TOXICOKINETICS OF DDE, BENZO(A)PYRENE, AND 2, 4, 5, 2', 4', 5'-HEXACHLOROBIPHENYL IN *PONTOPOREIA HOYI* AND *MYSIS RELICTA*

Marlene S. Evans[1]
Center for Great Lakes and Aquatic Sciences
The University of Michigan
Ann Arbor, Michigan 48109

Peter F. Landrum
Great Lakes Environmental Research Laboratory
National Oceanographic and Atmospheric Administration
Ann Arbor, Michigan 48105

ABSTRACT. The toxicokinetics of DDE, benzo(a)pyrene (BaP), and 2, 4, 5, 2', 4', 5'-hexachlorobiphenyl (HB) were followed for the amphipod, Pontoporeia hoyi, *and the mysid,* Mysis relicta. Pontoporeia *and* Mysis *had similar uptake clearances (K_u) for DDE (mean = 79.2 mL/g/h and 46.0 mL/g/h, respectively), BaP (mean = 75.9 mL/g/h and 39.9 mL/g/h, respectively), and HB (mean = 53.5 mL/g/h and 57.5 mL/g/hr, respectively) compounds with log octanol-water partition coefficients (K_{ow}'s) ranging from 5.7 to 6.7. Amphipods and mysids were most efficient at eliminating BaP (mean K_d = -0.0017/h and -0.0047/h, respectively) and least efficient at eliminating HB (mean = -0.0008/h and -0.0001/h respectively). Amphipods were more efficient than mysids in eliminating DDE (mean = -0.0010/h and -0.0005/h, respectively) and HB while mysids were more efficient at eliminating BaP. Because K_d's for DDE, BaP, and HB were substantially different between* P. hoyi *and* M. relicta, *there were substantial differences in the calculated bioconcentration factors (BCFs). Amphipods tended toward larger BCFs than mysids for BaP (mean = 48,582 and 8,496, respectively) but lower BCFs for DDE (mean = 95,629 and 138,760, respectively) and HB (mean = 101,663 and 442,231, respectively) than mysids.*
ADDITIONAL INDEX WORDS: Bioconcentration, toxic substances, partition coefficient, benthos, benthic environment.

INTRODUCTION

Chemicals which are toxic, persistent, and readily biomagnified from one trophic level to another are a major environmental concern and require regulation. Compounds which exhibit such properties, e.g., DDT and polychlorinated biphenyls (PCB), can adversely affect ecosystem health, particularly for higher trophic levels. Moreover, some persistent compounds are of continued concern years after implementation of production bans, e.g., PCB and DDT in regions such as the Great Lakes. Of particular importance is the recycling of persistent contaminants from sedimentary sinks to the

pelagic region through resuspension and benthic-based food web dynamics.

The offshore benthic community of the Great Lakes is dominated by two species of macroinvertebrates: the amphipod, *Pontoporeia hoyi*, and the mysid, *Mysis relicta*, (Mozley and Howmiller 1977, Nalepa 1987). Because *P. hoyi* and *M. relicta* are relatively large invertebrates and account for the major proportion of benthic biomass, they are an important component in the diets of many species of Great Lakes fish (Scott and Crossman 1973). Similarly, because these macroinvertebrates contain moderately high concentrations of organic contaminants such as DDT and PCB (Evans *et al.* 1982, Borgmann and Whittle 1983), *P. hoyi* and *M. relicta* may represent a major pathway for transfer of persistent toxic organics through the

[1]Present address: Aquatic Ecology Division, National Hydrology Research Institute, 11 Innovation Boulevard, Saskatoon, Canada S7N 3H5

food web to higher trophic levels (Jensen *et al.* 1982, Breck and Bartell 1988).

M. relicta and *P. hoyi* exhibit important differences in their ecology and physiology. Mysids are epibenthic in habitat, residing near the sediment-water interface during the day and migrating to shallower depths at night where they feed upon the plankton. Amphipods are benthic and feed primarily upon detritus (Mozley and Howmiller 1977). Respiration rates are higher in *M. relicta* than in *P. hoyi* (Frez and Landrum 1986). Similarly, *P. hoyi* and *M. relicta* may differ in their ability to take up, eliminate, and bioconcentrate xenobiotics.

Several studies have examined the toxicokinetics of selected polycyclic aromatic hydrocarbons (PAH) in *P. hoyi* (Landrum *et al.* 1985; Landrum 1982, 1988), including an investigation of the major physiological and environmental factors that influence the toxicokinetics of these contaminants. A similar study was conducted of the toxicokinetics of benzo(a)pyrene (a representative PAH) for *M. relicta* (Frez and Landrum 1986). Most of the *P. hoyi* studies were based on amphipods collected from an approximate station depth of 29 m. Toxicokinetics exhibited a strong dependence on K_{ow}, exposure temperature, and *P. hoyi* size and lipid content (Landrum 1988). As a result of these modifying factors, there were seasonal patterns in the toxicokinetics of *P. hoyi* for the PAHs investigated (Frez and Landrum 1986, Landrum 1988). While fewer variables were examined for *M. relicta* (collected from the 60–70 m depth region), seasonal patterns were observed in the toxicokinetics of BaP. Exposure temperature, molt cycle, and related physiological processes appeared to be important variables affecting toxicokinetics (Frez and Landrum 1986).

Both *P. hoyi* and *M. relicta* are found at greater depths in the Great Lakes than the populations investigated by Frez and Landrum (1986) and Landrum (1988). As water column depth increases, environmental conditions at the sediment-water interface become more stable, e.g., there is less seasonal variability in near-bottom water temperature at 100 m than at 15 m. Moreover, *P. hoyi* and *M. relicta* may exhibit significant differences in their physiology between the deep and shallower regions of the lake. For instance, *P. hoyi* completes its life cycle in 1 year at 15-m depth but in 2 years at 42 m (Winnell and White 1984). At 100 m, *P. hoyi* requires 2.5 to 3 years to complete its life cycle (Lubner 1979). Moreover, deepwater (100 m) *P.*

hoyi contains lower amounts of total lipid (on a non-lipid, dry weight basis) than *P. hoyi* collected at 45 m (W. S. Gardner, personal communication, Great Lakes Environmental Research Laboratory, Ann Arbor, Michigan).

In this work, we extend the variety of toxic organic compounds studied in *P. hoyi* and *M. relicta* and use benzo(a)pyrene as a reference compound for comparison with previous work. The toxicokinetics of DDE, benzo(a)pyrene (BaP), and 2, 4, 5, 2′, 4′, 5′-hexachlorobiphenyl (HB) were measured over the course of two field seasons. DDE (k_{ow} = 5.7) is the predominant form of DDT in Lake Michigan ecosystem (Evans *et al.* 1982). BaP (k_{ow} = 6.5) and HB (k_{ow} = 6.7) are a common polycyclic aromatic hydrocarbon and PCB congener, respectively, in the Great Lakes ecosystem (Eadie *et al.* 1983, Oliver 1984, Smith *et al.* 1985). Our experiments permit comparisons between deep and shallow water populations of *P. hoyi*, provide new data on the toxicokinetics of DDE and HB, and improve interspecies comparisons between *M. relicta* and *P. hoyi*. Previous toxicokinetics studies of these organisms have focussed on PAHs.

MATERIALS AND METHODS

Amphipods and mysids were collected in southeastern Lake Michigan at a 97-m deep station approximately 18 km west of Grand Haven, Michigan (Evans *et al.* 1982). Mysids were collected during daylight hours by towing a 1-m diameter, 656 μm-mesh plankton net mounted in a 1-m square frame near the sediment-water interface. Mysids were very patchy in distribution and could not be collected in sufficient numbers for experimental studies during all cruise months. Sufficient numbers of mysids were collected in good condition during the 18 August and 16 September 1985 cruises and the 15 May, 24 June, and 14 July 1987 cruises. Immediately upon recovery of the net, mysids were transferred to an ice chest filled with hypolimnetic water.

During some cruises, large numbers of *P. hoyi* were collected during the mysid tows while, during other cruises, amphipods were collected with a PONAR grab. When the grab was used, lake water was added to the sediment sample and amphipods gently sieved and transferred to the ice chest containing the mysids. Amphipods were collected during the 13 June, 18 August, 16 September, and 9 October 1986 cruises and the 15 May, 24 June, 14 July, and 12 August 1987 cruises. Lake water was

collected during each cruise for use in experimental studies.

Amphipods and mysids were returned to Ann Arbor within 1 day of collection and immediately placed in a dark, 4°C cold room until used in the experiments. Experiments were conducted within 2 to 4 days after sample collection.

Dual-labelled experiments using DDE (^{14}C, specific activity 13.4 mCi/mM, Pathfinders Laboratories) and BaP (^{3}H, specific activity 23.8 Ci/mM, Amersham) were conducted in 1986 and 1987: August 1986 experiments were run only with DDE. HB (^{14}C, specific activity 14.06 mCi/mmole, Pathfinders Laboratories) experiments were conducted separately and only in 1987. Compounds were examined for radiopurity by using a combination of thin layer chromatography (TLC) and liquid scintillation counting (LSC). ^{3}H-BaP was purified when necessary by TLC (hexane:benzene 8:2, V:V) on silica gel plates: no purification was required for DDE and HB. All compounds were >98% radiopure when used.

Lake Michigan water was filtered through either 3 μm membrane (Gelman, AN-3000) or 0.45 μm HA (Millipore HAWP 293 25) filters, cooled overnight to 4°C, and labelled in bulk (5 L). Test compounds were dosed in methanol or acetone carrier to replicate test chambers. The concentration of carrier was <0.5 mL/L and was not expected to affect the kinetic measurements (Landrum 1983).

Uptake Experiments

In 1986, *P. hoyi* uptake experiments were conducted in quadruplicate in temperature-controlled (4°C) flow-through 200-mL chambers. Due to equipment limitations in 1987, experiments were conducted statically in 1-L beakers containing 500 mL of water: beakers were placed in a 4°C incubator. Approximately 25–40 amphipods were added to each of the experimental containers. Experiments were run under low-level red light (1986) or in the dark (1987) to minimize the response of *P. hoyi* to light. Beakers (in 1987) were covered with aluminum foil to minimize evaporative losses. For the flow-through experiments, water was passed into the test chambers at approximately 100 mL/hr. Two animals were removed from each replicate chamber (or beaker) at 1, 2, 4, and 6 h, blotted dry, weighed on preweighed aluminum foil boats on a Cahn electrobalance, and then placed in a labelled scintillation vial. Scintillation cocktail (ACS or SafetySol) was then added. Scintillation

cocktail is an effective extractant for non-polar organics compared with more conventional tissue solubilization techniques (Landrum *et al.* 1985).

A similar procedure was followed for the mysid experiments with two minor exceptions. Experiments were conducted in 6-L aquaria filled with 3-L dosed Lake Michigan water rather than the smaller containers used for the amphipod experiments. Aquaria were covered with aluminum foil and were maintained in the dark in a 4°C incubator. Ten to fifteen mysids were placed in each experimental container: one mysid per replicate was removed at each sampling period.

Water samples (2 mL) were removed from each replicate chamber at 0, 1, 2, 4, and 6 h. Each sample was placed in a scintillation vial and 12–15 mL of cocktail added. These samples provided estimates of the total activity in water over the various time periods. In order to estimate the sorption of the organic contaminant to dissolved organic matter (DOC), the reverse-phase separation technique (Landrum *et al.* 1984) was used. After the 4-h water sample was collected from each aquarium, a second 2-mL aliquot of water was passed through a C-18 Sep Pak (Waters Associates) into a scintillation vial and cocktail added. The total activity in each of these samples provided an estimate of the amount of contaminant bound to DOC. The freely dissolved xenobiotic concentration was then calculated by subtracting the activity associated with the bound fraction from the total activity associated with the whole water sample.

Elimination Experiments

Elimination experiments were performed by transferring the remaining animals from the uptake experiments to 6-L aquaria containing undosed, unfiltered lake water. For the amphipod experiments, sieved, 97-m lake sediments were added shortly before the elimination studies commenced. As a benthic organism, *P. hoyi* is found primarily within the sediments (Krezoski *et al.* 1978). Sediments were added to more closely approximate the environmental conditions under which elimination would occur. Because *M. relicta* is an epibenthic species (Mozley and Howmiller 1977), no sediments were added to the mysid elimination chambers. Mysids were not fed although particulates in the unfiltered lake water may have provided a limited amount of nutrition. Aquaria were maintained in a dark, cold (4°C) incubator. Elimination exper-

iments were conducted over 10–26 days. Time periods were extended over the course of the study when it became apparent that DDE and HB were not readily eliminated. Four replicates, each consisting of one *M. relicta* or two *P. hoyi*, were removed at several (5 to 6) time intervals over the course of the study. Animals were analyzed as in the uptake experiments, i.e., weighed, placed in scintillation vials, and cocktail added.

[14]C and ^{3}H activities were assayed on a Packard 460C liquid scintillation counter. Samples were corrected for quench using the external standards ratio method after subtracting background. All concentrations of the experimentally-added xenobiotics in animals and water were based solely on ^{14}C or ^{3}H activity. Previous studies have shown no transformation of BaP by *P. hoyi* over the length of time required for measuring uptake (Landrum 1982, 1983, 1988).

Kinetics and Statistics

The uptake clearance constants for BaP, DDE, and HB were determined by using a one-compartment model.

$$C_a = K_u C_w t,$$

where C_a = concentration in animal (dpm/g wet weight)

C_w = corrected concentration in water (dpm/mL)

K_u = uptake clearance constant (mL/g wet weight/h)

t = time (h)

dpm = disintegrations per minute

Uptake clearance refers to the volume of water "cleared" of the test compound per hour per unit wet weight of the test organism. Thus, it is roughly analogous to the term used in quantifying filtering rate for zooplankton. The term and units are the proper expressions to relate the flux of contaminant into an organism from the water (Wilkinson 1987).

The model assumes that elimination is unimportant over the time period for which the clearance constant is calculated and that water concentration remains constant. Compound concentrations decreased only slightly over the course of the uptake experiments averaging 20.7%, 13.7%, and 11.5% for the BaP, DDE, and HB *P. hoyi* experiments respectively. Smaller average

losses (11.0%, 9.6%, and 4.0% for BaP, DDE, and HB respectively) were observed for the *M. replicta* experiments. Lower loss rates in the mysid experiments may have been associated with container effects, i.e., relatively less adsorptive losses occurred in the larger mysid aquaria than in the smaller chambers and beakers used for the amphipod studies. A proportionately lower adsorptive loss to container walls occurs with increasing container size because the surface area to volume ratio decreases with increasing volume. Elimination rates were very slow and less than 5% of the elimination half-life occurred over the course of the uptake experiment.

Plots of C_a versus $C_w t$ were linear for all 4-h data and for most 6-h data. In instances where the curves were not linear when 6-h data were included, K_u calculations were based only on the 1–4 h uptake measurements. Xenobiotic concentrations in water were corrected for the fraction bound to dissolved organic matter.

The elimination constants were determined by fitting the elimination data to a first order decay model

$$C_a(t) = C_a(0)e^{-K_d t}$$

where $C_a(0)$ = initial concentration in animal (dpm/g)

K_d = elimination rate constant (/h)

t = time (h)

The contaminant bioconcentration factor (BCF) was estimated by dividing K_u by K_d based on the two-compartment donor dependent

$$C_a = K_u C_w (1-e^{K_d t})/K_d$$

When biotransformation is negligible over large values of t, the equation simplifies to

$$C_a/C_w = K_u/K_d$$

and BCF is defined as C_a/C_w (or K_u/K_d) at steady state. Similarly, the contaminant half-life ($t_{0.5}$) is estimated from

$$t_{0.5} = 0.693/K_d$$

Differences in toxicokinetics were examined with nonparametric test procedures. The two-sided Mann-Whitney U-test (p = 0.05 and p = 0.10) was used for all comparisons of K_u's, K_d's, and

BCF's for the two organisms and three test compounds. A probability level of 0.10 was used (in addition to p = 0.05) because the various data sets contained a relatively low number of observations (6-14), many of which had tied (i.e., below detection limits) values. Interspecies (amphipods versus mysids) analyses were limited to those months in which both mysid and amphipod experiments were conducted. Similarly, for each species, intercompound comparisons by contaminant pair (e.g., BaP versus DDE) were limited to those months in which both xenobiotics were investigated. Nonparametric test procedures were used because of the small number of experimental observations. Such test procedures are relatively insensitive to departure from normality (a requirement for parametric test procedures) and are ideally suited for small sample sizes (Conover 1980).

RESULTS

There was relatively little seasonal variation in the mean sizes of *P. hoyi* and *M. relicta* used in the experiments. The mean wet weight of *P. hoyi* used in the experiments ranged from 6.4-10.7 mg and averaged 8.3 mg. Assuming a dry weight to wet weight conversion of 27% (Landrum 1988) and using the length-weight regression developed by Winnell and White (1984) for deep-water populations of *P. hoyi*, animals were approximately 6.8 mm in length. Such animals were immatures in their second year of life (Winnell and White 1984). The mean wet weight of *M. relicta* used in the experiments ranged from 34.1-52.6 mg and averaged 43 mg. Assuming an 21% dry weight to wet weight conversion (personal communication, W. Frez, Large Lakes Research Station, Grosse Ile, MI) and dry weight to length conversions developed by Sell (1982), such animals averaged 16.9 mm long and were approximately 16 months old (Sell 1982). Thus, mysids were larger but perhaps younger than amphipods used in the experiments.

The percent of contaminant bound to dissolved organic matter for the *P. hoyi* experiments averaged 13.2% for DDE, 15.2% for BaP, and 20.7% for HB. Similar mean values (12.9%, 14.2%, and 17.3%) were observed for the *M. relicta* experiments. Percent bound increased with K_{ow}.

Uptake Experiments

DDE and BaP uptake clearances for *P. hoyi* varied with sampling period (Fig. 1). Uptake clearances were very high in June 1986 but only moderately so
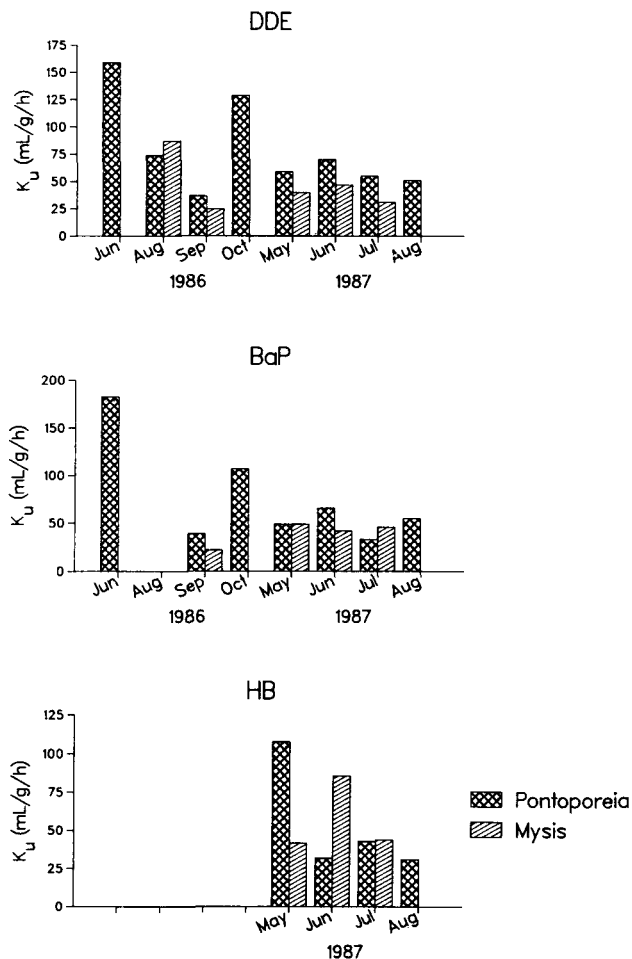

FIG. 1. **Pontoporeia hoyi** and **Mysis relicta** uptake clearance rates (K_u) for DDE, BaP, and HB for the 1986 and 1987 toxicokinetic experiments.

in June 1987. Uptake clearances were similar between May, June, July, and August 1987 both for *P. hoyi* and *M. relicta*.

Only a limited set of HB uptake experiments were conducted (Fig. 1). HB uptake clearance for *P. hoyi* was substantially higher in May 1987 than in June, July, and August: in contrast, *P. hoyi* uptake clearance for BaP and DDE varied little between May and August 1987. HB uptake clearance for *M. relicta* in June was approximately twice that observed in May and July: in contrast, there were less pronounced differences in BaP and DDE uptake measured in June versus that measured in May and July 1987.

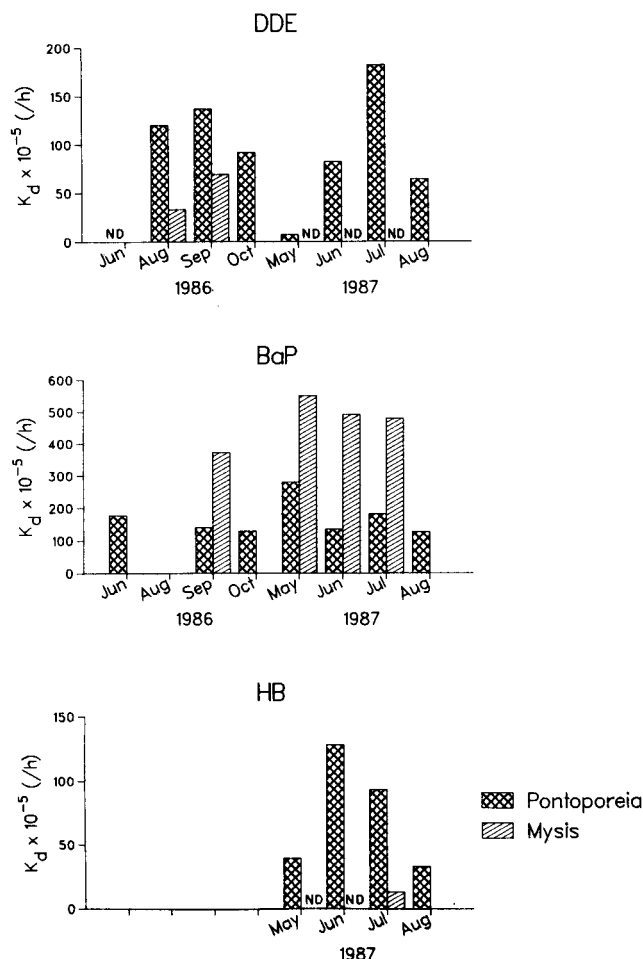Although uptake clearances varied between sampling intervals, there was no evidence of a strong

FIG. 2. Pontoporeia hoyi *and* Mysis relicta *depuration rates* (K_d) *for DDE, BaP, and HB for the 1986 and 1987 toxicokinetic experiments.* nd = *not detectable.*

seasonal trend for any of the three compounds investigated for either *P. hoyi* or *M. relicta*.

Elimination

Amphipods were capable of eliminating DDE although rates were, on occasion, not detectable (June 1986) or exceedingly low (May 1987) (Fig. 2). DDE elimination, although not detectable in June 1986, was detectable in June 1986. Mysids were considerably less capable than *P. hoyi* of eliminating DDE: no loss rate was detected during the May, June, and July 1987 experiments.

P. hoyi elimination rates for BaP were relatively high and more readily measured (Fig. 2) than for DDE. Mysid BaP elimination rates also were read-

ily measured during all cruises: elimination rates varied little between May and July 1987.

HB was not readily eliminated by *P. hoyi* over May–August 1987 (Fig. 2). There was no correspondence between HB and BaP elimination rates over this period. No HB elimination rate could be detected for *M. relicta* in the May and June experiments. A very low rate was measured in July 1987.

As with the uptake clearances, no strong seasonal trend was observed for any of three compounds for either *P. hoyi* or *M. relicta*.

Bioconcentration

Bioconcentration was estimated by dividing the uptake clearance by the elimination rate constant (Leversee *et al.* 1982). For months in which an elimination rate could not be measured, the lowest measured contaminant elimination rate was used.

DDE bioconcentration factor varied with sampling period both for *M. relicta* and *P. hoyi* (Fig. 3). BaP was bioconcentrated less than DDE. HB was the most strongly bioconcentrated compound.

While neither uptake clearance nor elimination rate constant showed any seasonal dependence, it was thought that the calculated BCF might exhibit such seasonality by amplifying small trends in K_u and K_d when the BCF was calculated. However, no such pattern emerged either for *P. hoyi* or *M. relicta* for any of the three test compounds (Fig. 3).

Statistics: Mean Values

Mean K_u's, based on all observations, were calculated to provide the best estimate of the compound uptake clearance for each species (Fig. 4). The same number of observations was not used for calculating each mean: a different number of experiments was conducted for each compound and test organism. The mean K_u's for *P. hoyi* were similar for DDE (79.2 mL/g/h, n = 8), BaP (75.9 mL/g/h, n = 7), and HB (53.5 mL/g/h, n = 4). The mean K_u's for *M. relicta* were similar for DDE (46.0 mL/g/h, n = 5), BaP (39.9 mL/g/h, n = 4), and HB (57.5 mL/g/h, n = 3). The lower mean value for BaP and DDE uptake by mysids compared with amphipods could be partly associated with the fact that *M. relicta* experiments were not conducted during months in which *P. hoyi* uptake rates were relatively high, i.e., June and October 1986.

Mean K_d rates (Fig. 4) are based only on those experiments in which an elimination rate could be measured. Thus, mean estimates are elevated by
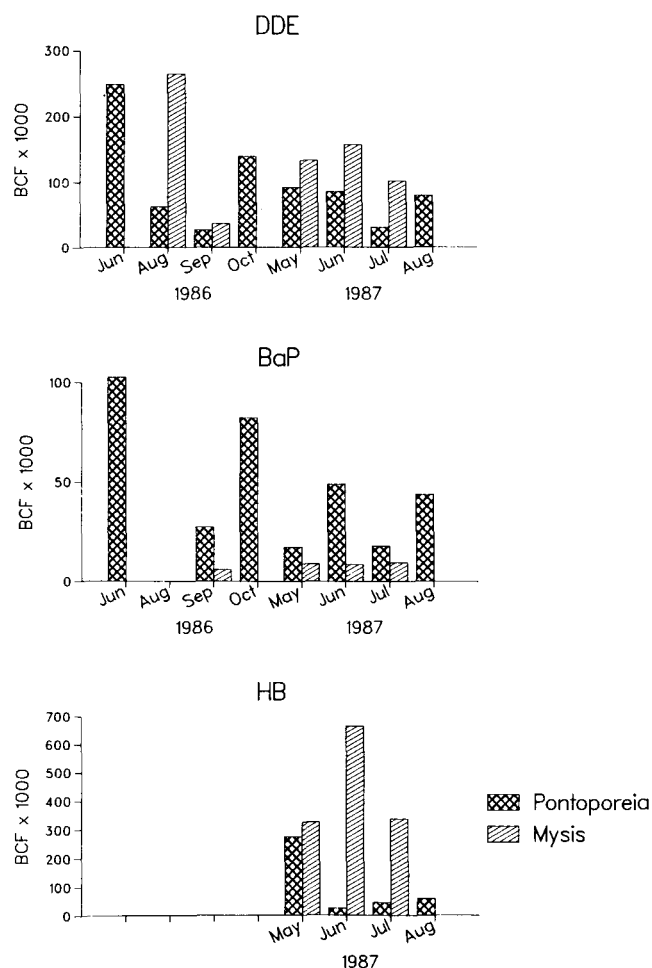
FIG. 3. **Pontoporeia hoyi** *and* **Mysis relicta** *bioconcentration factors (BCF) for DDE, BaP, and HB for the 1986 and 1987 toxicokinetic experiments. See text for additional explanations.*

FIG. 4. The mean (+ 1 standard deviation) uptake clearance rate (K_u), depuration rate (K_d), bioconcentration factor (BCF), and half-life exhibited by **Pontoporeia hoyi** *and* **Mysis relicta** *for DDE, BaP, and HB.*

the exclusion of experiments in which elimination rates were below detection limits. For *P. hoyi*, the highest mean K_d was observed for BaP (-0.00168/h, n = 7) and lowest for HB (-0.00078/h, n = 4), while DDE (-0.00098/h, n = 7) had an intermediate elimination rate. Similarly, for *M. relicta*, the highest mean K_d was observed for BaP (-0.00470/h, n = 4), the lowest for HB (-0.00013/h, n = 1), while DDE had an intermediate (-0.00051/h, n = 2) elimination rate.

Mean BCF (Fig. 4) for *P. hoyi* was greatest for HB (101,663), lowest for BaP (48,582), and intermediate for DDE (95,629). Similarly, for *M. relicta*, mean BCF was greatest for HB (442,231), lowest for BaP (8,496), and intermediate for DDE
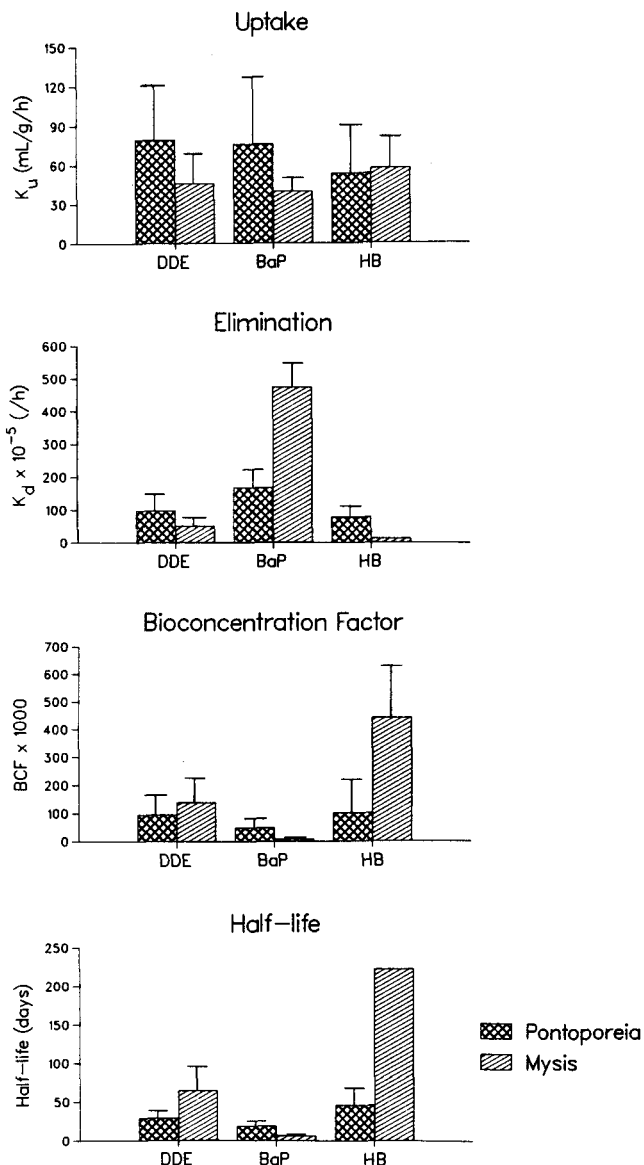
(138,760). Mean BCF estimates for DDE and HB are conservative because elimination was below detection limits for some experiments. For those months, BCF was conservatively estimated by using the lowest measured K_d for that compound and organism.

Compound half-life $(t_{0.5})$ (Fig. 4) could be estimated only for those experiments in which the appropriate K_d could be measured. HB had the

TABLE 1. Results of the Mann-Whitney U-test comparing uptake clearances (k_u), elimination rates (K_d), and bioconcentration factors (BCF) for Pontoporeia hoyi and Mysis relicta for DDE, BaP, and HB. ** = significant at $p < 0.05$; * = significant at $p < 0.10$; n.s. = not significant; n = number of observations.

	K_u	K_d	BCF	n
DDE	n.s.	**	*	10
BaP	n.s.	**	**	8
HB	n.s.	*	*	6

TABLE 2. Results of the Mann-Whitney U-test comparing the toxicokinetics of Pontoporeia hoyi and Mysis relicta for DDE, BaP, and HB. See Table 1 for explanation of abbreviations.

	K_u	K_d	BCF	n
Pontoporeia hoyi				
DDE/BaP	n.s.	**	n.s.	14
DDE/HB	n.s.	n.s.	n.s.	8
BaP/HB	n.s.	*	n.s.	8
Mysis relicta				
DDE/BaP	n.s.	**	**	8
DDE/HB	n.s.	n.s.	*	6
BaP/HB	n.s.	*	*	6

longest half-life, averaging 45.6 days in *P. hoyi*: a single HB $t_{0.5}$ estimate of 222.1 days was obtained for mysids. BaP had the shortest half-life, averaging 18.5 days in *P. hoyi* and only 6.2 days in *M. relicta*. The mean $t_{0.05}$ for DDE was intermediate to that of HB and BaP for both *P. hoyi* (28.8 days) and *M. relicta* (64.7 days).

Statistics: *P. hoyi* Versus *M. relicta* Comparisons

Differences in amphipod and mysid uptake clearances were not statistically significant (Table 1) for DDE, BaP, or HB when based on months in which both species were tested. However, differences in amphipod and mysid elimination rates were statistically significant for DDE, BaP, and HB: a zero elimination value was assumed for those months in which elimination could not be measured. Similarly, differences in amphipod and mysid BCFs were statistically significant for DDE, BaP, and HB.

Statistics: DDE/BaP, DDE/HB, and BaP/HB Comparisons

Intercompound differences in DDE/BaP uptake clearances were not statistically significant (Table 2) for either *P. hoyi* or *M. relicta* when based on months in which both compounds were tested. Similarly, differences in DDE/HB and BaP/HB uptake clearances were not statistically significant for either species.

DDE/BaP elimination rates were significantly different (Table 2) for *M. relicta* and for *P. hoyi* when based on months when both compounds were tested for a given species. Similarly, BaP/HB elimination rates were significantly different for *M. relicta* and for *P. hoyi*. In contrast, DDE/HB elimination rates were not significantly different either for either species.

Differences in BCFs for DDE/BaP, DDE/HB, and BaP/HB were not statistically significant for

P. hoyi (Table 2). However, differences in BCFs and DDE/BaP, DDE/HB, and BaP/HB were statistically significant for *M. relicta*. For *M. relicta*, statistically significant differences in BCFs were related to relatively large differences in K_d for BaP (which was readily eliminated) and HB (a compound with an exceedingly low K_d). Amphipods exhibited less pronounced differences in their ability to eliminate BaP and HB.

DISCUSSION

Ideally, environmental protection should be based on *a priori* and not a *posteriori* strategies. Consequently, regulatory agencies such as the U.S. Environmental Protection Agency are developing protocols to prevent the discharge of chemicals that are toxic, persistent, and biomagnified. Because thousands of new compounds are synthesized annually, and thus subject to regulatory scrutiny, it is important that cost-effective screening protocols be developed.

Laboratory studies have been shown linear relationships between bioconcentration, bioaccumulation, and biomagnification and the K_{ow}'s of various organic contaminants (Lu et al. 1977, Southworth et al. 1978). However, as K_{ow} exceeds 10^6, bioconcentration factors may decline (Oliver 1984). Moreover, toxicokinetic factors may vary between species (Lu et al. 1977, Moore et al. 1977, Leversee et al. 1981) and, within a species, be modified by factors such as animal size, feeding regime, and season (Murphy 1971, Corner et al. 1976, Hansen 1980, Landrum 1988). Thus, some ecosystem components may bioconcentrate or bioaccumulate organic contaminants more than others. The results of our experimental studies illustrate some

of the complex factors affecting the biocencentration of organic contaminants in the laboratory.

Uptake

The influence of biological factors affecting K_u is best illustrated by considering uptake clearances for BaP, the compound for which we have the largest data sets. The mean BaP k_u for *P. hoyi* collected at our 97-m station was 76 mL/g/h. In contrast, *P. hoyi* collected at a more nearshore 29-m station exhibited higher BaP uptake clearances, averaging 117 mL/g/h (Landrum 1988) to 137 mL/g/h (Frez and Landrum 1986). These differences in K_u estimates may be due to differences in the size frequency distribution of *P. hoyi* inhabiting the 29- and 97-m depth regions of Lake Michigan. *Pontoporeia* inhabiting the 97-m region of Lake Michigan exhibit little seasonal change in mean length (Evans *et al*. In press); animals used in our laboratory studies averaged a mean wet weight of 8 mg. In contrast, *P. hoyi* inhabiting shallower depths are more seasonally variable in size and are, on average, smaller than in the profundal region of Lake Michigan (Winnell and White 1984, Evans *et al*. In press). *P. hoyi* collected from the 29-m depth region of the lake and used for experimental toxicokinetic studies ranged from 4 to 7 mg in wet weight (Fig. 4 in Landrum 1988). Animal size and BaP K_u are inversely related in *P. hoyi* (Fig. 4 in Landrum 1988). Extrapolating Landrum's Figure 4 to a mean *P. hoyi* body weight of 8 mg gives an estimated BaP K_u of 78 mL/g/h, a value that agrees excellently with our mean estimate of 76 mL/g/h. Seasonal variations in size should contribute to a seasonal pattern in uptake for nearshore but not profundal populations of *P. hoyi*.

Fewer investigations have been made of *Mysis relicta* uptake kinetics. The mean BaP K_u for *M. relicta* collected at 97 m was 39 mL/g/hr versus a mean rate of 72 mL/g/hr observed by Frez and Landrum (their Fig. 2, 1986) using animals collected from a 65-m station. Differences in mean BaP K_u's for the two study sites are unlikely to be related to differences in animal size. Frez and Landrum (1986) reported that mysids used in their studies were 15–20 mm in length, a value similar to our estimate of a 17 mm mean length of the mysids used in our studies. Moreover, the K_u for BaP appears to be independent of mysid size (Frez and Landrum 1986). The reasons for these between-station differences in mysid BaP K_u's remain to be resolved.

There was no evidence that intercompound (DDE versus BaP, DDE versus HB, and BaP versus HB) differences in mean K_u's were statistically significant either for *P. hoyi* or *M. relicta*. Similarly, there was no evidence that interspecies (*P. hoyi* versus *M. relicta*) differences in mean K_u's were statistically significant either for DDE, BaP, or HB. Thus, a 97-m populations of Lake Michigan *P. hoyi* and *M. relicta* may have generally similar uptake clearances for organic compounds with K_{ow}'s ranging from 5.7–6.7.

Elimination

The mean BaP K_d observed for 97-m *P. hoyi* populations was –0.0017/h, a value very similar to that observed by Frez and Landrum (1986; –0.002/h) and Landrum (1988; –0.0016/h) using 29-m populations. This similarity in elimination rate was unexpected given the larger differences in K_u between the two populations. The similarity in K_d's between the nearshore and profundal populations may be due to the antagonistic effects of the smaller mean body size (affecting an increase in K_d) but higher lipid content (affecting a decrease in K_d) in 29-m versus 97-m populations of *P. hoyi*. As in the BaP uptake studies, 97-m populations of *M. relicta* had a lower mean BaP elimination rate (–0.0047/h) than observed for 65 m populations (–0.013/h; Frez and Landrum 1986).

In contrast to K_u, K_d differed significantly with respect to compound. However, intercompound differences in K_d were not directly related to K_{ow}. Both *P. hoyi* and *M. relicta* were more efficient at eliminating BaP than DDE. However, the K_{ow} for BaP is larger than that for DDE. The higher elimination rate for *M. relicta* for BaP is likely due, in part, to the ability of mysids to biotransform this compound (W. A. Frez, Large Lakes Research Station, Grosse Ile, Michigan, personal communication). The biotransformation explanation does not apply to *P. hoyi* as attempts to measure biotransformation have resulted in no measurable rate (Landrum *et al*. 1985, Landrum 1988). Both taxa were least efficient at eliminating HB, the compound with the highest K_{ow}. For both species, smaller differences were observed between DDE/HB elimination rates than between BaP/HB elimination rates. Overall, our results suggest that, for a given species and within the 5.7–6.7 range, K_{ow} does not appear to be a precise indicator of relative differences in K_d.

Mysids and amphipods exhibited strong differ-

ences in their abilities to eliminate xenobiotics. Mysids are considerably more capable of eliminating BaP than amphipods (this study, Frez and Landrum 1986). Similarly, *M. relicta* is more efficient than *P. hoyi* at eliminating other PAH compounds such as anthracene and phenanthrene (Frez and Landrum 1986). However, these elimination capabilities do not extend to all classes of compounds. Mysids are approximately 52% as capable as *P. hoyi* of eliminating DDE and 17% as capable of eliminating HB as amphipods. The reasons for the low elimination of these compounds by mysids is not evident and will require more research to elucidate.

Overall, our results suggest that, compared to uptake, elimination is the more critical process affecting bioconcentration. Elimination, in turn, is strongly dependent on organism metabolic and biochemical capabilities, parameters nor readily incorporated into consideration of K_{ow}-BCF relationships.

Bioconcentration Factors

Bioconcentration is a function of the uptake rate balanced against the elimination rate. Intercompound differences in the BCFs for DDE, BaP, and HB, as calculated from the ratio of K_u/K_d, were not statistically significant for *P. hoyi*. This fits in well with the absence of significance for differences for the DDE/HB, DDE/BaP, and BaP/HB K_u and K_d comparisons. In contrast, the BCFs of *M. relicta* were significantly different for DDE, BaP, and HB. This apparently was due to the fact that BaP was readily eliminated while HB had a very low elimination rate. Although our data sets are limited in the number of compounds studied, there was no evidence that BCF and K_{ow} were strongly interrelated. The lowest BCF was associated with BaP, the compound with the intermediate K_{ow}. Thus, lipophilicity, as expressed by K_{ow}, may not be the major factor affecting bioconcentration factors over a narrow range of K_{ow}'s. Rather, differences in other physical-chemical properties within and between classes of compounds and organisms (particularly those affecting K_d) must be important.

Like elimination, interspecies differences in the bioconcentration of xenobiotics were statistically significant. However, while *P. hoyi* bioconcentrated BaP to a greater extent than *M. relicta*, it bioconcentrated DDE and HB to a lesser extent than mysids. These relative differences in BCFs between the two species clearly could not have been predicted from a simple consideration of compound K_{ow}. Thus, it is important that toxicokinetic studies include the investigation of organisms appropriate to the environment of concern and not simply be restricted to a few, conveniently-studied taxa. It also is important to realize that toxicokinetic studies, by being conducted under simplified laboratory conditions, may not accurately predict the degree to which compounds are biomagnified by organisms living in a given lake. In the laboratory, we determined that *P. hoyi* bioconcentrated DDE and HB to a greater extent than *M. relicta*. However, the converse apparently occurs in the lake. Previous studies have shown that, in Lakes Michigan and Ontario, *P. hoyi* contain greater amounts of total DDT and PCB than *M. relicta* (Evans *et al.* 1982, Borgmann and Whittle 1983). Such differences between laboratory predictions and lake realities may be due to a number of factors related to amphipod and mysid ecology. Important factors may include habitat (benthic versus epibenthic), vertical migratory patterns, and diet.

Finally, we note that our estimates of mean BCFs are conservative. First, elimination of DDE and HB could not be detected in all months: we used the lowest K_d for that compound and species to provide a conservative estimate of BCF for that experiment. Second, mysid elimination rates were measured in the absence of strong feeding. Feeding is important in the elimination process for invertebrates: enhancements in elimination have been observed for *Hyalella azteca*, *M. relicta*, and *Chironomus riparius* when feeding (Leversee *et al.* 1982, Landrum and Scavia 1983, Frez 1988). Thus, K_d values obtained for DDE, HB, and BaP must be considered minimum values and would be expected to increase under more food-rich conditions.

CONCLUSIONS

Our study investigated the toxicokinetics of DDE, BaP, and HB for the benthic *P. hoyi* and the epibenthic *M. relicta*. Uptake rates for all three xenobiotics were similar for both species. However, there were strong interspecies and intercompound differences in elimination. BaP was the most readily eliminated compound while HB was the most slowly eliminated compound. Mysids were more capable than amphipods of eliminating BaP while the converse occurred for DDE and HB. Differences in contaminant toxicokinetics were not read-

ily estimated by K_{ow}: BaP, the least bioconcentrated compound, had the intermediate K_{ow}. Species differences in contaminant bioconcentration were primarily a function of the differential ability of mysids and amphipods to eliminate (or transform) these compounds. Thus, contaminant-BCF relationships developed for one species may not be appropriate for the other.

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