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Comparative Susceptibility of Cypermethrin in *Ornithodoros lahorensis* Neuman and *Argas persicus* Oken (Acari: Argasidae) Field Populations

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Abstract: The toxicity of cypermethrin was determined in five different soft tick strains of *Argas persicus* Oken and *Ornithodoros lahorensis* Neuman by topical application method. The *O. lahorensis* Bij, *O. lahorensis* west Ol, *O. lahorensis* Mesh, *A. persicus* Lor, *A. persicus* West Ap strains were collected from Bijar, Kurdistan province, Takab, Western Azerbaijan province, Meshkinshar, Ardebil province, Khoramabad, Lorestan province, Takab, Western Azerbaijan province of different areas of Islamic Republic of Iran, respectively during 2004 and 2005. In the topical application bioassay, the average LD₅₀ of *O. lahorensis* Bij, West Ol, Mesh and *A. reflexus* Lor and West AP strains were 0.03, 0.04, 1.7, 0.7 and 1.7 µg tick⁻¹, respectively and the steep slopes of dose-response curves indicated that the field population of these soft tick strains were homogenous in response to cypermethrin. Comparison of the resistance ratio of collected strains with susceptible strain showed a resistance ratio of 56.7 and 2.4-folds for cypermethrin in *O. lahorensis* Mesh and *A. reflexus* West Ap strains, whereas the *O. lahorensis* West Ol completely susceptible to cypermethrin.

Key words: *Ornithodoros lahorensis*, *Argas persicus*, cypermethrin, soft tick, Iran

INTRODUCTION

Ticks transmit a greater variety of pathogenic microorganisms than any other arthropod vector group and are among the most important vectors of diseases to livestock, humans and companion animals such as tick-borne encephalitis, Lyme disease, babesiosis and theileriosis (Jongejan and Uilenberg, 2004; Matias *et al.*, 2007). Infestations can also cause severe anemia, toxic conditions such as paralysis and local irritation and allergy (Hoogstraal, 1985; Matias *et al.*, 2007). In addition, tick infestations decrease the economic performance of livestock. External parasites annually cost the US beef cattle industry over US\$ 2.4 billion. Ticks cause harm to animals through: (1) blood loss, (2) general stress and irritation, (3) depression of immune function and (4) transmission of opportunistic pathogens.

Control of pests of agriculture, economic and medical importance usually focuses on the use of pesticides. Control of ticks (and hence, tick-borne diseases) is based mainly on the use of acaricides (Barnard *et al.*, 1994;

Schmidtman, 1994; Tolleson *et al.*, 2007). Current methods to control tick numbers are largely based on administration of organophosphorus or pyrethroid compounds. However, the main threat to the success of this strategy is the growing problem of tick resistance to acaricides (Jonsson *et al.*, 2000). Therefore, it is important to investigate the susceptibility that confers acaricide resistance to ticks, because this can provide significant information and tools to be used in management of acaricide resistance.

The development of resistance and cross-resistance are potential problems resulting from the over-use of any pesticides. One of the principal justifications for continued research on acaricide efficacy, longevity and application is to prevent the development of resistance in the target insect population. Pree *et al.* (2005) tested diagnostic concentrations of acaricide: Bifenazate, acequinocyl, spirodiclofen and etoxazole against Populations resistant of *Panonychus ulmi* (Koch) (Acari: Tetranychidae) to organochlorine, organotin or IGR-type acaricides. Resistance to etoxazole (4-fold) was identified

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in populations resistant to the IGRs clofentezine and hexythiazox (Pree *et al.*, 2005). Although, attempts to monitor resistance of Iranian field-collected vector and vector-borne disease strains at the adult and other stages have been conducted (Rashti *et al.*, 1992; Yaghoobi-Ershadi *et al.*, 2000; Ladonni, 2001; Enayati *et al.*, 2003; Vatandoost *et al.*, 2005; Yaghoobi-Ershadi *et al.*, 2006; Nasirian *et al.*, 2006; Djadid *et al.*, 2006; Limoe *et al.*, 2007), very little information is currently available on the toxicity of acaricides especially cypermethrin against *Ornithodoros lahorensis* Neuman and *Argas persicus* Oken for acaricide susceptibility level.

Monitoring ticks susceptibility to acaricide is required for effective use of acaricides. This requirement prompted us to evaluate the susceptibility of cypermethrin in five different soft tick strains of *Argas persicus* Oken and *Ornithodoros lahorensis* Neuman by topical application method.

MATERIALS AND METHODS

Tick strains: The toxicity of cypermethrin was determined in five different soft tick strains of *Argas persicus* Oken and *Ornithodoros lahorensis* Neuman by topical application method during 2004 and 2005. The *O. lahorensis* Bij, *O. lahorensis* west Ol, *O. lahorensis* Mesh, *A. persicus* Lor, *A. persicus* West Ap were collected from Bijar, Kurdistan province, Takab, Western Azerbaijan province, Meshkinshar, Ardebil province, Khoramabad, Lorestan province, Takab, Western Azerbaijan province of different areas of Islamic Republic of Iran, respectively during 2004 and 2005.

Tick collection and identification: Tick collection was carried out in human dwelling, animal and poultry shelters. Ticks were collected from cracks, crevices, ceiling and floor and transferred into the holding tubes (Telmadarrayi *et al.*, 2004). After collection, ticks were transported to laboratory of the Department of Medical Entomology and Vector Control, School of Public Health and Institute of Public Health Researches, Medical Sciences/University of Tehran.

The ticks were identified by morphological characteristics using tick identification keys available from different sources (Ruedisueli and Manship, 2006; Telmadarrayi *et al.*, 2004; Walker *et al.*, 2003; Bowman and Lynn, 1995). Important morphological features used in tick identification included size of the tick, position of head and mouthparts (capitulum) relative to the thorax and abdomen on dorsal and ventral views and shape of the body. Laboratory investigation of the collected ticks

revealed the ticks to be soft ticks belonging to the *Argas persicus* and *Ornithodoros lahorensis* (Acari: Argasidae) (Ruedisueli and Manship, 2006; Walker *et al.*, 2003; Bowman and Lynn, 1995).

Chemicals and reagents: Technical grade insecticide: Cypermethrin (97.5%) and acetone as a solvent were used.

Topical application bioassay: Technical grade cypermethrin (97.5%) was delivered in 0.1 μ L acetone to the tergum surface of adult soft ticks by topical application with a micro applicator equipped with a 1.0 mL hypodermic glass syringe fitted with a 27-gauge needle. Adult stage of soft ticks were treated with 5-6 concentration of acaricide giving >0% and <100% mortality and each concentration was replicated 3-6 times (10 ticks for each replicate). Control groups received acetone alone. Treated ticks were placed in plastic Petri dishes (150 mm diameter) and monitored for mortality for 14 days at $27\pm 2^\circ\text{C}$, $60\pm 10\%$ RH and a photo-period of 12: 12 (L: D) h. If ticks on their backs were unable to right themselves when prodded, they were considered dead.

Statistical analysis: Mortality data from the replicates were pooled and the dose-response was assessed by probit analysis (Finney, 1972), with a SPSS package on an IBM computer. Resistance ratios were calculated as the 50% response value (LD_{50}) of RR strain divided by the 50% response value of the SS strain.

RESULTS AND DISCUSSION

The toxicity of cypermethrin was determined in five different soft tick strains of *Argas persicus* and *Ornithodoros lahorensis* by topical application method. Table 1 shows data for the mortality assay topical application to the tergum surface of adult soft ticks. In the topical application bioassay, the average LD_{50} of *O. lahorensis* Bij, West Ol, Mesh and *A. reflexus* Lor and West AP strains were 0.03, 0.04, 1.7, 0.7 and 1.7 $\mu\text{g tick}^{-1}$, respectively and the steep slopes of dose-response curves indicated that the field population of these soft tick strains were homogenous in response to cypermethrin (Fig. 1).

Comparison of the resistance ratio of collected strains with susceptible strain showed a resistance ratio of 56.7 and 2.4-folds for cypermethrin in *O. lahorensis* Mesh and *A. reflexus* West Ap strains, respectively, whereas the *O. lahorensis* West Ol completely susceptible to cypermethrin. In conclusion resistance ratio of 56.7-fold is very high and is disheveled the experts of vector control. This is the first study from Islamic Republic of Iran reporting tick acaricide resistance. Although this

Table 1: Toxicity of cypermethrin topically applied to five different soft tick strains of *Ornithodoros lahorensis* and *Argas reflexus*

Strains	n	Slope±SE	χ^2	LD ₅₀ ^a (CI)	RR ₅₀ ^b
<i>Ornithodoros lahorensis</i>					
Bij	150	1.40±1.00	1.6	0.03 (0.01-0.05)	-
Mesh	150	0.02±0.01	2.3	1.7 (0.5-2.8)	56.7
West Ol	150	1.30±1.01	0.2	0.04 (0.01-0.06)	1.3
<i>Argas reflexus</i>					
Lor	150	0.50±0.20	2.2	0.7 (0.13-1.89)	-
West Ap	150	0.10±0.10	0.6	1.7 (0.61-3.3)	2.4

^a: LD₅₀ values in micrograms per insect (95% CI); ^b: Resistance ratio: LD₅₀ of feral strain/LD₅₀ of susceptible strain

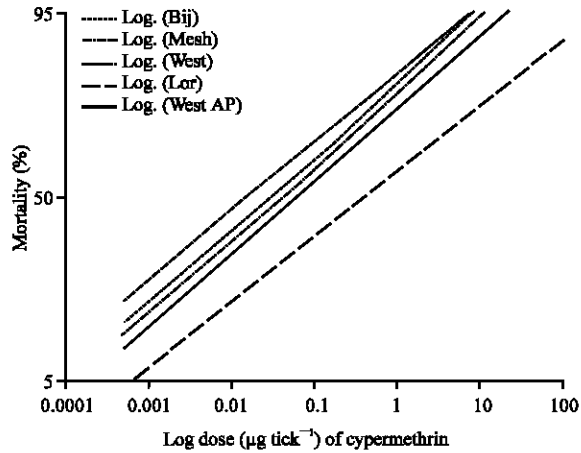


Fig. 1: Probit-regression lines of cypermethrin on mortality of five different soft tick strains of *Ornithodoros lahorensis* and *Argas reflexus*

study showed that a resistance ratio of 56.7 and 2.4-folds for cypermethrin in *O. lahorensis* Mesh and *A. reflexus* West Ap strains and a valuable results for tick acaricide resistance monitoring, this is the first step for acaricide resistance management in Islamic Republic of Iran. Because monitoring ticks susceptibility to acaricides is required for effective use of acaricides therefore, further studies are required to determine the susceptibility of soft tick and probably acaricide resistance, *A. persicus* and *O. lahorensis* to other acaricide groups in Islamic Republic of Iran and elsewhere in these and other tick species.

Monitoring ticks' susceptibility to acaricides is required for effective use of acaricides. Foil *et al.* (2004) revealed that populations of southern cattle ticks, *Boophilus microplus*, from Mexico have developed resistance to many classes of acaricide including chlorinated hydrocarbons (DDT), pyrethroids, organophosphates and formamidines (amitraz). According to present study, we cannot use cypermethrin and probably other pyrethroid acaricides to control the *O. lahorensis* and *A. reflexus* species in Islamic Republic of Iran and shows that *O. lahorensis* Mesh strain is closed up to pyrethroid insecticides for vector control or especially agriculture pests control. The use of

pesticides for the control of pests can affect the incidence of tick resistance and tick-borne diseases (Foil *et al.*, 2004).

We have to arm ourselves to control the pests especially ticks with new acaricides in different mode of action and new pest control methods. Hence, we propose further studies on toxicity of novel insecticides such as avermectins acting at GABA, gama-aminobutyric acid and glutamate receptors in proximity to chloride channels, diacylhydrazines mimicking the action of ecdysone, indoxacarb acting at a novel site in sodium channels and imidacloprid, thiamethoxam and spinosad acting at novel sites on the nicotinic acetylcholine receptor, several promising products acting at new target sites, which are currently not affected by resistance that will be used for pest control in Islamic Republic of Iran.

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