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Long-Term Effects of Dredging Operations

Sediment Extraction Using Deposit-Feeder Gut Fluids: A Potential Rapid Tool for Assessing Bioaccumulation Potential of Sediment-Associated Contaminants

Donald P. Weston, Rod N. Millward, Lawrence M. Mayer,
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July 2002

Environmental Laboratory

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US Army Corps of Engineers
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Environmental Effects of Dredging Progam

Long-Term Effects of Dredging Operations

Sediment Extraction Using Deposit-Feeder Gut Fluids: A Potential Rapid Tool for Assessing Bioaccumulation Potential of Sediment-Associated Contaminants (ERDC/EL TR-02-18)

ISSUE: Traditionally, measuring contaminant bioavailability from dredged material has involved a 28-day bioaccumulation test. This phase is often the most expensive component of dredged material testing, due to the time taken and the trained analytical technique required. Current testing guidelines include a screening tool, the theoretical bioaccumulation potential (TBP), to minimize the need to resort to such bioaccumulation tests. However, TBP is limited to nonpolar organic compounds. Experimental screening tools, such as sediment extraction using deposit-feeder gut fluids, might offer a reliable screening tool for assessing concentrations of both polar and nonpolar compounds potentially available for bioaccumulation.

RESEARCH OBJECTIVE: The objective of this project was to assess the efficacy and suitability of sediment extractions using natural and synthetic invertebrate gut fluid as a measurement of contaminant bioavailability and bioaccumulation.

SUMMARY: Recent studies have shown that contaminants released from sediment following in vitro incubation with deposit-feeder digestive fluid can provide a reliable measurement of bioavailability, and might be a good predictor of bioaccumulation. It is therefore conceivable that chemical analysis of gut-fluid extracts might be a rapid and cost-effective tool for screening potential bioaccumulation hazards associated with dredged sediments. This report outlines work to date on this technique, as well as current research goals. These goals include correlation analysis of bioaccumulation with gut-fluid extraction for a number of analytes, organisms, and sediments; development of a biomimetic (synthetic) gut fluid; examination of the importance of redox chemistry and digestive ligands in metal bioaccumulation; and the effects of contaminant and sediment matrix interactions on bioaccumulation.

AVAILABILITY OF REPORT: The report is available at the following Web site: *http://libweb.wes.army.mil/index.htm.* The report is also available on Interlibrary Loan Service from the U.S. Army Engineer Research and Development Center (ERDC) Research Library, telephone (601) 634-2355.

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Contents

List of Figures

Preface

This report was prepared as part of the Long-Term Effects of Dredging Operations (LEDO) program supported by the U.S. Army Corps of Engineers. The program monitor was Dr. Joe Wilson, Headquarters, U.S. Army Corps of Engineers; Program Manager was Dr. Robert Engler, U.S. Army Engineer Research and Development Center (ERDC), Vicksburg, MS.

The work was performed under the direction of the Environmental Laboratory (EL), ERDC. The principal investigators were Dr. Donald Weston (University of California, Berkeley), Dr. Lawrence Mayer (University of Maine), Mr. Ian Voparil (University of Maine), Dr. Rod Millward (Analytical Services Inc., Vicksburg, MS), and Dr. Guilherme Lotufo (EL, ERDC). Dr. Victor McFarland (EL, ERDC) and Dr. Todd Bridges (EL, ERDC) were technical reviewers of this report.

The work was conducted with the help of Mr. B. Maurice Duke and Mr. Cory McNemar (Analytical Services Inc., Vicksburg MS).

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1 Introduction

Objectives

This report discusses the feasibility of extracting contaminants from sediments using gut fluids collected from benthic invertebrates as a simple, cost-effective, and biologically relevant assessment of contaminant bioavailability. Potentially, such extractions are predictive of in vivo contaminant bioaccumulation and hence might offer a rapid screening tool for bioaccumulation.

This report introduces current guidelines for bioaccumulation assessment (U.S. Environmental Protection Agency (USEPA) and U.S. Army Corps of Engineers (USACE) 1991, 1998) and discusses how a currently adopted screening method applicable to nonpolar organic contaminants (theoretical bioaccumulation potential, or TBP) presently reduces the frequency of costly and laborious bioaccumulation studies for this group of contaminants. Also discussed is the potential value of gut fluid extraction as an alternate screening tool for a wider range of contaminants including metals. Finally, further research needs for development of this approach, and its potential contribution to current USEPA and USACE dredged sediment evaluation procedures (USEPA and USACE 1991, 1998), are presented.

Bioaccumulation Assessment for Toxicological Evaluation of Dredged Material

There are more than 40,000 km of navigation channels and over 400 harbors in the United States. Maintenance dredging generates approximately 400 million m^3 of dredged material for disposal annually, about 80 percent of which is placed in designated sites in the aquatic environment. Dredging and placement of dredged material are regulated in accordance with a number of environmental statutes including the National Environmental Policy Act (NEPA) of 1969, the Clean Water Act (CWA) of 1972, and the Marine Protection, Research and Sanctuary Act (MPRSA) of 1972. The

USACE has primary responsibility for permit issues for all dredging necessary to maintain commercial waterways throughout the United States. The permitting process requires a detailed evaluation of the specific dredged material in accordance with regulatory criteria using technical evaluation procedures developed jointly by the USEPA and USACE (USEPA and USACE 1991, 1998).

The primary evaluative endpoints are toxicity and bioaccumulation potential of sediment-associated contaminants to benthic organisms. Bioaccumulation assessment is used as a direct indicator of contaminant bioavailability in the dredged material. While contaminants might not be present at levels high enough to promote detectable biological effects in laboratory bioassays, organisms will bioaccumulate many contaminants present in the surrounding media. In addition, bioaccumulation may result in impacts that will not be reflected by the specific endpoints used in standard toxicity tests, but that may still lead to significant impacts at the population level. Moreover, many contaminants are biomagnified up through the aquatic food web, posing serious hazard to higher consumers, including humans.

The current guidance manuals (USEPA and USACE 1991, 1998) utilize a tiered approach designed to proceed from simple, cost-effective evaluations, which take advantage of available information, to more complex and costly assessments that fill data gaps and reduce uncertainty. An evaluation proceeds through the tiers until necessary and sufficient information is developed to make a decision about how the dredged material should be managed.

Tier I is primarily an evaluation of existing physical, chemical, or biological information. In many cases, a permit decision can be made in Tier I, thus providing a timely and cost-effective regulatory decision. However, in dredged material evaluations involving concerns about contaminants, Tier I will often indicate that further testing in subsequent tiers is warranted.

Tier II is designed to take advantage of predictive assessment models to make cost-effective decisions. The TBP model is used in Tier II to evaluate the potential for benthic impact. The TBP calculation in Tier II is applied to predict the magnitude of bioaccumulation of nonpolar organic contaminants in the dredged material and in the reference material. The TBP expresses the predicted steady-state concentration of nonpolar organic contaminants (which include all the priority pollutant polycyclic aromatic hydrocarbons (PAHs), chlorinated hydrocarbon pesticides, polychlorinated biphenyls (PCBs), dioxins, and furans) in benthic organisms exposed to sediment. When the TBP for nonpolar organic contaminants of concern in the dredged material exceeds the TBP for the reference sediment, or contaminants of concern other than nonpolar organics are present in the dredged material, bioaccumulation is evaluated experimentally in Tier III.

Tier III testing assesses experimentally the impact of contaminants in the dredged material on appropriately sensitive organisms to determine if there is the potential for an unacceptable impact at the disposal site. Tier III and

IV assessments include toxicity and bioaccumulation testing and generally represent a significant increase in both complexity of analysis and interpretation and expenditure in terms of both time and money.

Improved Screening Methods for Assessing Contaminant Bioaccumulation

Estimation of TBP in Tier II, i.e., steady-state concentration in benthic organisms exposed to sediment, is limited to nonpolar organic contaminants. At present there is no analogous methodology for predicting bioaccumulation from dredged material contaminated with polar organics, metals, organometals, organic acids, or salts. Bioaccumulation potential for contaminants other than nonpolar organics must currently be evaluated using bioaccumulation testing in Tiers III and IV. Bioaccumulation testing is typically the most costly component of the dredged material evaluation process, due mainly to costs associated with prolonged (e.g., 28-day) exposures and analysis of tissue concentrations at the conclusion of the test. Therefore, development of a rapid and more universal screening method for simultaneously predicting bioaccumulation of nonpolar organics and other classes of contaminants would result in considerable cost savings.

Assessment of Bioaccumulation Using Gut Fluid Extraction

It is well established that sediment-associated contaminants enter aquatic food webs through ingestion by deposit-feeding organisms. Uptake of contaminants via the diet is thought to be the main route of bioaccumulation in deposit feeders for many metals (Wang and Fisher 1999), PAHs (Weston, Penry, and Gulmann 2000), chlorinated hydrocarbons, and other hydrophobic contaminants (Lee et al. 2000). In order for a contaminant to be accumulated via ingestion, it must generally be desorbed from the ingested particle and solubilized in the fluids of the gut lumen. Recent studies have shown that in vitro incubation of contaminated sediment with deposit-feeder digestive fluid and quantification of contaminant that is extractable in that fluid often provides a reliable measurement of bioavailability (Mayer et al. 1996; Weston and Mayer 1998a, 1998b; Lawrence et al. 1999; Mayer, Weston, and Bock 2001). This measure is consistent with more traditional approaches for measuring bioavailability (Weston and Mayer 1998b) and is a good predictor of contaminant bioaccumulation (Weston and Maruya 2002). This report summarizes the progress made in the development of this in vitro technique and addresses its potential application to the current USEPA/USACE dredged material evaluation process.

2 Current Status of Digestive Fluid Extraction Approach

Conceptual Basis

Deposit-feeding and some suspension-feeding organisms accumulate many heavy metals and hydrophobic organic compounds via the ingestion of sediment (Landrum and Robbins 1989; Lee et al. 2000; Weston, Penry, and Gulmann 2000; Wang and Fisher 1999). However, a substantial proportion of any given contaminant is not desorbed from the particles while in the gut, and passes out of the organism via the feces. Ideally, environmental management decisions pertaining to contaminated sediments should include consideration of the bioavailable fraction rather than the total contaminant concentration. However, existing chemical methods of analysis include an extraction step that is intended to extract all of the targeted contaminant from sediments using a strong acid or strong organic solvent. As a result, these approaches overestimate the bioavailable fraction.

Several investigators have attempted to improve human health risk assessment by developing fluids that mimic human stomach fluid, and to use these fluids as in vitro extractants to estimate how much contaminant would be bioavailable from soil if incidentally ingested by humans (Ruby et al. 1993; Hack and Selenka 1996; Jin, Simkins, and Xing 1999; Oomen et al. 2000). For purposes of ecological risk assessment, attempts to extract the bioavailable fraction have not mimicked any natural digestive fluid, but instead have employed approaches that are simply weaker versions of exhaustive extraction procedures (e.g., Tessier and Campbell 1987; Smith and Flegal 1993; Tang and Alexander 1999). While such approaches may be preferable to an exhaustive extraction, the extraction conditions used are fundamentally unlike those in deposit-feeder guts (Mayer et al. 1997), and none of these weaker extraction protocols have become broadly adopted.

A new approach for assessment of the bioavailability of particle-associated contaminants has recently been proposed that employs the digestive fluid of deposit feeders to solubilize contaminants (Mayer et al. 1996). Digestive fluid of a deposit-feeding organism is removed from the gut lumen,

and the sediments of concern are incubated with that fluid in vitro. The amount of the particle-associated contaminant that is desorbed in the fluid is then quantified on the presumption that sediment-associated contaminants must first be solubilized in order to be bioavailable (excluding the potential for intracellular digestion in some taxa). While the approach does not address the subsequent absorption of the solubilized contaminant across the gut wall, the method at least places an upper limit on the contaminant that is likely to be made bioavailable from a given sediment during gut passage. The approach has the simplicity of a chemical extraction, but by using digestive fluid rather than an exotic solvent, the approach provides more biological realism than is achieved by conventional chemical methods.

Recent attempts to assess sediment risk using in vitro digestive fluid extraction have illustrated some advantages of the approach over conventional measures of bioavailability involving exposure of live organisms (Weston and Maruya 2002). First, it can be done much faster than conventional bioaccumulation testing (a few hours versus nearly a month), with associated cost savings and faster data availability, and thus offers a screening tool applicable to the USACE/USEPA tiered assessment protocol. Second, the digestive fluid approach to predict bioaccumulation eliminates the potential confounding effects of biotransformation, which can lead to significant metabolism of certain compounds in some test species, thus leading to an underestimation of bioaccumulation. Third, the technique allows evaluation of sediments by a consistent method over a wider range of abiotic parameters (e.g., grain size, salinity) than would be tolerated by any single bioaccumulation test species.

The digestive fluid extraction approach is probably not useful for compounds for which ingestion is likely to be a minor route of uptake (e.g., hydrophilic organic compounds, Weston and Mayer 1998b) or those for which intestinal absorption rather than solubilization constrains uptake (e.g., chromium). In addition, reliance upon natural populations of deposit feeders as a source of digestive fluid limits widespread adoption of the approach, but the use of commercially available substances having extraction properties similar to the natural constituents shows promise (Chen and Mayer 1999, Ahrens et al. 2001).

It is therefore theoretically defensible to suggest that in vitro gut fluid contaminant extraction provides a direct measurement of contaminant bioavailability to deposit feeders. In addition, the method may be useful as a predictive tool for contaminant bioaccumulation in sediment-dwelling invertebrates, dependant upon a number of conditions. To illustrate this extrapolation, the steps whereby a contaminant is released from sediment and accumulated ultimately within the tissues of a benthic organism must first be considered. The concentrations of sediment-associated contaminants bioaccumulated via ingestion by deposit feeders can be considered using a multistep model (Figure 1). The first two functions (a and b, Figure 1) consider the concentration of contaminants transferred from the sediment into the gut fluids, calculated as the concentration of contaminant solubilized by the gut fluid (a) minus the concentration subsequently reabsorbed back

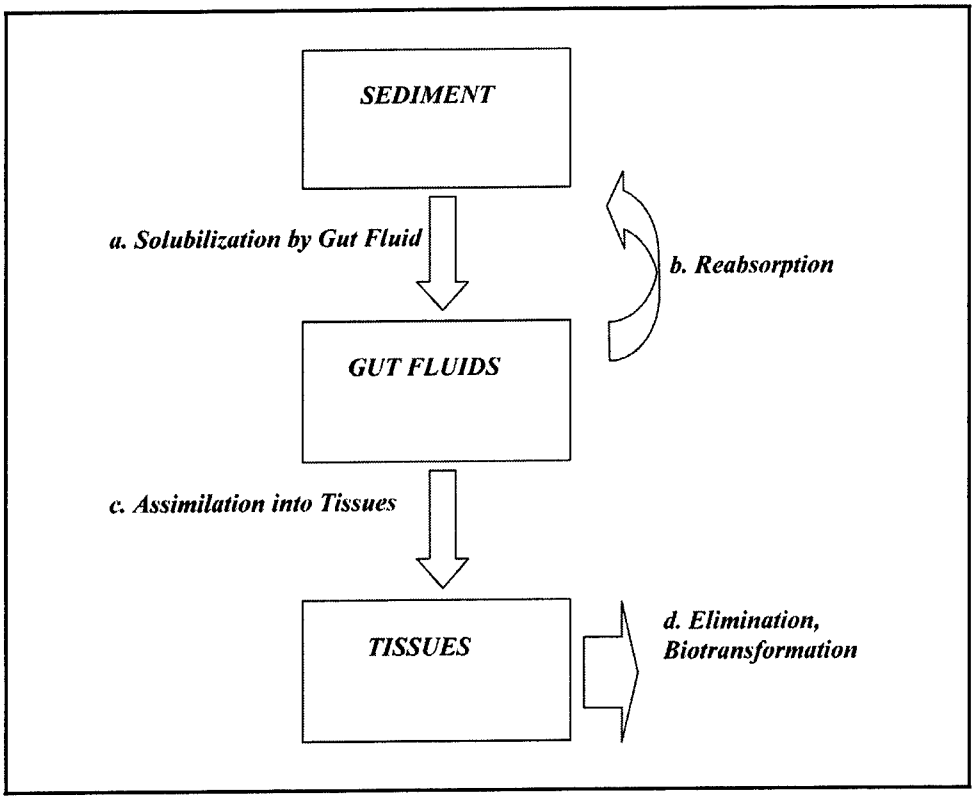

Figure 1. Conceptual model showing the bioaccumulation of contaminants from sediments via deposit feeding

into the sediment matrix (b). The difference, (a) – (b), equates to the net in vitro gut-fluid extracted concentration. The amount of contaminant absorbed from gut fluids into tissues (c) is proportional to the absorption efficiency. Final tissue burdens are a product of the absorbed fraction minus losses due to metabolism and elimination (d). Using this construct it can be seen that, in instances when the limiting factor for contaminant uptake is bioavailability and not absorption efficiency, bioaccumulation is likely to be proportional to bioavailability. Regression analyses of gut-fluid extracted contaminant concentrations versus bioaccumulated body burdens reveal strong positive correlations for a number of contaminants (cadmium, lead), suggesting that gut-fluid extractions might be considered as predictors of bioaccumulation (Weston and Maruya 2002).

Extraction Protocol

While conceptually the digestive fluid of any deposit-feeding species could be used, the need to maximize fluid volume has limited past attempts to large organisms. The vast majority of the work to date has been done with arenicolid polychaetes. *Arenicola brasiliensis* from the Eastern Pacific typically provides about 1 mL of fluid per individual (Weston and Mayer 1998a). *Arenicola marina* from the North Atlantic typically provides

0.5 mL per individual (unpub. data). Some other detailed work on contaminant extractions has been done with the holothuroid *Parastichopus californicus* (Mayer et al. 1996), the echiuran *Urechis caupo* (Weston and Mayer 1998a), the polychaetes *Nereis succinea* and *Pectinaria gouldii* (Ahrens et al. 2001), and a survey of 18 species reported in Mayer, Weston, and Bock (2001).

Following collection of organisms, a gut evacuation period in water (without sediment) may be useful, as loss of sediment from the gut during this period simplifies fluid removal and enhances the volume recovered. An evacuation period of about 30 hr for *A. brasiliensis* has been shown to have no effect on contaminant solubilization potential or most biochemical properties of the fluid, though there can be some loss in fluid surfactancy (Mayer, Weston, and Bock 2001).

Fluid recovery is accomplished by exposure of the gut by dissection and withdrawal of fluid through the gut wall with a pipette. In large organisms (e.g., *P. californicus* and *U. caupo*), it is possible to let the fluid simply drain from the gut by holding the open end of the gut over a collection vial. Any residual sediment in the fluid is removed by centrifugation, and the fluid is frozen at -80 °C until use. The maximum storage time has not been established, though holding periods of several months are commonly used.

If multiple extractions are to be made over which the data are to be compared, as would typically be the case, it is essential to composite fluid from a sufficient number of individuals to obtain a single homogeneous batch. Individuals of *A. brasiliensis* have shown a threefold variation in contaminant solubilization potential of their gut fluids (Weston and Mayer 1998a), and presumably other species would be equally variable. A gut fluid composite from 30 to 150 individuals has typically been used (Weston and Mayer 1998a; Weston and Maruya 2002). Recent studies on gut fluid composition and mechanisms of contaminant solubilization are leading to the development of a synthetic, biomimetic gut fluid. Such a gut fluid substitute would enable standardization and widespread adoption of this methodology.

Sediment extractions are made using wet sediment in order to avoid any bioavailability changes that may accompany drying. The sediment is placed in a centrifuge tube, and digestive fluid is added at a dry-sediment-to-fluid ratio of up to 0.3 g dry sediment per milliliter gut fluid (Voparil and Mayer 2000; Weston and Maruya 2002). As the extraction efficiency is dependent on the sediment:fluid ratio (see paragraph "Solid:fluid ratio"), the ratio should be held constant across all sediments to be tested.

Extractions are made under constant agitation (e.g., orbital or reciprocating shaker) for time periods typically ranging from 2 to 4 hr. For hydrophobic organic compounds that have been spiked into the sediment, the duration of extraction is largely irrelevant as most of the contaminant extraction occurs in the first few minutes (Ahrens et al. 2001; Weston, unpub. data). In-situ-contaminated, field-collected sediments may show slower extraction rates,

with extractions incomplete even after 4 hr for some compounds (Voparil and Mayer 2000). Trace metal extraction is particularly time-dependent, with extraction efficiencies tending to increase up to a period of about 1 hr, and then, for some metals, decreasing with greater durations as solubilized metal readsorbs to the sediment (see paragraph "Variables Influencing Extraction Efficiency"). Extractions have typically been made at room temperature, although recently collected data (Weston, unpub.) indicate cooling during the extraction may better maintain gut fluid conditions as found in vivo.

At the completion of the extraction, fluid is usually recovered by centrifugation. Some investigators have used centrifugal forces up to 8,000 g (Chen and Mayer 1998), while others have used 2,100 g (Weston and Mayer 1998a). Voparil and Mayer (2000) added a filtration step (0.45 μm) to the centrifugation. The definition of "solubilized contaminant" is operational, and the stated extraction efficiency probably decreases as centrifugation speeds increase or a filtration step is added.

Contaminant concentrations in the extractant are quantified by conventional chemical means, and have often been used to calculate an extraction efficiency as:

$$\%\ extracted = \frac{C_{df} \times \left(V_{df} + V_w\right) \times 100}{C_s \times M_s} \tag{1}$$

where

C_{df} = concentration of contaminant in digestive fluid

V_{df} = volume of digestive fluid used in the extraction

V_w = volume of water initially incorporated in the wet sediment extracted

C_s = pre-extraction concentration of contaminant in the sediment

M_s = mass of sediment extracted (dry weight)

Generally, it would be desirable to subtract the concentration of contaminant existing in the digestive fluid pre-extraction prior to doing these calculations. This correction is likely to be relatively small for organic compounds, but digestive fluid can have very high trace metal concentrations even in organisms obtained from relatively pristine areas (Chen et al. 2000).

Mechanisms of Solubilization

Gut fluid can enhance the solubility of metals above that of clean water if complexing agents are present to bind the metals in solution. Most con-

taminant metals of concern are toxic because of their interaction with biological molecules, especially proteins. These interactions are usually strong ones, involving covalent bonding. Gut fluid shows a great ability to increase solubility of metals because it is a protein-rich solution. In other words, gut fluid has a biochemical composition functionally similar to the tissues of the animal. Hence, a variety of metals are found to be enriched in gut fluids of deposit feeders even in uncontaminated sediments; furthermore, they are enriched in proportion to the proteinaceous compounds dissolved in the gut fluid (Chen et al. 2000; Mayer, Weston, and Bock 2001). The pattern of gut fluid enrichments among metals shows a peak for metals with strong capacity for covalent interactions – the so-called Irving-Williams order – in keeping with this prediction.

Contaminant metals are solubilized from sediments by gut fluid also in proportion to the amino acid content of the fluid (Mayer et al. 1996; Chen and Mayer 1999). Careful work identifying the actual binding sites – the specific ligand – has been successfully carried out only for copper, using a site-blocking approach in which the suspected candidate was inactivated by blocking its binding group. In this case, the amino acid histidine was found to be responsible for most of the solubilizing power of the gut fluid (Chen and Mayer 1998). Chen et al. (unpublished) have also worked on identifying the responsible ligands for lead, with little success so far. Several obvious amino acid candidates have been tested and found not to have primary responsibility for lead-solubilizing capacity. It is believed that lead has a more complicated chemistry of solubilization, probably involving a variety of ligand types rather than the fairly simple mechanism derived for copper. It seems likely that this complexity extends to other metals as well, though this behavior has not been explored.

The mechanisms of solubilization of hydrophobic organic chemicals (HOCs) are somewhat different. These compounds are nonpolar; that is, they have an evenly spaced electron distribution. When introduced into a polar environment like water, HOCs tend to aggregate and limit their interaction with the aqueous phase. These interactions are nonspecific, occurring not because of HOCs affinities for each other, but rather due to their aversion to water. A number of compounds in invertebrate gut fluids offer a nonpolar refuge for HOC solubilization, including digestive surfactants, proteins, and perhaps food hydrolysates such as membrane fragments. Most animals rely on complex aggregates of these compounds in order to shuttle hydrophobic compounds from the ingested material, across the bulk aqueous solution, to the digestive epithelium for absorption.

In deposit-feeding marine invertebrates, much of the solubilization of hydrophobic compounds is due to surfactant micelles. Presumably, this mechanism developed to gain access to nutritional lipids in sediment such as sterols. However, due to the nonspecificity of HOC interactions, this mechanism also solubilizes hydrophobic contaminants such as PAHs at concentrations much greater than aqueous solubility. For example, *A. marina* gut fluids can dissolve ~2 μg of benzo[a]pyrene (mL^{-1} of gut fluid) – over one thousand times seawater solubility for this PAH. Previous work

(Voparil and Mayer 2000) suggests that micelles are responsible for as much as 80 percent of the PAHs in *A. marina* gut fluid (Figure 2). Proteinaceous material in the gut is likely responsible for much of the remaining PAH solubilization. Gut fluids are very protein-rich (Mayer et al. 1997), and large globular proteins offer a hydrophobic interior environment that can solubilize HOCs (Backus and Gschwend 1990; Voparil and Mayer 2000).

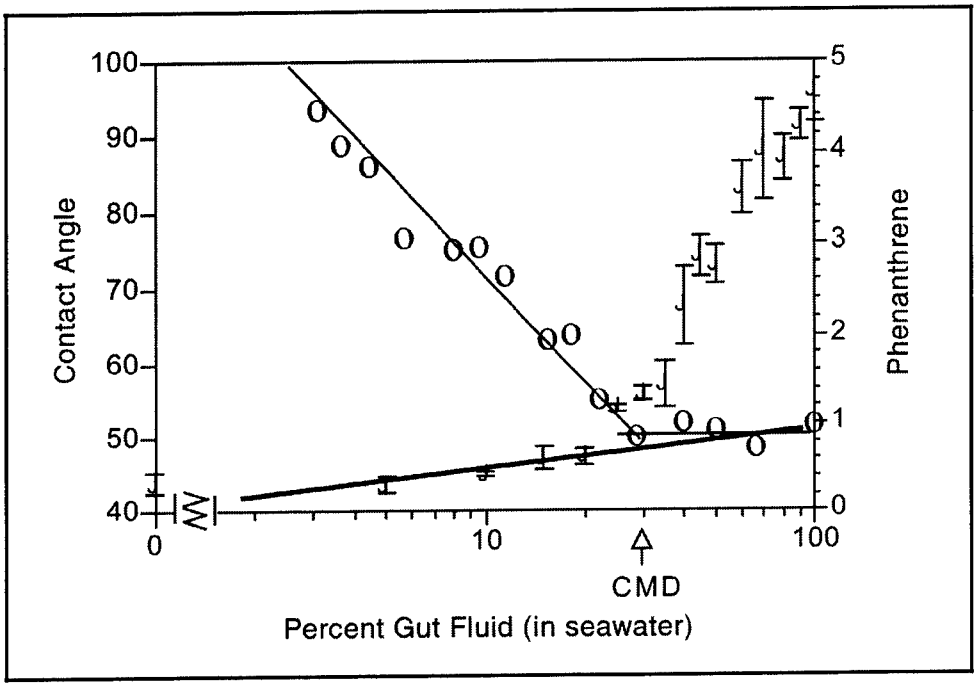

Figure 2. Contact angles and pure PAH solubilization by *A. marina* digestive fluid titrated with clean seawater. Abscissa represents the dilutions of the original solution. Left ordinate is the contact angle (O). Right ordinate is the concentration of phenanthrene solubilized (J). Thin solid lines fit to contact angle data intersect at the critical micelle dilution (CMD). Micelles are present when the gut fluid is at a higher percentage than the CMD. Thicker solid lines fit to PAH concentration data below the CMD are extrapolated to 100-percent gut fluid to determine PAH solubilization by nonmicellar components of the gut fluid. The nonmicellar components of gut fluid solubilize 23 percent of the total phenanthrene; thus, micelles are responsible for ~80 percent of the PAH solubilized by 100-percent gut fluid (Voparil and Mayer 2000)

Most HOC research has focused on PAH bioavailability. The nonspecific nature of HOC interactions suggests that micelles will be important for the availability of a number of other hydrophobic contaminants. Ahrens et al. (2001) found that gut fluid solubilization of tetrachlorobiphenyl and hexachlorobenzene was also related to surfactancy. Interactions with more polar, organic contaminants are unknown, but work with digestive surfactants used by vertebrates suggests that micellization is an important mechanism in the solubilization of compounds as polar as phospholipids.

Variables Influencing Extraction Efficiency

Species selection

The concept of bioavailability requires that attributes of the animal, as well as the contaminated matrix, be considered. The ability of an animal to digestively solubilize contaminants from sediments depends on various aspects of the animal's digestive physiology, such as the concentration of solubilizing agents, time of exposure, and sediment-gut fluid ratio. Aspects of these controls have received some attention, which has made it clear that large differences exist among species with regard to their ability to digestively solubilize contaminants from the same substrate.

The most important controlling parameter on extent of digestive solubilization appears to be concentration of solubilizing agent. While individuals of a single species typically show considerable variance in the concentration of various biochemicals in their gut fluids, the variances among species is often much greater. These trends lead to strong phyletic control on the potential for contaminant solubilization (Figure 3). In a study of 18 benthic invertebrate species, digestive fluids from echinoderms and a cnidarian tended to be relatively weak; those from polychaetes and echiurans were relatively strong and those from taxa such as sipunculans and molluscs were intermediate (Mayer, Weston, and Bock 2001). These trends correlated strongly

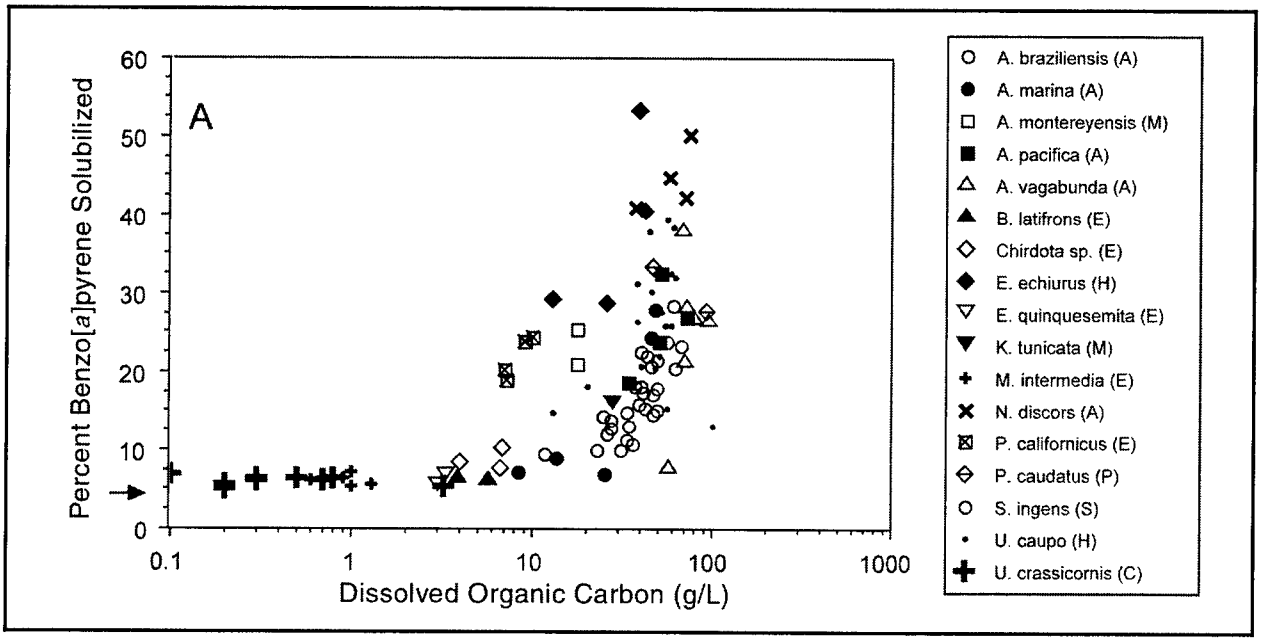

Figure 3. Percent of benzo[*a*]pyrene spike solubilized versus the organic carbon concentration in gut fluid. Phylum given by letter in parentheses following species name (C = cnidarian, E = echinoderm, A = annelid, S = sipunculid, P = priapulid, H = echiuran, M = mollusk). Arrow refers to percent of spike solubilized by the seawater control. X-axis values plotted on log scale to show detail at low concentrations. Only extractions using midgut fluids, which show greater solubilization capacity than fluid from other gut sections, are plotted (Mayer, Weston, and Bock 2001)

with concentrations or activities of digestive biochemicals such as dissolved amino acids, total dissolved organic matter, and enzyme activities, but not with pH. These experiments were carried out on relatively large benthic animals, due to the need for milliliter quantities of gut fluid for all of the analyses. Though experiments could not be conducted with very small invertebrates, the correlations with digestive parameters were strong enough to permit some predictive ability for contaminant solubilization of smaller species. Contaminant solubilization potential of gut fluid from even small species, which do not provide enough fluid for direct solubilization measurements, can probably be estimated from correlates such as dissolved amino acids for which only a few microliters of fluid are necessary for quantification.

Kinetics

Another important control on solubilization is the time of exposure of sediment to the gut fluid. The actual exposure times in vivo are often difficult to ascertain. Nominally, one might expect gut residence time to be the appropriate incubation period. This parameter has been measured for many species and, though it is variable even within an individual, it provides some bound on exposure times. However, as discussed above, solubilization depends on the concentration of solubilizing agents, and these concentrations vary strongly along the length of the gut of almost all species examined (e.g., Mayer et al. 1997). Thus, most reaction would be expected to occur within midgut regions where concentrations of active digestive agents are usually at their highest (Figure 4). Almost certainly a similar pattern would ensue for hydrophobic contaminant solubilization, due to presence of micelles only in digestively active midgut sections (Mayer, Weston, and Bock 2001). It might therefore be more accurate to consider only midgut residence times. The accuracy of this assumption is questionable given the episodic defecation behavior observed for many invertebrates, which are under pressure to minimize exposure time at the sediment-water interface due to potential predation.

How important are solubilization kinetics, relative to in vivo exposure times? Metal dissolution kinetics in gut fluid incubations often show incomplete reaction in the probable in vivo reaction times (e.g., Chen and Mayer 1999), implying considerable importance to assignment of incubation time for this class of contaminants. Most HOCs, such as PAHs and chlorinated hydrocarbons, on the other hand, appear to equilibrate, or at least reach some kind of steady-state, within 10 to 15 min, which is usually quicker than probable in vivo exposure times (Voparil and Mayer 2000; Ahrens et al. 2001). Some PAHs (e.g., pyrene) have shown longer times to maximum release. However, the time of exposure may be less critical for HOCs in general.

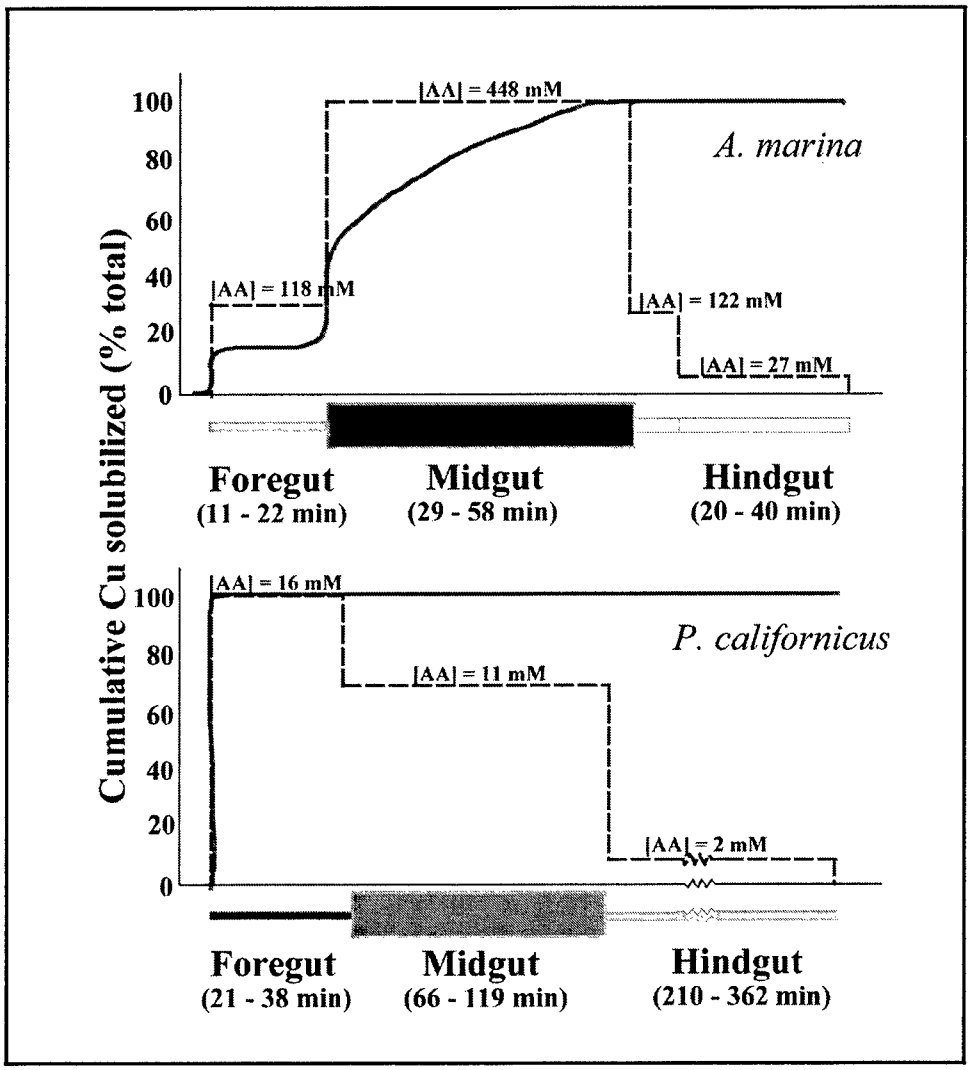

Figure 4. Extent of copper solubilization suggested by amino acid (AA) concentrations along the guts of *A. marina* (lugworm) and *P. californicus* (sea cucumber). Note that copper is predicted to be maximal in the gut segment containing the highest amino acid concentrations. This plot assumes that no absorption of solubilized copper occurs (Chen and Mayer 1999)

Solid:fluid ratio

The dietary solubilization of sedimentary contaminants is not an attribute of either the sediments or the organism alone. Rather it is a product of organism-sediment interactions, limited by either the amount of bioavailable contaminant ingested by the organism or by the amount of digestive solubilizing agent present to deliver contaminants to the digestive epithelia. The same factor is not necessarily always limiting. In fact, the limiting agent may switch between the two due to physiological adaptations of the organism. When using in vitro gut fluid incubations to measure the bioavailability of sedimentary contaminants, experimental parameters should approximate digestive conditions in order to determine digestive exposure during one gut

passage. By varying incubation conditions, additional information becomes available.

Varying the relative amounts of sediment and gut fluid (the solid:fluid ratio) in a set of incubations can shift the limiting phase from the amount of bioavailable sedimentary contaminant (at low solid:fluid ratios) to the amount of digestive ligand (at higher ratios). Voparil and Mayer (2000) found that the greatest fraction of an individual PAH, i.e. expressed as the percentage of the total amount of PAHs in the incubation, was always released during the most dilute incubation conditions. However, at higher solid-fluid incubation ratios, the concentrations of PAHs reached a plateau, i.e., appeared to saturate. Under such conditions, reporting the concentration of PAHs in gut fluid as opposed to the percentage of total PAHs released conveys more mechanistic information for understanding the organism-sediment interaction. These results were from sediments with very high PAH concentrations; gut fluids may not saturate when exposed to sediments with low to moderate contamination.

Another way of investigating the limiting phase of the interaction is with repeated extractions of the same aliquot of sediment with fresh gut fluid. Conceptually, this approach mimics dilute solid-fluid conditions. Chen and Mayer (1999) found that the total remobilizable copper in a contaminated sediment declined rapidly after the first incubation cycle providing a measure of the total amount of bioavailable copper in the sediment (Figure 4). Lead, on the other hand, was released at similar, high concentrations even after seven incubation cycles, indicating limitation of digestive ligands. Voparil and Mayer (2000) found that repeat incubations extended this conclusion to organic contaminants, showing that digestive agent saturation limited the release of PAHs from sediment. Clearly, an animal's physiology sets both upper and lower limits on the availability of contaminants traveling through its gut.

Total organic carbon

It is well recognized that sediment organic carbon plays a major role in determining the bioavailability of hydrophobic organic compounds and some trace metals. As sediment organic carbon content increases, contaminant bioavailability decreases (Di Toro et al. 1991). Thus, if digestive fluid extraction is a suitable measure of bioavailability, one would expect solubilization to be inversely proportional to sediment organic carbon content, and existing data show this to be the case.

In a study of six marine sediments, benzo[*a*]pyrene and phenanthrene extraction by digestive fluid was shown to be correlated with organic carbon content (Weston and Mayer 1998). As the percentage of organic carbon among the sediments increased from about 0.1 to 1.4 percent, benzo[*a*]pyrene solubilization decreased from 52 to 13 percent. Solubilization of methylmercury by digestive fluid has also been shown to be inversely related to organic content (Lawrence et al. 1999). As organic carbon content

among four sediments increased from 1 to 16 percent, methylmercury solubilization decreased from 38 to 3 percent. No similar relationship was seen for inorganic mercury, for which there would be less a priori expectation of an organic matter dependency.

Competition

Voparil and Mayer (2000) found that gut fluids were able to solubilize considerably more PAHs from pure PAH solids than was possible from highly contaminated sediments. They suggested that one reason for this discrepancy is competition for uptake sites in gut fluid, e.g., space in digestive micelles. Many HOCs and naturally occurring lipids should be able to compete for similar uptake sites. Likewise, metal-binding ligands in gut fluids may well be able to bind several different metals so that competition may affect bioavailable contaminants for this class of contaminants as well.

Contaminant concentration

In vitro desorption of contaminants under simulated gastric conditions has been shown to be dependent upon contaminant concentration (Jin, Simkins, and Xing 1999), but the subject of concentration dependency of bioavailability has in general received very little attention. Estimates of absorption efficiency from ingested sediment, for example, are routinely provided without any recognition of a potential concentration dependency, and estimates among multiple studies are routinely compared without consideration of whether differences may be due to variation in contaminant levels used among the studies (Wang and Fisher 1999). The limited data available, based only on preliminary experiments (Weston, unpub.), suggest contaminant bioavailability may indeed be concentration-dependent, though in a complex manner.

When a gradient of contaminant concentrations was obtained by spiking a single sediment with increasing amounts of contaminant, the proportion of contaminant extracted by digestive fluid has been found to increase or decrease as a function of spiked contaminant concentrations (Figure 5). Benzo[*a*]pyrene extraction tended to increase from about 50 percent to 70 percent as contaminant concentration increased over a four order-of-magnitude range. Although the precise mechanism was not investigated, the data suggest the compound may have, at low concentrations, partitioned into sedimentary phases from which extraction was relatively difficult. As those phases or sites become saturated, additional compound partitioned into less favorable sites (or more labile, reversible, or less desorption-resistant sites) within the matrix, and extraction efficiency increased. At least for some trace metals, the preliminary data suggest an opposite relationship, with extraction efficiencies decreasing as contaminant concentration increases (Figure 5). Metal solubilization can be strongly dependent upon the availability of complexing ligands within the gut fluid such as certain amino acids (Mayer,

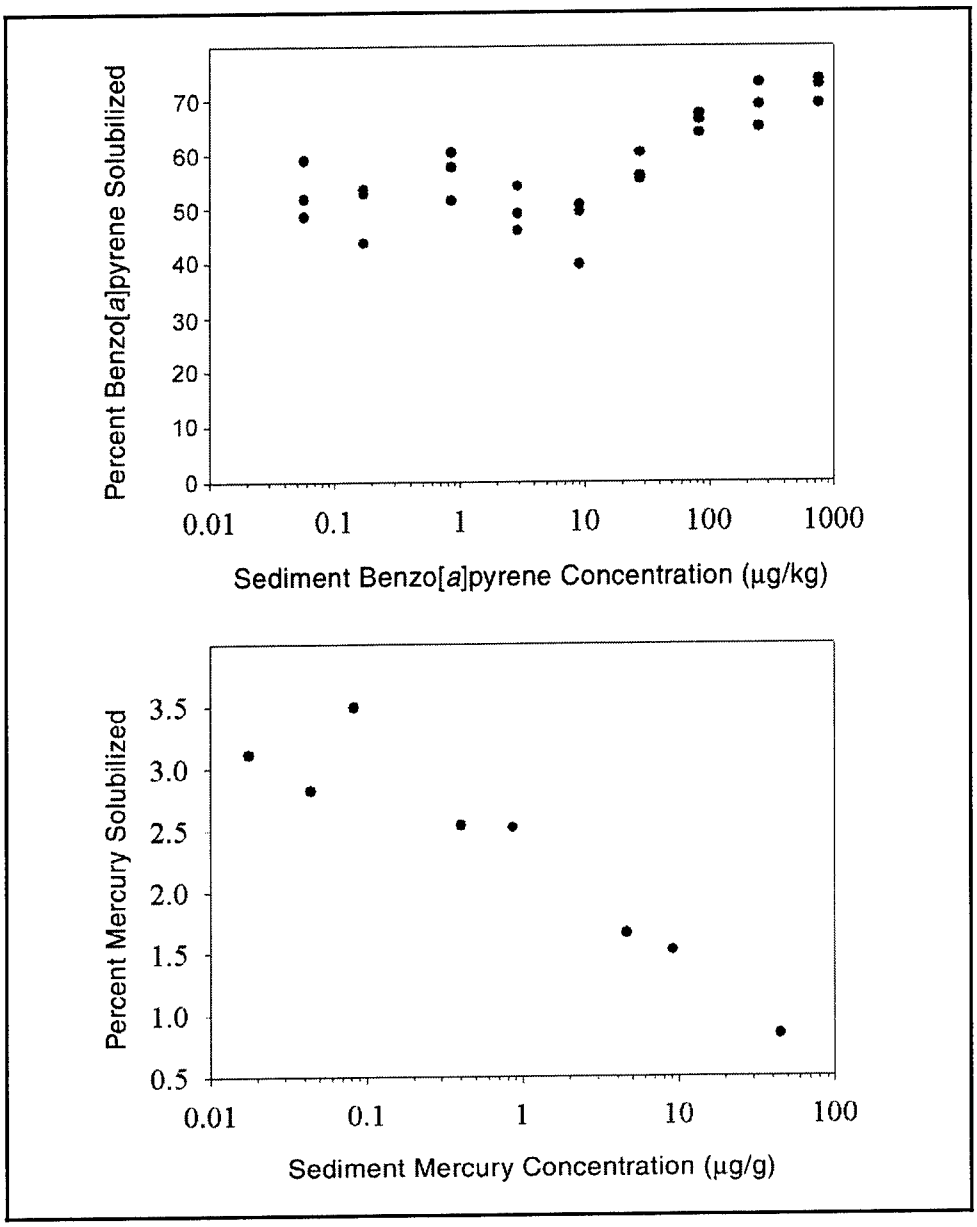

Figure 5. Efficiency of in vitro solubilization as a function of contaminant concentration in sediment. Data shown for sediments spiked with benzo[*a*]pyrene and mercuric chloride (Weston, unpubl)

Weston, and Bock 2001), and thus the proportion of contaminant solubilized may decrease as these ligands become saturated.

Existing data are inadequate to predict the effect of any given shift in contaminant concentration on digestive fluid solubilization or any other measure of bioavailability, but there appears to be a complex interplay between the availability of binding sites within the sediment matrix and fluid phase (Chen and Mayer 1999). Bioavailability is therefore not only a characteristic of a given contaminant in a given sediment, but a function of contaminant concentration within that sediment.

Adsorption

Contaminants from the solid phase solubilized by dissolved or colloidal (micellar) components of gut fluid might be prone to subsequent readsorption. From a biological perspective, it makes sense that animals should have evolved digestive agents that are not susceptible to adsorption by the sediment; they should otherwise suffer a loss of organic matter that counteracts the gain of digestive solubilization of nutritional organic matter. Indeed, experiments have shown that most enzyme activities and surfactant concentrations in gut fluid remain relatively constant upon exposure to physiologically normal levels of sediment (Mayer et al. 2001).

In the presence of contaminated sediments, however, at least two factors are introduced. First, there is the possibility that contaminated sediments will be more adsorptive of digestive agents than are uncontaminated sediments. Some indication of this possibility was found for surfactants apparently adsorbing onto oil-rich sediments by Voparil and Mayer (2000); this reaction seems logical due to uptake of the hydrophobic tails of surfactant molecules into oil-rich lipid phases. A second possibility is a change in solution-phase behavior of the solubilizing agents in seawater upon their association with contaminant materials. Some indication of this behavior has been found for certain metals (especially cadmium and mercury, so far), which show an initial dissolution from sediment followed by readsorption (Chen and Mayer 1999; Lawrence et al. 1999). This behavior might result from destabilization of protein ligands by the metals, resulting in greater adsorbability of the protein.

Comparison With Other Measures of Bioavailability

Absorption and assimilation efficiencies

Bioavailability of sediment-associated contaminants has traditionally been measured by absorption/assimilation efficiencies, uptake clearance rates, or by measures of steady-state bioaccumulation (Lee 1991). Of these, the most direct parallel to digestive fluid solubilization is the absorption and/or assimilation efficiency. Digestive fluid extraction is intended to provide an in vitro measure of the amount of contaminant that could be solubilized during gut passage in a deposit feeder, and thus be made available for digestive uptake. The approach does not explicitly predict that solubilized substances will be taken up across the gut wall (absorption) or incorporated into tissue (assimilation; sensu Penry 1998). However, if the approach is to have predictive value for risk assessment purposes, it is critical that solubilization rather than absorption be the process limiting uptake and that all or most of the solubilized contaminant be subsequently absorbed.

In those instances when absorption is the limiting factor, digestive fluid solubilization results can be highly misleading if used to predict bioavailability or bioaccumulation. For example, digestive fluid solubilization was shown to be a good predictor of bioaccumulation for many trace metals by the bivalve, *Macoma nasuta*, but predictions of chromium bioavailability were not reflected in the bivalve (Weston and Maruya 2002). This was believed to be due to the fact that Cr^{+3} is poorly absorbed from the gut by most organisms, and while the substance was solubilized in the *Macoma* gut, it was not absorbed.

For other contaminants for which absorption is not so clearly constrained, available information suggests there is a relationship between solubilization efficiency and absorption/assimilation efficiency (Figure 6), though further study is warranted. The best agreement between solubilization and assimilation has come from work involving exposure of two polychaete species, *Nereis succinea* and *Pectinaria gouldii*, to the chlorinated organic compounds, hexachlorobenzene (HCB) and tetrachlorobiphenyl (TCBP) (Ahrens et al. 2001). In an in vitro extraction, *N. succinea* gut fluid desorbed 72 and 79 percent of HCB and TCBP, respectively, while intact animals of the same species fed the same sediment assimilated 73 percent of both compounds. In vitro desorption and in vivo assimilation of HCB were both 37 percent using *P. gouldii* and its gut fluid.

Weston and Mayer (1998b) compared digestive fluid extraction to absorption efficiency of benzo[*a*]pyrene and phenanthrene by the polychaete *A. brasiliensis*. Absorption efficiency was determined by direct measurement of contaminant concentration in gut contents along the length of the digestive tract. In vivo absorption efficiencies for benzo[*a*]pyrene for three sediments ranged from 27 to 35 percent; in vitro solubilization from the same sediments using *A. brasiliensis* gut fluid ranged from 25 to 52 percent. Results for phenanthrene were similar with absorption efficiencies of 12 to 50 percent and in vitro solubilization of 22 to 49 percent. These results suggested that solubilization rather than absorption was the process limiting uptake, and that absorption of solubilized phenanthrene and benzo[*a*]pyrene approached 100 percent.

The only other data set available with which to compare solubilization and absorption is a study (Weston, unpub.) in which solubilization was measured with *A. brasiliensis* gut fluid and absorption efficiency was measured using the ^{14}C:^{51}Cr dual tracer technique (Klump et al. 1987) with the confamilial polychaete *Abarenicola pacifica*. In these experiments, solubilization of five PAHs (28 to 47 percent) by *A. brasiliensis* fluid consistently overestimated absorption efficiency in *A. pacifica* (4 to 24 percent, Figure 6), suggesting incomplete digestive absorption of solubilized contaminants. However, this disparity could be due to use of the dual tracer technique, which could have resulted in an underestimate of actual absorption efficiency. The approach contains more untested assumptions and potential artifacts than the more direct methods of the two previous studies. Alternatively, the use of different species for measurements of digestive fluid extraction and absorption efficiency measurements may have played

a role, although available data would discount this influence as *A. pacifica* gut fluid is a stronger extractant than that of *A. brasiliensis* (Mayer, Weston, and Bock 2001).

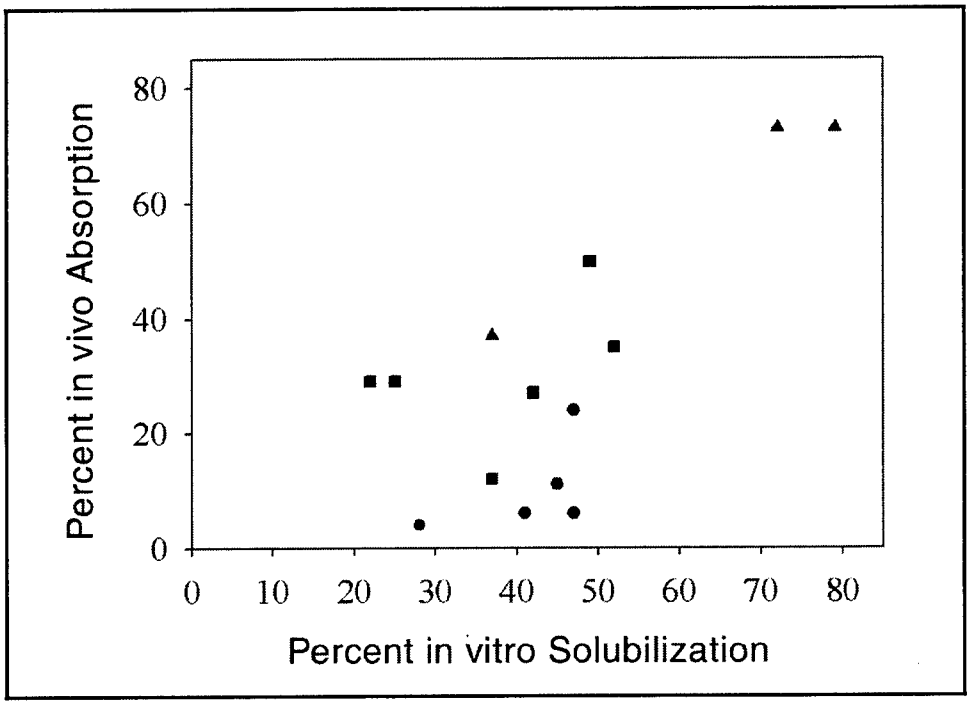

Figure 6. A comparison of the proportion of contaminant solubilized in an in vitro extraction with the proportion absorbed in vivo during gut passage. Dotted line indicates hypothetical perfect agreement. Data from the following studies.

Squares - Weston and Mayer (1998b). Contaminants studied were phenanthrene and benzo[*a*]pyrene. Absorption efficiency determined by direct measurement of contaminant concentration at points along the gut. In vitro and in vivo studies both using *A. brasiliensis*.

Circles - Weston (unpub.). Contaminants studied were five PAHs. Absorption efficiency determined by ^{14}C:^{51}Cr dual label technique. In vitro extractions done with *A. brasiliensis* gut fluid; in vivo adsorption measured in *Abarenicola pacifica*.

Triangles - Ahrens et al. (2001). Contaminants studied were hexachlorobenzene and tetrachlorobiphenyl. Absorption efficiency determined by pulse-chase methods. In vitro and in vivo studies done with same species, either *Nereis succinea* or *Pectinaria gouldii*.

Relationship to bioaccumulation

Bioaccumulation, to the extent that it reflects contaminant bioavailability, should be predictable by in vitro solubilization. Bioaccumulation is, however, subject to other confounding factors, most notably biotransformation. If a contaminant is rapidly biotransformed, digestive fluid extraction may predict high bioavailability and the compound may indeed be taken up

quite readily, but biotransformation of the compound could result in little or none of the substance being measured in the tissues. Bioaccumulation at steady-state should be a correlate of in vitro solubilization for substances that are not biotransformed (e.g., DDE), for taxa having poor biotransformation capabilities (e.g., bivalves), or when values for biotransformation can be empirically estimated.

Digestive fluid extraction, in all cases using gut fluid of *A. brasiliensis* or *A. marina*, has shown a relationship with bioaccumulation in a wide variety of taxa. In a study of sediments from San Francisco Bay (Weston and Maruya 2002), digestive fluid was unable to extract appreciable amounts of arsenic, copper, mercury, nickel, zinc, and low molecular weight PAHs from what were, for some of these substances, highly contaminated sediments. Similarly, the bivalve *Macoma nasuta*, when held in the sediments for 28 days, failed to bioaccumulate these same contaminants. Digestive fluid extractions found only cadmium, lead, chromium, PCB, and higher molecular weight PAH (HPAH) to be bioavailable from these sediments, and these were the same contaminants bioaccumulated by *M. nasuta* (with the exception of chromium for reasons discussed earlier). For cadmium and lead, sediments which produced a high contaminant concentration in the digestive fluid extract also yielded high bioaccumulation in the clam (Figure 7). For HPAH the relationship was marginal; for PCB there was no significant relationship between in vitro extractability and bioaccumulation. Less extensive tests have been conducted on the ability of arenicolid digestive fluid to predict bioaccumulation by the polychaete *A. pacifica* (Weston and Mayer 1998b) and the amphipod *L. plumulosa* (Lawrence et al. 1999). Both tests suggested a correlation between in vitro solubilization and bioaccumulation in these species, though the number of sediments tested was too few to draw statistically rigorous conclusions.

Uptake clearance rates

A third frequently used measure of bioavailability of sediment-associated contaminants is the uptake clearance rate, k_s, which is the rate of increase in body burden during the early stages of exposure prior to significant elimination, normalized to the sediment contaminant concentration (Landrum 1989). In a comparative study of six sediments, the proportion of benzo[a]pyrene solubilized by *A. brasiliensis* digestive fluid was significantly correlated to the uptake clearance rate of PAHs from the same sediments by *A. pacifica* (Weston and Mayer 1998b). A correlation between uptake clearance rate and in vitro solubilization would be expected to the extent they are both measures of bioavailability; however, uptake clearance rate is also a function of the feeding rate of the organism (Penry and Weston 1998) and may not correlate with in vitro solubilization if, for example, feeding rates vary substantially among the test sediments.

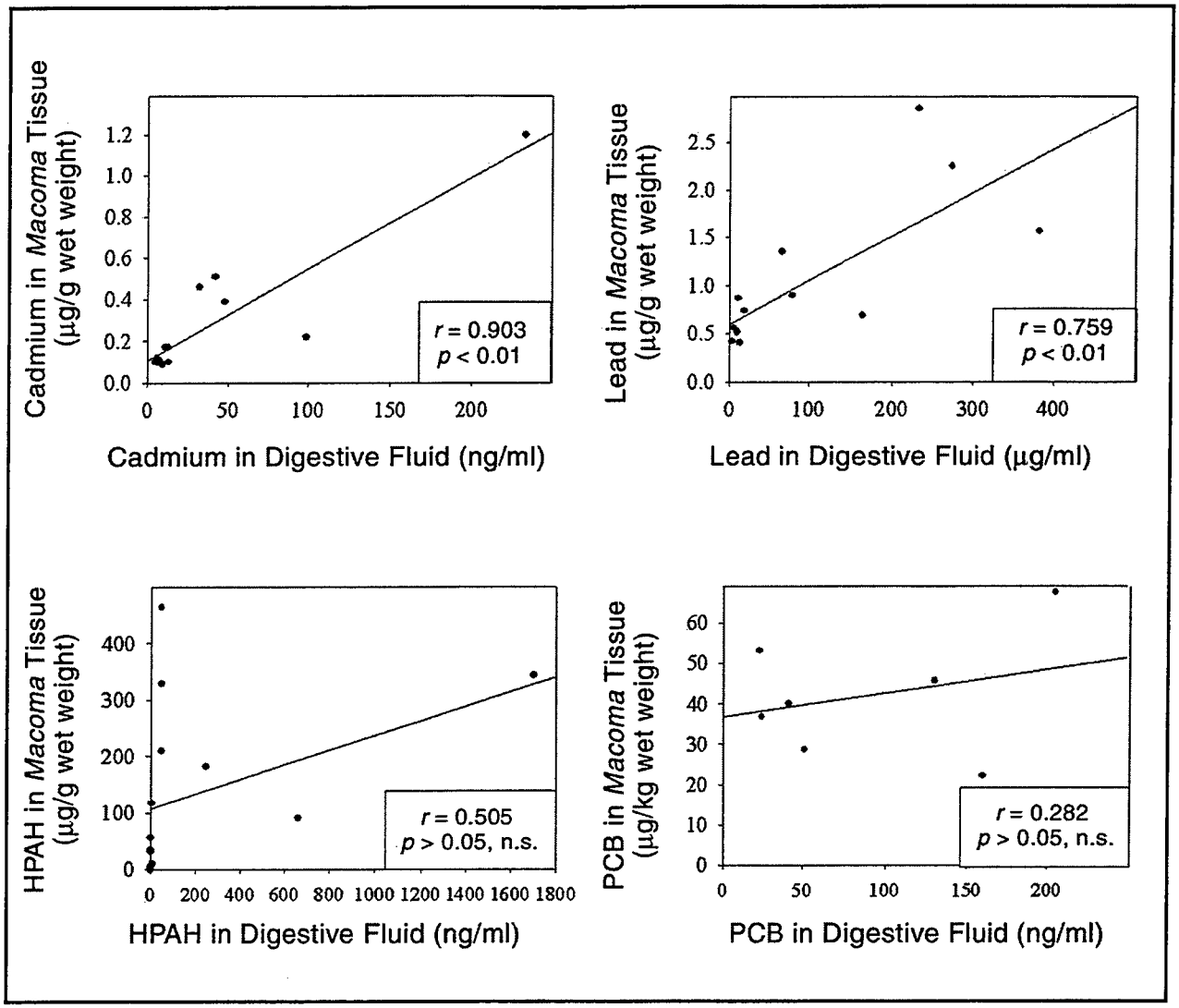

Figure 7. Concentration of contaminants in *A. brasiliensis* digestive fluid following a 3-hr in vitro extraction of 12 sediments in comparison to the concentrations attained in *Macoma nasuta* after 28 days exposure to same sediments. Statistic is Pearson product-moment correlation (from Weston and Maruya 2002)

3 Research Needs

Absorption of Solubilized Constituents

As previously discussed, digestive solubilization has value as a tool to assess bioavailability if the solubilization step of bioaccumulation rather than the intestinal absorptive step constrains uptake. Available data suggest this is the case (see paragraph "Comparison With Other Measures of Bioavailability"), with the possible exception of chromium. However, further study would be desirable to determine the fate of contaminants solubilized in the anterior portions of the gut and if they are in fact entirely absorbed in the more posterior portions.

Extension to Other Pollutant Types

In vitro digestive fluid extraction has only been studied extensively for PAH and copper, and there are some limited data for zinc, lead, cadmium, mercury, hexachlorobenzene, and tetrachlorobiphenyl. Conceptually, the technique should be applicable to any contaminant for which ingestion and digestion is a significant route of uptake. Thus, its applicability may extend to all or most trace metals, many organometallic compounds (e.g., tributyltin, methylmercury), a wide variety of chlorinated organic compounds, and hydrophobic pesticides (e.g., the pyrethroids, DDT). Validation of the technique as a predictor of bioavailability for some pesticides and PCBs is ongoing, and additional research with other contaminant classes is needed.

Development of Biomimetic Extractant

One of the principal constraints to broad utilization of in vitro digestive fluid extraction is the limited quantity of gut fluid that can be obtained from deposit-feeding organisms. Therefore, a near-term goal is the development of an artificial fluid that mimics the natural constituents of digestive fluid but can be prepared with commercially available substances.

tive fluid but can be prepared with commercially available substances. Considerable progress has been made in understanding how and to what extent digestive fluid solubilizes contaminants. Application of this understanding to the development of an artificial cocktail is a realistic goal in the near term, but there are many issues needing attention. Summarizing, the major needs are as follows:

a. Use of commercially available proteins and surfactants to mimic those in gut fluid must address potential adsorption of solubilizing agents onto sediment. While the in vivo versions of these compounds have evolved to avoid this adsorption, the extent of adsorption of commercial proteins and surfactants must be studied in order to avoid sediment adsorption of the commercial versions either prior to or after solubilization of contaminants. Such an adsorption would cause serious underestimates in apparent bioavailability.

b. Little attention has been given to the redox environment of gut fluid solubilization. Gut fluid contains redox-sensitive materials such as iron and manganese, and sedimentary matrices are also redox sensitive. The interactions of these two reactants, and the consequent extent of contaminant solubilization, are thus potentially subject to the presence or absence of oxygen during the incubation.

c. There needs to be a better understanding of the chemical mechanisms of metal binding by ligands in gut fluid. Only copper has had the relevant mechanisms determined to date. While a similar level of chemical determination for all contaminant metals is unrealistic in the near future, a clearer connection to ligand groups on protein molecules is at least called for. Further narrowing to certain types of ligand groups (e.g., thiol, imidazole) would be important for protein selection for artificial gut fluid cocktails, as various proteins are enriched or depaupurate in these groups.

d. Most work on solubilization of spiked contaminants has dealt with one contaminant at a time. However, most of the solubilization mechanisms have strong potential to interact with more than one contaminant. There is, therefore, strong possibility of positive and negative interactions that affect the spectrum of contaminants solubilized from contaminant mixtures. Complex mixtures are the rule rather than the exception in harbor sediments, so that systematic study of these interactions is indicated.

e. Finally, assembly of the mix of proteins and surfactants needs to be addressed. While most preliminary studies will use single extractant solutions to allow interpretation of experiments, cocktails will inevitably consist of complex mixtures of extractants. Interactions among these extractants and with sediment matrices will need attention.

4 Application to USEPA/USACE Tiered Evaluation Approach

Inherent within current guidelines for dredged material quality assessment (USEPA/USACE 1998) is an optimization of effort to ensure generation of an adequate, and not excessive, amount of data sufficient to allow a comprehensive assessment. This approach ensures the minimization of both time and resources while attaining an accurate factual determination of the quality of dredged material. Central to these considerations is the tiered approach used in the assessment, such that investigations only pass on to more complex and expensive tiers when previous tiers, utilizing simpler and cheaper methodologies, have shown to provide insufficient information.

The use of simple screening tools in the tiered evaluation is helpful in this regard, because these tools aid optimization of both time and resources. The TBP calculation – a screening tool currently employed in Tier II – utilizes the sediment contaminant concentrations and various derived constants to calculate a theoretical body burden. This approach is a rapid, predictive tool for contaminant bioaccumulation. Indications of no potential for significant bioaccumulation using TBP remove the necessity for further, expensive bioaccumulation tests in Tier III unless contaminants other than nonpolar contaminants are of potential concern. However, the major constraint of TBP is its limitation to nonpolar contaminants. Therefore, determination of benthic bioaccumulation of metals currently can only be assessed using costly and time-consuming bioaccumulation studies within Tier III. Clearly, the development of a more universal and rapid bioaccumulation-screening tool, which could be employed for metals, would reduce costs significantly. The use of gut fluid extractions may represent such a tool.

While the method has benefited from significant development in recent years, further research is necessary before this approach can be considered as a standard screening tool. In particular, development of a synthetic biomimetic gut fluid (BGF), with contaminant binding and sorption properties akin to those of natural gut fluids over a wide range of sediments and contaminants, is needed. Currently, development of this approach has

utilized gut fluids extracted from natural populations of a few deposit-feeding species. These studies have observed some degree of variability among batches of gut fluids collected in this manner. Clearly the widespread and routine application of this approach for dredged material quality assessment requires a more reliable and standardized source of solubilization fluid. Although these studies have achieved some success in the development of a standardized test method using the naturally derived gut fluid, there is a need for development of a definitive protocol applicable to the use of the BGF extraction to a range of sediments and contaminants.

Theoretically, should BGF extraction be accepted as a viable screening tool for bioaccumulation of metals and nonpolar organics, the method could be applied as part of Tier II alongside the current TBP approach. In this manner, BGF extractions of dredged material and reference material might be compared for a range of relevant contaminants. Cases where BGF concentrations following extraction of dredged material do not exceed those of the reference site would predict no significant bioaccumulation in the exposed biota, and would require no experimental bioaccumulation studies. Conversely, BGF-extracted concentrations in dredged material exceeding those of the reference sediment would necessitate further evaluation of bioaccumulation potential under Tier III.

In addition, should the BGF-extracted concentrations be found to be predictive of total body bioaccumulation, the dredged material BGF-extracted concentrations might be used to calculate theoretical body burdens. Such derived body burdens might then be compared to Food and Drug Administration Action or Tolerance Levels (USEPA and USACE 1998) and to critical body residue databases to assess potential for human health and environmental impact of the predicted bioaccumulation.

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Sediment Extraction Using Deposit-Feeder Gut Fluids: A Potential Rapid Tool for Assessing Bioaccumulation Potential of Sediment-Associated Contaminants

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14. ABSTRACT

Extraction of contaminated sediments using gut fluids from invertebrates has been used to estimate the biologically available fraction of contaminants. This report discusses how the technique might be used to estimate contaminant bioaccumulation and hence has potential as a universal bioaccumulation screening tool for use in the testing of dredged materials as part of the Inland Testing Manual. The report details the current status of the field and both the methods and theory of gut fluid extraction. The report then discusses significant factors that have been identified as significant influences upon gut fluid extraction efficiency and compares the method with other measurements of bioavailability. Finally, the report identifies current research needs and discusses how the technique may be applicable to the needs of U.S. Environmental Protection Agency and U.S. Army Corps of Engineers for a universal screening tool for sediment-associated contaminants in dredged material.

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