ARTICLES

LETHAL AND BEHAVIORAL IMPACT OF PERMETHRIN (NRDC 143) ON SELECTED STREAM MACROINVERTEBRATES

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ABSTRACT. In rapid through-flow laboratory tests on the acute toxicity of the synthetic pyrethroid, permethrin, to select stream macroinvertebrates, estimated LC_{90-95} values based on mortalities 24 hr after a 1 hr exposure ranged from 0.001 ppm in the case of *Baetis, Brachycentrus* and *Gammarus* to 0.1 ppm for *Hydropsyche*. *Simulium* larvae—late instar—occupied an intermediate position with the LC_{90-95} approximately 0.005 ppm.

Studies on behavioral reactions, with particular reference to insecticide-induced activation, detachment and downstream drift were carried out in a laboratory simulated-stream, using 30-min exposures to 2 critical concentrations, viz 0.0005 ppm and 0.005 ppm. These showed that on the one hand, *Baetis, Gammarus* and *Simulium* showed a high degree of activation, leading to drift, during the actual 30-min exposure period, and that with *Gammarus* at least this behavior pattern was consistent at both lethal and sublethal exposures. On the other hand *Brachycentrus* showed little sign of activation either during the exposure or subsequently up to 24 hr; at lethal concentrations over 90% of the *Brachycentrus* died in situ without drift.

Hydropsyche showed an intermediate behavior pattern, activation leading to downstream drift only beginning to appear after the end of the 30-min exposure period, then increasing steadily to a peak of over 50% after 2–4 hr. This reaction was consistent even though the concentration of permethrin was one from which over 95% of the test animals survived.

The implications of these findings are discussed in relation to the general problem of insecticide-induced downstream drift of stream benthos, and in relation to the findings, already reported, from similar laboratory studies on temephos and chlorpyrifos methyl.

INTRODUCTION

The synthetic pyrethroid permethrin (NRDC 143) (25% EC. 3-phenoxybenzyl (±) cis, trans 3-(2,2-dichlorovinyl) -2,2-dimethyl cyclopropane carboxylate (Elliott et al. 1975) has been shown to be highly lethal to *Simulium* larvae in laboratory simulated-stream experiments (Muirhead-Thomson 1977). As judged by the minimum lethal concentration sufficient to produce 90–95% mortality 24 hr after a 1-hr exposure, this insecticide was about 10 times more toxic than chlorpyrifos methyl and about 40 times more than temephos. The high toxicity of this compound (as FMC-33297) to mosquito larvae and several non-target organisms was established by Mulla and Darwazeh 1976 and Mulla et al. 1975.

The present investigation is a logical follow-up of the work on *Simulium* larvae and is aimed at establishing relative tolerance levels of some non-target macroinvertebrates. Behavioral reactions of organisms under simulated stream conditions are also reported.

In accordance with evaluation techniques developed in the course of similar investigations on stream invertebrates (Muirhead-Thomson 1977, 1978 a,b), this preliminary laboratory evaluation has taken place in two stages. (i) Establishing the relative tolerance levels on the basis of a standard 1-hr exposure in a through-flow test vessel, followed by a 24-hr holding period in a continuous through-flow of clean water, and (ii) Studying the behavioral responses of select stream organisms in a laboratory experimental channel or simulated stream, with particular reference to insecticide-induced

activation and detachment leading to downstream drift.

APPARATUS

The apparatus has been fully described in previous communications in this series. The same basic unit, a 10-L aspirator bottle with the top cut off, is used in the vertical position for the continuous through-flow tests, and in the near horizontal position for the simulated stream tests.

Stream macroinvertebrates regularly tested were as follows:

Amphipoda. *Gammarus pulex* Linn.
Ephemeroptera. *Baetis rhodani* Pictet[1]
Trichoptera. *Hydropsyche pellucidula* Curtis *Brachycentrus subnubilis* Curtis
Simuliidae. *Simulium equinum* L

METHODS

The test procedures conform to those previously reported (Muirhead-Thomson 1978 a,b) relative susceptibility levels first being established on the basis of an arbitrary standard 1-hr exposure in a continuous through-flow of insecticide-containing water in the test vessel, with the mortality recorded after a 24-hr holding period in a continuous flow of clean water. In case of doubt about mortality or moribundity observations were continued for a further 24 hr. Overall, 7 different concentrations, ranging from 0.01ppm down to 0.0001 ppm were tested, but only *Baetis* was tested over the complete range. The actual number exposed in each test was determined mainly by availability of specimens collected from natural streams, but normally ranged from 20–25 at a time, similar-sized late instars.

In the case of experiments conducted in the laboratory experimental channel or simulated stream, normally 20–25 specimens were introduced into the near horizontal channel and allowed a settling down period of about 24 hr—with aeration—before a flow of water was introduced and the simulated stream established. On the basis of their relative tolerance levels established in the through-flow tests above, the two concentrations selected for the simulated stream tests were 0.005 ppm and 0.0005 ppm (5.0μg/l and 0.5 μg/l).

The number of exposed test animals washed downstream as a result of activation and detachment during the 30min exposure period was recorded, as also was the subsequent mortality after a 24hr holding period in clean flowing water. This latter figure provided a measure of lethal impact in an actual stream of water, and also enabled a comparison to be made between the effect of a 30-min exposure versus the standard 1 hr exposure of the through-flow tests.

In those cases where drift was zero during the actual exposure period, observations were extended to 1-, 2-, 4- and 24-hr subsequent to 1st introduction of the chemical.

RESULTS

The results of the first phase acute toxicity tests (Table 1) shows a high susceptibility to permethrin on the part of all test species with the exception of the caddis larvae, *Hydropsyche*. The $LC_{90\text{-}95}$ 24 hr after a 1 hr exposure is approximately 0.001 ppm for *Baetis, Gammarus* and *Brachycentrus*. For *Simulium* the figure is approximately 0.005 ppm, and for *Hydropsyche* greater than 0.1 ppm.

Observations in the simulated stream (Table 2) show a wide difference in response, with *Gammarus, Baetis* and *Simulium* showing a considerable degree of activation and detachment leading to downstream drift during the short 30-min exposure; this contrasts with *Hydropsyche* and *Brachycentrus* which show lit-

[1] While *Baetis rhodani* was the dominant species in streams from which test material was collected, other species, in particular *B.vernus* Curtis and *B.buceratus* Etn, were recorded in varying proportions at other times. In this preliminary study it was not possible to check whether the reactions of those species differed significantly from those of *B.rhodani*.

Table 1. Relative susceptibility of selected stream macroinvertebrates to permethrin (NRDC 143) based on mortalities 24 hr after a 1 hr exposure in rapid through-flow test vessel at 17.5 ± 1.0° C

Concentration of chemical ppm	*Gammarus*		*Hydropsyche*		*Brachycentrus*		*Simulium*		*Baetis*	
	a	b	a	b	a	b	a	b	a	b
0.1			27	64*						
0.05			40	55						
0.01	40	100	30	30						
0.005	50	100	30	37	20	100	143	95	20	100
0.005 (30-mins)			30	7	98	99				
0.002	50	100	20	15	30	100			20	100
0.001	30	93			50	64	104	60	70	91
0.0005	20	40			57	30	80	0	78	88
0.0005 (30-mins)	40	20			29	41				
0.0002	40	10			45	0	460	2	70	40
0.0001									60	11

a. Total exposed (usually in 2–3 experiments at each concentration).
b. 24 hr mortality as percentage.
Control mortalities throughout remained at 0–2% level.
* (48 hr mortality. Many larvae inert and moribund at 24 hr had recovered completely after a further 24 hr).

Table 2. Impact of 30 min exposures to permethrin on selected stream macroinvertebrates in experimental channel (laboratory simulated-stream) with regard to activation, downstream drift, and subsequent 24 hr mortalities

Concentration of chemical	Test animal	Number of expts.	Total tested	Percentage drifting at intervals after beginning of 30 min exposure minutes						24-hour mortality
				5 min	10 min	15 min	20 min	25 min	30 min	
0.005 ppm	*Gammarus*	3	63	65	79	84	84	84	84%	100%
	Hydropsyche	3	37	0	0	0	0	0	0%	22%
	Simulium (late instar)	2	1050	–	–	9	–	–	46%	92%
	Brachycentrus	3	111	0	0	0	0	0	0%	99%
0.0005 ppm	*Gammarus*	5	73	52	88	91	94	94	94%	32%
	Hydropsyche	4	46	0	0	0	0	0	0%	0%
	Brachycentrus	5	89	0	0	0	0	0	0%	55%
	Baetis	5	143	4	14	24	49	54	63%	90%

Controls. All experiments based on zero detachment during 30 min pretreatment. Simultaneous controls gave maximum detachment during 30 min of 2–3% for *Baetis*, and 1% for *Gammarus*. All others zero.

tle sign of insecticide-induced drift during exposure.

In the case of *Gammarus*, activation which starts within a few minutes of exposure, reached a peak within about 15 min. By the end of 30-min, 84–94% of test animals have drifted downstream, by which time many of the animals exposed to the higher concentration are already dead or moribund. The activity pattern is much the same at the higher, lethal, concentration, and at the lower, less lethal one.

With *Baetis rhodani* detachment developed more slowly and reached a lower peak than with *Gammarus* at the single lower concentration tested. Many *Baetis* were dead or moribund by the end of this exposure period, and there was very little detachment beyond that peak. Many of those which died without detachment in the channel, remained in situ, so that even after 24 hr drift was not complete as some of the dead insects had died under parts of the stony substrate protected from the main current of water.

With *Simulium* larvae, already known to be readily activated by pyrethrins and pyrethroids (Muirhead-Thomson 1970), signs of irritation appeared within a few minutes of first contact with the insecticide-bearing stream, but did not lead to marked detachment and drift until about 15 min later. From that point onwards there was a rapid intensification of irritation and activation, leading to a degree of detachment approaching 50% at the lower concentration tested, 0.005 ppm. Previous observations (Muirhead-Thomson 1977) had shown that at a concentration of 0.01 ppm the same general pattern was shown, with a breakdown of figures showing 48% detachment and drift after 15 min, increasing to 53% at the end of the 30-min exposure period. Both of these concentrations produced mortalities of over 90% in exposed late instars.

With those macroinvertebrates which were not activated on exposure to permethrin, there was no obvious association between this absence of response and the degree of physiological susceptibility to

the insecticide. With both *Hydropsyche* and *Brachycentrus* exposure to 0.005 ppm permethrin failed to produce activation even though this concentration produced widely different lethal effects, viz, mortalities of 22% and 100% respectively.

When observations on these 2 test species were continued beyond the end of the exposure period, and they were once more exposed to a flow of clean running water, differences in behavior pattern appeared (Table 3). While *Brachycentrus* continued to show very little activation, with a drift of less than 10% after 24 hours (larvae which had left their cases), *Hydropsyche* on the other hand began to show signs of irritation and activation shortly after the end of the exposure period. As a result of this activation *Hydropsyche* left the shelter of its stony substrate and became caught in the flow of the current. Thirty min after the resumption of the flow of clean water, significant drift was taking place, reaching a maximum of 55% 2–4 hr after 1st contact with the insecticide. By the end of 24-hr, there was only a further slight increase in the drift. This delayed activation and drift of *Hydropsyche*, even to sublethal exposures of permethrin, is in contrast to its reactions, under similar test conditions, to the organophosphorus larvicides, temephos and chlorpyrifos methyl, in both of which cases drift remained negligible up to 24 hr at least after 1st exposure to the insecticides.

DISCUSSION

These behavioral reactions of stream macroinvertebrates in a laboratory experimental channel or simulated stream are sufficiently marked and consistent to indicate that somewhat similar responses would also be shown in natural streams or rivers exposed to permethrin treatment or contamination.

These observations confirm and extend the general conclusions reached after similar experiments with temephos and chlorpyrifos (Muirhead-Thomson 1977b) in showing that insecticide-induced activation, leading to downstream drift, is not necessarily correlated with susceptibility or physiological tolerance. Some test organisms consistently exhibit a low degree of activation on exposure to the insecticide-stream even at lethal concentrations. Others, which are readily activated to the 3 insecticides tested so far, may exhibit the same type of response both at lethal concentrations and at concentrations from which they completely recover.

The implications of these findings in relation to the phenomenom of 'catastrophic drift' in streams following insecticide application has been fully discussed in the report quoted above. Obviously a great deal more critical experimental work of this kind, using a wider range of stream macroinvertebrates, is indicated before we can begin to understand this complex situation.

In the overall assessment of permethrin impact on a stream ecosystem it would naturally be of considerable value to be able to make a direct comparison between the susceptibility levels of these stream macroinvertebrates, many of them important fish-food organisms, and those of the freshwater fish themselves. The extremely high toxicity of synthetic pyrethroids to a range of test fish species has

Table 3. Comparative drift of the caddis larvae, *Hydropsyche* and *Brachycentrus* during and subsequent to a 30 min exposure to 0.005 ppm permethrin in laboratory simulated stream.

			Percentage drifting					
			During exposure	Subsequent to exposure				
	Number of experiments	Total tested	30 min	30 min	2 hr	4 hr	24 hr	24-hr mortality
Hydropsyche	3	101	0	17	39	55	57	4.6%
Brachycentrus	3	120	0.8	0.8	4	5	9	99.0%

already been established by standard laboratory tests (Marking and Mauck 1975, Mauck et al. 1976), in which the 96-hr LC_{50} for the most sensitive species, rainbow trout (*Salmo gairdneri*), was found to be of the low order of 0.1–0.5 μg/l. Direct comparison between these figures and those in the present report is difficult because of the wide difference in exposure periods, 96 hr versus 1 hr or 30 min, and also because fish toxicity data are normally recorded as LC_{50} values in contrast to the $LC_{90\text{-}95}$ notation adopted in the present investigation. Nevertheless it is clear that the concentration range sufficient to produce 95% mortality in some stream macroinvertebrates after only 1 hr exposure overlaps the range which produces only 50% mortality of test fish after 96 hr exposure, and that *prima facie* the indications are that many fish food organisms are much more sensitive to permethrin than the most sensitive fish species tested.

Confirmation of this was provided through the courtesy of Mr. R. W. Hill of ICI Brixham Laboratory who tested rainbow trout to permethrin at the short exposures adopted in this investigation, that is, to the same brief exposure which fish and other aquatic fauna would be exposed in their natural stream or river habitats subjected to aerial application of insecticide, whether as a forest insecticide or as a *Simulium* larvicide. Using a 1-hr exposure period followed by transfer to clean aerated water, rainbow trout exposed to 50, 25, and 12.5 ppb of permethrin appeared to recover completely, no serious effects being noticed up till 100 ppb. Figures in the present test series showed that with the same 1 hr exposure period, mortalities over 95% were recorded for most macroinvertebrate test organisms at concentrations as low as 5–10 ppb, confirming the much greater sensitivity of typical fish food invertebrates as compared with the fish themselves.

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References Cited

Elliott, M., Farnham, A. W., Janes N. F., Needham, P. H., Pulman, D. A. and Stevenson, J. H. 1975. NRDC 143. A more stable pyrethroid. Proc. 7th Brit. Insect. Fungic. Conf. 1973:721–728.

Marking, L. L. and Mauck, W. L. 1975. Toxicity of paired mixtures of candidate forest insecticides to Rainbow Trout. Bull. Env. Cont. Toxic. 13(5): 518–523.

Mauck, W. L., Olson, L. E. and Marking L. L. 1976. Toxicity of natural pyrethrins and five pyrethroids to fish. Arch. Env. Cont. Toxic. 4:18–29.

Muirhead-Thomson, R. C. 1970. The potentiating effect of pyrethrins and pyrethroids on the action of organophosphorus larvicides in *Simulium* control. Trans. Roy. Soc. Trop. Med. Hyg. 64, 4, 895–906.

Muirhead-Thomson, R. C. 1977. Relative toxicity of temephos (Abate) and permethrin (NRDC 143) to *Simulium* larvae. Mosquito News. 37(2): 172–179.

Muirhead-Thomson, R. C. 1978 a. Relative susceptibility of stream macroinvertebrates to the *Simulium* larvicides temephos and chlorpyrifos methyl. Arch. Env. Cont. Toxic. 7(2): (in press).

Muirhead-Thomson, R. C. 1978 b. Lethal and behavioural impact of chlorpyrifos methyl and temephos on select stream macroinvertebrates. Arch. Env. Cont. Toxic. 7(2): (in press).

Mulla, M. S. and Darwazeh, H. A. 1976. Field evaluation of new mosquito larvicides and their impact on some non-target insects. Mosquito News. 36(3): 251–256.

Mulla, M. S., Darwazeh, H. A. and Majori, G. 1975. Field efficiency of some promising mosquito larvicides and their effects on non-target organisms. Mosquito News. 35(2) 179–185.