Bull. Environ. Contam. Toxicol. (1990) 44:790–797
© 1990 Springer-Verlag New York Inc.

Bulletin of Environmental Contamination and Toxicology

Effect of Sediment Contact and Uptake Mechanisms on Accumulation of Three Chlorinated Hydrocarbons in the Midge, *Chironomus riparius*

Deanna M. Fry and S. W. Fisher*

Environmental Biology Program, 103 Botany and Zoology Building, 1735 Neil Avenue, The Ohio State University, Columbus, Ohio 43210, USA

Chlorinated hydrocarbons (CHCs) are major contaminants of bottom sediments in many freshwater systems (Swartz and Lee 1980; Knezovich *et al.* 1986). The behavior and availability of sediment-sorbed compounds arouse much controversy due to the potential impact these contaminants could have on the ecosystem if they were to get into the food chain.

Benthic organisms are at great risk from sediment-sorbed contaminants since they inhabit bottom sediments. The primary medium from which uptake occurs is believed to be sediment interstitial water (Adams *et al.* 1985; Oliver 1987) and the uptake process is generally considered to be active (Landrum 1988; Murphy and Murphy 1971).

In this investigation, uptake of sediment-sorbed 5, 5', 6-trichlorobiphenyl (PCB), p,p'-DDE and PCP by the midge (*Chironomus riparius*) was examined under 3 conditions. Uptake from direct contact with contaminated sediment (sediment + water) was compared to uptake levels by the midge when it was screened from contaminated sediment contact (screened sediment) and to uptake in dead organisms exposed to contaminated sediment (passive).

MATERIALS AND METHODS

Fourth instar midge larvae, *Chironomus riparius*, were obtained from existing cultures maintained in the laboratory according to the method of Estenik and Collins (1979). [^{14}C]PCB (sp act=7.3 x 10^7dpm/mg) and [^{14}C]DDE (sp act=11.7 x 10^7dpm/mg) were obtained from Sigma Radiochemical Co. (St. Louis, MO) and [^{14}C]PCP (sp act=8.7 x 10^7dpm/mg) was obtained from California Bionuclear Corp. (San Diego, CA).

For all tests, 10 μL, 50 μL and 25 μL of DDE, PCB and PCP, respectively, were each applied to 5 g of Olentangy River sediment (3% OC) with 10 mL

*Correspondence and reprint requests

of distilled water. The mixture was centrifuged three times and allowed to sit for 24 hr. The mixture was then centrifuged once more, the supernatant poured off and the sediment with the compound was placed in a 1-L beaker with 500 mL of soft (40-48 mg/L) standard reference water (EPA 1975) adjusted to pH 6. The beaker with water and sediment settled for 24 hr after which 50 test organisms were added. In the screened-sediment tests the organisms were first placed in a screen apparatus which was then placed into the 1-L beaker; with the passive uptake tests, the organisms were first treated with a lethal dose of parathion before addition to the beaker.

The test beakers were held at 25°C on a diel photoperiod of 14 hr. After 24 hr, the organisms were separated, rinsed and analyzed by scintillation counting according to the method of Fisher and Wadleigh (1986). One mL water samples were taken initially and after 24 hr and were analyzed by the same method. At 24 hr the water was analyzed by the method of Fisher (1985) using hexane as the extraction solvent for DDE and PCB, and anhydrous diethyl ether for PCP. Extraction efficiencies for all compounds were greater than 95%. Sediment was also analyzed after 24 hr by placing dried, weighed sediment in a 200 mL brown jar with 100 mL of extraction solvent (cyclohexane for DDE and PCB, ethyl acetate for PCP). The jars were covered and stirred intermittently for 2 wk after which the extract was separated by filtration from the sediment, and a 1 mL sample was taken for scintillation analysis. The residual sediment was dried at 30°C, weighed and a subsample was taken for scintillation analysis in 10 mL ^{14}C-cocktail (dioxane/naphthalene/PPO, 1000:100:5) for 5 min per vial in a Beckman LS 7000 counter. The data were analyzed by analysis of variance (Crisp, 1986). Data from relative distribution of total radioactivity were transformed by arcsin square root before analysis of variance.

RESULTS AND DISCUSSION

Contact with sediment had a direct effect on uptake by the midges. With all three compounds (Tables 1-3), the midges in treatment A (water + sediment) accumulated significantly more parent compound than the midges in treatment B (screened sediment). The midges accumulated an average of 10 times more parent compound when sediment contact was made.

With the compounds PCB and DDE, the midges accumulated significantly more parent compound by active uptake (A) compared with passive uptake (C). For these chemicals, there were no significant differences between passive uptake (C) and uptake when the midges were screened from the sediment (B).

Passive uptake of PCP by the midge appeared to be a crucial mechanism for uptake (Table 3). Passive uptake by the midges (C) was greater than

uptake when the organism was alive (A), although there was no significant difference between these two values. Passive uptake of PCP by the midge was significantly greater than uptake when the midges were screened from the sediment (B).

Table 1. The concentrations (μg/L) of DDE in the initial water, 24-hr water, midge, extracted sediment and residual sediment.

	DDE Concentration (μg/L)		
Source	**A***	**B***	**C***
Initial Water	0.13[a**] $\pm$0.04	0.12[a] $\pm$0.02	0.07[a] $\pm$0.04
24-hr Water	0.11[a] $\pm$0.02	0.08[a] $\pm$0.03	0.12[a] $\pm$0.01
Midge	111.57[b] $\pm$7.39	12.59[a] $\pm$0.95	15.90[a] $\pm$12.73
Sediment Extract	153.03[a] $\pm$7.77	144.93[a] +8.95	182.80[b] $\pm$8.40
Sediment Residual	3.93[a] $\pm$0.36	5.10[a] $\pm$2.74	4.76[a] $\pm$1.31

*A=Sediment + Water, B=Sediment + Water + Screen, C=Passive Uptake

**Within a source, μg/L values followed by the same letter are not significantly different.

Sediment was the primary sink for PCB and DDE in every treatment (Tables 1-2). With PCP, however, the majority of the compound was found in the organism, except for the screened sediment treatment (B), where the levels in the sediment extract and midge were very close. PCB and DDE are both neutral lipophilic compounds whose log K_{ow}'s are 6.72 (Thomann and Connolly 1984) and 5.69 (Verschueren 1983) respectively. PCP, on the other hand, is an ionizable compound whose sorption is dependent on pH (Knezovich *et al.* 1986). The hydroxyl proton on PCP is dissociated at pH's greater than 4.8 and at pH 9.0 the compound is completely ionized (Kaiser and Valdanis 1982). These tests were run at pH 6. Therefore PCP was expected to predominate in the ionized form. In this form, PCP is not

lipophilic and would thus not be bound to the sediment in great amounts.

Table 2. The concentrations (μg/L) of PCB in the initial water, 24-hr water, midge, extracted sediment and residual sediment.

	PCB Concentration (μg/L)		
Source	**A***	**B***	**C***
Initial Water	0.30[a**] $\pm$0.05	0.16[a] $\pm$0.05	0.25[a] $\pm$0.00
24-hr Water	0.27[c] $\pm$0.03	0.07[a] $\pm$0.02	0.16[b] $\pm$0.03
Midge	370.40[b] $\pm$49.11	24.46[a] $\pm$3.01	56.19[a] $\pm$41.21
Sediment Extract	573.10[a] $\pm$52.77	649.67[a] $\pm$152.68	623.03[a] $\pm$35.36
Sediment Residual	18.27[a] $\pm$3.78	15.14[a] $\pm$9.37	36.20[b] $\pm$5.38

*A=Sediment + Water, B=Sediment + Water + Screen, C=Passive Uptake

**Within a source, μg/L values followed by the same letter are not significantly different.

The 24-hr water samples for DDE showed no significant difference between treatments. The levels of parent PCP and PCB in the 24 hr water samples, however, were significantly greater in treatment A (sediment + water), while treatment C (passive uptake) was significantly greater than treatment B (screened sediment). Karickhoff and Morris (1985) found pollutant transport of bottom sediment into the water column was increased 4-6 fold over a 90-d period. Larsson (1983) demonstrated a higher mobility of PCBs in aquatic systems when macroinvertebrates were present. Levels in the water column at 24-hr were highest when the midges were alive and in direct contact with the sediment. Although the midges were not alive in treatment C and bioturbation should have been at a minimum, when the midges were added to the system the sediment was slightly stirred to enhance midge cuticular contact with sediment interstitial water. This action may have suspended particulate material in the water column which was present at the 24 hr

sampling. Otherwise, one would expect these levels to be statistically similar to the results when the midge was isolated from sediment contact.

Table 3. The concentrations (μg/L) of PCP in the initial water, 24-hr water, idge, extracted sediment and residual sediment in the three different treatment types.

	PCP Concentration (μg/L)		
Source	**A***	**B***	**C***
Initial Water	2.61[b**] $\pm$0.22	1.90[a] $\pm$0.26	2.12[a] $\pm$0.12
24-hr Water	2.42[c] $\pm$0.10	0.16[a] $\pm$0.04	1.66[b] $\pm$0.06
Midge	374.83[b] $\pm$26.68	36.61[a] $\pm$1.44	561.43[b] $\pm$41.21
Sediment Extract	51.40[a] $\pm$4.83	51.54[a] $\pm$12.77	42.11[a] $\pm$10.74
Sediment Residual	7.57[a] $\pm$1.16	3.97[a] $\pm$0.63	4.44[a] $\pm$0.66

*A=Sediment + Water, B=Sediment + Water + Screen, C=Passive Uptake

**Within a source, μg/L values followed by the same letter are not significantly different.

The midges accumulated a significant amount of compound when they were in direct contact with the sediment, therefore the compounds in the sediment were not unavailable for uptake. Midges are deposit feeders and are known to rework sediments by feeding and burrowing (Lee and Swartz 1980). Through such activity, midges not only ingest contaminated particles but expose themselves to interstitial water. They also mix the interstitial water with the overlying water. When the midges were isolated from the sediment, such activity was inhibited, and as a result, not only did midge uptake levels decrease but contaminant release into the water column decreased. One exception was DDE, for which the 24-hr water levels were actually similar to the initial water samples. Perhaps, DDE forms stronger bonds with the sediment particles, or the particulate material that was suspended quickly

settled into the surficial sediments. It is also possible that DDE sorbed to DOC in the water column and was unavailable for uptake.

Although passive uptake by the midges did not appear to be a primary mechanism for uptake with DDE and PCB, it did occur. It is important to point out that when using dead organisms to measure passive uptake, normal activities of a midge do not occur. When the midges rework the sediment they expose themselves to greater levels of contaminant. Therefore, our uptake values can be considered an underestimation of uptake because the normal activities of the midge which may increase its risk did not occur.

Passive uptake appeared to be important in midge accumulation of PCP. Trujillo *et al.* (1982) found the half life of PCP in midges to be 4.7 d and within the first 4.5 hr, the rate of depuration was very accelerated. Perhaps, the midges accumulated high levels of PCP because with the dead organisms, depuration did not occur and as a result, the midges may have accumulated elevated quantities of PCP. If this was not the case, however, then passive uptake of PCP by the midge would be one of the primary mechanisms for uptake.

Direct sediment contact increased uptake for all three compounds. Neutral lipophilic compounds sorbed to sediment and became available when organisms interacted with the sediment. When organisms were screened from sediment contact, not only did uptake levels decrease but desorption of the contaminant from the sediment into the water column decreased. For neutral lipophilic compounds, uptake occurs primarily as an active process. Accumulation of xenobiotics in benthic organisms from direct sediment contact is believed to occur by adsorption to the body wall or exoskeleton; and/or absorption through the integument, gills or other respiratory surfaces; and/or ingestion of sediment, food and water followed by absorption through the gut (Swartz and Lee 1980). Adsorption or absorption of DDE and PCB to the integument or exoskeleton of the midge appeared to be unimportant for accumulation because of the low passive uptake levels.

Direct sediment contact also increased the uptake of ionizable compounds in the midge. Although ionizable compounds do not readily form hydrophobic interactions with sediment, as the neutral lipophilic compounds do, PCP was present in the sediments where the organisms could accumulate it. When the organisms were screened from sediment contact, uptake levels decreased as did desorption of contaminant from sediment to the water column. Uptake of PCP in the midge was primarily a passive process. The ionized compounds appeared to have a great affinity for the body wall of the midge because the uptake levels of PCP were so high in the dead organism.

The variability between the two chemicals types (neutral lipophilic and ionizable) was clearly demonstrated. Despite the fact that PCP has a log K_{ow} of 5.01 (Verschueren 1983), it did not behave like a neutral lipophilic compound. The activity and fate of PCP, therefore was not predictable from its K_{ow} value. With DDE and PCB, on the other hand, their fate and behavior were much more predictable from this value.

The results from this study suggest that the benthic organism, *Chironomus riparius*, accumulates significantly more neutral lipophilic and ionizable compounds when it is in direct contact with bottom sediments. The results also indicate that passive uptake occurs with neutral lipophilic and ionizable compounds, but is only a crucial mechanism of uptake for the latter.

REFERENCES

Adams WJ, Kimerle RA, Mosher RG (1985) Aquatic safety assessment of chemicals sorbed to sediments. In: Caldwell R, Purdy R, Bahner R (eds) Aquatic toxicology and hazard assessment: seventh symposium. ASTM STP 854, p 429-453

Coffman WP, Ferrington LC (1984) Chironomidae In: An introduction to the aquatic insects of North America, second edition. Merrit RW, Cummins KW (eds) Kendall/Hunt Publishing Co. Iowa p 551-643

Fisher SW, Wadleigh RW (1986) Effects of pH on the acute toxicity of PCP in the midge *Chironomus riparius*. Ecotoxicol Environ Safety 11:1-8

Kaiser KLE, Valdanis I (1982) Apparent octanol water partition coefficients of pentachlorophenols a function of pH. Canad J Chem 66:2104-2110

Karickhoff SW, Morris KR (1985) Impact of tubificid oliogochaetes on pollutant transport in bottom sediments. Environ Sci Technol 19:51-56

Knezovich JP, Harrison FL, Wilhelm RG (1986) The bioavailability of sediment-sorbed organic chemicals: a review. Water Air Soil Poll 32:233-245

Landrum PF (1988) Toxicokinetics of organic xenobiotics in the amphipod. *Pontoporeia hoyi*: role of physiological and environmental variables. Aquat Toxicol 12:245-271

Larrson P (1983) Transport of ^{14}C -labelled PCB compounds from sediment to water and from water to air in laboratory models systems. Water Res 17:1317-1326.

Lee H, Swartz RC (1980) Biological processes affecting the distribution of pollutants in marine sediments, part II, biodeposition and bioturbation. In: Contaminants and sediments, vol 2, Analysis,chemistry and biology. Baker RA (ed). Ann Arbor Sciences Publishers, Ann Arbor, Michigan, p 555-606

Murphy PG, Murphy JV (1971) Correlations between respiration and direct uptake of DDT in the mosquito fish *Gambusia affinis*. Bull Environ

Contam Toxicol 6:581-588
Oliver BG (1987) Biouptake of chlorinated hydrocarbons from laboratory-spiked sediments and field sediments by oligochaete worms. Environ Sci Technol 21:785-790
Swartz RC, Lee H (1980) Biological processes affecting the distribution of pollutants in marine sediments, part II, accumulation, trophic transfer, biodegradation and migration. In: Contaminants and sediments, vol 2, analysis, chemistry and biology. Baker RA (ed). Ann Arbor Sciences Publishers, Ann Arbor, Michigan, p 533-553
Thomann RV, Connolly JP (1984) Model of PCB in the Lake Michigan trout food chain. Environ Sci Technol 18:65-71
Trujillo DA, Ray LE, Murray HE, Giam CS (1982) Bioaccumulation of pentachlorophenol by killfish (*Fundulus similus*). Chemosphere 11:25-31
Verschueren K (ed) (1983) Handbook of environmental data on organic chemicals. Van Nostrant Press, New York, New York p 434-436, 953-956, 1008-1013
Received May 15, 1989; accepted September 17, 1989