# **Comparison of Permethrin and Fipronil Toxicity against German Cockroach (Dictyoptera: Blattellidae) Strains**

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#### Abstract

Control of insect pests of agriculture, economic, and medical importance usually focuses on the use of insecticides. The susceptibility levels of eleven strains of German cockroaches, *Blattella germanica* (L.), were compared with a standard susceptible strain against permethrin and fipronil. The strains were collected from nine infested students' dormitories and two infested hospitals in Tehran, capital of Iran. The susceptible strain showed  $LD_{50}$  of 0.43 µg and 0.96 ng for permethrin and fipronil respectively. Comparison of the resistance ratio of collected strain with susceptible strain showed resistance ratios of 8.6 to 17.7-folds for permethrin and 1.5 to 2.6-folds for fipronil, respectively. The result of this investigation indicated that the all strains were resistant to permethrin. Resistance spectrum of fipronil to permethrin showed different pattern of susceptibility to fipronil, indicating that there was no relationship between resistance to permethrin and fipronil.

Keywords: Blattella germanica, Fipronil, Permethrin, Iran

### Introduction

Control of insect pests of agriculture, economic, and medical importance usually focuses on the use of insecticides. The German cockroach, Blattella germanica (L.), has created substantial pest problems in many developing countries (1-4). The resistance of German cockroach to insecticides such as organochlorines, organophosphates, and pyrethroids has been investigated by several laboratories (2, 5, 6). The search for new insecticides and new methods of insecticide delivery to control the German cockroach continues because this insect remains one of the most economically and medically important pests of the urban environment (7). In this ongoing process, older chemicals are displaced because of insecticide resistance, increasingly strict regulations, and public demand for safer and more effective products. New active ingredients and innovative delivery tools emerge to

provide effective means of dealing with infestations (8). One of the newest insecticides is fipronil, a relatively fast-acting phenylpyrazole that blocks the transmission of signals by the inhibitory neurotransmitter gama-aminobutyric acid (9-11). Its higher toxicity of fipronil in insects than in mammals is due partially to the higher sensitivity of GABA receptors (12). It is highly toxic to the German cockroach (13) and other pests of public and medical importance (14). Fipronil is a highly effective, broad-spectrum insecticide with potential value for the control of a wide range of crop, public hygiene, amenity and veterinary pests. It can generally be applied at low to very low dose rates to achieve effective pest control.

The development of resistance and cross-resistance are potential problems resulting from the over-use of any pesticides and fipronil is relatively new and thus currently free from this problem. One of the principal justifications for continued research on insecticide efficacy, longevity, and application is to prevent the development of resistance in the target insect population. Although attempts to monitor resistance of field-collected strains at the adult stage have been conducted (15), in Iran very little information is currently available on the toxicity of fipronil to development of resistance German cockroach or no studies conducted.

The current study was designed to investigate the relationship between permethrin German cockroach strains resistant and fipronil.

# **Materials and Methods**

*Cockroach strains* Twelve German cockroach were used in this study: a standard susceptible (SS) strain was maintained since 1975 in the insectaria at the School of Public Health, Tehran University of Medical Sciences (TUMS), Iran, without exposure to insecticide; eight strains were collected from infested kitchen student dormitories at Sanatee Sharif, Tarbiat Modares, (TUMS), one strain from infested Habitable Convened (Saman) and two strains from infested hospitals in Tehran and colonized in the insectaria at the School of Public Health, (TUMS). Cockroaches were collected with a piece of a radiology film (10×10 cm) and transferred to a apparatus by hand catch. The apparatus manufactured from two parts, the upper inside surface of the upper part (5cm) was lightly greased with petroleum jelly to prevent cockroaches from escaping, after collecting cockroaches in the insectaria the lower part separated from the upper part and cockroaches transferred to glass rearing jars to prevent cockroaches from greasing. All cockroaches were maintained in an insectaria at  $27\pm 2$  °C,  $60\pm 10\%$  RH, and a photoperiod of 12:12 (L:D) h. Each strain was kept in separate labeled glass rearing jars of the same size (500 ml). The upper inside surface of the jars was lightly greased with petroleum jelly to prevent escape. Cockroaches were provided with cat food, water ad libitum and a cardboard shelter

Topical application bioassays Technical grade fipronil (95% [AI]; Rhone-Poulenc, Research Triangle Park, NC) and permethrin, 92.5% (Technical grade) cis: trans 60:40 (Zeneca, Haslemere, UK), were delivered in 0.5 ul acetone to the first abdominal sternum of briefly CO<sub>2</sub>-anesthetize adult male cockroaches by topical application with a micro applicator (Hamilton, Reno, NV) equipped with a 1.0-ml hypodermic glass syringe fitted with a 27-gauge needle (16). Adult male cockroaches (1-3 wk old) were treated with 5-6 concentration of insecticide, and each concentration was replicated 3-6 times (10 cockroaches for each replicate). Only adult males (1-3 wk old) were used in bioassays because their weight and physiology were more uniform than those of adult female cockroaches were (17). Moreover, female cockroaches were needed for further reproduction. Control groups received acetone alone. A 5-6 doses giving >0% and <100% mortality at 24 h for permethrin and 72 h for fipronil after insecticide treatment was used for each experiment. In addition, it should be mentioned that mortality after fipronil treated was recorded for 6 d but mortality after 72 h becoming stable.

Treated males were placed in 150 by 25-mm plastic petri dishes, provided with food and water, and monitored for mortality for 24 h (in permethrin treated) and 72 h (in fipronil treated) under the same temperature and photoperiod as the colony. If insects 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 (18), with an 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

Table 1 shows data for the mortality assay topical application on male adults (1-3 wk old). The susceptibility level of eleven strains of German cockroaches was evaluated and compared with a standard susceptible strain against permethrin and fipronil. The  $LD_{50}$  in male adults were measured by topical application. The susceptible strain showed  $LD_{50}$  of 0.43 µg and 0.96 ng for permethrin and fipronil, respectively. Com-

parison of the resistance ratio of collected strains with susceptible strain showed a resistance ratio of 8.6 to 17.7- folds for permethrin, whereas most of the strains showed 1.5 to 2.6folds very low level of resistance and a few strain a similar susceptibility to fipronil.

Strains		]	Permethrin					Fipronil		
	n	Slope±SE	LD <sub>50</sub> <sup>a</sup> (CI)	χ²	RR <sup>b</sup>	n	Slope±SE	LD <sub>50</sub> <sup>c</sup> (CI)	$\chi^2$	RR <sup>b</sup>
Susceptible	160	$5.01 \pm 0.67$	0.43 (0.38-0.48)	3.2	-	285	1.3± 0.13	0.96 (0.81-1.1)	3.0	-
Bustan-7	240	$0.33 \pm 0.04$	6.5 (5.9-7.1)	4.2	15.1	280	1.1±0.10	1.4 (1.2–1.6)	4.4	1.5
Bustan-8	160	$0.44 \pm 0.07$	4.5 (4.1-5.1)	1.0	10.5	314	$1.0 \pm 0.1$	1.4 (1.2–1.6)	5.0	1.5
Bustan-10	160	$0.33 \pm 0.05$	5.3 (1.8-9.7)	3.9	12.3	402	$0.9\pm0.1$	2.5 (2.2–2.8)	15.2	2.6
Kouye-Pezeshky	200	$0.35 \pm 0.04$	5.3 (1.8-9.7)	3.3	11.2	463	$1.3 \pm 0.1$	0.94 (0.82-1.1)	4.4	0.96
Shahmorady	160	$0.25 \pm 0.04$	5.9 (5.0-6.9)	3.4	13.7	279	$1.3 \pm 0.13$	1.0 (0.85-1.2)	2.6	1.0
Zanjan	160	$0.41 \pm 0.06$	3.7 (3.4-6.1)	3.8	8.6	400	$1.0 \pm 0.1$	2.5 (2.4-2.7)	4.6	2.6
Kouye-Tehran	200	$0.29 \pm 0.04$	7.6 (6.9-8.4)	4.3	17.7	362	$1.2 \pm 0.2$	1.5 (1.3-1.6)	4.6	1.6
Saman	200	$0.35 \pm 0.04$	6.1 (5.5-6.7)	3.0	14.2	406	$0.9\pm0.1$	2.3 (2.1-2.5)	9.8	2.4
Mogtameh	200	$0.41\pm0.05$	6.6 (6.0-7.2)	2.3	15.4	360	$1.2 \pm 0.11$	1.0 (0.96-1.2)	8.5	1.0
Kouye-Pezeshky	200	$0.32 \pm 0.04$	6.6 (6.0-7.3)	2.8	15.4	399	$1.0 \pm 0.1$	1.9 (1.8-2.1)	9.3	2.0
Fayyazbakhsh	160	$0.50 \pm 0.07$	4.2 (3.7-4.8)	2.3	9.8	355	$1.4 \pm 0.13$	1.0 (0.90-1.2)	4.2	1.0

Table 1: Toxicity of topic	ally applied permethrin an	d fipronil to laboratory-reare	d German cockroach strains
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### Discussion

In all field-collected strains, cockroaches showed different degrees of resistance to permethrin. Comparison of the resistance ratio of collected strains with susceptible strain demonstrated that permethrin resistance in German cockroach was not an isolated occurrence. The occurrence of insecticide resistance in feral German cockroach collected from different places in Tehran is probably caused by the exertion of selection pressure on these cockroaches by regular spraying with insecticides, including pyrethroides, organo phosphates, and carbamates. Our results confirm those of Kaakeh et al. (13) who reported that fipronil was very effective against laboratory-reared German cockroaches in nanogram quantities. Furthermore, German cockroaches collected from the field exhibited higher susceptibility level toward fipronil than the susceptible strain (13).

Fipronil was relatively slow-acting against German cockroaches, with the  $LD_{50}$  values decreasing until 72 h and becoming stable thereafter. For this reason, 72 h was used as the standard time for determination of toxicity for the remainder of the experiments. These data confirm a preceding study that determined the toxicity of fipronil in German cockroach and housefly (14).

Valles et al. (19) reported that fipronil effectively killed German cockroaches when applied in nanogram quantities; insecticide-susceptible, carbamate- and organophosphorus-resistant, pyrethroid- resistant strains were equally susceptible to fipronil with LD<sub>50</sub> values between 4.6 and 5.4 ng per insect. A pyrethroid- organophosphorus, and carbamate-resistant was 1.6fold tolerant to fipronil. German cockroaches strains collected from field were considerably tolerant to fipronil.

In another study (14), fipronil was highly toxic to German cockroaches. A cyclodiene-resistance strain (>17,000-fold) had cross- resistance (7.7-fold) to fipronil. A pyrethroid- resistant strain (59-fold), chlorpyrifos-resistant strain (22-fold) bendiocarb-resistant strain (89-fold), bendiocarb-resistant strain (270-fold) and propoxur-resistant strain (19-fold) were <2-fold different from the susceptible strains.

Fipronil is currently available only in bait formulations for use against cockroaches. The current moderate levels of resistance to fipronil in the German cockroach militate against its use as a residual insecticide, for the historical weight of evidence clearly indicates that cockroaches can become highly resistant to most insecticides used in this manner. It is indeed imaginable that German cockroaches could ultimately become as resistant to residual fipronil as they once were to the cyclodienes. If this were to occur, fipronil would almost certainly become ineffectual in either spray or bait formulations. It would be particularly troubling for fipronil to lose its efficacy in baits, for consumers and pest management professionals alike are increasingly turning to them in their efforts to control cockroaches. This trend is unlikely to let up, largely for regulatory reasons (20).

In conclusion, resistance spectrum of fipronil to permethrin showed different pattern of susceptibility to fipronil, indicating that there was no relationship between resistance to permethrin and fipronil.

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