Punjab Univ. J. Zool., Vol. 32 (2), pp. 167-171, 2017
ISSN 1016-1597(Print)
ISSN2313-8556 (online)

Original Article

Residual effect of lambda-cyhalothrin on abundance of insect pollinators in marigold field patch

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Article history
Received: January 08, 2017
Revised: November 19, 2017
Accepted: November 28, 2017

Authors' Contribution
HMT, ZIK: plan experimental research work, **SB, KA, SB:** perform experiments and collect and analyzed data and draft manuscript

Key words
Pollination
Honey bee
Insect pollinators

Abstract
In the present residual effects of lambda-cyhalothrin on the abundance of insect pollinators was recorded under field and a semi- field conditions. Recommended dose of lambda-cyhalothrin (0.093gm/ml) was used. The number of different pollinators that visited the marigold plant before and after insecticidal spray was recorded. In semi-field experiment, honey bees were exposed to insecticide treated plants for one hour. The mortality rate of honey bees in the control and insecticide exposed group was compared. Overall, a significant decline in plant pollinators was observed after application of lambda-cyhalothrin on the patch of marigold plants. Lambda-cyhalothrin caused significant mortality (15/20=75%) in honey bees in semi-field experiment. It is concluded that lambda-cyhalothrin is highly poisonous to insect pollinators; therefore its use should be minimized to protect the population of insect pollinators.

To cite this article: TAHIR, H.M., KHAN, Z.I., BATOOL, S., AHMAD, K. AND BEGUM, S., 2017. Residual effect of lambda-cyhalothrin on abundance of insect pollinators in marigold field patch. *Punjab Univ. J. Zool.,* **32**(2): 167-171.

INTRODUCTION

Pollination is an essential biological process which is accomplished through various ways. In angiosperms, 60-90% reproduction takes place through pollination (Richards, 1986; Renner, 2006; Kremen *et al.,* 2007). Insect pollinators not only maintain healthy plant populations (Ollerton *et al.,* 2011) but also contributes to economic value of over $150 billion per annum globally (Gallai *et al.,* 2009). Social bees (honeybees, bumblebees and stingless bees) are the key insect pollinators (Greenleaf and Kremen, 2006; Winfree *et al.,* 2007), but in recent years their populations have practiced noteworthy declines (Oldroyd, 2007; Goulson *et al.,* 2008; Van Engelsdorp *et al.,* 2008; Brown and Paxton, 2009; Cameron *et al.,* 2011; Burkle *et al.,* 2013). Recent agricultural practices greatly rely on chemical pesticides to maintain high crop yields. However, due to the application of insecticides, insect pollinators are exposed to various chemicals in the environment. When these pollinators visit insecticides treated crops for pollens or nectar, these are exposed to insecticides (Mullin *et al.,* 2010). Resultantly they returned to their nest with pollens or nectars that contain insecticide residues. These pesticide residues affect other colony members and brood (Johnson *et al.,* 2010). The intensity of pesticide to which bees are exposed depends on the amount which is applied to the target crop (Thompson and Maus, 2007). Lambda-cyhalothrin is a pyrethroid insecticide. It was registered in 1988 by the U.S. Environmental Protection Agency (EPA, 1988). Pyrethroids disrupt the sodium channel gates by keeping them it in the open position. Prolonged excitation of nerve fibers occur due to delayed closing of sodium channel gates (WHO, 1990). It causes rapid paralysis and death to an insect when ingested or exposed externally (Tomlin *et al.,* 1997). Aim of this study was to evaluate the

147-PUJZ-71028010/17/0167-0171
Part of thesis
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residual effects of lambda-cyhalothrin on common insect pollinators in the field and semi-field conditions (for *Apis mellifera*).

MATERIALS AND METHODS

The study was conducted at Department of Botany, University of Sargodha and University College of Agriculture, Sargodha, Pakistan. For the study, one patch of marigold "*Calendula officinalis*" (24 feet long and 10 feet wide) was selected. Residual effect of lambda-cyhalothrin on different types of pollinators (*i.e.,* honey bees, syrphid flies, bumble bees, butter flies) that visit marigold plant from 9-12am was recorded. The marigold plants are very beautiful and unique for their flower colours. Large number of pollinators visits these flowers. Before treating the marigold plants with insecticide, the number and types of pollinators that visited these plants from 9-12am was recorded for three days. Floral visits were also counted for each pollinator individually coming from outside into the patch. After recording data of three days, the marigold patch was sprayed with recommended dose of lambda-cyhalothrin (0.093gm/ml) using Knapsack Hand Sprayer*.* To study the residual effect of lambda-cyhalothrin on pollinators readings were taken continuously for ten days after insecticidal spray.

Toxic effects of lambda-cyhalothrin on honey bee, Apis mellifera

To examine toxic effect of lambda-cyhalothrin on *Apis mellifera*, a semi field experiment was conducted. For this study, two pots (30cm square each) containing three marigold plants with flowers (2.5 feet high) were used. The pots were covered with transparent sheet and a piece of net was adjusted on one side for ventilation. Flowers of one pot were sprayed with water and it was considered as control. However, the plants of second pot were sprayed with recommended dose of lambda-cyhalothrin (0.093gm/ml). After one hour of insecticidal spray 20 bees were released in each pot very carefully and the setups were closed again properly. Data of mortality was recorded after every 4 hours till 24 hours. The experiment was replicated thrice.

Statistical analyses

Analysis of variance (ANOVA) was used to compare the mortalities at various days after the treatment. Probit analysis was used to record the LT_{50} and LT_{90}. Two-sample t-test was used to compare the mortalities between control and experimental group. The results were considered significant if $p<0.05$. MINITAB (version 13.2) was used to analyze the data.

RESULTS

It is depicted in the Table I that before the application of lambda-cyhalothrin, the mean number of honey bees (mean of three days data) that visited the marigold field patch was higher than after the insecticidal spray. The number of honey bees progressively decreased after application of lambda-cyhalothrin.

Table I: Residual effect of Lambda-cyhalothrin on abundance of pollinators in marigold field patch.

Insect pollinator	Before insecticide application (Mean)	No. of insect pollinators									
		After insecticide application									
		Days									
		1	2	3	4	5	6	7	8	9	10
Honey bees	209	173	102	98	69	89	51	60	84	51	40
Syrphid fly	7	8	16	11	13	9	7	4	10	4	5
Bumble bees	2	3	3	2	3	3	2	4	3	1	0
Butter flies	20	25	12	13	22	14	4	11	16	2	3
Others	16	19	15	9	8	17	4	9	5	10	7
Total	**254**	**228**	**148**	**133**	**115**	**132**	**68**	**88**	**118**	**68**	**55**

The enduring effect of lambda-cyhalothrin sustained in field till 10th day after insecticidal spray (Table I). Effect of lambda-cyhalothrin on other pollinators (*i.e.,* syrphid fly, bumble bees and butterflies) was not obvious. When we compared the number of honey bees and other insect pollinators at different days after lambda-cyhalothrin spray, a considerable difference in their abundance was observed (Table II).

Table II. Results of analysis of variance showing comparison of honey bees number with other insect pollinators.

Difference	Sum of Squares	DF	Mean Square	F	P- Value
Between Pollinators	63689.382	4	15922.345	27.390	P<0.01
Within pollinators	29066.000	50	581.320		
Total	92755.382	54			

To record the effect of lambda-cyhalothrin on survival of honey bees, a semi-field experiment was performed. During this experiment, no mortality was recorded in the control group though; in the experimental group, two deaths were observed after 4 hours of insecticidal spray and this number increased to 15/20 after 12 hours. After 16 hours, all honey bees died in the experimental group (Figure I; $P < 0.05$ at all time intervals). The calculated LT_{50} and LT_{95} were 9.51 hours and 12.31 hours, correspondingly.

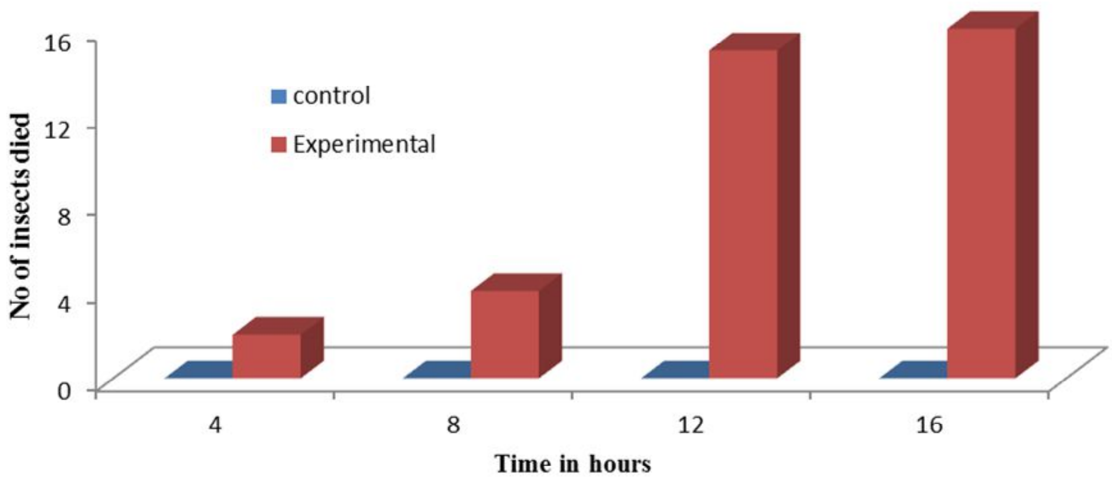

Figure 1. Effect of lambda-cyhalothrin on honey bees under semi-field conditions.

DISCUSSION

It has been observed during the study that application of insecticide in selected patch decreases the number of pollinators. Pollinators keep away from such plants which are treated with insecticide. The mode of action is not fully understood, but visual, olfactory, gustatory and chemical cues may be involved (Ramirez *et al.,* 2005). Such repulsive effects of insecticides on honeybee foraging have been reported several times (Pike *et al.,* 1982; Shires *et al.,* 1984). We found noteworthy change in the behaviour of pollinators. Before application of insecticide pollinators visit the selected patch regularly and but after application of insecticides a notable change was observed in their visiting activity (Vaidya *et al.,* 1996).

In the present study lambda-cyhalothrin was found to be highly toxic to the pollinators. Similar results that lambda-cyhalothrin is toxic to insect pollinators have also been reported by several other researchers in the fields and in the laboratory experiments (Arzone and Patetta, 1986; Prakash and Kumaraswami, 1984; Rieth and Kevin, 1987; Shivrana and Jain, 1994). The repellent action of lambda-cyhalothrin to honey bees was also observed by Fries and Wibran (1987) and reported that this insecticide is toxic to the honey bees. Our semi-field experiment showed that lambda-cyhalothrin caused 100% mortality after 16 hours. Similarly in field experiment, we observed that after spray number of insect pollinators in the experimental patch was reduced and the residual effect of lambda-cyhalothrin remained in field till 10th day. Our results are contradictory to Lewis *et al.* (1990), who carried out the studies to assess the effect of lambda-cyhalothrin residues on honey bees in semi field experiment and found non-significant mortality of honey bees.

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