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THE SUSCEPTIBILITY OF TWO HOUSEFLY (MUSCA DOMESTICA L.) STRAINS TO SOME CHLORINATED HYDROCARBON INSECTICIDES[1]

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The sucsceptibility of two- to three-day old adult females of two Philippine strains of the housefly, Musca domestica L.) to six chlorinated hydrocarbon insecticides in acetone solution was studied by topical application. The relative order of susceptibility of the College strain to the insecticides used was as follows: lindane > aldrin = dieldrin > DDT = heptachlor = chlordane. The toxicity to the Calamba strain was as follows: lindane > aldrin = dieldrin > DDT = heptachlor and chlordane. The College strain was found to be more resistant than the Calamba strain based on the LD_{50} values obtained. Both strains were found highly resistant to heptachlor, dieldrin, chlordane and aldrin, and moderately resistant to lindane and DDT.

The extent of researches and investigations in the fields and laboratories devoted to insecticide resistance in houseflies and other insect species is easily reflected by the availability of literature on the subject matter. The first report in 1946 that houseflies in Northern Sweden did not respond as expected to DDT seriously upset the advantages offered by chemical control. The first record on the ability of insects to develop tolerance or resistance to insecticidal action was made by Melander in 1914 with San Jose scale (*Aspidiotus perniciosus*) and lime sulfur sprays. This was followed by Quayle in 1916 with California red scale (*Aonidiella aurantii*) and hydrogen cyanide fumigation (Metcalf, 1955a). However, it was only after the discovery of widespread development of DDT resistance by the housefly that conclusive demonstration of the relevance of this phenomenon in insect control was recognized. Hence, housefly resistance has been much more thoroughly investigated than that of any other insect (Metcalf, 1955b).

The varied aspects of this complex phenomenon have been and are being investigated continuously by researchers in many parts of the world. Metcalf (1955b) compiled studies dealing with the physiological basis of resistance and Crow (1957) on the genetical aspect. Hoskins and Gordon

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(1956), Kearns (1956) and Brown (1958b) prepared review papers on the understanding of the problems posed by the phenomenon of resistance in insect control. Brown (1958a) prepared a monograph on arthropod resistance which includes the discovery of resistance in houseflies in various parts of the world involving chlorinated hydrocarbon insecticides and other groups of insecticidal compounds.

In the Philippines however, studies on insect resistance have not been conducted. A far from conclusive report on housefly resistance in the Philippines was cited by Brown (1958a) referring to a personal communication with a certain F. J. Dy about the "presence of DDT-resistance of houseflies in Manila and Quezon City, although DDT was still being broadcast from fog applicators in 1956."

This study was undertaken in the laboratory to test the susceptibility of two Philippine strains of the housefly, *Musca domestica* L. to six locally-available insecticides of the chlorinated hydrocarbon group. It was conducted from December, 1967 to March, 1968 in the Department of Entomology, University of the Philippines.

MATERIALS AND METHODS

Rearing Test Insects. Adult houseflies used as starter cultures were collected from pigpens in the Department of Animal Husbandry, University of the Philippines, and from the slaughterhouse and meat stalls in the public market of Calamba, Laguna. They were designated "College" and "Calamba" strains, respectively. Each strain was reared separately in 1 x 1 x 1 ft^3 rearing cages provided with food consisting of moist non-fat dry milk-refined sugar mixture. Cultures were maintained subsequently on a standardized moist poultry feed[3] as larval medium.

Gravid female flies were allowed to oviposit on larval medium in Petri dishes. Eggs were transferred to culture jars containing larval medium adequate to support larval development. Culture jars were made from several half-gallon capacity reagent bottles with the top cut off. Cheese cloth and fine wire-screen were used to cover the culture jars. Maggots about to pupate were isolated from the medium with the aid of a wire-mesh separator. Pupae were placed in rearing cages where refined sugar and tap water were made always available to emerging flies.

Preparation of Test Insecticides. The insecticides used and their chemical composition were as follows: aldrin (1, 2, 3, 4, 10, 10-hexachloro-1, 4, 4a, 5, 8, 8a-hexahydro-1, 4-endo, exo-5, 8-dimethanonaphthalene), technical, 95%; chlordane (1, 2, 4, 5, 6, 7, 8, 8-octachloro-2, 3, 3a, 4,

[3] "B-Meg" brand laying mash (San Miguel Poultry and Livestock Feed Plant) was used. Composition — crude protein, 18% Min.; nitrogen free extract, 47% Min.; crude fat, 8% Max.; crude fiber, 8% Max.; and moisture, 13% Max.

7, 7a-hexahydro-4, 7-methanoindene), refined (clarified) grade; p-p' DDT (1, 1, 1-trichloro-2, 2-bis [p-chlorophenyl], p-p' isomer), 100%; dieldrin (1, 2, 3, 4, 10, 10-hexachloro-6, 7-epoxy-1, 4, 4a, 5, 6, 7, 8, 8a-octahydro-1, 4-endo, exo-5, 8-dimethanonaphthalene), technical, 85%; heptachlor (1, 4, 5, 6, 7, 8, 8-heptachloro-3a, 4, 7, 7a-tetra-hydro-4, 7-endomethanoindene), technical, 95%; and lindane (1, 2, 3, 4, 5, 6-hexachlorohexane, gamma isomer), 100%. Stock solutions of 50 mg/ml concentration were prepared for each insecticide using redistilled acetone as solvent. Dilutions of the stock solutions were used to deliver the various dosages in decreasing order of concentration for each insecticide.

Preparation and Handling of Test Insects. The susceptibility of the two strains to six locally-available chlorinated hydrocarbon insecticides was determined, following the methods of topical application suggested by Metcalf (1958). Two- to three-day old female flies and as closely uniform in size were used in all tests. They were initially anesthetized in an improvized anesthetizing chamber with CO_2 at a pressure of 10 psi for 10 minutes and subsequently kept paralyzed for handling during the tests by passing CO_2 intermittently through a 5-inch Buchner funnel with a perforated bottom. This is a slight modification of the technique used by Webb (1959) in which the CO_2 was allowed to pass from dry ice through a perforated holding chamber. The test insects were held by the wings with a fine-pointed forceps and 0.001 ml-drops of the particular dosage of insecticide were applied on the notum of each test insect by means of a pre-calibrated micrometer-actuated tuberculin syringe. Acetone was alowed to evaporate before placing each treated fly in well ventilated, clean holding cages under a room temperature of 23.44 $\pm$ 0.30 C. Sugar-water in soaked cotton wads was made available in all holding cages to the flies.

Gathering and Analysis of Data. Dosage was calculated in terms of micrograms of test insecticide for each test fly. Ten female flies of uniform size and age were treated at each dosage level. All tests were replicated four times. Controls of ten female flies treated with pure redistilled acetone were assigned to every replicate. All tests were done within the time range of from 4:00 P. M. to 11:00 P. M.

Mortality counts were taken 24 and 48 hours after treatment. All individuals found dying were considered dead.

The observed mortalities were not corrected for natural mortality by Abbott's (1925) formula since no mortalities were observed in the controls. The LD_{50} for each insecticide was determined for each strain using probit analysis (Finney, 1952). Dosage-mortality curves, in which dosage in terms of micrograms of toxicant administered per insect is plotted against percent mortality, were constructed for each insecticide.

RESULTS AND DISCUSSION

Figures 1 and 2 present the relationship between insecticide dosage and percent mortality of the College and the Calamba strains respectively. Both strains were found to be homogenous in their response to all the insecticides used in this study as determined by the chi-square test at the 5% probability level.

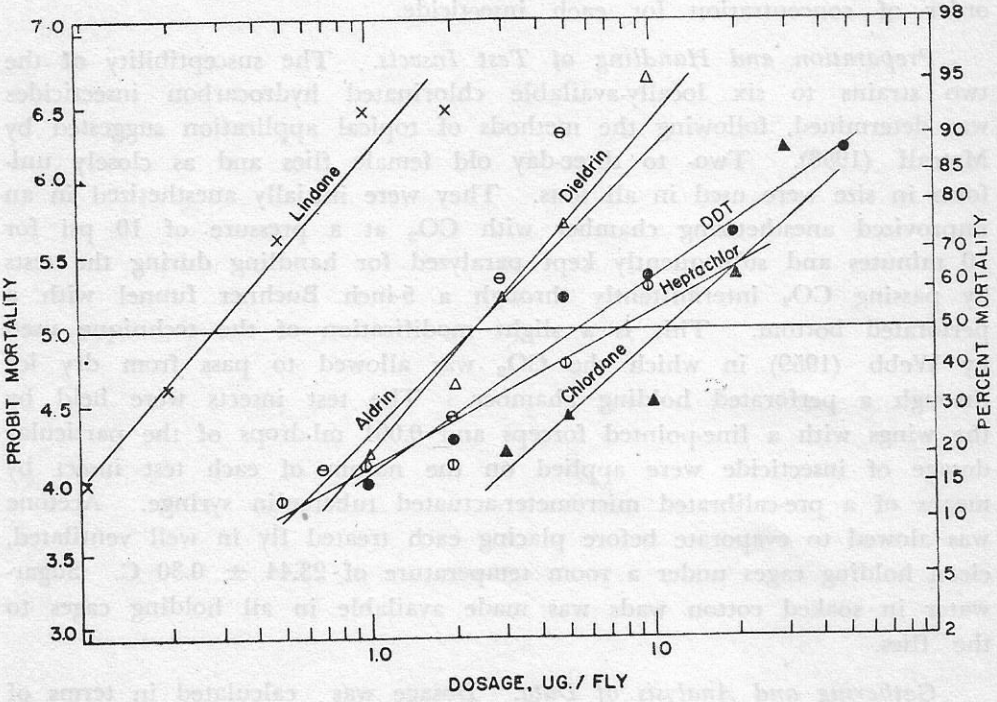

Fig. 1. The dosage-mortality regression lines of several insecticides to adult females of the College strain of *Musca domestica* L.

Comparison of the susceptibility of the two strains (Tables 1 and 2) shows that the College strain was more resistant to all the insecticides used than the Calamba strain. The comparative susceptibility of the College strain to that of the latter in terms of higher dosage required to cause 50% mortality was as follows: chlordane, 3.01X; DDT, 2.48X; heptachlor, 2.21X; dieldrin, 2.00X; aldrin, 1.94X; and lindane, 1.93X. These results collaborate the observations by Crow (1957) which states that higher resistance is to be expected in population with a history of exposure to poisons. The College strain has a history of non-periodic exposure to insecticidal chemicals while the Calamba strain has seldom been subjected to chemical control.

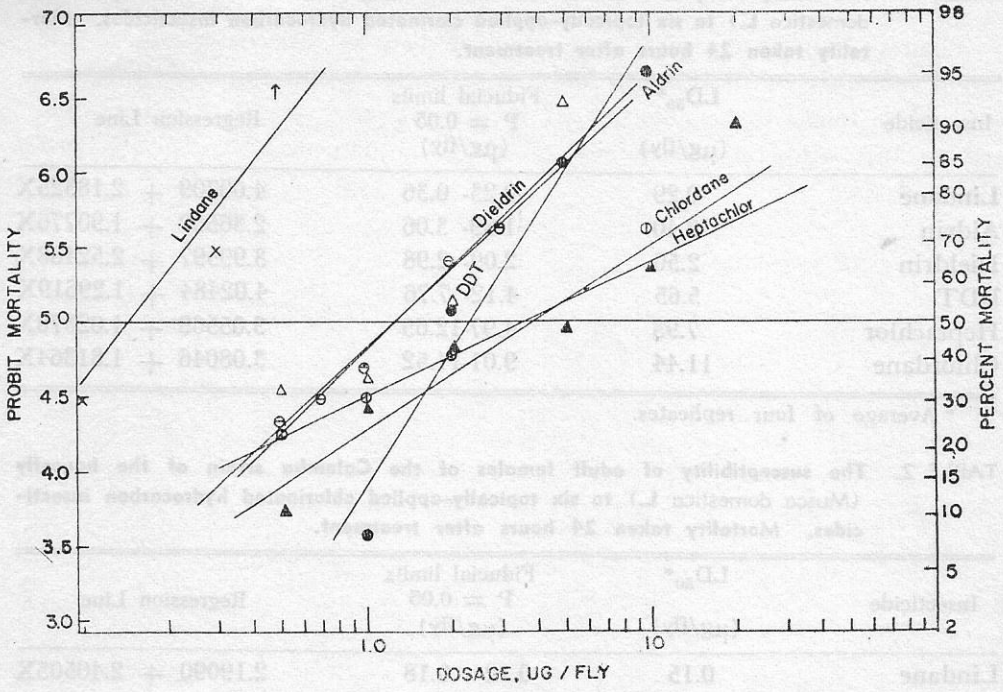

Fig. 2. The dosage-mortality regression lines of several insecticides to adult females of the Calamba strain of *Musca domestica* L.

The relative toxicity of the insecticides used to the adult females of the College strain in order of decreasing toxicity (Table 1) was a follows: lindane > aldrin = dieldrin > DDT = heptachlor = chlordane. The relative order of toxicity to the Calamba strain (Table 2) was as follows: lindane > aldrin = dieldrin > DDT = heptachlor and chlordane.

There is no prior study of this nature under local conditions which can provide a basis of comparison for the results of the present study. However, in a similar situation Webb (1959) in determining housefly resistance in nine areas in Germany and France used for comparison the LD_{50} values of 0.33 μg/fly for DDT, 0.03 μg/fly for lindane and 0.02 μg/fly for dieldrin as the "standard values for normally-susceptible flies" and the criteria of a five-to ten-fold increase in the amount of toxicant needed for equivalent mortality as an indication of resistance at least for houseflies as proposed by Decker and Bruce (1952). On this basis, Webb (1959) went on to conclude all nine strains studied to be significantly resistant to DDT, five strains lindane-resistant to a moderate degree and five strains resistant to dieldrin.

TABLE 1. **The susceptibility of adult females of the College strain of the housefly** (Musca domestica **L.**) **to six topically-applied clorinated hydrocarbon insecticides. Mortality taken 24 hours after treatment.**

Insecticide	LD_{50}* (μg/fly)	Fiducial limits P = 0.05 (μg/fly)	Regression Line
Lindane	0.29	0.23- 0.36	4.00709 + 2.18325X
Aldrin	2.40	1.89- 3.06	2.36560 + 1.90776X
Dieldrin	2.50	2.09- 2.98	3.99597 + 2.52488X
DDT	5.65	4.12- 7.76	4.02484 + 1.29619X
Heptachlor	7.93	4.97-12.65	3.05569 + 1.02378X
Chlordane	11.44	9.01-14.52	3.08046 + 1.81364X

* Average of four replicates.

TABLE 2. **The susceptibility of adult females of the Calamba strain of the housefly** (Musca domestica **L.**) **to six topically-applied chlorinated hydrocarbon insecticides. Mortality taken 24 hours after treatment.**

Insecticide	LD_{50}* (μg/fly)	Fiducial limits P = 0.05 (μg/fly)	Regression Line
Lindane	0.15	0.12 - 0.18	2.19090 + 2.40505X
Aldrin	1.24	0.97 - 1.59	3.00185 + 1.82678X
Dieldrin	1.25	0.95 - 1.64	3.07308 + 1.75613X
DDT	2.28	1.89 - 2.76	3.93078 + 2.98414X
Heptachlor	3.59	2.08 - 6.19	3.64829 + 0.86930X
Chlordane	3.80	2.68 - 5.38	3.06095 + 1.22758X

* Average of four replicates.

The LD_{50} values obtained in the present study were compared with those for susceptible strains reported from other works in other countries (Metcalf, 1955a) which indicated LD_{50} 0.44 μg/female fly for DDT, 0.16 for chlordane, 0.04 for aldrin, 0.03 for heptachlor, 0.02 for lindane and 0.02 for dieldrin. Determination of the degree of resistance in the two strains studied to the insecticides used was based on the criterion of a five-fold increase in resistance for adult houseflies required for significance as proposed by Decker and Bruce (1952). Comparison of the LD_{50} values obtained in this study with those of the designated susceptible strain (Table 3) showed that both strains studied were highly resistant to heptachlor dieldrin, chlordane, and aldrin, and moderately resistant to lindane and DDT. The ratios obtained for the College strain were: heptachlor, 264.3X; dieldrin, 125.0X; chlordane, 71.5X; aldrin, 60.0X; lindane, 14.5X and DDT, 12.8X. The ratios obtained for the Calamba strain were: heptachlor, 119.7X; dieldrin, 62.5X; aldrin, 31.0X; chlordane, 23.8X; lindane, 7.5X; and DDT, 5.2X.

TABLE 3. **Toxicity of several insecticides to two Philippine strains and a susceptible strain of the housefly** (Musca domestica **L.**).

Insecticide	Susceptible strain LD_{50} (μg/fly)	* College Strain		Calamba Strain	
		LD_{50} (μg/fly)	Ratio to normal strain	LD_{50}	Ratio to normal strain
Lindane	0.02	0.29	14.5	0.15	7.5
DDT	0.44	5.65	12.8	2.28	5.2
Aldrin	0.04	2.40	60.0	1.24	31.0
Chlordane	0.16	11.44	71.5	3.80	23.8
Dieldrin	0.02	2.50	125.0	1.25	62.5
Heptachlor	0.03	7.93	264.3	3.59	119.7

* All LD_{50} values refer to those for a susceptible Rome strain, except that for beptachlor obtained from a susceptible standard laboratory strain (Metcalf, 1955a).

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