SETAC PRESS

Environmental Toxicology and Chemistry, Vol. 22, No. 11, pp. 2719–2726, 2003

Printed in the USA
0730-7268/04 $12.00 + .00

EVALUATION OF POLYCHLORINATED BIPHENYL BIOACCUMULATION PATTERNS IN WHITE SEA URCHINS (*LYTECHINUS PICTUS*) USING MULTIPLE APPROACHES

JAMES A. NOBLET,* EDDY Y. ZENG, and KERRY J. RITTER
Southern California Coastal Water Research Project, 7171 Fenwick Lane, Westminster, California 92683, USA

(*Received* 1 *November* 2002; *Accepted* 10 *March* 2003)

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 on 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 within 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.

Keywords—Polychlorinated biphenyl Bioaccumulation Sea urchin Equilibrium partition theory Nonequilibrium modeling

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 [1–3]. 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 [1,4–7]. Benthic organisms accumulate POPs because they live in intimate contact with and often feed on 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 [8].

The most commonly used approaches for evaluating bioaccumulation data are based on the equilibrium partitioning (EqP) model [3,8–11], which assumes that chemcials partition between sediment organic carbon, pore water, 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,

$$\text{BSAF} = \frac{(C_{\text{biota}}/f_{\text{lipid}})}{(C_{\text{sed}}/f_{\text{OC}})} \quad (1)$$

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 to 2, or 0 to 0.3 on a log scale [1,11].

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 [1,12]. In addition, BSAFs frequently exhibit a parabolic dependence on K_{OW} [1,13–16]. 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 nondietary 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 [17,18]. 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 [19]. In cases where the equilibrium assumption is invalid, it

* To whom correspondence may be addressed (jnoblet@csusb.edu). The current address of J. A. Noblet is Chemistry Department, California State University-San Bernardino, San Bernardino, CA 92407, USA.

is necessary to employ the use of bioaccumulation models that make no assumptions regarding the thermodynamic state of the system [1,18,20–22].

Polychlorinated biphenyls are synthetic chemicals produced as mixtures of individual congeners. The production of PCBs was phased out in 1977 in the United States because of concerns about their potential toxicity to wildlife and humans, but approximately 600 million kg of PCBs had already been manufactured [23]. Because of their resistance to chemical, physical, and biological degradation, PCBs can remain intact for long periods after discharge to the environment. In southern California, USA, coastal sediments in Santa Monica Bay, Los Angeles and Long Beach Harbors, and San Diego Bay and on the Palos Verdes Shelf are known to contain elevated levels of PCBs [24–27]. The presence of these potential carcinogens in California's coastal sediments was an important factor in the development and implementation of a new regulatory initiative, the Total Maximum Daily Load program, in California (San Diego RWQCB 1998 303[d] Water bodies: http://www.swrcb.ca.gov/tmdl/docs/segments/region9/table1_1998_303d.xls). Obviously, quantitative information about the bioaccumulation of PCBs by marine invertebrates is highly relevant in both tracing the fate of PCBs in the environment and developing effective regulatory measures to minimize the effects of PCBs on ecological and human health.

The objectives of the present study were to determine whether the PCB bioaccumulation patterns in the sea urchins were the same as those observed in the source sediments, determine if the concentration of the contaminants affects bioaccumulation patterns, and identify, if possible, an appropriate model for predicting the bioaccumulation of hydrophobic organic contaminants from sediments by benthic marine organisms. To this end, we evaluated the bioaccumulation of PCBs in the white sea urchin (*Lytechinus pictus*) from amended field-contaminated sediments using data derived from a previously reported study [16]. The white sea urchin was selected because it is a common marine epibenthic invertebrate and a voracious omnivore that lives and feeds on virtually any organic matter at the interface between the sediments and the overlying water column. Moreover, a recent regional survey found that the white sea urchin was the most abundant megabenthic invertebrate in the Southern California Bight [28]. Several different statistical and theoretical approaches were used to evaluate the data, including similarity and affinity analysis, regression analysis and multivariate statistics, EqP theory, and a nonequilibrium bioaccumulation model.

MATERIALS AND METHODS

Sea urchin exposure experiments

The experimental setup consisted of 20 polyethylene chambers, approximately 29 cm long × 26 cm wide × 14 cm deep. 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 weight, respectively. The sediments were prepared by mixing highly contaminated sediment (~3,800 mg/g dry wt total PCBs) from New Bedford Harbor (MA, USA), 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/min, 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, it was calculated that the entire seawater volume in each chamber was replaced approximately four to five times each day.

The urchins used for these experiments were collected from a coastal reference area near Dana Point. Fifteen urchins were introduced into each chamber and left undisturbed for a period of 42 d (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 fed by ingesting sediment, which was amended with Argent Hatchfry microencapsulated food (Redmond, WA, USA) at a rate of 0.36 g per exposure chamber every other day. Five sets of animals, related sediment, and interstitial 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. The unexposed sea urchins contained no detectable PCBs, and the concentrations of PCBs measured at the beginning and the end of the exposure experiments were not statistically different [16]. Therefore, the results are not reported herein. Sea urchins exposed to the reference sediment (R) were analyzed along with the other samples and 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 [16] and will be only 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 into a flask and dried with sodium sulfate. The centrifuge bottles were extracted twice more with hexane and the solvent layers 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 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 a gas chromatograph/electron capture detector system with selected confirmation by a gas chromatograph/mass spectrometer system. The instrument was calibrated using a 1:1:1:1 mixture of Aroclors® 1242, 1248, 1254, and 1260 (Monsanto, St. Louis, MO, USA). 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.

The performance of the analytical methods was monitored through measurements of the recoveries of surrogate standards spiked into each sample prior to extraction. The recoveries of three surrogate standards, tetrachloro-*m*-xylene, PCB 65, and PCB 189, were 105 ± 40%, 120 ± 35%, and 99 ± 22% for sediment samples and 121 ± 13%, 133 ± 18%, and 114 ± 9% for urchin samples. Because some of the samples (sediments B and C and sea urchins exposed to them) often contained PCB concentrations that were out of the calibration range and hence dilution of the sample extracts was required to ensure proper quantitation, this process may have introduced unexpected variability in the surrogate standard recoveries.

Data analysis

Similarity and affinity analysis. The measured PCB patterns in the sea urchins were compared to those in the sediments using similarity analysis. In a recent study, Wood et al. [29] investigated the bioaccumulation of PCBs and polycyclic aromatic hydrocarbons in aquatic insect larvae by treating the groups of contaminants as if they were ecological communities. The individual compounds within a chemical mixture and the chemical concentrations were considered analogous to the number of species and abundance, respectively. 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 were quantified by calculating the Bray–Curtis similarity coefficients [29] for each sediment–urchin paired data set. The general equation used was

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

where S_{jk} is the percentage 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 (number of samples containing the congener/total number of samples). The parameter D was modified by using the relative concentration (C_i/C_{total}) 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. Apparently, similarity values tend to plateau at 82% based on numerous previous studies [29], and samples with similarity values ≥82% are considered essentially identical.

The difference in the relative uptake of individual PCB congeners or selective bioaccumulation was also investigated. Again, by analogy to ecology, affinity analysis was used to quantify the degree of selectivity for certain PCB congeners or groups of congeners [29]. The bioaccumulation affinity, A_i, for an individual PCB congener was calculated using the equation

$$A_i = \frac{\frac{r_i}{n_i}}{\frac{1}{m}\sum_{j=1}^{m}\frac{r_j}{n_j}} \quad (3)$$

where r_i is the proportion of contaminant i in the organism, n_i is the proportion of congener i in the source (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 log BSAF versus log K_{OW} curves for the three amended sediments were compared to ascertain whether concentration had any affect on PCB bioaccumulation. All the log K_{OW} values for the PCB congeners used in this study were obtained from Hawker and Connell [30]. 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 log BSAF 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 [1]. In this study, the form of the model developed specifically for detritivores was used:

$$\frac{f_B}{f_S} = \frac{E_W G_W \frac{f_W}{f_S} + E_D G_D \frac{f_D}{f_S} \delta_D \phi_D K_{OC}}{E_W G_W + E_D(1-\alpha)(1-\beta) G_D \delta_D \phi_D K_{OC} + V_B K_M K_{BW}} \quad (4)$$

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 α and β describe dietary assimilation efficiencies and are the fraction of organic carbon removed on ingestion and the fraction of the ingested diet absorbed by the organism, respectively. The parameters δ_D and ϕ_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 $0.41K_{OW}$ [31]. 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. [1] 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

Table 1. Results of the similarity analysis of polychlorinated biphenyl patterns between the indicated sediments and sea urchins. Similarities ≥82% indicate identical polychlorinated biphenyls patterns

Sample	Concn. (μg/g)	Similarity (%)	SD[a]	*n*
Sediment A	4	79.9	1.4	5
Sediment B	10	83.6	0.9	5
Sediment C	100	86.7	1	5

[a] SD = standard deviation.

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.

Affinity analysis was also performed to determine if 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 (International Union of Pure and Applied Chemistry number). To facilitate further comparison, the data for the PCB congeners were grouped according to homologs (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 (Fig. 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; Fig. 1a and b) and much less evident for the high concentration sediment C (Fig. 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 (Fig. 1c).

Only three congeners 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 (Fig. 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 coeluted 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 log BSAF values were calculated for each sediment–urchin congener pair and plotted versus their corresponding log K_{OW} values (Fig. 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.

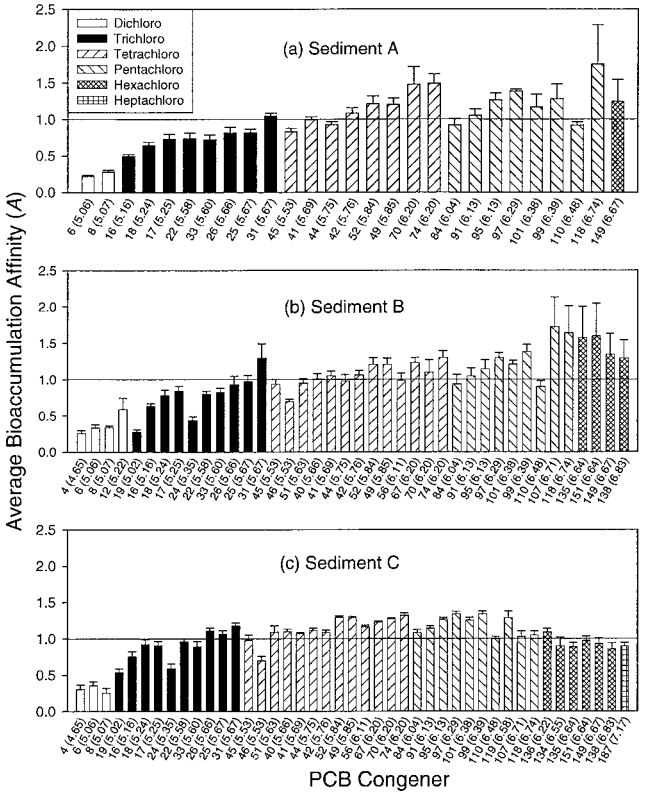

Fig. 1. Average bioaccumulation affinities with respect to polychlorinated biphenyl (PCB) congener number (numbers in parentheses are log K_{OW} values) for (**a**) sediment A, (**b**) sediment B, and (**c**) sediment C. The error bars are standard deviations of the five-replicate analyses.

The R^2 values for each of the replicates ranged from 0.61 to 0.70 (Fig. 2). A MANOVA was then performed on the three-dimensional response vector to test the effect of starting material on log BSAF profiles. The MANOVA resulted in a p value <0.0001 from Wilke's lambda. Pairwise 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} (Fig. 3). This result at first appears to be counterintuitive since the log K_{OC} values should increase with increasing log K_{OW} as predicted by well-established empirical relationships [31,32]. However, Di Toro et al. [9] have shown that plots of this appearance result from not accounting for dissolved organic matter (DOM) in the interstitial water. The DOM 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.

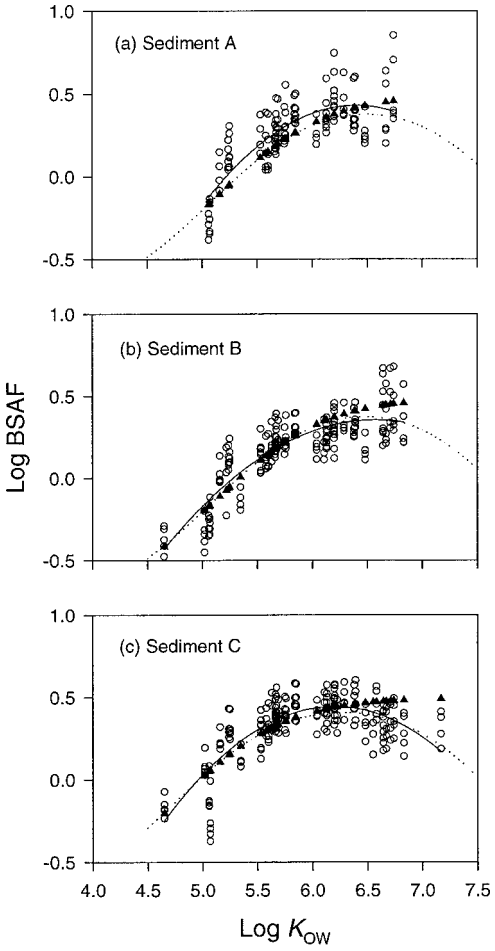

Fig. 2. Profiles of log biota–sediment accumulation factor (BSAF) versus log K_{OW} for white sea urchins on exposure to (**a**) sediment A, (**b**) sediment B, and (**c**) sediment C. ○ = measured log BSAF values; ▲ = log BSAF values predicted using the bioaccumulation model (Eqn. 4). The solid curve 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 ($af_{lipid}V_BK_{OC}^2$) related to some unknown hydrophobicity-dependent processes. Fitted equations describing the solid lines take the following forms: sediment A: $y = -0.31x^2 + 4.0x - 12.2$ ($R^2 = 0.61$); sediment B: $y = -0.21x^2 + 2.8x - 8.8$ ($R^2 = 0.70$); and sediment C: $y = -0.29x^2 + 3.6x - 10.6$ ($R^2 = 0.61$).

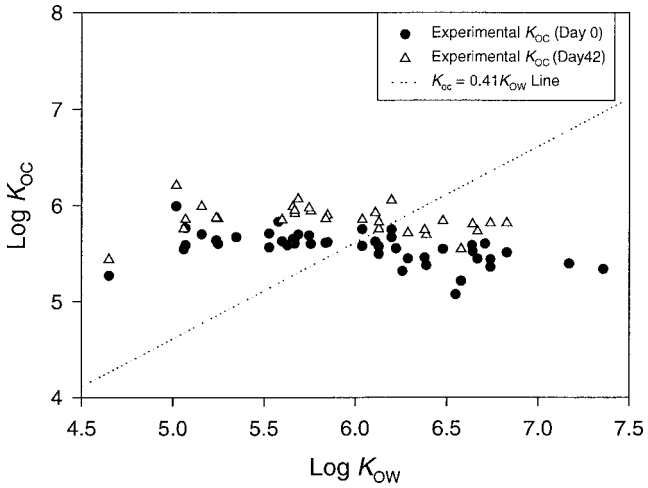

Fig. 3. Measured log K_{OC} values calculated from the sediment and interstitial water polychlorinated biphenyl concentrations for sediment C. Also shown is the predicted log K_{OC} line calculated from the K_{OW} values [30], using the empirical relationship $K_{OC} = 0.41K_{OW}$ [31].

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 3. Because the presence of DOM would enhance the measured concentrations of PCB congeners in the dissolved phase, correcting for the effect of DOM would have increased the log K_{OC} values. Therefore, apparent log K_{OC} data would be expected to plot below the empirical log K_{OC} line if the system was measured at equilibrium. The fact that about half 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 if the sea urchin bioaccumulation results could be correctly predicted using the benthic bioaccumulation model (Eqn. 4) developed by Morrison et al. [1]. A literature review was conducted, and this was identified as the most recent, most sophisticated and comprehensive model available for predicting the bioaccumulation of organic contaminants by benthic organisms. 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 determined 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 (Fig. 2).

All the final model parameters (Table 2) obtained by the iterative variation process were evaluated as to their reasonableness based on 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 [1]. Given that sea urchins have a greater surface area for ventilation (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 [33]. 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 con-

Table 2. Final model input parameters used to calculate biota–sediment accumulation factor values

Model parameters	Sediments A and B	Sediment C
PCB assimilation efficiency from water (E_W, %)[a]	100	100
Water ventilation rate (G_W, m^3/d)[b]	0.05	0.05
Water–sediment fugacity ratio (f_W/f_S)[b]	0.1	0.1
PCB assimilation efficiency from food (E_D, %)[c]	62	62
Food ingestion rate (G_D, m^3/d)[d]	2.3×10^{-6}	2.3×10^{-6}
Food–sediment fugacity ratio (f_D/f_S)[b]	0.93	0.93
Density of diet (δ_D, kg/L)[b]	1	1
Organic carbon fraction of diet (ϕ_D, %)[e]	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)[f]	0	0
Rate of PCB biotransformation (K_M)[f]	0	0
Biota–water partition coefficient (K_{BW})[f]	0	0

[a] Data from Morrison et al. [1].
[b] These parameter values were varied iteratively until a good fit to the experimental data was obtained.
[c] Data from Schweitzer et al. [35].
[d] Data from H. Moore [33].
[e] Data from Zeng et al. [34].
[f] The biotransformation rate (K_M) for polychlorinated biphenyls (PCBs) was assumed to be negligible [1], and therefore all parameters in the same model term were set equal to zero as well.

sidering that losses due to volatilization and uptake by the urchins were not taken into account.

The relative importance of dietary and nondietary PCB uptake can be estimated from evaluation of the model. If all terms related to dietary uptake are eliminated, the model reduces to $f_B/f_S = f_W/f_S$, or $f_B/f_S = 0.1$. Since BSAF = $1.6f_B/f_S$, the BSAF is reduced to a constant value of 0.16 (log BSAF = −0.79). If the model parameters are correct and the predicted extent of nonequilibrium is valid, then the results show that the uptake of PCBs by the sea urchins was due entirely dietary uptake and that bioaccumulation from the dissolved phases was negligible.

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 ≥80%; Table 1). This result is in contrast to the results of Wood et al. [29], 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 (Fig. 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 (Fig. 2). 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 (Fig. 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 complimentary paper [16], 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 log BSAF versus log K_{OW} curves (Fig. 2) dramatically shows the inadequacy of the EqP model for describing bioaccumulation data under these experimental conditions. The EqP model predicts that the log BSAF values should be approximately constant and in the range of 0.0 to 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. [1], 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 (Fig. 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 percentage organic carbon in the diet (ϕ_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 processes.

Modification of the bioaccumulation model

One apparent shortfall with the nonequilibrium bioaccumulation model is its inability to predict the drop-off of the BSAF profiles as log K_{OW} becomes ≥6.5 (Fig. 2), particularly for sediment C (Fig. 2c). Similar observations have been reported for the same bioaccumulation correlation with a deposit-feeding organism, *Yoldia limatula* [14], as well as for bioconcentration and biomagnification of PCBs in fish [13,15]. 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 [14]. 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 [13,15]. Interestingly, it appears that all the proposed explanations can be related to the hydrophobicity of the chemicals, and therefore a modification to the bioaccumulation model is presented here.

In modeling the bioaccumulation data (Fig. 2), the rate of PCB biotransformation (K_M) was assumed to be zero (Table 2). If we instead designate K_M as a term describing the apparent decrease in the rate of PCB bioaccumulation in the biota, a correction can be made to the model. Based on the reasons stated previously, we can assume K_M is proportional to K_{OW} and therefore to K_{OC} ($K_M \sim K_{OC}$). 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 (K_M) in Equation 4 with a hydrophobic term, $af_{lipid}V_BK_{OC}{}^2$, where a is a constant of proportionality. This term describes an unknown process(es) that seemingly prevents PCBs with K_{OW} >6.5 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 [16,34], and the average sea urchin volume (V_B) was estimated as 5.36 × 10^{-6} m^3. 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 = 2.0 × 10^{-5}/d for sediments A and B and 7.4 × 10^{-5}/d 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 $K_{OW} \geq 6.5$ (Fig. 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 nonequilibrium, steady-state approach of Morrison et al. [1] 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 $K_{OW} \geq 6.5$) 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. And 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.

Acknowledgement—The authors are grateful to Harold Stubbs and Dario Diehl for the collection of the local sediments and sea urchins used in this study. We thank Robert Burgess (U.S. Environmental Protection Agency, Narragansett, RI) for the New Bedford Harbor sediment. We also want to thank the following present and former SCCWRP personnel for their technical expertise and assistance in generating the data presented herein: S. Bay, C. Vista, C. Yu, D. Greenstein, J. Brown, A. Jirik, A. Khan, K. Tran, and L. Schweitzer. Finally, we are very grateful for the constructive comments provided by anonymous reviewers that helped improve the quality of this paper.

REFERENCES

1. Morrison HA, Gobas FAPC, Lazar R, Haffner GD. 1996. Development and verification of a bioaccumulation model for organic contaminants in benthic invertebrates. *Environ Sci Technol* 30:3377–3384.
2. Barron MG. 1990. Bioconcentration. *Environ Sci Technol* 24: 1612–1618.
3. Bierman VJ Jr. 1990. Equilibrium partitioning and biomagnification of organic chemicals in benthic animals. *Environ Sci Technol* 24:1407–1412.
4. Clark KE, Gobas FAPC, Mackay D. 1990. Model of organic chemical uptake and clearance by fish from food and water. *Environ Sci Technol* 24:1203–1213.
5. Connolly JP, Pedersen CJ. 1988. A thermodynamic-based evaluation of organic chemical accumulation in aquatic organisms. *Environ Sci Technol* 22:99–103.
6. Thomann RV. 1989. Bioaccumulation model of organic chemical distribution in aquatic food chains. *Environ Sci Technol* 23:699–707.
7. Sharpe S, Mackay D. 2000. A framework for evaluating bioaccumulation in food webs. *Environ Sci Technol* 34:2373–2379.
8. Shea D. 1988. Developing national sediment quality criteria. *Environ Sci Technol* 22:1256–1261.
9. Di Toro MD, Zarba CS, Hansen DJ, Berry WJ, Swartz RC, Cowan CE, Pavlou SP, Allen HE, Thomas NA, Paquin PR. 1991. Technical basis for establishing sediment quality criteria for nonionic organic chemicals using equilibrium partitioning. *Environ Toxicol Chem* 10:1541–1583.
10. Vanier C, Planas D. 2001. Equilibrium partition theory applied to PCBs in macrophytes. *Environ Sci Technol* 35:4830–4833.
11. Wong CS, Capel PD, Nowell LH. 2001. National-scale, field-based evaluation of the biota-sediment accumulation factor model. *Environ Sci Technol* 35:1709–1715.
12. Kraaij R, Seinen W, Tolls J, Cornelissen G, Belfroid AC. 2002. Direct evidence of sequestration in sediments affecting the bioavailability of hydrophobic organic chemicals to benthic organisms. *Environ Sci Technol* 36:3525–3529.
13. Fisk AT, Norstrom RJ, Cymbalisty CD, Muir DJ. 1998. Dietary accumulation and depuration of hydrophobic organochlorines: Bioaccumulation parameters and their relationship with the octanol/water partition coefficient. *Environ Toxicol Chem* 17:951–961.
14. Lamoureux EM, Brownawell BJ. 1999. Chemical and biological availability of sediment-sorbed hydrophobic organic contaminants. *Environ Toxicol Chem* 18:1733–1741.
15. Bremle G, Okla L, Larsson P. 1995. Uptake of PCBs in fish in a

contaminated river system: Bioconcentration factors measured in the field. *Environ Sci Technol* 29:2010–2015.
16. Zeng EY, Bay SM, Greenstein D, Vista C, Yu CC, Ritter KJ. 2003. Toxic effects of polychlorinated biphenyl bioaccumulation in sea urchins exposed to contaminated sediments. *Environ Toxicol Chem* 22:1065–1074.
17. Suffet IHM, Jafvert CT, Kukkonen J, Servos MR, Spacie A, Williams LL, Noblet JA. 1994. Synopsis of discussion session: Influences of particulate and dissolved material on the bioavailability of organic compounds. In Benson WH, ed, *Bioavailability—Physical, Chemical, and Biological Interactions.* Lewis, Boca Raton, FL, USA, pp 93–108.
18. Means JC, McElroy AE. 1997. Bioaccumulation of tetrachlorobiphenyl and hexachlorobiphenyl congeners by *Yoldia limatula* and *Nephtys incisa* from bedded sediments: Effects of sediment- and animal-related parameters. *Environ Toxicol Chem* 16:1277–1286.
19. Mackay D. 1982. Correlation of bioconcentration factors. *Environ Sci Technol* 16:274–278.
20. Lee H II. 1992. Models, muddles and mud: Predicting the bioaccumulation of sediment-associated pollutants. In Burton GA, ed, *Sediment Toxicity Assessment.* Lewis, Boca Raton, FL, USA, pp 267–293.
21. Boese BL, Lee H II, Echols S. 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*. *Environ Toxicol Chem* 16:1545–1553.
22. Newman MC. 1994. Bioaccumulation—Chapter 3. In *Quantitative Methods in Aquatic Ecotoxicology*. Lewis, Boca Raton, FL, USA, pp 448.
23. Weaver G. 1984. PCB contamination in and around New Bedford, Mass. *Environ Sci Technol* 18:22A–27A.
24. Fairey R, Downing J, Roberts C, Landrau E, Hunt J, Anderson B, Wilson CJ, Kapahi G, LaCaro F, Michael P, Stephenson M, Puckett M. 1998. Chemistry, toxicity and benthic community conditions in sediments of the San Diego Region. Final Addendum Report. California Department of Fish and Game, Sacramento, CA, USA.
25. Stull JK, Swift DJP, Niedoroda AW. 1996. Contaminant dispersal on the Palos Verdes continental margin: I. Sediments and biota near a major California wastewater discharge. *Sci Total Environ* 179:73–90.
26. Zeng EY, Bay SM, Tran K, Alexander C. 2001. Temporal and spatial distributions of contaminants in sediments of Santa Monica Bay, California. In Elmore D, ed, *Southern California Coastal Water Research Project Annual Report 1999–2000.* Southern California Coastal Water Research Project, Westminster, CA, USA, pp 96–109.
27. Anderson BS, Hunt JW, Phillips BM, Fairey R, Roberts CA, Oakden JM, Puckett HM, Stephenson M, Tjeerdema RS, Long ER, Wilson CJ, Lyons JM. 2001. Sediment quality in Los Angeles Harbor, USA: A triad assessment. *Environ Toxicol Chem* 20:359–370.
28. Allen MJ, Moore SL, Schiff KC, Weisberg SB, Diener D, Stull JK, Groce A, Mubarak J, Tang CL, Gartman R. 1998. Southern California Bight Pilot Project: Volume V—Demersal fishes and megabenthic invertebrates. Southern California Coastal Water Research Project, Westminster, CA, USA.
29. Wood LW, O'Keefe P, Bush B. 1997. Similarity analysis of PAH and PCB bioaccumulation patterns in sediment-exposed *Chronomus tentans* larvae. *Environ Toxicol Chem* 16:283–292.
30. Hawker DW, Connell DW. 1988. Octanol-water partition coefficients of polychlorinated biphenyl congeners. *Environ Sci Technol* 22:382–387.
31. Karickhoff SW, Brown DS, Scott TA. 1979. Sorption of hydrophobic pollutants in natural sediments. *Water Res* 13:241–248.
32. Schwarzenbach RP, Gschwend PM, Imboden DM. 1993. *Environmental Organic Chemistry*. John Wiley, New York, NY, USA.
33. Moore HB. 1966. Chapter 4: Ecology of Echinoids. In Boolootian RA, ed, *Physiology of Echinodermata.* Interscience, New York, NY, USA, pp 822.
34. Zeng EY, Bay SM, Vista C, Yu C, Greenstein D. 1997. Bioaccumulation and toxicity of polychlorinated biphenyls in sea urchins exposed to contaminated sediments. In Hallock D, ed, *Southern California Coastal Water Research Project*. Westminster, CA, USA, pp 79–89.
35. Schweitzer LE, Bay SM, Suffet IHM. 2000. Dietary assimilation of polychlorinated biphenyls in adult sea urchins (*Lytechinus pictus*) and maternal transfer to offspring. *Environ Toxicol Chem* 19:1919–1924.