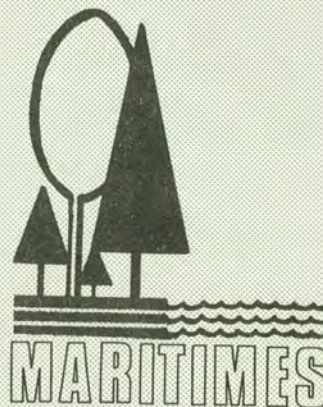


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FENITROTHION  
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STREAM**

BY  
D.C.EIDT



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**TOXICITY OF FENITROTHION TO INSECTS IN A WOODLAND STREAM**

by

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**Maritimes Forest Research Centre**

**Fredericton, New Brunswick**

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### ABSTRACT

A stream was injected with fenitrothion and the effects on aquatic arthropods were monitored. It was determined that a dose of insecticide of about 20  $\mu\text{g}/\text{L}$  for 4 h, resulting from injection at 49.6  $\mu\text{g}/\text{L}$  for 2.5 h, was enough to kill larvae of the stonefly *Nemoura (Amphinemura) wui* Claassen and to a lesser extent, larvae of the mayfly *Baetis* sp. prob. *herodes* Burks, but not other species. *Leuctra* spp. stonefly larvae in cages were killed at a site 59 m below the injection point of fenitrothion at 69.1  $\mu\text{g}/\text{L}$  for 2.5 h, but the effect diminished downstream. *Leuctra* spp. larvae in cages buried up to 34 cm in the substrate showed better survival than those resting on the stream bed.

### RESUME

Les effets subis par les arthropodes aquatiques d'un ruisseau traite au fenitrothion furent notes. On y determine qu'une dose d'insecticide d'environ 20  $\mu\text{g}/\text{L}$  pendant 4 h, etait suffisante pour tuer les larves de la perle *Nemoura (Amphinemura) wui* Claassen et dans une certaine mesure les larves de l'ephemere *Baetis* sp. prob. *herodes* Burks, mais aucunes autres especes. Des larves encagees des perles *Leuctra* spp. furent tuees a un point 59 m en aval de l'endroit injecte a raison de 69.1  $\mu\text{g}/\text{L}$  pendant 2.5 h, quoique l'effet toxique diminua plus en aval. Les larves des especes *Leuctra* contenues dans des cages enfouies jusqu'a 34 cm dans le lit du ruisseau, demontrèrent une meilleure survie que celles reposant sur le fond du ruisseau.

## INTRODUCTION

Since 1968 when fenitrothion was adopted as the principal chemical for control of spruce budworm, *Choristoneura fumiferana* (Clemens), the effects on other organisms have been intensively studied. Because of the number of organisms, the complexity of their associations, and innumerable other variables, this has proven to be an unending task. It has, however, been possible to identify specific concerns, one of which is the effect on aquatic systems through fish-food organisms. Most fish-food organisms in streams are insects, which logically are sensitive to insecticides in varying degrees.

The toxicity of fenitrothion to aquatic arthropods has been determined in the laboratory for larvae of one species of a caddisfly, a stonefly, a dragonfly, a dobsonfly, and a crane fly (Wildish and Phillips 1972), three species of mosquito larvae, one of water flea, and a scud (Flannagan 1975), and another mosquito larva (Tadano 1970). Unfortunately, these toxicities cannot be interpreted in terms of the rapidly changing concentrations found in field situations. The decline in fenitrothion concentration in streamwater follows a negative exponential curve (Eidt and Sundaram 1975, Sundaram 1974). Although Eidt (1975a) and Flannagan (1975) give data on kill of aquatic arthropods that relate to fenitrothion in streams, there was no control over dosage. There is no way, except by analyzing a great many water samples, to know either the maximum concentration attained after an operational spray or the concentrations between samples.

Symons (1977) has thoroughly reviewed the literature up to the beginning of 1976 and has evaluated the hazard of operational fenitrothion sprays. By synthesizing the observations of many workers, he calculated an acute ecological effective dosage that kills 50% of the aquatic insects ( $ED_{50}$ ) to be a spray rate of 450 g fenitrothion/ha. This figure, he admits, treats the insects in a stream as if they were all a single species, and he acknowledges the wide variation in insecticide concentrations that may occur in streams for a multitude of reasons besides spray emission rate. He also ignores the generally low confidence levels in the biomass reduction data he used to derive the  $ED_{50}$  but it is the best hazard index we have.

Both Eidt (1975a) and Flannagan (1975) consider the possibility that animals buried in the stream bed are relatively safe from fenitrothion exposure. Eidt (1977) attempted to check this point under operational spray conditions and found only weak evidence

that insecticide concentration diminished with depth in the stream bed and that survival of caged insects increased.

This paper describes a field experiment designed to further define the dose of fenitrothion that will cause drift and kill of aquatic arthropods, and to determine conclusively if arthropods deep in the stream bed are safer from insecticide poisoning than those at or near the top of the stream bed.

## METHODS

A stretch of Narrows Mountain Brook, New Brunswick, (46° 17'N, 67° 01'W; Middle Brook before 1976) was chosen for the experiment. The wooded drainage basin of the stream is characterized in a brochure published by the Nashwaak Experimental Watershed Project (Anon [1976]). The stream is a clear cold tributary in the headwaters of the Nashwaak River with usual flow rates ranging from 11 to 700 L/sec and a water temperature rarely exceeding 17°C. Other parameters were measured by Dr. H.H. Krause, Faculty of Forestry, University of New Brunswick (personal communication); median values varied little from 1972 to 1977: pH 7.0, Na 1.2 ppm, K 0.3 ppm, Ca 5 ppm, Mg 0.8 ppm,  $NO_3^-$ -N 0.03 ppm, Kjeldahl-N 0.5 ppm, P 7 ppb, and suspended sediment 0.4 ppm.

The stretch was treated twice with fenitrothion. The pH of the water was 6.7 and the water temperature rose from 11 to 15°C during the course of the first treatment on 1 June. The temperature remained steady at 11°C during the second treatment on 14 June. The average water speed in the experimental stretch was estimated, using drifting orange peel, to be 61 cm/sec on 1 June, or 2 min and 37 sec travel time at midstream from the barrier net to the lowest drift sampling station. The stream discharge on 1 June was steady at 37 L/sec and on 14 June dropped from 161 L/sec at 1100 h to 153 L/sec at 1400 h.

A suitable place was selected where a barrier net could be installed to filter out drift above the test stretch. An insecticide dosing station was located on a mid-stream boulder 50.7 m above the barrier net. This distance was greater than that previously determined visually, using rhodamine WT dye, to be adequate for complete mixing across the stream. Various sampling stations were located, according to mid-stream distances as a tape measure was carried by the current, above and below the injection site (Table 1). One drift sampling station was established above the insecticide dosing station and four were established

Table 1. Distance (m) from dosing site to barrier net, caged insects, and drift and water sampling stations, 1 June 1977. Five cages and five water samples were clustered within  $\pm 1.9$  m of the location indicated.

Barrier net	Drift		Cages		Water	
	Station number	Distance	Station number	Distance	Station number	Distance
50.7	1	-11.9				
			1	57.5		
		58.4			1	64.4
		74.0				
		95.7				
			2	102.1		
					2	107.9
		145.7				
					3	146.7
				3	152.3	
			4	177.8		
			5	220.6		

58.4, 74.0, 95.7, and 145.7 m below the injection site (7.7, 23.3, 45.0, and 95.0 m below the barrier net). Three water-sampling stations were established about 64, 108, and 147 m below the dosing station. Caged *Leuctra* spp. larvae were located approximately 58, 102, 152, 178, and 221 m below the injection site for the initial run of the experiment 1 June; the cages were located approximately 61, 103, 150, and 219 m below the injection site when the experiment was repeated 14 June.

#### Barrier net

The barrier net was constructed of saran cloth with 0.42 mm apertures. It was stapled to a pole along its top edge and weighted to the bottom with lead attached to the selva and with rocks. The entire stream passed through the net except the water that passed among the bottom materials. A barrier net was used during the 1 June treatment, but not during the 14 June treatment because drift was not sampled.

#### Drift sampling

Drift was sampled with nylon bolting cloth nets 31 cm wide, 66 cm long, with 0.6 mm apertures. They were affixed so they sampled a column of water from the stream bottom to and including the surface. To

take a sample, the net was set for 15 min so that an estimated 10 to 20% of the stream passed through the net. Samples were taken at half-hour intervals beginning before dosing began and continuing until 1.5 h after dosing ceased. The catches were placed in stream water in sealed jars which were kept in the stream to prevent warming. All catches were sorted 2 to 12 h later according to living and dead organisms and were preserved in alcohol.

#### Caged insects

Insects for bioassay were caged in the stream in a manner similar to that described by Eidt (1977). They were collected in early nighttime drift and *Leuctra* spp. stonefly larvae were carefully selected to avoid excessive handling. These larvae had been shown by Eidt (1975a) to be one of the stream animal species most sensitive to fenitrothion. Ten larvae and a leaf of speckled alder, *Alnus rugosa* (DuRoi) Spreng, were placed in each of several cages of black plastic pipe, 2.6 cm ID by 9 cm long. Nylon cloth, of aperture size 0.2 mm, covered each end. One cage was placed on the stream bottom and three were buried 5 days before the treatment of 1 June at each of five stations below the injection site. Cages were similarly placed 4 days before the treatment of 14 June at each of four

stations below the injection site. It was difficult to bury the cages in stony rubble at the planned depths of 10, 20, and 30 cm, so the depths were remeasured when the cages were removed the day after the 1 June treatment, and 2 days after the 14 June treatment. Cages were transported from the field in ice water and examined for insect survival on the day of removal. Dead larvae with no physical injuries and larvae showing the typical twitching of terminally organophosphate-poisoned insects were classified as insecticide-killed.

#### *Insecticide dosing*

The stream was treated with fenitrothion in the mix generally used by Forest Protection Limited to spray New Brunswick's spruce budworm-menaced forest. The formulation was 150 parts of 95-97% technical fenitrothion, 50 parts of the emulsifier Atlox 3409F, and 20 parts of a solvent oil CFTX 107 Aerotex 3470.

The insecticide was injected into the stream using a mariotte bottle to ensure a uniform rate of flow. First, the stream discharge was determined with an Ott propellor-type flowmeter (which was checked later against a gauging station operated by the Water Survey of Canada). Then the concentration of the emulsion in the bottle was adjusted to give the desired concentration in the stream. There was no difficulty caused by separation of the emulsion such as that experienced by Symons and Harding (1974). The first treatment was 1 June 1977 between 0755 and 1025 h AST at a calculated concentration of 49.6  $\mu\text{g}$  a.i./L. An interruption of unknown duration occurred before 0845 h when it was found that the insecticide flow had stopped. The second treatment was 14 June between 1030 and 1300 h AST at a calculated concentration of 69.1  $\mu\text{g}$  a.i./L. The insecticide was considered completely mixed by the time it reached the barrier (on the basis of a simple visual test performed earlier with rhodamine WT dye).

#### *Insecticide residue analysis*

Surficial water samples were collected from mid-stream and interstitial water samples from three depths in the stream bed at each of three stations below the insecticide injection site. They were collected in glass jars with aluminum foil cap liners, then wrapped in aluminum foil to omit light. Surficial water was sampled by moving the jar between the bottom and the surface to assure a representative sample each time. Stream bed interstitial water was sampled by siphoning

through lengths of black rubber tubing which were installed near planned depths of 10, 20, and 30 cm, in the manner described by Eidt (1977), but without replication. All samples were kept in the dark at 15.3°C or less, and the fenitrothion was extracted with chloroform within 15 h of collection. The samples were later analysed for fenitrothion and its degradation products by *in situ* fluorometry after thin-layer chromatography by Dr. Victorin Mallet, Université de Moncton, N.B. (Zakrevsky and Mallet 1977). Six duplicate samples were analysed by flame-photometric detection with gas-liquid chromatography by G. Brun, Water Quality Laboratory, Fisheries and Environment Canada, Moncton, N.B. The results were similar.

## RESULTS

#### *Fenitrothion concentrations*

Fenitrothion concentrations found in the water during and after the first dosing on 1 June are shown in Table 2. Since no operational spraying took place within 27 km of the stream, the concentration before treatment was assumed to be 0. Although the calculated concentration was 49.6  $\mu\text{g}/\text{L}$ , the highest concentration detected in water samples was 26.1  $\mu\text{g}/\text{L}$ . Concentrations of fenitrothion after treatment began were generally higher at 30 min than at 1 h, and were higher again at 2 h. This variation resulted from the interruption of flow from the mariotte bottle that was discovered 30 min after the treatment began. The reason for interruption was that cotton string supporting the nozzle got wet, shrank, and raised the nozzle thus reducing the head to zero and stopping the flow. The flow diminished gradually before ceasing, then the concentration rose substantially when flow resumed. The concentration of insecticide was thus not uniform during the dosing period and may have attained a much higher level during treatment than was recorded by sampling. The data in Table 2 do not indicate that concentrations or duration of the presence of the insecticide differed among stations.

Depths of tubes for sampling interstitial water were remeasured when they were removed, and found to be 11, 17, and 20 cm at station 1; 11, 20, and 28 cm at station 2; and 11, 18, and 21 cm at station 3. The concentrations at various depths (Table 2) follow no clear pattern although it seems from the 4-h samples that concentrations lingered at the greatest depths 1 h and 30 to 34 min after the treatment ceased. It certainly took no longer than 30 min after treatment began, to rise to a high level at most stations.

Table 2. Fenitrothion concentrations in stream water, 1 June 1977, using TLC with *in situ* fluorometry, unless otherwise indicated

Distance (m) below injection	Lapsed time (h:min) after		Concentrations of fenitrothion ( $\mu\text{g/L}$ )			
	Dosing began	Dosing ended	Surface water	Interstitial water depth in stream bed (cm)		
64	0:30	-2:00	16.3	11	17	20
	1:00	-1:30	16.9	4.8	13.7	23.5
	2:00	-0:30	25.5	6.3	11.3	3.8
	4:00	1:30	N.D. <sup>1</sup>	—	22.5	16.8
108	0:30	-2:00	9.8	11	20	28
	1:00	-1:30	11.8	5.4	4.4	22.4
	2:00	-0:30	13.5	3.8	9.1	11.3
	4:00	1:30	0.4 <sup>2</sup>	10.0	10.6	25.3
147	0:34	-1:56	16.0	11	18	21
	1:04	-1:26	6.0	22.8	7.3	18.1
	2:04	-0:26	26.1	6.0 <sup>3</sup>	5.4	7.5
	4:04	1:34	1.8	25.6	15.3	17.5
				0.5 <sup>2</sup>	8.4	3.8

<sup>1</sup> Not detectable, below the lowest limit of detectability of 0.6  $\mu\text{g/L}$ .

<sup>2</sup> Determined by GLC with flame-photometric detection.

<sup>3</sup> Estimated.

The water was not sampled for fenitrothion concentration during the 14 June treatment. There was, however, no interruption in insecticide flow and the prescribed 69  $\mu\text{g/L}$  was injected at a constant rate for 2.5 h while stream discharge remained constant.

#### Drift of invertebrates

The numbers of aquatic arthropods in drift samples were essentially uniform throughout the experiment at the control station above the treated stretch (Table 3); dead insects were found only in the first three samples. At the other stations, catches during the treatment period did not differ from the control station, nor were they different from the 0840 sample which was taken before injection started. An effect of the insecticide is clearly shown at 1110 and at 1140 at all the downstream stations when catches of dead but not of living insects increased several-fold (Fig. 1). The most abundant species in dead drift

following treatment was the stonefly *Nemoura (Amphinemura) wui* Claassen (Table 4). The mayfly, *Baetis* sp. prob. *herodes* Burks was the only other species killed in sufficient numbers to be sure of an effect by the insecticide.

Drift was not sampled during the 14 June treatment.

#### Cages

There was no evidence that any caged insects were killed by the 1 June insecticide treatment (Table 5). More than half the dead *Leuctra* spp. were crushed or otherwise injured in handling. In the 22-cm-deep cage at station 5, they were probably attacked by a *Leuctra*-sized *Rhyacophila* sp. larva which was inadvertently introduced. Others may have escaped. Other living insect larvae found in cages were 3 *Paraleptophlebia* sp., 1 *Ephemerella (Ephemerella)* sp., 1 chironomid, 2 very small *Rhyacophila* sp., 1 *Lepidostoma*



Table 3. Aquatic insects and mites in 15-min drift samples from Narrows Mountain Brook, 1 June 1977, L = living, D = dead

Station	Distance (m) below injection	Time of net set (AST)																	
		0740		0810		0840		0910		0940		1010		1040		1110		1140	
		L	D	L	D	L	D	L	D	L	D	L	D	L	D	L	D	L	D
1	-11.9	4	4	5	2	2	2	4	0	4	0	2	0	6	0	2	0	2	0
2	58.4	0	2	1	0	1	1	0	1	2	7	0	2	3	5	0	20	1	7
3	74.0	4	1	1	2	2	4	3	1	4	1	0	7	0	8	0	23	1	14
4	95.7	3	0	2	3	3	1	1	6	0	0	1	5	0	7	2	33	2	35
5	145.7	3	0	2	1	4	0	3	3	0	1	3	2	4	0	3	28	0	74

Table 4. *Nemoura (Amphinemura) wui* Claassen in 15-min drift samples from Narrows Mountain Brook 1 June 1977, L = living, D = dead

Station	Distance (m) below injection	Time of net set (AST)																	
		0740		0810		0840		0910		0940		1010		1040		1110		1140	
		L	D	L	D	L	D	L	D	L	D	L	D	L	D	L	D	L	D
1	-11.9	1	2	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
2	58.4	0	0	0	0	0	0	0	0	0	1	0	0	0	4	0	16	0	6
3	74.0	2	0	0	0	0	1	0	0	0	0	0	1	0	5	0	17	0	8
4	95.7	0	0	0	0	0	0	0	1	0	0	0	2	0	4	0	30	0	29
5	145.7	1	0	0	0	0	0	0	1	0	0	0	2	0	0	0	24	0	60

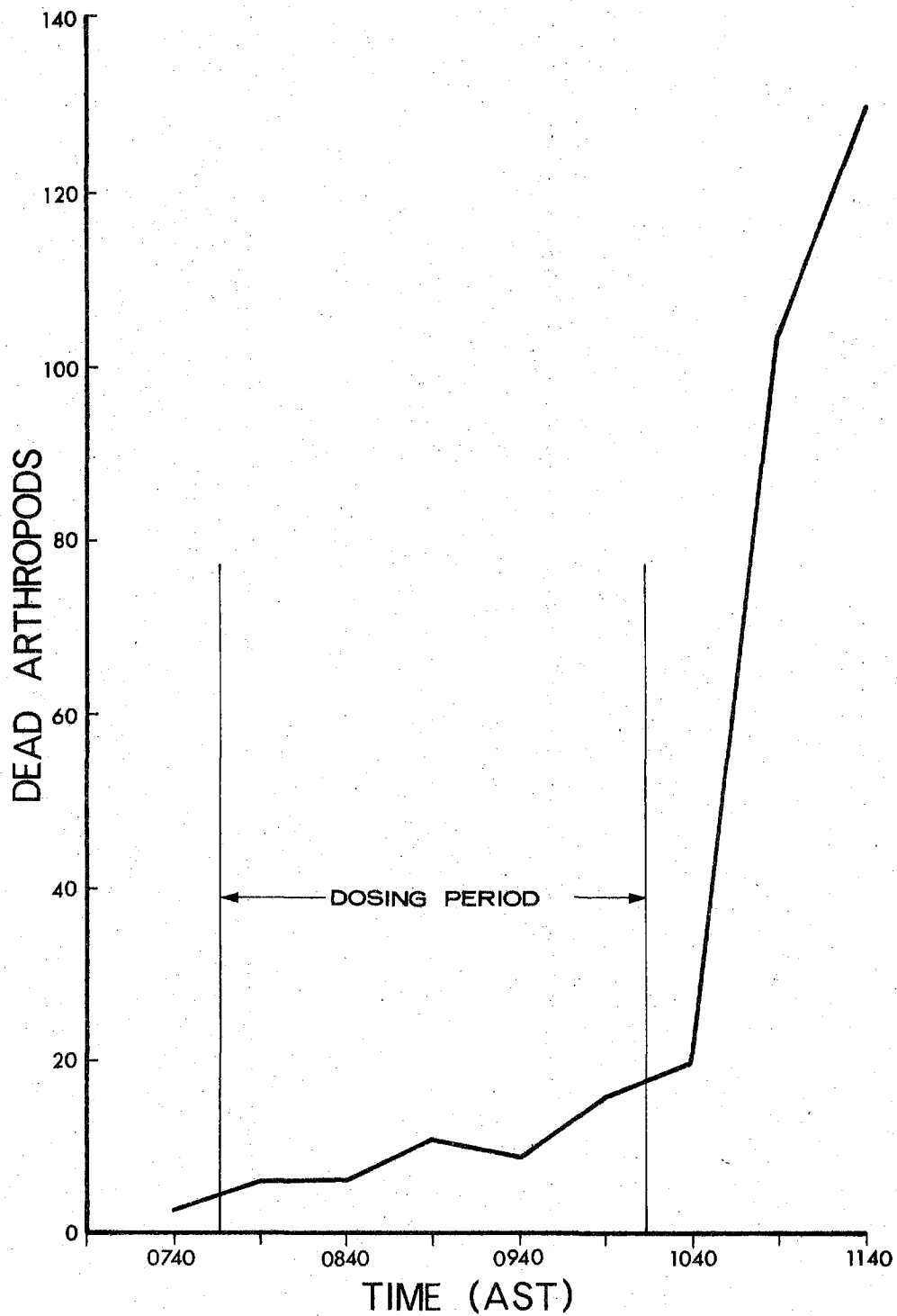


Figure 1. Dead arthropods collected in 15-min drift samples over starting times of net sets. Totals of four stations below injection site, Narrows Mountain Brook, 1 June 1977.

Table 5. Condition of caged *Leuctra* spp. following 1 June 1977 treatment at 49.6  $\mu\text{g}$  fenitrothion/L for 2.5 h

Station	Distance (m) below injection	Depth (cm) below stream bed	<i>Leuctra</i> spp.			
			Living	Dead		
				Cause unknown	Crushed or injured	Missing
1	57.7	0	10	0	0	0
	54.0	10	4	1	2	3
	56.5	20	8	1	0	1
	58.9	25	8	0	2	0
2	103.5	0	10	0	0	0
	100.7	10	8	0	0	2
	101.4	20	9	0	1	0
	102.1	20	4	1	1	4
3	150.8	0	7	1	0	2
	150.6	6	5	0	0	5
	152.2	25	8	0	0	2
	154.0	28	7	1	0	2
4	177.1	0	3	3	2	2
	176.4	12	12	0	1	0
	178.1	17	5	0	0	5
	179.2	25	5	0	2	3
5	220.1	0	8	0	1	1
	220.8	10	9	1	0	0
	221.3	22	3	pieces of unknown number		
	219.8	30	8	1	0	1

sp., 3 *Nemoura wuji*, 2 very small unidentified Plecoptera, and 3 additional *Leuctra* spp.

The increased insecticide dosage of 14 June was successful in inducing mortality in the cages (Table 6). Mortality was virtually complete at the upper levels at the two upstream stations except for one living specimen at station 1. The latter specimen was among the first examined and through inexperience unfortunately was not properly classified as healthy, sick, or twitching. There was considerable mortality at all levels at the first two stations but less at the two deeper levels. At station 3, there was mortality on the stream bed but not below it, and survival was complete at all levels. There was no mortality attributable to the insecticide at station 4, however, the cage on the stream bed had washed away. The other lost samples indicated in Table 6 were from cages damaged during their removal; they probably contained many survivors because 16 living

*Leuctra* larvae were found in the bucket in which the cages were transported from the field.

## DISCUSSION

The actual dose of insecticide received by the stream fauna on 1 June, whether on the stream bed or below it, is difficult to determine because of the interruption of flow from the mariotte bottle. Because of the timing of the samples relative to the dosing period, the highest concentration in surficial water undoubtedly exceeded the maximum measured (26.1  $\mu\text{g}/\text{L}$ ), and probably approached an average of 20  $\mu\text{g}/\text{L}$  over the 4 h following the start of treatment. This estimate was determined by graphically interpolating concentrations between the surficial water samples and averaging the means for each 15-min interval in the 4-h period.

Table 6. Condition of caged *Leuctra* spp. following 14 June 1977 treatment at 69.1 µg fenitrothion/L for 2.5 h

Station	Distance (m) below injection	Depth (cm) below stream bed	<i>Leuctra</i> spp.					
			Living		Dead			
			Healthy	Sick	Twitching	Not moving	Crushed	Missing
1	59.0	0	0	1	0	5	1	3
	61.3	15	0	0	2	5	0	3
	64.7	19	0	2	4	2	1	1
	58.7	29	1	2	4	2	1	0
2	103.8	0	0	0	5	4	0	1
	102.2	12	0	0	5	4	1	0
	103.7	22			sample lost			
	102.7	34	1	2	2	3	0	2
3	ca. 150	0	0	3	0	7	0	0
	ca. 150	11	0	10	0	0	0	0
	ca. 150	26	0	10	0	0	0	0
	ca. 150	32			sample lost			
4	—	0			cage lost			10
	219.4	11	7	0	0	0	2	1
	220.0	18	7	0	0	0	1	2
	217.9	27	5	0	0	0	0	5

There is no evidence in the data that the concentrations were any lower in the interstitial water. This is not surprising because the stream bed was very coarse and variable to start with and fine stream bed materials were washed out by the current when the tubes were installed, which would allow freer water movement from above. However, the 4-h sample data (Table 2) clearly show that the insecticide lingered longer at greater depths, and by the same process should have arrived later.

No streamwater samples were taken on 14 June for fenitrothion analysis because of budgetary limitations. However, the dose received should have approximated the calculated dose of 69.1 µg/L for 2.5 h. The total amount of insecticide injected agreed with the expected, based on the delivery rate of the mariotte bottle and the stream discharge data obtained from the Water Survey of Canada which used a concrete V-notch weir and a modern pressure-type water level recorder. Using a similar insecticide delivery method, Symons and Harding (1974) found less than ±5% variation in fenitrothion concentration in streamwater over an 8-h period.

There was no evidence from drift samples of insect poisoning during the dosing period 1 June, but an effect was clearly shown by the drift samples taken between 45 min and 1 h 15 min after dosing ceased, i.e. at 1110 and 1140 h (Fig. 1). The gradation in numbers in catches from stations 2 to 5 (Fig. 2) is explained by the increased area of origin of drift as the distance below the barrier net increased and does not indicate a greater insecticidal impact at downstream stations. The impact was brief because when the last sample was taken at 1140 h, which was between 1 h 15 min and 1 h 30 min after treatment ceased (Fig. 3), the peak of dead drift had passed at stations 2 and 3, 58.4 m and 74 m downstream from the injection site, although it had levelled off at station 4, 95.7 m downstream, and was still rising at station 5, 145.7 m downstream.

The stonefly *Nemoura wui* constituted the bulk of the insecticide-killed drift and the mayfly *Baetis* sp. prob. *herodes* was also affected, but *Leuctra* spp. were not. *Leuctra* spp. were abundant in fenitrothion-killed drift in the same brook in 1973 (Eidt 1975). The explanation may lie in the behaviour of *Leuctra* spp. which were abundant in normal drift between nightfall

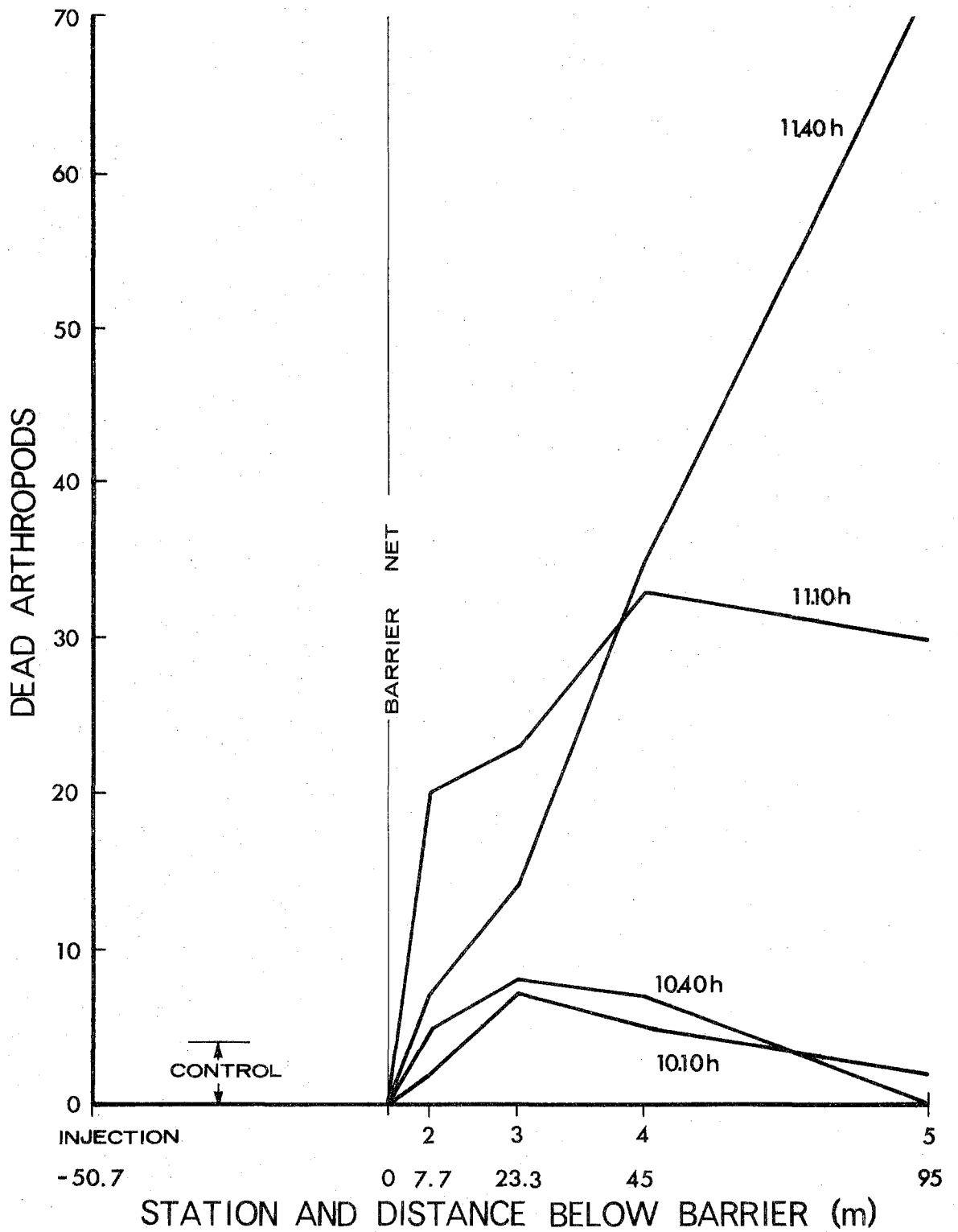


Figure 2. Dead arthropods collected in 15-min drift samples (by starting times of net sets) over distance (m) below insecticide injection site, Narrows Mountain Brook, 1 June 1977.

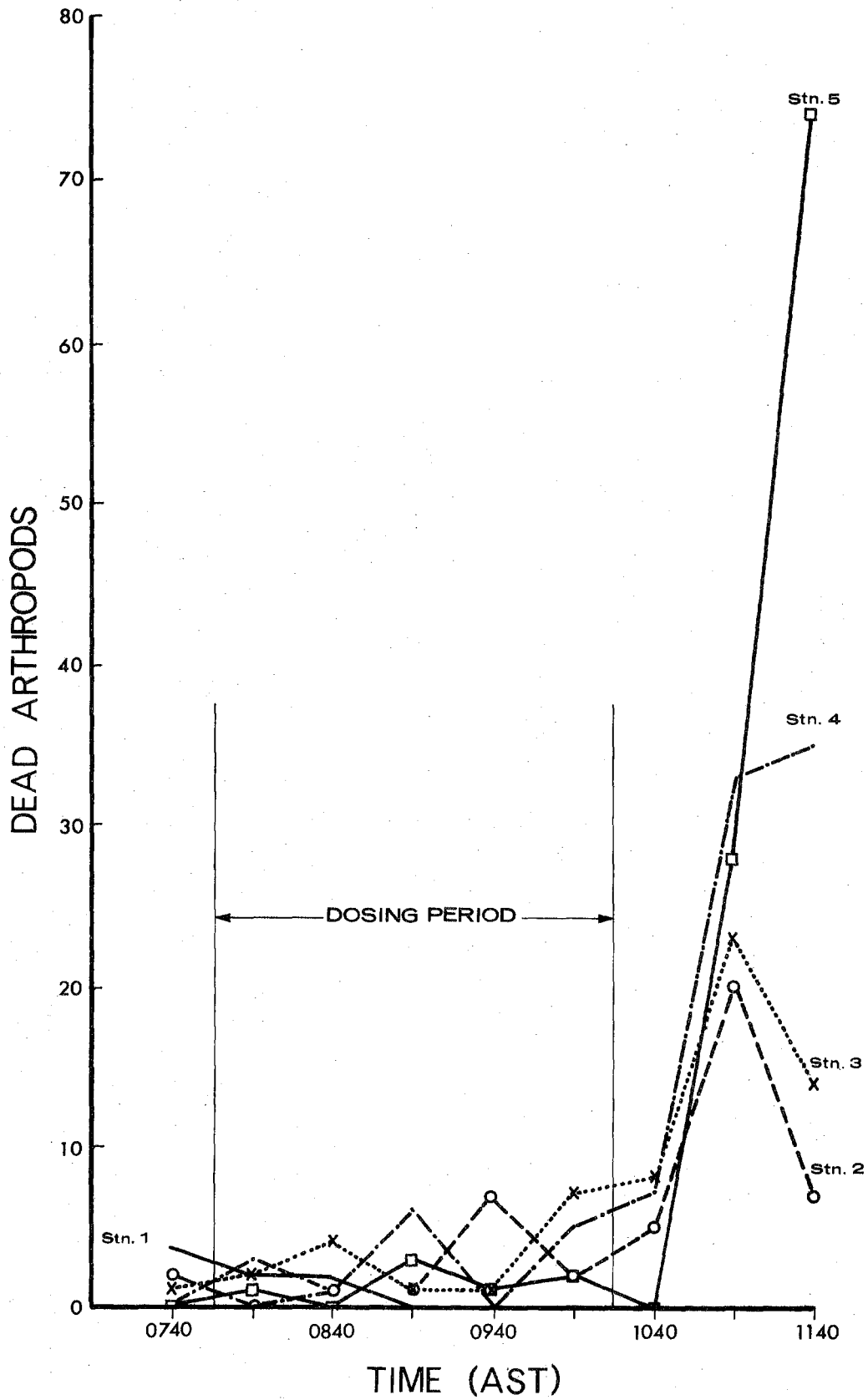


Figure 3. Dead arthropods collected in 15-min drift samples by station over time (AST) of net set, Narrows Mountain Brook, 1 June 1977.

and midnight about 100 m upstream. This may have resulted from differences in the drifting habits or vertical movements in the stream bed of the two species, but may also have reflected differences in the two stretches. The stretch where *Leuctra* was dominant in drift was undisturbed whereas the stretch where *N. wui* was dominant was still much altered from culvert construction four years earlier.

The drops in numbers in catches taken at 1140 from those taken at 1110 h at stations 2 and 3 (Fig. 2) were because the greatest impact on *N. wui* was past by 1140 h (Table 4). On the other hand, *N. wui* drift was still increasing at station 5 at 1140 h. Likewise, the effect on *Baetis* sp. prob. *herodes* was still increasing because the number of dead specimens at all treated stations was 9 at 1110 h and 18 at 1140 h, whereas only 4 dead specimens were found in all earlier samples and none at any time at the untreated station.

Sampling of drift ended too soon to observe the return to pretreatment levels. However, the process had already begun at stations 2 and 3 where catches were lower at 1140 than at 1110 and at station 4 where the rate of increase was much diminished (Fig. 3). It seems that one or two more drift samples at half-hour intervals would have shown declines at all stations.

That the treatment of 1 June did not affect larvae of *Leuctra* spp. in cages on the stream bed, when dead *N. wui* larvae appeared in drift, was unexpected. *Leuctra* spp. are at least as susceptible to fenitrothion poisoning as *N. wui* (Eidt 1975a). Thus, it seems that the cages, perhaps by restricting water flow, afforded some protection to the caged larvae.

There is good evidence from the results of this experiment that the hazard to benthos diminishes with depth in the stream bed. The data are inadequate to make a conclusive statement particularly since corollary differences in fenitrothion concentration with depth were not shown. However, with the experimental evidence from 1976 (Eidt 1977), the implications of the 1973 observations of Eidt (1975a) that insecticide-killed drift had the same periodicity as live drift, and Flannagan's [1975] failure to find fenitrothion in the fine substrate material of the treated stream, there seems little doubt that an undisturbed stream bed is indeed a refugium for benthos from the side effects of insecticides used in forest sprays.

The higher insecticide dose of 14 June (69.1  $\mu\text{g}/\text{L}$  for 2.5 h, based on injection rate) was high enough to kill *Leuctra* spp. in cages, but only at the three upstream stations. The lethality of a dose is a function of the concentration and the exposure time. The caged

insects would be exposed to essentially the same quantity of fenitrothion at all stations because sufficient time passed for a complete flushing of the stretch and in only 160 m with no tributaries entering and a water speed of 61 cm/sec it seems safe to speculate that not much insecticide would be lost. However, the downstream cages would be exposed to the lowest concentration for the longest time because of the tendency of downstream concentration curves to have lower peaks and longer tails (Morgan 1976). The toxicity of a given amount of fenitrothion is very much a function of how it is administered. For example, the kill of benthos observed in 1973 by Eidt (1975a) at much lower insecticide concentrations is explained by the longer exposure. The  $\text{LC}_{50}$  or median lethal concentration of a substance to a population of test animals, must always be qualified to account for exposure time. As far as I can determine, nobody has attempted to interpret these  $\text{LC}_{50}$ 's in terms of rapidly changing concentrations as found in some field situations. To the contrary, toxicologists strive to maintain fixed concentrations of chemicals during tests while decomposition, sorption, and assimilation tend to change the test medium.

To derive a statistic that incorporates both concentration and time, I arbitrarily multiplied the concentration of fenitrothion in  $\mu\text{g}/\text{L}$  by the time of exposure in hours (CT). Something similar was done by Fredeen (1974) who used a statistic 'ppm x minutes' to describe dosages of black fly larvicides injected into rivers. With changing concentrations the hourly means estimated from a hand drawn curve were added to get a value for CT. All of the situations where data were adequate to calculate CT, and the kill of aquatic insects was measured or estimated, are given in Table 7. In the two instances where the values were below 41, no kill occurred; five other examples, not included in Table 7, had CT values much lower than 15 and no kills occurred. When CT values were 48 or higher, kills occurred, involving increasing numbers and species as CT increased. The relationship is not a simple one because different values for CT resulted from the 1-h and 24-h carefully controlled experiments of Wildish and Phillips (1972). Fredeen (1974) suspected as much because he said that comparison of 96-h and 15-min exposures, both at 6 ppm x minutes dosage, was questionable. There is no way of knowing from these data what the relationship is between concentration and time but various suggestions have been made (Hynes 1966) based on constant concentrations. CT values are obviously not the answer, but some measure

Table 7. Comparisons between the effect of fenitrothion on aquatic insects and several values of CT

CT ( $\mu\text{g/L} \times \text{h}$ )	Peak concentration observed ( $\mu\text{g/L}$ )	Effect	Source
15	3.8	none	Narrows Mountain Brook, 1974 (Eidt 1975b)
41	6.38	none	Lake Brook, 1973 (Eidt 1975a)
48	3.75	slight kill	Narrows Mountain Brook, 1976 (Eidt 1977)
48	2.0	24-h $\text{LC}_{50}$ of <i>Acroneuria</i> sp.	Laboratory (Wildish & Phillips 1972)
66	5.25	substantial kill	Narrows Mountain Brook, 1973 (Eidt 1975a)
66	66.0	1-h $\text{LC}_{50}$ of <i>Acroneuria</i> sp.	Laboratory (Wildish & Phillips 1972)
95	26.1	kill	Narrows Mountain Brook, 1977
138	?	kill	Narrows Mountain Brook, 1977
152	21	kill	Lake Brook, 1976 (Eidt 1977)

is needed to relate toxicities to rapidly changing concentrations of pollutants and to relate these toxicities to those at fixed concentrations. The first step would be to conduct toxicology experiments with a sensitive species in rapidly changing, describable, concentrations of insecticide, such as are found in streams after forest spraying. Sources of variation, such as water chemistry, dissolved oxygen, temperature, species and stage of test animal, and other toxicants, would eventually have to be considered.

### CONCLUSIONS

It was determined from drift collections that a fenitrothion concentration of about 20  $\mu\text{g/L}$  for 4 h, resulting from injection at 49.6  $\mu\text{g/L}$  for 2.5 h, was adequate in Narrows Mountain Brook in early June to kill *Nemoura wui* and *Baetis* sp. prob. *herodes*, in order of sensitivity, but not other species. This dose was not enough to kill caged *Leuctra* spp., which may have been protected by their cages.

*Leuctra* spp. in cages were killed by injection of fenitrothion at 69.1  $\mu\text{g/L}$  for 2.5 h but the effect diminished rapidly downstream from the injection point.

There was insufficient evidence to say that concentrations of fenitrothion were lower in interstitial water, although the insecticide lingered longer at greater depths and by the same process should have arrived later.

It is evident, in spite of lack of corollary evidence on insecticide concentration, that benthos deep in a stream bed is less likely to be poisoned by incidental fenitrothion from aerial forest spraying than benthos at or near the surface of the stream bed.

In circumstances where insecticide concentration changes, a measure of dose is needed to relate kills in various field situations and to relate field toxicity to laboratory-determined toxicities.

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