

Susceptibility of malathion-resistant and susceptible *Tribolium castaneum* adults to abamectin, spinosad and indoxacarb

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ABSTRACT

Laboratory studies were carried out to determine the susceptibility of 10-day-old adults of malathion-resistant (PAK) and organophosphates-susceptible (FSS-II) strains of *Tribolium castaneum* to abamectin, spinosad and indoxacarb after 48 hours of treatment through residual film method. Adults of both strains were susceptible to abamectin and spinosad. The PAK strain was comparatively less susceptible to indoxacarb. Abamectin produced the lowest LC₅₀ values for PAK (88 mg L⁻¹) and FSS-II (109 mg L⁻¹), strains. These findings support the hypothesis that adult PAK and FSS-II, strains of *T. castaneum* were susceptible to abamectin, spinosad and indoxacarb.

Keywords: abamectin; indoxacarb; LC₅₀; spinosad; insecticide susceptibility; *Tribolium*.

INTRODUCTION

The red flour beetle, *Tribolium castaneum* (Herbst), is one of the most detrimental pests of stored grains and their products in Pakistan. Control of *T. castaneum* is becoming increasingly difficult due to the development of resistance to chlorinated hydrocarbons (e.g., DDT and BHC/lindane), organophosphates (e.g., malathion, fenitrothion, pirimiphos-methyl and dichlorvos), carbamates (e.g., carbaryl), pyrethroids (e.g., cypermethrin, deltamethrin, cyfluthrin, fenvalerate and permethrin) and fumigants (e.g., methyl bromide and phosphine) (Spiers *et al.* 1971, FAO 1975, Kumar & Bhatia 1982, Tyler *et al.* 1983, Saleem & Shakoori 1989, Collins 1990, Hasan *et al.* 1996, Hussain *et al.* 1996a & b, Chaudhry 1997). Resistance necessitates an escalation in application frequency and dosage, resulting in increased environmental damage and human health risks (Pimentel *et al.* 1992). Therefore, alternative means of control have been sought through the use of novel insecticides which not only control resistant insect pests, but also have low toxicity to vertebrates and beneficial organisms.

Abamectin and spinosad are rapidly eliminated *via* feces and urine in animals (EPA 1997, Dow 1997). According to Eisa & Ammar (1992), abamectin appears to be persistent in storage. The efficacy of spinosad against stored grain insect pests including *T. castaneum* has been reported by Toews and Subramanyam (2003), Toews *et al.* (2003) and Nayak *et al.* (2004). Spinosad residues were stable during storage for one year (Fang *et al.* 2002 a, b) or 9 months in warmer climates (Daglish and Nayak 2006). Indoxacarb is a reduced-risk pesticide (EPA 2000). There are no reports of abamectin, spinosad and indoxacarb effectiveness against adults of malathion-resistant (PAK) and organophosphate-susceptible (FSS-II) strains of *T. castaneum*. Therefore, the objective of the study was to evaluate the susceptibility of these two strains to the aforementioned insecticides.

MATERIALS AND METHODS

Maintenance of *T. castaneum*:

PAK and FSS-II strains of *T. castaneum* were obtained from the Department of Zoology, University of the Punjab, Lahore, Pakistan and the Ecotoxicology Centre, School of Biology, Faculty of Sciences, Agriculture and Engineering, University of Newcastle upon Tyne, UK, respectively. According to Saleem & Shakoori (1989), PAK strain adults have developed 56-fold resistance against malathion as compared to the FSS-II strain.

The beetles were reared in 300-ml glass jars covered with muslin cloth. Whole meal wheat flour was used as the culture medium. Prior to use, the culture medium was placed in an oven at 60°C for 3 hours to eliminate insect contamination (Rehman 2000). Both strains were maintained at 30±1°C and 60±5% r.h. New cultures were prepared every third day to obtain sufficient numbers of adults of the same age and size. Each bottle was filled with approximately 30 g of sterilized wheat flour (approximately ¼ the volume of the jar) and about 50 adult beetles were introduced into each. Adults collected 10 days after emergences were used in insecticidal bioassays as per Saleem & Shakoori (1989).

Toxicity Bioassays with Different Insecticides:

Commercially available formulations of abamectin, spinosad and indoxacarb were used in bioassays (Table 1). Specific concentrations were selected to yield LC₅₀ values for each insecticide for each strain on the basis of initial tests. For all experiments, insecticides were dissolved in acetone and solutions were then diluted to give a linear series of at least 5-6 concentrations. Concentrations

tested ranged from 7.8125 to 250 mg L⁻¹ for abamectin; 120 to 1920 mg L⁻¹ for spinosad and 187.5 to 3000 mg L⁻¹ for indoxacarb.

Table 1. Information regarding the toxicants used in the toxicity bioassays

Common Name	Brand Name	Main Group ³	Manufacturer/Distributor
Abamectin	Sure 1.8EC ¹	Chloride Channel activators	Pan Pacific (Pvt.) Ltd., Vehari, Pakistan
Spinosad	Tracer 240SC ²	Nicotinic Acetylcholine receptor agonists (allosteric)	SCL AgroSciences Pakistan (Pvt.) Ltd., Karachi, Pakistan
Indoxacarb	Steward 150SC	Voltage-dependent sodium channel blockers	DuPont Pakistan Operations (Pvt.) Ltd., Pakistan

¹EC = Emulsifiable Concentrate, ²SC = Soluble Concentrate; ³After IRAC MoA Classification v5.3

Insecticide dilutions (1.0 ml/dish) were applied to the centre of glass Petri dishes (130 cm²) and spread uniformly by gently rotating the dishes. Acetone alone was applied to the control dishes. Each treatment was replicated three times in a completely randomized design. After evaporation of acetone at room temperature, 20 healthy adults of similar size were released into each dish. The insects were held without food and mortality was assessed after 48 hrs. Insects were judged to be dead when the pressure from a brush failed to produce a response (as per Lloyd 1969). Percentage mortality in control was corrected using Abbott's formula for correcting control mortality (Abbott 1925).

Mortality data were subjected to probit analysis to produce LC₅₀ and LC₉₉ values as outlined by Busvine (1971) and described by Finney (1964). Resistance factors were calculated using the formula:

$$\text{Resistance factor} = \frac{\text{LC}_{50} \text{ for resistant strain}}{\text{LC}_{50} \text{ for susceptible strain}}$$

RESULTS

Characteristics of the concentration-mortality regression lines for the PAK and FSS-II strains of *T. castaneum* adults obtained by probit analysis are presented in Table 2. LC₅₀ values for PAK and FSS-II strains were 88 and 109, 724 and 854, and 1309 and 764 mg L⁻¹ for abamectin, spinosad and indoxacarb, respectively.

Table 2. Relative toxicities of some novel insecticides against malathion-resistant (PAK) and organophosphates-susceptible (FSS-II) strains of *Tribolium castaneum* adults

Insecticides	Strain	LC _{50s} (mg L ⁻¹)	95 % Fiducial limits	Slope ± S.E.	Regression equations	Chi-square (χ ²) at 2df	Resistance Ratios	LC _{99s} (mg L ⁻¹)	95 % Fiducial limits	Resistance Ratios
Abamectin (Sure 1.8EC)	PAK	88	72-109	1.83 ± 0.204	Y = 1.44 + 1.83x	1.32	0.81	41971	33884-51286	0.58
	FSS-II	109	87-137	1.74 ± 0.180	Y = 1.45 + 1.74x	0.54	-	71833	57679-90987	-
Spinosad (Tracer 240SC)	PAK	724	555-944	1.40 ± 0.065	Y = 0.996 + 1.40x	0.7	0.85	2290868	1757924-2985383	1.18
	FSS-II	854	654-1116	1.46 ± 0.057	Y = 0.720 + 1.46x	0.65	-	1939417	1492794- 2546830	-
Indoxacarb (Steward 150SC)	PAK	1309	1082-1604	2.05 ± 0.6	Y = -1.39 + 2.05x	2.27	1.71	321601	263998-391201	10.40
	FSS-II	774	528-1135	3.06 ± 0.161	Y = -3.88 + 3.08x	6.87	-	30917	21086-45290	-

Overlapping LC₅₀ fiducial limits revealed that the adults of both strains were equally susceptible to abamectin and spinosad. This was not true for indoxacarb. The PAK strain was less susceptible to indoxacarb than the FSS-II strain and the resistance ratio was 1.71. When the susceptibility of each strain was compared at the LC₉₉ level, the resistance ratio was reduced for abamectin but enhanced for spinosad and indoxacarb. It indicated the presence of some individuals in the PAK strain that were resistant to spinosad and highly resistant to indoxacarb.

DISCUSSION

The results of the research indicate that adults of PAK and FSS-II strains of *T. castaneum* are equally susceptible to abamectin by residual film exposure. However, Abro *et al.* (1988) showed that a field population of the diamondback moth, *Plutella xylostella* (L.) resistant to cypermethrin, malathion, and DDT (364- to >2100-fold) was 26-fold less susceptible to abamectin in a topical bioassay; however, the same population was equal in susceptibility in a 10 days foliar ingestion bioassay. Abamectin appears to be persistent at least in the absence of light and oxygen, e.g., in storage (Eisa & Ammar 1992).

Adults of PAK and FSS-II strains of *T. castaneum* were found susceptible to spinosad on the basis of LC₅₀ values. Similarly, spinosad showed no substantial levels of cross-resistance to organophosphate insecticides when tested on organophosphate resistant strains of house flies, *Musca domestica* L. (Scott 1998, Scott 2000, Kristensen & Jespersen 2004); obliquebanded leafroller, *Choristoneura rosaceana* (Harris) (Waldstein & Reissig 2000, Smirle *et al.* 2003); rice moth, *Corcyra cephalonica* (Stainton) (Huang & Subramanyam 2004). The only organophosphate-resistant species to show cross-resistance to spinosad has been *P. xylostella* (Sayyed *et al.* 2004). Spinosad has also been found effective against stored grain insect pests by Toews & Subramanyam (2003), Toews *et al.* (2003), Flinn *et al.* (2004), Daghish & Nayak (2006), Huang & Subramanyam (2007) and Daghish *et al.* (2008). Toews *et al.* (2003) observed excellent contact activity against adults of stored grain insects, especially on concrete, and suggested it be used as a general surface, spot, or crack/crevice spray to control insects, in empty bins, warehouses, food-processing facilities, and retail stores. Fang *et al.* (2002a, b) reported that spinosad residues were stable for one year when stored with wheat.

PAK strain adults were found less susceptible to indoxacarb than FSS-II strain adults. This indicates a presence of cross-resistance to indoxacarb in PAK populations. Ahmad *et al.* (2002) reported similar observations in studies of an organophosphate-resistant strain of *C. rosaceana*. Ahmad & Hollingworth (2004) indicated that enhanced detoxification, often mediated by cytochrome

P-450 monooxygenases, but with probable esterase and glutathione *S*-transferases contributions was the major mechanism imparting resistance in *C. rosaceana*.

We conclude that abamectin and spinosad may be of value in combating the growing threat of insecticide resistance in *T. castaneum*. PAK strain exhibits cross-resistance to indoxacarb, and as a result, indoxacarb will likely have little role in the management of this strain of organophosphate-resistant red flour beetles.

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حساسية مقاومة - الملاثيون وعرضة تريبوليوم كستانسيوم البالغة لمبيدات الآفات أبامكتين، سبائينوساد وندوكسكارب

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مختبر السمية الإيكولوجية - قسم الحشرات الزراعية
جامعة الزراعة - فيصل آباد - باكستان

خلاصة

تم القيام بالتجارب المختبرية لمعرفة حساسية كلا السلالتين مُقاومة - الميلاثيون (PAK) والمعرضة للفوسفات العضوي (FSS-II) من فصيلة تريبوليوم كستانسيوم البالغة من العمر 10 أيام تجاه ثلاثة أنواع من المبيدات هي أبامكتين، سبائينوساد وندوكسكارب على سلالتين من يافعات من خنفساء الدقيق بعد 48 ساعة من معالجتها خلال طريقة الفلم المتبقية. كان تأثير سمية المبيدات أبامكتين وسبائينوساد على البالغين من كلا السلالتين بنفس الكفاءة بينما تختلف كفاءة سمية المبيد نَدوكسكارب حيث كان أقل عرضة مقارنة بالسلالة الأخرى. لقد وجد أن أبامكتين ينتج أقل قيمة من LC50 لسلالتين PAK (88 ملجم/لتر) و FSS-II (109 ملجم/ لتر).

إن نتائج الدراسة الحالية تتفق مع الفرضية التي تعتقد بان المبيدات الثلاثة ذات كفاءة في مكافحة كلتا السلالتين من يافعات خنفساء الدقيق.

مجلة الشريعة والدراست الإسلامية

فصلية علمية محكمة تصدر عن مجلس النشر العلمي بجامعة الكويت
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- * تنوع الباحثون فيها، فكانوا من أعضاء هيئة التدريس في مختلف الجامعات والكليات الإسلامية على رقعة العالمين: العربي والإسلامي.
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