

Kinetic modeling of Ag, Cd and Co bioaccumulation in the clam *Macoma balthica*: quantifying dietary and dissolved sources

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ABSTRACT: A biokinetic model was used to better understand Ag, Cd and Co concentrations in a population of the clam *Macoma balthica* in San Francisco Bay. Model parameters included laboratory-derived uptake- and loss-rate constants from food and water and field measurements of metal concentrations in food, overlying water and oxidized pore water. Parameters used in modeling were taken from recent studies of metal influx from dissolved sources and metal assimilation from ingested sediment, and from laboratory experiments and field measurements in this study. Assimilation efficiencies from surface sediments ranged from 12 to 22% for Ag, 6 to 13% for Cd and 8 to 20% for Co. Assimilation efficiencies from phytoplankton were higher than metal assimilation efficiencies from sediment, ranging from 36 to 42% for Ag, 47 to 55% for Cd and 27 to 33% for Co. Influx of dissolved metals from overlying water increased with increasing ambient concentration, with uptake-rate constants for Ag ($0.34 \text{ l g}^{-1} \text{ d}^{-1}$) about an order of magnitude higher than for Cd and Co. Influx-rate constants for Ag and Cd from oxidized pore water were comparable to overlying water-rate constants, whereas the rate constant for Co influx from oxidized pore water was 3 times lower than that from overlying water. Efflux rates of all metals out of the clams ranged from 1 to 3% d^{-1} . To estimate the potentially bioavailable fraction of particle-bound metals, assumed to be the metal bound to particle surfaces, mean metal concentrations in shale were subtracted from metal concentrations in total sediment digestions. Metal accumulation was modeled for clams that were assumed to be surface deposit-feeding and those that were filter-feeding. By adding uptake from food (surface sediment or phytoplankton) and from dissolved sources (oxidized pore water or overlying water), the model-predicted ranges of concentrations of Ag, Cd and Co in deposit-feeding clams are shown to be directly comparable to tissue concentrations in field-collected clams from San Francisco Bay. Thus, it appears that the parameters experimentally derived for *M. balthica* are applicable to field conditions and that the model can account for the major processes governing metal concentrations in these clams. Further, through modeling, ingested sediment was shown to be a major source for Ag and Cd under all realistic environmental conditions, but Co accumulation was principally from the dissolved phase.

KEY WORDS: Metals · Bioaccumulation · Clams · Deposit-feeders · Modeling

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INTRODUCTION

Benthic animals are exposed to high concentrations of contaminants in industrialized estuaries. To under-

stand fully the toxic effect of contaminant metals on aquatic animals, one must know the concentration of metal in an organism's tissue. Much research and modeling has focused on predicting the bioavailability or the dose of metals to benthic organisms in relation to metal concentrations within the environment, yet no 1 method or principle has proved universal (Luoma & Bryan 1982, Bryan & Langston 1992, Ankley et al.

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1996, Hare & Tessier 1996, Luoma & Fisher 1997, Chen & Mayer 1999, US EPA 2000). Within the same habitat or experimental microcosm, metal concentrations in organisms can be highly variable with regard to taxa, mobility and feeding behavior (Maloney 1996, Kaag et al. 1997, Warren et al. 1998, Lee et al. 2000), suggesting the need to include biological parameters in a bioavailability model. Use of a bioenergetic-based toxicokinetic model has shown promise in predicting organic and inorganic contaminant concentrations in aquatic animals (Boese et al. 1990, Landrum et al. 1992, Luoma et al. 1992, Wang et al. 1996, Roditi et al. 2000). Mechanistically, the model considers contaminant bioaccumulation as a result of the balance between influx rate and efflux rate. Influx and efflux rates are influenced by biological, geochemical and physicochemical factors (Wang & Fisher 1997a) and all must be considered to understand metal bioavailability fully (Luoma & Fisher 1997). Modeling efforts to date suggest that when the biological nature and site-specific geochemical characteristics of the system are included in a model and the uptake terms are expressed by adjustable rate constants, measured metal concentrations in bivalves and copepods are generally within the same range as model-predicted concentrations (Luoma et al. 1992, Wang et al. 1996, Fisher et al. 2000, Roditi et al. 2000). Although the usefulness of the model has not been thoroughly exploited, these initial results suggest that the model could not only prove useful in predicting toxicity but may also be used in an iterative sense to further clarify mechanisms that control metal accumulation.

Metal influx into benthic organisms can result from both ingestion of food particles and from transport across tissues from dissolved sources. In the model used for this study, particle-bound and dissolved metal sources were considered separately so that each term could be manipulated to fit non-steady-state environmental conditions (Luoma & Fisher 1997, Wang & Fisher 1997a). The contributions of the 2 sources were considered to be additive. This separation and flexibility in the model is attractive for applicability to benthic invertebrates, for which there is still an inadequate understanding of the exposure an organism may experience living within a highly dynamic sedimentary environment.

Influx of metals from food is not simple to quantify. In surface sediments, most organisms live at sites of intense concentration gradients where heterogeneous distributions of trace metals exist in the solid phase. Metal concentrations in the food may vary greatly with the food source, which for some benthic invertebrates include the subsurface and surface sediments, suspended particles in overlying water, live prey, or a combination of these sources. Additionally, once in-

gested, metals are assimilated into soft tissues differently, depending in part on the metal, the particle composition, the tissue, and the intensity of digestion (Decho & Luoma 1994, Reinfelder & Fisher 1994, Wang & Fisher 1996, Lee & Luoma 1998, Griscom et al. 2000).

Influx of metals from the dissolved phase into benthic organisms is also complicated. Metal concentration gradients in the solid phase are magnified in pore waters. In the upper 1 cm of sediment, high dissolved metal concentrations exist from the migration and oxidation of metals remobilized from suboxic horizons and from the release of scavenged metals by newly deposited organic debris (Aller 1978, Tessier et al. 1994). In this thin surface microzone, dissolved metal concentrations may be substantially greater than concentrations deeper in the sediment or as much as 10× to 100× higher than that measured in the overlying water (Campbell et al. 1988, Zhang et al. 1995, Rivera-Duarte & Flegal 1997a,b) and it is within this zone that benthic activity is greatest. In addition, dissolved metal exposures within the burrow of a benthic organism are obtained from a variable mixture of surrounding pore water and overlying water, and the relative influence of any 1 dissolved source may change with pumping or ventilation activity (Griscom & Fisher 2002), the extent of burrowing, population density, and tidal cycle (Aller 1978).

Influx of metals from food and from water into an organism is balanced by the efflux (or loss) of metals out of the animal. In general, the efflux rate of metal from tissues with the slowest turnover rate exerts the greatest control on the steady-state concentration of metal in the organism. This assumption is applicable to most situations in which the largest proportion of tissue-bound metal is within this slowly exchanging compartment. However, using a single efflux-rate term may not be appropriate in all situations, and is discussed in detail below.

Given these issues, model parameters were quantified for *Macoma balthica*, a facultative deposit-feeder which can feed on surface sediment or filter particles from the overlying water column, depending on food availability (Brafeld & Newell 1961, Olafsson 1986). A range of environmental conditions was considered and combined with field measurements of metal concentrations to predict concentrations of metal in these clams. In general, bulk sediment characteristics often are not directly applicable to understanding the micro-environments in which indicator organisms live (Luoma 1996, Warren et al. 1998). With more focus on understanding the interplay between animal behavior and metal exposures, it may be possible to constrain more tightly the model parameters for specific populations of animals living in specific environmental conditions.

This study focused on a population of *Macoma balthica* in south San Francisco Bay at a site that has been continuously monitored for 30 yr (Hornberger et al. 2000). In experiments using clams collected from this location, many of the parameters in the model have been determined for Ag, Cd, and Co over a range of environmental conditions (Reinfelder et al. 1997, Lee & Luoma 1998, Lee et al. 1998, Griscom et al. 2000, Griscom & Fisher 2002). These metals have varying chemical properties which lead to different geochemical and biological behaviors in the environment (Nieboer & Richardson 1980) and are sometimes of concern as contaminants in various coastal regions, including San Francisco Bay. Further, at or near this location reliable field measurements of metal concentrations in surface sediments, overlying water, phytoplankton, and pore water metal concentrations have been made, although no measurements have been made of dissolved metal concentrations within the living burrow of *M. balthica*.

We present here the results of 2 series of experiments in which the assimilation and retention of Ag, Cd, and Co in the clam *Macoma balthica* were evaluated experimentally following dietary exposure to sediments and diatom food. We combine these new data with some recent literature values describing metal accumulation in this clam and use this compiled information to model the bioaccumulation of metals in *M. balthica*. We field-tested our model predictions in San Francisco Bay, for which we present metal concentrations in sediment, water, and a population of *M. balthica*. To examine how feeding mode could affect predicted metal accumulation, metal concentrations were calculated separately for filter-feeding and surface deposit-feeding clams using unique parameters for each (see equations below). An anticipated outcome of separately determining filter-feeding and deposit-feeding metal accumulation was insight into the extent to which clams in San Francisco Bay spend in one or the other feeding mode.

BIOKINETIC MODEL

The biokinetic model used in this study was initially developed to be generic in its mechanistic approach (Landrum et al. 1992, Thomann et al. 1995), and further development has shown the model to be effective in quantifying metal concentrations for specific aquatic organisms, as noted above. The kinetic model is a multiple pathway model in that it separately quantifies influx rates of metal from food, influx rates from water, and efflux rates of metal out of the organism. The model is flexible as the parameters can be adjusted to account for the effects of non-equilibrium conditions in the environ-

ment, such as variable water characteristics (e.g. salinity effects on metal speciation, complexation, and partitioning), changes in food concentrations or types (affecting assimilation efficiency and feeding behavior), and reproductive status (changing the bulk tissue metal concentrations and lipid content). All these factors may influence metal bioavailability, depending upon the metal or the organism in question.

Ignoring the effects of reproduction and growth, which for adult bivalves are negligible (Wang & Fisher 1997a,b), and assuming that metal is obtained from water and ingested material, the simplified equation used to describe soft tissue metal bioaccumulation in this study is:

$$dC/dt = [I_f + I_w] - (C_t \times k_e) \quad (1)$$

$$C_{ss} = \frac{I_f + I_w}{k_e} \quad (2)$$

where C = the metal concentration in the clam ($\mu\text{g metal g}^{-1}$ clam dry wt), C_{ss} = the metal concentration in the clam at steady state ($\mu\text{g metal g}^{-1}$ clam dry wt), t = time (d), I_f = the influx rate from food ($\mu\text{g metal g}^{-1}$ clam dry wt d^{-1}), I_w = the influx rate from water ($\mu\text{g metal g}^{-1}$ clam dry wt d^{-1}), and k_e = the rate constant of loss (from the slowest compartment) (d^{-1}).

The influx rates are further described by:

$$I_f = C_f \times \text{IR} \times \text{AE} \quad (3)$$

$$I_w = k_u \times [C_w]^b \quad (4)$$

where C_f = metal concentration in food ($\mu\text{g metal g}^{-1}$ food dry wt), IR = feeding rate (g food dry wt g^{-1} clam dry wt), AE = assimilation efficiency of ingested metal, C_w = metal concentration in dissolved phase ($\mu\text{g l}^{-1}$), k_u = uptake rate constant from dissolved phase ($\text{l g}^{-1} \text{d}^{-1}$), equal to the metal absorption efficiency times the filtration rate, and b = power of the relationship, typically = 1 ± 0.1 , unless metal uptake is biologically regulated (Wang et al. 1996).

Eq. (2) was modified to quantify separately predicted metal concentrations in filter-feeding or deposit-feeding clams. For these 2 different feeding modes there are correspondingly unique uptake rates, assimilation efficiencies and metal concentrations in food and water. For filter-feeding clams, metals are assumed to originate from food comprised of suspended particulate matter (SPM) and from dissolved metal in overlying water. The full equation used to predict concentrations in filter-feeding clams is:

$$C_{ss(\text{FF})} = \frac{[C_{(\text{SPM})} \times \text{IR} \times \text{AE}_{(\text{SPM})}] + [C_{w(\text{sw})} \times k_{u(\text{sw})}]}{k_e} \quad (5)$$

where $C_{ss(\text{FF})}$ = steady state metal concentration of filter-feeding clams ($\mu\text{g g}^{-1}$ clam dry wt), $C_{(\text{SPM})}$ = metal

concentration of suspended particulate matter ($\mu\text{g g}^{-1}$), $AE_{(\text{SPM})}$ = assimilation efficiency of metal from phytoplankton, $C_{w(\text{sw})}$ = metal concentration in overlying water ($\mu\text{g l}^{-1}$), and $k_{u(\text{sw})}$ = uptake rate constant of dissolved metal from overlying water ($\text{l g}^{-1} \text{d}^{-1}$).

The equation to describe steady-state metal concentrations in clams feeding solely on surface deposits is:

$$C_{\text{ss}(\text{SDF})} = \frac{[C_{(\text{sed})} \times \text{IR} \times AE_{(\text{sed})}] + [C_{w(\text{opw})} \times k_{u(\text{opw})}]}{k_e} \quad (6)$$

where $C_{\text{ss}(\text{SDF})}$ = steady-state metal concentration of deposit-feeding clams ($\mu\text{g g}^{-1}$), $C_{(\text{sed})}$ = metal concentration of surface sediment ($\mu\text{g g}^{-1}$), $AE_{(\text{sed})}$ = metal assimilation efficiency from sediment, $C_{w(\text{opw})}$ = metal concentration in oxidized pore water ($\mu\text{g l}^{-1}$), and $k_{u(\text{opw})}$ = uptake-rate constant of dissolved metal from oxidized pore water ($\text{l g}^{-1} \text{d}^{-1}$).

Assimilation efficiency. Assimilation efficiency is defined as the percent of ingested particulate metal that is physiologically taken up within the tissue of an animal during the course of a complete digestive cycle. The methods to measure assimilation efficiencies originated out of an interest in quantifying the amount of nutrition gained from food by certain marine invertebrates (Calow & Fletcher 1972, Kofoed 1975, Lopez & Cheng 1983). More recently, assimilation efficiencies have been determined to help quantify the bioavailability of ingested contaminants from diverse food sources in marine bivalves (Luoma et al. 1992, Gagnon & Fisher 1997, Wang & Fisher 1997a, Griscom et al. 2000, Fan & Wang 2001).

There are several methods that have been used to determine assimilation efficiencies, each with certain advantages and limitations. One method uses a mass balance approach in which labeled food is fed to an animal in a short pulse, after which the animals are fed non-labeled food. Feces are collected throughout depuration, and the original egested label is quantitatively analyzed. The amount of label recovered in the feces, after complete gut evacuation of the original pulse, is subtracted from the initial pulse of material ingested. The missing label is then considered assimilated. The limitations of this method include the potential loss of label by desorption from the feces into the overlying water. Label may also be excreted by the animal by other means (e.g. pseudofeces) so that the assimilation efficiency will be overestimated. A similar method using food labeled with gamma-emitting radioisotopes may be employed to measure loss of the pulse from the whole animal by non-destructive means (a whole animal may be gamma-counted non-destructively and returned to an aquarium to continue depurating). If loss is measured for a significant period (2 wk for clams and mussels), the physiological turnover portions of the radiotracer retention curve may be

traced back to the y-intercept and assumed to represent the amount that was physiologically assimilated during the initial passage of the radiolabeled food through the animal's gut. Comparisons of these approaches have shown that results can differ, especially for elements readily lost from feces, but in most cases differences are less than 2-fold, and careful experimental design can be used to avoid even this (Decho & Luoma 1994, Lee & Luoma 1998).

A third method for determining assimilation efficiencies has been derived from the 2 methods described above, whereby loss of gamma-emitting radiotracers is followed in individuals, and after the guts are assumed to be emptied of the initial pulse of radiolabeled food, the remaining radioisotope is assumed to be assimilated (approximately 3 to 4 d for the clam *Macoma balthica*; Decho & Luoma 1991, Luoma et al. 1992). Assimilation efficiency is simply measured as the radioactivity in the soft tissues at 3 to 4 d divided by the total radioactivity ingested during the pulse-feeding. Comparison of this short-cut method with the longer 2 wk depuration method has produced comparable results (S. B. Griscom & N. S. Fisher unpubl. data).

These radiotracer techniques were first developed for labeling phytoplankton food which could be uniformly radiolabeled with metals (Fisher & Teyssié 1986, Reinfelder & Fisher 1991). An important limitation of this approach when applied to understanding metal bioavailability from sediments lies in the fact that the adsorbed radiolabeled metal does not necessarily distribute within the sediment particle. It is likely that metals trapped within the lattice of fine-grained particles do not exchange with the radiotracer within the time frame of the radiolabeling period. Further, radiolabeling surface coatings requires weeks to months to ensure an even distribution of these coatings of fine-grained sediment. Experiments that addressed labeling time showed that the exposure time of sediment to radiotracer correlated inversely with metal assimilation efficiencies in marine bivalves; the extent varied among metals and differed between organisms (Wang et al. 1999, Griscom et al. 2000, Fan & Wang 2001).

Metal assimilation efficiencies vary considerably, depending upon the type of food particle, the specific metal, and the nature of the association between the particle and the metal (Fisher & Reinfelder 1995). In general, metals associated with living organic matter (phytoplankton, bacteria) are more bioavailable than from inorganic matter such as iron oxides, clay or humic matter (Decho & Luoma 1994, 1996, Gagnon & Fisher 1997, Griscom et al. 2000). Metals loosely sorbed to particle surfaces are more bioavailable than those metals trapped in precipitated mineral coatings (Griscom et al. 2000). The concentrations of metals associated with the cytosolic fraction within phyto-

plankton cells are generally related directly to metal assimilation efficiencies in marine animals, particularly those with short gut-residence times (Reinfelder & Fisher 1991, 1994).

Metal influx rates from dissolved phase (I_w). Metal influx rates and rate constants are derived by measuring the uptake of radioisotope during short-term exposure to radiolabeled overlying water (Wang et al. 1996). The exposure must be sufficiently short to ensure minimal efflux or recycling of metal from the animal back into the dissolved phase. Uptake-rate constants are quantified by measuring the metal uptake rate in animals exposed to several different concentrations. The uptake-rate constant is calculated from the relationship between metal influx rate and the dissolved metal concentration in overlying water, as expressed by Eq. (4).

Factors that affect metal uptake rates include various geochemical and biological conditions. Salinity, DOC concentrations and composition, pH and Eh may influence the speciation and complexation of metals and thereby affect the influx of dissolved metals into aquatic animals (Rashid 1985, Wright 1995, Wang et al. 1996, 1997, Wang & Fisher 1999, Guo et al. 2001). Various mechanisms of passive or facilitated cross-cell transport have been described (Simkiss & Taylor 1995) and experimental work has shown that uptake rates are dependent on the chemistry of the water and the metal. Additionally, the size of a bivalve (Wang & Fisher 1997b, Lee et al. 1998) and its filtration rate, or the rate that it moves water across a gill system or pumps into a burrow, also affect uptake rate (Lee et al. 1998).

Experiments that measure uptake rates from pore-water exposures are not as straightforward as those used to quantify uptake from overlying water. A large but poorly quantified amount of oxidized pore water is entrained with surface sediment as clams feed on deposits. Possibly 7 to 10 times more oxidized pore water is entrained than the amount of food ingested (Bubnova 1972, Black 1980, Hummel 1985). If the surface-sediment feeding rate is $0.5 \text{ g sediment g}^{-1} \text{ dry wt tissue d}^{-1}$ and the porosity of the sediment is approximately 0.95, the amount of oxidized pore water concurrently siphoned into the mantle cavity would be around $50 \text{ ml oxidized pore water g}^{-1} \text{ dry wt tissue d}^{-1}$, a rate that is more than $20\times$ lower than the measured filtration rate when clams are filter-feeding ($1 \text{ to } 4 \text{ l g}^{-1} \text{ d}^{-1}$; Harvey & Luoma 1984, Hummel 1985). In addition, clams are exposed continuously to metals in their burrow water, which is mixed to some degree with overlying water. The relative contribution of overlying water pumped into the burrow by the clams to the total burrow water is unknown; therefore, the actual metal concentration to which an organism is exposed while living within this habitat is unknown. In this study,

therefore, we only consider uptake from overlying water and oxidized pore water.

The chemical speciation of dissolved metals and the relative amount of overlying water in the burrow are affected by dynamic processes and will vary on a time-scale of hours to days. Other studies that attempted to measure bioavailability of metals from pore water into benthic organisms encountered problems characterizing metal speciation and concentrations in their experiments (Pesch et al. 1995). Short-term experiments that used radiolabeled pore water to measure the influx of metal from burrow water *in situ* showed high variability (Griscom & Fisher 2002).

Efflux rates (k_e). Efflux rate constants are best derived following prolonged exposure of the clams to metals, since long exposures ensure that the metals distribute uniformly among all tissue compartments. This not only better simulates conditions of chronic contamination but is also more relevant for understanding the role of bivalves in trace-metal cycling (Cutshall 1974). Nevertheless, measured efflux rates of metals out of organisms using radioactive tracers assume that the compartment with the slowest efflux rate can be measured without long-term exposures to metal (months to years; Cutshall 1974, Fisher et al. 1996, Wang et al. 1996). For most metals, the rate constant of loss is not affected by exposure route (food or water), although some differences have been noted (Fisher et al. 1996, Wang & Fisher 1998, Roditi et al. 2000). Rate constants of metal loss from bivalves are similar among metals and, with few exceptions, typically range from 1 to $5\% \text{ d}^{-1}$ (Wang & Fisher 1997a, Lee et al. 1998, Roditi et al. 2000).

Under some circumstances, metal-rich granules and metal-specific binding proteins such as metallothioneins are produced in animals (George 1990). These may represent storage compartments with much slower exchanging pools of metal, each with its own specific turnover rate (Mason & Jenkins 1995). It is not known, however, if radiolabels exchange with such compartments during short exposures (Wallace et al. unpubl.). If a significant proportion of an animal's total body burden of metal is contained in a very slowly exchanging compartment, then it is possible that the rate constants of loss observed in the present experiments are overestimates.

MATERIALS AND METHODS

Experimental data. The kinetic parameters used to model concentrations of Ag, Cd and Co in *Macoma balthica* were quantified in laboratory experiments in this study and those from Griscom et al. (2000) and Griscom & Fisher (2002).

Macoma balthica were collected in June 1999 from an exposed mudflat near the Palo Alto Baylands, California, in south San Francisco Bay, where this population of clams has been monitored for metal concentrations for over 25 yr (Hornberger et al. 2000). Clams were fed a continuous supply of the diatom *Thalassiosira pseudonana* and were acclimated to experimental conditions (12°C, salinity 25) for 10 d prior to experiments. Sediments fed to *M. balthica* for the efflux study were collected from the same location; 14 individual clams were used for each experiment, and 2 each were sacrificed at the beginning and end of the depuration period to determine the distribution of metals between shell and soft parts.

The protocols used to label the diatoms uniformly for use in assimilation efficiency experiments have been described elsewhere (Reinfelder & Fisher 1991, Wang & Fisher 1996). Briefly, *Thalassiosira pseudonana* was grown in 500 ml filtered f/2 seawater media as described elsewhere (Fisher & Wente 1993), radiolabeled with μl additions of $^{110\text{m}}\text{Ag}$, ^{109}Cd , and ^{57}Co together (added as $^{110\text{m}}\text{Ag}$ in 0.5 N HNO_3 , ^{109}Cd in 0.1 N HCl , and ^{57}Co in 0.1 N HCl). The radioactive diatoms were resuspended in non-radioactive seawater and fed in several pulses to actively filtering *Macoma balthica* so that a concentration of 4 mg dry wt phytoplankton l^{-1} was maintained. After 1 h, the clams were removed and rinsed with a supersaturated solution of EDTA followed by deionized water to remove metals adsorbed to the shell, and then whole clams ($n = 10$) were individually gamma-counted (typically 2 min) and returned to a non-radioactive aquarium. Clams were depurated in clean seawater for 4 d, during which time they were continuously fed unlabeled *T. pseudonana* at the same cell density as during the radioactive pulse-feeding. Fecal material was removed frequently in order to minimize potential recycling of the radioisotopes, and the aquarium water was changed after 1 d. Periodic gamma-counting of the water showed that radioactivity in the depuration aquarium was not different from background values throughout the experiment. The assimilation efficiency of each metal was determined as the amount retained after 3 d. A more thorough description of the methods used to perform assimilation efficiency experiments is given in Griscom et al. (2000).

Radioanalysis of samples was determined non-destructively with inter-calibrated gamma counters equipped with large well NaI(Tl) detectors. The gamma emissions of $^{110\text{m}}\text{Ag}$ were measured at 658 keV, of ^{109}Cd at 88 keV, and of ^{57}Co at 122 keV. Counting times were 1 to 5 min, sufficient to yield propagated counting errors <5%. The radioactivity in the soft parts of whole clams was determined by subtracting the radioactivity of dissected shells from whole body radio-

activity. Reported assimilation efficiencies are means of 10 individuals ± 1 SD.

Dissolved metal influx rate constants (k_u) for *Macoma balthica* were determined by measuring the metal uptake rates of 4 concentrations of radiolabeled metal in coastal seawater and 2 metal concentrations in oxidized pore water. The methods are described in detail elsewhere (Griscom & Fisher 2002). Briefly, approximately 4 l of surface sediment was oxidized by bubbling with air for 24 h. The oxidized pore water was extracted by centrifugation followed by 0.2 μm filtration of the decanted liquid. The experimental concentrations of metal in the oxidized pore water and the overlying coastal water were created by adding a mixture of stable and radioactive metals so that the resulting metal concentrations ranged from 0.2 to 10 $\mu\text{g l}^{-1}$ for Ag and from 0.5 to 50 $\mu\text{g l}^{-1}$ for Cd and Co (Griscom & Fisher 2002).

The efflux rates measured in this study were calculated in clams pulse-fed radiolabeled sediment for 2 h and then depurated for 7 d, similar to the approach used to measure assimilation efficiencies described above. Sediments were radiolabeled as described in Griscom et al. (2000). By using gamma-emitting tracers, loss was measured non-destructively in individuals. Loss rates were calculated using the data points from 4 to 7 d, well after the unassimilated material had been completely egested; this loss is taken to represent the physiological turnover from the slowest exchanging compartment. Efflux rates after uptake from the dissolved phase were not measured.

Field data. Measurements of metal concentrations in sediments, overlying water and pore water were used in the model to evaluate metal bioaccumulation in *Macoma balthica* in San Francisco Bay; model predictions were compared to measured metal concentrations in clam tissues. In this study, concentrations of Ag, Cd and Co were measured in surface sediments and clams collected from an exposed mudflat at the same site as that used for obtaining clams for metal bioaccumulation experiments.

Sediments were scraped from the surface layers (1 to 2 cm) of mud at low tide and immediately returned to the laboratory, where particles were sieved through a 100 μm mesh with distilled water. The mesh size was chosen to remove large grains that might bias interpretation of concentrations and to analyze the size fraction ingested by *Macoma balthica*. The sieved fraction was dried at room temperature and samples of 0.25 to 0.5 g were further dried at 60°C before re-weighing and extraction. Replicates were digested for 'total' metal by refluxing in 10 ml concentrated HNO_3 . After decomposition, samples were evaporated to dryness and reconstituted in dilute HCl for analysis. Another set of replicate sediment samples was subjected to a partial

weak acid extraction in 10 ml of 0.5 N HCl for 2 h at room temperature to provide a chemical estimate of bioavailable Ag. Extracts were filtered through a 0.45 μm filter before analysis.

Macoma balthica were collected simultaneously with the sediment samples. More than 40 individuals, ranging from approximately 7 to 30 mm, were collected and returned to the laboratory for gut evacuation for 48 h. The clams were then dissected and the tissue samples were dried, weighed and refluxed in concentrated HNO_3 until the digest was clear. Digests were dried and reconstituted in dilute HCl for trace-metal analysis.

Metal analysis for sediment and clams were conducted by inductively coupled plasma emission spectroscopy. Cd and Ag in sediments were analyzed by graphite furnace atomic absorption spectrometer (GFAAS) with Zeeman background correction. All glassware and field collection apparatus were acid-washed, thoroughly rinsed in ultra-clean deionized water, dried in a dust-free positive-pressure environment, and stored in a dust-free cabinet. With each analytical run, quality control was maintained by frequent analysis of blanks and NIST standard reference materials (tissues and sediments). Analysis of reference materials were within an acceptable range of certified values reported by NIST. For a more detailed description of the analytical procedure used to measure the concentrations of metals in clams and sediment, see Brown & Luoma (1995) and Hornberger et al. (1999).

The clams and sediments used in most of the experiments were collected in the fall of 1996. For comparisons between model-predicted metal concentrations in clams and concentrations measured in field collected clams, the averaged yearly concentrations of Ag and Cd from 1996 and of Co in 1998 were used.

RESULTS AND DISCUSSION

Assimilation efficiencies of Ag, Cd, and Co in *Macoma balthica* from the uniformly radiolabeled diatom *Thalassiosira pseudonana* were $39 \pm 3\%$, $51 \pm 4\%$, and $30 \pm 3\%$, respectively (Table 1); values are means and standard deviations for replicate clams for each metal. These values were lower than other assimilation efficiency results with *T. pseudonana* (Reinfelder et al. 1997) and 2 times higher than Ag and Co assimilation efficiencies and 5 times higher than the Cd assimilation efficiency from organic-poor San Francisco Bay sediment (Griscom et al. 2000) (Table 1). Metal assimilation efficiencies of Ag, Cd, and Co from *T. pseudonana* in *M. balthica* were generally comparable with metal assimilation efficiencies in the mussel *Mytilus edulis* (Wang et al. 1996). Results from other studies, including work with *M. balthica*, are shown in

Table 1. Assimilation efficiencies used in the model include results from *T. pseudonana* (Table 1) and from surface sediment particles collected from a mudflat in south San Francisco Bay (Palo Alto Baylands) (Griscom et al. 2000).

Results from pulse-feeding experiments (to measure assimilation efficiency and efflux rates) showed rapid loss of metal, as the initial radioactive bolus of food was egested by the clams, followed by a slower loss rate reflecting loss of assimilated metal from tissues (Figs. 1 & 2). This pattern is typical for diverse marine invertebrates and has been described before for *M. balthica* (Decho & Luoma 1991, Reinfelder et al. 1997, Griscom et al. 2000). The efflux rates used in the modeling ranged from $0.9\% \text{ d}^{-1}$ for Ag to $3\% \text{ d}^{-1}$ for Co (Fig. 2, Table 1); reported errors represent 1 SD of the slope of the regression line (Table 1). When all existing efflux data were analyzed (Table 1), no clear difference in loss rates were found in clams following short- or long-term exposure to Cd; Ag and Co loss was only measured following short-term (1 h) exposure.

Table 2 presents Ag, Cd, and Co concentrations measured in surface sediments and clams in south San Francisco Bay. For modeling purposes, metal concentrations in particulate matter (surface sediment from this study, suspended particulate matter and phytoplankton from other studies) that could be used as food by *Macoma balthica* are given in Table 3. Concentrations of these metals in overlying water and pore water in south San Francisco Bay were taken from literature values and are also shown in Table 3.

Total sediment concentrations of metals were not assumed to be equal to the bioavailable metal in these sediments. Potentially bioavailable Ag, Cd and Co concentrations bound to sediment were determined by subtracting the lattice-bound metal component from the total sediment and suspended particulate matter metal concentrations (Table 4). This determination was

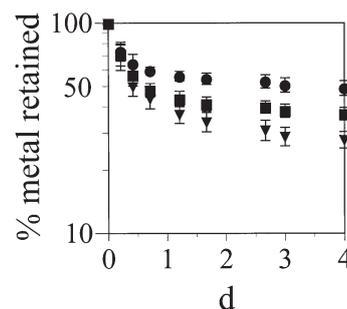


Fig. 1. *Macoma balthica*. Percentage of Ag (■), Cd (●) and Co (▼) retained by the clam over a 4 d depuration period following pulse-feeding on the uniformly radiolabeled diatom *Thalassiosira pseudonana*. Assimilation efficiencies were calculated as % retained at 3 d. Values are means for 10 individuals, error bars denote ± 1 SD

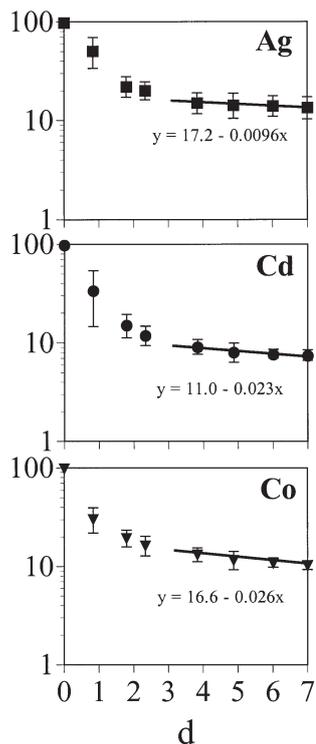


Fig. 2. *Macoma balthica*. The percentage of Ag (■), Cd (●) and Co (▼) retained by the clam (ordinate) over a 7 d depuration period following pulse-feeding on radiolabeled San Francisco bay surface-sediment. Efflux rates were calculated from 4 to 7 d using the regressions for the slowing exchanging compartment denoted by lines. Values are means for 10 individuals, error bars denote ± 1 SD

made because it is presumed that the lattice-bound metal in the core of the sediment particles is not assimilable by clams and because the assimilation efficiency measurements using radiotracers are applicable to metals sorbed to sediment (i.e. only those metals associated with surface layers around sediment particles). Thus, it is appropriate to apply these assimilation efficiency values to the surface-bound metals, not total metals associated with the sediment particles. Background (lattice-bound) concentrations of Ag, Cd, and Co were estimated from measurements of these metals in mean shale (IAEA 1985). The concentrations of Ag in the mean shale compared well with Ag concentrations in sediments at the bottom of meter-long cores collected in San Francisco Bay (Hornberger et al. 1999). No core data were available for Cd and Co concentrations in San Francisco Bay. A full discussion of the methods used to estimate the concentration of bioavailable metal in sediments is given in Campbell et al. (1988).

Influx rates of metals from overlying water (taken from Griscom & Fisher 2002) displayed a significant ($p < 0.01$) positive linear relationship with the dissolved

ambient metal concentrations. Calculated metal uptake rates from overlying water were highest for Ag; Cd and Co were comparable and were $10\times$ lower than Ag (Table 1). Uptake results for Cd were identical to those measured by Lee et al. (1998). Uptake of oxidized pore-water metal was not significantly different from the overlying water treatment except for Co, which was $3\times$ lower ($p < 0.01$).

A modeling sensitivity analysis was performed to evaluate the relative importance of dietary and dissolved source terms of metal accumulation in clams (Fig. 3). The equation used to differentiate the percentage of metal from food accumulated in filter-feeding clams was:

% from food =

$$\frac{C_{(sed)} \times IR \times AE_{(sed)}}{(C_{(SPM)} \times IR \times AE_{(SPM)}) + (k_{u(sw)} \times C_{w(sw)})} \times 100 \quad (7)$$

and in deposit-feeding clams was:

% from food =

$$\frac{C_{(sed)} \times IR \times AE_{(sed)}}{(C_{(sed)} \times IR \times AE_{(sed)}) + (k_{u(opw)} \times C_{w(opw)})} \times 100 \quad (8)$$

Parameter ranges were adjusted individually, while holding all other parameters constant at average values (Tables 1 & 2, ranges in Table 5). The parameters included AE, C_f , IR and k_u . A constant IR was used for

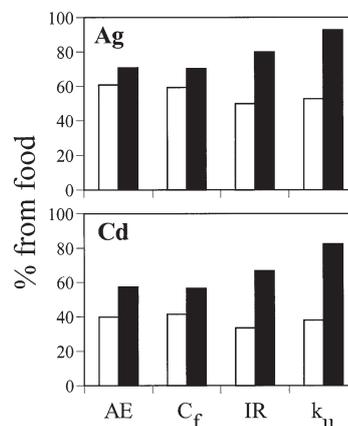


Fig. 3. *Macoma balthica*. Sensitivity analysis evaluating the fraction of Ag and Cd accumulated from food sources relative to water sources in deposit-feeding clams (calculated with Eq. 7). Parameter ranges were adjusted individually, while holding all other parameters constant at average values. AE: assimilation efficiency; C_f : metal concentration in food; IR: ingestion rate; k_u : uptake rate constant. Bars represent upper (solid bars) and lower (open bars) range of each parameter for deposit-feeding clams. The high and low values used for each parameter and for each metal are given in Table 5, with the exception of IR which was varied from 0.25 to 1.0 (g dry wt food g^{-1} dry wt clam). These values represent the range of IRs reported for this species (Bubnova 1972, Hummel 1985, Luoma et al. 1992)

the modeling but in the sensitivity analysis IR rates were varied from 0.25 to 1.0, which represent the range of IRs reported by others (Bubnova 1972, Hummel 1985, Luoma et al. 1992). A sensitivity analysis was not calculated for Co for which dietary uptake was minimal.

The general agreement between measured Ag, Cd, and Co concentrations in clams from south San Francisco Bay and the ranges predicted by the model (Table 6) suggests that the average rate parameters

experimentally derived for *Macoma balthica* adequately describe rates of metal uptake and loss in the clams living in these waters and that the model can account for the major processes governing metal concentrations in these clams. The range of Ag and Cd concentrations predicted for filter-feeding clams was higher than that predicted for deposit-feeding clams (Table 6). For Co the predicted concentration range was greater in deposit-feeding clams than in filter-feeding clams (Table 6). Metal concentrations in the

Table 1. *Macoma balthica*. Compilation of laboratory-derived values for parameters used in the biokinetic model to predict concentrations of Ag, Cd, and Co in clams living in San Francisco Bay (SFB). Parameters considered are assimilation efficiency, uptake-rate constant (k_u) and efflux rate (k_e). Values are means from replicate clams ± 1 SD. *Parameter values used in this study

Metal	Value	Comment	Source
Assimilation efficiency (%)			
Ag	18 \pm 4*	Sediment: oxic, organic-poor (SFB)	Griscom et al. (2000)
	21 \pm 4	Sediment: oxic, organic-rich (Flax Pond, Long Island)	Griscom et al. (2000)
	11 \pm 2	Sediment: anoxic, organic-rich (Flax Pond, Long Island)	Griscom et al. (2000)
	39 \pm 3*	Algae (<i>Thalassiosira pseudonana</i>)	This study
	38 \pm 1	Algae (<i>Isochrysis galbana</i>)	Reinfelder et al. (1997)
	49 \pm 1	Algae (<i>Thalassiosira pseudonana</i>)	Reinfelder et al. (1997)
Cd	9.7 \pm 3*	Sediment: oxic organic-poor (SFB)	Griscom et al. (2000)
	23 \pm 2	Sediment: oxic organic-rich (Flax Pond, Long Island)	Griscom et al. (2000)
	9 \pm 2	Sediment: anoxic organic-rich (Flax Pond, Long Island)	Griscom et al. (2000)
	13 \pm 2	Spring bloom particles (SFB) ^a	Lee & Luoma (1998)
	8.2 \pm 2	Sediment + benthic microalgae (SFB) ^b	Lee & Luoma (1998)
	33 \pm 7	Benthic microalgae (SFB) ^b	Lee & Luoma (1998)
	51 \pm 4*	Algae (<i>Thalassiosira pseudonana</i>)	This study
	69 \pm 2	Algae (<i>Isochrysis galbana</i>)	Reinfelder et al. (1997)
	88 \pm 10	Algae (<i>Thalassiosira pseudonana</i>)	Reinfelder et al. (1997)
Co	14 \pm 6*	Sediment: oxic, organic-poor (SFB)	Griscom et al. (2000)
	30 \pm 4	Sediment: oxic, organic-rich (Flax Pond, Long Island)	Griscom et al. (2000)
	16 \pm 6	Sediment: anoxic, organic-rich (Flax Pond, Long Island)	Griscom et al. (2000)
	30 \pm 3*	Algae (<i>Thalassiosira pseudonana</i>)	This study
	53 \pm 0	Algae (<i>Isochrysis galbana</i>)	Reinfelder et al. (1997)
	45 \pm 4	Algae (<i>Thalassiosira pseudonana</i>)	Reinfelder et al. (1997)
k_u (l g ⁻¹ d ⁻¹)			
Ag	0.354 \pm 0.076*	Salinity 30, overlying water (range 0.28–0.45)	Griscom & Fisher (2002)
	0.348 \pm 0.273*	Oxidized pore water (SFB)	Griscom & Fisher (2002)
Cd	0.033 \pm 0.005*	Salinity 30, overlying water	Griscom & Fisher (2002)
	0.032	Salinity 20, overlying water	Lee et al. (1998)
	0.0213 \pm 0.0138*	Oxidized pore water (SFB)	Griscom & Fisher (2002)
Co	0.035 \pm 0.005*	Salinity 30, overlying water	Griscom & Fisher (2002)
	0.0107 \pm 0.006*	Oxidized pore water (SFB)	Griscom & Fisher (2002)
k_e (d ⁻¹)			
Ag	0.0096 \pm 0.004*	Loss between 4 and 7 d after 1 h pulse of sediment	This study
	0.04 – 0.09	13 d loss after 1 h pulse-feed on algae	Reinfelder et al. (1997)
Cd	0.023 \pm 0.005*	Loss between 4 and 7 d after 1 h pulse of sediment	This study
	0.018	21 d loss after 7d uptake from food + water	Lee et al. (1998)
	0.0094	70 d loss after uptake from water only	Langston & Zhou (1987)
	≤ 0.01	13 d loss after 1 h pulse-feed on algae	Reinfelder et al. (1997)
Co	0.026 \pm 0.004*	Loss between 4 and 7 d after 1 h pulse of sediment	This study
	0.085	13 d loss after 1 h pulse-feed on algae	Reinfelder et al. (1997)
^a Approximately 80% <i>Thalassiosira rotula</i>			
^b <i>Achnanthes hungarica</i> , <i>Cylindrotheca gracilis</i> , <i>Navicula</i> sp. and <i>Nitzschia</i> sp.			

Table 2. Concentrations ($\mu\text{g g}^{-1}$) and annual means of Ag and Cd in surface sediments (<2 cm depth, <100 μm size fraction) and in the soft tissues of the clam *Macoma balthica* collected in 1996 from the Palo Alto mudflat in south San Francisco Bay. Errors represent standard deviation of 2 samples for sediment for each sampling date. For clams, the errors represent standard deviations of the mean of 10 replicate samples each containing 6 to 9 clams. Co concentrations in clams are derived from 1 sampling date in 1998 (using the same sampling scheme described above) at the Palo Alto site. Co concentrations in Palo Alto surface sediments are reported as a range of values (from Rivera-Duarte & Flegal 1997b). ns: no sample

Date	Ag		Cd		Co	
	Sediment	Clam	Sediment	Clam	Sediment	Clam
Jan 17	0.65 \pm 0.00	8.4 \pm 0.6	0.22 \pm 0.00	0.30 \pm 0.00	ns	ns
Feb 13	0.84 \pm 0.07	10.3 \pm 1.1	0.24 \pm 0.03	0.41 \pm 0.10	ns	ns
Mar 13	0.65 \pm 0.01	10.7 \pm 0.4	0.40 \pm 0.03	0.51 \pm 0.02	ns	ns
Apr 10	0.67 \pm 0.09	6.3 \pm 0.7	0.33 \pm 0.05	0.30 \pm 0.00	ns	ns
Jun 18	0.53 \pm 0.02	4.0 \pm 0.4	0.26 \pm 0.05	0.19 \pm 0.01	ns	ns
Sep 26	0.44 \pm 0.01	5.3 \pm 0.5	0.17 \pm 0.01	0.27 \pm 0.01	ns	ns
Dec 9	0.43 \pm 0.02	8.2 \pm 1.0	0.20 \pm 0.01	0.30 \pm 0.04	ns	ns
Annual mean	0.59 \pm 0.14	7.6 \pm 2.3	0.27 \pm 0.08	0.33 \pm 0.10	8.8–17.6	2.4 \pm 0.8

Table 3. Metal concentrations in food (surface sediment, suspended particulate matter and phytoplankton) and in water (overlying seawater and pore waters). All values are means \pm 1 SD from south San Francisco Bay except where noted

Metal	Concentration	Comment	Source
Surface sediment ($\mu\text{g g}^{-1}$)			
Ag	0.5 \pm 0.01	Total conc., yearly mean (1996)	This study
	0.24 \pm 0.06	HCl-extracted, yearly mean (1996)	This study
Cd	0.27 \pm 0.08	Total conc., yearly mean (1996)	This study
Co	8.8–17.6	Upper 5 cm, total conc.	Rivera-Duarte & Flegal (1997b)
Suspended particulate matter ($\mu\text{g g}^{-1}$)			
Ag	0.78	April 1989, San Francisco Bay	Smith & Flegal (1993)
	1.25	August 1989, San Francisco Bay	Smith & Flegal (1993)
	1.86	December 1989, San Francisco Bay	Smith & Flegal (1993)
Cd	0.27 \pm 0.07	Pre-spring bloom 1996	Luoma et al. (1998)
	0.55 \pm 0.08	Spring bloom 1996	Luoma et al. (1998)
	0.35 \pm 0.07	Post-spring bloom 1996	Luoma et al. (1998)
Co	1.5	Ligurian Sea (coastal) 1998	Fisher et al. (2000)
Phytoplankton ($\mu\text{g g}^{-1}$)			
Ag	0.1–2.0	Lower conc. limit, location unknown	Eisler (1981)
	0.2	Monterey Bay	Martin & Knauer (1973)
Cd	1.4	Estimated during spring bloom 1996	Luoma et al. (1998)
	1.5	Monterey Bay	Martin & Knauer (1973)
Dissolved: overlying water (nM) (ng l ⁻¹ in parentheses)			
Ag	0.075 (8.1)	January 1994	Sañudo-Wilhelmy et al. (1996)
	0.060 (6.5)	1989	Flegal et al. (1991)
	0.024 (2.6)	April 1989	Smith & Flegal (1993)
	0.085 (9.2)	August 1989	Smith & Flegal (1993)
	0.089 (9.6)	December 1989	Smith & Flegal (1993)
Cd	0.81 \pm 0.02 (91)	Pre-spring bloom 1996	Luoma et al. (1998)
	0.4 \pm 0.04 (45)	Spring bloom 1996	Luoma et al. (1998)
	0.5 \pm 0.03 (56)	Post-spring bloom 1996	Luoma et al. (1998)
	0.5–1.5 (56–168)	May–August 1992	Rivera-Duarte & Flegal (1997b)
	1.1 (123)	January 1994	Sañudo-Wilhelmy et al. (1996)
Co	0.2–1.0 (12–60)	May–August 1992	Rivera-Duarte & Flegal (1997b)
	0.26 \pm 0.04 (16)	Ligurian Sea (coastal)	Fisher et al. (2000)
Dissolved: pore water (nM) (ng l ⁻¹ in parentheses)			
Ag	0.54 (58)	Average conc.	Rivera-Duarte & Flegal (1997a)
	0.02–1.1 (22–120)	Upper 5 cm	Rivera-Duarte & Flegal (1997a)
Cd	0.05–1.5 (5.6–168)	Upper 5 cm	Rivera-Duarte & Flegal (1997b)
Co	0.3–60 (20–3500)	Upper 2 cm	Rivera-Duarte & Flegal (1997b)

Table 4. Mean concentrations (± 1 SD) of Ag, Cd and Co in average shale (IAEA 1985), in surface and suspended sediments collected from south San Francisco Bay (Smith & Flegal 1993, Rivera-Duarte & Flegal 1997b, Luoma et al. 1998) or, where not available for San Francisco Bay, metal concentrations in coastal suspended particles from elsewhere (Martin & Knauer 1973, Eisler 1981, Fisher et al. 2000). Metal concentrations are from total digested sediment and suspended particulate matter and include lattice-bound metals in detrital grains as well as labile metals in surface coatings. Potentially bioavailable metal concentrations were calculated by subtracting from the total digested concentrations the 'background' metal concentration measured in sediments from the mean shale concentration. The corrected concentrations in the food (C_f) (Table 5) were applied to assimilation efficiencies derived from experiments that radiolabeled the Csurface-bound metals in sediment coatings. All values are in $\mu\text{g g}^{-1}$

	Ag	Cd	Co
Mean shale	0.07	0.22	19
Surface sediment	0.59 ± 0.14	0.27 ± 0.08	18
Potentially bioavailable metal	0.52	0.05	0
Suspended particles	1.32 ± 0.54	0.41 ± 0.14	1.5
Potentially bioavailable metal	1.25	0.19	0

field-collected clams agreed best with the calculation for deposit-feeding clams, consistent with observations that the morphology of the clam is similar to those of other detritus-feeding animals and the observation that clams living in sandy muds feed on surface deposits (Gilbert 1977, Hummel 1985).

Overall, correcting the total metal concentrations by subtracting the mean shale concentration resulted in an excellent match between the predicted and field concentrations of the 3 metals in clams. It is noteworthy that Ag was the most enriched metal relative to the mean shale concentration in San Francisco Bay sediment and suspended particulate matter (7 to $20\times$ greater than mean shale values) and Co the least enriched (Table 4). Assimilation efficiencies derived in the laboratory are most appropriately applied to the surface-bound, bioavailable fraction for the purpose of modeling, as mentioned above. Sediment particles, amended with Ag, Cd and Co (added as isotope), include lattice-bound metals that exchange minimally with the isotope. It is unlikely that the lattice-bound metals are available to animals (Luoma 1989).

Table 5. *Macoma balthica*. Parameter ranges used to model metal concentrations in clams in 2 different feeding modes: filter-feeding and surface deposit-feeding. AE: % assimilation efficiency of metals from south San Francisco Bay surface sediment or from phytoplankton; IR: ingestion rate ($\text{g g}^{-1} \text{d}^{-1}$); C_f : bioavailable metal concentration ($\mu\text{g g}^{-1}$) in suspended particulate matter for filter-feeders and surface sediment for deposit-feeders corrected by subtracting the mean shale metal concentration. k_u : uptake rate ($\text{l g}^{-1} \text{d}^{-1}$) from seawater (SW) and surface oxidized pore water (OPW); C_w : dissolved metal concentration (ng l^{-1}) in SW and OPW; k_e : efflux rate constant (d^{-1}) in SW and OPW

Parameter	Ag		Cd		Co	
	Filter-feeding	Deposit-feeding	Filter-feeding	Deposit-feeding	Filter-feeding	Deposit-feeding
AE	36–42	12–22	47–55	6.4–13	27–33	8–20
IR	0.5	0.5	0.5	0.5	0.5	0.5
C_f	0.73–1.75	0.38–0.66	0.125–0.255	0.035–0.065	0	0
k_u SW	0.278–0.43	–	0.028–0.038	–	0.03–0.04	–
k_u OPW	–	0.076–0.622	–	0.008–0.035	–	0.005–0.017
C_w	2.6–9.6	16–120	45–168	6–220	12–60	20–3500
k_e	0.0092–0.01	0.0092–0.01	0.018–0.028	0.018–0.028	0.022–0.03	0.022–0.03

Table 6. Model-predicted metal concentrations in *Macoma balthica* compared with measured tissue concentrations in clams from south San Francisco Bay. Data ranges used in the biokinetic model are presented in Table 5. Metal concentrations are expressed as $\mu\text{g g}^{-1}$ dry wt of soft tissue (data are averages of 7 collection times throughout 1996—see Table 2). Error is expressed as ± 1 SD

	Ag ($\mu\text{g g}^{-1}$)		Cd ($\mu\text{g g}^{-1}$)		Co ($\mu\text{g g}^{-1}$)	
	Filter-feeding	Deposit-feeding	Filter-feeding	Deposit-feeding	Filter-feeding	Deposit-feeding
Predicted (range)	7–80	1.3–21	0.6–8	0.02–0.9	0.001–0.01	0.03–2.7
Predicted (mean)	25.4	6.3	2.2	0.2	0.005	0.7
Measured	7.6 ± 2.3		0.33 ± 0.01		2.4 ± 0.8	

In our study, experimental sediments were radiolabeled for 14 d. The time period necessary to label the surface coatings of detrital particles 'uniformly' may be as long as 6 mo. The required time differs among metals (Griscom et al. 2000) and could be dependent on the intensity of digestion of the experimental organism (Griscom et al. 2002). In general, increased 'aging' of metals in a sediment proportionally decreases the bioavailable fraction of the radiolabel for bivalves (Griscom et al. 2000, Fan & Wang 2001). Assimilation efficiency experiments using radiolabeled natural sediment should consider the effects of radiolabeling time on metal lability in quantifying metal accumulation appropriately from ingested sediment. In 2 similar studies that modeled metal concentrations in bivalves, assessing Cr concentrations in suspended particulate matter with a weak acid extraction brought predicted Cr concentrations in the organisms within the range of field measurements (Wang et al. 1996, Roditi et al. 2000). Again, these studies, like the present one, provided a first-order accounting for the potentially bioavailable fraction by operationally considering the material sorbed to the particle surfaces rather than considering the core of the sediment particle as assimilable material.

By setting the potentially bioavailable Co concentration in sediment to zero (because the sediment concentration of Co did not exceed shale concentrations of this metal) in the model, the predicted metal source was presumed to be solely from the dissolved phase. However, it is recognized that some small amount of Co, possibly not easily detectable above the lattice-bound sediment Co, may be bound in algal and bacterial detritus associated with sediment coatings. If, for example, 10% of the Co found associated with suspended particulate matter was bioavailable (a difference that would not be measurable), then the predicted concentration of Co in filter-feeding clams would increase from 0.005 to 0.87 $\mu\text{g g}^{-1}$ and in deposit-feeding clams the predicted Co concentration would increase from 0.7 to 3.6 $\mu\text{g g}^{-1}$. Thus, the Co concentration in clams is very sensitive to small changes in the fraction of particulate Co that is potentially bioavailable.

Lastly, sediment and suspended particles also contain an important and variable living fraction that has a distinctly higher bioavailability of metal than sediment alone (Decho & Luoma 1996, Lee & Luoma 1998, Griscom et al. 2000). During spring blooms or periods of high benthic algal growth, clams may accumulate greater amounts of Ag, Cd, and Co from food due to both higher assimilation efficiencies and enrichment of metals in algae compared to bulk surface sediment (Lee & Luoma 1998, Luoma et al. 1998). *Macoma balthica* can be a selective feeder on surface sediment, indicating a capability for ingesting material of higher nu-

tritional value (Hylleberg & Gallucci 1975, Shumway et al. 1985). This complication was not considered in our study, although it may influence metal accumulation if, for example, a high proportion of sediment-bound metals were associated with living material (Lee & Luoma 1998).

Differences in predicted metal bioaccumulation due to feeding mode in *Macoma balthica* could be explained by distinct variations in the model parameters. For all 3 metals, assimilation efficiencies from *Thalassiosira pseudonana* were higher (2× higher for Ag and Co, 5× higher for Cd) than assimilation efficiencies from San Francisco Bay sediment (Tables 1 & 5). Also contributing to higher Ag and Cd accumulation in filter-feeding clams were higher C_f values; Ag was 2 to 3× higher in suspended particles and Cd was up to 2× higher. Because there was little or no bioavailable Co in food, ingestion was not an important source of Co uptake for *M. balthica*.

The modeling results also allowed for the differentiation of the relative importance of food vs water as sources of metal for the clams (Eqs. 7 & 8). In deposit-feeding clams, approximately two-thirds of the Ag and half of the Cd were accumulated from food, with estimated ranges of 49 to 93% for Ag and 33 to 82% for Cd (Fig. 3). In the filter-feeding mode, over 98% of the Ag and 90% of the Cd in clams were accumulated from food (data not shown graphically). Higher concentrations of Ag and Cd in suspended particulate matter than in sediments and higher metal assimilation efficiencies from phytoplankton relative to San Francisco Bay sediment partly explain the greater importance of food as a source of Ag and Cd for filter-feeding clams (Fig. 3 & Table 5). For deposit-feeding clams, high concentrations of Ag and Cd, entrained during siphoning of surface deposits, were responsible for the increased importance of dissolved Ag and Cd as a source.

Metal uptake from pore water remains a challenging parameter to quantify, as does defining the bioavailable fraction of sediment-bound metals. As in any laboratory experiment, our experiments designed to quantify the model parameters used here do not fully duplicate environmental conditions or the behavior of clams living in natural waters. Nevertheless, it is noteworthy that the ranges of predicted metal concentrations for deposit-feeding clams are close to values measured in field-collected clams, as noted previously.

The kinetic model treats the organism as a single compartment, although of course there are several (at least) pools of metals distributed within the organism among the different tissues, each with its own biological half-life (Fisher et al. 1996). In all animals, metal distributions among tissues and biochemical compartments are undoubtedly complex and subject to feedback (as demonstrated in larger animals: Reinfelder et

al. 1998). Still, this model, which treats the clam as a single compartment, describes the key processes governing trace metal (or organic contaminant) accumulation in whole individuals in a way that matches model predictions well with independent field observations.

Models offer the greatest promise for flexibly taking into account the complex geochemical, biological and physicochemical factors that influence bioaccumulation. Simple models can address broad questions, and, if their results are consistent with field observations, as here, then additional complexities can be added. For example, a recent study that used this modeling approach added a 'selectivity factor' term to the model to take into account feeding behavior that selected more organic-rich particles by benthic invertebrates (Weston et al. 2000). If the validity of the model overall can be established, then, even where predicted concentrations differ from field measurements, insight may be gained about unanticipated mechanisms controlling metal bioaccumulation.

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