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Effects of Sediment Organic Carbon on Distribution of Radiolabeled Fluoranthene and PCBs among Sediment, Interstitial Water, and Biota

J. M. Brannon,[1] C. B. Price,[1] F. J. Reilly Jr.,[2] J. C. Pennington,[1] and V. A. McFarland[1]

[1]U.S. Army Engineer Waterways Experiment Station, 3909 Halls Ferry Road, Vicksburg, Mississippi 39180, USA and [2]AScI Corporation, 1720 Clay Street, Suite 3, Vicksburg, Mississippi 39180, USA

The relationship between concentrations of non-polar organic contaminants in sediments, sediment organic matter, and concentrations of these compounds in aquatic organisms has been an active research area. The objective of much of this work has been to develop equilibrium based prediction of bioaccumulation potential (organism concentration resulting from exposure to contaminated sediment) (McFarland 1984; McFarland and Clarke 1986; Rubinstein et al. 1987; McElroy and Means 1988; Bierman 1990; Ferraro et al. 1990). When equilibrium is assumed, bioaccumulation potential is a thermodynamic concept independent of rates of desorption, transport, uptake, or elimination (McFarland et al. 1989). McFarland (1984) related empirically derived equations for the relationship between K_{ow} and K_{oc} (Karickhoff 1981) and K_{ow} and bioconcentration factor (BCF). These equations permitted calculation of a theoretical accumulation factor (AF) describing proportional difference in equilibrium concentrations of non-polar organic chemicals in sediment organic carbon and organism lipid. In theory, the actual AFs for individual chemicals are not expected to differ greatly from the theoretical value of 1.72. Subsequent research has been undertaken to test the validity of this concept (Rubinstein et al. 1987; 1990; McFarland and Dorkin 1988; Ferraro et al. 1991; Young et al. 1991).

True biological and chemical equilibria are difficult, if not impossible, to achieve because organisms and sediments change over time and additional complications related to chemical properties of sediment contaminants arise (Bierman 1990). Furthermore, exposure conditions differ as a function of feeding modes for organisms (Bierman 1990; McElroy and Means 1988). The studies reported herein were designed to examine the relationship between sediment organic carbon, sediment interstitial water, and bioaccumulation of polychlorinated biphenyl (PCB) and polynuclear aromatic hydrocarbon (PAH) in two organisms having different feeding modes.

MATERIALS AND METHODS

Two experiments were conducted to 1) examine the chemical and biological dynamics in the bioassay system, and 2) examine the effect of organic carbon on contaminant uptake by organisms and on contaminant concentrations in interstitial water. Experiments were conducted using a sediment bioassay apparatus (Figure 1) similar to that developed by McElroy and Means (1988). This apparatus was selected because of small size (1 L),

Send reprint requests to J. M. Brannon at the above address.

simplicity, and the ability of the foam plugs on the air outlet to trap PCB-52 volatilizedor stripped from the water by aeration (McElroy and Means 1988). During both experiments, exposure vessels were randomly distributed and maintained in a constant temperature water bath.

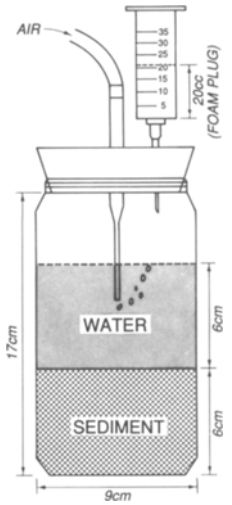

Figure 1. Experimental Apparatus

In the first experiment, sediment amendment was accomplished by coating the sides of the glass jars with sufficient PCB-52 ([UL-^{14}C]-2,2',5,5'-tetrachlorobiphenyl, specific activity of 5.25 Becquerel/mole, purity > 98% by high performance liquid chromatography) to yield sediment concentrations of either 1 or 10 μg PCB/g dry weight. This spiking method was chosen to avoid addition of solvent or carrier to the sediment. Sediment (Oakland Harbor, California, 1.06% total organic carbon) and sufficient saline water (31 ppt) were added to the jars to give a water:-sediment ratio of 3:1. The jars were shaken for 3 d and allowed to settle for 4 d before the excess water was siphoned off and replaced with fresh portions of saline water. The test apparatus was allowed to aerate for 1 d prior to addition of clams (*Macoma nasuta*). Test apparatus were maintained at 17.5°C, the temperature at which the clams were collected near Dillon Beach, California. Five clams, averaging 1.75 cm in length, were added to each jar. Four replicate jars of each exposure condition per sampling time and three controls for each sampling time were prepared. The overlying water, interstitial water, foam plugs, and clams were sampled at 2, 10, 15, and 23 d after introduction of clams. PCB trapped in the foam plugs was extracted by drawing three successive 5-mL aliquots of hexane into the syringe holding the plug, allowing the hexane to stand for 15 sec, then slowly eluting the hexane. Aliquots of hexane were then pooled and 1 mL was counted in a Beckman Liquid Scintillation Counter using the external standard method (Beckman Instruments 1974). Interstitial water was sampled by centrifuging 30 g of sediment in a 25-mL stainless steel centrifuge tube for 1 hr at 7,400 relative centrifugal force (RCF). Overlying water was also centrifuged prior to analysis. A portion of the interstitial water sample (1 mL) was counted immediately; 5 mL were passed through a C-18 Sep-Pak and a 1 mL aliquot of the eluant counted to determine the concentration of bound PCB-52 (Landrum et al. 1984). Clams were placed into containers of clean, aerated saline water with a false bottom in the container to prevent reingestion of depurated sediment. The clams were allowed to depurate for 24 hr following sampling, shucked, and the soft tissue frozen. Clams (3-5 g wet weight) were homogenized in stainless steel centrifuge tubes before extracting three times with a 1:1 acetone:hexane (V/V) mixture. At the end of each extraction, the homogenate was centrifuged at 4,162 RCF for 10 min to separate the extract from the clam tissue. All three extracts were pooled, measured (mL), and counted by LS. Lipids were determined by measuring 0.1 mL of the pooled extract into a tared aluminum pan, air drying to remove the solvents and weighing the residue on a microbalance (Rubenstein et al. 1987). This method consistently yields lipid concentrations about half (0.51 to 0.54) those obtained using the chloroform/methanol (modified Bligh-Dyer) method. Dry weight was also determined

on a tissue subsample by oven drying to constant weight (24 hr) at 90°C. Sediment total organic carbon content was determined using an Oceanographic International Model 700 organic carbon analyzer (American Public Health Association 1985).

In the second experiment, three sediments collected with a Ponar grab sampler were used: 1) Oakland Inner Harbor from Oakland, California; 2) Red Hook from the New York Bight, New Jersey; and 3) a mixture of sediment from Brown's Lake, which is a freshwater lake in Vicksburg, Mississippi, and sediment from a salt marsh channel in Louisiana. The mixed sediment provided a test of organic matter different from that in the two saline sediments (Oakland and Red Hook).

Two organisms having different feeding modes were used in this study: clams (*Macoma nasuta*) from northern California, which burrow into and deposit feed on surficial sediments via an incurrent siphon, and worms (*Nereis virens*) from the Maine coast, which burrow into and ingest the sediment. The clams and worms averaged 2.0 cm and 12.5 cm in length, respectively. Clams and worms were exposed to each of the three sediments amended with sufficient PCB-153 ([UL-^{14}C]2,2',4,4',5,5'-hexachlorobiphenyl, specific activity of 7.4 x 10^{11} Bequerels/mole, purity > 98% by high performance liquid chromatography) or fluoranthene ([3-^{14}C]fluoranthene, specific activity of 2.0 x 10^{12} Becquerel/mole, purity > 98% by thin layer chromatography) to yield a sediment concentration of 4 μg PCB-153 or fluoranthene/g dry weight using apparatus and methods described in the previous section.

Experiments were maintained in a water bath at 15° C, the optimum temperature for the worms and clams. Four replicate jars of each exposure condition, containing either three clams or two worms per sampling time and three control replicates for each sampling time were prepared. Overlying water salinity was maintained at 30 ppt. Organisms were harvested at 2, 10, and 15 d after initiation of the study. Overlying water, interstitial water, foam plugs, and clams and worms were sampled, processed, and counted using the methods described for the first study.

Means and standard errors were calculated using Quattro Pro®, Borland International, Scotts Valley, California. Analysis of variance, and differences between means (t-test following ANOVA) were determined using the Stat-Packets® Statistical Analysis Package, Walonick Associates, Minneapolis, Minnesota.

RESULTS AND DISCUSSION

Free PCB-52 concentrations (not bound to dissolved organic matter or microparticulates) in interstitial water from the first experiment, as indicated by radioactivity, significantly ($p < 0.05$) decreased following the 2 d sampling period in both the 1 and 10 μg/g treatments (Table 1). At 23 d, free PCB-52 levels in interstitial water were approximately 15 times higher in the 10 μg/g than in the 1 μg/g treatments.

Other system components were less stable. Overlying water concentrations increased through the 15 d sampling. Losses of PCB-52 from the system through volatilization accounted for 0.54% and 1.44% of the total mass of PCB-52 added in the 1 and 10 μg/g treatments, respectively. Tissue concentrations consistently increased (Table 2). After 23 d of exposure, clams had accumulated an average of 0.35 and 4.24 μg PCB/g wet

Table 1. Concentrations of PCB-52 ([^{14}C]2,2',5,5'-tetrachlorobiphenyl) [mean (standard error)] in the overlying water and interstitial water and mass loss of volatile PCB-52.

	1 μg PCB-52/g dry weight treatment			
	Concentration, μg/L			
Time, d	Overlying water	Interstitial water free	Interstitial water bound	Volatile loss, μg
2	0.031(0.009)	1.86(1.12)	0.06(0.01)	0.062(0.034)
10	0.045(0.012)	0.14(0.02)	0.02(0.01)	0.302(0.17)
15	0.093(0.011)	0.34(0.06)	0.09(0.01)	0.716(0.05)
23	0.080(0.034)	0.23(0.06)	0.02(0.01)	0.950(0.47)
	10 μg PCB-52/g dry weight treatment			
	Concentration, μg/L			
Time, d	Overlying water	Interstitial water free	Interstitial water bound	Volatile loss, μg
2	0.16(0.01)	5.45(0.34)	0.11(0.18)	1.81(.030)
10	0.50(0.09)	1.35(0.40)	0.22(0.03)	4.34(2.29)
15	0.67(0.16)	1.91(0.80)	0.10(0.01)	7.87(1.57)
23	0.61(0.04)	3.94(0.90)	0.25(0.04)	19.9(1.56)

weight tissue in the 1 and 10 μg/g treatments, respectively. Lipid concentrations in the organisms were relatively constant, averaging 9.4 (0.10) mg/g over both treatments and time. Accumulation factors at each time interval were calculated according to Rubinstein et al. (1987) as:

$$AF = (PCB_O/\%lipid)/(PCB_S/\%TOC)$$

Table 2. PCB-52 ([^{14}C]2,2',5,5'-tetrachlorobiphenyl) concentrations in clam tissue [mean (standard error)], bioaccumulation factors (BAF) (organism concentration μg/g wet wt./sediment concentration μg/g dry wt.), and accumulation factors (AF).

	1 μg PCB-52/g dry weight treatment Time, d			
	2	10	15	23
Tissue conc., μg/g	0.11(0.01)	0.19(0.01)	0.20(0.04)	0.35(0.08)
AF	8.26(1.31)	3.93(0.09)	3.91(0.84)	9.06(4.23)
BAF	0.11(0.01)	0.19(0.01)	0.20(0.04)	0.35(0.08)
	10 μg PCB-52/g dry weight treatment			
Tissue conc., μg/g	0.95(0.10)	2.05(0.14)	3.25(0.44)	4.24(1.00)
AF	19.2(3.1)	3.54(0.16)	2.34(0.42)	1.67(0.42)
BAF	0.10(0.01)	0.21(0.01)	0.33(0.04)	0.42(0.1)

where
PCB_O = PCB concentration in clams, μg/g wet weight
%lipid = Percent lipid in organism extracts, g/g wet wt.
PCB_S = PCB concentration in sediment, μg/g dry weight
%TOC = Percent total organic carbon, g/g dry weight

These non-equilibrium AFs were compared over time for each chemical and against the theoretical equilibrium AF of 1.72.

The AF for the 10 μg/g treatment decreased as exposure time increased (Table 2). The greatest decrease was between days 2 and 10 due to anomalous organism lipid concentrations in the day 2 exposure. Use of the average lipid concentration changes the AF to 9.3. No further significant ($p < 0.05$) decrease in AF was noted after 15 days of exposure. The AF value after 10 and 15 d of exposure in the 1 μg/g treatment was 3.9, which was similar to the AF value in the 10 μg/g treatment. Analysis of variance showed that day 15 values of AF did not differ significantly from AF values after 23 d of exposure indicating a rapid approach to steady-state bioaccumulation for this chemical. Accumulation factors were higher and more variable in the 1 μg/g treatment than in the 10 μg/g treatment. This agrees with findings that accumulation factors from areas of low contamination are higher and more variable than those from areas of higher PCB contamination (> 1.1 ppm) (Clarke et al. 1988).

In the second experiment, trends in interstitial water concentrations, organism bioaccumulation, and volatile losses paralleled results observed in the first experiment. Red Hook sediment, one of those used in the second experiment, contained numerous small lumps of shiny black coal. The TOC value (2.92%) obtained following passage through a 40 mesh sieve was used to compute measured K_{oc} and accumulation factors. The values of 15-day AFs for fluoranthene and PCB-153 (Table 3) as well as those for PCB-52 were within the range of values observed for other empirical determinations reported in the literature (McFarland and Clarke 1986; Bierman 1990; McElroy and Means 1988; Ferraro et al. 1991) for both field and laboratory studies and studies using both spiked and "naturally" contaminated sediment. However, accumulation factors were variable among sediments for the same organism as well as among organisms. Accumulation factors for *Nereis* showed a sixfold range of values (0.8 to 4.79) while those for *Macoma* showed an almost tenfold range of values (0.49 to 4.79). These values bracket the theoretical value of 1.72 (McFarland and Clarke 1986) with a range of variability higher than the two to fourfold variability in AF's reported by Lee (1992) when data from single experiments were analyzed. The reasons for this variability are not immediately apparent.

Table 3. Accumulation factors (n = 4) from experiment two following 15 d of bioaccumulation testing, mean (standard error)

	Fluoranthene		PCB-153	
Sediment	*Nereis*	*Macoma*	*Nereis*	*Macoma*
Oakland	0.8(0.17)	3.77(3.4)	4.78(0.65)	4.79(1.77)
Mixed	1.05(0.18)	2.47(0.79)	1.41(0.29)	Samples lost
Red Hook	3.31(1.27)	0.55(0.84)	4.79(2.50)	0.49(0.19)

Table 4. Measured Log K_{OC} values for fluoranthene and PCB-153 following 15 d of incubation.

	Fluoranthene		PCB-153	
Sediment	*Nereis*	*Macoma*	*Nereis*	***Macoma***
Oakland	5.28(0.06)	4.67(0.07)	6.15(0.08)	5.70(0.05)
Mixed	4.72(0.03)	5.47(0.35)	5.11(0.15)	5.38(0.12)
Red Hook	4.47(0.11)	4.62(0.17)	5.54(0.16)	5.26(0.11)

The ability of equilibrium partitioning to predict interstitial water PCB-153, PCB-52, and fluoranthene concentrations in sediment was tested by comparing estimated K_{oc} with measured K_{oc} values. Estimated K_{oc} values were computed by substituting values of log K_{ow} (octanol/water partition coefficient) for fluoranthene (5.5) (Tetra Tech 1985), PCB-52 (5.85), and PCB-153 (6.92) (Hawker and Connell 1988) in the equation, $K_{oc} = 0.411K_{ow}$, (Karickhoff 1981). Measured values of K_{oc} were determined by dividing the TOC normalized sediment concentration by the free interstitial water concentration.

Estimated log K_{oc} values for fluoranthene and PCB-153 were 5.09 and 6.5, respectively. Measured log K_{oc} values for the 15 day sampling (Table 4) differed substantially from estimated log K_{oc} values for both fluoranthene and PCB-153. Similar results were observed for PCB-52. Measured log K_{oc} was consistently lower than estimated log K_{oc} for PCB-153, but showed no consistent pattern for fluoranthene. Measured values of K_{oc} for PCBs and fluoranthene differed from estimated values by an average factor of 12 and 4, respectively.

Differences in measured and estimated K_{oc} values can be due to many factors. Grathwohl (1990) found that K_{oc} values and K_{ow} derived values for K_{oc} in the literature fail to account for variations in the composition of natural organic matter and are likely to be misleading. Gauthier et al. (1987) reported that K_{oc} can vary by a factor of 10 as a function of organic carbon aromatacity. Steinberg et al. (1987) reported that measured concentrations in soil pore waters differ from predicted values for contaminants that have been associated with the soil for extended periods of time.

Accumulation factors bracketed the theoretical value of 1.72 for these organisms and showed pronounced variability across sediments with different values and kinds of sediment organic carbon. Sediment concentrations of PCB and fluoranthene normalized to sediment organic carbon gave relatively poor estimates of free interstitial water concentrations.

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