AJAB

Asian J Agri & Biol. 2017;5(3):126-132.

Original Research Article

Effect of imidacloprid and bifenthrin on predation efficiency of *Coccinella septempunctata* (Coleoptera: Coccinellidae) under laboratory conditions

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Received:
May 19, 2017

Accepted:
July 17, 2017

Published:
September 30, 2017

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Abstract

Coccinella septempunctata is an important natural enemy of several pest species attacking various crops in Pakistan. Mostly pests are managed by the use of pesticides in crops and ornamentals, thus, impact of pesticides needs to be evaluated on voracity of predatory beetle. The effect of Imidacloprid and Bifenthrin on predation of adult beetles predating on Russian wheat aphid, *Diuraphis noxia* (Mordvilko) was evaluated under laboratory conditions (25 ± 2 ºC and 65 ± 5 % RH) at Systematics and Pest Management Laboratory, Department of Zoology University of Gujrat. The significant differences in the mortality of *C. septempunctata* were observed amongst control and other treatments. The adult beetles showed highest mortality (91.66 % and 83.83%) when subjected to Bifenthrin (0.7%) after 48 and 24 h of exposure, respectively. Whereas the adult beetles demonstrated highest mortality (75 % and 72.66 %) when exposed to Imidacloprid (0.7%) after 48 and 24 h of exposure, respectively. The statistical analysis yielded non-significant differences were recorded at lower concentrations of both pesticides. *C. septempunctata* showed lower mean predation of 20.66, 18.66 and 16 when exposed to 0.01, 0.02 and 0.03 % Bifenthrin after 24 h, respectively. The voracity of *C. septempunctata* was significantly affected by Imidacloprid and Bifenthrin. The study emphasized that non target beneficial species are affected due to residual insecticidal spray effects reducing their population and increasing more dependency on hazardous chemical toxicants.

Keywords: Pesticides, Voracity, Non-target species, Coccinellids

Introduction

Environmental hazards and costs are the key considerations while devising pest management strategies. Biological control agents, such as predators and parasitoids, are major component of integrated management of insect pests (Ahmad et al., 2011). Integrated pest management practices have been considered the best option for the management of insect pests. Pesticides as an effective and rapid control option is the most preferred strategy of pest management by the end users despite it poses serious threats to environment by developing resistance in pests, resurgence of various pests, pest outbreaks and a big menace to non-target beneficial insects (Solangi et al., 2007; Khaliq et al., 2007).

The integrated pest management programs are in the limelight from last three decades as the exclusive dependence on pesticides has been denting biodiversity and environment seriously. Coccinellid predators are the amongst major biological control agent that are naturally controlling pest populations in all natural terrestrial ecosystems and agro-ecosystems (Rain et al., 2016). In the current scenario of extensive use of pesticides in agro-ecosystems, the efficacy of predation and impact of pesticides on non-target species need to be evaluated time and again in order to develop sustainable IPM programs for agricultural and horticultural pests. This scenario explains the significance of evaluating existing and new pesticides for their effectiveness and environmental implications in the natural systems where biological control agents are acting as the friends of human by reducing the pests to their threshold levels. Coccinellidae family consists of about 5200 described species worldwide (Hawkeswood, 1987; Sarmad et al., 2015).

The perusal of literature indicated that about 75 species of predatory Coccinellids have been reported (belonging to 37 genera; 2 subgenera; 14 tribes and 05 subfamilies) from Pakistan (Rafi et al., 2005; Arif et al., 2011). Coccinellid beetles are amongst the effective predators of sucking insect pests (Dixon, 1998; Inayat et al**,** 2011).

Aphids, the destructive sucking pests of agricultural and horticultural crops, distributed worldwide with recorded species of about 4000 infesting over 250 crops (Ali and Rizvi, 2007). The ladybird beetles serve as natural control agents against insect pests of crops and in many cases are being utilized for the effective control of aphids and other insect pests all over the world. The perusal of literature indicated the harmful impact on the beetles predating on aphids but little work have been found on efficacy of predation, life cycle of predators and mortality of predators (Razaq et al., 2005; Cabral et al., 2011).

The study was undertaken to evaluate the impact of insecticidal spray on the mortality of adult beetles and efficacy of predation of *C. septempunctata* under laboratory conditions. The effect of Imidacloprid and Bifenthrin was evaluated on the predation efficiency of adults of *C. septempunctata* when sprayed on plant parts hosting aphid preys. The study emphasized on the impact non target species while managing pests through chemical control agents.

Materials and Methods

The experiment was conducted in March-April, 2016 in the Laboratory of Systematics and Pest Management, Department of Zoology, University of Gujrat, Punjab, Pakistan. The research work was carried out to evaluate the impact of different concentrations of Imidacloprid and Bifenthrin on the mortality of *C. septempunctata* and its predatory potential of Russian Wheat Aphid, *Diuraphis noxia* (Mordvilko). The experiment was carried out under laboratory conditions (Table 1 & 2). Toxicity of Imidacloprid and Bifenthrin against adult ladybird beetles was evaluated by contact method using wheat leaves in the laboratory bioassay under laboratory conditions (25 ± 2 ºC and 65 ± 5 % RH)

Adults of Coccinellid beetles, *C. septempunctata* were collected from fields adjacent to the Hafiz Hayat Campus, University of Gujrat, in 2016. These beetles were reared on wheat aphid under conditions suggested by earlier researchers to establish colonies in the laboratory (Naveed et al., 2007).

Specimen collection and identification

The specimens of adult ladybird beetles and aphids were collected from wheat fields adjacent to the Hafiz Hayat Campus, University of Gujrat. The beetles were collected by using sweep net whereas aphids were collected by removing twigs from the plants. The beetles as well as aphids were collected from the same fields as to maintain the predator-prey preference. The specimens were brought in the laboratory soon after collection.

The experimental animals were kept in petri dishes (9 x 1.5 cm) covered with plastic lid having proper ventilation openings. The specimens were subjected to identification by following Taxonomic keys and using microscope (ZMS-9). The adult of *C. septempunctata* were exposed to dry residues of Imidacloprid and Bifenthrin the way suggested earlier (Cabral et al., 2011).

Preparation of insecticidal treatments

The experimented was conducted in Completely Randomized Design (CRD) with three replications. The doses were prepared for the selected insecticides Imidacloprid and Bifenthrin by following formula:

$$\textbf{Volume of Insecticide}(\textbf{ml}) = \frac{\text{Total volume (L) x Percentage of insecticde required (\%)}}{\text{Formulation of insecticide}}$$

Table 1: Treatments of Bifenthrin and Imidacloprid to Determine Mortality (%) of *C. septempunctata*

Bifenthrin		Imidacloprid	
Treatments	Concentrations (%)	Treatments	Concentrations (%)
T_1	0.0 (Control)	T_1	0.0 (Control)
T_2	0.1	T_2	0.1
T_3	0.2	T_3	0.2
T_4	0.3	T_4	0.3
T_5	0.4	T_5	0.4
T_6	0.5	T_6	0.5
T_7	0.6	T_7	0.6
T_8	0.7	T_8	0.7

Table 2: Treatments of Bifenthrin and Imidacloprid to Work out Voracity of *C. septempunctata*

Bifenthrin		Imidacloprid	
Treatments	Concentrations (%)	Treatments	Concentrations (%)
T_1	Control (no insecticidal treatment)	T_1	Control (no insecticidal treatment)
T_2	0.01	T_2	0.01
T_3	0.02	T_3	0.02
T_4	0.03	T_4	0.03

Exposure to insecticidal treatments

The pre counted and treated aphids and beetles were placed in petri dishes along with wheat leaves as per procedure used by Desneux et al., (2007) with little modifications. Each replication contained 20 beetles and adequate number of pre-counted aphids. The treatment was applied on each group by topical exposure procedure to control individual dosage (and prohibited possible anti-feedant consequence of insecticides. For both pesticides, fresh solutions were prepared by diluting with distilled water at the doses recommended by the manufacturer for the control of aphids (Bifenthrin 250 ml/Acre; Imidacloprid 100- 250 ml / Acre). The solutions were prepared as following Fatima et al., (2016).

Data collection and analysis

The data of the mortality of beetles and aphids and predation was recorded after every 24 and 48 h of insecticidal exposures (Fatima et al., 2016). The data collected on the mortality of beetles was corrected by subjecting it to the Abbotts's Formula (Abbott, 1925):

Corrected Mortality

$$= \frac{\text{Mortality in Treatment [\%]} - \text{Mortality in Control [\%])}}{(100\% - \text{Mortality in control [\%])}}$$

Whereas the predation potential of *C. septempunctata* was determined by using voracity model (Soares et al., 2003).

The data collected on mortality and aphid voracity of adult coccinellid beetle, *C. septempunctata* was subjected to ANOVA by using SPPSS 21 and means were compared for significance (Zar, 1996).

Results and Discussion

The series of concentrations of Imidacloprid and Bifenthrin were tested against the mortality of *C. septempunctata* in order to calculate the LC_{50} and LC_{90} by Probit Analysis using SPSS 21. On the basis of preliminary screening of pesticide concentrations, seven concentrations of each Imidacloprid and Bifenthrin were applied to find out mortality of beetles after 24 and 48 h in comparison with control (Table 3).

Table 3: LC_{50} and LC_{90} Values for Imidacloprid and Bifenthrin at Different Times of Exposure

Insecticides		Bifenthrin	Imidacloprid
24 h	LC_{50}	0.053452	0.093287
	LC_{90}	0.193536	0.33777
48 h	LC_{50}	0.029664	0.043858
	LC_{90}	0.098883	0.146199
Overall	LC_{50}	0.039593	0.062787
	LC_{90}	0.145805	0.231222

The beetles were also subjected to insecticidal treatments to evaluate the impact of these pesticides and their concentrations on the non-target species (*C. septempunctata*) and predation efficiency.

Mortality (%) of *C. septempunctata* after 24 and 48 h

The data on mortality of adult *C. septempunctata* at 24 and 48 h after the application of treatments was given in (Figure 1). The data showed significant differences between various two duration were observed ($F_{(1, 89)}$ = 86.15, $P < 0.05$). The higher mortality was recoded at 48 h at all concentrations of both pesticides. There was no mortality in the control after 24 h, however, after 48 h non-significant mortality (2.66 & 4.16 % in Bifenthrin and Imidacloprid, respectively) after 48 h was observed which was non-significant with T_2 and T_3 (Figure 1). Overall mean mortality (%) of adult *C. septempunctata* at 24 (25.88 %) and 48 (45.55 %) h of insecticidal exposure indicated significant differences. The data on mortality exhibited significant differences between different concentrations ($F_{(14, 89)} = 33.89$, $P < 0.05$). Adult *C. septempunctata* showed highest mortality (91.66%) the when Bifenthrin (0.7%) after 48 h followed by beetle mortality (83.83%) after 24 h when exposed to same pesticide and concentration. Whereas lower mortality was recorded at lower concentrations of both pesticides and which was non-significant at T_1, T_2 and T_3 in Imidacloprid but significantly different from other treatments (Figure 1).The data indicated significant differences between two pesticides ($F_{(1, 89)} = 139.89$, $P < 0.05$).The highest values of mortality of 75 % were recorded followed by 72.66 % when beetles were exposed to Imidacloprid (0.7%) for 48 and 24 h, respectively.

The data indicated higher mortality (%) of adult *C. septempunctata* when the concentration was increased for both pesticides after 24 and 48 h. The findings of this study are in confirmation with earlier results presented by various researchers that Imidacloprid and Bifenthrin showed toxicity to both target and non-target species (Amer et al., 2010). Higher toxicity levels were observed in Bifenthrin as compared to Imidacloprid (Amin et al., 2014; Devee and Baruah, 2010). Predation rate decreased and mortality rate increased with the increase of insecticidal concentrations (Fernandes et al., 2016). Imidacloprid showed higher values of LC_{50} and LC_{90} as compared to Bifenthrin are in confirmation with the results presented in this study that depicted higher mortality in the beetles exposed to Bifenthrin. Higher mortality rates were observed after 24 and 48 h by exposure of Bifenthrin and Imidacloprid when mealy bugs were exposed to different concentrations (Fatima et al., 2016).

In addition to the direct effects presented in these results in the form of beetle mortality induced by pesticides, there may be some indirect effects of lethal and sub-lethal concentrations and repeated exposures on insect populations surviving leading to alterations in the longevity, reproduction, fecundity, development time, feeding behavior, mobility, and prey acceptance, etc.(Ruberson et al., 1998). Such sub-lethal effects more pronounced in the case of newly developed insecticide (Wright and Verkerk, 1995; Ochiai et al., 2007).

Mean voracity by *C. septempunctata*

The data on the voracity of *C. septempunctata* adults was worked out for comparison between treatments by applying ANOVA ($P < 0.05$). The results presented in Fig. 2 showed significant results for treatments and duration ($F_{(2, 43)} = 23.57$, $P = 0.0000$, $F_{(1, 43)} = 10.40$, $P = 0.0024$). Predation rate decreased with increase in the concentration of insecticidal concentration highest predation was showed in control group (56 and 82.66 at 24 and 48 h, respectively) where there was no exposure of insecticides to the experimental unit. The voracity of *C. septempunctata* was higher when predators were added to control group where no insecticidal spray was carried out whereas lower voracity was observed when topical insecticidal spray was applied. The results showed positive increase in the voracity in control whereas a decreasing trend was noticed in all other treatment which may be attributed to non-feeding repellency and poisonous hazards caused by the pesticides. This may also have indirectly influenced their non-preference to their prey due to insecticidal effects on prey.

C. septempunctata showed significantly low predation by 20, 16, 18.66 after 24 h when exposed to Bifenthrin (0.01, 0.02 % & 03 %) respectively, whereas 36.33, 29.33, 21.66 after 48 h when exposed to Bifenthrin (0.01, 0.02 % & 03 %) respectively, as compared to control. This indicated that predation was lowest at higher concentrations of Imidacloprid and Bifenthrin (Figure 2). The aphid consumption of *C. septempunctata* decreased under insecticidal spray. The decrease in the aphid consumption with the increase in the concentrations of insecticidal from 0.01 to 0.03 % solutions of both pesticides may be attributed to physiological alterations caused by the pesticides in both predator and prey. This may be due to non-preference of Coccinellid beetles of the insecticidal sprayed aphids. The earlier studies endorsed the fact that the reduced consumption of prey resulted when insecticide treated prey were fed and changes in the consumption of aphids by Coccinellid beetles were recorded (Thornham et al., 2007; Qi et al., 2001). This may also be due to modified habitat and limited space for countering the stress factor due to insecticides. Topical exposure to different pesticides decreases significantly the rate of aphid consumption (Provost et al., 2005; Liu and Stansly, 2004). The research works have been carried out in various parts of the world on the ingestion insecticide treated prey by Coccinellid beetles that had demonstrated negative effects of various pesticides at different concentrations on non-target species directly by causing mortality or indirectly by reducing longevity, fecundity, oviposition period and behavior (Angeli et al., 2005; Wang et al., 2005; Thornham et al., 2008; Ujjan et al., 2014). The studies also demonstrated that the efficacy of predation decreases with the use of insecticides to control pests. The result showed that there is serious decline in the aphid consumption in the insecticide treated groups which may be due to knockdown effect, locomotory and behavioral recorded by Yuxian et al., 2012. The work carried out by them reported sublethal effects of imidacloprid affect adversely development and reproductive capacity and may lead to lower population of the coccinellid predator. Imidacloprid residue lowers predator voracity.

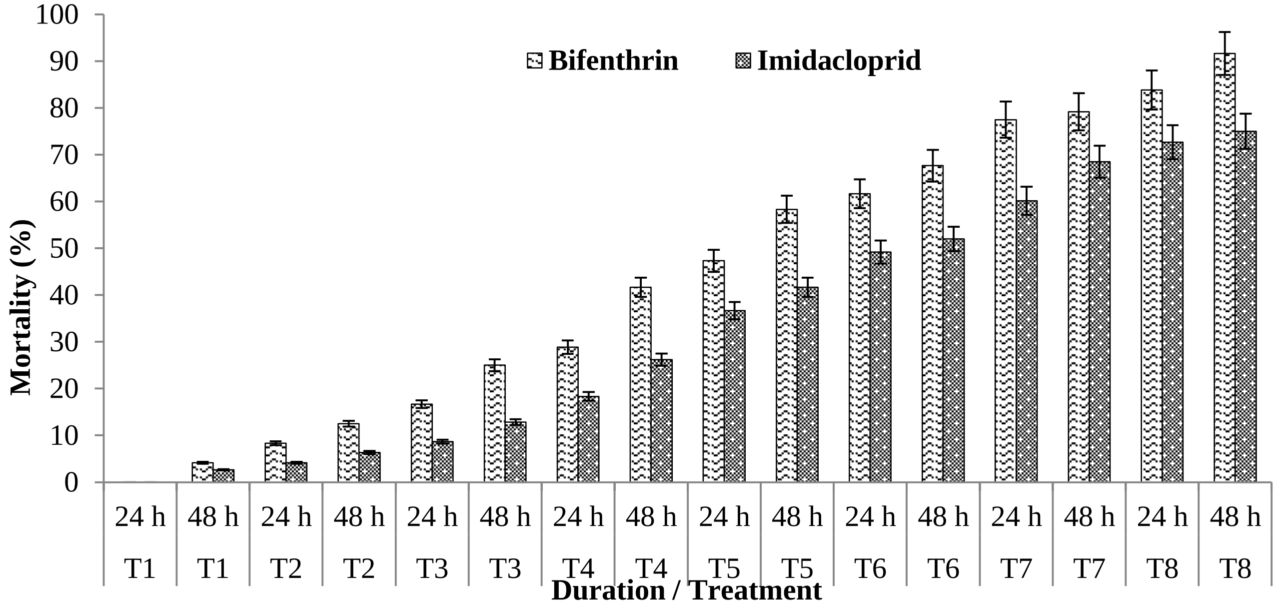

Figure 1: **Mortality (%) of Adult *C. septempunctata* after 24 and 48 h of Exposure to Different Concentrations of Imidacloprid and Bifenthrin**

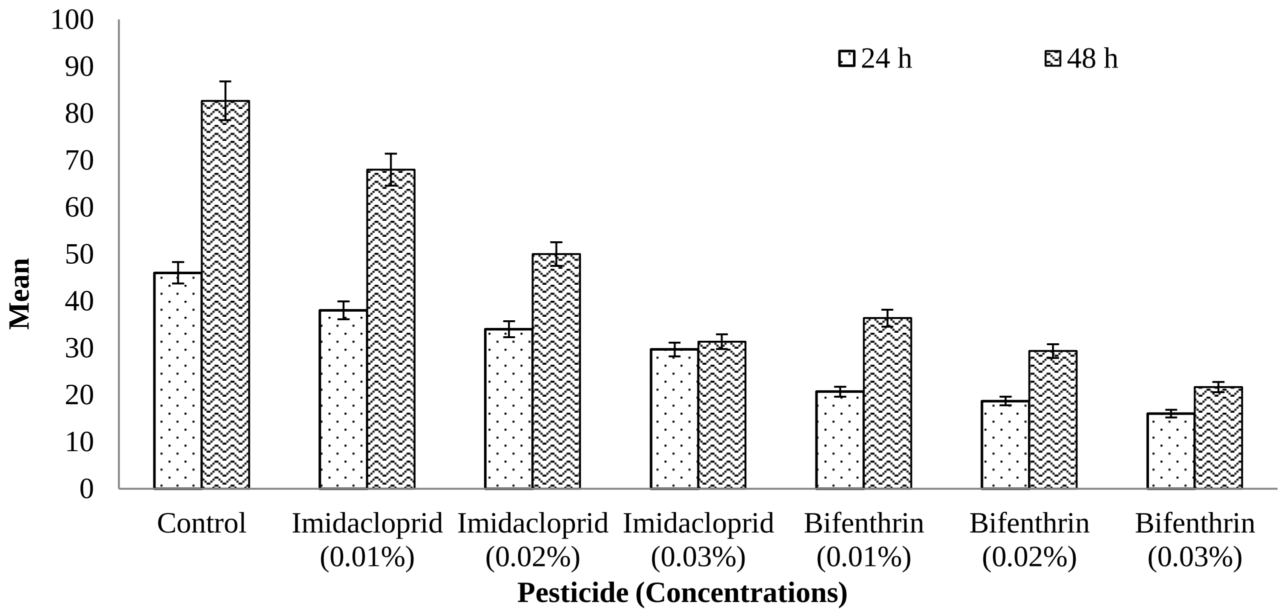

Figure 2: **Mean Voracity of *C. septempunctata* adults when treated with Bifenthrin and Imidacloprid or distilled water (control) using *Diuraphis noxia* as prey**

Conclusion

C. septempunctata showed negative response to the insecticidal treatments for both beetle mortality and decreased aphid consumption. The exposure of coccinellid beetles to Imidacloprid and Bifenthrin, may affect physiology and behavior of beetles as a result of ingestion of treated prey. Our study focused on the two aspects of insecticidal effects on non-target organisms at higher concentrations of pesticides in accordance with the LC_{50} and LC_{90} (LC_{50} for Bifenthrin 0.039593and LC_{50} 0.062787for Imidacloprid whereas LC_{90} for Bifenthrin 0.145805 and LC_{90} 0.231222 for Imidacloprid). The study suggested that there is need to protect natural enemies from residual effect of chemical sprays for the protection, management and conservation of biodiversity. The sub-lethal doses may also affect the biology of Coccinellid beetles significantly, thus, biological control agents need to be reintroduced and augmented in the areas where integrated pest management programs are dominated by chemical control of insect pests. The toxic effects of insecticidal exposures to ladybird beetles have agricultural and ecological importance due to their predatory significance in crop protection (Yu et al., 2014).

References

Abbott WS, 1925. A method of computing the effectiveness of an insecticide. J. Econ. Entomol. 18: 265-267.

Ahmad M, Rafiq M, Arif MI and Sayyed AH, 2011. Toxicity of Some Commonly Used Insecticides against *Coccinella undecimpunctata* (Coleoptera: Coccinellidae). Pak. J. Zool. 43(6): 1161-1165.

Ali A and Rizvi PQ, 2007. Development and Predatory Performance of *Coccinella septempunctata* L. (Coleoptera: Coccinellidae) on Different Aphid Species. J. Biol. Sci. 7(8): 1478-1483.

Amer M, Aslam M, Razaq M and Ali SS, 2010. Effect of Conventional and Neonicotinoid Insecticides against Aphids on Canola, *Brassica napus* L. at Multan and Dera Ghazi Khan. Pak. J. Zool. 42(4): 377-381.

Amin MA, Hameed A, Rizwan M and Akmal M, 2014. Effect of Different Insecticides against Insect Pests and Predators Complex on *Brassica napus* L., Under Field Conditions. Int. J. Sci. Res. Environ. Sci. 2(9): 340-345.

Angeli G, Baldessari M, Maines R and Duso C, 2005. Side-effects of pesticides on the predatory bug *Orius laevigatus* (Heteroptera: Anthocoridae) in the laboratory. Biocont. Sci Tech. 15(7): 745–754.

Arif MJ, Gogi MD, Abid AM, Imran M, Shahid MR, Husain S and Arshad M, 2011. Predatory potential of some native coccinellid predators against *Phenacoccus solenopsis*, Tinsely (Pseudococcidae: Hemiptera). Pak. Entomol. 33(2): 97-103.

Cabral S, Soares A and Garcia P, 2011. Voracity of *Coccinella undecimpunctata*: effects of insecticides when foraging in a prey/plant system. J. Pest. Sci. 84: 373–379.

Desneux N, Decourtye A and Delpuech JM, 2007. The sub-lethal effects of pesticides on beneficial arthropods. Annu. Rev. Entomol. 52: 52–81.

Devee A and Baruah AA, 2010. Relative toxicity of Imidacloprid and Bifenthrin against *Lipaphis erysimi* (Kalt.). Pesticide Res. J. 22(1): 86-87.

Dixon AFG, 1998. Aphid Ecology, 2nd edn. Chapman and Hall, London, pp. 300.

Fatima S, Hussain M, Malik MF, Noureen N and Abbas Z, 2016. Field efficacy of some insecticides against hibiscus mealybug, *Maconellicoccus hirsutus* (Hemiptera: Pseudococcidae). J. Entomol. Zool. Stud. 4(1): 240-244.

Fernandes MES, Alves FM, Pereira RC, Aquino LA, Fernandes FL and Zanuncio JC, 2016. Lethal and sub-lethal effects of seven insecticides on three beneficial insects in laboratory assays and field trials. Chemosphere. 156: 45-55.

Hawkeswood T, 1987. Beetles of Australia. Augus and Robertson, Sydney, Australia.

Inayat TP, Rana SA, Rana N, Ruby T, Sadiqui MJI and Abbas MN, 2011. Predation rate in selected coccinellid (Coleoptera) predators on some major aphidid and cicadellid (Hemiptera) Pests. Int. J. Agric. Biol. 13: 427–430.

Khaliq A, Attique MNR and Sayyed AH, 2007. Evidences of resistance to pyrethroids and organophosphate in *Plutella xylostella* from Pakistan. Bull. Entomol. Res. 97: 191-200.

Liu TX and Stansly PA, 2004. Lethal and sub-lethal effects of two insect growth regulators on adult *Delpgatus catalinae* (Coleoptera: Coccinellidae), a predator of whiteflies (Homoptera: Aleyrodidae). Biol. Control. 30: 298–305.

Naveed M, Salam A and Saleem MA, 2007. Contribution of cultivated crops, vegetables, weeds and ornamental plants in harboring of *Bemisia tabaci* (Homoptera: Aleyrodidae) and associated parasitoids (Hymenoptera: Aphelinidae) in cotton agroecosystem in Pakistan. J. Pestic. Sci. 80: 191–197.

Ochiai N, Mizuno M, Mimori N, Miyake T, Dekeyser M, Canlas LJ and Takeda M, 2007. Toxicity of Bifenazate and its Principal Active Metabolite, Diazene, to *Tetranychus urticae* and *Panonychus citri* and Their Relative Toxicity to the Predaceous Mites, *Phytoseiulus persimilis* and *Neoseiulus californicus*. Exper. App. Acarol. 43: 181-197.

Provost C, Coderre D, Lucas E, Chouinard G and Bostanian NJ, 2005. Impact of intraguild predation and lambda-cyalothrin on predation efficacy of three acarophagous predators. Pest Manag. Sci. 61: 532–538.

Qi B, Gordon G and Gimme W, 2001. Effects of neem-fed prey on the predacious insects *Harmonia conformis* (Boisduval) (Coleoptera: Coccinellidae) and *Mallada signatus* (Schneider) (Neuroptera: Chrysopidae). Biol. Control. 22: 185–190.

Rafi MA, Irshad M and Inayatullah M, 2005. Predatory ladybird beetles of Pakistan. PARC/NWFP Agric. Univ. Roohani Art Press, Islamabad, Pakistan, pp. 105.

Rain FF, Abu Faiz MD. Aslam, Sringki S, Sultana N, Akhter N and Howlader AJ, 2016. Coccinellid predators of aphid and their phylogenetic analysis using coi gene sequences. Int. J. Appl. Sci. Biotechnol. 4(3): 408-416.

Razaq M, Suhail A, Aslam M, Arif J, Saleem, AM, and Khan MHA, 2005. Evaluation of neonicotinoids and conventional insecticides against cotton jassid, *Amrasca devastans* (Dist.) and cotton whitefly, *Bemisia tabaci* (Genn.) on cotton. Pak Entomol.. 27(1):75-78.

Ruberson JR, Nemoto H, Hirose Y, 1998. Pesticides and Conservation of Natural Enemies in Pest Management. In: Barbosa P. (ed.) Conservation Biological Control. Academic Press, San Diego, CA. pp. 207-220.

Sarmad SA, Afzal M, Raza AM, Khalil, MS, Khalid H, Aqueel MA and Mansoor MM, 2015. Feeding efficacy of *Coccinella septempunctata* and *Propylea quatuordecimpunctata* against *Macrosiphum rosae*. Sci. Agri. 12(2):105-108.

Soares A, Coderre D and Schanderl H, 2003. Effect of temperature and intraspecific allometry on predation by two phenotypes of *Harmoniaaxyridis pallas* (Coleoptera: Coccinellidae). Environ Entomol. 32(5): 939–944.

Solangi BK, Lanjar AG and Lohar MK, 2007. Comparative toxicity of some insecticides on 4th instar grub of *Coccinella septempunctata* under laboratory conditions. Sarhad J. Agric. 23(4): 1091-1096.

Thornham DG, Blackwell A, Evans KA, Wakefield M and Walters KFA, 2008. Locomotory behaviour of the seven-spotted ladybird, *Coccinella septempunctata*, in response to five commonly used insecticides. Ann. Appl. Biol. 152:.349–359.

Thornham DG, Stamp C, Walters KFA, Mathers JJ, Wakefield M, Blackwell A and Evans KA, 2007. Feeding responses of adult seven-spotted ladybirds, *Coccinella septempunctata* (Coleoptera; Coccinellidae) to insecticide contaminated prey in laboratory arenas. Biocontrol Sci. Techn. 17(10): 983–994.

Ujjan ZA, Khanzada MA and Shahzad S, 2014. Insecticide and papaya leaf extract toxicity to mustard aphid, Lipaphis erysimi (kaltenbach). J Agri Food App.Sci. 2(2): 45-48.

Wang L, Huang J, You M, Guan X, Liu B, 2005. Effects of toxins from two strains of *Verticillium lecanii* (Hyphomycetes) on bioattributes of a predatory ladybeetle *Delphastus catalinae* (Col., Coccinellidae). J Appl. Entomol. 129 (1):32–38. doi:10.1111/j.1439-0418.2005.00929.32-38

Wright DJ and Verkerk RHJ, 1995. Integration of Chemical and Biological Control Systems for Arthropods: Evaluation in a Multitrophic Context. Pesticide Sci. 44: 207-218.

Yu C, Fu M, Lin R, Zhang Y, Yongquan L, Jiang H and Brock TCM, 2014. Toxic effects of hexaflumuron on the development of *Coccinella septempunctata*. Environ. Sci. Poll.Res. Int. 21(2): 1418–1424.

Yuxian, H., Juanwei, Z., Yu, Z., Desneux, N., Kongming, W, 2012. Lethal effect of imidacloprid on the Coccinellid predator Serangium japonicum and sublethal effects on predator voracity and on functional response to the whitefly *Bemisia tabaci*. Ecotoxicity, 21(5): 1291-1300. doi: 10.1007/s10646-012-0883-6

Zar JH, 1996. Biostatistical analysis. 3. London, UK: Prentice. pp. 662.