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Research Article

An exploratory study on the insecticidal potential of various chemical agents against *Bactrocera cucurbitae*

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ABSTRACT

Bactrocera cucurbitae is the most destructive species of fruit fly. It is a serious pest of several fruits in South Asian nations like as Pakistan, India, and Malaysia. During these studies, toxicity of fipronil, spinosad, emamectin-benzoate, bifenthrin, cypermethrin and deltamethrin were observed by two different methods i.e., diet incorporate method and contact method against laboratory reared population of *B. cucurbitae*. The culture was reared at 28 ± 2°C temperature with 65 ± 5% relative humidity and 12:12 (D/L) photoperiod in the laboratory. According to the results in diet incorporate method, spinosad was found to be the most efficient insecticide against *B. cucurbitae* as it showed the least LC_{50} i.e., 2.86 after 72 hours. It was followed by emamactin-benzoate. The LC_{50} value of emamectin-benzoate was 2.98 after 72 hours against *B. cucurbitae*. The third most efficient LC_{50} was 4.95 of spinosad after 48 hours. According to the results in contact method, cypermethrin was found as most efficient insecticide against the *B. cucurbitae* as it showed least LC_{50} i.e., 0.62 after 72 hours. It was followed by insecticide of the same group, i.e., bifenthrin. The LC_{50} value of bifenthrin was 0.63 after 72 hours against *B. cucurbitae* and the third most efficient insecticide was deltamethrin which had 1.06 LC_{50} values after 72 hours. Hence, the results showed that LC_{50} values of contact method were lower than diet incorporated method; so it is the most efficient method to control melon fruit fly among the two tested methods and cypermethrin is the most effective insecticide.

Keywords: Toxicity; Bactrocera cucurbitae; contact method; diet incorporate method.

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INTRODUCTION

Horticultural crops have made a significant contribution to global food production. These crops have high importance because of their high export and yield values. In 2005 the horticulture production was 150.73 million tons (Choudhary, 2021). The import of horticultural commodities were 1,769.25 million USDs in 2015 (Jha et al., 2019). These major crops face various challenges in the field. Many insect pests damage horticultural crops, however, fruit flies are the most common insect pest with excessive eating habit, reproduction rate and adoption in different regions like attributes make fruit flies serious insect pest of vegetables and fruits all over the world (Sarwar, 2015). Family Tephritidae of order Diptera comprises nearly 4500 species, out of which about 700 are fruit flies and the largest genus is *Bactrocera* with 552 species (Freidberg, 2006) with one-third species being economically important. These are found everywhere in tropical, subtropical & temperate zones on the globe (Christenson and Foote, 1960). *Bactrocera* genus comprises 43 species out of which *Bactrocera cucurbitae* (Diptera: Tephritidae) is the supreme and harmful insect of cucurbits (Shah et al., 1948) and in Asia it is the most damaging one (Nagappan et al., 1971).

Depending upon the season and crop, these fruit flies cause 30-100% economic losses (Hollingsworth et al., 1997). Fruit invasion by *B. cucurbitae* has been observed from 41-89% in *M. charantia* (Rabindranath and Pillai, 1986). Damage to bitter gourd in India had been observed 31.27 % due to this pest and on watermelon 28.55 % damage was recorded (Singh et al. 2000). In Papua New Guinea; the damage by *B. cucurbitae* was recorded as 95 % on bitter gourd and on snake gourd was 90 % and 60-87 % on pumpkin respectively in Solomon Islands (Hollingsworth et al., 1997).

B. cucurbitae is found everywhere in the world but is native to India (Shah et al., 1948). It is a major insect pest of South Asia particularly in Pakistan (Fletcher, 1987), China, Malaysia and Indonesia (Weems and Heppner, 2001). The melon fruit fly is also present in other Asian nations, including Nepal, Myanmar, Sri Lanka, Singapore (Narayanan, 1953), the Philippines (Hardy, 1949), Japan, Vietnam, Thailand, and Bhutan (Vasudha and Agarwal, 2019). Reports indicate its presence in Cameroon (Fontem et al., 1999), Egypt, Kenya, Tanzania, Mauritius, and East Africa (Weems and Heppner, 2001). The Australian Ocean melon fruit fly is present in Australia (Fletcher, 1987), the Solomon Islands (Hollingsworth et al., 1997), the Mariana Islands, the Hawaiian Islands, and Papua New Guinea (Weems and Heppner, 2001). Nonetheless, there are countries free of fruit flies, such as Chile, which accounts for almost 50% of global fruit output (Retamales and Sepúlveda, 2011).

B. cucurbitae survives in the field all over the year because a large number of host plants is available for survival. During the harsh winter, they take shelter under dried leaves of different plants. When the temperature rises, they survive by seeking refuge in humid and shaded areas. They also feed on the honeydew secreted by aphid on the different fruit plants. According to Weems et al. (2012) there are 125 plants species known as host of *B. cucurbitae*. Among which bitter gourd (*Momordica charantia*), snake gourd and muskmelon (*Trichosanthes cucumeria and Trichosanthes anguina*) are heavily damaged (Doharey, 1983).

B. cucurbitae is widely distributed key insect pest throughout the World. Most of the major cucurbit vegetable crops are severely attacked by this single pest. As the cucurbit fruits are harvested on a frequent basis for trade and domestic use, it's impossible to depend on any insecticide to control this insect due to its cryptic nature. Therefore, there is a basic need to curb this devastating menace by initiating control measures with some new and safer chemicals such as acephate or chlorfenapyr having antiovicidal effect or spinosad having rain fastness characteristics.

Being the major pest of horticultural crops melon fly cause severe damage in vegetable and fruits which results in decline of per acre yield of the horticultural crops. So, control is necessary to save crops and production of vegetables and fruits. Different control tactics are being used in the field to control the *b. cucurbitae* but chemical control is preferred by the farmer.

Few insecticides have been proved effective as cover sprays against melon fly depending upon the plant diamethoate, fenthion and trichlorfon are efficient to suppress melon fly population (Dominiak, 2013). The bio efficacy of different commercially available insecticides against melon fruit fly was determined during this study.

MATERIALS AND METHODS

Insecticides

Commercial formulations fipronil (Agenda 25EC, Bayer Crop Science (Pvt.) Ltd.), spinosad (24SC Tracer®, Dow Agro Science, Pakistan), emamectin-benzoate (Ticket®, 1.9% EC, Sona Agro Chemicals, RYK, Pakistan), cypermethrin (Cypermethrin 10 EC, Sun crop), bifenthrin (Talstar®, 10% EC, FMC United (Pvt.) Ltd. Lahore, Pakistan) and deltamethrin (Deltavan, 1.5% EC, Vantage chemicals (Pvt.) Ltd. Lahore, Pakistan) were used.

Insect Collection and Rearing

Melon fruit flies were collected from infested fruits. These infested cucumber fruits were collected from the vegetable market of Bahawalpur and were taken to toxicology laboratory, department of Entomology. These infested fruits were cleaned with the 10% ethanol (McGovern et al., 2004) and were kept in plastic boxes (30×15×15 cm^3) with moist sand beneath, to collect the pupae. The maggots were fed on cucumbers and pupated in the sterilized sand placed below which were shifted into new plastic ventilated cages (45×40×40 cm^3).

After adult emergence, the files were fed on semi-synthetic diet which contained banana, eggs without yolk, honey, white sugar, yeast and multi vitamins syrup (Hamzah et al., 2021). All the ingredients were mixed well in the blender and kept in the refrigerator overnight at 4 ℃ for fermentation.

For oviposition, fresh cucumbers were placed inside the cages of adult flies on alternate days. To keep cucumber safe from fungus attack, these were washed and coated with 10% sodium hypochlorite solution (Andrews, 1996). Cucumbers were then placed on the dishes and allowed air to dry for one hour before placing inside the cages for oviposition. To get next generation of flies, cucumbers were removed from the cages after oviposition and kept in separate boxes (30×15×15 cm^3) with sterilized sand beneath. Sand beneath the egg laid cucumbers is necessary for pupation which also absorbs excessive water oozing from infested cucumbers and kept maggots safe from sink until they became pupae. After pupation, the pupae were separated by sieve and kept in the Petri dishes and were kept inside the cages for adult emergence. The culture was reared at 28 ± 2°C temperature with 65 ± 5% relative humidity and 12:12 (D/L) photoperiod in the laboratory (Rana et al., 2015).

Bioassays

Diet Incorporate method

Bioassays for melon fruit fly (adult stage) were conducted through diet exposure method. Adults of *B*. *cucurbitae* (male and female) ten to fifteen days old were obtained from the laboratory culture. The serial dilutions of selected insecticides were prepared in distilled water. There were five to six concentrations for each insecticide and each concentration had three replications. For a single replication, ten pairs of adult flies were exposed. Dilutions of 2 ml selective insecticides were incorporated in 20 ml of semi-synthetic diet in one ratio of ten. Cotton balls were dipped into an insecticide mixed diet and suspended in each jar for adult feeding. For control, insecticide free diet was provided. Post application mortality percentage was record at different time of intervals (24, 48 and 72h). The flies were counted dead if they didn't show any movement by gentle touch with camel hair brush (Mosleh, Moussa, and Mohamed 2011).

Contact method

Stock solution and serial dilutions of selected insecticides were prepared in distilled water with five to six concentration levels for each insecticide. Each concentration had three replications and three untreated replications as control. Insecticide solution was coated inside a glass jar of 4.5 cm diameter and 10cm long and allowed to evaporate by swirling the jar so that it spreads uniformly across the whole surface, providing a residual coating. After coating, the jars were allowed to air dry. Then, ten adult flies (male and female) were added on to the coated surface and thus exposed to residual film. Sugar solution was provided by soaking cotton balls for adult feeding and survival requirements. Jars were examined after 24, 48 and 72 hours for dead flies (Rana et al., 2015; Paramasivam, 2017). The flies were considered dead if they didn't show any movement by gentle touch with camel hairbrush.

Statistical Analysis

Data for adult mortality was corrected by Abbott's formula (Abbot, 1925) and then subjected to Probit analysis (Hasanuzzaman et al., 2019). Parameters like lethal concentration, fiducial limits, chi square, probability and degree of freedom were calculated using Polo Plus 1.0 LeOra Software.

RESULTS AND DISCUSSION

Diet Incorporate Method

The results of different insecticides viz., bifenthrin, deltamethrin, cypermethrin, fipronil, spinosad and emamectin-benzoate by diet incorporate method against B. cucurbitae collected from Bahawalpur has been described below. The LC_{50}'s of bifenthrin were 96.63 ppm, 68.56 ppm and 16.88 ppm and LC_{90}'s were 672.81 ppm, 742.18 ppm and 742.98 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of deltamethrin were 124.26 ppm, 67.82 ppm and 10.37 ppm and LC_{90}s were 974.38 ppm, 1931.3 ppm and 376.67 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of cypermethrin were 497.58 ppm, 150.08 ppm and 9.50 ppm and LC_{90}'s were 7609.8 ppm, 4485.8 ppm and 171.87 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of fipronil were 110.08 ppm, 49.33 ppm and 19.13 ppm and LC_{90}'s were 972.42 ppm, 661.28 ppm and 317.18 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of spinosad were 58.63 ppm, 4.95 ppm and 2.86 ppm and LC_{90}'s were 719.38 ppm, 48.99 ppm and 13.33 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of emamectin-benzoate were 14.75 ppm, 9.53 ppm and 2.98 ppm and LC_{90}'s were 76.59 ppm, 71.02 ppm and 16.12 ppm after 24, 48 and 72 hours, respectively (Table 1). According to the result, in diet incorporated method spinosad was found to be the most efficient insecticide against *B. cucurbitae* as it showed the least LC_{50} i.e. 2.86 ppm after 72 hours. It is followed by emamectin-benzoate. The LC_{50} value of emamectin-benzoate was 2.98 ppm after 72 hours against *B. cucurbitae*. And the third most efficient LC_{50} was 4.95 ppm of spinosad after 48 hours. Different field and laboratories experiments have proven that spinosad is the most effective control measure and emamectin-benzoate is also effective measure for the control of fruit fly. Field studies were done by Bhowmik et al. (2014) to evaluate the efficiency of various insecticidal treatments on bitter gourd against *B. cucurbitae*. Spinosad was the most effective treatment in decreasing fruit infestation of melon fruit fly. The highest yield of bitter gourd was recorded in spinosad. In another experiment, Gogi et al. (2007) tested nineteen insecticides to determine the ideal insecticide based on its attract-and-kill capability and periodicity.

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Table 1. Effect of insecticides against *B. cucurbitae* by diet incorporate method.

Insecticides	Hours	LC_{50} (ppm)	95% FL	LC_{90} (ppm)	95% FL	Slope ± SE	χ^2	P
Bifenthrin	24	96.63	51.81-579.62	672.81	200.64-59915	1.52 ± 0.29	4.86	0.30
	48	68.56	43.87-146.48	742.18	281.17-5587.9	1.23 ± 0.23	3.81	0.43
	72	16.88	8.96-31.89	742.98	199.81-26825	0.78 ± 0.19	1.17	0.88
Deltamethrin	24	124.26	49.42-5532.17	974.38	182.67-0.15	1.43 ± 0.46	0.82	0.93
	48	67.82	28.45-791.81	1931.3	279.57-0.80	0.88 ± 0.23	1.60	0.80
	72	10.37	6.08-23.49	376.67	95.11-14248	0.82 ± 0.19	2.40	0.66
Cypermethrin	24	497.58	161.31-39477	7609.8	922.15-0.44488	1.08 ± 0.33	0.97	0.91
	48	150.08	68.59-1187.04	4485.8	721.88-0.10516	0.86 ± 0.22	2.29	0.68
	72	9.50	5.20-14.73	171.87	79.28-868.34	1.01 ± 0.20	0.64	0.95
Fipronil	24	110.08	59.80-446.27	972.42	288.43-21496	1.35 ± 0.30	0.38	0.98
	48	49.33	30.09-120.65	661.28	219.46-7384.7	1.13 ± 0.22	1.10	0.89
	72	19.13	12.49-33.76	317.18	121.61-2420.66	1.05 ± 0.20	1.80	0.77
Spinosad	24	58.63	29.60-299.99	719.38	177.11-29142	1.17 ± 0.27	1.79	0.77
	48	4.95	2.72-8.64	48.99	21.56-364.02	1.28 ± 0.20	1.28	0.86
	72	2.86	1.28-5.00	13.33	7.03-78.43	1.91 ± 0.26	8.15	0.08
Emamectin Benzoate	24	14.75	9.51-28.86	76.59	36.20-450.30	1.79 ± 0.26	4.64	0.32
	48	9.53	6.91-13.96	71.02	38.79-204.99	1.46 ± 0.22	2.24	0.69
	72	2.98	1.73-4.37	16.12	10.52-32.36	1.75 ± 0.30	3.42	0.48

The end result revealed that spinosad was the most effective among all the tested insecticides. Revis et al. (2004) performed trails to examine attractiveness and toxicity of GF-120 fruit fly bait, against melon fruit fly to evaluate the impact of concentrations and aging.They evaluated dilutions of 20, 40, and 80 ppm spinosad and used water as control; the treatments at 80 and 40 ppm were substantially more attractive than the 20 ppm and control treatments.

Contact Method

The results of different insecticides viz., bifenthrin, deltamethrin, cypermethrin, fipronil, spinosad and emamectin-benzoate by contact method against *B. cucurbitae* collected from Bahawalpur has been described. The LC_{50}'s of bifenthrin were 8.98 ppm, 1.37 ppm and 0.63 ppm and LC_{90}'s were 140.23 ppm, 11.92 ppm and 4.99 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of deltamethrin were 16.39 ppm, 7.49 ppm and 1.06 ppm and LC_{90}'s were 101.49 ppm, 379.10 ppm and 11.45 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of cypermethrin were 3.39 ppm, 3.06 ppm and 0.62 ppm and LC_{90}'s were 10536.65 ppm, 39.35 ppm and 3.39 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of fipronil were 55.79 ppm, 41.92 ppm and 17.006 ppm and LC_{90}'s were 318.87 ppm, 625.67 ppm and 1215.3 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of spinosad were 51.89 ppm, 23.94 ppm and 10.07 ppm and LC_{90}'s were 938.78 ppm, 558.26 ppm and 177.59 ppm after 24, 48 and 72 hours, respectively. The LC_{50}'s of emamectin-benzoate were 25.80 ppm, 10.83 ppm and 5.35 ppm and LC_{90}'s were 269.46 ppm, 87.95 ppm

Table 2. Effect of insecticides against *B. cucurbitae* by contact method.

Insecticides	Hours	LC_{50} (ppm)	95% FL	LC_{90} (ppm)	95% FL	Slope ± SE	χ^2	P
Bifenthrin	24	8.98	5.40-16.01	140.23	54.89-1099.97	1.07 ± 0.22	3.38	0.49
	48	1.37	0.62-2.18	11.92	7.41-27.23	1.36 ± 0.25	2.88	0.57
	72	0.63	0.323-1.255	4.99	2.56-9.83	1.55 ± 0.15	0.86	0.83
Deltamethrin	24	16.39	10.79-35.12	101.49	43.90-608.49	1.61 ± 0.31	2.19	0.70
	48	7.49	4.10-24.98	379.10	70.50-55221	0.75 ± 0.19	0.54	0.96
	72	1.06	0.59-1.58	11.45	6.61-31.63	1.24 ± 0.22	3.60	0.46
Cypermethrin	24	30.39	6.26-147.47	10536.65	2171.59-51124.246	0.50 ± 0.35	0.26	0.96
	48	3.06	1.610-5.834	39.35	20.677-74.922	1.25 ± 0.14	0.25	0.99
	72	0.62	0.31-0.99	3.39	1.96-10.97	1.74 ± 0.25	4.98	0.28
Fipronil	24	55.79	32.39-214.23	318.87	111.63-5765.2	1.69 ± 0.41	1.18	0.87
	48	41.92	22.48-160.89	625.67	162.40-17111	1.09 ± 0.24	0.78	0.94
	72	17.006	8.76-78.22	1215.3	175.25-0.77817	0.69 ± 0.19	1.24	0.87
Spinosad	24	51.89	25.44-293.19	938.78	198.10-64384	1.01 ± 0.24	2.55	0.63

	48	23.94	13.56-75.84	558.26	138.08-18761	0.93 ± 0.21	3.15	0.53
	72	10.07	6.52-18.28	177.59	65.47-1538.12	1.02 ± 0.20	1.81	0.76
Emamectin	24	25.80	11.69-85.41	269.46	82.75-42520	1.25 ± 0.21	7.76	0.10
Benzoate	48	10.83	3.24-22.91	87.95	35.92-2562.99	1.40 ± 0.21	8.88	0.06
	72	5.35	1.84-9.11	29.64	16.36-137.08	1.72 ± 0.27	6.14	0.18

and 29.64 ppm after 24, 48 and 72 hours, respectively (Table 2). According to the results, in contact method cypermethrin was found as most efficient insecticide against the *B. cucurbitae* as it showed least LC_{50} i.e. 0.62 ppm after 72 hours. It is followed by insecticide of the same group i.e. bifenthrin. The LC_{50} value of bifenthrin was 0.63 ppm after 72 hours against *B. cucurbitae* and the third most efficient insecticide was deltamethrin which has 1.06 ppm LC_{50} values after 72 hours. So, the results indicate that synthetic pyrethroids are the most effective insecticides in residual contact method. After 72 hours of time synthetic pyrethroids insecticides showed their maximum efficacy. Among the tested pyrethroids cypermethrin was most effective followed by bifenthrin and deltamethrin and in past cypermethrin was also proved most effective against fruit fly in various experiments. According to (Rana et al., 2015) Effect of cypermethrin against melon fruit fly noted with different concentrations against their third instar larvae showed highest 87% mortality at 0.025 percent concentration. Cypermethrin was also found to be best control when the effectiveness of ten organic formulations and one synthetic insecticide against fruit fly in cucumber crop was studied. The cypermethrin treatment resulted in the lowest fruit infection (Sharma et al., 2016). In another case the insecticides cypermethrin and chlorantraniliprole were tested against two Bactrocera species and results showed cypermethrin appears to be a better insecticide to employ for controlling certain fruit fly species than chlorantraniliprole, although with precaution (Hasanuzzaman et al., 2019).

CONCLUSION

According to the LC_{50}'s values of tested insecticides of both methods it is concluded that contact method was found as the most efficient method to control the adult melon fruit fly. Furthermore, among the tested insecticides, synthetic pyrethroids were the most effective in contact method. Specifically, cypermethrin showed maximum mortality at low dose followed by bifenthrin and deltamethrin. The other insecticides, i.e. fipronil, spinosad and emamectin-benzoate showed higher LC_{50}'s values than cypermethrin. In diet incorporate method, spinosad was most efficient against melon fruit fly with high mortality at low dose as it showed least LC_{50} 2.86 ppm which was followed by emamectin benzoate.

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