INVESTIGATIONS IN FISH CONTROL

72. Toxicity of Rotenone to Fish in Standardized Laboratory Tests



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- Residues of Quinaldine and MS-222 in Fish Following Anesthesia with Mixtures of Quinaldine Sulfate:MS-222, by Joe B. Sills, John L. Allen, Paul D. Harman, and Charles W. Luhning. 1973. 12 pp.

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INVESTIGATIONS IN FISH CONTROL

72. Toxicity of Rotenone to Fish in Standardized Laboratory Tests

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Contents

Page

Abstract	1
Materials and Methods	1
Results	2
Toxicity to various species of fish	2
Effects of temperature, water hardness, and pH on toxicity	2
Toxicity of Noxfish to green eggs of rainbow trout	2
Persistence of Noxfish in water	6
Detoxification of Noxfish	8
Toxicity of different formulations of rotenone	8
Toxicity in flow-through tests	8
Discussion	10
References	11

Toxicity of Rotenone to Fish in Standardized Laboratory Tests

by

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Abstract

Noxfish[®], which contains 5% rotenone, was toxic to a variety of freshwater fish at concentrations ranging from 21.5 to 497 μ g/l in 96-h laboratory exposures. Goldfish (*Carassius auratus*) and black bullheads (*Ictalurus melas*) were the most resistant species, and the Atlantic salmon (*Salmo salar*) was the most sensitive. Toxicity was influenced little by temperatures of 7 to 22 C, by water hardnesses of 10 to 300 mg/l, or by pH's of 6.5 to 9.5. In exposures of rainbow trout (*Salmo gairdneri*), newly fertilized eggs were much more resistant than fingerlings. Noxfish detoxified in water solutions; the half-life of biological activity was 22 days at 12 C and 13 days at 17 C. Potassium permanganate was an excellent detoxifier; chlorine was less efficient. Noxfish was consistently more toxic in static tests than in flow-through tests.

Rotenone, a crystalline ketone found in several plants of the Leguminosae, has been used as a toxicant by fishery managers since the 1930's for removing undesired fish populations in lakes (Baker and Cordone 1969). Ideal conditions for the reclamation of static waters with rotenone include temperatures between 16 and 21 C, alkalinities between 150 and 200 mg/l(ppm), pH's of 8 or less, and surface areas of less than 8.1 ha (Spitler 1970). The piscicide has been used extensively under a wide variety of conditions and is relatively harmless to most nontarget organisms (Schnick 1974: Lennon et al. 1970). Twenty-nine formulations of rotenone from 18 companies had been registered for aquatic or agricultural use by 1974 by the Environmental Protection Agency. Because the registrations are old, data supporting the labels must be updated to conform to present requirements (Lennon 1967). A guide or protocol for evaluating the toxicity of candidate fishery chemicals for registration was published by Marking (1975).

The present study was designed to determine (1) the toxicity of rotenone to fish in standardized static and flow-through tests, (2) the toxicity of rotenone to newly fertilized trout eggs, (3) the residual toxicity of rotenone in water after selected periods of aging, (4) the efficiency of two compounds used to detoxify rotenone, and (5) the comparative toxicities of three rotenone formulations.

Materials and Methods

Three formulations of rotenone (furnished by S. B. Penick & Co.) were used in the tests described here: Noxfish[®], an emulsifiable concentrate containing 5% rotenone; Pro-Noxfish[®], a synergized emulsifiable concentrate containing 2.5% rotenone; and rotenone, a powder containing 33% rotenone. Except for tests in which the toxicities of the different formulations were compared or evaluated, Noxfish was used in all tests, and concentrations were based on the total amount of Noxfish formulation rather than on the amount of rotenone in the formulation. Stock solutions of the toxicants were prepared daily, and the portions needed to yield the desired concentrations were added to test chambers.

Static and flow-through test procedures followed those of Lennon and Walker (1964) and The Committee on Methods for Toxicity Tests with Aquatic Organisms (1975). In static tests, 10 fish were exposed to each concentration in glass jars containing 15 liters of oxygen-saturated test water prepared from deionized water (Marking 1969). Waters of four levels of hardness were used (total hardness as mg/l of CaCO₃ in parentheses): very soft (10), soft (44), hard (170), and very hard (300).

In separate tests to assess the effect of pH on toxicity, chemical buffers were added to control the pH (Marking 1975). Test temperatures were regulated by immersing the test jars in constant temperature water baths. In flow-through toxicity tests, 20 fish were exposed to each concentration in a system similar to that described by Mount and Brungs (1967). Modifications included electronic microswitches to control cycling, pressure regulators, and an automatic pipette (Micromedic®) for injecting the toxicants into dilution water. Municipal water used for flow-through exposures was carbon filtered and had a total hardness of 320 mg/l and a pH of 7.5. Flow rates were maintained at about five chamber volumes per 24 h. Temperature was maintained with a water bath around the test chambers.

Fish weighing 1 to 1.5 g each were obtained from Federal hatcheries and maintained according to standardized procedures of the Fish Control Laboratory (Hunn et al. 1968). Scientific names for all species used are listed in Table 1. Fish were acclimated to the test conditions for 4 days before they were exposed to the toxicant. Mortalities were recorded at 1, 3, and 6 h on the first day of exposure and daily thereafter for the remainder of the test. Trout eggs were exposed to Noxfish in a manner similar to that used for fish, except that the static test vessel contained 2.5 liters of test solution. Details of methods for exposing eggs were those outlined by Olson and Marking (1973).

The methods of Litchfield and Wilcoxon (1949) were used in computation of the LC $_{50}$'s (concentrations producing 50% mortality) and 95% confidence intervals. Regressions were drawn and inspected for each set of data. All data reported fulfilled the chi-square test requirement for acceptability.

Deactivation indices for Noxfish were derived in soft water at temperatures of 12 and 17 C. Aged solutions of the toxicant were bioassayed to determine the biological activity remaining after selected time periods. The deactivation index was determined by dividing the LC_{50} of aged solutions by the LC_{50} of unaged solutions under corresponding test conditions (Marking 1972). The deactivation index was plotted against aging time on semilogarithmic coordinates to estimate the half-life of biological activity. Detoxification procedures with potassium permanganate (KMnO₄) and chlorine were those used by Marking and Bills (1975).

Results

Toxicity to Various Species of Fish

Noxfish was toxic to a wide variety of fish at concentrations ranging, in 96-h exposures, from 21.5 μ g/l (or parts per billion) for Atlantic salmon to

497 μ g/l for goldfish (Table 1). The 96-h LC ₅₀ was less than 100 μ g/l for bowfins, all six salmonids, northern pike, carp, longnose and white suckers, smallmouth bass, yellow perch, and walleyes. The 96-h LC₅₀ was greater than 100 μ g/l for goldfish, fathead minnows, black bullheads, channel catfish, green sunfish, bluegills, and largemouth bass. Goldfish and black bullheads were most resistant-10 times as resistant as most other species. Generally, the resistant species required longer exposures than did the sensitive species, before they succumbed. None of the goldfish died in 24 h of exposure to high concentrations of Noxfish and none of the black bullheads in 6 h. On the other hand, most of the sensitive species died in 3-h exposures to much lower concentrations.

Effects of Temperature, Water Hardness, and pH on Toxicity

Noxfish was generally less toxic to rainbow trout, channel catfish, and bluegills at the lower than at the higher temperatures in 3- and 6-h exposures (Tables 2, 3, and 4). After 96 h this trend remained but the differences in toxicity associated with most 5° differences in temperature were insignificant. The difference was significant for rainbow trout, however, at 7 and 12 C. Trout were consistently more sensitive than channel catfish or bluegills.

Water hardness had no effect on toxicity of Noxfish (Tables 2, 3, and 4), with one exception; in 96-h exposures the LC₅₀'s for channel catfish were 277 μ g/l in soft water and 328 μ g/l in very soft water (Table 3). Water at each hardness contained 384 mg/l of sodium bicarbonate to equalize the pH at about 8.0, and that quantity of bicarbonate in the soft water presumably resulted in slightly decreased toxicity to rainbow trout and channel catfish.

The toxicity of Noxfish was not influenced by differences in pH within the range of 6.5 to 9.5 (Tables 2, 3, and 4). Noxfish appeared to be more toxic to rainbow trout at pH 9.5 than at lower pH's, but the increased sensitivity might have been due to an inability of the trout to acclimate fully to the high pH. Bluegills responded uniformly at the three different pH's in soft water at 12 C; the 96-h LC ₅₀'s ranged only from 122 to 138 μ g/l.

Toxicity of Noxfish to Green Eggs of Rainbow Trout

Newly fertilized eggs of rainbow trout were 47 to 106 times more resistant than fingerlings to Noxfish; the 96-h LC₅₀ ranged from 5.60 mg/l in very soft water to 2.50 mg/l in very hard water (Table 5).

Species		₀ and 95% confidence	interval (μ g/l) at	·
	3 h	6 h	24 h	96 h
Bowfin	141	106	57.5	30.0
Amia calva	114-174	82.5-136	50.4-65.5	23.7–38.0
Coho salmon	358	152	71.6	62.0
Oncorhynchus kisutch		105–219	63.1–81.3	54.8-70.2
Chinook salmon	212	156	49.0	36.9
O. tshawytscha	171-262	137–177	44.3-54.2	33.9–40.2
Rainbow trout	175	86.9	68.9	46.0
Salmo gairdneri	160–191		56.2-84.4	32.6–64.9
Atlantic salmon	61.5	40.0	35.0	21.5
S. salar	53.4-70.8	33.6-70.8	29.7-41.2	15.5–29.8
Brook trout	141	79.7	47.0	44.3
Salvelinus fontinalis	124-160	69.2–91.8	42.2–52.3	41.1–47.7
Lake trout	50.0	28.3	26.9	26.9
S. namaycush	38.6-64.7	21.0-38.0	19.8-36.5	19.8–36.5
Northern pike	181	58.2	44.9	33.0
Esox lucius	160-204	52.5-64.5	31.4 - 64.3	26.6–41.0
Goldfish Carassius auratus		_	_	497 412–600
Carp	_	270	84.0	50.0
Cyprinus carpio		254–287	74.7-94.4	41.1 -6 0.8
Fathead minnow	_	1,190	400	142
Pimephales promelas		917-1,453	291–549	115–1 76
Longnose sucker	388	218	67.2	57.0
Catostomus catostomus	332-454	141–337	59.3-76.1	51 .9-6 2.6
White sucker	630	238	71.9	68.0
C. commersoni	452–878	186-304	64.0-80.8	54.0-85.6
Black bullhead Ictalurus melas	_	_	665 516-856	389 298–507
Channel catfish	1,410	840	400	164
I. punctatus	1,139–1,745	717–984	234-684	138-196
Green sunfish	389	332	218	141
Lepomis cyanellus	332-456	249-443	197–241	114–174
Bluegill	424	336	149	141
L. macrochirus	335-537	245-461	124–178	13 3-149
Smallmouth bass	277	165	93.2	79.0
Micropterus dolomieui	219-350	—	85.1-102	70.7-88.2
Largemouth bass	514	360	200	142
M. salmoides	449–588	305-425	131–305	115 - 176
Yellow perch	150	134	92.0	70.0
Perca flavescens	126-179	120-149	80.1-106	59.8–82.0
Walleye	135	52.4	16.5	
Stizostedion vitreum vitreum	103 - 176	46.8–58.7	15.2–17.9	

Table 1. Toxicity of Noxfish® to fish in standardized laboratory tests at 12 C.

Temp	Water	Water		LC $_{50}$ and 95% confidence interval ($\mu g/l$) at			
(°C)	hardness	pH	3 h	6 h	24 h	96 h	
7	Soft	7.5	>400	276 237-322	158 134-186	70.0 62.5-78.4	
12	Soft	7.5	175 160–191	86.9	68.9 56.2-84.4	46.0 32.6–64.9	
17	Soft	7.5	73.0 59.8–89.1	73.0 59.8-89.1	43.4 30.9–60.9	43.4 30.9-60.9	
12	Very soft	8.0	122 108–138	61.9 54.9-70.2	54.4 45.9–64.4	54.4 45.9-64.4	
12	Soft	8.0	90.0 81.1-99.9	62.0 51.6-74.5	56.5 48.2–66.3	56.5 48.2-66.3	
12	Hard	8.0	112 95.1–132	81.9 64.5–104	55.1 43.9-69.	55.1 43.9-69.1	
12	Very hard	8.0	113 92.7–138	66.9 55.0-81.4	53.0 44.3-63.4	53.0 44.3-63.4	
12	Soft	6.5	169 151–189	129 107–155	78.5 69.6-88.6	69.5 63.7-75.9	
12	Soft	8.5	133 108–163	98.0 87.4–110	80.0 70.3-91.0	62.1 51.7-74.5	
12	Soft ,	9.5	124 106–145	75.0 63.2-89.0	54.0 45.9–63.6	35.5 28.7–43.9	

Table 2. Toxicity of Noxfish® to rainbow trout in water of different temperatures, hardnesses, and pH's.

Temp	Water		LC $_{50}$ and 95% confidence interval (μ g/l) at				
(°C)	hardness	pH	3 h	6 h	24 h	96 h	
12	Soft	7.5	1,720 1,381-2,141	1,000 784–1,276	539 377-770	200 164-244	
17	Soft	7.5	1,410 1,139-1,745	840 717-984	400 234-684	164 138–196	
22	Soft	7.5	739 672-813	449 352–572	164 137-196	164 137–196	
12	Very soft	8.0	1,420 1,080-1,868	640 464-883	476 346-655	328 290–370	
12	Soft	8.0	1,640 1,341-2,005	890 677-1,171	450 341-593	237 199-282	
12	Hard	8.0	1,220 1,023-1,454	942 756-1,173	400 312-513	308 237–400	
12	Very hard	8.0	1,160 996-1,351	1,000 782-1,279	359 282-457	$\begin{array}{c} 318\\ 240\text{-}421\end{array}$	
12	Soft	6.5	1,530 1,175-1,991	865 692–1081	500 326-767	200 158-254	
12	Soft	8.5	899 689-1,173	735 608-889	565 454-704	309 237–403	
12	Soft	9.5	629 548-721	625 546-715	550 444–681	248 184-335	

 $Table \ 3. \quad Toxicity \ of \ Nox fish \ {\ e} \ to \ channel \ catfish \ at \ selected \ temperatures, \ hardnesses, \ and \ pH's.$

Temp	Water		LC_{50} and 95% confidence interval ($\mu g/l$) at					
(°C)	hardness	pH	3 h	6 h	24 h	96 h		
12	Soft	7.5	450 353–573	270 217-335	141 114–174	141 114 - 174		
17	Soft	7.5	424 335–537	336 245-461	149 124–178	141 133–149		
22	Soft	7.5	268 240-300	227 194–266	140 107-183	132 122–143		
12	Very soft	8.0	334 194–575	219 183–262	142 129–157	132 118-147		
12	Soft	8.0	450 317–639	$\begin{array}{c} 319\\241\text{-}422\end{array}$	152 135–172	137 123–153		
12	Hard	8.0	300 196–460	284 200–403	146 131-162	132 118-147		
12	Very hard	8.0	295 211-413	194 148-255	138 125–152	$132 \\ 121-144$		
12	Soft	6.5	291 207–409	228 194–267	150 124–181	138 110-173		
12	Soft	8.5	255 204–319	192 170-217	122 108–138	122 108-128		
12	Soft	9.5	196 162–237	152 134–173	122 108–138	122 108–138		

Table 4. Toxicity of Noxfish® to bluegills at selected temperatures, hardnesses, and pH's.

Although the difference in toxicity was not significant at each hardness increment, the difference was significant in very soft as compared to hard or very hard water.

Persistence of Noxfish in Water

The toxicity to bluegills of Noxfish solutions aged for 1, 2, and 3 weeks decreased through each week of aging. At 12 C the 96-h LC₅₀'s were 133 μ g/l in freshly prepared solutions and 254 μ g/l in solutions aged for 3 weeks (Table 6). The toxicity decreased

Table 5. Toxicity of Noxfish® to newly fertilizedeggs of rainbow trout in reconstituted water ofdifferent hardnesses at 12 C.

Table 6.	Toxicity (96-h LC_{50} 's and 95% confidence
	in $\mu g/l$ to bluegills of fresh and aged
solutions	of Noxfish [®] in soft water (deactivation
	indices shown in parentheses).

Water hardness	96-h LC $_{50}$ (mg/l) and 96% confidence interval
Very soft	5.60
	3.55-8.83
Soft	4.42
	3.28-5.96
Hard	3.20
	2.31-4.43
Very hard	2.50
	2.16-2.90

Temp		Half-life			
(°C)	0	1	2	3	(days)
12	133 117-151 (1.00)		213 193-236 (1.60)	254 215–300 (1.91)	22
17	90.0 64.1-126 (1.00)	117 92.3–148 (1.30)	190 159-228 (2.11)	288 247-336 (3.20)	13

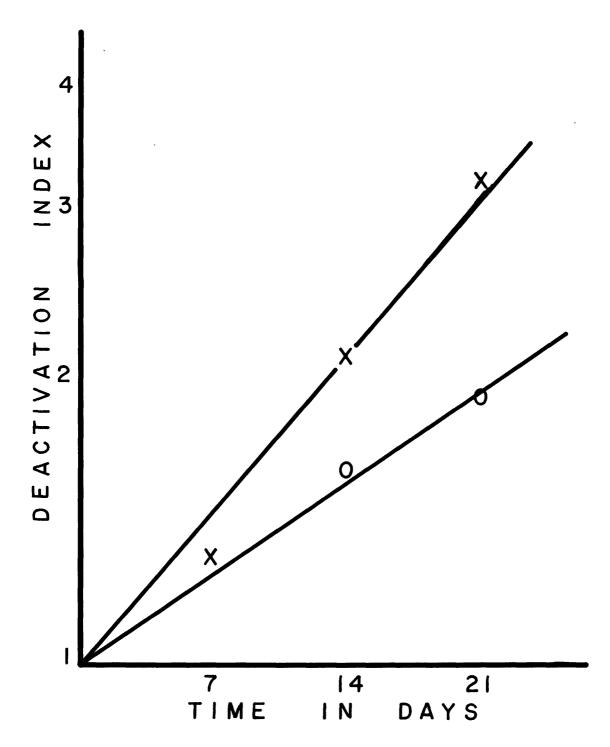


Fig. 1. Detoxification rate for Noxfish® in soft water at 12 C [O] and 17 C [X].

more rapidly at 17 C than at 12 C; the half-lives were 13 and 22 days, respectively (Fig. 1).

Detoxification of Noxfish

Static tests with green sunfish showed that potassium permanganate immediately detoxified Noxfish, as indicated by the marked loss of activity in unaged solutions of Noxfish and KMnO₄ (Table 7). For instance, the 96-h LC₅₀ at pH 7.5 was 0.241 mg/l for Noxfish alone and 1.71 mg/l for Noxfish plus 1.0 mg/l of KMnO₄. The solutions of Noxfish and KMnO₄ that were aged for 50 min before the fish were added had lost some additional activity (96-h LC₅₀, 3.09). The immediate detoxification in unaged solutions was probably a reflection of the effective exposure time, i.e., the time required for Noxfish to produce a lethal effect. The KMnO₄ detoxified Noxfish in water at all pH's tested.

Chlorine was far less effective than KMnO₄ for detoxifying Noxfish in laboratory tests. There was little immediate detoxification in unaged solutions and little detoxification during 6 h of aging. For instance, at pH 7.5 the 96-h LC $_{50}$ was 0.293 mg/l for Noxfish alone and 0.429 mg/l for Noxfish plus

0.5 mg/l of chlorine (Table 8). In a 6-h aging period, Noxfish was detoxified more efficiently at pH 9.5 than at pH's 7.5 and 8.5.

Toxicity of Different Formulations of Rotenone

Pro-Noxfish, a synergized formulation containing 2.5% rotenone, was more toxic to rainbow trout than the Noxfish formulation or powdered rotenone when concentrations were calculated on the basis of rotenone content. The comparative 96-h LC $_{50}$'s (μ g/l) were as follows: Pro-Noxfish, 1.02; Noxfish, 3.05; and powdered rotenone, 4.20 (Table 9). There was no significant difference in toxicity between Noxfish and rotenone powder formulations.

Toxicity in Flow-through Tests

In 4-day flow-through exposures, Noxfish was more toxic to chinook salmon and yellow perch than to carp or white suckers (Table 10). Toxicity did not increase with exposure time after 4 days, except for carp (in which toxicity increased through 20 days). Noxfish

Table 7. Toxicity and detoxification of Noxfish[®] in static tests with green sunfish in water containing 1.0 mg/l of $KMnO_4$ at 12 C.

Compound and (for compounds combined)	96-h LC $_{50}$ and 95% confidence interval (mg/l) at					
interaction time (min) ^a	pH 6.5	pH 7.5	pH 8.5	pH 9.5		
KMnO₄	3.47	3.03	1.41	3.08		
	3.12–3.87	2.69-3.41	1.14–1.74	2.32-4.08		
Noxfish	$0.184 \\ 0.161 - 0.211$	0.241 0.203-0.287	0.158 0.114-0.219	0.378 0.317-0.451		
Noxfish + KMnO4						
0	1.32	1.71	2.10	1.55		
	1.04-1.68	1.49-1.96	1.91-2.31	1.16-2.08		
10	1.17	1.41	1.81	1.36		
	0.948-1.44	1.14-1.74	1.54-2.13	1.07-1.70		
20	1.41	2.89	2.38	1.64		
	1.14-1.74	2.28–3.36	2.24–2.53	1.38-1.94		
30	1.82	2.89	3.10	1.91		
	1.54-2.15	2.28–3.66	2.69–3.58	1.59-2.29		
40	2.00	2.28	3.09	1.41		
	1.64-2.44	1.82-2.85	2.88-3.32	1.14–1.74		
50	1.93	3.09	3.59	1.81		
	1.61–2.31	2.74-3.49	3.19–4.03	1.53 - 2.13		

^aLength of time Noxfish and KMnO₄ were added before fish were introduced.

Compound and (for compounds combined)	96-h LC $_{50}$ and 95% confidence interval (mg/l) at				
interaction time (h) ^a	pH 7.5	pH 8.5	pH 9.5		
Chlorine	0.840	0.820	0.709		
	0.703-1.00	0.588-1.14	0.567-0.887		
Noxfish	0.293	0.338	0.329		
	0.264-0.325	0.304–0.376	0.294-0.368		
Noxfish + chlorine					
0	0.429	0.300	0.488		
	0.359-0.512	0.242-0.372	0.426-0.559		
0.5	0.348 0.299-0.405	0.492 0.427-0.567			
1.0	0.483	0.380	0.900		
	0.406-0.575	0.318-0.455	0.677-1.20		
2.0	0.412	0.400	0.689		
	0.337-0.504	0.339-0.473	0.584-0.813		
4.0	0.494	0.489	0.770		
	0.438-0.557	0.432-0.554	0.689–0.862		
6.0	0.494	0.454	1.37		
	0.438-0.557	0.390-0.529	0.988–1.90		

Table 8.	Toxicity and detoxification of Noxfish [®] in static tests with green sunfish in water containing
	0.5 mg/l of chlorine at 12 C.

^aLength of time Noxfish and chlorine were added before fish were introduced.

Table 9.	Toxicity of three	formulations of	^f rotenone to	rainbow tro	ut in sof	ft water at 12 C.
i ubic b.	TOWNERS OF UNICE	1011111111110110 01	TOTENONE TO	1411000 110	u u u u u u u	

Preparation	% active rotenone	LC $_{50}$ and 95% confidence interval ($\mu g/l$) at				
		1 h	3 h	6 h	24 h	96 h
Pro-Noxfish®	2.5	13.0 8.15–20.7	4.53 3.28–5.65	2.98 2.43-3.65	1.82 1.60-2.08	1.02 0.917–1.15
Noxfish®	5.0	25.5 16.1-40.5	8.70 6.90–11.0	5.50 4.87-6.20	3.25 2.78–3.81	3.05 2.85-3.27
Powdered rotenone	33.0	16.5 14.3–19.1	8.09 6.37–10.3	6.60 5.41-8.02	3.82 3.30-4.46	3.20 2.09–3.70

Species	LC $_{50}$ and 95% confidence interval (μ g/l) at						
	1 day	4 days	10 days	20 days	30 days		
Chinook	112	71.0	62.0	59.0	_		
salmon	97.7-128	55.3-99.1	52.1-73.7	49.5-70.3			
Carp	-	142 122-165	96.0 78.0–118	67.0 57.4–78.3	68.0 57.7-80.1		
White	_	144	129	112	112		
sucker		122–170	118-141	95.5-131	95.5–131		
Yellow	160	60.0	50.0	46.0			
perch	121-211	53.3–67.6	36.8-68.0	32.7-64.8			

Table 10. Toxicity of Noxfish® to four species of fish in flow-through toxicity tests at 12 C.

was consistently and significantly more toxic to all four species in static than in flow-through tests (Table 11).

Discussion

The literature on toxicity of rotenone to fish suggests that concentrations used in fishery management are generally higher than those known to be lethal in laboratory tests; that toxicity depends on temperature, water hardness, pH, and physical characteristics; and that many different application rates may be effective for the same target species of fish (Schnick 1974; Meyer 1966).

Since laboratory procedures are usually more standardized than field procedures, laboratory data are expected to be more consistent than field data. Although the LC_{50} 's of less than 0.2 mg/l for

Table 11. Comparison of acute toxicities of Noxfish® to four species of fish in 96-h flow-through and static tests in carbon filtered municipal water at 12 C.

Species -	LC_{50} and 95% confidence interval (μ g				
	Static	Flow-through			
Chinook salmon	34.7	71.0			
	26.9 - 44.7	55.3-99.1			
Carp	19.0	142			
	12.1 - 29.9	122-165			
White sucker	17.9	144			
	12.8 - 25.1	122 - 170			
Yellow perch	30.0	60.0			
-	23.6-38.2	53.3-67.6			

Noxfish against rainbow trout, channel catfish, and bluegills reported by Bridges and Cope (1965) were similar to ours, applications of at least 1 mg/l have been repeatedly recommended for eliminating these species. Spitler (1970) reported that 1.6 mg/l of Noxfish was not effective and that as much as 5 mg/lwas sometimes needed. The difference in laboratory and field data is due to several factors. Laboratory data generally indicate concentrations that produce 50% mortality (LC $_{50}$), whereas field concentrations are based on eliminating 100% of the target fish. Organisms, particulate matter, and sunlight contribute to the tendency toward faster detoxification of chemicals in natural waters than in the laboratory. Furthermore, because uniform concentrations are much more difficult to obtain in the field. additional amounts of toxicants are generally applied to ensure a lethal concentration throughout a body of water.

Although some of the reports are conflicting, rotenone is generally more effective at high than at low temperatures (Gersdorff 1943; Almquist 1959; Ball 1948; Hooper 1955), in acid than in alkaline waters (Leonard 1939; Foye 1964), and in soft than in hard water (Foye 1964). In many of these studies, however, efficacy was based on survival time of the fish rather than on the concentration of the toxicant. Our laboratory data show only slight changes in the toxicity of rotenone at different temperatures, hardnesses, or pH's. Consequently, concentrations used in the field should be based on the results of onsite toxicity tests (Burress 1975) rather than on extrapolations of laboratory or field data.

Most studies—in laboratory or field—have shown that goldfish and black bullheads are the species most resistant to rotenone. Individual fish of a species may be exceptionally resistant (Meyer 1966) an observation that may explain some incomplete fish kills and the need to apply a concentration greater than that indicated in laboratory tests.

The detoxifiers KMnO₄ and chlorine were toxic to fish at concentrations only slightly greater than those needed to detoxify rotenone. For example, against green sunfish in water at pH 8.5, the 96-h LC_{50} for KMnO₄ was 1.41 mg/l and that for chlorine was 0.82 mg/l. These results support Engstrom-Heg and Loeb (1968) and Engstrom-Heg (1972), who cautioned that high concentrations of KMnO₄ may become toxic and may have to be reduced with sodium thiosulfate or other agents. Therefore, detoxifiers should be used only when necessary and in only the quantities needed.

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