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Biological Studies on PCBs

Acute toxicity to fish The toxicity of the Aroclor® series was determined using the standard 96-hr static bioassay and cutthroat trout. Additional tests were also made with bluegills and channel catfish (Table 1). The solubilities of the Aroclor® formulations in water are reported in the range of 0.1-0.3 µg/l.

The results of these tests indicate a decrease in acute toxicity for Aroclor® 1254 and higher. The PCB toxicity indicated by these tests is misleading in that most fish mortalities occurred in the last 24 hours of the test and the concentrations of the test solutions were considerably above the true solubilities. The toxicities of these compounds became more apparent in long term, intermittent-flow bioassays.

The acute oral toxicity of Aroclor® 1242 and 1260 to rainbow trout was greater than 1500 mg/kg (given *per os* for 5 days at 300 mg/kg/day).

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MONS 041403

Table 1. Acute toxicity[1] of several Aroclors® to selected fishes.

Aroclor®	Species	Temperature (C)	96-hr TL_{50} (µg/l)
1221	Cutthroat trout	8.9[2]	1,170
1232	" "	8.9	2,500
1242	" "	8.9	5,430
1248	" "	8.9	5,750
1254	" "	8.9	42,500
1260	" "	8.9	60,900
1262	" "	8.9	> 50,000
1268	" "	8.9	> 50,000
4465	" "	8.9	> 50,000
5442	" "	8.9	> 50,000
5460	" "	8.9	> 50,000
1248	Channel catfish	18.3[3]	6,000
1254	" "	18.3	12,000
1248	Bluegills	18.3[3]	278
1254	"	18.3	2,740

1/ Static bioassay

2/ Alkalinity, 159 ppm; pH 7.6

3/ Alkalinity, 35 ppm; pH 7.1

Chronic toxicity to fish Intermittent-flow tests were conducted with rainbow trout, channel catfish, bluegills, and yellow perch and Aroclors® 1242, 1248, 1254 and 1260. Rainbow trout were the most susceptible to the Aroclor® compounds, then yellow perch, bluegills, and channel catfish (Table 2). Aroclor® 1248 was the most toxic to all species tested. Aroclor® 1242 was equal or perhaps slightly more toxic than 1254. The test with yellow perch and 1248 serves as a model for estimating time-independent toxicity (Figure 1).

Table 2. Toxicity of four Aroclors® and DDT to four species of fish in dynamic exposures

Species	Compound	TL_{50}s (ppb) after 5 to 35 days						
		5	10	15	20	25	30	35
Rainbow trout	DDT	4.2	1.4	0.5 (0.3-0.9)	0.4	0.2	-	-
	Aroclor 1242	67	48	18 (11-31)	10	12	-	-
	Aroclor 1248	54	38	16 (9-31)	6.4	3.4	-	-
	Aroclor 1254	-	160	64 (29-140)	39	27	-	-
	Aroclor 1260	232	326	143 (33-610)	78	49	51	-
Channel catfish	DDT	-	-	2.3	1.3	0.86	0.57 (0.28-1.1)	-
	Aroclor 1242	-	125	219	150	132	87 (46-164)	-
	Aroclor 1248	-	121	121	115	104	75 (29-197)	-
	Aroclor 1254	-	303	286	293	181	139 (94-349)	-
	Aroclor 1260	-	535	482	512	465	433 (185-1010)	-
ucgills	DDT	1.2	1.4	1.6	1.3	0.59 (0.39-0.89)	0.35 (0.21-0.58)	-
	Aroclor 1242	-	-	164	125	120	84 (48-150)	-
	Aroclor 1248	136	115	111	106	100	78 (42-143)	-
	Aroclor 1254	-	-	303	260	239	177 (97-321)	-
	Aroclor 1260	-	-	-	-	-	-	-

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Table 2. Continued

Species	Compound	TL_{50}s (ppb) after 5 to 35 days						
		5	10	15	20	25	30	35
Yellow perch	Aroclor 1242	-	-	-	135	94 (78-114)	92	-
	Aroclor 1248	-	-	110	90	78	72 (58-88)	-
	Aroclor 1254	-	-	128	92	92	92 (76-110)	-
	Aroclor 1260	-	-	-	-	-	240	185 (139-247)

Water: pH = 7.4, alkalinity=237 ppm, hardness=272 ppm, and 17° C.
No value means insufficient data for computation of TL_{50}.

AROCLOR® 1248 - YELLOW PERCH

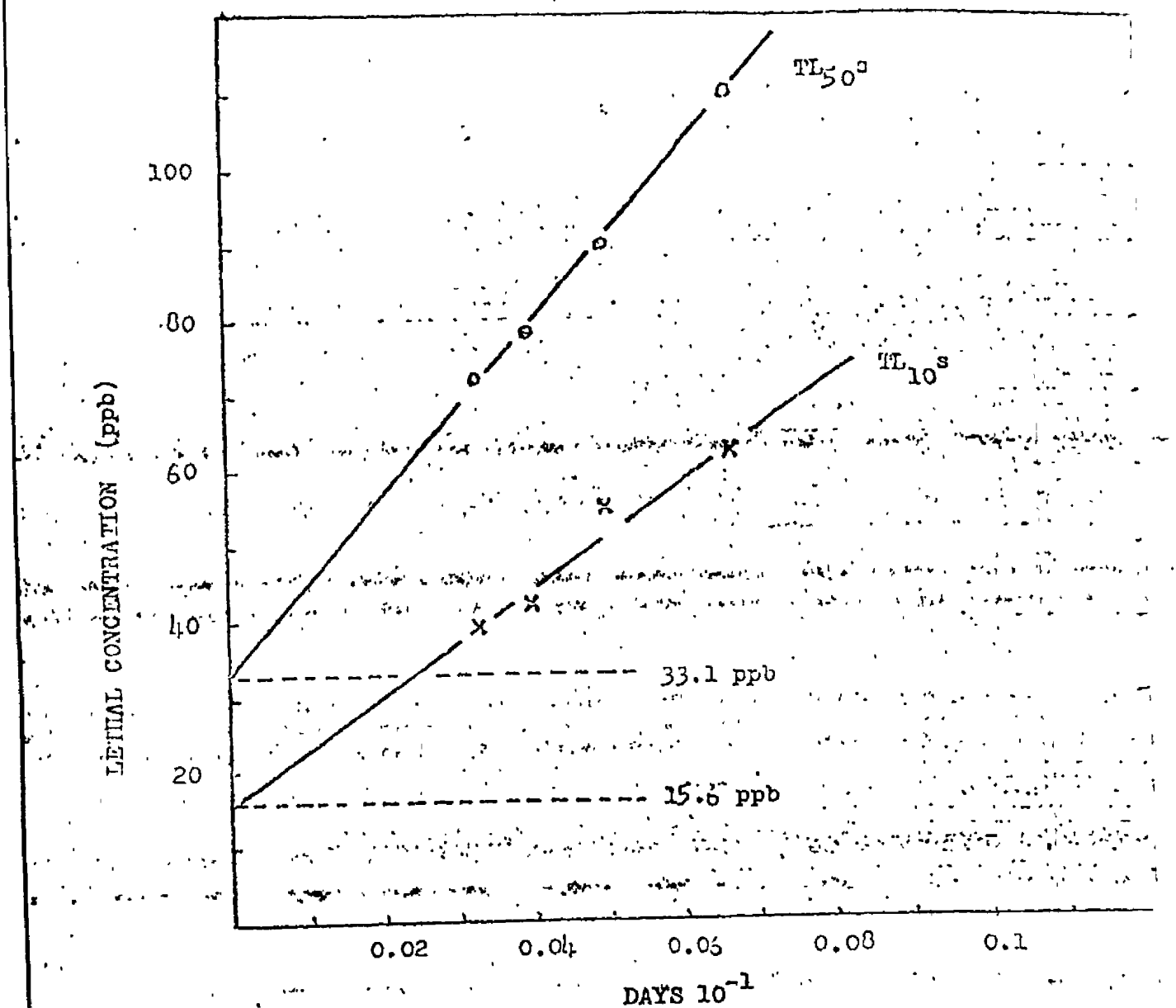

Figure 1. Estimation of time-independent toxicity by intermittent-flow bioassay.

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Toxicity to Invertebrates Static and continuous-flow tests with invertebrates did not differ as much as those with fish. In general, insects were much more resistant to the Aroclors® than were crustacea (Table 3). Scud (Gammarus fasciatus) were also exposed to Aroclor® 1254 for 7 days in an intermittent-flow diluter at 17° C and the TL_{50} value was 34 µg/l (24-45). Homologues having lower chlorine numbers were more toxic to scud than those having higher chlorine numbers (Table 4).

Bioconcentration by aquatic microorganisms Aroclors®, like some of the organochlorine insecticides, appear to be rapidly and greatly concentrated by various organisms. For example, preliminary experiments showed that Daphnia magna concentrated ^{36}Cl-Aroclor® 1254 by 45,000 times after only 48 hours of exposure to a concentration of 300 ng/l. These data closely parallel that obtained in DDT and aldrin investigations with Daphnia magna.

Bioconcentration by aquatic invertebrates The potential of 1254 to accumulate in fish-food organisms was investigated by exposing several species of aquatic invertebrates to a ^{36}Cl-labeled formulation of this chemical in a continuous-flow system. All invertebrates exposed to 1254 showed an initial period of rapid uptake and residue magnification many times greater than the concentrations in water. For example, late instar mosquito larvae Culex tarsalis exposed to 1.5 µg/l of 1254 accumulated a residue of 19 mg/kg within 24 hours. This represents a 12,600-fold increase over the residues in water. The tissue residues continued to accumulate until the seventh day when the mosquito larvae transformed into pupae.

Table 3. Aroclor® and DDT toxicity to invertebrates.

Compound	Organism[1]	Bioassay type[2]	Exposure (days)	TL_{50} (ug/l)
Aroclor® 1242	Crayfish	static	7	30
Aroclor® 1254	Crayfish	static	7	100
Aroclor® 1254	Crayfish	continuous-flow	7	80
DDT	Crayfish	static	4	100
Aroclor® 1242	Scud	continuous-flow	4	10
Aroclor® 1242	Scud	continuous-flow	10	5.0
Aroclor® 1248	Scud	static	4	52
Aroclor® 1254	Scud	static	4	2,400
DDT	Scud	static	4	3.2
DDT	Scud	continuous-flow	5	0.6
Aroclor® 1254	Glass shrimp	continuous-flow	7	3.0
DDT	Glass shrimp	static	5	1.0
DDT	Glass shrimp	continuous-flow	5	1.3
Aroclor® 1242	Dragonfly	static	7	800
Aroclor® 1254	Dragonfly	static	7	1,000
Aroclor® 1242	Damselfly	continuous-flow	4	400
Aroclor® 1254	Damselfly	continuous-flow	4	200
DDT	Damselfly	static	4	56

1/ Crayfish (*Orconectes nais*), Scud (*Garmarus fasciatus*), Glass shrimp (*Palaemonetes kadiakensis*), Dragonfly (*Macromia* sp.), Damselfly (*Ischnura verticalis*).

2/ Temperature, 15.6° C; alkalinity, 35 ppm; pH, 7.1

Table 4. Toxicity[1/] of five PCB homologues to scud[2/] at 21 C.

PCB isomer	TL_{50} (mg/l) and 95% confidence intervals at -- 24 hr	96 hr
2,3,4' trichlorobiphenyl	0.10 (0.08-0.14)	0.07 (0.05-0.12)
4,4' dichlorobiphenyl	0.16 (0.11-0.23)	0.10 (0.06-0.15)
2,4' dichlorobiphenyl	0.15 (0.10-0.20)	0.12 (0.08-0.18)
2,4,6,2', 4', 6' hexachlorobiphenyl	0.20 (0.16-0.26)	0.15 (0.09-0.21)
2,4,5,2', 5' pentachlorobiphenyl	0.43 (0.38-0.50)	0.21 (0.12-0.36)

[1/] Static bioassay.

[2/] Gammarus pseudolimnaeus.

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In some tests, we were able to continue the exposure until an apparent plateau or equilibrium concentration was reached within the organism. For example, scud were exposed continuously to 1.6 µg/l of 1254 in water until residues reached an apparent equilibrium concentration at 14 days; total body residues at this time were 38 mg/kg, a concentration 27,000 times that in water. Once this equilibrium was reached within the organism, no further residue magnification was recorded after an additional 21 days of exposure (Figure 2). Waterfleas accumulated a residue 30,000 times the water concentration (1.1 µg/l) in 3 days. In a similar study early instar crayfish Orconectes nais exposed to 1.2 µg/l of 1254 accumulated residues at a slower rate. The crayfish were accumulating 1254 at a linear rate after 28 days of exposure when the experiment was terminated (Figure 3).

Scud exposed to 1254 were analyzed by gas-liquid chromatography to investigate potential shifts in the residue composition which may have occurred as a result of metabolism. However, we found no significant alterations in the chemical, but some isomers were accumulated more than others. The most notable shift of residue components was a two-fold increase in concentrations of the lower chlorine isomers (tri- and tetrachlorobiphenyl isomers). The relative concentration of higher chlorinated isomers (penta- and hexachloro biphenyls) were decreased by one half.

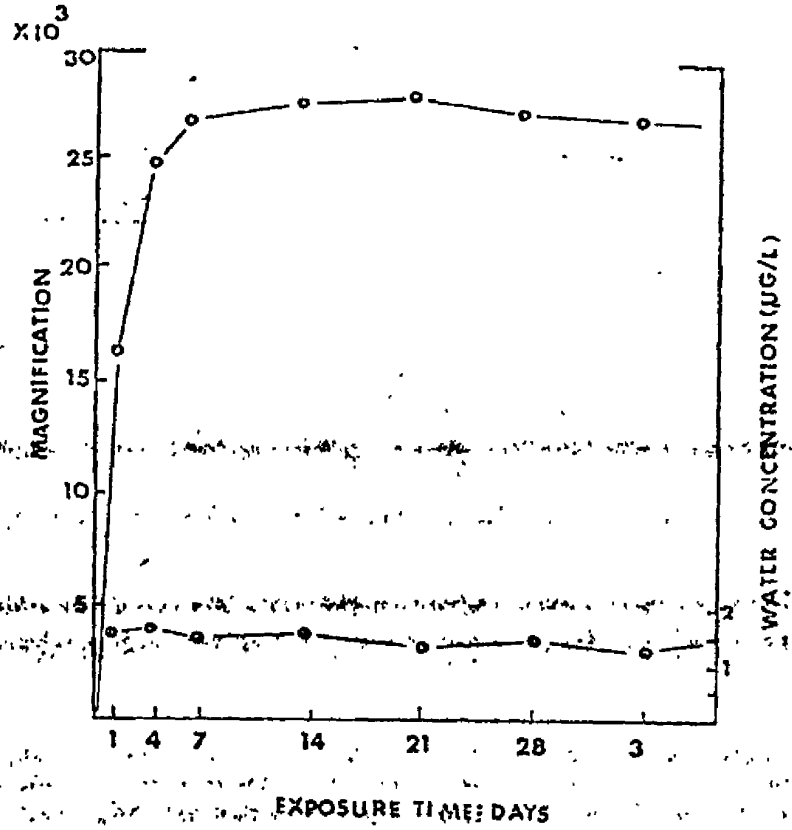

Figure 2. Biological magnification by scud, Gammarus pseudolimnaeus of ^{36}Cl-Aroclor® 1254 from water during 35 days exposure.

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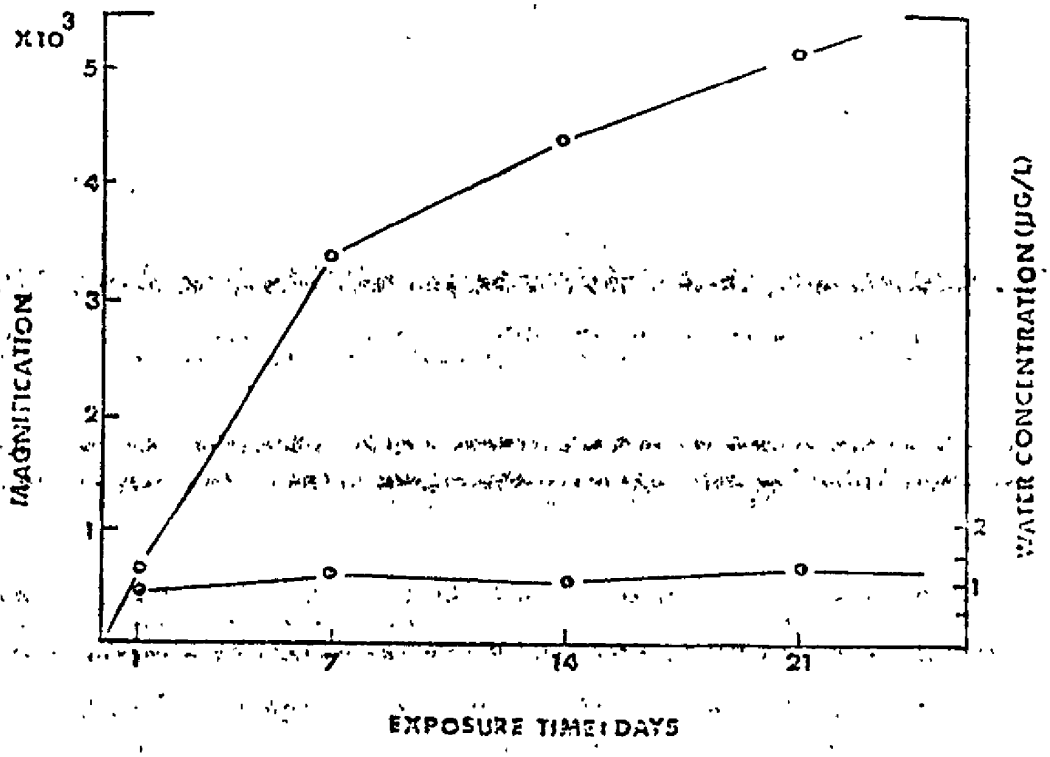

Figure 3. Biological magnification by crayfish Orconectes nais of ^{36}Cl-Aroclor® 1254 from water during 21 days of exposure.

Physiologic effects of PCBs Because the thyroid plays an integral role in reproduction, osmoregulation, and oxidative metabolism, we studied its activity in five species of fish exposed to organochlorine chemicals by diet or bath. Aroclors® 1248 and 1254 had significant effects on thyroid activity (72-hr ^{125}I-uptake) (Table 5). The lowest dietary concentrations that affected thyroid activity were 0.48 µg/gm (ppm in dry food) of Aroclor® 1254. These concentrations are comparable to those found in natural fish foods.

Table 5. Thyroid activities in five species of fish exposed to polychlorinated biphenyls (PCBs) or DDT.

Chemical	Dosage (µg/kg bw/day)	Concentration (ppm)	Species	Exposure (days)	Changes in thyroid activity[1]	Whole body residue[2]
Aroclor 1232	40	1.2	Channel Catfish	160	NC	3.7
Aroclor 1232	400	12	Channel Catfish	160	NC	8.3
Aroclor 1248	40	1.2	" "	160	NC	4.2
Aroclor 1248	400	12	" "	160	NC	14.0
Aroclor 1254	40	1.2	" "	160	+56	5.4
Aroclor 1254	400	12	" "	160	+91	21.0
Aroclor 1260	40	1.2	" "	160	NC	5.5
Aroclor 1260	400	12	" "	160	NC	45.0
Aroclor 1254	1.45	0.05	Coho salmon	260	NC	0.40
Aroclor 1254	14.5	0.48	" "	260	+52	0.59
Aroclor 1254	145	4.8	" "	260	+35	3.87
Aroclor 1254	1450	48	" "	260	+87	54.5
Aroclor 1254	14,500	480	" "	260	+119	646.0

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Table 5. Continued.

Chemical	Dosage (ug/kg bw/day)	Concentration (ppm)	Species	Exposure (days)	Changes in thyroid activity[1]	Whole body residue[2]
Aroclor 1248	-	4	Bluegills	74	NC	NA
Aroclor 1248	-	7.3	" "	74	-29	NA
Aroclor 1254	-	4.6	" "	74	NC	NA
Aroclor 1254	-	7.5	" "	74	+54	NA
Aroclor 1248	-	6.7	Channel catfish	77	+50	NA
Aroclor 1248	-	11.8	" "	77	+50	NA
Aroclor 1254	-	3.5	" "	77	+44	NA
Aroclor 1254	-	5.3	" "	77	+105	NA

1/ Percent change in 72-hr ^{125}I-uptake from controls; NC = no significant difference.

2/ NA = not analyzed; residue concentration = ug/g of wet body weight.

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Growth of lake trout was retarded by Aroclor® 1248 in the diet for 3 months. Concentrations of 1.2, 3.8, and 12 ppm decreased weight gain by 6, 10, and 28%, respectively, below that of controls. Growth of the high-exposure group was only about half that of the controls after 6 months. Conversely, thyroid activity, as indicated by 72-hr radioiodine uptake, was stimulated by PCB exposure (Figure 4). Serum cortisol concentrations ranged from 33 to 46% below that of the controls. Depressed growth rates and elevated thyroid activity imply metabolic inefficiency producing a "loading" stress that can adversely affect viability and reproductive success.

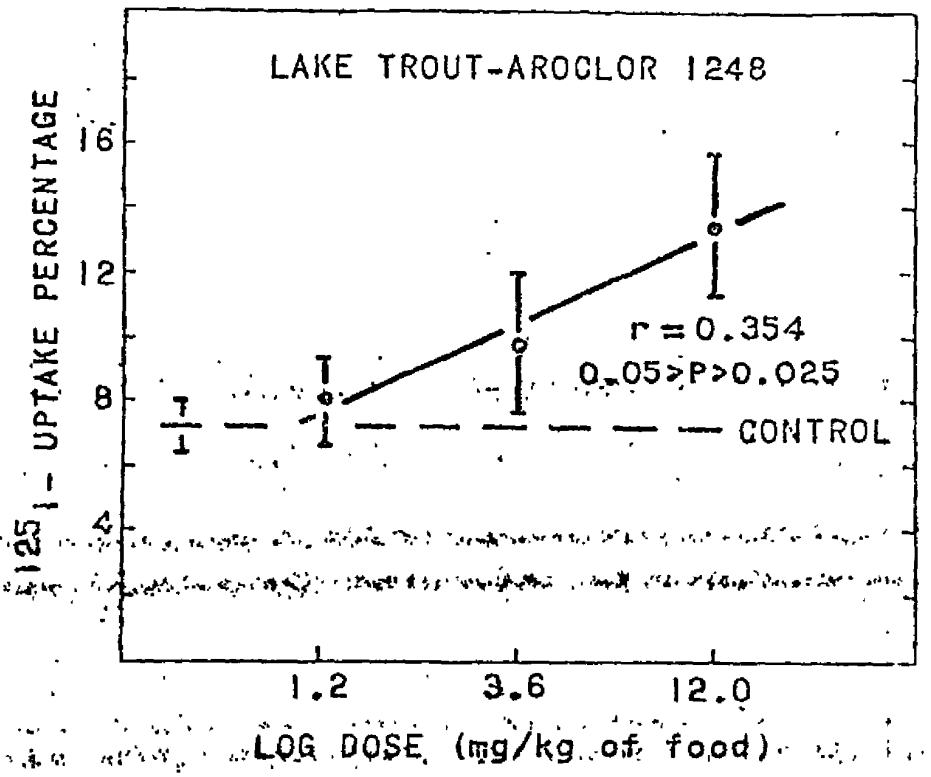

Figure 4. Thyroid stimulation by 6 month dietary exposure to Aroclor® 1248.

Chronic dietary toxicity of PCBs

Aroclor® 1248 was incorporated in diets of juvenile lake trout at concentrations of 0.1, 1.8 and 6.0 mg/kg of food (concentrations comparable to those found in natural foods). These concentrations depressed growth and serum cortisol levels, but stimulated thyroid activity during the first 160 days of the experiment (Table 6a). After 320 days, however, there were few differences between control and treated fish.

Dose-dependent pathologic changes were observed in the gills and livers of PCB-dosed fish. Gill lesions consisted of hyperplasia and hypertrophy of the respiratory epithelium and edema and intercapillary congestion of the lamellae. Changes in the liver included parenchymal degeneration, cytoplasmic vacuolization of the hepatic cells, and pleomorphism of the liver cells.

Whole-body residues increased during the exposure (Table 6b), but after the trout were fed uncontaminated food for 60 days, residues declined an average of 31%, 15%, and 12% in the 0.6-, 1.8-, and 6.0-mg/kg lots, respectively.

Also, lake trout growth was retarded by dietary Aroclor® 1248 and chlordane in combination. Fish that consumed food containing 0.94 µg/g of Aroclor® 1248 and 0.26 µg/g chlordane for 80 days grew 15% less than the controls.

Table 6a. Growth, serum cortisol concentrations, and thyroid activity in juvenile lake trout exposed by feed to Aroclor® 1248.

	Percent growth			Serum cortisol		Thyroid activity	
	160 days	310 days	total growth	160 days	320 days	160 days	320 days
Control	887	2,930	3,817	6.5±0.8	10.3±0.6	3.6±0.4	1.2±0.2
0.6 mg/kg	708	3,017	3,724	3.9±0.4	7.8±1.4	4.0±0.7	1.3±0.1
1.8 mg/kg	777	3,032	3,810	4.3±0.8	11.2±1.4	4.9±1.1	1.2±0.1
6.0 mg/kg	522	3,262	3,783	3.5±0.2	13.7±1.3	6.7±1.1	1.0±0.1

Table 6b. Whole body residues (µg/g) in juvenile lake trout fed Aroclor® 1248.

Exposure (days)	Doses (mg/kg of dry food)			
	0	0.6	1.8	6.0
0	0.17	0.17	0.17	0.17
70	0.11	0.26	0.59	2.00
145	0.09	0.42	1.20	3.70
246	0.15	0.81	2.40	6.80
320	0.13	1.60	2.70	9.70
Recovery[1/]				
60	0.12	1.10	2.30	8.50

1/ Fed non-contaminated food.

230 days mean half-life

MONS 041421

Uptake of dietary PCBs

Dietary doses of 1.45, 14.5, 145, 1,450, and 14,500 µg Aroclor® 1254/kg body wt. per day to coho salmon had no effect on growth, nor produced observable toxicosis until death occurred in the highest dosage group after 260 days. The two highest dosage groups achieved extremely high concentrations of residues (Table 7). Fish fed 14,500 µg/kg began dying on the 260th day, and 5 days later all fish had died. Thyroid activity was increased significantly after 260 days of treatment (Table 5). A detectable stimulation of thyroid activity occurred in salmon dosed with 1/1000th of that causing mortality.

Dietary doses of Aroclors® 1232, 1248, 1254, and 1260 to channel catfish at concentrations of 2.4 and 24 mg/kg (dry wt) caused no mortality nor had any effect on growth. Thyroid activity was altered significantly only by Aroclor® 1254. The low dose increased uptake by 56%, whereas a 91% increase was induced by the high dose. Residue accumulation was greatest in the groups exposed to the high dose of Aroclor® 1260; i.e., as the percent chlorine content of the PCB molecule increased, residue accumulation increased (Table 8).

Table 7. Whole body residues of Aroclor® 1254 (PCB) in coho salmon exposed to dietary concentrations of Aroclor® 1254 at 18° C.

Exposure (days)	PCB residues[1] (µg/g) after exposure to theoretical dietary dosages[2] (µg/kg body wt per day) of --					
	0	1.45	14.5	145	1,450	14,500
14	0.15	0.19	0.28	0.96	8.9	81.7
28	0.15	0.23	0.42	1.32	11.4	114.5
56	0.12	0.18	0.69	2.90	21.8	300.0
112	0.17	0.47	0.50	3.83	21.6	384.0
142	0.16	0.35	0.61	3.52	34.18	361.0
200	0.14	0.36	0.68	2.48	57.14	659.0
260	0.17	0.40	0.59	3.87	54.5	645.5

1/ Pooled samples of four fish.

2/ Dosage corresponds to 0.19 (background in feed), 0.25, 0.64, 6.4, 64, and 640 µg/gm food dry wt., respectively.

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Table 8. Whole body residues of Aroclors® 1232, 1248, 1254, and 1260 in channel catfish after 193 days exposure to dietary concentrations of the PCBs at 25° C.

Aroclor	Dosage[1] (µg/kg bw/day)	Exposure (Days) 17	45	87	150	193
1232	480	9.0[2]	5.4	5.2	11.3	14.5
1232	48	0.8	1.9	1.7	4.5	3.2
1248	480	13.0	2.3	4.3	NA	13.0
1248	48	0.6	0.9	1.3	NA	1.9
1254	480	10.0	0.8	0.8	14.2	21.0
1254	48	0.8	0.03	0.2	2.3	4.8
1260	480	6.2	5.0	7.5	17.0	32.0
1260	48	0.7	1.2	1.7	2.3	3.5

[1] Dosages of 480 and 48 µg/kg body wt. per day corresponds to 24 and 2.4 µg/gm food (dry wt.), respectively.

[2] Mean of three samples (µg/gm).

Uptake of PCBs from water by fish

After 77 days, bluegills and channel catfish exposed to two concentrations of Aroclors® 1248 and 1254 in intermittent-flow diluters, magnified the Aroclors® by a factor of 20,000 to 60,000. Although the mean Aroclor® residue in the fish seemed dependent on water concentration, the magnification factor from water did not. For example, bluegills exposed to 2.6 and 7.3 µg/l of Aroclor® 1254 in water magnified those concentrations by 26,560- and 26,300-times and contained mean residues of 68.3 and 192 µg/g of whole fish, respectively.

Chromatograms of Aroclor® residues in bluegills and catfish showed a difference in storage rates for various Aroclor® components. Uptake appeared biased toward the later eluting components from the GC. After Aroclor® exposure was terminated, PCB elimination by fish was more rapid for the early eluting components.

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