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TOXICITY OF SELECTED INSECTICIDE AGAINST DIAMONDBACK MOTH *PLUTELLA XYLOSTELLA* (L.) UNDER CONTROLLED CONDITION

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ABSTRACT

Plutella xylostella L. commonly known as diamondback moth, is the major pest of crops belonging to family *Brassicaceae* all over the world. The diamondback moth reduces the economical production of the crucifers, while several insecticide and IPM techniques have been applied for its better management, but it has developed resistance against insecticides. Five insecticides, including Chlorantraniliprole, Lufenuron, Flubendamide, Trichlorophon, and Pyriproxifen, were tested against *P. xylostella* (DBM) at the 2nd and 3rd larval stages to evaluate mortality by using the leaf dip bioassay method after a time interval of 24, 48, and 72 hours. All the insecticides were found toxic to the studied insects; however, mortality was found to be time-dependent. Flubendiamide proved to be more effective with 96.6 percent corrected mortality after 48hrs and LC₅₀ 7.035 µl/ml against *P. xylostella* immatures, while chlorantraniliprole was found to be the second most effective insecticide. The present study reveals the high potential of insecticides, so these insecticides could be evaluated further under field conditions.

Keywords: Cabbage; Insecticide; Leaf dip bioassay method; *Plutella xylostella*

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INTRODUCTION

The *Brassicaceae* (Cruciferae) family comprises approximately 350 genera, with their species comprising 3500 roundabouts for all the dicotyledonous wild herbs with hypogenous, comprehensive, and cruciform blossoms (Warwick et al., 2003). Diamondback moth (DBM) is a major crop pest universally, with some of its inhabitants being resilient to pesticides (Agboyi et al., 2016; Attique et al., 2006; Ayalew, 2006; Balasubramani et al., 2008; Bautista et al., 2009). It can cause injury to flourishing fragments of cauliflower, and due to considerable damage, it reduces about 50–80% of yield throughout unembellished infestations on the crops (Ayalew, 2006; Prashant et al., 2007). Due to insecticidal control failures, reasonable production of these crucifers has become nearly terrible in several parts worldwide. As a result, considerable efforts have been made around the world to progress IPM

(Integrated Pest Management) sequencers, which are mostly centered on manipulating natural enemies. Though it has been concluded that approximately 130 natural enemies (parasitoids) species of the DBM have been identified as attacking on its various stages, most of the species have been managed by a small number of hymenopteran species belonging to the following genera: the *Ichneumonid* genera *Diadegma* and *Diadromus*, the *Braconid* genera *Microplitis* and *Cotesia*, and the *Eulophid* genus *Oomyzus*. Hence, the population dynamics of *P. xylostella* (DBM) vary in the sense of their genetic and biological modification from different localities, although the specific diamondback moth strains lie with their specific parasitoids. Hence, the adherents of this large family contain parsimoniously vital crops such as canola and mustard from oilseeds, cabbage and cauliflower from Cole crops, and some of the root vegetables, including radish and turnips, etc. These are the

most prevalent and vital components of diverse cultures' diets, which can be cultivated in tropical and temperate regions (Sarfraz and Keddie, 2005). Regarding the entire cost of DBM to the Brassica crop production and management, we need to make sure that how much is spent on its management per hectare on its host plant (*Brassica* crop), production, and how much yield is lost despite this treatment. These costs are determined in part by the localities of diamondback moths, in spite of the fact that their lavishness diverges geographically. It also needs to be determined how lavishness is disseminated comparatively to agricultural host plants, and the economic damage caused by diamondback moths by nourishing their host crops is also monitored. The DBM larvae proliferate on all segments of their host plants, interpreting them as ailing for assimilation. The first instars are leaf miners who feed on the lower surface of plant leaves after moulting, and the last stage of their larval instars nibbles on the host portions, causing uneven patches (Golizadeh et al., 2009).

The use of synthetic chemicals for their management is a basic component of IPM, which is a shrewd plant fortification approach to diminishing the hazardous effects of chemicals on the environment and human and animal health. Hence, resistance is an inherited progression in populations that declines the efficiency of synthetic insecticides and pesticides due to their recurrent application (Trumble, 1998). This phenomenon ultimately has an undesirable influence on the IPM programs (Jensen, 2000).

MATERIALS AND METHODS

Insect culture

The diamondback moth (*P. xylostella*) culture was collected from cabbage fields in Texlia, Pakistan. Approximately 150–200 larvae and pupae of the *P. xylostella* were collected and transported to the Biocontrol Laboratory Department of Entomology at PMAS Arid Agriculture University Rawalpindi. The cabbage leaves (*Brassica oleracea* cv. *Asha*) were placed in a cage as a larval diet. Each population's adults were released into cages that

measured 70 × 70 cm, and a 10% honey solution was provided. For the purpose of egg-laying, 40-day-old cabbage plants were provided to 15 pair of adults, offered plants were changed every day to homogenize the egg-laying batch. The cages were reserved in a room with a controlled temperature and relative humidity of 25 °C, 50-60% RH, and a 13L: 11D photoperiod.

Insecticides bioassays

In the experiment, five commercial insecticides were used: chlorantraniliprole (Coragen 20% SC; FMC (Pvt.) Ltd. Pakistan) at 50 ppm, Trichlorphon (Diptrex 80% SP; ICI (Pvt.) Ltd. Pakistan) at 50 ppm, Flubendamide (Belt 480 SC; Bayer (Pvt.) Ltd. Pakistan) at 32 ppm, Pyriproxifen, and Lufenuron. The insecticides were applied to the population using the method described by Sayyed et al. (2008). In a nutshell, the leaf dip bioassay approach was applied to assess the insect pests' susceptibility and level of resistance to pesticides. The pesticides were first tested on each population to determine the concentration range at which the concentration-response connection occurs. For the preliminary testing, insecticides were tested in at least five different concentrations, and five concentrations were used for the final bioassays. To obtain the desired dose of insecticide, given chemical was diluted in distilled water. *Brassica oleracea* crop leaves were gathered from unsprayed fields, washed with water, dried, and soaked in an insecticide solution for 30 seconds before drying at room temperature for two hours. The leaves were then dried on tissue before being placed in Petri dishes coated with moistened filter paper. The leaf discs were placed in petri plates with moistened filter paper after drying. Ten second-instar larvae were transferred per Petri dish using a soft camel hair brush. Each treatment was repeated three times and was meant to be absolutely random. Each bioassay was carried out at least twice in a growth chamber (BOD) (27 °C; 65% RH). The death rate was measured 24 hours, 48 hours, and 72 hours after the larvae were placed on the treated leaves. Larvae were declared dead if their complete bodies did not respond when touched (Tabashnik et al., 1990).

Active Ingredient	Trade Name	Company	Class	Years
Pyriproxifen	Admiral 10EC	FMC	New Chemistry	2023
Lufenuron	X-Tall	Kanzo	New Chemistry	2023
Chlorentaneliprol	Coragen 20% SC	FMC	New Chemistry	2023
Flubendamide	Belt Expert® 480 SC	Bayer	New Chemistry	2023
Trichlorfon	Diptrex 80% SP	ICI	Organophosphate	2023

Analysis

Data for each pesticide and population were subjected to

probit analysis to adjust for control mortality (Finney, 1971). The replicated data were merged and poloPlus was

used for probit analysis to construct dose-mortality regressions analysis. When appropriate, bioassay results were corrected for control mortality (Abbott, 1925). Because bioassays have inherent variability, pair-wise LC50 value comparisons were performed at the 1% significant level (where individual 95% FL for two treatments do not overlap) (Litchfield and Wilcoxon, 1949). For each product, the resistance ratio was computed by dividing the LC50 value for the susceptible population by the LC50 value for the field populations.

RESULTS AND DISCUSSION

Integrated pest management (IPM) strategies have proven to be successful against *P. xylostella* in both laboratory and field settings. Many people have used cultural, biological, botanical, and chemical procedures on a national and international scale. Chemical or pesticide control is the best and most successful approach for controlling *P. xylostella* (Kumar and Yadav, 2009; Parsaeyan et al., 2013).

The percentage corrected mortality was evaluated after the time interval of 48 and 72 hours of the applied insecticides against the *P. xylostella*. In this experiments the highest

percentage mortality 90%, 86.6% were obtained against Chlorentaneliprol and Flubendamide after 48 hours at 62 and 50ppm. While Lufenuron, Trichlorphon and Pyriproxifen treatments against Diamondback moth observed 80%, 93.2% & 86.6% respectively after 72 hours at the concentration rate of 50ppm.

All the insecticides tested in this present studies caused concentration-dependent mortality in 3rd instar larvae of *P. xylostella*. Toxicity of tested insecticides against 3rd instars of *P. xylostella* after 24, 48 and 72 hours is given in Table 2. The LC50 values of Chlorentaneliprol (Coragen 20% SC) after 24 and 48 h were 50.79 and 9.34 µg/ml, respectively. While the LC90 values vary from 292.54 and 91.94 after subsequent time interval. Approximately all the tested larvae were found dead after 72h therefore Lc50 was not estimated for this time. The Chlorentaneliprol was observed high lethal insecticide than Flubendamide and Lufenuron. The LC50 and LC90 values of Flubendamide after 24 and 48 were 41.70, 7.03, 286.34 and 87.45 respectively as shown in table 2. As Leufenuron is insect growth regulator therefore it acts slowly thereby The LC50 values of Lufenuron after 72 hours was also calculated and represented in the table 1.

Table 1: corrected mortality of insecticides after 48/72 hours of post-treatment application.

Insecticide	Dose applied (ppm)	Corrected Mortality (%)
Chlorentaneliprol (48 h)	62	90
Flubendamide (48 h)	50	96.6
Lufenuron	50	80
Trichlorphon	50	93.2
Pyriproxifen	50	86.6

Table 2. Exhibits the toxicity of pesticides against the third larval instar of *P. xylostella* L. in the laboratory using the leaf dip bioassay method.

Insecticide	Time	LC50 µg/ml	LC90 µg/ml	Slope± S.E	χ ²	df	N
Chlorentaneliprol	24	50.794	292.54	1.685±0.497	1.948	3	180
	48	9.334	91.948	1.290±0.361	1.013	3	180
Flubendamide	24	41.707	286.34	1.532±0.438	0.717	3	180
	48	7.035	87.454	1.171±0.369	0.341	3	180
Lufenuron	24	59.626	604.142	1.274±0.454	0.717	3	180
	48	29.309	396.00	1.133±0.383	0.908	3	180
Trichlorphon	72	14.071	174.907	1.171±0.369	0.341	3	180
	24	82.151	1736.913	0.976±0.208	0.959	3	180
Pyriproxifen	48	31.308	228.975	1.544±0.130	0.548	3	180
	72	10.597	68.874	1.926±0.103	0.974	3	180
Lufenuron	24	75.159	1873.237	0.918±0.216	1.000	3	180
	48	19.164	547.788	0.880±0.207	0.997	3	180
Pyriproxifen	72	7.708	55.970	1.165±0.160	0.991	3	180

The calculated LC50 values of Lufenuron at 24, 48 and 72h were as 59.62, 29.30, 14.07 and LC90 were 604.14, 29.30 and 174.90 were evaluated. The LC50 values of Trichlorophon after 24, 48 and 72 hours were calculated and it represent that was 82.15, 31.30, 10.59 and LC90 were 1736.91, 228.97 and 68.87 were evaluated. The LC50 values of Pyriproxifen after 24, 48 and 72 hours were calculated and it represent that the LC50 values followed according to mentioned time duration of recording the mortality. The calculated LC50 values at 24, 48 and 72h were as 75.15, 19.16, 7.70 and LC90 were 1873.23, 547.78 and 55.97 were evaluated.

The assessment of insect vectors' susceptibility to commonly employed insecticides is a crucial factor in determining the optimal and efficacious insecticide for use (Abbas et al., 2021). In present study insecticidal bioassays were conducted to estimate the toxicity induced in larval population of *P. xylostella*. Previously Hill and Foster (2000) and Sarfraz and Keddie (2005) have conducted trial to estimate conserving efficacy of insecticides against DBM. However, these studies are the older one, recent study provide updated outlook on the efficacy of modern insecticides against *P. xylostella*. All the insecticides were found toxic to studied insect however mortality was found to be time depended. The toxicity reaches to peak value after 48 or 72 hrs of post treatment application. Flubendamide was found to be more potent as compared to other insecticides. The findings of Hirooka *et al.* (2007), suggest that Flubendiamide is more effective on the larvae of *P. xylostella*. Similarly, Sunitha and Pandurang (2015) and do Carmo *et al.* (2023) also observed that Flubendamide exerted more effective chemical toxicity as compared to other chemicals applied. This might be because Flubendamide is a Diamide insecticide which targets ryanodine receptors-sensitive cytosolic Ca²⁺ transients leading to impairment with muscle contraction (Troczka *et al.*, 2017). The second effective insecticide was chlorantraniliprole in the present study with 50.79 and 9.33 median lethal concentrations after 24 and 48hrs of treatment application. The findings of Han *et al.* (2012) are consistent with the current findings, indicating that this chemical has a considerable insecticidal potential against DBM. The lowest median lethal concentration of Lufenuron was 14.07 µg/ml representing its effectiveness after 48hrs of post treatment application while Flubendamide and Chlorantraniliprole showed maximum toxicity after 48hrs. The findings of Akhtar *et al.* (2021) also suggested that neonicotinoids and Diamides were highly toxic to *C. flavipes* adults and caused 100% mortality at 48 h of exposure while Lufenuron

toxicity was slower. As mentioned earlier formers are the Diamides insecticides ceasing the mobility while later one is the IGR, affecting the developmental stages making it chronic chemical. However, in a long run the effect of Lufenuron can be observed on coming generations and within developing instars (Hafez and Abbas, 2021; Kopit and Pitts-Singer, 2018). The current study demonstrates the high potential of pesticides that can be used in the field to successfully manage *P. xylostella*. However, additional investigations pertaining to resistance development mechanisms and target-site mutations are required to estimate the sustainability of these chemicals against insects.

CONFLICTS OF INTEREST

The authors declare conflicts of interest.

AUTHOR'S CONTRIBUTION

All authors contributed and supported towards writing of this manuscript.

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