8 INSECTICIDES IN FRESHWATER

L C V Pinder, W A House, I S Farr & J A B Bass

8 INSECTICIDES IN FRESHWATER.

8.1 Introduction

The preceding report in this series Pinder et al (1990) (In Davis, 1990) emphasised laboratory studies of acute toxicity of low levels of insecticides to aquatic invertebrates, both with and without the presence of organic sediment. Contamination levels were selected to be representative of the range of concentrations that could realistically result from contamination of shallow water bodies by spray drift, using data contained in the first report in the series (Williams et al 1987). Field experiments to investigate the impact of actual drift events are a logical and desirable development of these studies. Two such experiments were carried out during the present phase of the work. One of these was concerned with the impact of cypermethrin drift from an experimental, ground based application, on the air breathing Hemiptera, *Corixa* sp and *Notonecta* sp. The second, which is separately reported in Chapter 7, investigated the effects of deltamethrin drift following aerial spraying, on a range of target organisms.

However, spray drift is only one of the routes whereby surface freshwater bodies may become contaminated, run-off and subsurface drainage are also important. Synthetic pyrethroids and organo-chlorine insecticides, being lipophilic adsorb readily onto organic particles and are therefore likely to reach water bodies in an already adsorbed condition or are rapidly adsorbed and incorporated into sediment once they enter the water body. Aquatic sediments are the ultimate repository for a wide range of pollutants and although the toxicity of individual compounds in the adsorbed condition is generally regarded as being low there is little information on the pollutant loading of aquatic sediments or of the possible synergistic effects of multiple contamination. A major emphasis in the present phase of the study has therefore been the analysis of a range of organic sediments from river and land drain sediments and a bioassay of the toxicity of 3, previously analysed sediments was carried out using nymphs of the burrowing mayfly, *Ephemera danica*. Pesticide residues in water samples were also determined as were residues in the tissues of a range of invertebrates and fish from sites in East Anglia. A supplementary study on the persistence of pesticide in sediment under controlled temperature conditions was also carried out over a period of one year.

8.2 Methods.

8.2.1 Cypermethrin drift experiment.

The original intention had been to carry out an aerial spraying experiment but this had to be cancelled at a late stage. In order to avoid a total waste of the effort that had gone into the preparation a ground spraying experiment was substituted.

Spraying was carried out using tractor based equipment and trays of water containing the experimental animals. *Corixa* sp. nymphs were placed on short grass at distances downwind of the strip being sprayed of 1 metre, 5 metres, 7 metres, 10 metres and 15 metres. Since fewer specimens of *Notonecta* nymphs were available these were placed only at distances of 1 metre, 5 metres and 7 metres. The animals were exposed to spray drift arising from a single pass of the sprayer with a wind speed of 3 metres sec^{-1}. Numbers of dead animals were recorded at intervals over a 96 hour period. Controls were provided by maintaining animals in similar conditions but without exposure to spray drift.

8.2.2 Analysis of water samples.

Water samples were analysed using the methodology previously described by Pinder et al. (1990).

8.2.3 Estimation of pesticide residues in sediment.

Samples of sediment were obtained using a standard pond net, operated either from the bank or from a boat depending on accessibility. The net is operated so as to remove only the surface layers of sediment and samples from several positions within the sampling reach were combined and well mixed, to minimise the effects of local variability in pesticide concentrations. The sediment was then sieved, first through a 1 cm mesh to remove large molluscs, fragments of plant and lumps of clay, stones etc. and then through a 1 mm mesh before being stored in a 500 ml, glass tobacco jar. Sodium azide was added immediately, to prevent any breakdown of pesticide in the stored sample and the screw-on lids were lined with aluminium foil to avoid the possibility of pesticide becoming adsorbed onto the lid.

Within 2 to 3 days the samples were taken to the River Laboratory, for analysis, where they were immediately freeze dried to await extraction. Freeze drying was carried out over a period of 5 days, until the weight loss was $< 0.1\%$ in a 48 hour period. The dried sediments were then further sieved through a 0.5 mm brass sieve and well mixed once again to assure homogeneity. The samples were stored as necessary, in the dark at a temperature of 4°C in a nitrogen gas atmosphere.

Sediments were analysed using the following procedure:

(i). Analysis was carried out on 25 g sub-samples of dried sediment, extracted with pesticide grade acetone, with blanks being included with each batch of analyses. A two stage isolation, or clean up, procedure was followed, using adsorption chromatography with magnesium silicate, Florisil. The final stage of this procedure involves the elution of the pesticides from the adsorption column using 5% acetone in hexane. Three eluate fractions of 2 ml volume are collected and stored for subsequent analysis by glc with electron capture detector.

(ii). The extracts are concentrated by a factor of 10, using a stream of dry nitrogen i.e. 10 ml of each of the eluate fractions is reduced to 1 ml. The concentrated extracts are then analysed using glc / mass spectrometer detector, both in full scan mode, to identify major organic components, and also in selected ion monitoring mode (SIM). This mode is used for the confirmation of the glc/ecd results.

8.2.4 Stability trials.

Sediments were obtained from the lower part of the Swavesey drainage channel system on 12 December, 1990 when autumn spraying of crops was substantially completed, using the method described previously, but sodium azide preservative was not added in this case. The sample was taken to the River Laboratory the following day when it was transferred to a stainless steel tray and stored in the dark at 4°C. One litre capacity aluminium trays were prepared for the stability trial. They were washed in hot water and distilled water, followed by acetone and hexane. The lids were swabbed with acetone and hexane. On December 14 the sediment was thoroughly homogenised and then distributed between 6 trays. One tray was frozen immediately, one prepared for freeze drying and the remaining 4 trays placed in an incubator at a temperature of 15°C. The lids were crimped in place during storage.

The sample prepared for freeze drying was immediately dried and then stored at -20°C until finally sieved and sub-sampled for analysis of pesticides.

After 1 month's storage, one of the trays was removed from the incubator and its contents were freeze dried. On 22 January, 1991, the 2 dry samples (i.e. taken at the start of the trials and after 1 month storage) were sieved and sub-sampled for pesticide analysis. Further analyses were carried out after 6 months and 1 year from the date of sampling.

The samples were extracted and cleaned up using the procedures described previously and the 3 eluate fractions from each of the batches and from the blank are being were analysed as already described.

8.2.5 Bioassay using natural sediments.

Nymphs of the burrowing mayfly, *Ephemera danica* were selected as the test organism for these trials. *Ephemera* species feed on finely particulate organic particles that are obtained by filtering sediment in the manner described by Ladle & Radke (1990). Four nymphs were placed in each of 20, 10 cm diameter, glass jars containing 1 cm of washed river sand overlain by a depth of 4 to 5 cm of filtered pond water. Sediments from batches that had been previously analysed (Table 8.10) were added to the jars which were then maintained in the dark, in incubators at 12°C. Five jars received sediment collected from Wicken Lode, 5 received sediment from Reach Lode and 5 received sediment from the River Great Ouse near Godmanchester. The remaining 5 were supplied with sediment collected from a pond at the Institute's River laboratory site that was known from earlier analysis to be virtually free from pesticide contamination. The jars were examined periodically over a period of 4 months and the presence of any dead nymphs was noted.

8.3 Results

8.3.1 Cypermethrin drift experiment.

The number of live *Corixa* at intervals following possible exposure to cypermethrin spray drift is shown in Table 8.1. The number of animals at each distance at the start of the experiment varied between 9 and 14, as indicated in the Table.

Table 8.1 Numbers of live *Corixa* sp at intervals following possible exposure to cypermethrin drift as a result of ground-based spraying.

	Distance from spray (metres)			
Time (hrs)	1	5	10	15
0	10	9	14	12
0.5	10	9	14	12
1.5	10	9	14	12
3	10	9	13	12
4	10	9	13	12
5	10	9	13	12
6.5	10	9	13	12
10	10	9	12	12
21	8	9	12	12
32	7	9	12	12
42	7	8	12	12
55	6	8	12	12
65	6	8	9	12
77	4	4	6	9
96	4	4	6	9

No mortality was observed during the first few hours after spraying except, at 10 metres where 1 animal was found to be dead after 3 hours and a second after 10 hours. No further mortality occurred at this distance until 65 hours when a further 3 animals were dead. At 1 metre deaths were recorded from 21 hours onwards while the first mortality at 5 metres occurred after 42 hours. There was little difference in mortality between the 1 metre and 5 metre treatments.

The proportion of animals surviving over the course of the experiment is shown in Figure 8.1 and mortality at each distance, expressed as probits, are shown in Figure 8.2. The 96 hour LD50 was around 8 metres while extrapolation of the probit regression line suggests that zero mortality, the absolute safe distance would have been about 28 metres.

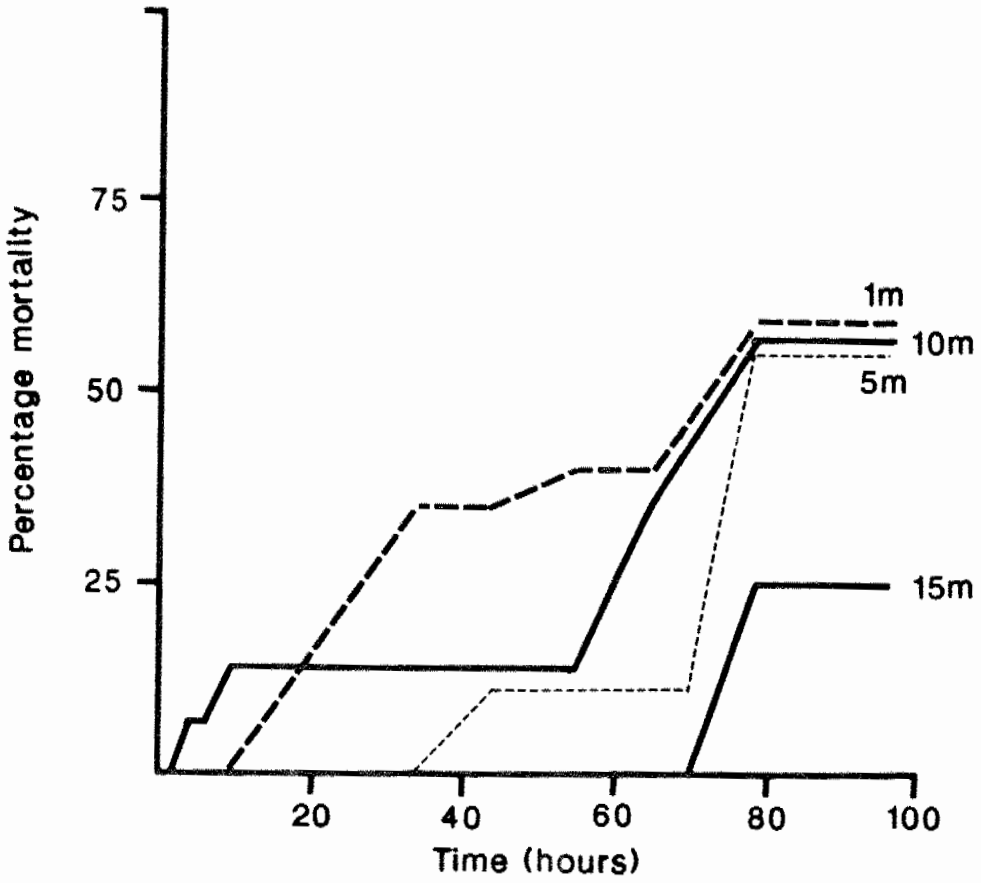

Figure 8.1 Proportion of dead *Corixa* at intervals following exposure to cypermethrin drift at distances of 1, 5, 10 and 15 metres from sprayer.

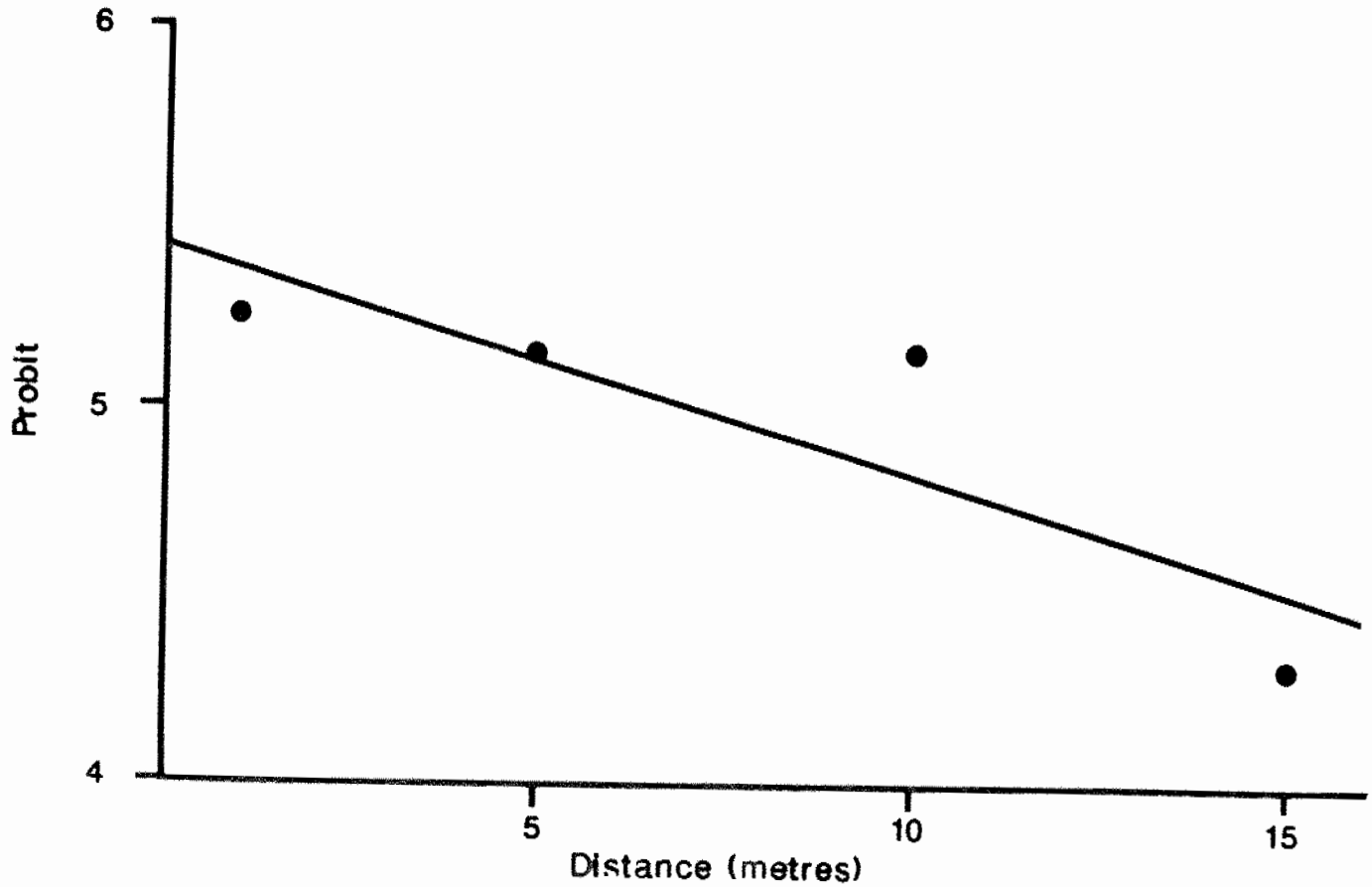

Figure 8.2 Proportion (probits) of *Corixa* dead 96 hours after exposure to cypermethrin drift at distances of 1, 5, 10 and 15 metres, with fitted regression line.

Table 8.2 shows the number of *Notonecta glauca* at the start of the experiment and the number surviving at intervals after spraying. In this case targets were placed at 1 metre, 5 metres and 7 metres only. No mortality was noted at 7 metres, until 77 hours had elapsed, when 1 individual was found to be dead. Mortality was also relatively slight at the closer distances although the first deaths were recorded after 2.5 hours at 1 metre and 4 hours at 5 metres.

The proportions of dead *Notonecta* at intervals over the period of the experiment are shown in Figure 8.3. *Notonecta* was clearly more resistant to cypermethrin than *Corixa* with only about 30% of animals dead after 96 hours at both 1 and 5 metres. Only 1 animal died at 7 metres and for *Notonecta* 10 metres therefore appears to be an adequate safe distance. It is worth noting that *Notonecta* also proved to be relatively tolerant of deltamethrin contamination in the aerial spraying experiment (Chapter 7).

Table 8.2 Numbers of live *Notonecta* sp at intervals following exposure to cypermethrin drift as a result of ground-based spraying.

	Distance from spray (metres)		
Time (hrs)	1	5	7
0	13	14	16
0.5	13	14	16
1.5	13	14	16
3	12	14	16
4	12	13	16
5	12	13	16
6.5	12	13	16
10	11	13	16
21	11	13	16
32	11	13	16
42	11	13	16
55	11	13	16
65	11	11	16
77	10	10	15
96	10	10	15

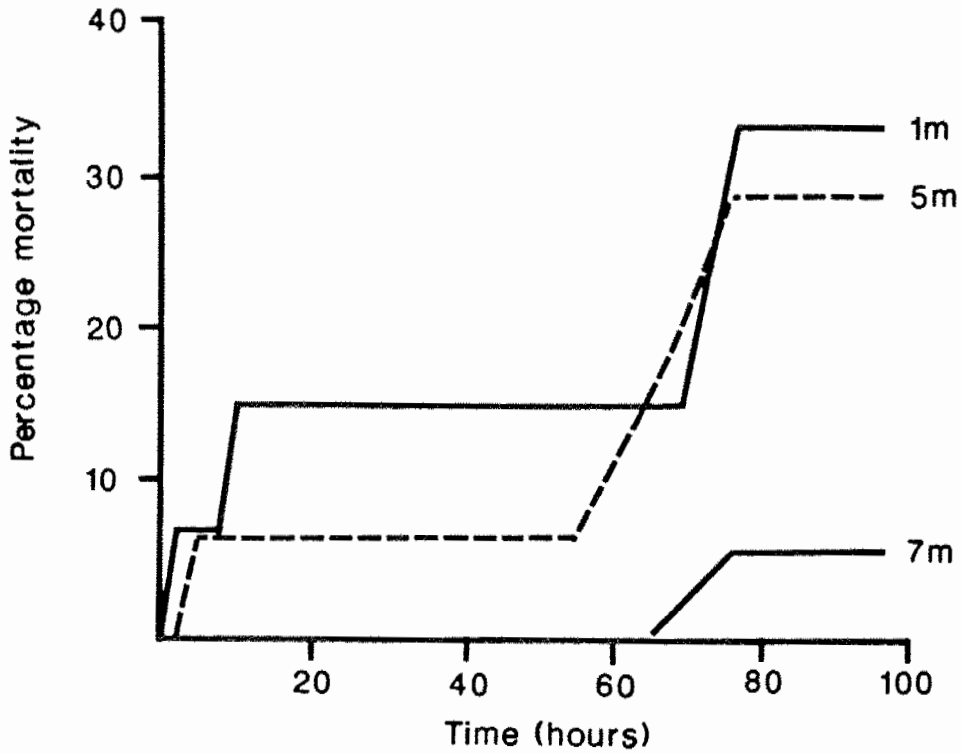

Figure 8.3 Proportion of dead *Notonecta* at intervals following exposure to cypermethrin drift at distances of 1, 5, and 7 metres from sprayer.

8.3.2 Pesticide residues in water samples.

Samples of water were taken for analysis from 7 sites associated with the Great Ouse system. These were, Wicken Lode and Reach Lode, on the Ely Ouse system and Lees Brook, and the River Great Ouse at Brampton, Hartford, Godmanchester and Needingworth as well as a marina at Needingworth. Analyses were carried out for the following organo-chlorine and synthetic pyrethroid insecticides.

α BHC, Lindane, Heptachlor, DDE, Dieldrin, Endrin, TDE, DDT, cis-Permethrin, trans-Permethrin, Cypermethrin (4 isomers), Fenvalerate (2 isomers) and Deltamethrin.

Of these DDE, Dieldrin, TDE, and isomer 2 of cypermethrin were not detected in any of the water samples.

Results of the analyses for individual samples are shown in Tables 8.3 - 8.10.

Table 8.3 Results of water sample analysis, 1- Wicken Lode.

Pesticide	Concentration ng l^{-1}	Std Dev.	Mass Spectrometer Result
αBHC	1.1	0	BDL
Lindane	1.6	0.1	C
cis Permethrin	40	1	C;42
Cypermethrin 1	11	_	BIC
Cypermethrin 3	3	_	ND
Deltamethrin	3	_	BDL

BDL = below detection limit
C = confirmed - base ion + confirmation
ND = not detected
BIC = Base-ion confirmed only

Table 8.4 Results of water sample analysis, 2- Reach Lode.

Pesticide	Concentration ng l^{-1}	Std Dev.	Mass Spectrometer Result
αBHC	1.1	0	BDL
Lindane	5.4	0.1	C
Endrin	2.1	1.8	ND
c-Permethrin	16	2	ND
Cypermethrin 3	20	21	ND
Cypermethrin 4	13	4	ND

Table 8.5 Results of water sample analysis, 3- Lees Brook.

Pesticide	Concentration ng l^{-1}	Std Dev.	Mass Spectrometer Result
αBHC	3.6	0.3	BIC
Lindane	19.6	0	C
Endrin	1.2	_	ND
Cypermethrin 1	103	_	BIC
Cypermethrin 3	26.4	_	ND
Fenvalerate 1	4.8	_	ND
Fenvalerate 2	1	_	ND

Table 8.6 Results of water sample analysis, 4- Brampton.

Pesticide	Concentration ng l^{-1}	Std Dev.	Mass Spectrometer Result
αBHC	3.7	0.4	BIC
Lindane	20.7	0.4	C
Endrin	1.0	0	ND
DDT	8.6	0.8	ND
Cypermethrin 1		_	Trace

Table 8.6 Results of water sample analysis, 5- Hartford.

Pesticide	Concentration ng l^{-1}	Std Dev.	Mass Spectrometer Result
αBHC	2.2	0	BIC
Lindane	13.4	0.1	C
Endrin	1	-	ND
DDT	8.6	0.8	ND
cis-Permethrin	8.8	5	ND
Cypermethrin 1	111	1	BIC
Cypermethrin 3	15.4	1.1	ND

Table 8.7 Results of water sample analysis, 6- Godmanchester.

Pesticide	Concentration ng l^{-1}	Std Dev.	Mass Spectrometer Result
αBHC	2.0	0.7	BIC
Lindane	19.1	0.3	C
Heptachlor	0.6	0	ND
Dieldrin	4.3	0.3	ND
Endrin	1.1	0.1	ND
c-Permethrin	6	-	ND
Tr-Permethrin	1	-	ND
Cypermethrin 1	} Problems in integrating chromatogram		
Cypermethrin 3	}		
Cypermethrin 4	}		
Fenvalerate 1	}		
Fenvalerate 2	}		
Deltamethrin	}		

Table 8.8 Results of water sample analysis, 7- Needingworth, river.

Pesticide	Concentration ng l^{-1}	Std Dev.	Mass Spectrometer Result
αBHC	1.3	1.6	BIC
Lindane	19.5	1.1	C
Endrin	1.5	0.3	ND
c-Permethrin	30.5	5	ND
Tr-Permethrin	2.7	1.1	ND
Cypermethrin 1	2200 *	210	C; 102

* ECD is an overestimation, because of poor integration of the peak.

Table 8.9 Results of water sample analysis, 6- Needingworth, marina.

Pesticide	Concentration ng l^{-1}	Std Dev.	Mass Spectrometer Result
αBHC	3.6	0.2	BIC
Lindane	14.3	0.8	C
Cypermethrin 3	35	2	ND
Fenvalerate 1	24	5	ND

The generally low concentrations found in water are to be expected because of the propensity, which all of these compounds have, to be readily adsorbed onto organic sediment particles. They and are generally similar to the range of values reported by other sources, such as the NRA. αBHC and Lindane were found in all samples and cypermethrin occurred in most. the occurrence of other compounds was more sporadic.

8.3.3 Pesticides in sediment.

Samples of sediment for analysis were taken from 4 of the sites from which water samples were analysed, Wicken Lode, Reach Lode, River Ouse at Godmanchester and River ouse at Brampton. An additional analysis was carried out on sediment from the lower part of the Swavesey drainage system, near Needingworth.
The pesticides for which analyses were carried out were as for water samples with the addition of Simazine and Phosalone. Heptachlor, Endrin, isomer 2 of cypermethrin and Fenvalerate were not detected.

The results of the analyses are shown in Table 8.10. in all cases strict criteria were applied to interpretation of the analysis. Relative retention times within ± 0.001 of the standard for organo-chlorines and within ± 0.002 for pyrethroids wre demanded. A total of 14 compounds, including isomers were detected. The DDT derivatives TDE and DDE were found at all sites, as was Deltamethrin, while α BHC, Lindane, Simazine and Dieldrin were found at 4 of the 5 sites. DDT was found to be present at 2 river

sites, in one case, Brampton, at the relatively high concentration of 20.8 μg kg^{-1}. Total pesticide concentrations ranged from 19μg kg^{-1} in the Swavesey sediments to 88.9 μg kg^{-1} in the river at Brampton. The concentration in Wicken Lode was only half that in the adjacent Reach Lode.

Figure 8.10 Concentrations of pesticides in river and land drain sediments, μg kg^{-1} dry wt.

Pesticide	Wicken Lode		Reach Lode		Brampton R.Ouse		Godman. R.Ouse		Swavesey Drain	
	ECD	MSD	ECD	MSD	ECD	MSD	ECD	MSD	ECD	MSD
α BHC	2.0		0.2		1.9		0.6		ND	
Lindane	2.3	C	1.9		1.0		2		ND	
Simazine	-	1.8C	ND		-	18C	0.7	C	1.9	C
DDE	2.3	C	1.8	C	5.2	C	2	C	3.1	C
Dieldrin	0.8		5.3		ND		8.4	C	6.8	C
TDE	3.1	C	3	C	4.8	C	3.5	C	3	C
DDT	ND		ND		20.8	C	3.2	C	ND	
Phosalone	1.3		ND		3.4		ND		ND	
c-Permethrin	Tr		D	2.3	ND		ND		ND	
tr-permethrin	9.2	C	ND		ND		10		ND	
Cypermethrin-1	ND		16.5	C	ND		ND		2.1	C
Cypermethrin-3	ND		ND		D	27.3C	ND		ND	
Cypermethrin-4	ND		D	7 C	ND		ND		ND	
Deltamethrin	Tr		5.9		6.4		2.1		2.1	

Tr = Trace detected
D = Detected but could not be quantified because of interference
C = Confirmed by mass spectrometry
ND = Not detected by GLC with electron capture detector (ECD).

8.3.4 Pesticide stability trials.

The initial analysis of the sediment obtained from the Swavesey drainage system was disappointing in that only 2 pesticides were present in sufficiently high concentrations for a stability trial to be carried out. One of these was Dieldrin and the other was Simazine. This is in spite of the sample having been taken in early December, when the autumn spraying was expected to have taken place and pyrethroids were expected to be present. However the autumn of 1990 was extremely dry and emergence of autumn sown crops was consequently much delayed, along with the associated spraying programme.

The results of the analyses of Simazine are shown in Figure 8.4. Initial concentrations of simazine in the Swavesey sediments samples were low at about 1µg kg^{-1}. After 1 month the measured concentration was almost 4µg kg^{-1} while about 11µg kg^{-1} was estimated after 6 months. However, as Figure 8.4 shows, the standard deviations for the analyses were large and differences between means were not significant. Simazine was not detectable after 12 months, using the MSD (Mass selective detector but a peak with an acceptable retention time was detected using the Nitrogen phosphate detector (NPD). This gave an estimated concentration of 8 µg kg^{-1}. as a result of the erratic nature of the data it was not possible to estimate a rate of die-away for simazine under the experimental conditions.

Good data were obtained for dieldrin (Figure 8.5). The initial concentration was 29.1 µg kg^{-1} (SD± 2.5) and this declined progressively with time (Figure 8.5). Most of the dieldrin had disappeared after one year. The half life was estimated from the regression of log concentration on time (Figure 8.6) as 73 days (SD ±9).

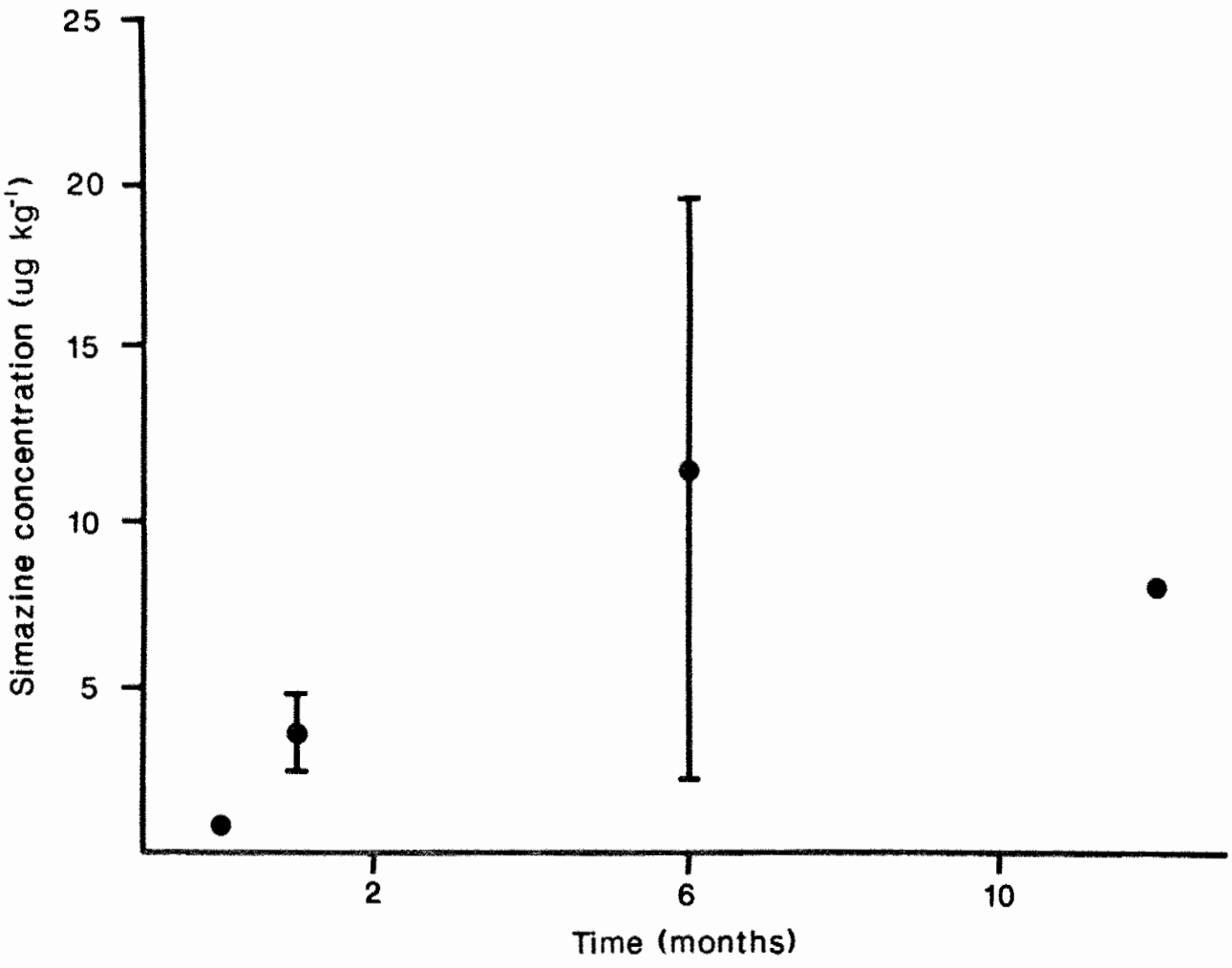

Figure 8.4 Results of Simazine die-away experiment. (Vertical bars indicate standard deviations).

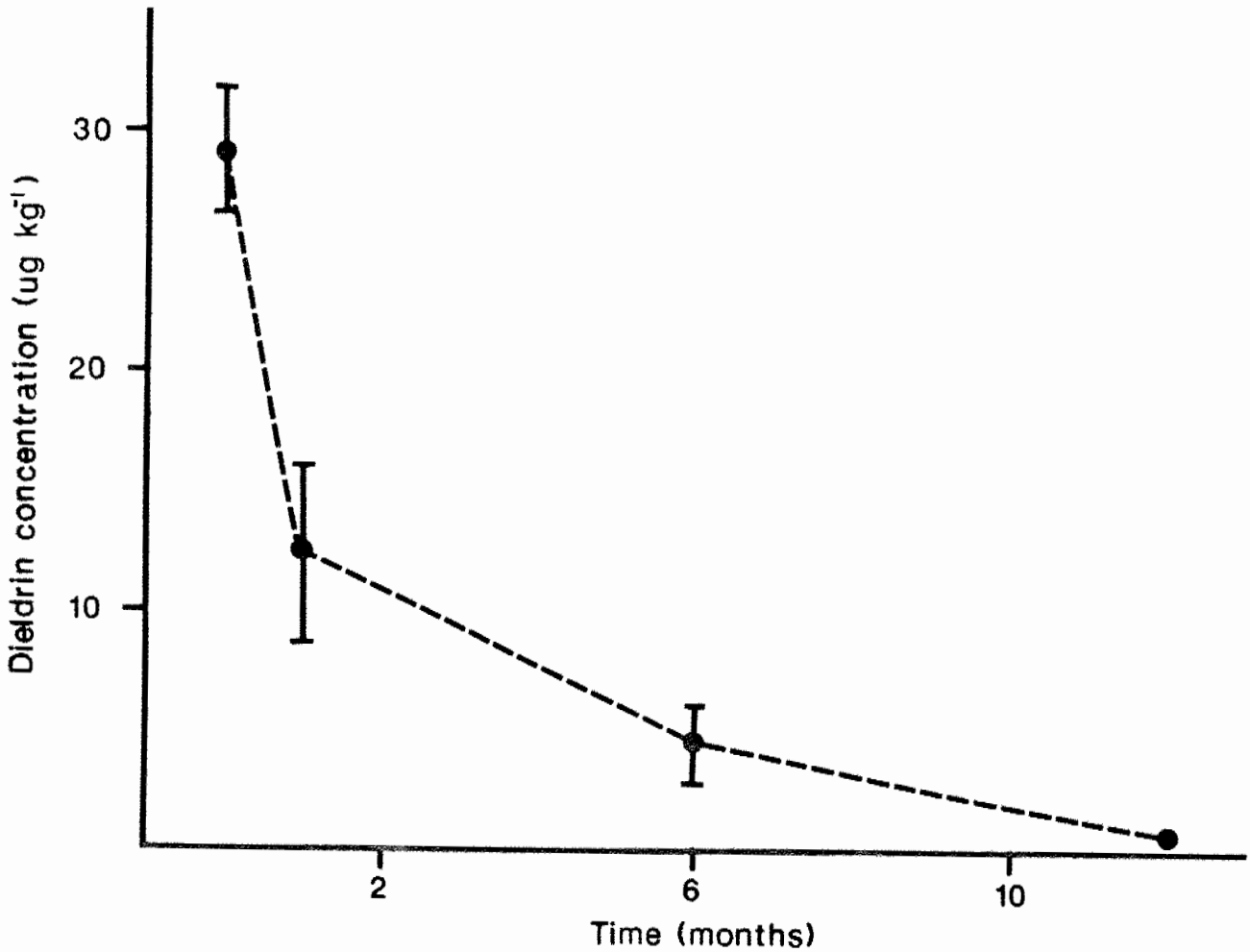

Figure 8.5 Die-away of dieldrin in sediment.
(Vertical bars indicate standard deviation).

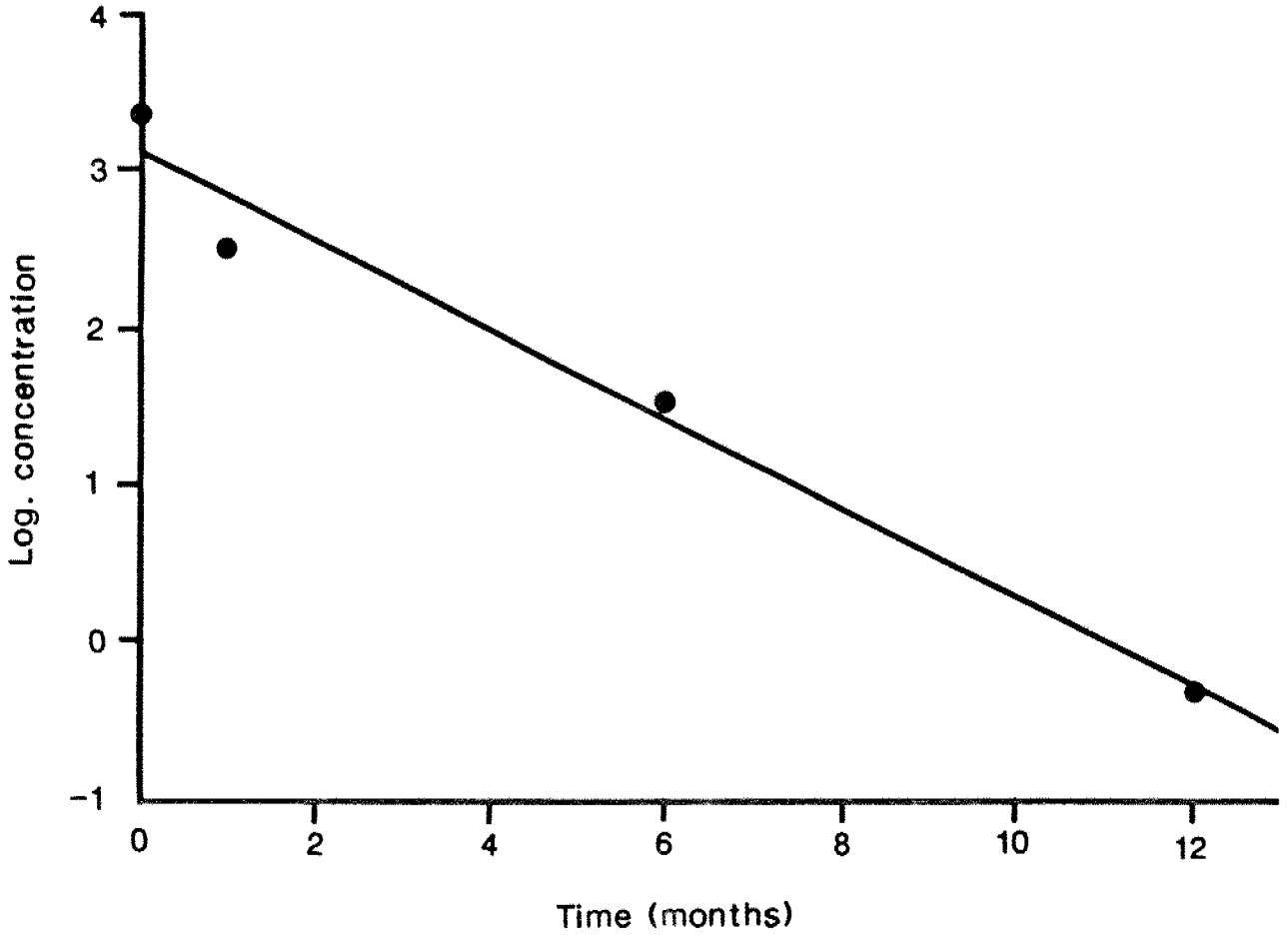

Figure 8.6 Log concentration of dieldrin in sediment over a one year period.

8.3.4 Pesticides in tissues of aquatic animals.

Analyses were carried out on the tissues of 5 species of invertebrate, 2 damselflies and 3 molluscs and 5 species of fish from the River Great Ouse and Lees Brook, near Godmanchester. The results of analyses of sediment and water from this same area are shown in Table 8.7 and 8.10. Dieldrin was detected in every sample at concentrations ranging from 9.3 μg kg^{-1} wet weight in 3 to 4 year old *Anodonta* (Mollusca) to 90.4 μg kg^{-1} wet weight a nymph of the damselfly, *Coenagrion* sp. Although Lindane occurred commonly in water and sediment samples it was not detected in any of the invertebrate tissues, although it did occur, at concentrations of between 6.0 and 15.9 μg kg^{-1} wet weight in all 5 fish. DDE also occurred in all of the fish and 2 of the invertebrates, while DDT was detected in only 1 of the fish, a chub from Lees Brook. TDE, Heptachlor, Permethrin and Fenvalerate were also detected in a small number of specimens. α-BHC, Endrin, Cypermethrin and Deltamethrin were not detected in any of the tissue samples, in spite of occurring regularly in water and sediment samples. Full details of the analyses are shown in Table 8.11. Species codes in the Table are as follows:-

1. *Calopteryx* sp (damselfly nymph), Great Ouse, Godmanchester;
2. *Coenagrion* sp (" "), " " " ;
3. *Unio tumidus* (bivalve mollusc), " " " ;
4. *Anodonta* sp. (" "), " " " ;
5. *Unio pictorum* (" "), " " " ;
6. *Gobio gobio* (Gudgeon), " " " ;
7. *Perca fluviatilis* (Perch), " " " ;
8. *Alburnus alburnus* (Bleak), " " " ;
9. *Rutilus rutilus* (Roach), " " " ;
10. *Leuciscus cephalus* (Chub), Lees Brook, Godmanchester.

Table 8.11 Concentrations of pesticide in animal tissues, μg kg^{-1} wet weight. (See above text for interpretation of species codes).

Species code	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.
Lipid (% w/w)	1.96	0.8	0.78	0.22	0.45	0.88	1.6	4.5	1.13	2.61
Pesticide										
Lindane	ND	ND	ND	ND	ND	6.0	9.7	15.9	7.2	11.1
Dieldrin	22.5	90.4	17.0	9.3	10.9	11.5	23.3	34.6	12.7	25.1
DDT	ND	ND	ND	ND	ND	ND	ND	ND	ND	11.4
DDE	ND	ND	6.5	ND	3.6	13.5	18.0	42.8	9.8	15.3
TDE	ND	ND	ND	ND	ND	ND	10.8	17.3	ND	ND
Heptachlor	ND	ND	ND	ND	ND	ND	ND	ND	ND	2.6
Permethrin	ND	ND	ND	ND	ND	12.4	ND	ND	21.0	ND
Fenvalerate	ND	8.7	ND	Tr	ND	ND	ND	19.0	ND	ND

ND = not detected; Tr = trace.

Most pesticide would be expected to be associated with fatty tissue and a close correlation in fact exists between the pesticide concentration in fish and their lipid contentPesticide concentrations expressed as μg kg^{-1} of lipid are given in Table 8.12. The concentration of Dieldrin expressed in this way ranged from 11337.2 μg kg^{-1} in *Coenagrion* to 754.5 in bleak, *Alburnus alburnus*.

Table 8.12 Pesticide content of fish and invertebrates, expressed as μg kg^{-1} of lipid. (Species codes as in Table 8.11)

	Pesticide							
	Lind.	Dield.	DDT	DDE	TDE	Hepta.	Perm.	Fenval.
Species code	Concentration μg kg^{-1} lipid							
1	ND	1147.1	ND	ND	ND	ND	ND	ND
2	ND	11337.2	ND	ND	ND	ND	ND	1090.1
3	ND	2182.5	ND	833.3	ND	ND	ND	ND
4	ND	4137.9	ND	ND	ND	ND	ND	ND
5	ND	2441.9	ND	814.0	ND	ND	ND	ND
6	679.4	1306.6	ND	1541.8	ND	ND	1444.1	ND
7	604.8	1503.5	ND	1123.3	674.0	ND	ND	ND
8	347.1	754.5	ND	935.6	377.3	ND	ND	415.0
9	635.9	1122.2	ND	860.3	ND	ND	1851.6	ND
10	425.0	962.5	437.5	587.5	ND	100.0	ND	ND

8.3.5 Bioassay of sediment using *Ephemera danica* nymphs.

Samples of the macroinvertebrate fauna from Wicken Lode revealed the presence there of a population of the burrowing mayfly, *Ephemera vulgata*. This observation is of interest since this genus of mayfly is not found in any of the adjacent lodes and is generally very scarce throughout the rivers and drains of the region. A further comparison of the fauna of Wicken Lode and the adjacent Reach Lode (Table 8.12 showed that they were otherwise very similar, although the indices of biological water quality, BMWP (Biological Monitoring Working Party) score and ASPT (Average Score per Taxon) were slightly higher in Wicken than in Reach Lode. Nymphs of *Ephemera* feed on fine particles of organic sediment. The total load of pesticide in the sediment of Reach Lode was twice that in Wicken Lode (Table 8.10). These observations led to the hypothesis that *Ephemera* was absent from Reach Lode as a result of the higher concentration of pesticide there.

Nymphs of the closely related species *Ephemera danica*, collected from a site where they were abundant and where pesticide levels were relatively low were used for the experiment in preference to *Ephemera vulgata*. Nymphs were provided with sediment from 4 sites with differing pesticide loads (see Table 8.10 for analysis) but no significant differences in mortality occurred over a period of 4 months. The hypothesis was therefore not supported.

Table 8.12 Macro-invertebrate taxa recorded from Wicken Lode and Reach Lode.

		Reach Lode	Wicken Lode
MOLLUSCA			
	Bivalvia		
	Sphaeriidae	+	+
	Gastropoda		
	Valvatidae	+	-
	Hydrobiidae	+	+
	Lymnaeidae	+	+
	Physidae	+	-
	Planorbidae	+	+
PLATYHELMINTHES			
	Turbellaria		
	Dendrocoelidae	+	-
ANNELIDA			
	Oligochaeta	+	+
	Hirudinea		
	Piscicolidae	+	-
	Glossiphonidae	-	+
	Erpobdellidae	+	-
ARTHROPODA			
	Crustacea		
	Gammaridae	+	+
	Asellidae	+	+
	Ostracoda	+	+
	Arachnida		
	Hydrachnellae	-	+
	Insecta		
	Ephemeridae	-	+
	Caeniidae	+	+
	Baetiidae	+	+
	Coenagriidae	+	+
	Sialiidae	+	+
	Molannidae	+	-
	Leptoceridae	+	+
	Polycentropidae	+	+
	Limnephilidae	+	+
	Haliplidae	-	+
	Dytiscidae	-	+
	Gyrinidae	-	+
	Elmidae	-	+
	Notonectidae	+	+
	Corixidae	+	+
	Chironomidae	+	+
Total taxa		24	25
Scoring taxa		23	23
BMWP Score		107	112
ASPT		4.65	4.89

8.4 Discussion

Pesticide contamination of water bodies may occur from a wide variety of sources other than from spray drift. For example, through erosion of soils containing adsorbed pesticide, through industrial effluent and sewage, as well as through direct contamination through accidental spillage. Atmospheric fall out may also be a significant source of contamination. Low level contamination of rainfall by pesticides has been demonstrated in all parts of the world. In a recent study Larsson et al (1991) considered that the source of PCBs, DDT DDE and Lindane in eels (*Anguilla anguilla*) in a lake in southern Scandinavia was most likely to be the atmosphere. Furthermore they suggested that release of these pollutants from fatty tissues during the long migration to the spawning grounds could account for the decline in eel populations in northern Europe during recent decades.

It is probable that sources other than drift account for most of the pesticides that are found in water bodies. Field experiments have frequently shown that the amount of deposition of spray drift onto water surfaces may be considerably less than might be expected in theory. In ground spraying experiments with cypermethrin, in which tractor mounted sprayers passed within 2 metres of ponds, 1 to 2 metres deep (Crossland et al 1982), deposition onto the water surface was 4 to 5 orders of magnitude less than onto the crop. No impact on aquatic populations was discernible, except for some species of air breathing insects, in which knockdown, but no mortality was observed. In another series of experiments, Shire and Bennett (1984) found that maximum deposition of cypermethrin onto field drains surrounding an area that was aerially sprayed was 6% of the application rate, giving a maximum concentration in the water of 0.03µg l^{-1}. Only some air breathing corixids and highly susceptible species of mite were reported to show any effects and depletions in their populations were only short term.

Nevertheless, modern pyrethroids in particular are a legitimate cause for concern in respect of their potential impacts on aquatic organisms. Coats et al (1989) point out that modern synthetic pyrethroids are designed to produce enhanced residual activity, through greater photostability, and through greater resistance to chemical and biological degradation. In addition they are exceptionally toxic to many aquatic organisms at very low concentrations. For example 24 hour LC_{50}, for deltamethrin, cypermethrin, fenvalerate and permethrin in respect of a range of aquatic insects varied between 0.02 and 13 µg l^{-1}. Most LC_{50} values for insects and crustacea are in fact below 1µg l^{-1}.

Unusually, synthetic pyrethroids are more toxic to both insects and fish at low temperatures than at high temperatures. A variety of other factors also influence toxicity. These include differences in hardness and salinity of water, bio-availability, mode of uptake, rates of elimination and bio-concentration. These factors are discussed in some detail by Coats et al (1989) but a particular point of relevance to the present investigation is the influence of suspended solids. Pyrethroids, because they are

highly lipophilic, adsorb readily onto organic particles and in general, studies have shown that when incorporated into sediments their toxicity is greatly reduced. It has been demonstrated, for example, that fenvalerate LC_{50} were 40 times higher in channel catfish when the pesticide was first applied to soil particles than when introduced directly into clean water. Similarly, a concentration of cypermethrin that caused 100% mortality in rainbow trout in micro-filtered water did not cause mortality when water containing 14.5 mg l^{-1} of suspended solids was employed. However sediment-living invertebrates may accumulate relatively large concentrations of pesticide. *Chironomus tentans* larvae, for example accumulated significantly more permethrin when allowed to enter sediment than when held above the sediment surface.

Detritivorous aquatic insects often exhibit very high rates of feeding activity. Gut throughput rates of 30 minutes or less are not unusual and uptake of pesticide, either from dissolved or adsorbed sources is a possibility. However, pyrethroids have been shown to inhibit feeding activity in some cases and cuticular uptake may in fact be the more important route. After feeding for an hour and attaining high concentrations of fenvalerate in their bodies mosquito larvae became restless and stopped feeding (Coats et al., 1989).

Coats et al (1989) conclude their paper by stating that although the photostable pyrethroids are innately quite toxic to fish and aquatic arthropods there are many factors that influence the degree of hazard that they present in practice. In considering the results of the experiment on ground spraying by cypermethrin and the aerial spraying experiment described in Chapter 7 this should be kept in mind. These experiments demonstrate very clearly the potential toxicity of pyrethroids to insects and crustacea but they were designed to represent, in many respects, worst case situations. The impact of a similar spray drift event on field populations is likely to be much less, at least in the case of invertebrates that live fully submerged. There are several reasons for this, including the effects of water depth and presence of sediment. Experiments reported by Pinder et al (1990), and reexamined in the discussion of the results of the aerial spraying, in Chapter 7, consider the likely effect of adsorption of the pesticides onto sediment.

Numerous studies have noted an increased incidence of invertebrate drift in streams contaminated by pyrethroid drift. In the experiments carried out within the context of the present studies and in the earlier laboratory based studies, invertebrates were unable to drift. However, two effects were observed that are likely to increase the probability of individuals entering the drift in flowing water. The initial response of several species, though this was not quantified, was an increase in activity. This was noted for example in *Sigara dorsalis*, *Asellus aquaticus* and *Centroptilum pennulatum* in the aerial spraying experiment, following exposure to spray drift. It was also noted in *A. aquaticus* in laboratory experiments (Pinder et al , 1990). In the latter case animals exposed to

higher concentrations of pyrethroid frequently crawled above the water surface and came to rest on the sides of the beaker in which they were maintained.

This phase of behaviour was followed by a proportion of the animals becoming partially immobilised. In this condition in flowing water they would not only have an increased likelihood of being passively swept into the stream drift but would also be prone to being eaten by fish and other invertebrates. The tendency to become immobilised was particularly noticeable in the case of *Asellus aquaticus* which frequently remained in this condition for several days before dying or more rarely, recovering. This behaviour may be useful in determining whether significant spray contamination of a site has occurred up to several hours, or even a few days, after the event, especially in still waters, where the animals could not drift away.

Water bodies are inevitably the final repository for many pollutants and the analyses of water and sediment samples, although mainly restricted to pyrethroids and organochlorine pesticides, have shown the variety of pesticides that may be present. As expected of lipophilic, relatively insoluble chemicals, concentrations in water were generally very low.

It is particularly disturbing to note the widespread occurrence in sediments of organochlorine compounds that have been withdrawn from the market and banned from use for some years, such as DDT and Dieldrin. The laboratory experiment on the persistence of Dieldrin shows that it is relatively quickly broken down, at least under the prevailing experimental conditions. This leads to the conclusion that continuing recontamination of water by dieldrin is probably still occurring, though from what source is uncertain. Similarly, DDT would be expected to be degraded into DDE and TDE relatively quickly yet in one sample a concentration of almost 21 $\mu g\ kg^{-1}$ of DDT was detected.

The potential for bio-accumulation of pyrethroids appears to be very small. Only 2 such pesticides, Permethrin and Fenvalerate were detected in animal tissues and then only in odd individuals. However, Fenvalerate was not detected in any water or sediment analyses and some bioaccumulation may be occurring in this case.

Neither cypermethrin nor deltamethrin were found in any of the animal tissues. The absence of the former is particularly noteworthy since it occurred, often at relatively high concentrations, in virtually all water and sediment samples.

In contrast dieldrin was found in the tissues of all the invertebrates and all of the fish that were analysed. Other organochlorine pesticide residues also occurred widely in fish tissues but only exceptionally in invertebrate tissues. This is surprising, particularly in the case of the bivalve molluscs in which residues would have been expected to be present.

In view of the wide range of pollutants, only a few of which have been reported in this study, that may be present in aquatic sediments and the multiplicity of combinations and synergistic effects that are possible, the use of bioassay to assess the toxicity of sediments appears to be a sensible, perhaps the only sensible, option. Although, in the present case no effect of sediment chemistry on the mayfly *Ephemera danica* was detected the possibility of long term, chronic effects, especially in view of the annual or in some cases biennial life cycles of *Ephemera* species, cannot be ruled out. Whole life cycle studies are highly desirable and the further development of this type of bioassay is highly desirable to identify useful indicator organisms and develop methods. It is interesting to note that laboratory experiments with another species of burrowing mayfly, *Hexagenia rigida*, in which nymphs were exposed to sediment, contaminated with permethrin up to 8 days previously produced substantial mortality (Friesen et al. 1983).

The subject of sediment toxicity testing was reviewed by Burton (1991). He points out that every test site is a unique ecosystem and the toxicity of the sediment will be a function of both independent and integrative natural factors, such as patch dynamics, and their biological, physical and chemical relationships with the contaminants (as well as unknown contaminant interactions). As he rightly points out the use of multiple assays improves the probability of detection. He also correctly states that the detection of toxicity in the laboratory must be validated by removing the possible effect of collection and laboratory manipulation and then relating the effect to an ecosystem perturbation. Of equal importance is determining the appropriate level of sensitivity, that is a level that is ecologically relevant. This task, he states is difficult, if not impossible at this time, and will likely always be a point of debate. Clearly the subject of sediment toxicity testing is in need of development. Equally as more persistent pyrethroids are developed and marketed it will be increasingly important to determine their fate in freshwater and assess their true effects on aquatic organisms. These effects cannot necessarily be accurately predicted from knowledge of their physical and chemical properties alone, and bioassay using indicator organisms and "natural sediments", has the potential to be very useful.

8.5 References

Burton, G.A. (1991). Assessing the toxicity of freshwater sediments. Environmental Toxicology and Chemistry. 10, 1585-1627.

Coates, J.R. (1989). Toxicology of synthetic pyrethroids in aquatic organisms; a review. Environmental Toxicology and Chemistry 8, 671-679.

Crossland, N.O., Shires, S.W. & Bennett, D. (1982). Aquatic toxicology of cypermethrin. III. fate and biological effects of spray drift deposits in fresh water adjacent to agricultural land. Aquatic Toxicology 2, 253-270.

Davis, B.N.K. (Editor) (1990). Pesticide drift and impact. Final Report to Nature Conservancy Council and Department of the Environment, March 1990.

Friesen,M.K., Galloway, T.D. & Flannagan, J.F. (1983). Toxicity of the insecticide permethrin in water and sediment to nymphs of the burrowing mayfly *Hexagenia rigida* (Ephemeropter: Ephemeridae). Canadian Entomologist, 115, 1007-1014

Ladle, M. & Radke, R. (1990). Burrowing and feeding behaviour of the larva of *Ephemera danica* Müller (Ephemeroptera: Ephemeridae). Entomologists Gazette 41, 113-118.

Larsson, P., Hamrin, S. & Okla, L. (1991). Factors Determining the Uptake of Persistent Pollutants in an Eel Population (*Anguilla anguilla* L.) Environmental Pollution 69, 39-50.

Pinder, L.C.V., House, W.A. & Bass, J.A.B. (1990). Effects of insecticide drift on aquatic invertebrates. In Davis (Ed) Pesticide drift and impact, Final Report to NCC and DoE.

Shires, S.W. & Bennett, D. (1985) Contamination and Effects in Freshwater Ditches Resulting from an Aerial Application of Cypermethrin. Ecotoxicology and Environmental Safety, 9, 145-158.

Williams, C.T., Davis, B.N.K., Marrs, R.H. & Osborn, D. (1987). Impact of pesticide drift. Unpublished report to NCC.