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Partitioning of 2,4,5,2′,4′,5′-Hexachlorobiphenyl between Seawater and Air

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Polychlorinated biphenyl (PCB) residues have been detected in many different marine species during the past three decades. While use has declined, incineration has occurred on vessels in the Gulf of Mexico since the early 1980s (Zurer 1985), and incineration off the California coast has been proposed (California Air Resources Board 1981). In order to better understand the adverse effects of PCBs on marine organisms, it has been important to describe the toxic responses of species most likely impacted by disposal or incineration.

PCBs are generally characterized by low vapor pressures (P_vs, 4 x 10^{-6}-2.3 atm; Mackay et al. 1983); they would not usually be considered volatile. However, they are also characterized by low water solubilities and high log K_{ow}s (4-9; Rapaport and Eisenreich 1984; Woodburn et al. 1984); water solubility is inversely related to chlorine content (Geyer et al. 1984). Thus, they exhibit significant volatility from aqueous systems (Geyer et al. 1984). PCBs move between liquid and gaseous phases by dissolution and volatilization until chemical potential (fugacity) is equalized and equilibrium is attained between the two. The equilibrium is described by the Henry's law constant (H'), which relates the partial pressure of a chemical in the gas phase to its concentration in the liquid phase. H' is dependent upon both vapor pressure and water solubility.

Studies addressing PCB toxicology have usually employed open aquaria (Vodicnik and Peterson 1985). Therefore, the objective of this investigation was to describe the gas-liquid partitioning of a single PCB isomer, 2,4,5,2',4',5'-hexachlorobiphenyl (HCBP), in seawater within glass aquaria commonly used for toxicological testing. Partitioning may reduce desired exposure levels during testing, seriously effecting results. However, it may be utilized to realistically model environmental conditions during testing (Tjeerdema and Jacobs 1987). Therefore, the measured H' will be compared to others obtained with specialized purging equipment. 2,4,5,2',4',5'-HCBP was chosen for its presence in common PCB mixtures and high chlorine content, thus low water solubility and high bioaccumulation potential (Hutzinger et al. 1974).

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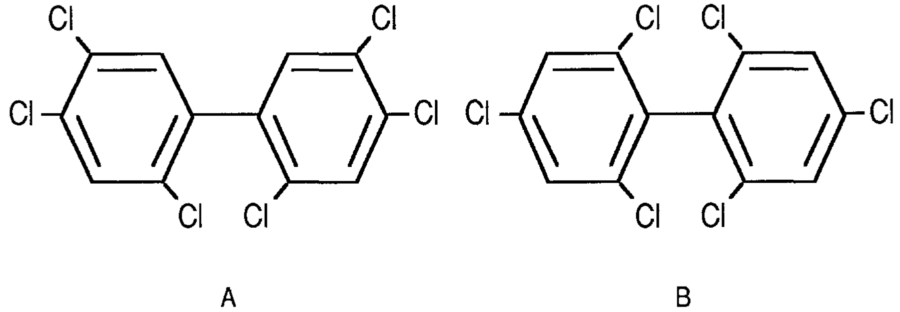

Figure 1. Structures of the two PCBs: (A) 2,4,5,2',4',5'-HCBP and (B) 2,4,6,2',4',6'-HCBP.

MATERIALS AND METHODS

Both 2,4,5,2',4',5'- and 2,4,6,2',4',6'-HCBP (Fig. 1) were purchased from Analabs-Foxboro Analytical (North Haven, CT). In a flowing seawater (18°C) fiberglass tank were placed six 2-gal glass jars (21 cm dia) filled with 6 L of seawater and equipped with glass aerators (2 L min^{-1}). Into each of three jars was pipetted 60.0 ± 0.3 μg of 2,4,5,2',4',5'-HCBP in acetone to produce an initial concentration of 10.0 ± 0.1 μg L^{-1}; the level being similar to those measured near sewage outfalls (Young et al. 1976). Three jars, with clean seawater only, served as controls. All jars were sealed with aluminum foil, leaving a small vent for air flow. Sampling commenced immediately upon chemical addition; after 6 h the jars were emptied and refilled with clean seawater to check for glass adsorption by facilitating desorption during an additional 6 h period. After 0, 0.5, 1, 1.5, 2, 2.5, 3 h, and thereafter every hour until 12 h, a 2-mL water sample was removed from each jar; they were pipetted into 1-dr vials and frozen; all glassware was disposed of or decontaminated (Bevenue et al. 1971).

Seawater was extracted sequentially with 300 μL of 15% diethylether-in-hexane, 6% diethylether-in-hexane, and hexane; extracts were aspirated, combined, volume-reduced to 100 μL at ambient temperature with N_2 gas, and passed through Florisil® (E. Merck, Darmstadt, FRG) columns for cleanup. Columns were pasteur pipets packed at the tips with glass wool, filled with activated Florisil®, and topped with anh. sodium sulfate; Florisil® was activated by heating at 130°C for 5 h, then immersed in hexane. Following elution, columns were flushed with 6% diethylether-in-hexane; the eluent and wash from each column were combined, evaporated to dryness, and adjusted to 25 μL with hexane.

Samples were analyzed with a Hewlett-Packard Model 5840A gas chromatograph, equipped with a 50.0 x 0.2 mm (i.d.) capillary column coated with methyl silicone and an electron capture detector. Helium and argon-methane (95:5; 31 mL min^{-1}) served as carrier and detector make-up gasses, respectively. Column temperature was programmed to rise from 80-250°C at 10°C min^{-1} for the first 10 min, and 3°C min^{-1} thereafter. Injection was splitless (180°C), and the inlet pressure was 25 psi. Retention times were 38.2 min for 2,4,5,2',4',5'- HCBP and 32.5 min for 2,4,6,2',4',6'-HCBP, which served as a method internal standard

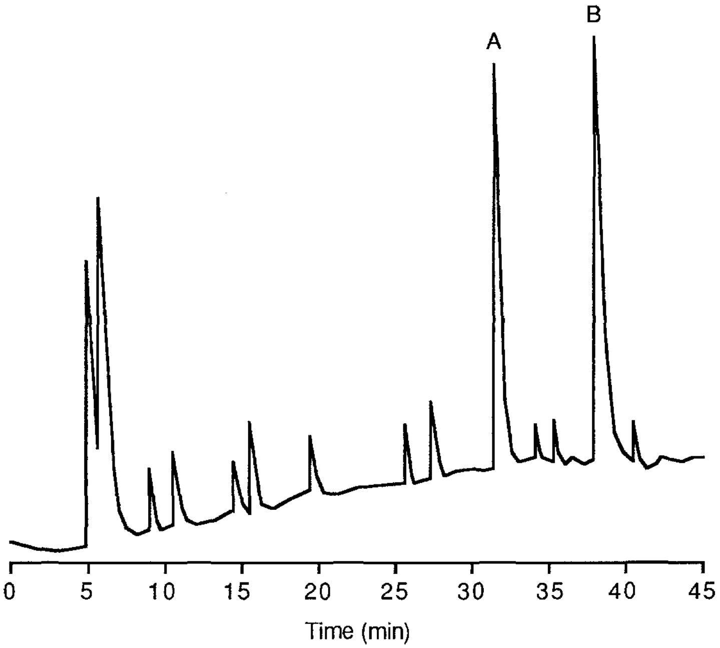

Figure 2. Chromatogram from a seawater sample: (A) 2,4,6,2',4',6'-HCBP, and (B) 2,4,5,2',4',5'-HCBP (see text for conditions).

(Fig. 2). Amounts as low as 0.01 µg L^{-1} could be detected accurately and reproducibly, and recoveries above 90% were routine.

H's were calculated from the depletion-rate data based on a model developed by Mackay et al. (1979) and modified by others (Dunnivant et al. 1988). Values were calculated as follows: $H' = D_r(RTV)G^{-1}$; D_r, depletion rate constant; R, ideal gas law constant; T, temperature (°K); V, aqueous solution volume; and G, gas flow rate. D_r may be calculated by at least three different methods: (1) the fraction of chemical remaining in solution with time; (2) the fraction purged from solution with time; and (3) the cumulative mass purged from solution with time (Dunnivant et al. 1988). Mathematically, all three approaches give identical values for D_r when data fits the model well; we calculated D_r using method 1. Comparisons were made with a one-tail *t* test (Goldstein 1964).

RESULTS AND DISCUSSION

During the 6 h depletion period, seawater levels diminished by more than 60%; decrease was most rapid during the first hour (Fig. 3). The H', $1.52 \times 10^{-4} \pm 0.05 \times 10^{-4}$ atm·m^3·mol^{-1}, was within those previously reported for 2,4,5,2',4',5'-HCBP in distilled water at 25°C, which range between 1.23-1.77 atm·m^3·mol^{-1} (Coates 1984; Burkhard et al. 1985; Dunnivant et al. 1988). H' may also be written as a function of both vapor pressure (V_p) and water solubility (S): $H' = V_pS^{-1}$. Assuming most organic compounds, including PCBs, are less soluble in seawater than

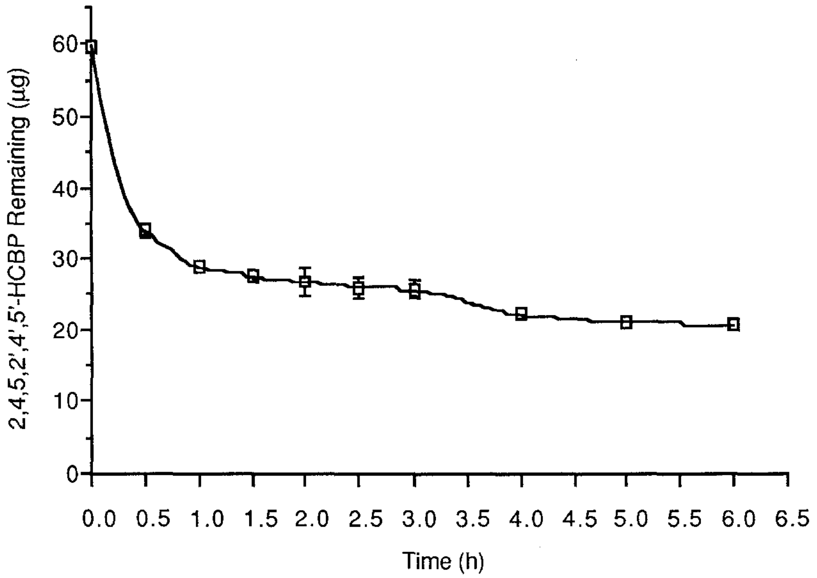

Figure 3. Depletion curve for 2,4,5,2',4',5'-HCBP in seawater without sea urchins; intervals are in SD (n=3).

fresh water, due to greater ionic strength, a larger H' was anticipated. However, the solutions were not equilibrated prior to aeration (to mimic toxicological testing), and the temperature (18°C) was well below that commonly used (25°C).

While depletion in seawater was most likely due to partitioning between it and air, glass adsorption (and subsequent desorption following loss from air-water partitioning) would decrease the measured H' (Dunnivant et al. 1988). Therefore, a partial H' (H_p') of $8.62 \times 10^{-4} \pm 0.16 \times 10^{-4}$ atm·m^3·mol^{-1} for the first three time points (the period of most rapid PCB loss) was calculated. The larger H_p' indicates glass adsorption/desorption did occur, particularly early on. Also, following the 6-h initial period, glass-adsorbed 2,4,5,2',4',5'-HCBP rapidly desorbed into the clean replacement seawater. Concentrations stabilized within 1 h, measuring 0.40 ± 0.09 µg L^{-1} for the 6-h phase.

Generally, the H's measured for both polycyclic aromatic hydrocarbons and halogenated aromatics range from 10^{-5}-10^{-3} atm·m^3·mol^{-1} (Thomas 1982). In this range, both liquid- and gas-phase resistances to partitioning are important; volatilization, while not rapid, is significant. 2,4,5,2',4',5'-HCBP falls within this range. However, losses due to air-water partitioning are influenced by both the high salinity of the aqueous phase, reducing solubility, and the low temperature, reducing both solubility and vapor pressure. Burkhard et al. (1985) previously has shown that measured H's for PCBs increase approximately an order of magnitude with a 25°C increase in temperature.

Utilizing both static and flow-through exposure systems, the disposition

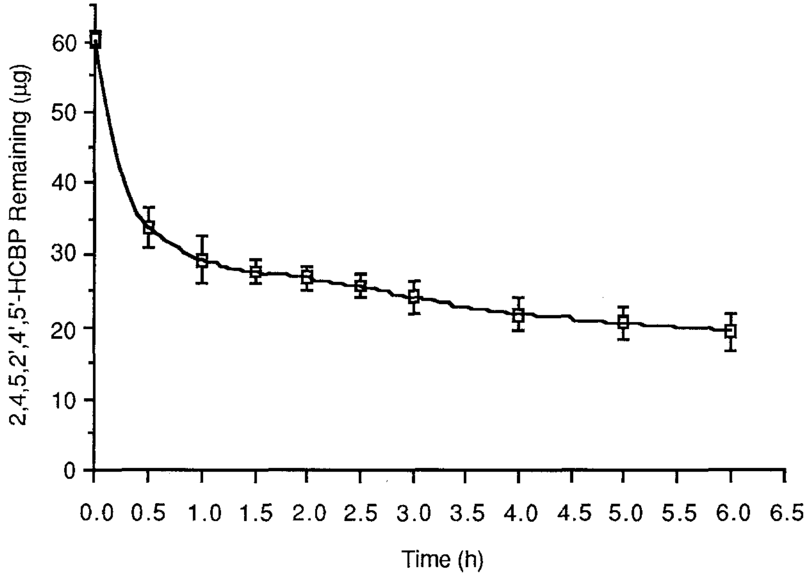

Figure 4. Depletion curve for 2,4,5,2',4',5'-HCBP in seawater in the presence of sea urchins; intervals are in SD (n=9; Tjeerdema and Jacobs 1987).

and toxic effects of several PCB isomers have been described in both invertebrates and fishes (Lowe et al. 1972; Guiney et al. 1977). When using PCBs in glass systems, adsorption and desorption will occur. Similarly, when using open systems, air-water partitioning must be considered. In an open static system, maintaining a constant PCB level is virtually impossible; significant losses will occur from both air-water partitioning and surface adsorption. In a closed system, maintaining proper aeration becomes an additional problem. When using open or closed flow-through systems, time must be allowed for saturation of glass surfaces prior to organism exposure. Also, PCB mixtures, like all complex organic mixtures, are subject to differential partitioning of the various components. Therefore, closed flow-through systems are a necessity when using such mixtures; single isomers may be effectively used in either open or closed flow-through systems. However, when using an open system, isomer concentration must be closely monitored.

Open static systems may be used with single PCB isomers when stable water levels are not desired. Previously, elimination of 2,4,5,2',4',5'-HCBP was described in the purple sea urchin (*Strongylocentrotus purpuratus*) with the same protocol as in this study (Tjeerdema and Jacobs 1987). The decrease in water levels during exposure was used to mimic a single chemical pulse, and thus model the periodic nature of environmental exposure (Fig. 4); contributing to chemical loss may be uptake by organisms. However, both the H' ($1.67 \times 10^{-4} \pm 0.29 \times 10^{-4}$ atm·m^3·mol^{-1}) and H_p' ($8.59 \times 10^{-4} \pm 1.36 \times 10^{-4}$ atm·m^3·mol^{-1}) did not significantly differ from those without organisms ($P>0.05$); water levels following a similar clean water flush also did not differ (0.44 µg L^{-1}, P>

0.05). Generally, when stable water levels are required, closed flow-through systems are superior. One such system using a syringe pump to deliver chemical directly to the water flow, eliminating need for a complex dilution apparatus, has been recently developed for use with volatile pesticides (Tjeerdema and Crosby 1987).

In conclusion, toxicological testing of chemicals possessing low water solubilities and/or high vapor pressures, such as PCBs, should incorporate closed flow-through exposure systems; open systems may be effective for single compounds with close analytical monitoring. Open static systems are rarely suitable for single compounds unless declining water levels are desired, and unsuitable for complex chemical mixtures due to differential physical properties. If cost is not an issue, systems made of teflon should be considered.

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