

ROTENONE TOLERANCES OF STREAM-BOTTOM INSECTS¹

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ABSTRACT

Stream-bottom insects varied greatly in their tolerance to rotenone. Tolerances did not adhere closely to taxonomic lines, but showed a possible relationship to oxygen requirements. The more sensitive genera tended to have short life cycles, high mobility and a potential for rapid repopulation. In many streams an exposure of 5 per cent emulsifiable rotenone formulation would eliminate undesired fish with only mild and temporary damage to the bottom fauna. Very few immature aquatic insects could survive a 48-hour exposure to 3 ppm of 5 per cent formulation.

Responsible resource management requires that any chemical treatment of natural waters be conducted in a way that will accomplish the desired ends with minimal damage to non-target biota. In the case of pesticide treatment of streams, there is an additional reason for minimizing damage to aquatic insect populations. It is highly probable that maximum benefits from such treatments could be obtained if they were made in the late summer and followed almost immediately by the stocking of fall fingerling game fish (trout or smallmouth bass). Benthic food production would then be funneled into the rapid growth of a single year class in a competition-free environment. This would, of course, not be practical if the fish toxicant had also eliminated most of the benthic insect fauna, even though repopulation might occur the next year. Binns (1967) and Swan (1965) found that prolonged treatment of streams with high rotenone concentrations effectively eliminated most aquatic insects and that, while some species became reestablished within a month or two, full restoration of the original fauna might require as long as 2 years.

In August and September of 1970, a segment of the Ten Mile River, a tributary of the Delaware River in Sullivan County (N.Y.), received three successive treatments with 5 per cent emulsifiable rotenone at concentrations of 1 ppm, 3.71 ppm and 4.75 ppm (6.0, 8.7 and 11.7 ppm-hours). A portion of the stream received only the 1 ppm treatment. On September 28, a series of nine bottom samples were obtained with a Surber square-foot bottom sampler at stations in parts of the stream that

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TABLE 1. SUMMARY OF AQUATIC INSECTS AND OTHER INVERTEBRATES IN BOTTOM SAMPLES TAKEN FROM THE TEN MILE RIVER, ACCORDING TO DEGREE OF ROTENONE EXPOSURE, SEPTEMBER 28, 1970*

Insect or other invertebrate	Rotenone exposure		
	None	1.0 ppm	1.0, 3.71 and 4.75 ppm
Ephemeroptera			
Baetidae§	5.0	13.7	4.0
Other families	0.3	0.0	0.0
Trichoptera			
Hydropsychidae	19.3	39.0	19.2
Other families	2.3	4.7	3.0
Diptera			
Chironomidae	9.7	6.3	7.0
Other families	0.0	0.3	0.0
Other insects	0.7	1.3	1.7
Total insects	37.3	64.4	34.9
Gastropoda	14.6	44.5	55.0
Oligochaeta	5.3	13.5	12.2
Total	57.2	122.6	67.2

* Values in animals per square foot (mean of three samples).

§ Predominantly *Paraleptophlebia*.

had had no treatment, only the 1 ppm treatment, and all three treatments. Analysis (Table 1) showed no significant differences between the three parts. The bottom fauna, dominated by hydropsychid caddisflies of the genera *Hydropsyche* and *Cheumatopsyche* and the mayfly *Paraleptophlebia* sp., had apparently not been affected to any appreciable extent. The total rotenone dosages applied to the Ten Mile River were considerably less than those reported by Binns (1967) for the Green River in Wyoming (65 ppm-hours to several times that dosage at downstream stations) and by Swan (1965) for Fall Creek in Oregon (over 48 ppm-hours of 2.5 per cent synergized formulation), suggesting that it might be possible to find a level of exposure that would kill undesired fish without seriously affecting the bottom fauna of most streams. To test this hypothesis, a series of experiments were conducted at the DeBruce laboratory in 1971, 1972 and 1973.

METHODS

Since the object of the experiments was to find tolerable levels of rotenone exposure for a wide variety of aquatic insects in the context of limited time and manpower, the methods used departed to a large extent from the standard canons of bioassay procedure as described by Doudoroff et al. (1951). Rather than determine exact LD₅₀'s for a few taxa,

a decision was made to study as wide a range of insects as was feasible and to estimate the exposure levels at which rotenone caused significant or total mortality.

In early tests, each experiment began with a series of similar bottom samples from a stream, usually Mongaup Creek, near the laboratory. Each sample was obtained by collecting the bottom fauna from 3 to 5 square feet of substrate with a Surber sample and a bottle brush. Samples were placed in 5-gallon glass jars partly filled with the stream water and taken to the laboratory where springwater was added to adjust the volume to 10 liters. The jars were then floated in a bath adjusted to 65° F. and aerated with compressed air via an air stone. A few small rocks were placed in the bottom of each jar for cover. Jars were held 24 hours before treatment. Treatment consisted of adding 5 per cent emulsifiable rotenone at 1 ppm or 3 ppm. At the end of 1, 3, 6, 16, 24 or 48 hours, 4 ppm of potassium permanganate (KMnO₄) were added to each jar to detoxify the rotenone. After 20 minutes the potassium permanganate was reduced with 6 ppm of sodium thiosulfate (Na₂S₂O₂), as would occur in a typical stream treatment (Engstrom-Heg, 1972). Jars were held an additional 24 hours and then removed from the bath. Most of the water was poured off through a Surber sampler, and the remainder, containing most of the insects, was poured out onto a large white enamel tray. The insects were removed from the tray and sampler and placed in one of two jars of 70 per cent alcohol for live and dead specimens, respectively. Each experiment included at least one control jar that was treated with potassium permanganate and sodium thiosulfate, but not with rotenone. At a later date insects from each jar were counted and identified, usually to genus, according to Pennak (1953) and Usinger (1956).

Later experiments were conducted in an identical fashion, except that individuals from a particular genus or group of genera, for which earlier tests had yielded insufficient information, were specifically collected and tested. Animals were collected from several Sullivan and Delaware County streams.

RESULTS

Typically, the control insects suffered an approximately 20 per cent mortality. This was to be expected in view of the stresses of collection and the fairly prolonged holding in an essentially abnormal situation with increased population density, reduced cover and no current. Mortality in the experimental insects began to depart significantly (chi-square at 5 per cent level with 1 d.f.) from that of the control insects at a wide range of exposures, varying from 1 ppm-hours (for *Baetis*) to over 144 ppm-hours (for *Cheumatopsyche*). Findings on individual taxa are listed in Table 2.

TABLE 2. SUMMARY OF RESULTS OF ROTENONE TOLERANCE TESTS ON AQUATIC INSECTS AND RELATED INFORMATION

Order	Family and genus	Number of animals		Exposure levels (in ppm-hours) according to degree of mortality			Survival (per cent)		
		Test	Control	Not significant	Significant*	Apparently total	Control	Non-significant level	Significant level*
Ephemeroptera	Baetidae	46	52	None	1, 3	6	80.8	..	4.9
	<i>Baetis</i> †	137	125	3, 6	9, 16, 48, 72	144†	84.8	88.2	47.2
	<i>Ephemerella</i> †	37	27	3, 6, 9, 16, 24	48, 72	144†	85.1	86.7	9.5
	<i>Paraleptophlebia</i> †								
	Heptageniidae	25	18	3, 6, 9	None	16†	94.4	68.0	..
	<i>Stenonema</i>	10	29	3, 6	None	16†	82.7	60.0	..
Other (mixed)	35	47	3, 6, 9	None	16†	86.6	65.7	..	
Total†									
Plecoptera	Pteronarcidae					None	92.9	89.9	73.9
	<i>Pteronarcys</i> †	101	42	3, 6, 16, 24, 48	72				
	Perlidae					16	76.7	..	38.9
	<i>Neophasganophora</i> †	36	30	None	6, 9				
Perlodidae					6, 9, 24	57.1	100.0	23.0	
<i>Isogenus</i> †	53	35	1	3					
Chloroperlidae					48	76.5	58.3	20.0	
Chloroperlinae (mixed)†	97	17	3, 6	9, 16					
Odonata	Aeschnidae					None	100.0	100.0	..
	<i>Aeschnia</i>	10	10	3	None				
Megaloptera	Corydalidae					None	96.7	100.0	..
	<i>Chauliodes</i> †	46	30	3, 10, 16, 48	None				
Tricoptera	Rhyacophilidae					16	61.5	73.3	14.3
	<i>Rhyacophila</i> †	56	39	3, 6	9	72	58.1	55.6	..
	<i>Glossosoma</i> †	57	43	3, 6, 9, 16, 24, 48	None				

	Philopotomidae	9	10	3, 9	None	None	50.0	55.6	..
	Psychomyiidae								
	<i>Psychomyia</i>	14	19	None	None	6†, 9†, 16, 58, 72	68.4
	Hydropsychidae								
	<i>Hydropsyche</i> †	116	79	3, 9, 48	6, 16, 24, 72, 144	None	69.6	69.2	28.9
	<i>Cheumatopsyche</i> †	19	13	3, 6, 9, 16, 24, 48, 72, 144	None	None	100.0	89.5	..
	Limnephilidae								
	<i>Limnephilus</i> †	80	19	3, 6, 9, 16	24	None	100.0	90.0	55.0
	<i>Pycnopsyche</i> †	50	30	3, 6	9, 16	24	100.0	100.0	70.0
	Brachycentridae								
	<i>Brachycentrus</i> †	213	222	None	1, 3, 6, 9	16, 24, 48, 72, 144	83.3	..	9.0
	Odontoceridae								
	<i>Psilotreta</i>	13	13	9, 24	None	None	92.3	69.2	..
Coleoptera	Elmidae								
	<i>Limnius</i>	6	2	9, 16, 48	None	None	100.0	83.4	..
Diptera	Tipulidae								
	<i>Eriocera</i> †	62	25	3, 6, 9, 16, 24	48	72	88.0	72.9	10.0
	<i>Antocha</i>	17	3	3, 6	None	9†, 16, 24	100.0	50.0	..
	Simuliidae (mixed)†	449	92	1	3	6, 24	83.7	77.5	73.2
	Chironomidae								
	<i>Pentaneura</i> †	63	40	3, 6, 9, 16	24	None	92.3	75.5	42.9
Other (mixed)	123	10	3	6, 9, 16	None	70.0	100.0	53.7	
Rhagionidae									
<i>Atherix</i>	6	5	3, 16	None	48†	80.0	33.3	..	

TABLE 2. (Continued)

Order	Family and genus	Estimated rotenone tolerance	Life cycle	Mobility	Time to reestablish§
Ephemeroptera	Baetidae <i>Baetis</i> † <i>Ephemerella</i> † <i>Paraleptophlebia</i> †	Variable Very low Intermediate to high†† High	Variable Short Intermediate Intermediate	High High High	Short Intermediate
	Heptageniidae <i>Stenonema</i> Other (mixed) Total†	Intermediate to high Intermediate to high Intermediate to high Intermediate to high		Intermediate Intermediate Intermediate	ND ND
	Plecoptera				
Plecoptera	Pteronarcidae <i>Pteronarcys</i> †	Very high	Long	High	Intermediate
	Perlidae <i>Neophasganophora</i> †	Low		High	Intermediate
	Perlodidae <i>Isogenus</i> †	Low		High	Intermediate
Odonata	Chloroperlidae Chloroperlinae (mixed)†	Intermediate	Intermediate	High	ND
	Aeschnidae <i>Aeschnia</i>	May be high	Long	Intermediate	ND
	Megaloptera				
Megaloptera	Corydalidae <i>Chauliodes</i> †	High	Long	High	ND
Tricoptera	Rhyacophilidae <i>Rhyacophila</i> † <i>Glossosoma</i> †	Variable Low High	Intermediate†† Intermediate†† Intermediate††	Low Intermediate Low	Long (?) ND Long
	Philopotomidae	Intermediate	Intermediate††	Low	ND
	Psychomyiidae <i>Psychomyia</i>	Low	Intermediate††	Low	ND

Hydropsychidae <i>Hydropsyche</i> † <i>Cheumatopsyche</i> †	Very high	Intermediate††	Low	Long	
	High	Intermediate††	Low	Long	
	Very high	Intermediate††	Low	Long	
Limnephilidae <i>Limnephilus</i> † <i>Pycnopsyche</i> †	Intermediate to high	Intermediate††	Intermediate	ND	
	High	Intermediate††	Intermediate	ND	
	Intermediate				
Brachycentridae <i>Brachycentrus</i> †	Very low	Intermediate††	Intermediate	Intermediate	
Odontoceridae <i>Psilotreta</i>	High	Intermediate††	Intermediate	ND	
Coleoptera	Elmidae <i>Limnius</i>	High	Long (?)	High	ND
Diptera	Tipulidae <i>Eriocera</i> † <i>Antocha</i>	Variable High Low to intermediate	(?) (?)	Low Low	Long Long Long
	Simuliidae (mixed)†	Low	Short	Intermediate	Short
	Chironomidae <i>Pentaneura</i> † Other (mixed)	Intermediate Intermediate	Short Short	Intermediate Intermediate	Short Short
	Rhagionidae <i>Atherix</i>	Intermediate	Short (?)	Low	Short

* Significantly different from mortality of controls, based on chi-square (adjusted values) at 5 per cent level of confidence with 1 degree of freedom.

§ After Binns (1967); ND indicates he gave no data on time to reestablish.

† Included in Figure 1.

‡ Based on total mortality to small, statistically insignificant number of insects. These taxa may be more tolerant than indicated.

†† The genus *Ephemerella* includes a number of species with different ecological adaptations and, quite possibly, with a considerable range of rotenone tolerances. At least three species were included in the tests.

‡‡ Life cycles of caddisflies tend to vary greatly with food and other habitat conditions. Many species ordinarily produce one or two broods per year.

Use of ppm-hours as a unit of exposure is based on the approximately hyperbolic character of most concentration-response curves. The product of toxicant concentration and response time is often nearly constant through a fairly wide range of values. It should be recognized, however, that this relationship is merely approximate and that it may be invalidated by threshold effects at very high or low concentrations (Burdick, 1957). The data indicate that its use was justified for this set of experiments, in that mortality generally increased with increased exposure in ppm-hours, whether the toxicant was added to 1 ppm or 3 ppm.

The estimated tolerances should probably be regarded as minimal, in that the experiments were run on stressed animals uprooted from microhabitats that in many cases may be quite exacting. It is likely that most of the insects would have tolerated somewhat greater exposures in their natural stream setting.

Further analysis of the data (Figure 1) indicates a rather sharp increase in the number of affected genera at approximately the 10-ppm-hour level. Interestingly, the three dominant genera in the Ten Mile River fauna turned out to be highly resistant. It can be seen that the insects of the orders Ephemeroptera, Plecoptera and Diptera are highly variable in their tolerance to rotenone and that strong differences within a single family are not unusual. Limited information, including fragmentary data not listed in Table 2 as well as scattered references in the literature (Claffey and Ruck, 1967; Zischkale, 1952; Leonard, 1939), suggests that members of the orders Odonata, Megaloptera and Coleoptera and burrowing mayflies of the family Ephemeridae may all be quite resistant. Binns (1967) found that adult air-breathing aquatic insects successfully withstood prolonged rotenone exposures that killed most immature forms. Observations on the few adult coleopterans and hemipterans in the present samples tend to confirm this finding. Observations on non-insect benthos indicate that turbellarian worms are very sensitive to rotenone and that the aquatic isopod *Asellus militaris* ranks with the more resistant insects.

DISCUSSION

In fish, rotenone tolerance tends to vary inversely with oxygen requirements, as would be expected for a respiratory poison. A similar pattern may hold for immature aquatic insects. Roback (1962) found that the rotenone-resistant caddisfly genera *Hydropsyche* and *Cheumatopsyche*, while occurring in a wide range of habitats, were tolerant of polluted waters with high BOD levels. The work of Degan (1973), as reviewed by Schnick (1974), suggests that antimycin, which has a mode of action similar to that of rotenone, may have similar effects on aquatic insects. Populations of *Baetis*, *Brachycentrus* and *Antocha*, all rotenone-sensitive genera, were eliminated or drastically reduced after treatment of a stream

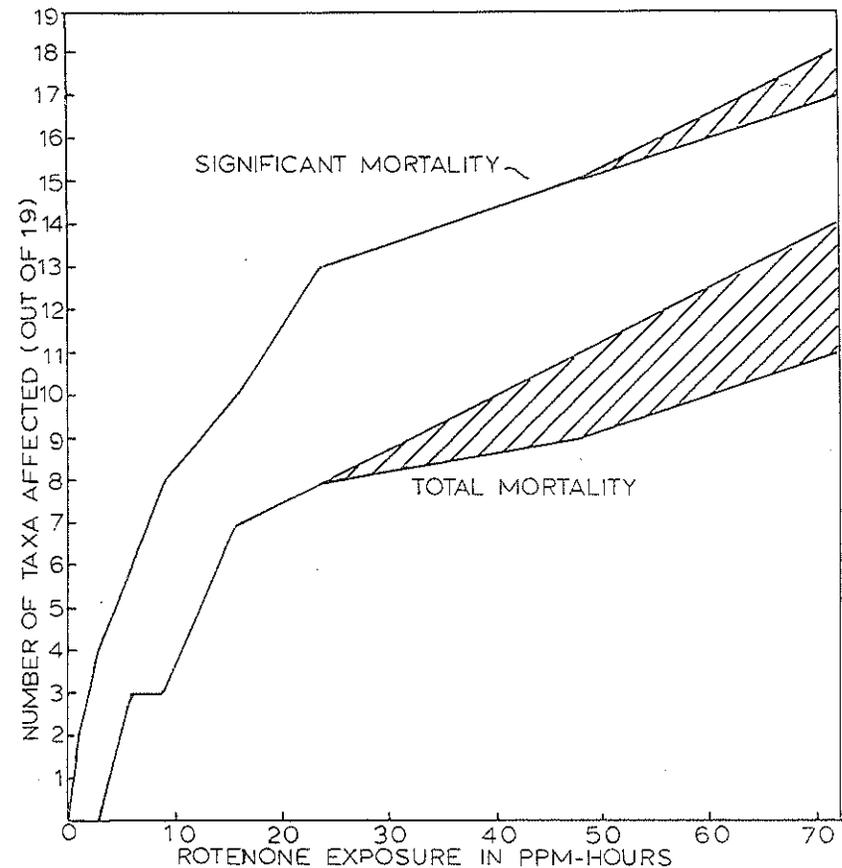


Figure 1. Number of insect taxa suffering significant (chi-square at 5 per cent level with 1 d.f.) or total mortality at various rotenone exposures. Shaded areas represent range between maximum and minimum estimates resulting from missing or inadequate tests at high concentrations. Only those taxa (19 as noted in Table 2) for which 30 or more insects were tested are included.

with 10 to 40 ppb of the chemical. The abundance of *Hydropsyche* was less severely depressed, and the elmid beetle *Stenelmis* was apparently not affected.

Rotenone treatment of a stream to eliminate rough fish generally requires application of at least 6 ppm-hours of 5 per cent rotenone formulation, even for relatively short segments (Engstrom-Heg and Loeb, 1971). Lennon and Parker (1959) found that a dose of 10⁶ ppm-hours, introduced as 5 ppm for 1 hour, followed by 1 ppm for 5 hours, effectively killed carp. Calculations based on measurement of stretch-out in pool-and-riffle streams indicate that a slightly modified version of this

pattern, in which introduction began and ended with 30-minute bolts at 5 ppm, would effectively treat a stream segment several miles long without undesirable attenuation.

The bioassay results show that there is no level of application at which rough fish can be eliminated without at least minor damage to the bottom fauna. However, it is possible to minimize damage by avoiding excessively prolonged or concentrated treatment. Fortunately, there is a strong tendency for the more sensitive insects to be highly mobile and/or to have short life cycles, and thus to repopulate depleted areas rapidly through drift, migration and oviposition (Table 3). It is probable that

TABLE 3. RELATIONSHIP OF ROTENONE TOLERANCE TO RATE OF RECOLONIZATION FOR VARIOUS AQUATIC INSECTS

Rotenone tolerance	Rate of recolonization*		
	Rapid§	Intermediate	Slow†
Low (significant at 1 or 3 ppm-hours)	<i>Baetis</i> <i>Simuliidae</i>	Perlodidae Brachycentridae	..
Intermediate (significant at 6, 9 or 16 ppm-hours)	Chironomidae Rhagionidae	<i>Ephemera</i> Heptageniidae Perlidae <i>Antocha</i>	..
High (significant at 24 or more ppm-hours)	..	<i>Paraleptophlebia</i> <i>Pteronarcys</i> Elmidae	<i>Glossosoma</i> Hydropsychidae <i>Eriocera</i> <i>Chauliodes</i>

* After Binns (1967).

§ Recolonization occurred at three or more stations within 2 months.

† Still absent at three or more stations at 1 year, or requiring 2 years at any station.

in most streams, treatment at 10 ppm-hours would result in mild and temporary damage. This, however, should not be taken as a hard-and-fast dictum. For instance, in the late summer in 1971 and 1972, the bottom fauna of Mongaup Creek was dominated by rotenone-sensitive caddisfly larvae of the genus *Brachycentrus*, which probably would have been eliminated by treatment at this level. Streams should be evaluated individually, and estimates of probable damage to the bottom fauna should be considered in planning any extensive rotenone treatments. If animals not listed in this paper should be important as fish forage, it would not be difficult in most cases to determine their approximate rotenone tolerance.

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