

US EPA ARCHIVE DOCUMENT

CASE GS0315

LINDANE

PM PM# 04/05/84

CHEM 009001

Lindane (gamma isomer of benzene hexac

BRANCH EEB

DISC 40 TOPIC 05054543

FORMULATION 00 - ACTIVE INGREDIENT

FICHE/MASTER ID 00105344

CONTENT CAT 01

Katz, M. (1961) Acute toxicity of some organic insecticides to three species of salmonids and to the threespine stickleback. Trans. Am. Fish Soc. 90(3):264-268. (Also in unpublished submission received Nov 1, 1970 under unknown admin. no.; submitted by Hercules, Inc., Agricultural Chemicals, Wilmington, DE; CDL:005106-0)

SUBST. CLASS = S.

DIRECT RVW TIME = 3 (MH) START-DATE 5/10/85 END DATE 5/10/85

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10. Materials and Methods:

- A. Test Animals: Spring chinook (*Oncorhynchus tshawytscha*); coho salmon (*O. kisutch*); rainbow trout (*Salmo gairdneri*)-- all age group 0; adult threespine stickleback (*Gasterosteus aculeatus*). The chinooks were obtained from the McKenzie Salmon Hatchery of the Oregon Fish Comm.; the coho salmon were collected from the headwaters of the Yaquina River in Lincoln County, OR; rainbow trout were from the Roaring River Hatchery of the Oregon Fish Comm.; and sticklebacks were collected from the tidal sloughs adjacent to Yaquina Bay, OR.

	<u>Weights</u>	<u>Lengths (TL)</u>
Chinook	1.45 - 5 g	2 - 4 1/4 in
Coho	2.7 - 4.1 g	2 1/4 - 3 in
Rainbow	mean 3.2 g	2 - 3 1/8 in
Stickleback	0.38 - 0.77 g	7/8 - 1 3/4 in

- B. Dose: The following pesticides and the percent AI were tested:

Lindane	- 100%	Methoxychlor	- 89.5%
Aldrin	- 88.4%	DDT	- pure
Toxaphene	- 67-69% as Cl	Carbaryl	- 95%
Dieldrin	- 90%	Guthion	- 93%
Endrin	- 98.2%	Malathion	- 57%
Chlorodane	- 100%	Co-Ral	- 98%
Heptachlor	- 72%		

Stock solutions were prepared by dissolving 100 mg of the pesticide to a volume of 100 ml with pure acetone. Highly toxic chemicals were diluted at the rate of a ml of the original stock solution to 100 ml with pure acetone. With Co-Ral, weighed amounts were added directly to the aquaria. Control aquaria contained acetone in the diluted water.

- C. Study Design: Fish were acclimatized overnight before the tests began. Each aquarium contained 15 liters of soft water at 20 °C and pH 6.8 to 7.4. There were 3 to 5 salmonids per aquarium and 11 to 20 sticklebacks per aquarium; the loading per aquarium was 1 g fish/liter water. There were 10 to 20 fish per test concentration. The sticklebacks were tested at 5 and 25 ppt salinity. The test solutions were aerated to maintain an adequate level of D.O.
- D. Statistical Analysis: The method used to calculate the TLM was not given.

11. Reported Results:

The attached tables list the TLM's for the four species and several pesticides. The 96-hr TLM's for lindane were:

Chinook	- 40 ppb ai
Rainbow	- 38 ppb ai
Coho	- 50 ppb ai
Stickleback	- 44 ppb ai at 5 ppt
	- 50 ppb ai at 25 ppt

12. Author's Conclusions/QA Statement:

Endrin was consistently the most toxic insecticide and Co-Ral was the least toxic. In general the cohos and rainbow were more tolerant than the chinooks. Salinity did not play a significant role in the response of the sticklebacks.

No QA statement.

13. Reviewer's Discussions:

A. Test Procedures:

The test procedures generally follow those recommended by EPA except for two major points—the use of aeration and the high temperature (20 °C). Salmonids should be tested at 12 °C.

B. Statistical Analysis:

Without the raw data, the reported results could not be verified.

C. Discussion/Results:

We cannot accept the results if aeration is used and the concentrations are not measured.

D. Adequacy of the Study:

1. Category: Invalid
2. Rationale: Aeration, high test temperature, lack of raw data, no description of statistical methods.
3. Repairability: None

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TOXICITY OF ORGANIC INSECTICIDES TO SALMONIDS AND STICKLEBACK

TABLE 1.—Estimated median tolerance limits (TL₅₀) of some insecticides for chinook salmon, coho salmon, and rainbow trout
[Expressed in parts per billion active ingredients]

Insecticide	Chinook salmon				Coho salmon				Rainbow trout			
	24	Hours		96	24	Hours		96	24	Hours		96
Toxaphene	7.9	3.3	2.7	2.5	13.0	10.5	10.0	9.4	11.5	8.4	8.4	8.4
Aldrin	12.4	10.6	8.7	7.5	-	61.0	48.6	45.9	42.4	23.9	20.3	17.7
Dieldrin	7.9	6.7	6.1	6.1	17.5	15.3	14.4	10.8	15.7	13.0	9.9	9.9
DDT	34.0	17.0	14.0	11.5	66.0	46.0	41.0	41.0	42.0	42.0	42.0	42.0
Lindane	56.0	42.0	42.0	40.0	60.0	56.0	56.0	50.0	42.0	41.0	39.0	36.0
Methoxychlor	23.0	27.9	27.9	27.9	66.2	66.2	66.2	66.2	62.6	62.6	62.6	62.6
Heptachlor	32.4	26.6	23.0	17.3	61.9	60.4	60.4	39.0	36.7	33.8	23.9	19.4
Chlordane	59.0	59.0	57.0	57.0	100.0	86.0	82.0	36.0	56.0	44.0	44.0	44.0
Endrin	2.0	1.2	1.2	1.2	1.3	0.8	0.52	0.51	0.79	0.58	0.58	0.58
Guthion	6.8	6.2	4.3	4.3	7.0	5.0	4.8	4.2	4.7	3.8	3.8	3.2
Malathion	24.5	23.9	23.6	23.0	-	-	-	-	-	-	-	-
Co-Ral	-	-	-	-	22,000.0	20,000.0	18,000.0	15,000.0	-	1,800.0	1,500.0	1,500.0
Sevin	-	-	-	-	1,230.0	997.0	997.0	997.0	-	1,600.0	1,350.0	1,350.0

maintained adequately by surface reeration, the test solutions were aerated with compressed air. Glass tubing of 7 millimeters outer diameter was used to introduce the air into the test aquaria. The tubes could be removed at the end of each test and cleaned thoroughly. There was no indication that aeration changed the toxicity of the toxicant dilutions.

The insecticides were introduced into 15 liters of water taken from the same source as the water in which the fish were held. The pH values of the diluent water varied between 6.3 and 7.1 but usually were near the higher value. Total alkalinity as CaCO₃ varied between 15 and 57 p.p.m. The diluent water was stored in the constant temperature room from 3 to 14 days before use. Test aquaria were 5-gallon wide-mouth glass jars.

Three to five salmonids were tested in each jar, while 11 to 20 sticklebacks were used per jar. The numbers of test fish per jar were varied to insure that not more than 1 gram of fish per liter of water was present. Ten to 20 fish were used at each concentration of toxicant at which some fish survived for 48 hours or more. To conserve experimental fish, only five fish were used at the concentrations at which all fish survived for 96 hours or at which all died within the first 48 hours in the initial tests. Test concentrations were selected in accordance with Table 1 of Doudoroff *et al.* (1951). Each compound was tested in replicate during successive weeks with different samples of fish. Tests were terminated at the end of 96 hours.

The insecticides tested included nine chlorinated hydrocarbon compounds, three organic

phosphorus compounds, and one carbamate. The formulations of chlorinated hydrocarbons were:

- Aldrin, technical—83.1 percent aldrin, 4.6 percent related compounds;
- Dieldrin, technical—90 percent dieldrin;
- Endrin, purified—93.2 percent endrin;
- Chlordane, reference standard—100 percent chlordane;
- Heptachlor, technical—72 percent heptachlor, 28 percent related compounds;
- Methoxychlor—49.5 percent p, p' isomer;
- Toxaphene, reference standard—highest purity, containing 67-69 percent chlorine;
- DDT, reference standard—pure p, p' isomer; and
- Lindane—100 percent lindane.

The insecticides used in this study, with the exception of the endrin, were sent to the author by Croswell Henderson (R. A. Taft Sanitary Engineering Center, Cincinnati) and were subsamples of the toxicants used in the bioassays reported by Henderson *et al.* (1959). Their chemical composition and useful physical properties are adequately discussed by those authors and in published material distributed by manufacturers.

The organic phosphorus compounds tested were malathion, Guthion, and Co-Ral. The formulations tested, the active ingredients, and the compositions of these compounds were:
Malathion—57 percent malathion, 43 percent inert ingredients. (The chemical formulation and pertinent physical properties are discussed in some detail by Henderson and Pickering, 1958).
Guthion, technical—93 percent active ingredient. Chemical name: O,O-dimethyl

TABLE 2.—The 96-hour median tolerance limits of some insecticides for rainbow trout, chinook salmon, coho salmon, fathead minnow, bluegill, goldfish, and guppy
[Expressed in parts per billion active ingredients]

Insecticide	Chinook salmon	Coho salmon	Rainbow trout	Fathead minnow ¹	Bluegill ²	Goldfish ²	Guppy ²
Toxaphene	2.5	9.4	8.4	7.5	3.5	5.6	20.0
Aldrin	7.5	45.9	17.7	33.0	13.0	24.0	33.0
Dieldrin	6.1	10.8	9.9	18.0	7.9	37.0	22.0
DDT	11.5	44.0	42.0	32.0	18.0	27.0	43.0
Lindane	40.0	50.0	38.0	62.0	77.0	152.0	134.0
Methoxychlor	27.9	66.2	62.6	64.0	62.0	56.0	120.0
Heptachlor	17.3	59.0	19.4	94.0	19.0	230.0	177.0
Chlordane	57.0	54.0	44.0	52.0	22.0	82.0	190.0
Endrin	1.3	0.51	0.58	1.0	0.8	1.9	1.5
Malathion	23.0	—	—	12,500.0 ³	95.0 ³	—	—
Co-Ral	—	15,000.0	1,500.0	18,000.0 ³	180.0 ³	—	—
Sevin	—	997.0	1,350.0	6,700.0 ³	5,300.0 ³	—	—
Guthion	4.3	4.2	3.2	83.0 ³	5.2 ³	—	—

¹Henderson et al. (1959), unless otherwise designated.
²Henderson and Pickering (1958).
³Henderson et al. (1960).

S-(4-oxo-1, 2, 3-benzotriazinyl-3-methyl) phosphoro-dithionate. Properties: solid, soluble in water at approximately 1:30,000, soluble in many organic solvents.²

Co-Ral—98 percent active ingredient. Chemical name: O,O-diethyl O-(3-chloro-1-methylumbelliferone) phosphorothionate. Properties: white crystalline powder with characteristic pungent odor, slightly soluble in water and readily soluble in acetone (O'Brien and Wolfe, 1959).

Sevin, which was also tested, may be classed as a carbamate. The following information about Sevin is available:

Sevin—95 percent active ingredient. Chemical name: N-methyl-1-naphthyl carbamate. Properties: slightly colored, light to lavender, nearly odorless, crystalline, solubility less than 0.1 percent in water.³

Stock solutions of the above insecticides were prepared by dissolving 100 milligrams of the toxicant to a volume of 100 milliliters with pure acetone. In order to make accurate dilutions of a compound which was highly toxic to fish, a weaker stock solution was prepared by diluting 1 milliliter of the original stock solution to 100 milliliters with pure acetone. The only exception to the above procedures was with Co-Ral, of which some test solutions were prepared by adding weighed

²Chemagro Corporation. (1957) Guthion. New York 16, New York.
³Union Carbide and Chemicals Company. (1958) Technical information on Sevin for research workers. New York 17, New York. 12 pp.

amounts of the insecticide directly to the water in the test aquaria.

Control aquaria contained water to which was added the largest amount of acetone used in any experiment in the bioassay series. No mortalities were observed in the control. The experimental data are expressed in parts per billion of the active ingredient. No attempt was made to test any of the special formulations commonly used in insect-control procedures.

RESULTS AND DISCUSSION

The estimated median tolerance limits (TL₅₀) for chinook and coho salmon and rainbow trout fingerlings of the insecticides tested are listed in Table 1. The 21-, 48-, 72-, and 96-hour median tolerance limits of the insecticides are expressed in parts per billion (p.p.b.) of active ingredient. The 96-hour TL₅₀ estimates for chinook and coho salmon and rainbow trout are listed in Table 2, along with the estimates obtained by Henderson et al. (1959) and by Henderson and Pickering (1958) for warm-water and aquarium species. The median tolerance limits (TL₅₀) for marine threespine sticklebacks in salinities of 5 and 25 parts per thousand are listed in Table 3.

Of the compounds tested, endrin was consistently the most toxic to all species of salmonids, with a 96-hour TL₅₀ for chinooks of 1.2 p.p.b., for cohos of 0.51 p.p.b., and for rainbow trout of 0.58 p.p.b. Co-Ral is the least toxic of the compounds tested, with an estimated 96-hour TL₅₀ of about 15,000 p.p.b. for cohos and 1,500 p.p.b. for rainbows. In general, the cohos and rainbows were more

TABLE 1

Insecticide
Toxaphene
Aldrin
Dieldrin
DDT
Lindane
Methoxychlor
Heptachlor
Chlordane
Endrin
Guthion
Malathion
Co-Ral
Sevin

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TOXICITY OF ORGANIC INSECTICIDES TO SALMONIDS AND STICKLEBACK

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TABLE 3.—Estimated median tolerance limits for sticklebacks of some insecticides in waters of 5 and 25 parts per thousand salinity

[Expressed in parts per billion active ingredients]

Insecticide	5 parts per thousand salinity				25 parts per thousand salinity			
	24	48	72	96	24	48	72	96
Triazophos	—	12	9.8	8.8	—	—	8.8	7.8
Aldrin	—	48.3	41.5	39.8	—	44.2	37.4	27.4
Dieldrin	20.7	18.9	18.0	15.3	—	—	13.5	13.1
DDT	22.0	21.0	18.5	18.0	18.0	15.0	14.5	11.5
Lindane	50.5	45.0	45.0	44.0	66.0	51.0	50.0	50.0
Methoxychlor	93.6	86.4	86.4	86.4	—	89.1	69.1	69.1
Heptachlor	120.8	111.9	111.9	111.9	111.9	111.9	111.9	111.9
Chlorolane	118.0	118.0	90.0	90.0	180.0	170.0	170.0	160.0
Endrin	—	0.45	0.45	0.44	—	—	0.58	0.50
Guthion	15.8	15.8	14.9	12.1	8.9	5.0	4.8	4.8
Malathion	96.9	94.0	94.0	94.0	76.9	76.9	76.9	76.9
Co-Ral	—	2,234.0	1,862.0	1,862.0	—	1,764.0	1,568.0	1,470.0
Sevin	—	16,625.0	6,172.0	3,990.0	—	10,450.0	4,910.0	3,990.0

tolerant to most of the insecticides tested than were the chinooks. Chinooks were more tolerant to endrin than were the cohos and rainbows, but the difference was minor. The differences in tolerance of the three species to most of the insecticides are not large.

A comparison of the 96-hour TL_{50} estimates for the salmonids with the data obtained by Henderson *et al.* (1959) and Henderson and Pickering (1958) shows that the salmonids, especially chinook salmon, were usually somewhat more sensitive than the warm-water and aquarium species, although the salmonids were tested at 20° C. instead of 25° C., at which the warm-water species were tested.

Coho salmon were able to tolerate relatively high concentrations of Co-Ral and Sevin, with the respective 96-hour TL_{50} values being 15,000 and 977 p.p.b. These data suggest that useful insecticides can be developed that would not be highly toxic to fish. Co-Ral, which is the least toxic of the compounds tested, is used principally as a systemically active insecticide and is in no sense, at least at present, a replacement for the more versatile insecticides. Sevin, with a 96-hour TL_{50} for coho salmon of about 1,000 p.p.b., is reported to have a wide spectrum of insecticidal activity. This compound therefore might be suggested as a substitute for substances of pronounced lethality to fish in insect-control programs where possible damage to fish populations is a consideration.

No insecticide differed much in its toxicity to sticklebacks in water of 5 and 25 parts per thousand salinity. Eight of the 13 substances tested were slightly more toxic, while three substances were slightly less toxic in the more

saline water. The greatest difference was noted in the case of Guthion. The median tolerance limits obtained with the marine sticklebacks at salinities of 5 and 25 parts per thousand (Table 3) did not show any striking differences from the data obtained with the salmonids in fresh water.

Those who may wish to use these data as a guide in planning an insecticide spraying program should exercise some caution. The termination of these experiments at 96 hours does not imply that no deaths would occur after this period. For example, no mortalities were observed at concentrations of 10,000 p.p.b. Co-Ral at the end of the 4-day test period. The fish were allowed to remain in the toxicant solution for 5 additional days, and at the end of the 9-day period, all of the fish had died. There were instances of similar delay of mortality with some other insecticides, particularly endrin.

Weiss (1959) showed convincingly that the known undesirable effect of some organic phosphorus insecticides (i.e., the inhibition of acetylcholinesterase activity) persists as long as 45 days after contact in some fish. Even if mortalities do not occur following insecticide applications, the depression of normal activity patterns may make some fish unable to carry on their functions with the competitive efficiency required for survival in their natural environment.

ACKNOWLEDGMENTS

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