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TOPICAL TOXICITY OF TEN INSECTICIDES TO THE LARVAE OF OSTRINIA FURNACALIS (GUENEE) (LEPIDOPTERA: PYRALIDAE)[1]

C. P. Rodriguez and F. F. Sanchez[2]

The median lethal doses (LD_{50}) in ug/g body weight of ten selected insecticides on the different larval stages of O. furnacalis **were determined in the laboratory. The insecticides used were: tetrachlorvinphos, chlorpyrifos, malathion, DDT, endosulfan, diazinon, methyl parathion, lindane, carbaryl and dichlorvos.**

The larvae were relatively susceptible to tetrachlorvinphos, chlorpyrifos, malathion, methyl parathion, lindane and carbaryl at all instars, but not to dichlorvos (during the first instar), endosulfan (during the second, third and fourth instars), and diazinon and DDT (during the third instar). Generally, LD_{50} values increased with age up to the third instar and then decreased starting from the fourth instar up.

The peak resistance of O. furnacalis **at the third instar and the observation that it is the last stage that could be reached by insecticides before the larvae bore into the plants clearly indicate that the third-instar larvae are the selected test insects in any screening of insecticides against the corn borer.**

Infestation of the corn borer, *Ostrinia furnacalis* (Guenee) has been a limiting factor to corn production in the Philippines. The insect also feeds on sorghum and mungbean.

Under local conditions, early reports on the importance of *O. furnacalis* were made by Jones (1913) and Navarro (1917). Buligan (1929) made a detailed study on the biology of the pest. Egg laying and larval habits were described by Guerrero (1965).

Screening of several insecticides for the control of *O. furnacalis* was done by Banaag and Viado (1954). They reported the effectiveness of DDT and dieldrin against the pest. In different investigations, it was shown that DDT and endrin provided good control against the insect. (Dizon 1958, Estores

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[2] Research Associate, National Institute of Science and Technology, NSDB, Manila, and Associate Professor and Director of Rodent Research Center, University of the Philippines at Los Baños, College, Laguna, respectively.

1958, and Deang 1959). Viado, *et al.* (1963) combined DDT with endrin and found the mixture to give adequate control. Calora and Ferino (1963) reported that aside from DDT and endrin, Thiodan is also effective. The application of insecticidal granules and spray for the corn borer was done by Sanchez and Reyes (1969). Other studies on chemical control are those of Gibe (1958), Ferino (1959), Mendoza (1964) and Rigor (1964).

This study was conducted at the Department of Entomology, U.P. College of Agriculture from November, 1970 to March, 1972 to determine the LD_{50} values and relative toxicities of the commonly used insecticides. These information may serve as a guide in selecting insecticides for the control of the insect under field condition; and may form a basis for determining the development of resistance of the corn borer in the future.

MATERIALS AND METHODS

Mass Rearing. Pupae of *O. furnacalis* were collected from field corn at the Central Experiment Station to start the laboratory culture. The emerging adults were placed in rearing cages measuring 32 x 46 x 61 cm. as modified from Guthrie, *et al.* (1965). Sides of the cages are made up of wooden frames covered with 16-mesh screen; the top is covered with 4-mesh screen. A 56 x 28 cm. sheet of waxed paper, which was replaced daily, was placed on top of the cage for oviposition. A thin layer of foam was placed above the wax paper to hold it in place. Jute sacks were wrapped around the cage to induce mating because the adults usually mate under subdued light. The cages were sprinkled with water at least twice a day to maintain high humidity. A cotton wad saturated in five percent honey solution contained in half petri dish was placed inside the cage as adult food. One rearing cage accommodated 150-200 adults. The mean temperature inside the oviposition room was 79.6 ± 0.1°C with a mean relative humidity of 82.2 ± 0.3 percent.

The waxed paper containing egg masses was cut and placed in petri dishes. The eggs at black head stage were transferred and allowed to hatch in screen-covered glass rearing jars with bean pods. One rearing jar contained five egg masses with approximately 30 eggs per mass. Before changing the food, fresh bean pods were placed on top of old ones for the larvae to transfer. After most of the larvae had transferred, the fresh pods were placed in other rearing jars. A fine camel's hair brush was used in picking up the larvae which failed to transfer. Food was changed at least three times a week. When the larvae were about to pupate, strips of corrugated paper were placed vertically inside the jars for pupation. The pupae were then transferred to the previously described mass rearing cages.

The cages were cleaned thoroughly after each batch of insect culture to minimize disease occurrence. The oviposition and rearing room was also kept clean.

Toxicological Study. Toxicological evaluation of ten insecticides was done using all the larval stages of *O. furnacalis.* The percentage purity and chemical composition of the insecticides used are as follows: DDT [100% 1,1,1-trichloro 2,2-bis (p-chlorophenyl) ethane], malathion [96% S-(1,2,-dicarbethoxyethyl) 0,0-dimethyl) phosphorodithioate], lindane [100% 1,2,3,4,5,6- hexachlorohexane, 99% or more gamma isomer], tetrachlorvinphos, [99.5% 2-chloro-1-(2,4,5-trichlorophenyl) vinyl dimethyl-phosphate], dichlorvos [97%-0-dimethyl 2,2-dichlorovinyl phosphate], Diazinon [97% 0-0-diethyl 0-(2-isopropyl-4-methyl-6-pyrimidinyl) phosphorothioate], carbaryl [97% 1-naphthyl N-methylcarbamate], endosulfan [97% 6,7,8,9, 10-hexachloro 1,5,5a,6,9,9a hexahydro-6,9-methano-2,4,3-benzodioxathienpin 3-oxide], chlorpyrifos [99% 0-0-diethyl 0-(3,5,6,-trichloro-2 pyridyl phosphorodithioate], and methyl parathion [80% 0,0-diethyl 0-p-nitrophenyl phosphorothioate.].

Stock solutions of each insecticide were prepared at 50 mg/ml using acetone as solvent. Serial dilutions of these were used in all tests.

Larvae of relatively uniform age and size were used in the test. The larvae were weighed in groups of ten before each treatment using a semi-micro Mettler Balance.

An automatic microapplicator with a calibrated tuberculin syringe was used in applying insecticides on the thoracic dorsum of each test insect. Technical acetone was used in the control. The treated larvae were placed in batches of ten in petri dishes with bean pods. A fine camel's hair brush was used for transferring the larvae to the petri dishes. The tests were done under a mean temperature of 78.3 ± 0.1°C. There were five replications per treatment. Moribund individuals were considered dead. The data were statistically analyzed using probit analysis (Finney, 1952).

RESULTS

Mass Rearing. Enough test insects were produced using the mass rearing techniques previously described. Sufficient numbers of fertile eggs were produced daily.

The first instar larvae were too soft and minute, so it was more advantageous to place black head stage eggs on the mass rearing jars than to handle the newly-hatched larvae. It was found that 100-150 larvae per jar was ideal.

Toxicological Study. The median lethal doses (LD_{50} values) in µg/g body weight of ten insecticides on all larval stages of *O. furnacalis* are presented in table. I The relative toxicity of the insecticides used in decreasing order was as follows:

First instar: tetrachlorvinphos = chlorpyrifos> malathion = methyl parathion = lindane = carbaryl =DDT> endosulfan> diazinon≥ dichlorvos

TABLE 1. **The median lethal doses (LD_{50}), confidence limits and regression lines of ten insecticides topically applied on the different larval instars of O. furnacalis.**

Insecticide	LD_{50} (μg/gm larva)	Confidence Limit (p = .05) lower	upper	Regression line
	FIRST INSTAR			
Tetrachlorvinphos	0.898	.645	1.129	3.738+1.691 x
Chlorpyrifos	1.079	.807	1.413	2.137+1.655 x
Malathion	4.292	3.437	5.292	2.719+1.735 x
M-Parathion	5.560	4.00	6.00	2.580+1.729 x
Lindane	7.140	6.00	8.00	2.156+1.826 x
Carbaryl	8.500	6.78	10.66	3.936+1.690 x
DDT	8.984	7.113	11.89	3.738+1.691 x
Endosulfan	48.360	36.00	64.00	2.073+1.226 x
Diazinon	82.432	66.216	104.054	0.558+1.593 x
Dichlorvos	208.333	179.166	274.91	-0.115+1.682 x
	SECOND INSTAR			
M-Parathion	1.529	1.345	2.018	1.672+1.810 x
Tetrachlorvinphos	2.184	1.800	2.800	1.486+1.718
Malathion	6.667	5.333	8.444	0.998+1.609 x
Diazinon	15.494	12.40	18.667	3.176+1.887 x
Dichlorvos	26.782	21.00	33.00	-0.876+1.714 x
Lindane	26.940	21.938	33.324	-0.127+1.716 x
DDT	50.840	41.525	52.355	-0.408+1.55 x
Carbaryl	54.667	42.175	69.438	2.951+1.472 x
Chlorpyrifos	99.080	80.00	120.00	3.733+1.823 x
Endosulfan	306.500	250.00	378.00	-7.167+1.794 x
	THIRD INSTAR			
Malathion	7.701	6.323	9.379	3.329+1.988 x
M-parathion	8.568	7.000	10.00	-0.973+1.847 x
Carbaryl	10.000	7.826	12.265	1.382+1.766 x
Chlorpyrifos	26.492	21.516	32.701	-1.534+1.743 x
Dichlorvos	62.723	50.057	78.688	-1.656+1.613 x
Tetrachlorvinphos	81.774	63.384	94.710	3.299+1.710 x
Lindane	92.790	63.876	113.821	-2.173+1.768 x
Diazinon	173.900	140.859	219.13	2.631+1.809 x
DDT	323.33	227.572	402.796	4.020+1.730 x
Endosulfan	353.157	281.753	439.297	3.932+1.766 x

TABLE 1. **Continued . . .**

Insecticide	LD_{50} (μg/gm larve)	Confidence Limit (p = .05) lower	upper	Regression line
		FOURTH INSTAR		
Malathion	2.224	1.740	2.780	3.292+1.632 x
Dichlorvos	5.962	4.882	7.306	-2.947+1.776 x
M-Parathion	6.440	5.425	7.575	-1.836+2.01 x
Chlorpyrifos	14.374	11.810	17.554	-2.117+1.862 x
Carbaryl	31.117	25.933	37.466	2.652+2.047 x
Lindane	36.138	28.457	44.565	2.833+1.775 x
Tetrachlorvinphos	43.204	35.396	52.735	-1.703+1.580 x
Diazinon	64.770	51.673	79.959	3.641+1.774 x
Endosulfan	101.364	84.459	121.567	-4.494+2.033
DDT	211.988	172.974	260.147	-3.621+1.726 x
		FIFTH INSTAR		
M-Parathion	1.874	1.424	2.384	3.578+1.466 x
Chlorpyrifos	2.880	2.204	3.526	2.674+1.852 x
Malathion	4.727	3.870	5.764	-2.67 +1.772 x
Tetrachlorvinphos	6.338	5.174	7.772	-2.508+1.695 x
Lindane	9.042	7.246	11.240	-2.191+1.538 x
Endosulfan	9.737	7.654	12.181	3.755+1.592 x
Dichlorvos	12.310	10.060	15.065	-3.340+1.69 x
Diazinon	17.496	13.869	21.846	3.311+1.633 x
DDT	28.00	22.93	34.213	-3.940+1.737 x
Carbaryl	36.747	29.47	44.302	2.749+1.751 x
		SIXTH INSTAR		
M-Parathion	3.360	3.164	4.769	-4.36 +1.802 x
Malathion	8.120	7.458	11.246	4.276+1.601 x
Chlorpyrifos	12.164	10.017	15.146	2.593+1.926 x
DDT	14.107	11.448	16.941	2.472+1.726 x
Dichlorvos	17.500	14.672	22.424	2.174+1.816 x
Tetrachlorvinphos	19.440	16.519	23.941	-4.936+1.863 x
Endosulfan	29.370	23.371	36.645	3.302+1.780 x
Diazinon	32.111	25.342	40.331	3.668+1.650 x
Carbaryl	32.49	26.50	39.871	-5.067+1.811 x
Lindane	34.115	27.93	41.669	-5.681+1.830 x

Second instar: Methyl parathion = tetrachlorvinphos> malathion> diazinon> dichlorvos = lindane> DDT = carbaryl> chlorpyrifos> endosulfan
Third instar: Malathion = methyl parathion = carbaryl> chlorpyrifos> dichlorvos = tetrachlorvinphos = lindane> diazinon> DDT = endosulfan
Fourth instar: Malathion> dichlorvos = methyl parathion> chlorpyrifos> carbaryl = lindane = tetrachlorvinphos = diazinon> endosultan> DDT
Fifth instar: Methyl parathion = chlorpyrifos> malathion> tetrachlorvinphos = lindane = endosulfan = dichlorvos = diazinon > DDT = carbaryl
Sixth instar: Methyl parathion> malathion = chlorpyrifos = DDT = dichlorvos = tetrachlorvinphos = endosulfan = diazinon> carbaryl = lindane

Table 2 shows the relative responses of the different larval instars to each insecticide. The relative toxicity of the chemical to the different instars in decreasing order was as follows:
Tetrachlorvinphos: First> Second> Fifth> Sixth> Fourth> Third
Chlorpyrifos: First> Fifth> Sixth = Fourth> Third> Second
Malathion: Fourth> First = Fifth = Second = Third = Sixth
DDT: First = Sixth> Fifth> Second> Fourth = Third
Endosulfan: Fifth> Sixth = First> Fourth> Second = Third
Diazinon: Second = Fifth> Sixth = Fourth = First> Third
Lindane: First = Fifth> Second = Sixth = Fourth> Third
Carbaryl: First = Third > Fourth = Sixth = Fifth = Second
Lindane: First = Fifth> Second = Sixth = Fourth> Third
Carbaryl: First = Third> Fourth = Sixth = Fifth = Second
Dichlorvos: Fourth> Fifth = Sixth = Second> Third> First

DISCUSSION

The larvae were found relatively susceptible to tetrachlorvinphos, chlorpyrifos, malathion, methyl parathion, lindane and carbaryl at all instars, but not to dichlorvos (during the first instar), endosulfan (during the second, third and fourth instars), diazinon and DDT (during the third instar). Generally, the LD_{50} values increased with age up to the third instar and increased susceptibility was observed starting from the fourth instar. Since the larvae, in nature, start to bore during the third instar, it may be possible that the insect at this stage has a high level of physiologic activity (e.g. increased production of certain enzymes) resulting in a greater ability to degrade out certain insecticides.

The different larval stages exhibited no definite pattern of susceptibility to the insecticides used. Similar observations were reported by Cadapan (1971), Deang (1971) and Ratisoontorn (1971) on *Pseudaletia separata* (Walker), *Helicoverpa armigera* (Hubner) and *Chilo suppressalis* (Walker), respectively. They attributed the change in the level of susceptibility to the variations on the physiological activity of the larvae at different stages of development. It is therefore important to specify the larval instars used in an insecticidal testing. Further research on the physiologic explanation of the result is imperative.

The finding that the third instar larvae of *O. furnacalis* was generally the most resistant stage has an important practical implication in the screening of insecticides against the species. This, and the observation that the third instar larvae is the last stage that could be reached by insecticides outside the plant are clear indications that the third instar larvae should be the test insects in screening insecticides against *O. furnacalis*.

TABLE 2. **The susceptibility of the different larval instars of** O. furnacalis **to ten topically applied insecticides.**

Larval Instar	LD_{50} (µg/gm larva)	Confidence Limit (p = .05) Lower	Upper	Regression Lie
		TETRACHLORVINPHOS		
I	0.898	.645	1.129	3.738 + 1.691 x
II	2.184	1.800	2.800	1.486 + 1.718 x
III	81.774	63.384	94.710	3.299 + 1.710 x
IV	43.204	35.396	52.735	-1.703 + 1.580 x
V	6.338	5.174	7.772	-2.508 + 1.695 x
VI	19.440	16.519	23.941	-4.936 + 1.863 x
		CHLORPYRIFOS		
I	1.079	.807	1.413	2.137 + 1.655 x
II	99.080	80.00	120.00	3.733 + 1.823 x
III	26.492	21.516	32.701	-1.534 + 1.743 x
IV	14.374	11.810	17.554	-2.117 + 1.862 x
V	2.880	2.204	3.526	2.674+ 1.852 x
VI	12.164	10.017	15.146	2.593 + 1.926 x
		MALATHION		
I	4.292	34.437	5.292	2.719 + 1.735 x
II	6.667	5.383	8.444	0.998 + 1.609 x
III	7.701	6.323	9.379	3.329 + 1.988 x
IV	2.224	1.740	2.780	3.292 + 1.632 x
V	4.727	3.870	5.764	-2.67 + 1.722 x
VI	8.120	7.458	11.246	4.276 + 1.601 x
		D D T		
I	8.984	7.113	11.89	3.738 + 1.691 x
II	50.840	41.525	52.355	-0.408 + 1.55 x
III	323.33	227.572	402.796	4.020 + 1.730 x
IV	211.988	172.974	260.147	-3.621 + 1.726 x
V	28.00	22.93	34.213	-3.940 + 1.737 x
VI	14.107	11.448	16.941	2.472 + 1.726 x

TABLE 2. **Continued...**

Larval Instar	LD_{50} (μg/gm larva)	Confidence Limit (p = .05) Lower	Upper	Regression Line
		ENDOSULFAN		
I	48.360	36.00	64.00	2.073 + 1.226 x
II	306.500	250.00	378.00	-7.167 + 1.794 x
III	353.157	281.753	439.297	3.932 + 1.766 x
IV	101.364	84.459	121.567	-4.494 + 2.033 x
V	9.737	7.654	12.181	3.755 + 1.592 x
VI	29.370	23.371	36.645	3.302 + 1.780 x
		DIAZINON		
I	2.43	66.216	104.054	0.558 + 1.593 x
II	15.494	12.40	18.664	3.176 + 1.887 x
III	173.900	140.859	219.13	2.631 + 1.809 x
IV	64.770	51.673	79.959	3.641 + 1.774 x
V	17.496	13.869	21.846	3.311 + 1.633 x
VI	32.111	25.342	40.331	3.668 + 1.650 x
		METHYL PARATHION		
I	5.560	4.00	6.00	2.580 + 1.729 x
II	1.529	1.345	2.018	1.672 + 1.810 x
III	8.568	7.00	10.00	-0.973 + 1.847 x
IV	6.440	5.425	7.575	-1.836 + 2.01 x
V	1.874	1.424	2.384	3.578 + 1.466 x
VI	3.630	3.164	4.769	-4.136 + 1.802 x
		LINDANE		
I	7.140	6.00	8.00	2.156 + 1.826 x
II	26.940	21.938	33.324	-0.127 + 1.716 x
III	92.790	63.876	133.821	-2.173 + 1.768 x
IV	36.138	28.457	44.565	2.883 + 1.775 x
V	9.042	7.246	11.240	-2.191 + 1.538 x
VI	34.115	27.930	41.669	-5.681 + 1.830 x
		CARBARYL		
I	8.5	6.78	10.66	3.936 + 1.69 x
II	54.667	42.175	69.438	2.951 + 1.472 x
III	10.00	7.826	12.265	1.382 + 1.766 x
IV	31.117	25.933	37.466	2.652 + 2.047 x
V	36.747	29.47	44.302	2.749 + 1.751 x
VI	32.49	26.50	39.871	-5.067 + 1.811 x
		DICHLORVOS		
I	208.333	179.166	274.91	-0.115 + 1.682 x
II	26.782	21.00	33.00	-0.876 + 1.714 x
III	62.723	50.057	78.688	-1.656 + 1.613 x
IV	5.962	4.882	7.306	-2.947 + 1.776 x
V	12.310	10.060	15.065	-3.340 + 1.69 x
VI	17.500	14.672	22.424	2.174 + 1.816 x

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