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Research Article

Toxicity of Cypermethrin and Chlorpyrifos Against German Cockroach [*Blattella germanica* (Blattaria: Blattellidae)] Strains from Hamadan, Iran

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Abstract

Background: German cockroach has relatively short life cycle and reproduce rapidly. It is the most common medically and public health pest. As a result, it is essential to combat this pest. Cypermethrin and chlorpyrifos are used by private companies in Hamadan to control *Blattella germanica*. It seems necessary to determine its susceptibility levels to these insecticides. **Objective:** The aim of this study was to determine the susceptibility levels of *B. germanica* strains to cypermethrin and chlorpyrifos in Hamadan. **Materials and Methods:** In this study, the German cockroach strains were collected from two hospitals (Fatemiyeh and Atiyeh) in Hamadan and transferred to the insectarium. The cockroach strains were reared under the same laboratory condition. Then their sensitivity levels were considered to 1, 2, 4, 8 and 16 mg m⁻² for cypermethrin and 0.82, 1.65, 3.31, 6.63, 9.945 and 13.26 mg m⁻² for chlorpyrifos using surface contact method. **Results:** Results based on insecticide treated doses, *B. germanica* strains showed different percent mortality to the insecticides ranged from 13.3-100. The LD₅₀ and LD₉₀ and regression lines of the treated insecticides against German cockroach strains indicate that Fatemiyeh Hospital strain is more susceptible to the treated insecticides than Atiyeh Hospital strain. The LD₅₀ and LD₉₀ of chlorpyrifos are also lower than cypermethrin, indicated that chlorpyrifos is more effective than cypermethrin against German cockroach. As the slopes of the regression lines are observed mild in this study indicate that the population of the cockroach strains is very heterogeneous. It can be a symbol of insecticides resistance to cypermethrin and chlorpyrifos. **Conclusion:** As chlorpyrifos and cypermethrin insecticides are also used for residual spraying by private companies and the doses which provide more than 90% mortality are below the WHO recommended insecticide doses. Therefore, chlorpyrifos and cypermethrin insecticides can be used for *B. germanica* control in Hamadan within regular monitoring and preventive measures of resistance.

Key words: German cockroach, chlorpyrifos, cypermethrin, *Blattella germanica*, toxicity

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

German cockroach [*Blattella germanica* (Blattaria: Blattellidae)] is the most common medically and public health pest^{1,2}. German cockroach has relatively short life cycle and reproduce rapidly. *Blattella germanica* can infest homes, restaurants, hotels, hospitals and food storage. It can transmit pathogenic agents such as bacteria, viruses, fungi, protozoa and parasite eggs to human mechanically or through the digestive tract. Therefore, it is the most important vector of the diseases such as cholera, plague, leprosy, typhoid and diarrhea, etc. Moreover, German cockroach is the most important sources of allergen. So, it has a significant role in the development and exacerbation of allergic diseases and asthma². As a result, it is essential to combat this pest and has a particular importance.

This is essential to use insecticides for German cockroach control. Repeat application and extensive use of insecticides has caused German cockroach resistant to insecticides in the world. In Iran, German cockroach is the most important public health problem. It has been resistant to different groups of insecticides and actually its control is very difficult²⁻¹⁰.

Although, good food hygiene practices, including preventing access to food by sealing it in containers with close-fitting lids and preventing cockroaches from reaching water by repairing dripping taps and covering plug holes in sinks and drains are effective in restricting cockroach populations and the use of gel bait insecticides can be a strategy to control it¹¹⁻¹⁴. Whereas it is commonly used residual insecticide spraying by private companies that are responsible for its fighting. Therefore, it is necessary to consider the susceptibility levels to the consumed insecticides such as chlorpyrifos and cypermethrin. So far, several studies have been done about susceptibility of German cockroach to cypermethrin¹⁵⁻²⁵ and chlorpyrifos^{5,15,16,20,23,26-30} in the world. These insecticides are already used by private companies in Hamadan, a study design to consider the susceptibility levels of *B. germanica* to cypermethrin and chlorpyrifos in Hamadan. The aim of this study was to determine the susceptibility levels of *B. germanica* strains to cypermethrin and chlorpyrifos in two hospitals of Hamadan from Iran.

MATERIALS AND METHODS

Site of study: In this study, wild German cockroach strains were collected from Fatemiyeh and Atiyeh Hospitals in

Hamadan, the Center of Hamadan province. Hamadan is one of the oldest cities in the world and a metropolitan of Iran now. It composed an area about 4118 km² with two parts and 8 districts. Hamadan locates on the Alvand mountain ranges in the Western and mountainous of Iran with an elevation of 1820 m above the sea level. The city in terms of cultural heritage, history and handicrafts has abundant tourist attractions. Every year a large number of Iranian and foreign tourist visit Hamadan.

Fatemiyeh Hospital locates in the West of Hamadan and has 6,000 m² building space with 300 approved beds it has 158 active beds now. Fatemiyeh Hospital consists of different parts includes: Ten medical units, 9 clinical and diagnostic units and 12 specialized and ultra-specialized clinics. It is the first degree hospital and the first ones in Hamadan province which has received the 'ISO 9001-2000 certificate'. It is an already women top pole of health care, education and study in the West of Iran and provides the medical care to the people and educational and research services to the students.

Atiyeh Hospital locates in the North of Hamadan and has 2,400 m² building space with 256 approved beds, it has 116 active beds now. Atiyeh Hospital consists of different parts includes: 16 medical units, 16 clinical and diagnostic units and 13 specialized and ultra-specialized clinics. It provides the medical care to the people.

Cockroaches collecting and rearing: Cockroaches were collected by Nasirian *et al.*^{4,31} methods. After collecting each cockroach strains were transferred into large glass jar, maintained and colonized at 24±2°C, 40±5% RH and a photoperiod of 12:12 (L:D) h in the insectary at Department of Medical Entomology, School of Medicine, Hamadan University of Medical Sciences. Cockroaches were provided with rodent diet, a cotton plugged water vial and a cardboard as a harborage.

Chemicals and reagents: Cypermethrin (technical grade, 93.0%) and chlorpyrifos (technical grade, 97.0% crystal) as insecticides, CO₂ as an anesthetic and acetone as a solvent were used.

Surface contact bioassay: In this study a glass jar was used for surface contact bioassay. It calculated the glass jars inner surface area, it was 188.4 cm². Technical grade of the insecticides were diluted by acetone solvent. So, this study designed to dilute insecticide concentrations which 1 mL volume contains considered insecticide doses. Then, 1 mL of diluted insecticide was pipetted into the glass jar. To deposit

the insecticide evenly over the inner surface of the glass jars, the glass jars were rolled horizontally over a flat surface until all of the acetone had evaporated. In a series of contact experiments, a concentration of technical grade at a 30 min exposure time was found to be a discriminating dose for male adults. Then adult male cockroaches were treated with 5-6 insecticide dose exposures at a 30 min time and each dose exposures were replicated 3 times (10 cockroaches for each replicate). This study used the insecticide doses of 1, 2, 4, 8 and 16 mg m⁻² for cypermethrin and 0.82, 1.65, 3.31, 6.63, 9.945 and 13.26 mg m⁻² for chlorpyrifos. Control groups received acetone alone. All insecticide dose exposures were given >0% and <100% mortality at 24 h after insecticide dose exposures. Insecticide exposed males were placed in plastic dishes, provided with food and water and monitored for mortality for 24 h under the same temperature and photoperiod as the colony. If insects on their backs were unable to right themselves, they were considered dead.

Statistical analysis: Mortality data from the replicates were pooled and the dose exposure mortality was assessed by probit regression analysis with a PASW statistics (18 version for windows) on a computer. Microsoft Office Excel (2007 version) were used to calculate equation and draw the regression lines.

RESULTS

Results based on the insecticide treated doses, German cockroach strains showed different percent mortalities to the insecticides ranged from 13.3-100 (Table 1).

The LD₅₀ and LD₉₀ of the treated insecticides against German cockroach strains indicate that Fatemiyeh Hospital strain is more susceptible to treated insecticides than Atiyeh Hospital strain (Table 2). The LD₅₀ and LD₉₀ of chlorpyrifos are lower than cypermethrin indicated that chlorpyrifos is more effective than cypermethrin against German cockroach (Table 2).

Regression lines of Fatemiyeh Hospital strain to the treated insecticides are also in front of Atiyeh Hospital strain that demonstrate Fatemiyeh Hospital strain is more susceptible than Atiyeh Hospital strain (Fig. 1). The slopes of the regression lines were observed mild (Fig. 2 and 3) indicate that the population of the cockroach strains is very heterogeneous.

DISCUSSION

German cockroach is the most common medically and public health pest^{1,2}. It is commonly used residual insecticide spraying by private companies that are responsible for its fighting. Chlorpyrifos and cypermethrin insecticides are already used by private companies in Hamadan, the current study designed to consider the susceptibility levels of *B. germanica* strains to cypermethrin and chlorpyrifos in two hospitals of Hamadan from Iran. Results based on the insecticide treated doses German cockroach strains showed different percent mortality to the insecticides ranged from 13.3-100 (Table 1).

The slope of the regression line is a scale for the heterogeneity and genetic population variation. In this study, although the resistance of German cockroach was not studied but the slopes of the regression lines are mild (Fig. 2 and 3) indicate that the population of the cockroach strains is very heterogeneous. It can be a symbol of insecticides resistance to cypermethrin and chlorpyrifos. A degree of chlorpyrifos

Table 1: Toxicity of cypermethrin and chlorpyrifos against German cockroach strains by WHO glass jar method

Cypermethrin (mg m ⁻²)	Cockroach mortality (%)		Chlorpyrifos (mg m ⁻²)	Cockroach mortality (%)	
	FH	AH		FH	AH
1	23.3	16.7	0.82	20.0	13.3
2	46.7	36.7	1.65	50.0	43.3
4	66.7	60.0	3.31	70.0	63.3
8	83.3	73.3	6.63	86.6	80.0
16	97.0	93.0	9.945	96.6	93.3
-	-	-	13.26	100	100

FH: Fatemiyeh Hospital strain and AH: Atiyeh Hospital strain

Table 2: Probit regression analysis of cypermethrin and chlorpyrifos against German cockroach strains by WHO glass jar method

Strain	n ^a	Intercept±SE	Slope±SE	χ ²	df	p	LD ₅₀ ^b (CI)	LD ₉₀ ^b (CI)
Cypermethrin								
FH	150	-0.53±0.18	0.18±0.03	4.9	3	0.18	3.03 (1.38-4.35)	10.36 (8.28-14.54)
AH	150	-0.69±0.17	0.15±0.03	5.2	3	0.16	4.49 (2.89-6.00)	12.81 (10.37-17.45)
Chlorpyrifos								
FH	180	-0.66±0.19	0.28±0.05	4.9	4	0.29	2.37 (1.34-3.21)	6.97 (5.78-9.02)
AH	180	-0.80±0.18	0.24±0.04	7.1	4	0.13	3.28 (0.91-5.13)	8.53 (6.35-14.71)

^aNo. of cockroach treated, ^bLD₅₀ and LD₉₀ values in mg m⁻² (95% CI), FH: Fatemiyeh Hospital strain and AH: Atiyeh Hospital strain

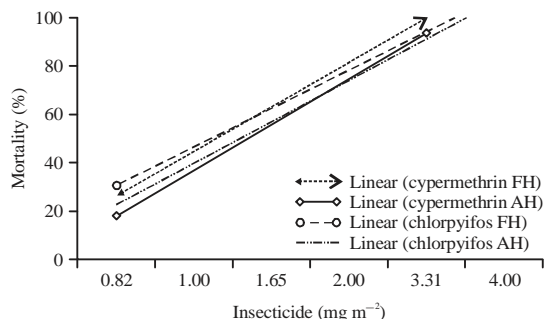


Fig. 1: Regression line of cypermethrin and chlorpyrifos against German cockroach strains by WHO glass jar method, FH: Fatemiyeh Hospital strain and AH: Atiyeh Hospital strain

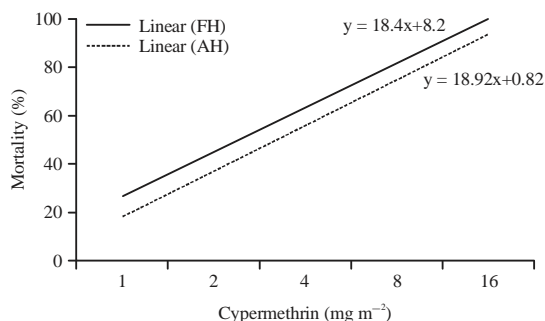


Fig. 2: Regression line of cypermethrin against German cockroach strains by WHO glass jar method, FH: Fatemiyeh Hospital strain and AH: Atiyeh Hospital strain

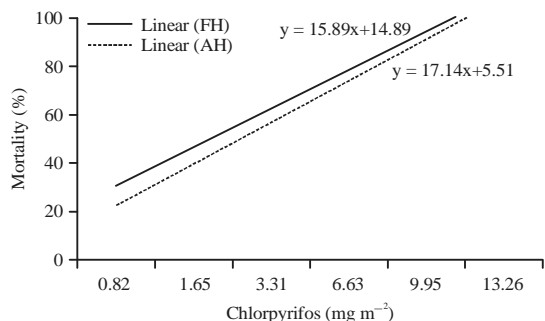


Fig. 3: Regression line of chlorpyrifos against German cockroach strains by WHO glass jar method, FH: Fatemiyeh Hospital strain and AH: Atiyeh Hospital strain

resistance was reported in *B. germanica*³⁰ in Iran. So far, worldwide cases of resistance were reported to cypermethrin^{15-25,32,33} and chlorpyrifos^{5,16,20,23,27-30,32}.

Recently, it has been proved that the essential oil of *Pogostemon cablin* (Lamiales: Lamiaceae) leaves; *Artemisia*

arborescens and *A. santolina* (Asterales: Asteraceae); Apiaceae (Apiales) plants and their major constituents have good potential as a source for natural insecticides or repellents³⁴⁻³⁶ for German cockroach control, until they will be legally and officially used take a lot of time.

The use of gel bait insecticides can also be a strategy for control of the German cockroach¹¹⁻¹⁴. Whereas, it is commonly used the residual insecticide spraying by private companies that they are responsible for German cockroach fighting. In this study, as the doses of chlorpyrifos and cypermethrin insecticides provide more than 90% mortality (Table 1) are below the WHO recommended insecticide doses. Therefore, they can be used for *B. germanica* control in Hamadan. But precautions such as using rotationally different group of insecticides, etc. must be observed.

CONCLUSION

The slope of the regression line is a scale for the heterogeneity and genetic population variation and they are observed mild in this study, indicate that the population of the cockroach strains is very heterogeneous. It can be a symbol of insecticides resistance to cypermethrin and chlorpyrifos. As chlorpyrifos and cypermethrin insecticides are commonly used for residual spraying by private companies and the doses which provide more than 90% mortality are below the WHO recommended insecticide doses. Therefore, they can be used for *B. germanica* control in Hamadan. But precautions such as using rotationally different group of insecticides, etc., must be observed.

SIGNIFICANCE STATEMENT

- The susceptibility levels of *B. germanica* strains to cypermethrin and chlorpyrifos in Hamadan determined
- The LD₅₀ and LD₉₀ and regression lines of the treated insecticides against German cockroach strains indicate that Fatemiyeh Hospital strain is more susceptible to treated insecticides than Atiyeh Hospital strain. The LD₅₀ and LD₉₀ of chlorpyrifos are also lower than cypermethrin indicated that chlorpyrifos is more effective than cypermethrin against German cockroach
- Chlorpyrifos and cypermethrin insecticides can be used for *B. germanica* control in Hamadan within regular monitoring and preventive measures of resistance

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