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DIGESTIVE BIOAVAILABILITY TO A DEPOSIT FEEDER (*ARENICOLA MARINA*) OF POLYCYCLIC AROMATIC HYDROCARBONS ASSOCIATED WITH ANTHROPOGENIC PARTICLES

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Abstract—Marine sediments around urban areas serve as catch basins for anthropogenic particles containing polycyclic aromatic hydrocarbons (PAHs). Using incubations with gut fluids extracted from a deposit-feeding polychaete (*Arenicola marina*), we determined the digestive bioavailability of PAHs from fly ashes, coal dusts, diesel soots, tire tread materials, and urban particulates. We found that gut fluids solubilize significant concentrations of PAHs from two tire treads, two diesel soots, and the urban particulates. However, PAHs in fly ashes and coal dusts were not available to the digestive agents in gut fluid. Potential digestive exposure to PAHs is much greater than that predicted to be available from these materials using equilibrium partitioning theory (EqP). Amending an already-contaminated sediment with fly ash decreased phenanthrene solubilization by gut fluid. In contrast, addition of tire tread to the sediment resulted in increased solubilization of four PAHs by gut fluid. Therefore, addition of certain types of anthropogenic particles to sediments may result in an increase in bioavailable PAHs rather than a net decrease, as predicted by EqP. Difficulty in predicting the amount of change due to amendment may be due to interactions occurring among the mixture of compounds solubilized by gut fluid.

Keywords—Gut fluid   Solubilization   Bioaccumulation

INTRODUCTION

Marine sediments around urban areas serve as repositories for anthropogenic particles (AP) including aerosols from the combustion or pyrolysis of organic materials (e.g., soot carbon), and particles derived from asphalt, brake-linings, tire treads, and material from construction sites. Delivery of these particles via atmospheric deposition and surface-water runoff leads to their elevated concentrations in sediments. These particles constitute the majority of nonmineral matter in sediments around cities. For example, up to 30% of total organic carbon in coastal sediments is from soot [1]. Furthermore, tire tread debris approaches 15% of the total sediment (by mass, including minerals) in areas surrounded by heavy automobile traffic (C. Reddy, Woods Hole Oceanographic Institution, Woods Hole, MA, USA, personal communication and [2,3]).

Organic solvent extractions of many types of AP (e.g., soot, coal, and tire treads) release polycyclic aromatic hydrocarbons (PAHs), which are a class of hydrophobic contaminants that can have toxic and carcinogenic effects on marine animals (see review in [4]). Sediments close to urban areas show characteristic PAH enrichments, and presumably much of the PAH is associated with AP, although petrogenic sources of PAHs also may be present [5]. Though clearly a global trend and problem [6], PAH contamination of sediments surrounding the United States is particularly well documented and pervasive [7,8]. In Boston Harbor (USA), for example, sedimentary concentrations of individual PAHs range as high as 120 µg g⁻¹ [9,10].

Though available to organic solvents, much of the PAHs associated with AP are sequestered in forms that are not thought to be bioavailable (i.e., not available to animals). Bioavailability is a function of both the geochemical characteristics of the AP as well as the specific physiological pathways that expose a particular organism to contaminants. For example, pyrogenic materials like soot carbon and fly ashes sorb PAHs very strongly and, when present in sediments, can depress interstitial water concentrations of PAHs [11,12]. For animals exposed to PAHs only via interstitial water, the PAHs associated with pyrogenic AP ought to be less bioavailable than petrogenic PAHs. Empirical studies of soot carbon [13–15] and coal-associated PAHs [16–18] in sediments generally have conformed; the biological effects of PAHs are suppressed.

Deposit feeders conspicuously are rare in areas with high concentrations of PAHs, even when associated with pyrogenic AP [19], suggesting an additional route of exposure besides interstitial water for these animals. Recent results suggest that deposit feeders receive the majority of their exposure to sedimentary organic contaminants via digestion [20–23]. Surfactant micelles are responsible for the bulk of sedimentary PAH solubilization in a deposit-feeder's gut fluids [10] and form a nonpolar pseudophase for the mobilization of hydrophobic compounds. As a result of micelle formation, digestive fluids may be better able than interstitial water, both kinetically and thermodynamically, to solubilize PAHs from AP.

We studied extractability of 12 PAHs from nine types of AP by digestive fluids of a deposit-feeding polychaete, *Arenicola marina*. Then, we determined whether patterns of PAH release from AP alone hold under more realistic conditions in

Table 1. Sample descriptions; Standard Reference Material (SRM)

Sample	Source	Description
GF80A tire tread	Rouse Rubber (Vicksburg, MS, USA)	Extremely fine, black powder
GR16 tire tread	Baker Rubber (South Bend, IN, USA)	Coarse, black powder
Diesel soot	Interstate Diesel Equipment Service (North Kingstown, RI, USA)	Fine, black powder scraped from exhaust systems of diesel vehicles
SRM 1650 diesel soot	National Institute of Standards and Technology (Gaithersburg, MD, USA)	Fine, black powder collected from a heat exchanger fed by four diesel engines
SRM 1649 urban particulates	National Institute of Standards and Technology (Gaithersburg, MD, USA)	Very fine, atmospheric particulate material collected with large-diameter filters
Valley Power Plant coal dust	Wisconsin Electric (Milwaukee, WI, USA)	Unburned, fine, black, bituminous coal powder from Pennsylvania
Dal-Tex coal dust	New England Power Company (Somerset, MA, USA)	Unburned, coarse, black, bituminous coal powder from Pennsylvania/West Virginia
Class F fly ash	Wisconsin Electric (Milwaukee, WI, USA)	Burned, very fine, gray-black, bituminous coal powder from Colorado
Modified fly ash	U.S. Generating Company (Somerset, MA, USA)	Burned, very fine, high-carbon powder prepared in proprietary process used to separate bituminous Venezuelan coal fly ash into high- and low-carbon fractions

which AP are only a fraction of a contaminated sediment (by wt) by using a sediment amended with either a tire tread or a fly ash. These in vitro incubations mimic the digestive solubilization of compounds, but not their uptake across the digestive tract (assimilation) or metabolic transformations that occur once inside the animal (biotransformation). The importance of the digestive pathway of exposure is made clear by comparison of the amount of PAHs solubilized by digestive fluids to the amount predicted to be dissolved freely in interstitial water by equilibrium partitioning theory.

MATERIALS AND METHODS

Collection of *A. marina*

Arenicola marina (lugworms) were dissected to remove midgut digestive fluids as described previously [10], except that gut fluids were extracted on the day of collection and were clarified by filtration (0.45-μm polytetrafluorethylene membrane) instead of centrifugation. *Arenicola marina* has surfactant micelles in the digestive tract [24], which are responsible for the bulk of PAH solubilization, though other compounds such as proteins probably also are involved [10]. The gut fluid used in this study was surfactant-rich, having a critical micelle dilution of approximately 20% using the contact angle dilution method [24]. In other words, gut fluids required 80% dilution with artificial seawater before surfactant micelles would disassociate into individual surfactant monomers.

AP characterization

A variety of AP were acquired (Table 1). Samples were donated by the company named in the Source column of Table 1, except for Standard Reference Material (SRM) 1650 Diesel Soot and SRM 1649 Urban Particulates, which were purchased from the U.S. National Institute of Standards and Technology (www.nist.org). Standard Reference Material 1649 was collected from urban atmospheres with a large-diameter filter and presumably is weathered. The SRM 1650 was collected from heat exchangers after 200 h of diesel engine operation. The other samples were collected directly from their sources, without environmental mixing or weathering, and were used without additional modification.

All particles were analyzed for total organic carbon (TOC) using either a Perkin-Elmer Series II CHNO/S 2400 elemental analyzer (Wilton, CT, USA) or Carlo Erba NA 1500 elemental analyzer (Fisons Instruments, Beverly, MA, USA). Samples were treated by direct acidification with 1 M HCl to remove calcium carbonate. Soot carbon for all samples was determined using a thermal fractionation method (375°C for 24 h with excess oxygen, [25]). Surface area was measured by N_2 adsorption and multipoint Brunauer, Emmett, and Teller (B.E.T.) analysis of freeze-dried samples, except for the tire tread samples. Both tire tread materials liquefied when subjected to the high temperatures (150°C) required for complete outgassing of N_2; thus, measured values did not represent the samples' true environmental state.

To measure total PAH concentrations in these particles, approximately 0.5 g of each dry sample was extracted with 50 ml of acetone/dichloromethane in a wrist-action shaker (Burrell, Pittsburgh, PA, USA) for 48 h at a medium mixing rate. After extraction, the solvent was passed through a glass fiber filter ([GF/C], Whatman International, Maidstone, UK), dried with anhydrous sodium sulfate, and the contents transferred to a turbo-evaporation tube (Zymark, Hopkinton, MA, USA) for volume reduction and exchanged into hexane. Final extracts were stored at 4°C in the dark until analyzed by gas chromatography/mass spectrometry (GC/MS) using a Hewlett-Packard 5890 Series II gas chromatograph (Avondale, PA, USA) equipped with a 7673A autosampler, electronic pressure control, and 5971A mass selective detector monitoring ions at 188, 240, 264 (internal standards phenanthrene-10d, benzo[*a*]anthracene-12d and perylene-12d, respectively), and target PAH mass units. Polycyclic aromatic hydrocarbon concentrations in the samples were adjusted for the recovery of internal standards and quantified by comparison to peak areas from calibration standards of authentic compounds (Supelco, Belafonte, PA, USA). A multipoint (4–5 level) internal standard calibration curve was used to determine calibration response factors. Limits of detection were approximately 0.01 μg g^{-1} for individual PAHs.

Collection of sediment

During a low tide, sediment was collected with a shovel from Little Mystic Channel (LMC), Boston Harbor (MA, USA). Within a day of collection, sediment was twice washed to remove salt with artificial seawater/deionized water (5/95

by volume), centrifuged at 8,000 *g* for 15 min, freeze-dried, and stored in the dark at 5°C. Sediment from this location is highly contaminated with PAHs and has elevated organic carbon:specific surface area ratios that suggest an already heavy loading with anthropogenically derived organic material [10]. Although a more pristine sediment may have provided a clearer test of how release of PAHs from AP occurs in sediment, our choice of LMC sediment was an effort to maintain environmental realism with regards to the types of sediment that actually are impacted by AP.

Gut fluid incubations

Arenicola marina gut fluid was incubated in triplicate for 4 h in the dark with approximately 0.05 g of each sample at a solid-fluid ratio of approximately 0.05 g ml^{-1}. This solid-fluid ratio was lower than the sediment-fluid ratio normally found in a deposit-feeder's gut [26], but ensured that enough fluid would be available for analysis after incubation with these low-density materials. Gut fluids were clarified by filtration (0.45 µm) and then liquid/liquid extracted with nanopure water and dichloromethane (DCM), partitioning PAHs into the DCM phase. Deuterated PAHs (phenanthrene-d10, benzo[*a*]anthracene-d12, and benzo[*a*]pyrene-d12) were added to the DCM extracts to serve as internal standards. Typical recoveries were 56 ± 11%, 103 ± 14%, and 111 ± 28%, respectively, and reported concentrations are corrected for their recovery. Dichloromethane extracts were purified in the dark by passage through sodium sulfate and ENVI-Florisil columns (Supelco 5–7,058) to remove polar and sulfur compounds, and dried under nitrogen gas at 38°C. Dried samples were reconstituted in 1:1 acetonitrile:water (v/v), passed through a 0.45-µm syringe filter, and injected into a Hitachi D-7000 high-pressure liquid chromatograph ([HPLC], Tokyo, Japan). A Vydac 201 TP (Interchim, Montlucon, France), 5-µm, 250 × 4.6 mm column was used under the following operational conditions: Flow rate = 1.0 ml min^{-1}; temperature = 29°C; injection volume = 250 µl; mobile phase = 1:1 acetonitrile:water (v/v) for 5 min, ramping to 100:0 in 15 min, and holding for 8 min. Polycyclic aromatic hydrocarbons were identified using retention time and absorbance spectrum when concentrations permitted and quantified using fluorescence detection. Fluorescence detection limits were approximately 0.1 µg (L gut fluid)$^{-1}$ for individual PAHs.

Amended sediment incubations

To prepare amended sediments, freeze-dried LMC sediment was mixed with GF80A tire tread at 3.25% and modified fly ash at 3.48% (by wt). This amount is reasonable for sediments close to the source of contamination as 0.6% soot was measured in outer Boston Harbor sediments [25]. Gut fluids were incubated with just sediment (control) and each of the amended sediments (treatments) at a solid-fluid ratio of approximately 0.25 g (ml)$^{-1}$. This solid-fluid ratio is greater than that used for pure AP incubations, but mimics in vivo conditions in deposit-feeder guts [26]. Incubations and subsequent PAH measurements were performed as described above.

Statistics

All values reported are means and standard deviations of triplicate samples. The effects of added AP on gut fluids' release of PAHs from sediment were determined by analysis of variance of contrasts between the unadulterated sediment (control) and each of the amended sediment treatments using Systat

9 (SPSS, Chicago, IL, USA) statistical software on a PC computer. Risk of type I error was controlled at 0.05.

RESULTS

AP characteristics

Organic solvents extracted PAHs from all of the samples except for the fly ashes (Table 2). In the other AP, most individual PAH concentrations were on the order of several µg g^{-1} (dry wt). Coal dusts were relatively enriched in phenanthrene; diesel soots were enriched in phenanthrene and pyrene; tire treads were enriched in pyrene; and SRM 1649 had a uniform distribution of PAH concentrations. With GF80A tire tread, matrix interferences apparently caused the benzofluoranthenes to elute as one peak during GC/MS analysis. As a result, concentrations of the individual compounds benzo[*b*]fluoranthene and benzo[*k*]fluoranthene were not determined and the sum of the benzofluoranthenes is reported in Table 2 for GF80A tire tread. We believe this chromatographic aberration affected only this particular sample.

Tire treads, diesel soots, and coal dusts all contained more than 50% organic carbon, with significant fractions as soot carbon. Our measurements of soot carbon may be biased positively, as the thermal oxidation method recently has been accused of overestimating soot carbon [25]. These biases are believed to result from both the protection of organic components by the mineral matrix of sediments and condensation reactions occurring among labile organic components such as lipids, proteins, and carbohydrates.

Gut fluid release of PAHs from AP

Polycyclic aromatic hydrocarbon release to gut fluids varied widely among the nine particles (Table 3). No PAH was detected from fly ash after incubation with gut fluid, which was expected as extraction with organic solvents indicated that there were no PAHs in these samples (Table 2). No PAHs were released from either of the coal dust samples, except for traces of phenanthrene and pyrene at levels close to analytical detection limits (0.1 µg L^{-1}).

Gut fluids released PAHs from GF80A and GR16 tire treads, diesel soot, SRM 1649, and SRM 1650, but the amounts released did not correlate strongly with the total PAHs present in any of the samples, except for GR16 tire tread (i.e., Spearman nonparametric rank coefficients were all nonsignificant with $p > 0.05$, except for GR16 tire tread [0.90, $n = 5$]). The greatest concentration of PAHs released was benzo[*a*]pyrene (B[*a*]P) from GF80A tire tread (50.6 µg L^{-1}) (Table 3). Gut fluid PAH concentrations were greater than seawater solubility only for PAHs with molecular weight ≥252. Large variations were associated with our measurements of PAHs in gut fluids as reflected by the standard deviations of the data. The limited amount of gut fluid available from *A. marina* forced little gut fluid (1.0 ml) and small sample sizes (0.05 g) to be used during incubations, which may have resulted in this variability. If PAHs were highly concentrated on only a few particles, then strong intersample heterogeneity would be expected to influence the amount of PAHs released by gut fluids. Close inspection of the results did not reveal a systematic bias. For example, there was no evidence of particular incubations having unusually elevated gut fluid concentrations of all of the PAHs.

Significant fractions of some of the PAHs associated with these particles were extractable in gut fluid. When calculated

Table 2. Geochemical characteristics of the anthropogenic particles from Little Mystic Channel ([LMC], Boston, MA, USA). Reported values are the mean of analytical duplicates, unless followed by a ±, which indicates that values are the mean of triplicates ± a standard deviation; Standard Reference Material = SRM

Sample	GF80A tire tread	GR16 tire tread	Diesel soot	SRM 1650	SRM 1649[a]	Valley Power Plant coal dust	Dal-Tex coal dust	Class F fly ash	Modified fly ash	LMC sediment
Total organic carbon (% sample dry wt)	98.0	81.9	61.2 ± 3.5	60.7	18.2	60.5 ± 16.3	73.7	30.0 ± 7.5	19.2 ± 0.2	71.7
Soot carbon (% sample dry wt)	23.1	10.3	28.9 ± 6.1	53.1	5.2	42.4 ± 0.1	15.3	26.4 ± 6.4	20.9 ± 0.1	NM[b]
Surface area (m²g⁻¹)			17.18	48.00	3.55	1.43	0.98	7.16	9.33	
Polycyclic aromatic hydrocarbon concentrations (µg g⁻¹)										
Phenanthrene	3.81 ± 0.07	12.80 ± 0.25	29.90 ± 5.87	49.9 ± 0.10	4.14 ± 0.37	16.50 ± 0.78	18.30 ± 3.65	ND[c]	ND	120.0
Anthracene	0.32 ± 0.05	1.25 ± 0.05	0.71 ± 0.23	0.86 ± 0.16	0.43 ± 0.08	13.50 ± 0.61	0.39 ± 0.08	ND	ND	18.2
Fluoranthene	8.70 ± 0.08	6.11 ± 0.43	8.81 ± 1.34	37.7 ± 2.03	6.45 ± 0.18	1.86 ± 0.11	2.53 ± 0.33	ND	ND	146.0
Pyrene	42.80 ± 0.76	35.90 ± 2.50	23.20 ± 8.16	33.4 ± 1.99	5.29 ± 0.25	3.70 ± 0.21	3.35 ± 0.49	ND	ND	74.2
Benzo[a]anthracene	1.04 ± 0.08	0.80 ± 0.07	1.17 ± 0.41	9.25 ± 0.72	2.21 ± 0.07	2.45 ± 0.11	2.12 ± 0.45	ND	ND	37.6
Chrysene	6.73 ± 0.07	3.80 ± 0.17	5.03 ± 1.57	39.2 ± 1.36	3.05 ± 0.06	3.10 ± 0.15	3.69 ± 0.71	ND	ND	50.9
Benzo[b]fluoranthene	6.60 ± 0.36[c]	NM	3.28 ± 0.36	NM	6.45 ± 0.64	NM	3.55 ± 0.64	NM	NM	36.2
Benzo[k]fluoranthene	[c]	NM	3.37 ± 0.42	2.1[a]	1.91 ± 0.03	NM	3.36 ± 0.60	NM	NM	16.8
Benzo[a]pyrene	2.49 ± 0.19	0.77 ± 0.08	0.32 ± 0.05	1.12 ± 0.25	2.51 ± 0.09	1.96 ± 0.12	1.78 ± 0.44	ND	ND	43.0
Dibenzo[a,h]anthracene	0.62 ± 1.08	0.09	ND	0.85 ± 0.57	0.29 ± 0.02	0.12 ± 0.02	ND	ND	ND	2.5
Benzo[ghi]perylene	18.10 ± 1.83	2.85 ± 0.18	0.45 ± 0.09	2.53 ± 0.69	4.01 ± 0.91	0.52 ± 0.04	2.08 ± 0.62	ND	ND	24.4
Indeno(1,2,3-cd)pyrene	1.11 ± 1.08	0.30 ± 0.01	0.32 ± 0.14	1.50 ± 0.54	3.18 ± 0.72	0.18 ± 0.01	0.42 ± 0.39	ND	ND	18.2

[a] Concentrations supplied by the National Institute of Standards and Technology (USA) but not measured by us.
[b] ND = not detected; NM = not measured.
[c] The benzofluoranthenes co-eluted during gas chromatography/mass spectroscopy analysis of GF80A. Therefore, the reported concentration is for both compounds.

as the fraction of the total PAHs in the samples, 40 and 33% of the B[a]P and dibenzo[a,h]anthracene, respectively, in GF80A tire tread, 24% of B[a]P in Diesel soot 1, and 44 and 28% of the anthracene in SRM 1650 and SRM 1649, respectively, were released to gut fluid (Table 4). These values were calculated using the average concentration of each PAH released by gut fluid and do not reflect the variability of measurements as noted in the previous paragraph. Gut fluids appeared to solubilize more dibenzo[a,h]anthracene than ought to have been present during the incubation of diesel soot (245%). Re-examination of both the GC/MS and HPLC data did not resolve this conundrum; both peaks appeared to be identified correctly according to retention time, mass spectrum (during GC/MS analysis), and fluorescence characteristics (during HPLC analysis). Mass spectrum data are more discriminative than fluorescence characteristics, therefore, we suspect that a co-eluting peak (with fluorescence characteristics similar to dibenzo[a,h]anthracene, e.g., another PAH) artificially may have elevated the concentration measured by HPLC in gut fluid. In addition, sample heterogeneity again may have been responsible for the underestimation of dibenz[a,h]anthracene present in the incubation. Releases of these PAHs were exceptional, however, as less than a few percent of most other PAHs were released from these samples.

Release of PAHs from amended sediment

Arenicola marina gut fluids solubilized PAHs from unadulterated LMC sediment at a range of concentrations spanning 0.29 µg L⁻¹ (anthracene) to 35 µg L⁻¹ (B[a]P; Fig. 1). Previous extractions of LMC sediment with a different *A. marina* gut fluid [10] released similar concentrations of phenanthrene and B[a]P, but an order of magnitude more pyrene than the current study. Adding modified fly ash to the sediment decreased the amount of phenanthrene released to gut fluids by 31%, but had no effect on other PAHs. Addition of GF80A tire tread significantly ($p < 0.05$) increased gut fluid solubilization of fluoranthene, benzo[a]anthracene, benzo[k]fluoranthene, and dibenz[a,h]anthracene by 16, 1.6, 54, and 0.62 µg L⁻¹, respectively, compared to sediment alone.

DISCUSSION

Biological aspects of bioavailability

The bioavailability of sedimentary organic chemicals depends upon both the geochemical characteristics of the sediment as well as the biochemical pathways through which animals are exposed. Benthic organisms can accumulate contaminants through interstitial water and dietary exposures. Ingestion of contaminated sediment is a major route of exposure to hydrophobic organic contaminants for deposit feeders [20,22,27,28]. Dissolved organic material in guts is composed of food hydrolysates, digestive enzymes, and surfactants [24] that render xenobiotic compounds available for absorption. In vitro gut fluid incubations offer a simple way of predicting deposit-feeder bioaccumulation of hydrophobic sedimentary contaminants [29,30]. Though our results are directly applicable only to one animal, *Arenicola marina*, the common use of surfactant micelles to solubilize lipids in deposit-feeder gut fluids [24], suggests that this work also is applicable to other deposit feeders.

We can compare exposure to PAHs via digestion (as measured by gut fluid incubations) and via interstitial water (as predicted by equilibrium partitioning theory ([EqP], [31]).

Table 3. Concentrations of polycyclic aromatic hydrocarbon (PAH) in *Arenicola marina* gut fluid after incubation. Standard Reference Material = SRM

PAH (μg L⁻¹)	Molecular weight (g)	Seawater[a] solubility	GF-80A tire tread[b]	GR16 tire tread[b]	Diesel soot[b]	SRM 1650[b]	SRM 1649[b]	Valley Power Plant coal dust[c,b]	Dal-Tex coal dust[c,b]	Class F fly ash[b]	Modified fly ash[b]
Phenanthrene	178.2	725.0	6.9 ± 0.5	4.5 ± 0.8	14.9 ± 1.2	ND[d]	ND	0.1	0.1	ND	ND
Anthracene	178.2	29.0	0.5 ± 0.1	0.7 ± 0.2	0.3 ± 0.1	5.7 ± 1.7	3.5 ± 1.3	ND	ND	ND	ND
Fluoranthene	202.3	152.0	2.0 ± 0.1	1.1 ± 0.1	1.0 ± 0.6	5.5 ± 0.6	2.0 ± 0.3	ND	ND	ND	ND
Pyrene	202.3	83.0	11.7 ± 1.7	17.7 ± 1.6	2.0 ± 0.4	19.3 ± 1.8	15.2 ± 5.3	0.3	ND	ND	ND
Benzo[a]anthracene	228.3	6.7	2.4 ± 0.1	ND	3.3 ± 0.4	ND	3.1 ± 0.7	ND	ND	ND	ND
Chrysene	228.3	1.2	0.4 ± 0.1	1.9 ± 0.6	1.1 ± 0.1	4.4 ± 1.1	2.6 ± 0.3	ND	ND	ND	ND
Benzo[b]fluoranthene	252.3	0.9	17.6 ± 7.5	ND	8.0 ± 2.0	ND	ND	ND	ND	ND	ND
Benzo[k]fluoranthene	252.3	0.5	12.5 ± 1.1	2.3 ± 1.2	4.0 ± 2.5	ND	5.0 ± 0.9	ND	ND	ND	ND
Benzo[a]pyrene	252.3	2.3	50.6 ± 27.77	ND	3.8 ± 1.0	3.5 ± 0.2	5.3 ± 0.6	ND	ND	ND	ND
Dibenzo[a,h]anthracene	278.4	0.3	10.2 ± 1.1	ND	1.2 ± 0.1	ND	ND	ND	ND	ND	ND
Benzo[ghi]perylene	276.3	0.2	3.2 ± 0.4	ND	0.4 ± 0.1	ND	ND	ND	ND	ND	ND
Indeno(1,2,3-cd)pyrene	276.3	0.7	ND	ND	ND	ND	ND	ND	ND	ND	ND

[a] Values from Xie et al. [51].
[b] See Table 1 for sample sources.
[c] Single measurements, not replicated.
[d] ND = not detected.

Normal sediment-water partitioning (K_d; L kg⁻¹) behaviors of organic contaminants such as PAHs are described as

$$K_d = C_s/C_d = f_{oc}K_{oc} \tag{1}$$

which has been extended by [25] to account for sedimentary soot partitioning as

$$K_d = C_s/C_d = f_{oc}K_{oc} + f_{sc}K_{sc} \tag{2}$$

where C_s is the PAH sediment concentration (μg kg⁻¹), C_d is the PAH dissolved concentration (μg L⁻¹), f_{oc} is the weight fraction of organic carbon in the sediment (g g⁻¹), K_{oc} is the partition coefficient between sediment organic carbon and interstitial water (L kg⁻¹; often predicted by the octanol–water partitioning coefficient K_{ow}), f_{sc} is the fraction of soot carbon (g g⁻¹), and K_{sc} is the soot-carbon normalized partition coefficient (L kg⁻¹; predicted by activated carbon-water partitioning coefficients found in [32,33]). The equilibrium-dissolved concentration of PAHs and other nonionic organic compounds is

$$C_d = C_s/(f_{oc}K_{oc} + f_{sc}K_{sc}) \tag{3}$$

Digestive fluids released considerably more PAHs than these calculations predict to be dissolved from AP, and this trend increases with molecular weight of the PAHs (Fig. 2). For example, gut fluid solubilizes 20 times more pyrene and almost 1,000 times more B[a]P from SRM 1650 than EqP predicts would be available from this material. Because hydrophobic compounds adsorb even more strongly to soot than to normal organic material in sediments (as reflected in soot partitioning coefficients that are greater than organic carbon partitioning coefficients), soot-corrections caused EqP to underestimate the amount of PAH released to gut fluid by an additional two orders of magnitude.

Gut fluids do not solubilize very hydrophobic compounds particularly strongly. In fact, *A. marina* digestive fluids solubilize a wide size-range of PAHs at similar extent [10,34]. In contrast, EqP theory assumes that compounds available for bioaccumulation come from a freely dissolved state and, therefore, uses partition coefficients between some phase and water (e.g., the organic carbon–water partition coefficient [K_{oc}]), to determine how much is available. Because water is an increasingly poor solvent for PAHs of increasing molecular weight [35,36], EqP theory predicts that less of the very hydrophobic compounds are available.

Geochemical aspects of bioavailability

Fly ashes. Neither organic solvents (Table 2) nor gut fluids (Table 3) were able to mobilize PAHs from either of the fly ashes. Though these materials actually may have associated PAHs, as PAHs have been found in other fly ashes [37–39], our extractions were unable to free them. Indeed, recoveries of PAHs used as internal standards were low, suggesting that fly ashes removed them from solution in organic solvents via adsorption.

Because of their capacity for sorbing low molecular weight organic contaminants in aqueous solutions, fly ashes have been proposed as barriers to contaminant transport in groundwater systems [39,40]. Our results suggest limited utility as an adsorbent in sediment systems in which concern is for deposit-feeders' digestive exposure to PAHs. The addition of 3% of modified fly ash did decrease gut fluid's release of phenanthrene from Boston sediment by 31%. The toxicological significance of this reduction is unknown and none of the other PAHs were affected by fly ash. We emphasize that we made

Table 4. Percentage of total polycyclic aromatic hydrocarbon (PAH) in the sample released to *Arenicola marina* gut fluids. Standard Reference Material = SRM

	GF-80A tire tread	GR16 Baker tire tread	Diesel soot	SRM 1650	SRM 1649
Phenanthrene	3.6	0.7	1.0	0.0	0.0
Anthracene	2.9	1.2	0.8	43.8	28.3
Fluoranthene	0.5	0.4	0.2	1.0	1.1
Pyrene	0.6	1.0	0.2	3.9	10.9
Benzo[*a*]anthracene	4.6	0.0	5.7	0.0	3.5
Chrysene	0.1	1.0	0.5	0.7	1.2
Benzo[*b*]fluoranthene	9.1[a]	[b]	4.9	[b]	0.0
Benzo[*k*]fluoranthene	[a]	[b]	2.4	0.0	5.2
Benzo[*a*]pyrene	40.6	0.0	24.4	14.2	5.1
Dibenzo[*a,h*]anthracene	33.0	0.0	245.4[c]	0.0	0.0
Benzo[*ghi*]perylene	0.4	0.0	1.9	0.0	0.0
Indeno(1,2,3-*cd*)pyrene	0.0	0.0	0.0	0.0	0.0

[a] We compared the gut fluid solubilization of benzo[*b*]fluoranthene and benzo[*k*]fluoranthene to the sum of the benzofluoranthenes present in the sample.

[b] These PAHs were measured in gut fluid, but their total concentrations in the samples were not measured.

[c] Dibenzo[*a,h*]anthracene concentrations in gut fluid were greater than should be possible, considering that the total concentration of this PAH in diesel soot was below detection limits (0.01 $\mu g\ g^{-1}$). We suspect a co-eluting compound may have artificially elevated our measurement of this PAH in gut fluid. Calculated using the average concentration of each PAH released by gut fluid.

no attempt to quantify PAHs smaller than phenanthrene, though fly ashes would be suspected of having an especially strong impact on the availability of these compounds.

Coal. Unburned coal can be a significant source of total PAHs in marine coastal sediments [41–43]. Our coal dusts contained PAHs, as measurements of total PAHs indicate (Table 2), but they are sequestered apparently in ways that make them unavailable to the digestive agents in gut fluid. Polycyclic
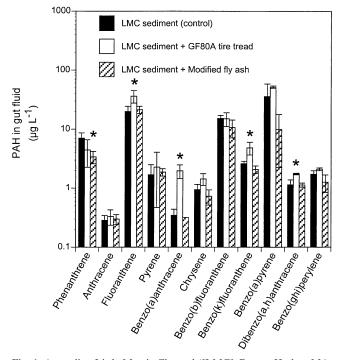

Fig. 1. Amending Little Mystic Channel ([LMC], Boston Harbor, MA, USA) sediment with GF80A tire tread increased the amount of polycyclic aromatic hydrocarbon (PAH) solubilized by *Arenicola marina* gut fluids and there was a significant increase in fluoranthene, benzo[*a*]anthracene, benzo[*k*]fluoranthene, and dibenzo[*a,h*]anthracene concentrations. The addition of modified fly ash decreased the amount of phenanthrene released to gut fluids. No other PAH was affected. An asterisk indicates a difference from control at $p < 0.05$.

aromatic hydrocarbons sorbed to coal particles have high desorption activation energies [44] that inhibit dissolution into water and digestive fluids. The weak digestive fluids of suspension feeders like *Crassostrea virginica* and *Mytilus edulis* and even the stronger fluids of the deposit feeders *A. marina*, *Rhepoxynius abronis*, and *Neanthes arenaceodentata*, seemingly cannot desorb PAHs from coal particles ([16,17] this study). Therefore, while coal may have some detrimental biological effects on benthic animals by diluting sediment and decreasing its volumetric nutritional content, PAHs associated with this bituminous coal do not appear to be bioavailable.

Diesel soot. We found significant fractions of some PAHs to be released to gut fluid from diesel soot samples. These results are at odds with other research on the interstitial water solubility (and assumed bioavailability) of diesel soot–PAHs. Soot-associated PAHs generally are thought to be less bioavailable than PAHs associated with sedimentary organic matter [13,14,18]. Diesel soot is a much stronger PAH adsorbent than natural organic matter [1] and causes enhanced partitioning of sedimentary PAHs to the particulate phase rather than interstitial water [12]. However, our deposit feeder's digestive fluids solubilized PAHs from diesel soot at concentrations greater than seawater solubility (Table 3) and much greater than equilibrium partitioning would suggest (Fig. 2). These results suggest that digestive micelles of deposit feeders may access PAHs in diesel soot that are not available to other animals.

Two factors may have influenced our results. First, approximately 50% of the carbonaceous material in diesel soot and 12% of that in SRM 1650 is removed by oxidation at 375°C and thus does not fit the analytical definition of soot-like material proposed by [25]. This other fraction of organic carbon in these soots may be responsible for part or all of the bioavailable PAHs. Second, our clarification of gut fluid (passage through a 0.45-μm filter) may not have removed all soot particulates from the fluid-phase after incubation; for example, soot particles as small as 180 nm are discerned when suspended in ethanol [25]. If such particles were present in gut fluid after clarification, then PAH measurements would include PAHs associated with colloidal particles, rather than only solubilized
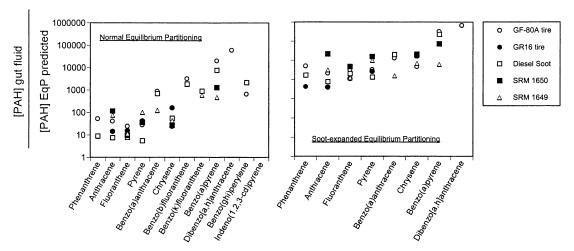
Fig. 2. Gut fluids solubilize more polycyclic aromatic hydrocarbon (PAH) from anthropogenic particles than would be predicted by equilibrium partitioning theory. The *y*-axis is the amount of PAH measured in gut fluids divided by that predicted to be dissolved by equilibrium partitioning theory ([EqP], Eqn. 3). Values greater than 1 on the *y*-axis represent more digestively available PAH than EqP would have predicted. Soot-partitioning coefficients were not available for all PAHs; therefore, fewer PAHs are in the graph on the right. Standard Reference Material (SRM).

PAHs, and thus overestimate bioavailability. It is not clear whether particles this small would be present in an aqueous system like gut fluid, as soot cannot be suspended in water (Ö. Gustafsson, University of Stockholm, Stockholm, Sweden, personal communication). Calculations indicate that 2.14 mg of SRM 1650 diesel soot would have to pass through a 0.45-μm filter in order to deliver enough solid-phase to match the amount of B[*a*]P we measured in gut fluids. This mass of diesel soot is equivalent to 5% of the amount in the incubation, which likely would be visible. However, gut fluid filtrates were clear to the naked eye. In fact, the gut fluid solubilization of PAHs we noted from soots was not remarkable, actually quite reasonable, when compared to amounts of release from contaminated sediments [10,21]. Therefore, though we cannot discount solid-phase contamination of the gut fluids, we suspect little bias due to the clarification step.

Tire tread materials. We believe this paper to be the first report on the bioavailability of PAHs associated with tire tread materials. Gut fluids incubated with tire treads alone released many PAHs from these matrices. From GF80A tire tread, all PAHs with molecular weight ≥252, except for indeno(1,2,3-*cd*)pyrene, were solubilized at concentrations above aqueous solubility. From GR16 tire tread, chrysene and benzo[*k*]fluoranthene were solubilized at the same concentration as aqueous solubility (Table 3). As both types of tire treads contained PAHs (Table 2), the differences in PAH solubilization likely are due to compositional differences of the tires. However, we have no other metrics with which to characterize these samples. In general, tires contain a melange of organic compounds from synthetic polymers and pitches, oils, tars, rubber, and carbon black that have been mixed together to modify the life and workability of the tire. Polycyclic aromatic hydrocarbons generally make up over 200 μg g^{-1} of the final products [45,46].

The number of cars and trucks in urban areas suggests that tire tread material may be a significant source of bioavailable PAH contamination to surrounding sediments. Reddy and Quinn [47] estimated that roughly 1.3×10^9 kg of tire is released each year into the environment around the United States. From the concentrations of benzothiazoles, the estimated amounts of tire tread material in sediments can approach 15% by mass in areas surrounding heavy automobile traffic

such as the San Francisco–Oakland Bay Bridge (USA) [3] and metropolitan Tokyo [2]. As benzothiazoles are water soluble and photolytic [47], they may underestimate actual amounts of tire particles in sediments. The relative bioavailability of PAHs from different tire tread formulations, therefore, may be an important area for future research.

Urban particulates. Although PAHs associated with urban dusts long have been known to have biological effects on terrestrial mammals, we believe that this is the first test of their digestive bioavailability to a marine deposit feeder. Though PAH contamination in sediments around urban areas often is thought to derive from urban dusts, only recently has a molecular fingerprint been used to confirm this hypothesis; thia-arene ratios characteristic of SRM 1649 were found in bottom and suspended sediments from Hamilton Harbor (ON, Canada) [48]. We found that SRM 1649 contained seven digestively bioavailable PAHs (Table 3), which suggests a potential biological impact of these particles on benthic communities.

AP in sediments

Our amended sediment experiments were meant to address the implications of AP additions to real world systems. A number of conceptual models could be used to interpret our results (e.g., EqP). According to EqP, addition of either GF80A tire tread or modified fly ash to LMC sediment ought to reduce the interstitial water concentration of all PAHs and presumably would decrease their bioavailability. The additional sorptive power of the added organic carbon phase has greater influence than the added PAHs. However, these predicted changes in PAH solubility are small and probably not detectable as they are less than the standard deviations of our PAH measurements. Therefore, for the fly ash treatment, in which there were no changes of PAH concentrations except for phenanthrene, we cannot rule out the validity of the soot-amended EqP model.

However, when tire tread material is added, measured changes in some PAH digestive bioavailabilities are opposite EqP predictions; that is, fluoranthene, benzo[*a*]anthracene, benzo[*k*]fluoranthene, and dibenzo[*a,h*]anthracene concentrations increased. We tried to predict the effects of tire tread amendment with an additive model in which concentrations of PAHs solubilized from LMC sediment were increased by

the bioavailable percentage of each PAH in GF80A tire tread (Table 4) multiplied by the weight of tire tread added to these incubations. The additive model underpredicted actual changes in fluoranthene and benzo[*a*]anthracene solubilization, but overpredicted dibenzo[*a,h*]anthracene concentrations (data not shown).

The differences between the actual and predicted changes in gut fluid solubilization from amended sediments may result from the presence of an unmeasured constituent in GF80A tire tread, or from the many component interactions of compounds solubilized by the digestive micelles in gut fluid. Experiments with pairs of lipids have shown both synergistic and antagonistic interactions between solubilizates in gut fluid [49]. Surfactant micelles dynamically adjust sizes and shapes depending on their constituents [50]. For example, fatty alcohols serve as cosurfactants, increasing micelle size and capacities for more nonpolar compounds like cholesterol [50].

CONCLUSION

This paper demonstrates the potential for harmful biological impacts of anthropogenic particles to a deposit-feeding animal. Organic contaminants associated with these particles tend to be bound strongly, resulting in increased solid-water partitioning coefficients relative to natural organic matter in sediments, and decreased predictions of bioavailability using EqP. However, the gut fluids of *A. marina* solubilize much greater concentrations of PAHs from some anthropogenic particles than are available to water. This enhanced exposure likely is due to surfactant micelles in the digestive fluids of this animal.

REFERENCES

1. Bucheli T, Gustafsson Ö. 2000. Quantification of the soot-water distribution coefficient of PAHs provides mechanistic basis for enhanced sorption observations. *Environ Sci Technol* 34:5144–5151.
2. Kumata H, Takada H, Ogura N. 1997. 2-(4-Morpholinyl) Benzothiazoles (24 MoBT) as an indicator of tire wear particles and road dust in the urban environment. In Eganhouse RP, ed, *Molecular Markers in Environmental Geochemistry*. Symposium Series 671. American Chemical Society, Washington DC, pp 291–305.
3. Spies RB, Andresen BD, Rice DW. 1987. Benzthiazoles in estuarine sediments as indicators of street runoff. *Nature* 327:697–699.
4. Meador JP, Stein JE, Reichert WL, Varanasi U. 1995. Bioaccumulation of polycyclic aromatic hydrocarbons by marine organisms. *Rev Environ Contam Toxicol* 143:79–165.
5. Lake JJ, Norwood C, Dimrock C, Bowen R. 1979. Origins of polycyclic aromatic hydrocarbons in estuarine sediments. *Geochim Cosmochim Acta* 43:1847–1854.
6. LaFlamme RE, Hites RA. 1978. The global distribution of polycyclic aromatic hydrocarbons in recent sediments. *Geochim Cosmochim Acta* 42:289–303.
7. Johnson AC, Larsen PF, Gadbois DF, Humason AW. 1985. The distribution of polycyclic aromatic hydrocarbons in the surficial sediments of Penobscot Bay (Maine, USA) in relation to possible sources and to other sites worldwide. *Mar Environ Res* 15:1–16.
8. National Oceanic and Atmospheric Administration. 1989. A summary of data on individual organic contaminants in sediments collected during 1984, 1985, 1986, and 1987. Technicol Memorandum. NOS OMA 47. Rockville, MD, USA.
9. McLeese CW, Ray S, Burridge LE. 1987. Accumulation of polynuclear aromatic hydrocarbons by the clam *Mya arenaria*. In Ketchum BH, Capuzzo JM, Brut WV, Duedall IW, Park PK, Kes-

ter DR, eds, *Wastes in the Ocean,* Vol 6—Nearshore Waste Disposal. John Wiley, New York, NY, USA, pp 81–88.
10. Voparil IM, Mayer LM. 2000. Dissolution of sedimentary polycyclic aromatic hydrocarbons into the lugworm's (*Arenicola marina*) digestive fluids. *Environ Sci Technol* 34:1221–1228.
11. McGroddy SE, Farrington JW. 1995. Sediment pore water partitioning of polycyclic aromatic hydrocarbons in three cores from Boston Harbor, Massachusetts. *Environ Sci Technol* 29:1542–1550.
12. McGroddy SE, Farrington JW, Gschwend PM. 1996. Comparison of the in situ and desorption sediment-water partitioning of polycyclic aromatic hydrocarbons and polychlorinated biphenyls. *Environ Sci Technol* 30:172–177.
13. Knutzen J. 1995. Effects on marine organisms from polycyclic aromatic hydrocarbons (PAH) and other constituents of wastewater from aluminum smelters with examples from Norway. *Sci Total Environ* 163:107–122.
14. Maruya KA, Risebrough RW, Horne AJ. 1997. The bioaccumulation of polynuclear aromatic hydrocarbons by benthic invertebrates in an intertidal marsh. *Environ Toxicol Chem* 16:1087–1097.
15. Naes K, Hylland K, Oug E, Forlin L, Ericson G. 1999. Accumulation and effects of aluminum smelter–generated polycyclic aromatic hydrocarbons on soft-bottom invertebrates and fish. *Environ Toxicol Chem* 18:2205–2216.
16. Bender ME, Roberts MH, deFur PO. 1987. Unavailability of polynuclear aromatic hydrocarbons from coal particles to the eastern oyster. *Environ Pollut* 44:243–260.
17. Chapman PM, Downie J, Maynard A, Taylor LA. 1996. Coal and deodorizer residues in marine sediments—contaminants or pollutant? *Environ Toxicol Chem* 15:638–642.
18. Paine MD, Chapman PM, Allard PJ, Murdoch MH, Minifie D. 1996. Limited bioavailability of sediment PAH near an aluminum smelter: Contamination does not equal effects. *Environ Toxicol Chem* 15:2003–2018.
19. Oug E, Naes K, Rygg B. 1998. Relationship between soft-bottom macrofauna and polycyclic aromatic hydrocarbons (PAHs) from smelter discharge in Norwegian fjords and coastal waters. *Mar Ecol Prog Ser* 173:39–52.
20. Leppanen M. 1995. The role of feeding behaviour in bioaccumulation of organic chemicals in benthic organisms. *Ann Zool Fennici* 32:247–255.
21. Mayer LM, Chen Z, Findlay RH, Fang J, Sampson S, Self RFL, Jumars PA, Quetel C, Donard OFX. 1996. Bioavailability of sedimentary contaminants subject to deposit-feeder digestion. *Environ Sci Technol* 30:2641–2645.
22. Mayer LM, Weston DP, Bock MJ. 2001. Benzo[*a*]pyrene and zinc solubilization by digestive fluids of benthic invertebrates—a crossphyletic study. *Environ Toxicol Chem* 20:1890–1900.
23. Weston DP, Mayer LM. 1998. Comparison of in vitro digestive fluid extraction and traditional in vivo approaches as measures of polycyclic aromatic hydrocarbon bioavailability from sediments. *Environ Toxicol Chem* 17:830–840.
24. Mayer LM, Schick LL, Self RFL, Jumars PA, Findlay RH, Chen Z, Sampson S. 1997. Digestive environments of benthic macroinvertebrate guts: Enzymes, surfactants and dissolved organic matter. *J Mar Res* 55:785–812.
25. Gustafsson Ö, Haghseta F, Chan C, MacFarlane J, Gschwend PM. 1997. Quantification of the dilute sedimentary soot phase: Implications for PAH speciation and bioavailability. *Environ Sci Technol* 31:203–209.
26. Plante CJ, Mayer LM. 1994. Distribution and efficiency of bacteriolysis in the gut of *Arenicola marina* and three additional deposit feeders. *Mar Ecol Prog Ser* 109:183–194.
27. Weston DP. 1990. Hydrocarbon bioaccumulation from contaminated sediment by the deposit-feeding polychaete *Abarenicola pacifica*. *Mar Biol* 107:159–169.
28. Boese BL, Lee H, Specht DT, Randall RC, Winsor MH. 1990. Comparison of aqueous and solid-phase uptake for hexachlorobenzene in the tellinid clam *Macoma nasuta* (Conrad): A mass balance approach. *Environ Toxicol Chem* 9:221–231.
29. Ahrens MJ, Hertz J, Lamoureux EM, Lopez GR, McElroy AE, Brownawell BJ. 2001. The role of digestive surfactants in determining bioavailability of sediment-bound hydrophobic organic contaminants to 2 deposit-feeding polychaetes. *Mar Ecol Prog Ser* 212:145–157.
30. Weston DP, Mayer LM. 1998. In vitro digestive fluid extraction

as a measure of the bioavailability of sediment-associated polycyclic aromatic hydrocarbons: Sources of variation and implications for partitioning models. *Environ Toxicol Chem* 17:820–829.

31. Di Toro DM, 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.

32. Luehrs DC, Hickey JP, Nilsen PE, Rogers TN. 1996. Linear solvation energy relationship of the limiting partition coefficient of organic solutes between water and activated carbon. *Environ Sci Technol* 30:143–152.

33. Walters RW, Luthy RG. 1984. Equilibrium adsorption of polycyclic aromatic hydrocarbons from water onto activated carbon. *Environ Sci Technol* 18:395–403.

34. Voparil IM. 2003. Lipid solubilization by benthic marine invertebrates. PhD thesis. University of Maine, Orono, ME, USA.

35. Whitehouse BG. 1984. The effects of temperature and salinity on the aqueous solubility of polynuclear aromatic hydrocarbons. *Mar Chem* 14:319–332.

36. Afghan BK, Chau ASY. 1989. *Analysis of Trace Organics in the Aquatic Environment.* CRC, Boca Raton, FL, USA.

37. Zou D, Liu K, Pan W-P, Riley JT, Xu Y. 2003. Rapid analysis of PAHs in fly ash using thermal desorption and fast GC-TOF-MS. *J Chromatogr Sci* 41:245–250.

38. Liu K, Xie W, Zhao ZB, Pan YP, Riley JT. 2000. Investigation of polycyclic aromatic hydrocarbons in fly ash from fluidized bed combustion systems. *Environ Sci Technol* 34:2273–2279.

39. Mott HV, Weber WJ. 1992. Sorption of low molecular weight organic contaminants by fly ash: Considerations for the enhancement of cutoff barrier performance. *Environ Sci Technol* 26:1234–1242.

40. Banerjee K, Cheremisinoff PN, Cheng SL. 1995. Sorption of organic contaminants by fly ash in a single solute system. *Environ Sci Technol* 29:2243–2251.

41. Barrick RC, Furlong ET, Carpenter R. 1984. Hydrocarbon and azarene markers of coal transport to aquatic sediments. *Environ Sci Technol* 18:846–853.

42. Dickhut RM, Canuel EA, Gustafsson KE, Liu K, Arzayus KM, Walker SE, Edgecombe G, Gaylor MO, MacDonald EH. 2000. Automotive sources of carcinogenic polycyclic aromatic hydrocarbons associated with particulate matter in the Chesapeake Bay region. *Environ Sci Technol* 34:4635–4640.

43. Tripp BW, Farrington JW, Teal JM. 1981. Unburned coal as a source of hydrocarbons in surface sediments. *Mar Pollut Bull* 12:122–126.

44. Talley JW, Ghosh G, Tucker SG, Furey JS, Luthy RG. 2002. Particle-scale understanding of the bioavailability of PAH in sediment. *Environ Sci Technol* 36:477–483.

45. Jungclaus GA, Games LM, Hites RA. 1976. Identification of trace organic compounds in tire manufacturing plant wastewaters. *Anal Chem* 48:1892–1904.

46. Rogge WF, Hildemann LM, Mazurek MA, Cass GR, Simoneit BRT. 1992. Sources of fine organic aerosol. 3. Road dust, tire debris, and organometallic brake lining dust: Roads as sources and sinks. *Environ Sci Technol* 27:1892–1904.

47. Reddy CM, Quinn JG. 1997. Environmental chemistry of benzothiazoles derived from rubber. *Environ Sci Technol* 31:2847–2853.

48. Marvin CH, McCarry BE, Villella J, Allan LM, Bryant DW. 2000. Chemical and biological profiles of sediments as indicators of sources of genotoxic contamination in Hamilton Harbor. Part I: Analysis of polycyclic aromatic hydrocarbons and thia-arene compounds. *Chemosphere* 41:979–988.

49. Voparil IM, Mayer LM, Place AR. 2003. Interactions among contaminants and nutritional lipids during mobilization by digestive fluids of marine invertebrates. *Environ Sci Technol* 37:3117–3122.

50. Carey MC, Small DM. 1970. The characteristics of mixed micellar solutions with particular reference to bile. *Am J Med* 49:590–608.

51. Xie W-H, Shiu W-Y, Mackay D. 1997. A review of the effects of salts on the solubility of organic compounds in seawater. *Mar Environ Res* 44:429–444.