Evaluation of PCB bioaccumulation patterns in white sea urchins (*Lytechinus pictus*) using multiple approaches

James A. Noblet, Eddy Y. Zeng, and Kerry J. Ritter

ABSTRACT - The bioaccumulation of polychlorinated biphenyls (PCBs) from three amended field-contaminated sediments (with total PCB concentrations of ~ 4, 10, and 100 µg/g-dry wt) by white sea urchins (*Lytechinus pictus*) was evaluated using multiple statistical and theoretical approaches. Similarity analysis of the PCB bioaccumulation patterns, based upon the concept of ecological communities, showed that the PCB patterns in the sea urchins and source sediments were essentially identical for all three sediment concentrations. However, affinity analysis did show some preference for bioaccumulation of higher molecular weight and more hydrophobic congeners by the urchins. The affinity analysis also showed that with a homologous series, bioaccumulation increased with increasing hydrophobicity. The biota-sediment accumulation factor (BSAF) profiles for the two lower concentration sediments (A and B) were found to be statistically different from the high concentration sample (Sediment C) by a multivariate analysis of variance (MANOVA). The relationship between the measured apparent organic carbon normalized partition coefficients (K_{OC}) and octanol-water partition coefficient (K_{OW}) (log-based) suggested a significant departure from thermodynamic equilibrium. A nonequilibrium, steady-state bioaccumulation model was found to correctly predict the observed experimental bioaccumulation patterns. To improve the model performance, a hydrophobic term was introduced to account for the drop-off in BSAF profiles with log $K_{OW} \geq 6.5$. This study showed that nonequilibrium, steady-state models are far superior to equilibrium partitioning-based models for understanding the bioaccumulation of organic chemicals by sea urchins.

INTRODUCTION

Sediments contaminated with persistent organic pollutants (POPs) are of concern because of their role as a reservoir from which these compounds can be transferred to benthic organisms via bioaccumulation and/or bioconcentration (Barron 1990, Bierman 1990, Morrison *et al.* 1996). While direct toxicity to benthic organisms is certainly a concern, a more important aspect of sediment contamination by POPs is their propensity for bioaccumulation and biomagnification in organisms at higher trophic levels within aquatic food webs (Connolly and Pedersen 1988, Thomann 1989, Clark *et al.* 1990, Morrison *et al.* 1996, Sharpe and Mackay 2000). Benthic organisms accumulate POPs because they live in intimate contact with, and often feed upon, aquatic sediments. Therefore, a thorough understanding of the transfer of potentially toxic POPs from sediments to aquatic organisms is a necessary prerequisite for establishing meaningful sediment quality criteria (Shea 1988).

The most commonly used approaches for evaluating bioaccumulation data are based on the equilibrium partitioning (EqP) model (Shea 1988, Bierman 1990, Di Toro *et al.* 1991, Vanier and Planas 2001, Wong *et al.* 2001), which assumes that chemicals partition between sediment organic carbon, porewater, and the lipid fraction of benthic organisms, and that thermodynamic equilibrium is approximately achieved among these phases. Bioaccumulation data for benthic organisms are often used to calculate the BSAF, which is simply the ratio of the lipid-normalized organism concentration to the organic carbon-normalized sediment concentration,

$$BSAF = \frac{\left(C_{biota} \big/ f_{lipid} \right)}{\left(C_{sed} \big/ f_{oc} \right)} \qquad (1)$$

where C_{biota} is the concentration in the organism , f_{lipid} is the fraction of lipid in the organism, C_{sed} is the sediment concentration, and f_{oc} is the fraction of organic carbon in the sediment. One of the most attractive aspects of the EqP model is that it is conceptually straightforward and easy to use. If the assumption of equilibrium among all phases is true, then the BSAF should be a constant independent of chemical, organism, and sediment properties. It has been suggested that the theoretical constant value for BSAF should be in the range of 1–2, or 0–0.3 on a log scale (Morrison *et al.* 1996, Wong *et al.* 2001).

In contrast to the predictions of the EqP model, reviews of the literature have shown that BSAF values can differ by several orders of magnitude (Morrison *et al.* 1996, Kraaij *et al.* 2002). In addition, BSAFs frequently exhibit a parabolic dependence on K_{OW} (Bremle *et al.* 1995, Morrison *et al.* 1996, Fisk *et al.* 1998, Lamoureux and Brownawell 1999, Zeng *et al.* 2003). Deviations from the EqP model can be attributed to two general causes-differences in bioavailability and nonequilibrium. The bioavailability of POPs is a function of complex biological, ecological, and physicochemical factors such as habitat; feeding behavior; dietary and non-dietary assimilation efficiency; biotransformation and depuration rates; sorption/desorption behavior; organic carbon content and nature of the sediments; and binding to suspended solids, colloids, and dissolved organic matter (Suffet *et al.* 1994, Means and McElroy 1997). Similarly, the attainment of equilibrium for POPs requires prolonged contact between the multiple phases in a system. It has long been recognized that the assumption of equilibrium is at best an approximation for many natural waters, and that complex environmental systems would be more accurately described using nonequilibrium models (Mackay 1982). In cases where the equilibrium assumption is invalid, it is necessary to employ the use of bioaccumulation models that make no assumptions regarding the thermodynamic state of the system (Lee 1992, Newman 1994, Morrison *et al.* 1996, Boese *et al.* 1997, Means and McElroy 1997).

The PCBs are synthetic chemicals produced as mixtures of individual congeners. The production of PCBs was phased out in 1977 in the United States due to concerns about their potential toxicity to natural species and humans, but approximately 600 million kg of PCBs had already been manufactured (Weaver 1984). Because of their resistance to chemical, physical, and biological degradation, PCBs can remain intact for extended periods upon discharge to the environment. In southern California, coastal sediments in Santa Monica Bay, Los Angeles/Long Beach Harbor, and San Diego Bay, and on the Palos Verdes Shelf are known to contain elevated levels of PCBs (Stull *et al.* 1996, Fairey *et al.* 1998, Anderson *et al.* 2001, Zeng *et al.* 2001). Quantitative information about the bioaccumulation of PCBs by marine invertebrates is obviously relevant for both tracing the fate of PCBs in the environment and assessing the effects of PCBs on ecological and human health.

This study was intended to provide such quantitative information by evaluating the PCB bioaccumulation patterns in the white sea urchin (*Lytechinus pictus*), a typical marine invertebrate in southern California, upon exposure to contaminated sediments. The objectives were to (1) determine whether the PCB bioaccumulation patterns in the sea urchins were the same as those observed in the source sediments, (2) determine whether the concentration of the contaminants affects bioaccumulation patterns, and (3) identify, if possible, an appropriate model for predicting the bioaccumulation of hydrophobic organic contaminants from sediments by benthic marine organisms. All experimental data used herein were derived from a previously reported study (Zeng *et al.* 2003).

METHODS
Sea Urchin Exposure Experiments

The experimental setup consisted of 20 polyethylene chambers, approximately 29 cm (L) × 26 cm (W) × 14 cm (D) in dimension. Each chamber was filled with sediment to a depth of approximately 3 cm. Three different amended sediments designated A, B, and C were prepared at total PCB concentrations of approximately 4, 10, and 100 µg/g-dry wt, respectively. The sediments were prepared by mixing highly contaminated sediment (~3,800 µg/g-dry weight total PCBs) from New Bedford Harbor, Massachusetts, with uncontaminated local sediments from coastal sites near Dana Point and/or Newport Beach, California, in the appropriate ratios. In addition, a reference sediment (R) was prepared from only the clean local sediments. Five replicate chambers were prepared with each of the three different amended sediments and the reference sediment. The exposure chambers were allowed to come to steady-state at 15°C, with seawater flowing through each chamber

at 8 to 10 mL/minute, at a depth of about 4 cm, for a period of four weeks prior to introduction of the test animals. Based on the volumes and the nominal flow rate, the seawater volume in each chamber was replaced approximately 4 to 5 times each day.

The urchins used for these experiments were collected from a coastal reference area near Dana Point, California. Fifteen urchins were introduced into each chamber, and left undisturbed for a period of 42 (Sediments R, A, B) to 43 d (Sediment C). The depth of the seawater was kept to a minimum so that the urchins were forced to remain in constant contact with the sediment. Gentle aeration was provided to each chamber. The animals were fed Argent Hatchfry microencapsulated food at a rate of 0.36 g every other day. Five sets of animals, related sediment, and insterstitial water samples were collected at the end of the exposure experiments and analyzed for PCBs on a conger-specific basis. Sea urchin, sediment, and interstitial water samples (three replicates each) were also analyzed at the beginning of the exposure experiment to establish baseline PCB concentrations, but the results are not reported. Sea urchins exposed to the reference sediment (R) were analyzed along with the other samples, but they contained no detectable PCBs. Therefore, they were not included in the data analysis. The food materials were also analyzed, and no detectable PCBs were found.

Analytical Methods

The details of the analytical procedures used in this study were previously described (Zeng *et al.* 2003), and will only be briefly reviewed here. Sediment samples were extracted three times with methylene chloride using a roller table. The combined extracts were dried using sodium sulfate, concentrated, solvent-exchanged into hexane, and sulfur removed by standing overnight with activated copper granules. Tissue samples were extracted three times by homogenization and sequential addition of acetonitrile, hexane, and water. After centrifugation, the solvent layer was decanted, poured into a flask and dried with sodium sulfate. The centrifuge bottles were extracted twice more with hexane, and the solvent layers were dried and added to the previous extract. The sediment interstitial water was isolated by centrifugation in 250 mL glass bottles, and extracted three times with methylene chloride. The extracts were combined, dried with sodium sulfate, and solvent exchanged into hexane. All of the

extracts were concentrated to about 1 mL under a gentle stream of nitrogen.

Extracts from all samples were cleaned and fractionated by column chromatography using alumina and silica gel. The eluted fractions containing PCBs were concentrated and analyzed by gas chromatography with dual electron capture detection (GC/ECD) with selected confirmation by gas chromatography/ mass spectrometry (GC/MS). The instrument was calibrated using a 1:1:1:1 mixture of Aroclors 1242, 1248, 1254, and 1260. This calibration standard was characterized prior to the study with 120 individual PCB congener standards. Sediments were analyzed for total organic carbon (TOC) using a Carlo Erba 1108 CHN Elemental Analyzer (Carlo Erba Instruments, Milan, Italy). The sea urchin lipid content was measured by taking a small aliquot of the tissue extract (~5 μL), transferring it to a small aluminum boat, and allowing it to evaporate to constant weight. The residual weight was assumed to be due to lipids, and was used to calculate the lipid content of the original tissues.

Data Analysis

Sediment and sea urchin concentration data (Zeng *et al.* 2003) were evaluated with several different statistical and theoretical approaches, including similarity and affinity analysis, regression analysis and multivariate statistics, EqP theory, and a nonequilibrium bioaccumulation model. A detailed description of these approaches is given below.

Similarity and Affinity Analysis

The similarity analysis was based upon the concept of ecological communities. The individual compounds within a chemical mixture and the chemical concentrations were considered analogous to the number of species and abundance, respectively (Wood *et al.* 1997). Thus, by assigning these attributes to the chemicals, the bioaccumulation data are made amenable to ecological similarity and preference analysis. Similarity between the urchin and sediment PCB patterns was quantified by calculating the Bray-Curtis Similarity Coefficients (Wood *et al.* 1997) for each sediment-urchin paired data set:

$$S_{jk} = \frac{2\left[\sum \min(X_{jk})\right]}{\sum(X_j + X_k)} \times 100 \qquad (2)$$

where S_{jk} is the percent of similarity of the PCB patterns in the urchins and sediment; X_j and X_k are the prominence values $D\sqrt{F}$ for the individual congeners in samples j and k, respectively; D is the concentration of a given congener; and F is the frequency of its occurrence in all samples (i.e., number of samples containing the congener/total number of samples). The parameter D was modified by using the relative concentration (C_i/C_{total}), in order to account for the inherent difference in concentration between the urchins and the sediments. Chemical concentrations below the detection limit were assigned a value of zero. Based on numerous previous studies, Wood *et al.* (1997) concluded that paired samples with similarity values ≥82% are considered essentially identical in terms of distribution pattern. This criterion was also used in the present study. The affinity analysis was also derived from ecology, and was used to quantify the degree of selectivity for certain PCB congeners or groups of congeners accumulated by an organism (Wood *et al.* 1997). The bioaccumulation affinity, A_i, for an individual PCB congener was calculated using the equation,

$$A_i = \frac{\dfrac{r_i}{n_i}}{\dfrac{1}{m}\displaystyle\sum_{j=1}^{m}\dfrac{r_j}{n_j}} \tag{3}$$

where r_i is the proportion of contaminant i in the organism, n_i is the proportion of congener i in the source (i.e., sediment), and m is the total number of congeners. Affinity values greater than 1 indicate a greater than average tendency to bioaccumulate, whereas affinities less than 1 imply a below average tendency for bioaccumulation.

Statistical Analysis of BSAF Profiles

The magnitude and shape of the logBSAF versus log K_{OW} curves for the three amended sediments were compared to ascertain whether concentration had any affect on PCB bioaccumulation. Data were log-transformed to facilitate the statistical analysis. To determine whether the congener uptake patterns differed among the three different PCB concentrations, a multivariate analysis of variance (MANOVA) was performed on profile summaries for logBSAF as a function of log K_{OW}. Profile summaries consisted of coefficient estimates from least squares regression where a quadratic curve was fit to the data. This was done to reduce the multivariate response for

each experimental unit to three simple summaries (intercept, linear coefficient, and quadratic coefficient) representing the overall shape of the curve.

Bioaccumulation Modeling

The bioaccumulation of PCBs in the sea urchins was modeled using a nonequilibrium, steady-state model developed specifically for the bioaccumulation of organic chemicals from sediments by benthic organisms (Morrison *et al.* 1996). In this study, the form of the model developed specifically for detritivores was used, i.e.,

$$\frac{f_B}{f_S} = \frac{E_W G_W \dfrac{f_W}{f_S} + E_D G_D \dfrac{f_D}{f_S} d_D f_D K_{OC}}{E_W G_W + E_D (1-\boldsymbol{a})(1-\boldsymbol{b}) G_D \boldsymbol{d}_D \boldsymbol{f}_D K_{OC} + V_B K_M K_{BW}} \tag{4}$$

where f_B, f_S, f_W, and f_D are the fugacities of the biota, sediment, water, and diet, respectively. E_W and E_D are the efficiencies of chemical transfer across the respiratory surface, and between the gut contents and the organism, respectively. G_W is the rate of water ventilation across the respiratory surface, and G_D is the food ingestion rate. The parameters $\boldsymbol{a}$ and $\boldsymbol{b}$ describe dietary assimilation efficiencies, and are the fraction of organic carbon removed upon ingestion and the fraction of the ingested diet absorbed by the organism, respectively. $\boldsymbol{d}_D$ and $\boldsymbol{f}_D$ are the density and the fraction of organic carbon in the diet, respectively. K_M is the chemical biotransformation rate constant, V_B is the volume of the biota, and K_{OC} and K_{BW} are the organic carbon-water and biota-water partition coefficients, respectively. The K_{OC} for each congener was calculated as $0.41K_{OW}$. The relationship between the biota-sediment fugacity ratio and the BSAF ($f_B/f_S = 0.62$BSAF) follows directly from the model assumptions, and the fugacity ratios were converted to BSAF by multiplication by 1.6. Morrison *et al.* (1996) argued that given the natural variability in the densities and sorptive properties of sediments, that BSAF values between 1 and 2 probably reflect near thermodynamic equilibrium conditions.

RESULTS
Similarity and Affinity Analysis

Similarity analysis using Equation (2) indicated that the PCB bioaccumulation patterns in the sea urchins were essentially identical to the patterns in the sediment, and that the urchins were able to uptake

most PCB congeners from the sediment with similar efficiency (Table 1). The trend of increasing similarity with increasing concentration (Table 1) suggests that the small differences in similarity values among sediments were related to analytical factors and not reflective of any change in bioaccumulation efficiency.

Table 1. The results of the similarity analysis of PCB patterns between the indicated sediments and sea urchins. Similarities ≥ 82% indicate identical PCB patterns.

Sample	Concentration (µg/g)	Similiarity (percent)	Standard Development	N
Sediment A	4	79.9	1.4	5
Sediment B	10	83.6	0.9	5
Sediment C	100	86.7	1	5

Affinity analysis was also performed to determine whether any individual PCB congeners were bioaccumulated preferentially by the urchins. Although the results of the similarity analysis suggest that selective bioaccumulation was not occurring to a great extent (Table 1), it was still possible that a few congeners would exhibit high or low propensities for bioaccumulation. Affinities were calculated for each congener, for each of the five replicates, and the results were averaged. The resulting average bioaccumulation affinities were plotted versus their respective log K_{OW} values and PCB congener number (IUPAC number). To facilitate further comparison, the data for the PCB congeners were grouped according to homologs (i.e., congeners with the same number of chlorine atoms), and ranked within each homologous series by log K_{OW} values. The analysis was limited to sediment-urchin paired data with at least three replicates.

Several trends are apparent in results of the affinity analysis (Figure 1). First, the di- and trichloro homologs had affinity values ≤1, whereas the tetra- and higher homologs mostly had affinities ≥1. Since the degree of chlorination and molecular weight are generally related to the hydrophobicity of a PCB congener, this implies that hydrophobicity may be a primary factor influencing the bioaccumulation of PCBs in these benthic invertebrates. This was further supported by the apparent increase in affinity with increasing K_{OW} within each homolog group. These trends are most noticeable in the sediments with lower concentrations (A and B; Figures 1a and 1b), and much less evident for the high concentration Sediment C (Figure 1c). In addition, Sediment C resulted in affinities that were mostly greater than 1 for the tetrachloro- and pentachlorobiphenyls, and drop to less then 1 again for the hexa- and heptachlorobiphenyls. Moreover, the variability in the data, as measured by the standard deviation of the replicates, is significantly reduced for the high concentration sediment (Figure 1c).

There were only three congeners that seemed to not fit the observed trends. Congeners 24, 46, and 110 had affinities that were noticeably lower than the other congeners within their respective homologous series (Figure 1). No obvious cause could be identified based on chemical, physical, or structural properties, and therefore the observed deviations from the trends may be due to analytical anomalies. Congener 110 co-eluted with two other PCB peaks, but Congeners 24 and 46 were eluted as single compounds. Thus, the reasons for the deviation of these three congeners from the overall trends are unclear.

BSAF Profiles

The logBSAF values were calculated for each sediment-urchin congener pair and plotted versus their corresponding log K_{OW} values (Figure 2). Again, the analysis was limited to data sets with at least three replicates. The curve resulting from the least squares regression analysis is shown on each plot. For comparison, the quadratic regression curves for the three different test sediments are plotted together in Figure 3. In addition, regression analysis was performed on the pooled mean BSAF data from all three sediment concentrations (Figure 4).

The R-squared values for each of the replicates ranged from 0.61 to 0.70 (Figure 3). A MANOVA was then performed on the three-dimensional response vector to test the effect of starting material on logBSAF profiles. The MANOVA resulted in a p value <0.0001 from Wilke's Lambda. Pairwise
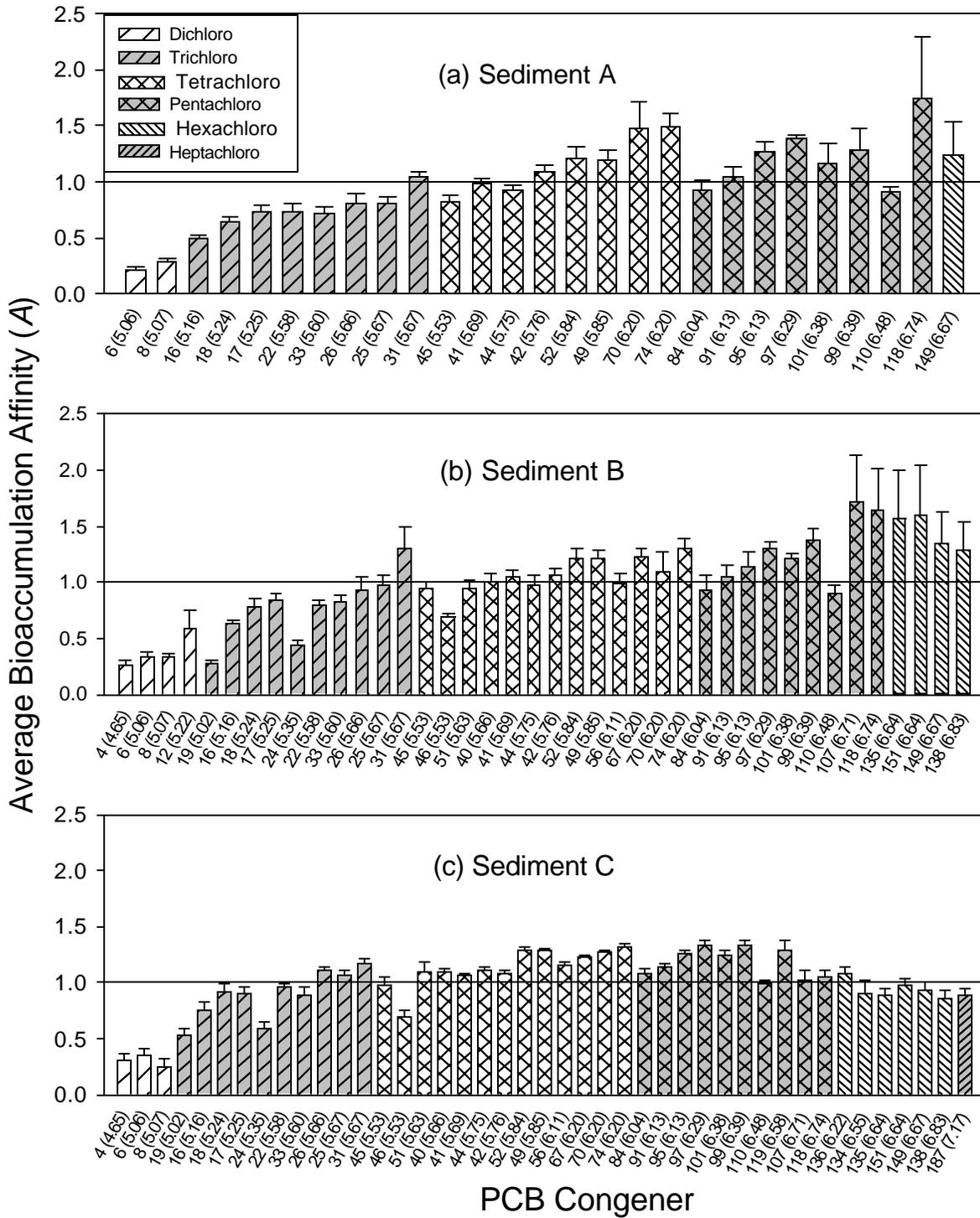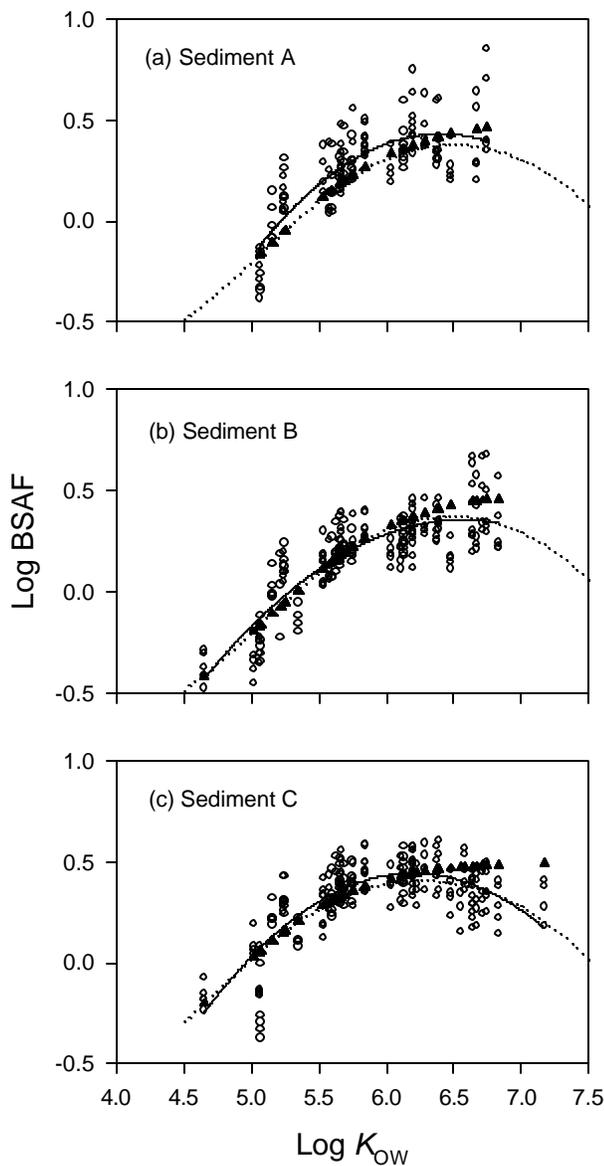
Figure 1. Average bioaccumulation affinities with respect to PCB congener number (numbers in parentheses are log K_{ow} values) for (a) Sediment A; (b) Sediment B; and (c) Sediment C. The error bars are standard deviations of the five-replicate analyses.
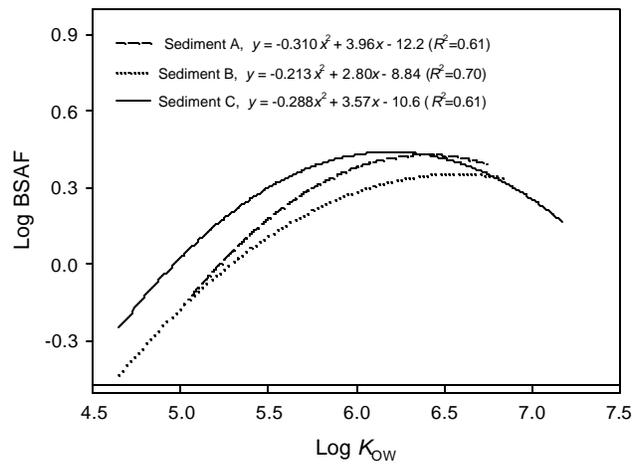
Figure 3. Sea urchin BSAF regression curves from all three sediments.

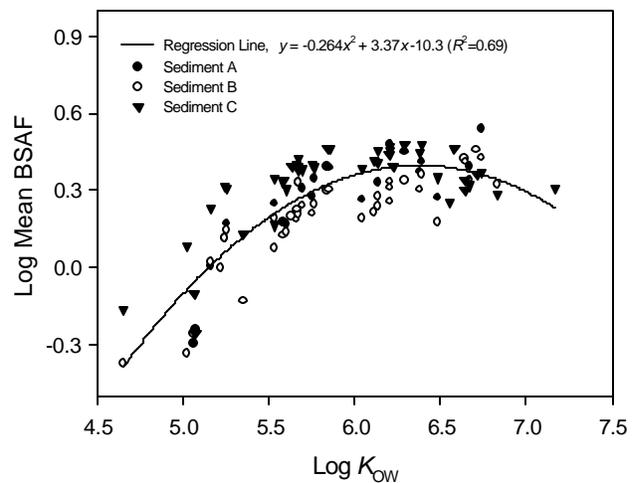**Figure 4. Profile of the log mean BSAF data for Sediments A, B, and C. The curve is the result of the least-squares polynomial regression analysis.**

C. ○: measured logBSAF values; ▲: BSAF values predicted using the bioaccumulation model (Equation (4)); the solid curve is the best fit to the experimental data using least-squares polynomial regression analysis; and the dotted curve represents a modification of the bioaccumulation model by introducing a hydrophobic term ($af_{lipid}V_B K_{OC}{}^2$) related to some unknown hydrophobicity-dependent processes.

comparisons among the three starting materials resulted in statistically significant differences (0.05 level after Bonferroni adjustment) between Sediment C and both Sediments A and B (p values = 0.003 and 0.008, respectively). Sediments A and B, however, were not statistically different (p value = 0.047).

Interstitial Water Analysis

The analytical results from the interstitial water and the corresponding sediments were used to calculate log K_{OC} values, which were then plotted versus log K_{OW}. The resulting plots are nearly horizontal with a slightly negative slope, indicating a decrease in the calculated log K_{OC} values with increasing log K_{OW} (Figure 5). This result at first appears to counterintuitive, since the log K_{OC} values should increase with increasing log K_{OW} as predicted by well-established empirical relationships (Schwarzenbach *et al.* 1993). However, DiToro *et al.* (1991) have shown that plots of this appearance result from not accounting for dissolved organic matter (DOM) in the interstitial water. The DOM
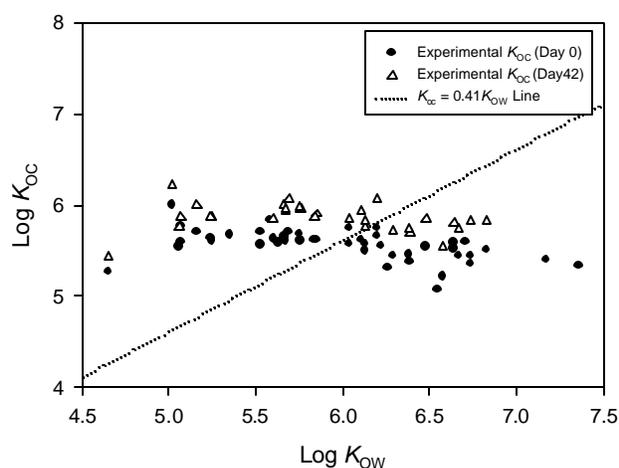
Figure 5. Measured log K_{OC} values calculated from the sediment and interstitial water PCB concentrations for Sediment C. Also shown is the predicted log K_{OC} line calculated from the K_{OW} values, using the empirical relationship, $K_{OC} = 0.41 K_{OW}$ (Mackay and Paterson 1981, Means and McElroy 1997).

binds hydrophobic organic compounds, and increases their apparent solubility in the interstitial water. This, in turn, lowers the calculated log K_{OC} values, and the effect becomes more pronounced as log K_{OW} increases.

Unfortunately, DOM was not measured in the interstitial water phase, and therefore the data cannot be corrected. However, some valuable information can still be obtained from Figure 5. Apparent log K_{OC} data would be expected to plot below the empirical log K_{OC} line. The fact that about half of the data points are above the empirical log K_{OC} line suggests that the system is not at thermodynamic equilibrium. This inference is consistent with the flow-through design of the exposure chambers, wherein the attainment of equilibrium would not be expected. Although not at equilibrium, the results do show that steady-state conditions prevailed in the chambers for the 42-d duration of the exposure.

BSAF Modeling

Efforts were undertaken to determine whether the sea urchin bioaccumulation results could be correctly predicted using the benthic bioaccumulation model (Equation [4]) developed by Morrison *et al.* (1996). The nonequilibrium, steady-state model requires the input of 13 different parameters relating to the nature of the sediment, water column, and the organism. Wherever possible, experimentally deter-

mined values from the literature were used for the model parameters. For parameters that were unavailable or unknown, initial estimates were made and then the values were varied iteratively until the model produced an accurate prediction of the observed BSAF values (Figure 2).

All of the final model parameters (Table 2) obtained by the iterative variation process were evaluated as to their reasonableness based upon knowledge about the experimental conditions. For example, the final water ventilation rate (G_W) obtained was about three times higher than that known for crayfish (Morrison *et al.* 1996). Given that sea urchins have a greater surface area for ventilation (i.e., not just gills), and the flow-through nature of the exposure chambers, the resulting value was considered reasonable. Also, the final feeding rate used was about half that of a field grazing rate reported in the literature (Moore 1966). However, the estimated feeding rate from the literature was for urchins grazing on *Thallasia*, and therefore the lower feeding rate was considered reasonable for the more efficient food source used in this study. Perhaps the most interesting result was the low water-sediment fugacity ratio ($f_W/f_S = 0.1$) required to obtain a good fit to the experimental data. This value suggests a significant degree of disequilibrium between the sediment and the overlying water, which is consistent with the flow-through experimental conditions. In order to evaluate whether this fugacity ratio was reasonable, calculations were performed to estimate the mass of PCBs that would be lost from the sediment to the water due to a steady-state fugacity ratio of 0.1 over the 42-d duration of the exposure experiments. Assuming that PCBs were lost only to the flowing water, the calculations predicted about a 2% loss of total PCB mass in the sediments. This result is quite reasonable relative to the 3.6%, 4.6%, and 5.8% change in sediment concentration observed, especially considering that losses due to volatilization and uptake by the urchins were not taken into account.

DISCUSSION
Assessment of PCB Bioaccumulation Patterns

Each of the different approaches used to evaluate the sea urchin PCB bioaccumulation data provided valuable information about the nature of interactions between the organism and sediments. Similarity analysis showed that the pattern of PCB congeners in the urchins was a direct reflection of the PCB concentrations in the source sediment (similarities

Table 2. Final model input parameters used to calculate BSAF values.

Model Parameters	Sediments A, B	Sediment C
PCB Assimilation Efficiency from Water (E_W, %)[a]	100	100
Water Ventilation Rate (G_W, m^3 day^{-1})*	0.05	0.05
Water-sediment Fugacity Ratio (f_W/f_S)*	0.1	0.1
PCB Assimilation Efficiency from Food (E_D, %)[b]	62	62
Food Ingestion Rate (G_D, m^3 day^{-1})[c]	2.3×10^{-6}	2.3×10^{-6}
Food-sediment Fugacity Ratio (f_D/f_S)*	0.93	0.93
Density of Diet (δ_D, kg L^{-1})*	1	1
Organic Carbon Fraction of Diet (ϕ_D, %)[d]	0.33	0.75
Organic Carbon Assimilation Efficiency (α)[a]	0.5	0.5
Fraction of Ingested Diet Absorbed (β)[a]	0.05	0.05
Volume of Organism (V_B)[e]	0	0
Rate of PCB Biotransformation (K_M)[e]	0	0
Biota-water Partition Coefficient (K_{BW})[e]	0	0

*These parameter values were varied iteratively until a good fit to the experimental data was obtained.
[a]Data from (Morrison *et al.* 1996).
[b]Data from (Schweitzer *et al.* 2000).
[c]Data from (Moore 1966).
[d]Data from (Zeng *et al.* 1997).
[e]The biotransformation rate (K_M) for PCBs was assumed to be negligible (Morrison *et al.* 1996), and therefore all parameters in the same model term were set equal to zero as well.

≥80%) (Table 1). This result is in contrast to the results of Wood *et al.* (1997), who found significant differences (similarities ≤61%) between PCB patterns in aquatic insect larvae and those in the source sediments. This discrepancy suggests that epibenthic organisms, like sea urchins, may bioaccumulate contaminants differently than benthic infuanal organisms. It also highlights the importance of considering bioaccumulation on species-specific basis. Although the overall PCB patterns in the sediments and urchins were found to be virtually identical, affinity analysis did reveal some slight preferences for higher molecular weight and more hydrophobic congeners. Moreover, hydrophobicity appeared to be the primary driving force affecting the degree of bioaccumulation (Figure 1).

The multivariate statistical analysis of the overall shape of the BSAF curves provided another method for comparing the bioaccumulation results for the different sediments tested. The MANOVA analysis showed that the two lower concentration sediments (A and B) were statistically similar, and that both were statistically different from the higher concentration Sediment C (Figure 3). This was consistent with the results of the affinity analysis, which showed similar patterns for Sediments A and B, and significantly different pattern for Sediment C (Figure 1). This result may reflect an actual difference in

bioaccumulation patterns as a function of concentration, or it may reveal that the urchins had not yet reached their ultimate steady-state PCB concentrations for the higher concentration sediment. The latter explanation is supported by the results of a complementary paper (Zeng *et al.* 2003), which shows that the higher molecular weight PCBs had not yet reached their maximum steady-state concentration over the 42-d time frame of the exposure experiments.

Modeling of PCB Bioaccumulation in Sea Urchins

The general shape of the logBSAF versus log K_{OW} curves (Figure 2) dramatically shows the inadequacy of the EqP model for describing bioaccumulation data under these experimental conditions. The EqP model predicts that the logBSAF values should be approximately constant, and in the range of 0.0–0.3. The problem lies in the assumption of thermodynamic equilibrium, which is clearly not appropriate for the experiments in this study and also not appropriate for many natural environmental systems as well.

The model developed by Morrison *et al.* (1996) specifically to predict bioaccumulation of organic contaminants in benthic organisms did an excellent job of predicting the BSAF patterns of PCBs in the sea

urchins (Figure 2). Moreover, by adjusting the parameters to fit the experimental data, the model was able to accurately estimate the thermodynamic state of the system. Another important result of the model is that it was able to accurately predict the PCB bioaccumulation patterns in the urchins from the three different sediments using virtually the same model parameters, indicating a consistency of fundamental processes. The one parameter that was changed for the higher concentration sediment was the percent of organic carbon in the diet (f_D), which was twice as high in Sediment C compared to Sediments A and B. The higher organic carbon content of Sediment C was due to a higher percentage of New Bedford Harbor sediment in the sample required to produce a 10-fold higher total PCB concentration. Thus, the model shows that the differences in the BSAF curves are due to a change in the organic carbon content of the sediment, but not to any change in the fundamental bioaccumulation process.

Modification of the Bioaccumulation Model

One apparent shortfall with the nonequlibrium bioaccumulation model is its inability to predict the dropoff of the BSAF profiles as log K_{OW} becomes ≥6.5 (Figure 2), particularly for Sediment C (Figure 2c). Similar observations have been reported for the same bioaccumulation correlation with a deposit-feeding organism, *Yoldia limatula* (Lamoureux and Brownawell 1999), as well as for bioconcentration and biomagnification of PCBs in fish (Bremle *et al.* 1995, Fisk *et al.* 1998). It has been postulated that slow desorption of high-K_{OW} compounds from sediment may lower the bioavailability of the more hydrophobic PCBs, resulting in higher measured sediment concentrations than actually available for bioaccumulation (Lamoureux and Brownawell 1999). Other factors, such as lipid quality, molecular size (thus membrane permeability), elimination into feces, and inaccurate K_{OW}s, have been proposed to explain the observations (Bremle *et al.* 1995, Fisk *et al.* 1998). Interestingly, it appears that all of the proposed explanations are related to the hydrophobicity of the chemicals, and therefore a modification to the bioaccumulation model is presented here.

In modeling the bioaccumulation data (Figure 2), the rate of PCB biotransformation (K_M) was assumed to be zero (Table 2). If we instead designate K_M as an apparent decrease in the rate of PCB bioaccumulation in the biota, a correction can be made to the model. Based on the reasons stated above, we can assume K_M is proportional to K_{OW}, and therefore to K_{OC} (i.e., $K_M \sim K_{OC}$). The biota-water partition coefficient (K_{BW}) is also related to K_{OC}, and thus $K_{BW} \sim f_{lipid}K_{OW} \sim f_{lipid}K_{OC}$. Therefore, we can replace the biotransformation term in the Equation (4) with a hydrophobic term, $af_{lipid}V_B K_{OC}^2$, where a is a constant of proportionality. This term describes the unknown process(es) that seemingly prevent PCBs from bioaccumulating in proportion to their hydrophobicity. In this context, even the desorption of PCBs from sediments can be regarded as one of the processes affecting the actual PCB concentrations in the sea urchins by dictating the amount of bioavailable PCBs.

In the present study, f_{lipid} = 2.65%, 2.82%, and 2.02% for sea urchins exposed to Sediments A, B, and C, respectively (Zeng *et al.* 1997), and the average sea urchin volume (V_B) was estimated as 5.36x10⁻⁶ m³. Using the model parameters in Table 2 (except for V_B, K_M, and K_{BW}) and an iterative procedure, a new fit to the experimental data was obtained with a = 2.0x10⁻⁵ day⁻¹ for Sediments A and B and 7.4x10⁻⁵ day⁻¹ for Sediment C, respectively. The predicted curves from the modified bioaccumulation model essentially overlap with those from the unmodified model before log K_{OW} reaches 6.5, but the modified model was better at predicting the BSAF profiles with log K_{OW} ≥6.5 (Figure 2). The difference in a values, needed to best fit the different experimental data sets, suggests that the unconfirmed processes accounting for the observed drop-off in the BSAF profiles may be concentration dependent. This further supports the previously mentioned possibility that the higher molecular weight PCBs had not yet reached their maximum steady-state concentration in the urchins, and thus kinetic factors played a major role in shaping the bioaccumulation patterns.

The results from this study clearly show that the nonequlibrium, steady-state approach of Morrison *et al.* (1996) is technically sound, and is effective for predicting the bioaccumulation of organic chemicals in benthic organisms from sediments under any conditions, provided the appropriate input parameters are available or can be estimated. For compounds such as PCBs that are resistant to biotransformation within biota, this study demonstrates that a minor modification to the bioaccumulation model may be necessary to account for the lower bioaccumulation of very hydrophobic chemicals (log K_{OW} ≥6.5) due to some unidentified process(es) apparently related to the

hydrophobicities and concentrations of the chemicals. Such nonequilibrium, steady-state models should provide an effective tool for understanding which parameters and processes are most important for the bioaccumulation of organic contaminants from aquatic sediments. A thorough understanding of bioaccumulation processes is a necessary prerequisite to developing effective measures and criteria to eliminate or minimize any potential harm stemming from POPs.

LITERATURE CITED

Anderson, B.S., J.W. Hunt, B.M. Phillips, R. Fairey, C.A. Roberts, J.M. Oakden, H.M. Puckett, M. Stephenson, R.S. Tjeerdema, E.R. Long, C.J. Wilson and J.M. Lyons. 2001. Sediment quality in Los Angeles Harbor, USA: A triad assessment. *Environmental Toxicology and Chemistry* 20: 359-370.

Barron, M.G. 1990. Bioconcentration. *Environmental Science and Technology* 24: 1612-1618.

Bierman, V.J., Jr. 1990. Equilibrium partitioning and biomagnification of organic chemicals in benthic animals. *Environmental Science and Technology* 24: 1407-1412.

Boese, B.L., H. Lee, II and S. Echols. 1997. Evaluation of a first-order model for the prediction of the bioaccumulation of PCBs and DDT from sediment into the marine deposit-feeding clam *Macoma nasuta. Environmental Toxicology and Chemistry* 16: 1545-1553.

Bremle, G., L. Okla and P. Larsson. 1995. Uptake of PCBs in fish in a contaminated river system: Bioconcentration factors measured in the field. *Environmental Science and Technology* 29: 2010-2015.

Clark, K.E., F.A.P.C. Gobas and D. Mackay. 1990. Model of organic chemical uptake and clearance by fish from food and water. *Environmental Science and Technology* 24: 1203-1213.

Connolly, J.P. and C.J. Pedersen. 1988. A thermodynamic-based evaluation of organic chemical accumulation in aquatic organisms. *Environmental Science and Technology* 22: 99-103.

Di Toro, M.D., C.S. Zarba, D.J. Hansen, W.J. Berry, R.C. Swartz, C.E. Cowan, S.P. Pavlou, H.E. Allen, N.A. Thomas and P.R. Paquin. 1991. Technical basis for establishing sediment quality criteria for nonionic organic chemicals using equilibrium partitioning. *Environmental Toxicology and Chemistry* 10: 1541-1583.

Fairey, R., J. Downing, C. Roberts, E. Landrau, J. Hunt, B. Anderson, C.J. Wilson, G. Kapahi, F. LaCaro, P. Michael, M. Stephenson and M. Puckett. 1998. Chemistry, toxicity and benthic community conditions in sediments of the San Diego Region. Final Addendum Report. San Diego, CA.

Fisk, A.T., R.J. Norstrom, C.D. Cymbalisty and D.J. Muir. 1998. Dietary accumulation and depuration of hydrophobic organochlorines: Bioaccumulation parameters and their relationship with the octanol/water partition coefficient. *Environmental Toxicology and Chemistry* 17: 951-961.

Kraaij, R., W. Seinen, J. Tolls, G. Cornelissen and A.C. Belfroid. 2002. Direct evidence of sequestration in sediments affecting the bioavailability of hydrophobic organic chemicals to benthic organisms. *Environmental Science and Technology* 36: 3525-3529.

Lamoureux, E.M. and B.J. Brownawell. 1999. Chemical and biological availability of sediment-sorbed hydrophobic organic contaminants. *Environmental Toxicology and Chemistry* 18: 1733-1741.

Lee, H., II. 1992. Models, Muddles and mud: Predicting the bioaccumulation of sediment-associated pollutants. pp. 267-293 *in*: G.A. Burton (ed.), Sediment Toxicity Assessment. Lewis. Chelsea, MI.

Mackay, D. 1982. Correlation of bioconcentration factors. *Environmental Science and Technology* 16: 274-178.

Mackay, D. and S. Paterson. 1981. Calculating fugacity. *Environmental Science and Technology* 15: 1006-1014.

Means, J.C. and A.E. McElroy. 1997. Bioaccumulation of tetrachlorobiphenyl and hexachlorobiphenyl congeners by *Yoldia Limatula* and *Nephtys Incisa* from bedded sediments: Effects of sediment- and animal-related parameters. *Environmental Toxicology and Chemistry* 16: 1277-1286.

Moore, H.B. 1966. Chapter 4: Ecology of echinoids. pp. 822 *in*: R.A. Boolootian (ed.), Physiology of Echinodermata. Interscience Publishers. New York, NY.

Morrison, H.A., F.A.P.C. Gobas, R. Lazar and G.D. Haffner. 1996. Development and verification of a bioaccumulation model for organic contaminants in benthic invertebrates. *Environmental Science and Technology* 30: 3377-3384.

Newman, M.C. 1994. Chapter 3: Bioaccumulation. pp. 448 *in*, Quantitiative Methods in Aquatic Ecotoxicology. Lewis Publishers. Boca Raton, FL.

Schwarzenbach, R.P., P.M. Gschwend and D.M. Imboden. 1993. Environmental Organic Chemistry (1st edition). John Wiley & Sons, Inc. New York, NY.

Schweitzer, L.E., S.M. Bay and I.H.M. Suffet. 2000. Dietary assimilation of polychlorinated biphenyls in adult sea urchins (*Lytechinus pictus*) and maternal transfer to offspring. *Environmental Toxicology and Chemistry* 19: 1919-1924.

Sharpe, S. and D. Mackay. 2000. A framework for evaluating bioaccumulation in food webs. *Environmental Science and Technology* 34: 2373-2379.

Shea, D. 1988. Developing national sediment quality criteria. *Environmental Science and Technology* 22: 1256-1261.

Stull, J.K., D.J.P. Swift and A.W. Niedoroda. 1996. Contaminant dispersal on the Palos Verdes continental margin: I. Sediments and biota near a major California wastewater discharge. *The Science of the Total Environment* 179: 73:90.

Suffet, I.H.M., C.T. Jafvert, J. Kukkonen, M.R. Servos, A. Spacie, L.L. Williams and J.A. Noblet. 1994. Synopsis of discussion session: Influences of particulate and dissolved material on the bioavailability of organic compounds. pp. 93-108 *in*: W.H. Benson (ed.), Bioavailability - Physical, Chemical, and Biological Interactions. Lewis Publishers, CRC Press, Inc. Boca Raton, FL.

Thomann, R.V. 1989. Bioaccumulation model of organic chemical distribution in aquatic food chains. *Environmental Science and Technology* 23: 699-707.

Vanier, C. and D. Planas. 2001. Equilibrium partition theory applied to PCBs in macrophytes. *Environmental Science and Technology* 35: 4830-4833.

Weaver, G. 1984. PCB contamination in and around New Bedford, Mass. *Environmental Science and Technology* 18: 22A-27A.

Wong, C.S., P.D. Capel and L.H. Nowell. 2001. National-scale, field-based evaluation of the biota-sediment accumulation factor model. *Environmental Science and Technology* 35: 1709-1715.

Wood, L.W., P. O'Keefe and B. Bush. 1997. Similarity analysis of PAH and PCB bioaccumulation patterns in sediment-exposed *Chronomus tentans* Larvae. *Environmental Toxicology and Chemistry* 16: 283-292.

Zeng, E.Y., S.M. Bay, D. Greenstein, C. Vista, C.C. Yu and K.J. Ritter. 2003. Toxic effects of polychlorinated biphenyl bioaccumulation in sea urchins exposed to contaminated sediments. *Environmental Toxicology and Chemistry* (in press).

Zeng, E.Y., S.M. Bay, K. Tran and C. Alexander. 2001. Temporal and spatial distributions of contaminants in sediments of Santa Monica Bay, California. pp. 96-109 *in*: S.B. Weisberg and D. Elmore (eds.), Southern California Coastal Water Research Project Annual Report 1999-2000. Southern California Coastal Water Research Project. Westminster, CA.

Zeng, E.Y., S.M. Bay, C. Vista, C. Yu and D. Greenstein. 1997. Bioaccumulation and toxicity of polychlorinated biphenyls in sea urchins exposed to contaminated sediments. pp. 79-89 *in*: S.B. Weisberg, C. Francisco and D. Hallock (eds.), Southern California Coastal Water Research Project. Westminster, CA.

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