate treatments. *Mitt. Dtsch. Ges. Allg. Angew. Entomol*., 4: 98–101

Moulton, J.K., D.A. Pepper and T.J. Dennehy, 1999. Studies of resistance of beet armyworm (*Spodoptera exigua*) to spinosad in field populations from the Southern USA and Southeast Asia. *Univ. Arizona, College Agric. 1999* Veg. Rep. address 'http://ag.arizona.edu/pubs/crops/az1 143/'

Moulton, J.K., D.A. Pepper and T.J. Dennehy, 2000. Beet armyworm (*Spodoptera exigua*) resistance to spinosad. *Pest Manag. Sci*., 56: 842–848

Muda, R. and B.W. Cribb, 1999. Effect of uneven application of azadirachtin on reproductive and anti–feedant bahaviour of *Rhyzopertha dominica* (Coleoptera: Bostrichidae). *Pestic. Sci*., 55: 983–7

Nakakita, H. and R.G.Winks, 1981. Phosphine resistance in immature stages of a laboratory selected strain of *Tribolium castaneum* (Coleoptera: Tenebrionidae). *J. Stored Prod. Res*., 17: 43–52

Pasquier, D. and P.J. Charmillot, 2003. Effectiveness of twelve insecticides applied topically to diapausing larvae of the codling moth, *Cydia pomonella* L. *Pest Manag. Sci*., 60: 305–8

Paul, C.F., R.S. Dixit and P.N. Agarwal, 1963. Evaluation of the insecticidal properties of the seed oil and leaf extract of the common Indian neem, *Azadirachta indica* L. *Sci. Cult*., 29: 412–3

Pospischil, R., K. Szomm, M. Londershausen, I. Schroder, A. Turberg and R. Fuchs, 1996. Multiple resistance in the larger house fly, *Musca domestica* in Germany. *Pestic. Sci*., 48: 333–41

Rehman, R.A., 2000. Studies on the phosphine–induced changes in esterase levels and their role in development of resistance in 6^{th} instar larvae of red flour beetle *Tribolium castaneum*. *M.Sc. Thesis.* Department of Zoology, University of Punjab, Pakistan

Saleem M.A., A.R. Shakoori and D. Mantle, 1998. *In vivo* Ripcord induced macromolecular abnormalities in *Tribolium castaneum* larvae. *Pakistan J. Zool.,* 30: 233–43

Saleem, M.A. and A.R. Shakoori, 1989. Toxicity of malathion, permethrin, and cypermethrin against resistant and susceptible strains of *Tribolium castaneum* (Herbst.). *Pakistan J. Zool*., 21: 347–9

Saleem, M.A. and A.R. Shakoori, 1990. Toxicity of eight insecticides to sixth instar larvae and adult beetles of *Tribolium castaneum* (Herbst.). *Pakistan J. Zool*., 22: 207–16

Savitri, P. and S.C. Rao, 1976. Studies on the admixture of neem seed kernel powder with paddy on the control of important storage pests of paddy. *Andhra Agric. J*., 23: 137–43

Sayyed, A.H., D. Omar and D.J. Wright, 2004. Genetics of spinosad resistance in a multi–resistant field–selected population of *Plutella xylostella*. *Pest Manag. Sci*., 60: 827-832

Schuster, D.J. and P.A. Stansly, 2000. Response of two lacewing species to biorational and broad–spectrum insecticides. *Phytoparasitica*, 28: 297–304

Scott, J.G., 1998. Toxicity of spinosad to susceptible and resistant strains of house flies, *Musca domestica*. *Pestic. Sci*., 54: 131–3

Scott, J.G., T.G. Alefantis, P.E. Kaufman and D.A. Rutz, 2000. Insecticide resistance in house flies from caged–layer poultry facilities. *Pest. Manag. Sci*., 56: 147–53

Shakoori, A.R., M.A. Saleem and D. Mantle, 1998. Some macromolecular abnormalities induced by a sublethal dose of Cymbush 10EC in adult beetles of *Tribolium castaneum*. *Pakistan J. Zool.,* 30: 83–90

STEDEC (Scientific and Technological Development Corporation), 2004. Tenekil 100EC. STEDEC House, Lahore

Studebaker, G.E. and T.J. Kring, 2003. Effects of insecticides on *Orius insidiosus* (Hemiptera: Anthocoridae), measured by field, greenhouse and Petri dish bioassays. *Florida Entomol*., 86: 178–185

Tabassum, R., M. Jahan, S.N.H. Naqvi, 1994. Determination of toxicity of malathion and RB–a formulation against *Tribolium castaneum*. *Neem Newsl*., 11: 7–9

Tillman, P.G., G.G. Hammes, M. Sacher, M. Connair, E.A. Brady and K.D. Wing, 2001. Toxicity of a formulation of the insecticide indoxacarb to the tarnished plant bug, *Lygus lineolaris* (Hemiptera: Miridae), and the big–eyed bug, *Geocoris punctipes* (Hemiptera: Lygaeidae). *Pest Manag. Sci*., 58: 92–100

Toews, M.D. and B. Subramanyam, 2003. Contribution of contact toxicity and wheat condition to mortality of stored–product insects exposed to spinosad. *Pest Manag. Sci*., 59: 538–44

Toews, M.D., B. Subramanyam and J.M. Rowan, 2003. Knockdown and mortality of adults of eight species of stored–product beetles exposed to four surfaces treated with spinosad. *J. Econ. Entomol*., 96: 1967–73

Tyler, P.S., R.W. Taylor and D.P. Rees, 1983. Insect resistance to phosphine fumigation in food warehouses in Bangladesh. *Int. Pest Control*, 25: 10–3

Vassarmidaki, M.E., P.C. Harizanis and S. Katsikis, 1999. Effects of Applaud on the growth of silkworm (Lepidoptera: Bombycidae). *J. Econ. Entomol*., 93: 290–2

Wei, S.H., A.G. Clark and M. Syvanen, 2001. Identification and cloning of a key insecticide–metabolizing glutathione S–transferase (MdGST–6A) from a hyper insecticide–resistant strain of house fly, *Musca domestica*. *Insect Biochem. Mol. Biol.,* 31: 1145–53

White, N.D.G. and J.G. Leesch, 1995. Chemical control. *In*: Integrated Pest Management of Insects in Stored Products. Marcel Dekker, Inc. New York

Yadav, T.D., 1993. Olfactory, oviposition and developmental response of four species of pulse beetles to four neem oil treated legumes. *In*: Singh, R.P., M.S. Chari, A.K. Raheja and W. Kraus (eds.). *Neem and Environ.*, Vol. I, pp. 344–8. Oxford and 1BH Publ. Co. Pvt. Ltd., New Delhi.

Zahid, M., S. Ahmad, M.A. Murtaza and M. Hamed, 2000. Comparison of neem oil solution with malathion and Actellic in relation to the efficacy against flour beetles, *Tribolium castaneum* (Herbst.) and *T. confusum* (Duv.) infesting stored sunflower seed. *Pakistan Entomol*., 22: 51–3

Zettler, J.L., 1991. Pesticide resistance in *Tribolium castaneum* and *T. confusum* (Coleoptera: Tenebrionidae) from flour mills in the USA. *J. Econ. Entomol*., 84: 763–7

Zhao, J.Z., Y.X. Li, H.L. Collins, L.G. Minuto, R.F.L. Mau, G.D. Thompson and A.M. Shelton, 2001. Monitoring and characterization of diamondback moth (Lepidoptera: Plutellidae) resistance to spinosad. *J. Econ. Entomol*., 95: 430–6

(Received 01 February 2005; Accepted 20 July 2005)