EFFECTS OF ROTENONE ON MACROBENTHIC INVERTEBRATES OF A PENNSYLVANIA STREAM

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Abstract: Although the use of rotenone as a fish toxicant is a common freshwater fisheries management technique, little is known of its effect on stream invertebrates. In this study pre- and post-treatment bottom samples of benthic invertebrates, collected from 4 study stations, were compared to determine the short-term effects of rotenone. Continuous drift samples collected throughout the treatment period provided additional qualitative information on the vulnerability of the benthic invertebrates to the toxin. Of the 4 major orders of macrobenthic invertebrates represented in Sinking Creek (Trichoptera, Ephemeroptera, Plecoptera, and Diptera), all exhibited substantial decreases in numerical abundance 11 days after rotenone treatment. Populations of Plecoptera (stoneflies) and Diptera (blackflies and midge larvae) were nearly exterminated, while densities of the 2 remaining taxa, Trichoptera (caddisflies) and Ephemeroptera (mayflies) were reduced to 50% of the pretreatment levels. All 4 orders appeared in increased numbers in the drift samples taken during the treatment period.

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Rotenone ($C_{23}H_{22}O_{2}$), a toxic chemical of derris root, has been used for centuries by man to obtain fish (Leonard 1939). During the past 35 years, rotenone has been widely employed as a fish toxicant to control undesirable fish populations (Kiser et al. 1963; Lennon et al. 1971; Smith 1940; Solman 1950). At present, the use of rotenone to rehabilitate waters for sportfish production is a common procedure in freshwater fisheries management.

Early published research concerning the effects of rotenone in aquatic systems focused primarily on fish mo ality (Ball 1948; Hooper 1955; Smith 1940; Surber 1936). Subsequent studies have emphasized its effects on zooplankton (Anderson 1970; Kiser et al. 1963; Neves 1975) and benthic invertebrates in lakes (Almquist 1959; Brown and Ball 1942; Cushing and Olive 1957). However, except for the work of Binns (1967) and Cook and Moore (1969), both on streams in western U.S. and laboratory bioassays by Claffey and Ruck (1967) and Engstrom-Heg et al. (1978), little attention has been directed toward the effects of rotenone on benthic invertebrates inhabiting running waters.

Despite the apparent effectiveness of rotenone as a fish toxicant and its widespread popularity as a fisheries management tool, failure to evaluate adequately its potentially detrimental effects on non-target components of lotic communities has been justifiably criticized (Hubbs 1963). Binns (1967) found that prolonged treatment of streams with high rotenone concentrations effectively eliminated most aquatic insects and suggested that complete restoration of the original fauna may require several years. Engstrom-Heg et al. (1978) concluded that there is no level of rotenone application at which rough fish can be eliminated without at least minor damage to bottom fauna.

The purpose of this study was to examine the short-term effects of rotenone on stream-dwelling macrobenthic invertebrates. The data presented here represent an attempt to determine major, short-term effects; neither time nor facilities permitted a detailed taxonomic analysis or extensive collections of stream invertebrates before and after rotenone treatment. The present investigation was conducted in conjunction with a fisheries management project designed to eliminate undesirable rough fish species from the upstream waters of a newly-constructed mainstream sportfishing reservoir.

STUDY AREAS AND METHODS

Sinking Creek is a small coolwater mountain stream located in Centre County, Pennsylvania. The stream is approximately 25.6 km in total length, of which the upper

12.8 km drains a fairly extensive forested watershed before continuing for an additional 12.8 km through open farmland. The stream originates from Bear Meadow Pond, a relic sphagnum bog, and flows in a north-easterly direction before entering Penn's Creek in the village of Spring Mills (Fig. 1). The area selected for rotenone treatment included the upper 9.7 km of stream from a point just below Bear Meadows to Colyer Dam. Stream side vegetation in the study area is characterized by a dense understory of rhododendron (*Rhododendron* sp.) and mountain laurel. Stream side vegetation in the study area is characterized by a dense understory of rhodedendron (*Rhododendron* sp.) and mountain laurel (*Kalmia latifolia*) thickets. Prior to rotenone treatment, this section of stream supported a reproducing population of brook trout (*Salvelinus fontinalis*). Green sunfish (*Lepomis cyanellus*), rock bass (*Ambloplites rupestris*) and suckers (*Catostomus* spp.), the major targets of the stream reclamation project, dominated the fish fauna of the downstream waters.

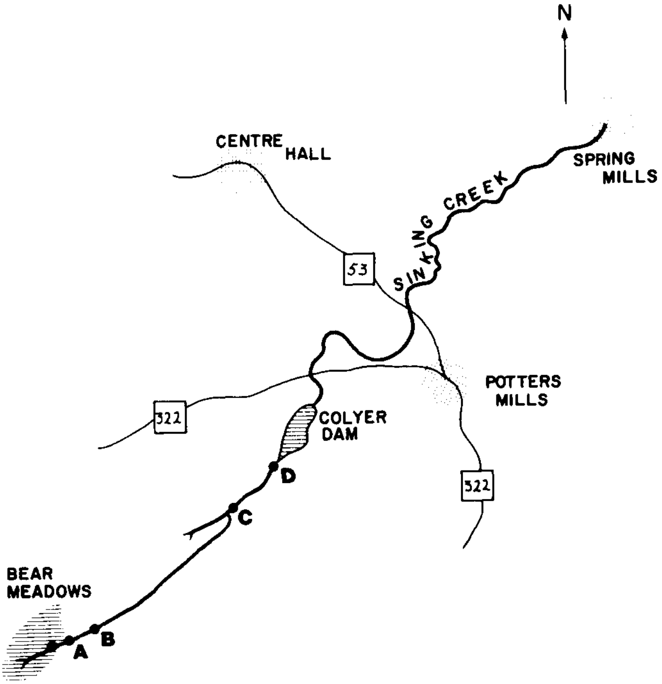

Fig. 1. A diagrammatical map of the study area indicating the locations of the four study sites.

Four study sites were selected to make pre- and post-treatment comparisons of benthic invertebrates in areas that were physically, chemically and biologically similar. An additional upstream site, initially chosen as an untreated reference area, proved too dissimilar to the downstream sites to make valid comparisons and was disregarded. All 4 study sites chosen were located in riffle areas with water velocities ranging from 15 to 25 cm/sec, depths between 15 and 35 cm, and bottom rubble sizes between 1 and 15 cm in greatest dimension. Replicate bottom samples were collected at each riffle site 1 day before and 11 days after rotenone treatment with a Surber square foot samples (Surber 1936). To supplement the benthic collections, drift samples were taken at Station D with square foot drift nets supported vertically by pipes driven into the stream bottom. To determine typical pretreatment drift rates, a 24-hour drift sample was collected just prior to rotenone treatment. For comparative purposes, a series of drift samples was collected at 15-minute intervals during the 6 hours that rotenone passed Station D. This series of samples provided a qualitative record of the successive vulnerability of benthic invertebrates to the toxin.

All macroinvertebrate samples were preserved in 45% isopropyl alcohol. Organisms were separated from detritus, identified to approximate taxonomic levels and counted under a binocular microscope at 30x. Identifications were made using Pennak (1953) and Unsinger (1968). The dominant macroinvertebrates encountered in this study are listed in Table 1.

Table 1. A list of the dominant macrobenthic invertebrates collected in Sinking Creek.

Plecoptera (stoneflies)	*Ephemeroptera (mayflies)*
Acroneuria spp.	*Ephemera simulans*
Diploperla	*Ephemerella rotunda*
Leuctra sp.	*Habrophlebia* sp.
Neophasganophora sp.	*Pseudocloeon* sp.
Pteronarcys sp.	*Stenonema* sp.
Trichoptera (caddisflies)	Diptera
Cheumatopsyche sp.	Chironomidae (midges)
Hydropsyche sp.	*Prosimulium* sp.
	(blackfly)
Micrasema sp.	*Antherix variegata*
Molanna sp.	*Tabanus* sp.
Neophylax sp.	
Nyctiophylax sp.	
Oligostomis sp.	
Polycentralis centralis	
Pycnopsyche sp.	
Rhyacophila spp.	
Trentonius distinctus	

Water chemistry, monitored at each site throughout the study period, showed a distinct downstream acid-alkaline gradient. The pH values ranged from 6.8 in the headwaters near Bear Meadows Bog (Station A) to 7.8 in the open pasture land just above Colyer Dam (Station D). Similarly, total hardness was correlated with stream gradient, ranging from 9.1 mg/ l in the headwaters to 27.0 mg/ l in the downstream waters. Water temperature and dissolved oxygen concentrations were relatively uniform between stations during the study. Diel temperature and oxygen variations were greater within stations than between stations. Dissolved oxygen levels were characteristically high (9.0 to 10.6 mg/ l), and water temperatures cool (15 to 18 C). At the time of rotenone application, dissolved oxygen concentration averaged 9.5 mg/ l, and water temperature averaged 16.7 C in the study area.

The upper 9.7 km of Sinking Creek was treated with rotenone on August 28, 1966 under the direction of the Pennsylvania Fish Commission. A total of 18.9 l of 5% rotenone, calculated to provide a concentration of at least 1 mg/ l through the study area, was applied at the major drip site located 5 m above Station A. Because of dilution and low flow a second dose of rotenone was administered approximately 1.6 km above

Station D. The arrival of rotenone at each study site was indicated by fluorecein dye which was introduced along with the toxicant. Although no test was designed to evaluate the potential effects of the dye on the macroinvertebrate community, it is probable that there was none since this substance is generally considered biologically inert.

RESULTS

The major groups of benthic invertebrates encountered in Sinking Creek during the study were representatives of the orders Diptera, Plecoptera, Trichoptera, and Ephemeroptera. On the basis of pre-treatment benthic samples, the dominant taxa of invertebrates, in order of their abundances, were Chironomidae (midges), *Prosimulium* (black fly), *Ephemerella* (mayfly), *Leuctra* (stonefly), *Hydropsyche* (caddisfly), *Stenonema* (mayfly), and *Cheumatopsyche* (caddisfly). Of the remaining orders, too few individuals were collected to evaluate treatment effects. These included species of Odonata (Zygoptera), Coleoptera (Elmidae and Psephenidae), Hemiptera (Gerridae and Corixidae), Crustacea (Amphipoda and Decapoda), Mollusca (Gastropoda and Pelecypoda), and Annelidae (Hirudinea and Oligochaeta). Collectively these groups represented less than 10% of the invertebrate fauna sampled.

The immediate effect of the treatment was a substantial increase in the numbers of drifting dead and dying invertebrates. The total number of organisms collected as stream drift during the 6 hour period that rotenone passed station D was more than 500 times greater than the pretreatment drift taken during the same hours one day before treatment (Table 2). These exceptionally high values for total stream drift were associated with increased representation by all major orders, particularly stoneflies and mayflies, which were notably absent from the pretreatment drift. This increase in total drift was attributed largely to the stoneflies *Leuctra* and *Diploperla*, the mayfly *Ephemerella*, and the caddisfly *Hydropsyche*; these 4 genera constituted 79% of the total drift during the treatment period.

Table 2. A comparison of the total numbers of invertebrates per square foot collected as stream drift over a 6-hour period (11 am to 5 pm) 1 day before (8/28) and on the day of rotenone treatment (8/29) at Station D.

	Drift Rates	
Order	*Pre-treatment*	*Treatment*
Plecoptera	0	2635
Trichoptera	8	1570
Ephemeroptera	0	1494
Diptera	3	383
Others	1	251
	12	6333

Continuous collections of stream drift sampled throughout the treatment period provided a qualitative estimate of the successive vulnerability of benthic invertebrates to the toxin. It should, however, be noted that the relationship between the arrival times of the rotenone dosages and the abundances of certain invertebrates in the drift is merely an approximate index of their susceptibility that may be invalidated by clumped distributions, relative buoyancies and other factors. The response times of the 4 dominant orders to rotenone, as reflected by drift rates, were variable (Fig. 2). Shortly after the arrival of rotenone the numbers of drifting stoneflies and caddisflies increased sharply; both orders attained peak densities within 2 hours after the arrival of the first dose. In contrast, the numbers of drifting mayflies remained low after the initial dose, but gradually increased to a maximum approximately 2 hours after the arrival of the second dose of rotenone. Of the 4 orders, the dipterans were the least abundant and exhibited only a slight peak during the first 2 hours of the treatment. The major constituents of stream drift, in order of their appearance after treatment, were *Diploperla, Hydropsyche, Leuctra, Cheumatopsyche, Prosimulium,* and *Ephemerella.*

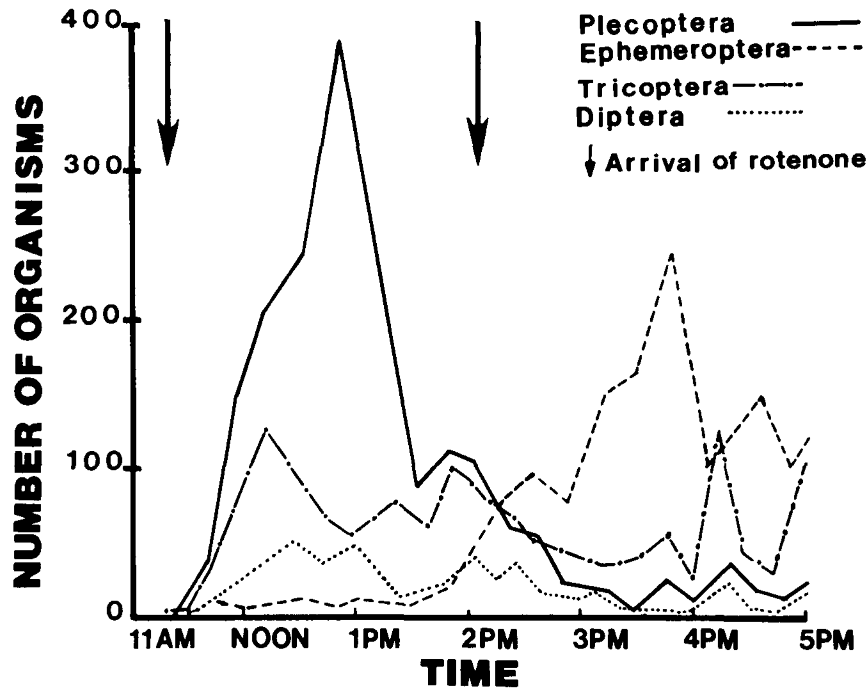

Fig. 2. Changes in the abundances of the four major orders of benthic invertebrates collected as stream drift at station D as a function of time.

The densities of all 4 major orders of insects collected in the benthic samples were severely reduced following rotenone treatment (Table 3). After 11 days, the total number of benthic invertebrates was less than 25% of the pre-rotenone abundance. The marked decrease of the total invertebrate fauna occurred at all four sampling sites. Populations of Plecoptera (stoneflies) and Diptera (black flies and midges) were nearly exterminated, while the total numbers of Trichoptera (caddisflies) and Ephemeroptera (mayflies) dropped sharply to approximately 50% of their pretreatment levels.

Individual genera within these major insect orders varied considerably in their tolerance to rotenone. Populations of three of the eight major taxa represented in the benthic collections, Chironomidae (midges), *Leuctra* (stonefly) and *Prosimulium* (blackfly) were virtually eliminated; all suffered mortalities greater than 95% after the treatment. The caddisflies (*Cheumatopsyche, Hydropsyche,* and *Neophylax* appeared less sensitive to rotenone (mortalities about 40%). The mayflies *Ephemerella* and *Stenonema* exhibited the greatest tolerance (mortalities about 30%) to the toxin.

Table 3. Total number of benthic invertebrates per square foot collected at 4 downstream study stations 1 day before (8/28) and 11 days after (9/8) rotenone treatment.

Stations	*A*		*B*		*C*		*D*		*Total*		
Sampling Dates	8 28	9 8	8 28	9 8	8 28	9 8	8 28	9 8	8 28	9 8	Percent
Number of Samples	2	2	2	2	4	4	4	4	12	12	Reduction
ORDER											
Diptera	712	7	42	4	148	21	16	9	918	41	96
Plecoptera	0	0	15	0	52	0	46	3	113	3	97
Trichoptera	116	31	20	10	47	59	42	7	225	107	52
Ephemeroptera	0	0	0	0	327	119	83	29	410	148	64
Others	35	23	23	5	22	21	15	1	95	50	47
Totals	863	61	100	19	596	220	202	49	1761	349	76

DISCUSSION

The application of rotenone at a concentration of 1 mg/l resulted in an immediate, massive increase in stream drift, and a corresponding, sharp reduction in the abundance of invertebrates. The effect of rotenone on the total invertebrate fauna was catastrophic. The total number of drifting dead and dying insects increased 500 fold during the treatment, while post-treatment benthic invertebrate densities decreased by 76%. All 4 major orders of insects represented were adversely affected. Of these orders, the Plecoptera and Diptera were nearly eliminated; although less severely depressed, the Trichoptera and Ephemeroptera also suffered heavy mortalities. Cook and Moore (1969) reported similar short-term effects, noting that rotenone treatment virtually annihilated the total insect fauna (predominantly stoneflies, dipterans, caddisflies and mayflies) in a California stream.

Although the precise mechanism by which rotenone kills gill-breathing aquatic organisms has not yet been established, the death of both fishes and gill-breathing aquatic insects is most likely caused by a disruption of respiratory metabolism in the citric acid cycle (Claffey and Ruck, 1967). The susceptibility of fishes and, presumably, gill-breathing invertebrates to rotenone appears to be directly related to their oxygen requirements, as would be expected for a respiratory toxin. For example, game fish, especially trout, tend to be less resistant to rotenone than rough fishes, particularly carp (*Cyprinus carpio*) and suckers. Similarly, stream-dwelling aquatic insects with high dissolved oxygen requirements may be more vulnerable to rotenone than taxa inhabiting polluted or standing water. The results presented in this study indicate insects of the orders Plecoptera, Diptera, Trichoptera and Ephemeroptera, all of which are generally considered intolerant of polluted waters with low dissolved oxygen levels, were also extremely sensitive to rotenone.

Since the modes of respiration and individual oxygen requirements of immature aquatic insects may vary widely, even within closely related taxonomic groups, an attempt was made to estimate the relative vulnerability of the major insect taxa to rotenone. On the basis of differential mortalities in the post-treatment benthic samples and varied response times in the drift, the stoneflies *Leuctra* and *Diploperla*, and the blackfly *Prosimulium* were considered the most rotenone-sensitive genera present. Of the remaining major taxa, the caddisflies *Hydropsyche* and *Cheumatopsyche*, and the midges Chironomidae exhibited intermediate sensitivity, while the mayflies *Ephemerella* and *Stenonema* appeared to be the least sensitive to rotenone. With the exception of the caddisfly genera *Hydropsyche* and *Cheumatopsyche*, which were considered to be rotenone-resistant by Engstrom-Heg et al. (1978), scattered references in the literature tend to confirm these observations (Claffey and Ruck 1967; Cook and Moore 1969; Engstrom-Heg et al. 1978).

On the basis of the data obtained in this study it is obvious that stream reclamation programs designed to eliminate nuisance fish species with rotenone can seriously reduce populations of non-target benthic invertebrates, many of which are important fish food organisms. It should be pointed out, however, that the results presented here were limited in scope to short-term effects only and do not necessarily reflect permanent damage to the invertebrate fauna. Cook and Moore (1969) reported a great resurgence of the insect fauna after their initial near annihilation and concluded that rotenone application has little effect on aquatic insects. A similar sequence of events, a severe reduction followed by a nearly complete recovery, usually within 2 years, has been reported by others studying the effects of rotenone on zooplankton (Anderson 1970; Kiser et al. 1963).

Despite the fact that rotenone is a rapidly degradable toxin, and general observations that many aquatic insects have the innate capacity for rapid recolonization, the results presented here clearly indicate the need for careful consideration and adequate evaluation of the potential damage to non-target aquatic life. The ability of stream invertebrates to recover and swiftly recolonize treated areas through drift or other forms of migration appears to be largely a function of the mobility, fecundity, timing of oviposition, life span, resistant life stages and tolerances of the individual species, all of which are often poorly characterized and somewhat unique for each stream system.

In view of the foregoing results, fisheries management agencies should promote the use of on-site bioassays to establish the lowest effective rotenone concentration required to eradicate rough fish and, thereby, at least minimize the risk to non-target aquatic life. Where possible, consideration should be given to partial stream poisoning, or timing applications to correspond with emergence and oviposition of rotenone-sensitive species to minimize damage and maximize the availability of fish forage following treatment (Cook and Moore 1969). For additional recommendations, the reader is directed to the excellent review of fish toxicants provided by Lennon et al. (1971). Clearly, there is a considerable need for continued research on all aspects of stream reclamation with fish toxicants.

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