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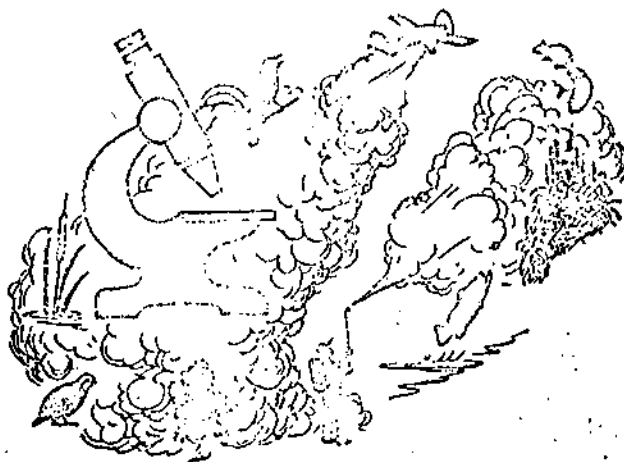
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ALVIN L. YOUNG

PESTICIDE-WILDLIFE STUDIES

A Review of Fish and Wildlife Service
Investigations During 1961 and 1962



UNITED STATES DEPARTMENT OF THE INTERIOR

Stewart L. Udall, Secretary

FISH AND WILDLIFE SERVICE

Circular 167

Abbreviations routinely used by research workers in toxicology may not always be clear to non-technical people. These include:

ppm, parts per million, is the number of parts of toxicant per million parts of the substance in question; these may include residues in whole animals, certain tissues of animals or plants, or amount of toxicant in natural or artificial foods. Residues in this publication, unless otherwise noted, are expressed in terms of ppm dry weight. For animal tissues averaging 90 percent water, 1 ppm wet or live weight would average about 10 ppm dry weight. The amount of moisture in a specimen, and therefore the wet weight, varies after death. Therefore, wet weights are most meaningful when taken of live or freshly-killed animals.

ppb, parts per billion, is the number of micrograms per kilogram of the substance in question.

mg/kg, milligrams per kilogram, is the same ratio as ppm but is used to designate the amount of toxicant required per kilogram of body weight of test organism to produce a designated effect, usually the amount necessary to kill 50 percent of the test animals.

µg/g, micrograms per gram, is much the same as mg/kg except that amounts involved, 1/1,000,000 of a gram per gram, are 1/1,000 those of mg/kg, 1/1,000 of a gram per 1,000 grams; but the ratio (ppm) is the same.

µg/l, micrograms per liter, is another expression much the same as the ratio of ppb, as the weight of a liter of water is approximately 1,000,000,000 micrograms, but the weight in micrograms of toxicant per volume of material is given.

LD₅₀, lethal dose 50, is the amount of toxicant lethal to 50 percent of the test animals to which it is administered under the conditions of the experiments. Usually the dosage necessary to kill 50 percent of test animals will vary with time, greater total amounts being needed to kill over longer periods of time. When only one oral administration is made to produce a LD₅₀, it is usually referred to as the single oral LD₅₀.

LC₅₀, lethal concentration 50, is the concentration of toxicant in the environment which kills 50 percent of the test organisms exposed to it. The expression is used by biologists working with aquatic organisms in water contaminated with the toxicant.

ED₅₀, effective dose 50, is the amount of toxicant, usually measured in mg/kg, that produces a designated effect to 50 percent of the population of test organisms receiving the dose. The term is used when the effect of the toxicant is measured in terms other than mortality or lethal effect.

EC₅₀, effective concentration 50, is the concentration of toxicant in the environment which produces a designated effect to 50 percent of the population of test organisms exposed to it.

TLm, median tolerated limit, is the median amount or concentration which produces mortality to 50 percent of the tested population in a given period of time. Commonly used periods in fisheries experiments are 24 hours (24-hour TLm), 48 hours (48-hour TLm), or 96 hours (96-hour TLm).

Table 6A. Concentration of pesticides in sea water causing 50 percent mortality, 24- and 48-hour TLm, to juvenile white mullet (M) and longnose killifish (K)

Pesticide	Kind of fish	24-hour TLm ppm	48-hour TLm ppm	Mean water temperature °C.
<u>Chlorinated hydrocarbons</u>				
Aldrin	M	0.0031	0.0028	28
BHC	M	0.8	0.8	28
Chlordane	M	0.043	0.0055	22
DDT	M	0.0008	0.0004	26
DDT	K	0.0055	0.0055	24
Dieldrin	M	0.0078	0.0071	28
Endrin	M	0.0026	0.0026	29
Endrin	K	0.0003	0.0003	25
Heptachlor	M	0.0048	0.003	26
Kepone	M	0.5	0.055	31
Kepone	K	0.3	0.084	31
Lindane	M	0.03	0.03	16
Lindane	K	0.3	0.24	29
Methoxychlor	M	0.055	0.055	24
Mirex	M	No mortality at 2.0	10% mortality at 2.0	25
Thiodan	M	0.005	0.0006	29
Toxaphene	M	0.0055	0.0055	19
<u>Fungicides</u>				
Ferbam	K*	1.0	0.8	28
Phaltan	M*	1.56	1.56	28
Phaltan	K*	2.5	2.5	29
<u>Herbicides</u>				
2,4-D acid	M*	No effect at 50.0	No effect at 50.0	20
2,4-D propylene glycol butyl ether ester	K*	5.0	4.5	20
2,4-D butoxy-ethanol ester	K*	5.0	5.0	20
Diuron	M*	10.8	6.3	29

(Continued)

*Tests performed in standing water aquaria.

Table 6A. Concentration of pesticides in sea water causing 50 percent mortality, 24- and 48-hour TLM, to juvenile white mullet (M) and longnose killifish (K)
(continued)

Pesticide	Kind of Fish	24-hour TLM ppm	48-hour TLM ppm	Mean water temperature °C.
<u>Herbicides</u>				
Eptam	M*	10% mortality at 20.0	10% mortality at 20.0	19
Eptam	K*	Irritated at 20.0	Irritated at 20.0	28
Esteron 99 (formulation)	M*	1.5	1.5	20
Esteron 99 (formulation)	K*	3.5	3.0	19
MCP amine weed killer (formulation)	K*	No effect at 75.0	No effect at 75.0	28
Monuron	M*	20.0	16.3	28
Radapon (formulation)	M*	No effect at 50.0	No effect at 50.0	27
Radapon (formulation)	K*	No effect at 50.0	No effect at 50.0	29
2,4,5-T acid	K*	No effect at 50.0	No effect at 50.0	19
Tillam (formulation)	M*	6.25	6.25	21
Tillam (formulation)	K*	7.78	7.78	29
<u>Carbamates</u>				
Bayer 37344	K	0.55	0.55	16
Sevin	M*	4.25	2.5	24
Sevin	K*	1.75	1.75	28
<u>Organophosphorus insecticides</u>				
Bayer 29493 (Baytex)	M	1.73	1.59	29
Bayer 25141	K	0.085	0.055	17
Diazinon	M	0.25	0.25	29

(Continued)

*Tests performed in standing water aquaria.

Results of whole-body analyses are presented in table 6B and table 7B. Table 6B shows residues of DDT, DDD, and DDE in the fish fed DDT weekly in the diet, and demonstrates the relatively large amounts stored in the fish fed 3 and 1 mg/kg of body weight. The fish fed the three lowest levels contained the insecticide in amounts not much different than those in the control group. Table 7B shows residue measurements in fish given DDT monthly in baths. It is seen that the fish bathed in the two highest levels stored the most insecticide, and that the lower three levels resulted in stored amounts similar to those in the control fish.

Variation from fish to fish was high in both fed fish and in those given DDT baths.

Mortalities occurring in the different lots of trout because of stresses of various kinds are summarized in table 8B. The data show that the highest day-to-day mortalities took place in the two highest treatment lots of both the fed and the contact fish, with the greatest number of deaths in the lot fed 3 mg/kg of body weight.

Histopathological studies on these fish revealed no pathology attributable to DDT. Microhaematocrit measurements made on the trout throughout the experiment showed no differences between treated and untreated fish.

Differences among lots in average sizes of fish developed during the experiment. Table 9B presents average weights of trout, and shows that fish in lots receiving the greatest amounts of DDT had the greatest average weights. Since the highest mortalities also occurred in the lots receiving the most DDT, it is suggested that selection of less vigorous individuals accounts for the differences in growth. The DDT was probably responsible for mortality in the smaller or weaker fish, leaving the larger or stronger ones to survive and grow at faster rates than the untreated fish.

2,4-D and Bluegill Sunfish (G. H. Wallen)

An experiment to measure chronic effects of 2,4-D on bluegill sunfish in treated ponds was begun at Tishomingo, Oklahoma, in July, 1961. Three ponds were partitioned with polyvinyl chloride sheeting to provide six testing spaces for fish. Each subdivision measured 1/10 acre. One space was used for the untreated control, and each of the others was treated with one of five concentrations of Esteron 99, propylene glycol butyl ether ester of 2,4-D, the treatment concentrations being 10, 5, 1, 0.5, and 0.1 ppm.

The herbicide remained in the ponds near treatment concentrations for about 3 weeks, and all disappeared from the water after 12 weeks.

The control of the aquatic weeds in the treated ponds varied in effectiveness. The pond treated at 10 ppm had 80-100 percent control of Chara, Potamogeton, Najas, Digitaria, Salix, and Typha. Some regrowth of Chara took place after 12 weeks. Death of weeds in other

ponds ranged from 0-100 percent, depending upon the kind of weed and the treatment level.

Mortality among fish was 19 percent in the 10 ppm pond in the first week, but there were essentially no deaths in the other ponds. Applications at the same rates in adjacent ponds in 1962 resulted in 100 percent mortality at 10 and 5 ppm. Spawning was delayed for 2 weeks in the 10 ppm pond; all other lots spawned at the normal time. Fry production appeared to be essentially the same in all lots.

Whole-body analyses of fish for 2,4-D were performed, and the results are reported in table 10B. It is seen that residues were found in fish from only the ponds with the two highest concentrations, and in relatively small amounts.

Histopathological examination of sampled bluegill was made by Dr. E. M. Wood (1962, unpublished). Three kinds of pathology were found in the fish, involving the liver, vascular system, and brain. Liver glycogen was markedly depleted, accompanied by shrinkage, irregular staining characteristics, and loss of normal morphology of liver parenchyma cells. Globular deposits suggestive of glycoproteins appeared throughout the vascular systems. A marked stasis and engorgement of the circulatory system of the brain occurred. Surviving fish recovered completely, and after 112 days no pathological changes were seen.

Microhaematocrit readings did not differ among the lots of fish.

DDT in Blackburn Pond (W. R. Bridges, B. J. Kallman, A. K. Andrews)

A $\frac{1}{2}$ -acre pond on the Blackburn property near Denver was treated with DDT by our personnel at the relatively low rate of 0.02 ppm in July, 1961, for a study on the breakdown and distribution of the compound in a warm-water pond. Sampling of the existing fish population, rainbow trout and bullheads, and of water, crayfish, aquatic vegetation, and bottom sediments, was carried on after the application of DDT, and the samples were analyzed for residues of the insecticide and its metabolites. Sampling was continued until the termination of the study in November, 1962.

The concentration of DDT in the pond water was at its highest point 30 minutes after treatment. A decline in DDT levels then took place; none could be detected 21 days later. Aquatic vegetation contained 6-30 ppm of DDT plus DDE plus DDD in the first week after treatment and declined to 1 ppm in 65 days. In the bottom mud there was 8.3 ppm of the DDT complex after 24 hours; after the third day the concentrations were 1.5 ppm and lower.

Bullheads and trout contained the greatest amounts of chlorinated hydrocarbon 30-40 days after treatment, with concentrations over 4 ppm. Levels slowly declined after that, averaging 3.5 ppm in samples taken 9 and 10 months after treatment in both species, and 3 ppm in rainbow trout taken 14 months after treatment. Crayfish developed lower DDT residues than did trout, and contained 0.33 ppm after 14 weeks.

Table 2B. Toxicity expressed as 24-, 48-, or 96-hour LC₅₀ of various herbicides, fungicides, and antibiotics to rainbow trout, bluegill, and redear sunfish

Toxicant	Species	Weight or length	Temperature °F.	Estimated LC ₅₀ , µg per liter ^{1/}		
				24-hour	48-hour	96-hour
Amchem's silvex	Bluegill	2 in.	65	700 (18 hrs.)	600 (32 hrs.)	-
Amitrol-T	Bluegill	2 in.	65	(No mortality at 10,000 g/l over 100 hrs.)		
Antimycin A.	Rainbow	2-3 in.	55	0.25 (18 hrs.)	-	-
Casoron	Redear	3 g.	65	(No mortality at 20,000 g/l at 48 hrs.)		
Diquat	Rainbow	2-3 in.	55	(No mortality at 10,000 g/l over 100 hrs.)		
Esteron 99	Bluegill	2 in.	65	1,200 (18 hrs.)	-	-
Esteron 99, EC	Bluegill	0.6 g.	75	700	-	-
Fenac, sodium salt	Redear	3 g.	75	(No mortality at 12,000 g/l at 48 hrs.)		
Fenac, sodium salt	Rainbow	0.6 g.	65	10,000	7,500	-
Kurasol	Bluegill	0.6 g.	75	120,000	-	-
Weedar MCP	Bluegill	2 in.	65	(No mortality at 10,000 g/l over 100 hrs.)		

^{1/} Times of exposure are indicated in parentheses where they deviate from times in column heads.