

# Short-term accumulation of Cd and Cu from water, sediment and algae by the amphipod *Melita plumulosa* and the bivalve *Tellina deltoidalis*

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**ABSTRACT:** The benthic organisms *Melita plumulosa* (amphipod) and *Tellina deltoidalis* (bivalve) are currently being used as test species for assessing contamination of estuarine–marine sediments in whole-sediment toxicity tests in Australia. Radiotracer techniques were used to determine the rates of accumulation (at  $21 \pm 3^\circ\text{C}$ ) of cadmium and copper by these organisms from water, algae and sediment sources. A biokinetic model was then developed to separately quantify influx and efflux rates from the metal sources. For exposures to seawater, metal uptake ( $\text{l g}^{-1} \text{d}^{-1}$ ) and efflux rates ( $\% \text{d}^{-1}$ ) were 0.028 and 6.0 (Cd) and 0.12 and 16 (Cu) for *M. plumulosa* and 0.012 and 8.5 (Cd) and 0.19 and 11 (Cu) for *T. deltoidalis*. Net accumulation of metals from the dissolved phase was greater for Cu than Cd for both organisms. For sediment exposures, metal assimilation efficiencies (AEs, %) and efflux rates ( $\% \text{d}^{-1}$ ) were  $22 \pm 10$  and  $19 \pm 3$  (Cd) and  $7.8 \pm 3$  and  $31 \pm 4$  (Cu) for *M. plumulosa*, and  $25 \pm 3$  and  $17 \pm 3$  (Cd) and  $30 \pm 2$  and  $20 \pm 4$  (Cu) for *T. deltoidalis*. AEs and efflux rates for algae were  $56 \pm 4$  and  $9.8 \pm 1$  (Cd) and  $33 \pm 5$  and  $16 \pm 4$  (Cu) for *M. plumulosa*, and  $73 \pm 3$  and  $5.0 \pm 2$  (Cd) and  $49 \pm 4$  and  $4.3 \pm 1$  (Cu) for *T. deltoidalis*. For *M. plumulosa*, the presence of algae increased the rate of accumulation of both Cu and Cd from sediments, and indicated that *M. plumulosa* fed more rapidly, but not necessarily selectively, in the presence of algae. Ingestion rates of sediments were estimated as  $0.20$  and  $0.08 \text{ g g}^{-1} \text{d}^{-1}$  for *M. plumulosa* and *T. deltoidalis*, respectively. The biokinetic models indicated that metal accumulation by the organisms would increase greatly as partitioning to the pore waters increased or if organisms increased their ingestion of metal-contaminated algae in preference to sediments. If it is assumed that the toxic effect of accumulated metals is independent of accumulation pathway, then changes in sediment properties or in an organism's feeding behaviour would be expected to greatly affect the toxicity of metals to the organism. The study highlights the need for careful scrutiny of metal partitioning in sediments when data are to be used to estimate toxicity thresholds or effects concentrations, before such values are adopted in sediment quality guidelines.

**KEY WORDS:** Sediment · Metals · Bioaccumulation · Biokinetic modelling

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## INTRODUCTION

For most sediment-dwelling organisms, sediments, pore waters and overlying waters may all contribute to the accumulation of metal contaminants (Wang & Fisher 1999a, Griscom & Fisher 2002). Furthermore,

the metal speciation in each of these compartments, and the presence of different food sources, will modify metal uptake rates (Rainbow 1995, Wang & Fisher 1999a, Wang et al. 1999, Griscom et al. 2000, Fan et al. 2002). Whole-sediment bioassays are often used to assess the toxicity of contaminated sediments (ASTM

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2003, Simpson et al. 2005). Criteria for selecting test species include a demonstrated sensitivity to contaminants and direct contact and association of the organism with the sediment (ASTM 2003). Understanding the contaminant exposure pathways of a given species is, however, not listed in these criteria. For most species that are used routinely in toxicity tests, the relative importance of the different contaminant exposure pathways (overlying waters, pore waters, sediment, detritus) has not been evaluated (Batley et al. 2004).

The assessment of ecological effects occurring in contaminated sediments is likely to be more effective if the contaminant exposure pathways for individual species are known (Hare et al. 2003, Batley et al. 2004). Ignoring sediment- and food-exposure pathways could severely underestimate metal exposures for some benthic organisms. The combined use of exposure-pathway models, knowledge of effects concentrations and bioaccumulation following metal exposures from different sources (sediment–water partitioning) is expected to improve our understanding of cause–effect relationships (Simpson & King 2005).

Whole-sediment bioassays have recently been developed using 2 invertebrates, *Melita plumulosa* and *Tellina deltoidalis*, both endemic to Australia (Simpson et al. 2005, R. V. Hyne, S. A. Gale, C. K. King unpubl. data). *M. plumulosa* (Fam. Melitidae) is an epibenthic deposit-feeding amphipod commonly found in freshwater, estuarine and marine environments throughout SE Australia. The species lives in close association with sediments, inhabiting estuarine tidal mudflats ranging from silty to sandy sediments and seagrass beds, in waters up to 25 m depth. *M. plumulosa* are an important source of food for higher trophic levels. The bivalve *T. deltoidalis* (Fam. Tellinidae) lives infaunally in silty to sandy estuarine and coastal sediments from south Queensland to Tasmania and south Western Australia. It grows to approximately 25 mm in length, and although the biology of this species has not been studied, it is thought to be a deposit feeder like other tellinids, collecting organic material and particles from surface sediments.

In this study, we used radiotracer techniques to determine the rates of accumulation of cadmium and copper from water, sediment and algal sources by *Melita plumulosa* and *Tellina deltoidalis*. Biokinetic models were used to separately quantify influx and efflux rates from the metal sources. Detailed reviews have been published on this kinetic modelling approach (Wang & Fisher 1999a,b) and its application has been demonstrated (Wang & Fisher 1999a, Wang et al. 1999, Griscom et al. 2000, Griscom & Fisher 2002). These models were used to investigate the effects of changes in sediment properties and organism feeding behav-

our on the exposure pathways and the toxicity of cadmium and copper to *M. plumulosa* and *T. deltoidalis* residing in metal-contaminated sediments.

## MATERIALS AND METHODS

**Organisms.** *Melita plumulosa* were obtained from laboratory-maintained cultures originally established from organisms collected from intertidal mud flats at Brooklyn in the Hawkesbury River, north of Sydney. Procedures for maintaining cultures of *M. plumulosa* in the laboratory have been described previously (Hyne et al. 2005). *Tellina deltoidalis* of 5 to 8 mm in length were collected during low tide from mud flats at Boronia Park, Lane Cove River, Sydney, Australia, during 2002 and 2003. Procedures for the collection and holding of *T. deltoidalis* in the laboratory have been described previously (Simpson et al. 2005). *T. deltoidalis* were used in tests within 7 d of collection. All organisms were isolated from the holding sediment 24 h before test commencement by gentle sieving through 500  $\mu\text{m}$  sieves into holding trays containing 2 cm of filtered seawater. Overlying water in the holding trays was continuously gently aerated and trays were covered with foil to minimise light disturbances to the organisms. The organisms were not fed during this period.

**General analytical.** Deionised water (18 M $\Omega$  cm, Milli-Q) was used for washing and preparing all metal stock solutions. Seawater (32.5‰ salinity) was collected from Cronulla, Sydney, New South Wales and filtered through 0.45  $\mu\text{m}$  filters. All glass and plastic ware were cleaned by soaking in 10% (v/v) HNO<sub>3</sub> (Trace Pur, Merck) for >24 h, followed by rinsing with deionised water. All chemicals were analytical reagent grade or equivalent analytical purity. Measurements of pH (calibrated against pH 4 and 7 buffers) were made using a pH meter equipped with a pH electrode as described previously (Simpson & Batley 2003).

Cd-109 ( $t_{1/2}$  = 464 d) purchased from Perkin Elmer Life Sciences had a specific activity of 121 GBq g<sup>-1</sup> Cd at calibration. Cu-64 (specific activity = 140 TBq g<sup>-1</sup> Cu) in 0.02 M HCl was purchased from ANSTO Radio-pharmaceuticals and Industrials (ARI). The <sup>64</sup>Cu ( $t_{1/2}$  = 12.7 h) contains approximately 2% <sup>67</sup>Cu ( $t_{1/2}$  = 61.7 h). Hence <sup>67</sup>Cu emission was used to monitor copper distribution at 36 h post <sup>64</sup>Cu calibration time. Both materials contained less than 0.1% contamination of other radioisotopes. The <sup>109</sup>Cd and <sup>64</sup>Cu were carrier-free and, throughout all experiments, concentrations of Cd and Cu associated with the radioisotope additions were less than 0.5  $\mu\text{g l}^{-1}$ . The laboratory temperature was 21  $\pm$  3°C during all organism radioisotope exposure experiments.

**Sediment metal-spiking and radiolabelling.** Sediment used in these experiments was collected from Bonnet Bay, Woronora River, Sydney. The sediment was a hydrous (68% water) silty sediment (99% particles <63  $\mu\text{m}$ ), pH  $7.3 \pm 0.2$ , salinity 29‰, and was suboxic (acid-volatile sulphide < 0.5  $\mu\text{mol g}^{-1}$ , redox potential =  $-40 \pm 50$  mV). The sediment had  $12 \pm 2\%$  organic carbon and acid-extractable (30 min, 1 M HCl) metal concentrations (in  $\mu\text{g g}^{-1}$ ) of 6000 Fe, 50 Mn, 160 Zn, 66 Pb, 30 Cu, 4 Ni and <1 Cd (Simpson et al. 2004). Sediments were stored in the dark at 4°C for a maximum of 30 d prior to use.

Metal-spiked sediments were prepared in a nitrogen-filled glove box 48 h prior to radiolabelling, using the procedures described in Simpson et al. (2005). In a 30 ml Teflon centrifuge tube (Oak Ridge, Cole Parmer) 3 g of sediment (dry weight) was combined with sufficient deoxygenated (<0.2 mg  $\text{l}^{-1}$  dissolved oxygen) seawater to give a final water content of 80% (w/w). Metal concentrations of 0, 10, 40, 160 and 460  $\mu\text{g g}^{-1}$  Cd and 0, 20, 80, 320, and 960  $\mu\text{g g}^{-1}$  Cu in the sediments were achieved by spiking the sediments with 1000 mg  $\text{l}^{-1}$  standard solutions (Spectrosol, BDH) of Cd or Cu. The pH of the final sediment was adjusted to  $7.9 \pm 0.1$  using NaOH (15 M). This pH was chosen to promote the partitioning of metals to sediment particles, to ensure that the majority of iron(II) displaced to the pore waters by the added metals would oxidise and precipitate as iron hydroxide phases, and to minimise metal fluxes to the overlying water (Simpson et al. 2004). The pH-adjusted metal-spiked sediments were sealed, rapidly shaken for 30 s, then rolled on a bottle roller for 2 h before returning to the nitrogen glove box to equilibrate for 48 h before addition of radioisotopes.

Radioisotope additions of 330 and 5000 kBq  $\text{g}^{-1}$  sediment (dry weight) for  $^{109}\text{Cd}$  and  $^{64}\text{Cu}$ , respectively, were made 48 h before commencing the uptake experiments. Following the 48 h radiolabelling period, the sediments were centrifuged and the radioactive water (isolated pore water) was removed and replaced with non-radioactive filtered seawater. The water and sediments were then mixed for 30 s using a vortex mixer (Ratek Instruments). This procedure was performed 4 times and the radioactivity of the waste waters measured each time. The pH of the sediment slurries was measured before experiments to confirm that it was  $7.9 \pm 0.1$ . Based on radioisotope activity measured in the sediments and pore waters, sediment–water partition coefficients for the metals,  $K_d = [\text{metal in sediment}]/[\text{metal in water}]$ , were  $\sim 10^3$  and  $\sim 10^5$   $\text{l kg}^{-1}$  for Cd and Cu, respectively.

**Algae radiolabelling.** The marine diatom *Phaeodactylum tricornutum* (Bohlin) was chosen for the pulse-chase feeding experiments because it has previously been shown to accumulate copper (Franklin et al.

2001), is used as a food source for laboratory cultures of *Melita plumulosa*, and is easy to count and does not clump or adsorb to the walls of the test containers. *P. tricornutum* was obtained from CNR Istituto di Biofisica, Pisa, Italy and cultured in half-strength f medium (Guillard & Ryther 1962). Cultures were maintained on a 12 h light:12 h dark cycle (Philips TL 40 W fluorescence daylight, 72  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ ) at 21°C. Cell density measurements made using a particle analyser with 70  $\mu\text{m}$  aperture (Coulter Multisizer II, Beckman Coulter), were corrected for the background.

Radioisotope additions of 10 and 300 kBq  $\text{ml}^{-1}$  for  $^{109}\text{Cd}$  and  $^{64}\text{Cu}$ , respectively, were made to algae resuspended in 50 to 150 ml of filtered seawater ( $300 \times 10^4$  cells  $\text{ml}^{-1}$ ) maintained at light and temperature conditions used for the culturing. Following a 48 h radiolabelling period, the algae were washed by filtering through 0.45  $\mu\text{m}$  filters (25 mm glass microanalysis filter holder, Millipore), discarding the radioactive filtrate water and resuspending the algae retained on the filter in non-radioactive filtered seawater. The washing procedure was performed 4 times and the radioactivity of the filtrate water was measured each time to evaluate metal uptake by the algae.

**Metal uptake from the dissolved and sediment phases.** Metal uptake from the dissolved phase was determined by exposing the organisms to concentrations of 0.4, 2, 10 and 50  $\mu\text{g l}^{-1}$  Cd or Cu over time. Dissolved metal concentrations were prepared by spiking exposure wells with 20 to 100  $\mu\text{l}$  volumes of 0.2 or 5 mg  $\text{l}^{-1}$  metal solutions prepared from 1000 mg  $\text{l}^{-1}$  stock solutions (Spectrosol, BDH). Both radioisotope and stable metals were added to the wells containing 10 ml of filtered seawater 1 h before commencing the uptake experiments. The amounts of radioisotopes added were 2 and 15 kBq  $\text{ml}^{-1}$  for Cd and Cu, respectively. The pH of the well waters was  $7.9 \pm 0.1$  after metal additions.

Metal uptake from the sediment phase was determined by exposing the organisms to metal-spiked (0 to 460  $\mu\text{g g}^{-1}$  Cd, 0 to 960  $\mu\text{g g}^{-1}$  Cu) and radiolabelled (400 kBq  $\text{g}^{-1}$   $^{109}\text{Cd}$ , 5000 kBq  $\text{g}^{-1}$   $^{64}\text{Cu}$ ) sediments. Approximately 50 mg (dry weight equivalent) of the sediment was delivered to each exposure well (containing 10 ml of non-radioactive filtered seawater) using a pipette with a disposable plastic tip, widened (cut off) to aid sediment dispensing.

Twelve organisms were placed in individual wells for each treatment. At intervals of 2, 5, 7, 10 and 24 h, each organism was removed from the well containing the radiolabelled water or sediment, rinsed 3 times with non-radioactive filtered seawater and then returned to the respective well following counting of its radioactivity. The organisms were out of the wells for 15 min during this period. After 24 h of exposure to

the radioactive water or sediment, the organisms were depurated in non-radioactive wells containing clean, filtered seawater, or filtered seawater and sediment, respectively. During the depuration period, the radioactivity of the organisms was counted at intervals of 2, 5, 7, 10 and 24 h and the organisms returned to trays containing 10 ml of new, filtered seawater (replaced after each depuration period), or filtered seawater and sediment (not replaced), respectively. After the depuration period, organisms were dried at 60°C for 48 h and their dry weights determined. For the experiments with *Tellina deltoidalis*, an additional set of 6 wells were prepared for each of the 5 uptake-time treatments and for the depuration treatments, and identical experiments were undertaken. For these organisms, 6 bivalves were processed at each uptake period and each depuration period. For each, the adductor muscle between the 2 shell valves was cut, and water, algae and sediments were thoroughly washed out from within the shells, before the radioactivity of the organism was counted. The data from 'open shell' organisms, with individuals monitored at a single time point only, was used to compare results from 'closed shell' organisms that were monitored through time. The portion of radioactivity that was contained within the shell, but was not attributed to uptake by the organism into its tissues or shell, could therefore be determined. The 'open shell' organisms were then dissected into shell and tissue portions and the activity of each portion determined.

**Radioactive pulse-chase feeding and depuration.** Pulse-chase experiments were undertaken to determine metal assimilation efficiency from algae and sediment food sources and to obtain information on uptake and depuration rates. Pulse-chase feeding experiments were undertaken by placing organisms individually in exposure wells containing 10 ml of filtered seawater and the desired amount of radiolabelled algae ( $5 \times 10^5$  cells ml<sup>-1</sup>, 20 kBq Cd or 60 kBq Cu) or sediment (50 mg dry weight, 20 kBq Cd or 250 kBq Cu). Twenty-seven organisms were placed in individual wells for each treatment (3 uptake time periods  $\times$  9 organisms) and were allowed to feed for periods (pulses) of 2, 4 or 6 h with the radiolabelled algae, and for 4, 6 or 8 h with the radiolabelled sediments. At the end of each time period, 9 organisms were removed from the well water, rinsed 3 times with new, filtered seawater and their radioactivity was counted. The organisms were then placed in individual wells containing 10 ml of clean, filtered seawater and the same quantity of non-radioactive algae or sediments, respectively, that they were fed previously, to depurate. During the depuration period, the radioactivity of the organisms was counted (following washing) at intervals of 2, 5, 7, 10 and 24 h and they were

then returned to wells containing 10 ml of new, filtered seawater with non-radioactive algae or sediments. After the 24 h depuration period, organisms were dried at 60°C for 48 h and their dry weights determined. An additional set of 6 wells containing *Tellina deltoidalis* were prepared for each of the 3 algae pulse-feeding periods and the 3 sediment pulse-feeding periods, and identical experiments were undertaken to obtain uptake data for organisms that were dissected, as described earlier.

**Influence of food type on metal assimilation.** Experiments were undertaken to determine whether the presence of algae would affect the rate of metal uptake by *Melita plumulosa* from sediments. *M. plumulosa* were exposed to radiolabelled particulate phases individually: sediments (50 mg, 400 kBq g<sup>-1</sup> <sup>109</sup>Cd, 5000 kBq g<sup>-1</sup> <sup>64</sup>Cu) and radiolabelled algae ( $5 \times 10^5$  cells ml<sup>-1</sup>, 20 kBq Cd or 60 kBq Cu); and combinations of radiolabelled particulate phases: Cu-sediments/Cd-algae and Cd-sediments/Cu-algae at the same activity as individual exposures. The exposure conditions were the same as those used for investigating uptake from dissolved and sediment phases (exposure wells containing 10 ml of non-radioactive filtered seawater, 12 organisms per treatment). These experiments were also used as replicate experiments for determining Cd and Cu influx and efflux rates from sediment and algae phases.

**Radioactivity measurements.** Radioactivity in water, sediments, algae and organisms was measured using a gamma counter (Wizard 1470, Wallac Oy, Finland) with a 3.30  $\times$  5.08 cm end well-type NaI crystal (Wallac Oy). The gamma emissions of <sup>109</sup>Cd, <sup>64</sup>Cu and <sup>67</sup>Cu were measured at 88, 511, and 93 and 184 keV, respectively. All samples measured were decay-corrected and only data 3-fold greater than background was used. The concentration of metal (Cd or Cu) present in each sample was correlated to radioactivity measured. Final concentrations of each metal ion in the organisms ( $C_O$ ,  $\mu\text{g g}^{-1}$ ) were calculated by multiplying the ratio of activity accumulated in the organisms ( $A_O$ , cpm) to the total well activity ( $A_{\text{Well}}$ , cpm), by the amount of metal in the well ( $C_{\text{Well}}$ ,  $\mu\text{g}$ ), and dividing by the organism dry weight ( $O_{\text{Wt}}$ , g):

$$C_O = (A_O/A_{\text{Well}}) \times C_{\text{Well}}/O_{\text{Wt}} \quad (1)$$

Throughout all experiments, 50  $\mu\text{l}$  samples of overlying water were taken and the radioactivity present measured. Non-specific binding of radioactivity on the wells was determined by sampling 3 wells per treatment, as representative of each experiment. Typically, at the completion of both the sediment and algae experiments, the entire contents of 3 representative wells per treatment were filtered and the activity of the filtrate measured. The calculated metal partition co-

efficients ( $Kds$ ) between the sediments or algae and the well water were then corrected for this non-specific binding.

**Modelling metal bioaccumulation from water, sediment and algae exposure pathways.** The metal bioaccumulation in the organisms from filtration of water or ingestion of food (sediment, algae) was described using a bioenergetic-based kinetic model (Thomann 1981, Wang & Fisher 1999b, Wang et al. 1999). In this model, the metal influx rate from water was determined by multiplying the metal concentration in the dissolved phase by an uptake rate constant that reflects the filtration rate and the adsorption efficiency of the dissolved metal in the organism. The metal influx rate from sediment or algae phases was determined by multiplying the metal concentration in the ingested sediment or algae by the ingestion rate and assimilation efficiency of the ingested metal. The loss rate of metal following uptake from each pathway was also considered. The steady-state expression for this model is:

$$C_O = k_{u-W} \times C_W/k_{e-W} (\text{water}) + AE_S \times IR \times C_S/k_{e-S} (\text{sediment}) + AE_A \times IR \times C_A/k_{e-A} (\text{algae}) \quad (2)$$

where  $C_O$ ,  $C_W$ ,  $C_S$  and  $C_A$  are the metal concentrations in the organism ( $\mu\text{g g}^{-1}$ ), dissolved phase ( $\mu\text{g l}^{-1}$ ), sediment ( $\mu\text{g g}^{-1}$ ) and algae ( $\mu\text{g g}^{-1}$ ), respectively,  $k_{u-W}$  and  $k_{e-W}$  are the uptake and efflux rate constants from the dissolved phase, respectively,  $AE_S$  and  $AE_A$  are the assimilation efficiencies from sediments and algae phases, respectively,  $IR$  is the organism's ingestion rate ( $\text{g g}^{-1} \text{d}^{-1}$ ),  $k_{e-S}$  and  $k_{e-A}$  are the metal efflux rates ( $\% \text{d}^{-1}$ ) following uptake from sediment and algae food phases, respectively. Growth of the organisms during the experiment was expected to be negligible and was not considered in the model. The model assumed that uptake from dissolved, sediment and algae phases were additive.

Investigations of the uptake of Cd and Cu from sediments were undertaken for a range of metal concentrations and allowed the calculation of an influx rate constant from the sediment phase,  $k_{u-S}$ . Uptake from the sediment phase was also modelled by replacing  $AE_S \times IR \times C_S/k_{e-S}$  (sediment) of Eq. (2) with  $k_{u-S} \times C_S/k_{e-S}$  (sediment), where  $k_{u-S}$  is the uptake rate constant from the sediment phase.

## RESULTS

### Metal partitioning during uptake experiments

During the 24 h exposure of *Melita plumulosa* to Cd and Cu from water, sediment and algal sources, total metal concentrations in the wells declined by 1 to 7%

(1 to 5% in 2 to 8 h assimilation efficiency experiments). This was attributed to metal uptake by *M. plumulosa* (major component) and metal adsorption or adhesion of sediment/algae to the well walls (minor component). Cadmium sediment–water partition coefficients ( $Kds$ ) decreased (~35%) in the presence of *M. plumulosa* from  $3.1 \times 10^3$  to  $2.0 \times 10^3 \text{ l kg}^{-1}$  as sediment cadmium concentrations increased from 10 to  $460 \mu\text{g g}^{-1}$ .  $Kds$  for copper were  $4.1 \times 10^4$  to  $5.0 \times 10^4 \text{ l kg}^{-1}$  and were independent of copper concentration in sediments.

During the 24 h exposure of *Tellina deltoidalis* to Cd and Cu from water, sediment and algal sources, total metal concentrations in the wells declined by varying amounts. For the 3 exposure sources, metal concentrations declined by 30 to 45% for waters, 10 to 40% for algae and 8 to 15% for sediments. The decline was attributed mostly to metal uptake by *T. deltoidalis*, and the extent of decline reflected the dominant exposure pathway. No adjustment was made for concentration decline during the *T. deltoidalis* exposure experiments, and nominal concentrations were used in the development of the biokinetic model. Sediment–water partition coefficients ( $Kds$ ) in the presence of *T. deltoidalis* were  $4.7 \times 10^3$  to  $8.8 \times 10^3 \text{ l kg}^{-1}$  for cadmium and  $1.4 \times 10^5$  to  $1.6 \times 10^5 \text{ l kg}^{-1}$  for copper, and were independent of metal concentration in sediments.

During the 20 min washing period, the shell of *Tellina deltoidalis* may not have been sufficiently open to allow all water, sediment and algal materials to discharge. At the completion of experiments, the adductor muscles were cut to allow thorough washing out of materials within the shells. This thorough washing process resulted in concentrations decreasing by  $9 \pm 2\%$  (water),  $14 \pm 1\%$  (sediments) and  $6 \pm 2\%$  (algae) for Cu, and by  $11 \pm 4\%$  (water) for Cd (not measured for algae and sediment exposures). For the determination of influx and efflux rate constants and AE, total metal concentrations in the thoroughly washed *T. deltoidalis* were used.

Following these measurements, *Tellina deltoidalis* were then dissected into tissue and shell fractions. The ratios of metals in the tissues to total metals (tissue and shell, mean  $\pm$  SD) were  $57 \pm 9\%$  (waters),  $59 \pm 3\%$  (sediments) and  $91 \pm 2\%$  (algae) for Cu, and  $91 \pm 6\%$  (waters),  $82 \pm 14\%$  (sediments) and  $98 \pm 1\%$  (algae) for Cd. Tissue/shell metal ratios appeared to be independent of the metal concentration in the waters or sediments. The modelling calculations did not consider the ratio of metal in the tissue and shell fractions.

### Metal influx and efflux from the dissolved phase

The uptake of Cd and Cu from the dissolved phase by *Tellina deltoidalis* and *Melita plumulosa* is shown in Fig. 1. Although rapid surface adsorption of metals by

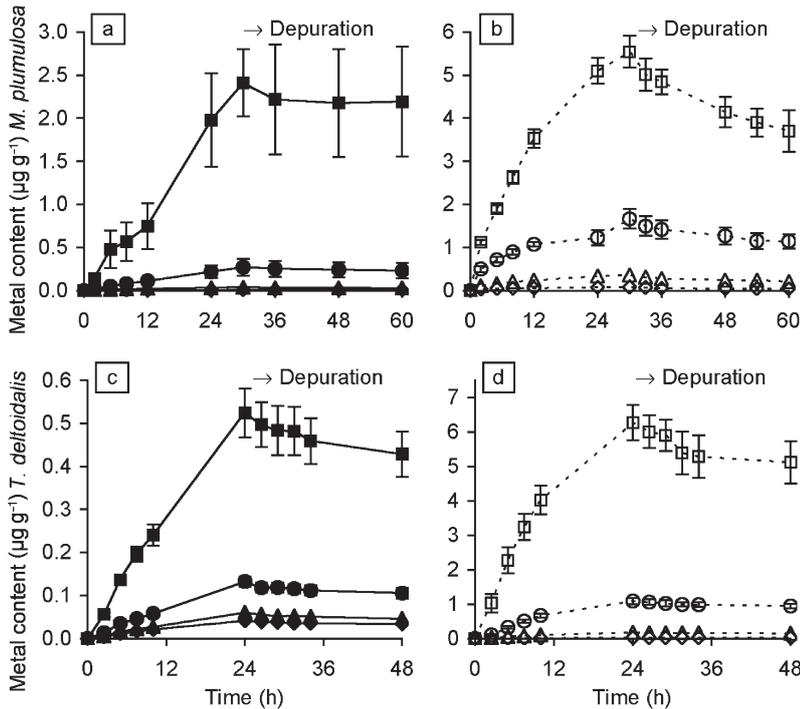


Fig. 1. *Melita plumulosa* and *Tellina deltoidalis*. Metal uptake, retention and depuration by (a,b) *M. plumulosa* and (c,d) *T. deltoidalis* exposed to dissolved cadmium (◆ 0.4, ▲ 2.0, ● 10, ■ 50  $\mu\text{g l}^{-1}$ ) and copper (◇ 0.4, △ 2.0, ○ 10, □ 50  $\mu\text{g l}^{-1}$ ). Error bars (shown for 10 and 50  $\mu\text{g l}^{-1}$  concentrations) represent standard errors for 12 replicate experiments at each concentration and exposure time

the organisms was expected, the rate of metal uptake during the initial 2 h was generally similar to the uptake rate for the initial 12 h, after which it decreased. Influx rates from the dissolved phase ( $I_W$ ,  $\mu\text{g g}^{-1} \text{d}^{-1}$ ) were calculated using the data from the 2 to 12 h and 2.5 to 10 h time periods for *M. plumulosa* and *T. deltoidalis*, respectively. The correlation between the  $I_W$  values and dissolved Cd and Cu concentrations for each organism is shown in Fig. 2. The uptake rate constants of Cd and Cu from the dissolved phase ( $k_{i-w}$ ,  $\text{l g}^{-1} \text{d}^{-1}$ ) were calculated from a regression of  $I_W$  and  $C_W$ , with the y-intercept set through zero. The choice of the zero intercept was based on theoretical considerations, and the rate constants were within 2% of values calculated without a zero intercept. For both species, there was greater rate of uptake of Cu than Cd from the dissolved phase.

Efflux rates were calculated from the decline in radioactivity of the organisms during the 24 h depuration period following the 24 h water uptake experiments. During the first 6 h of the depuration period, the loss of radioactivity was rapid and probably reflected clearance of the gut passage. Efflux rate constants ( $k_{e-w}$ ,  $\% \text{d}^{-1}$ , mean  $\pm$  SD), calculated using the data (5 experiments, 60 organisms) from the 7.5 to 24 h depuration period, were  $6.0 \pm 0.6$  (Cd) and  $16 \pm$

$0.8$  (Cu) for *Melita plumulosa*, and  $8.5 \pm 0.5$  (Cd) and  $11 \pm 1$  (Cu) for *Tellina deltoidalis*. Efflux rates did not appear to be dependent on metal concentrations in the waters.

### Metal influx and efflux from sediment particles

The uptake of Cd and Cu by *Melita plumulosa* and *Tellina deltoidalis* from sediments with increasing metal concentrations is shown in Fig. 3. For *M. plumulosa*, the rate of Cu uptake appeared to be greatest during the first 2 h, indicating that surface adsorption of Cu, perhaps associated with fine particles, may have contributed to the initial Cu uptake. The ventral body surface and appendages of *M. plumulosa* contain many fine hairs that are expected to offer many sites for metal binding and collection of fine particles. Metal influx rates and uptake rate constants of Cd and Cu from the sediments were calculated by the procedure used for uptake from the dissolved phase (Fig. 4).

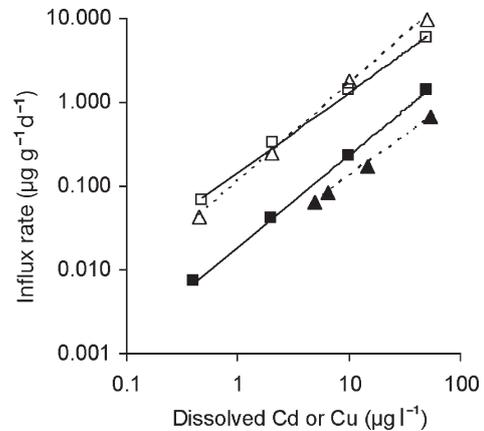


Fig. 2. *Melita plumulosa* and *Tellina deltoidalis*. Influx rates of (■, ▲) cadmium and (□, △) copper into (■, □) *M. plumulosa* and (▲, △) *T. deltoidalis* at different dissolved metal concentrations. The relationships describing metal influx rate ( $I_W$ ) and metal concentrations in the dissolved phase were:  $I_W = 0.0192[\text{Cd}]^{1.085}$  ( $r^2 = 1.00$ ) and  $I_W = 0.144[\text{Cu}]^{0.9565}$  ( $r^2 = 1.00$ ) for *M. plumulosa*, and  $I_W = 0.0136[\text{Cd}]^{0.9668}$  ( $r^2 = 1.00$ ) and  $I_W = 0.112[\text{Cu}]^{1.153}$  ( $r^2 = 1.00$ ) for *T. deltoidalis*. The calculated uptake rate constants ( $k_{i-w}$ ,  $\text{l g}^{-1} \text{d}^{-1}$ ) from the dissolved phase were 0.0278 (Cd) and 0.115 (Cu) for *M. plumulosa*, and 0.0120 (Cd) and 0.192 (Cu) for *T. deltoidalis*, calculated by linear regression of  $I_W$  and  $C_W$  without log-transformation

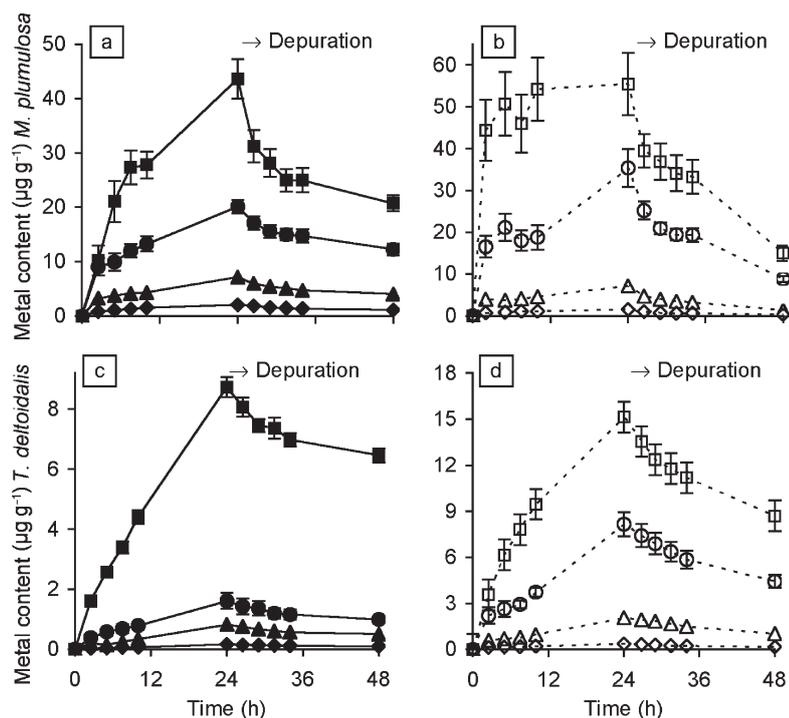


Fig. 3. *Melita plumulosa* and *Tellina deltoidalis*. Metal uptake, retention and depuration by (a, b) *M. plumulosa* and (c, d) *T. deltoidalis* exposed to sediment-bound cadmium (◆ 10, ▲ 40, ● 160, ■ 480  $\mu\text{g g}^{-1}$ ) and copper (◇ 20, △ 80, ○ 320, □ 960  $\mu\text{g g}^{-1}$ ). Error bars (shown for the highest concentration only) represent standard errors for 12 replicate experiments at each concentration and exposure time

Analogous to uptake from the dissolved phase, the uptake of Cd and Cu from the sediments increased with increasing metal concentration in the sediments. This indicates that metal uptake was not limited by the number of metal uptake sites in the organisms' digestive systems. In contrast to uptake from water, the uptake rate of *Melita plumulosa* from the sediment phase was greater for Cd ( $0.12 \text{ g g}^{-1} \text{ d}^{-1}$ ) than for Cu ( $0.023 \text{ g g}^{-1} \text{ d}^{-1}$ ). This may reflect the stronger binding to sediment particles of Cu (i.e.  $K_{ds}$  were  $10^5 \text{ l kg}^{-1}$  for Cu and  $10^3 \text{ l kg}^{-1}$  for Cd). However, the rapid uptake of Cu by *M. plumulosa* during the first 2 h of exposure was ignored in the calculation of the uptake rate constant. It was assumed that the Cu accumulated during this period was non-specific surface binding (e.g. to the body surface). Further studies are required to determine whether ignoring this fraction may have contributed to the low uptake rate constant calculated for Cu. For *Tellina deltoidalis*, the uptake rate of Cu from the sediment phase ( $0.034 \text{ g g}^{-1} \text{ d}^{-1}$ ) was greater than that of Cd ( $0.017 \text{ g g}^{-1} \text{ d}^{-1}$ ). Differences in organism physiology (e.g. gut chemistry) are likely to have contributed to the differences in metal uptake by the 2 species.

Although efflux rates are commonly considered to be the same following uptake from dissolved and particu-

late sources, individual efflux rates were calculated for each metal source in this study. Efflux rate constants ( $k_{e-s}$ , %  $\text{d}^{-1}$ , mean  $\pm$  SD), calculated using the data (5 experiments, 60 organisms) from the 7.5 to 24 h depuration period following 24 h sediment uptake experiments, were  $19 \pm 3$  (Cd) and  $31 \pm 4$  (Cu) for *Melita plumulosa*, and  $17 \pm 3$  (Cd) and  $20 \pm 4$  (Cu) for *Tellina deltoidalis*. The efflux rates did not appear to be dependent on metal concentrations in the sediments, but were approximately twice as great as the efflux rates following water exposures and were also more variable between individual organisms. Incomplete excretion of particles may have contributed to these differences to some extent. For both species, efflux rates were greater for Cu than Cd.

#### Metal assimilation efficiency following uptake of sediment and algae

In the metal assimilation experiments, short uptake periods were used before starting depuration so that metal efflux

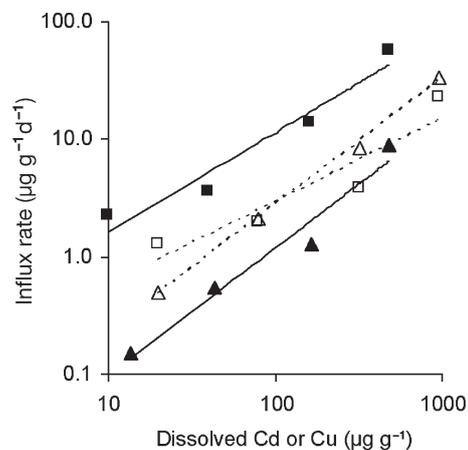


Fig. 4. *Melita plumulosa* and *Tellina deltoidalis*. Influx rates of (■, ▲) cadmium and (□, △) copper into (■, □) *M. plumulosa* and (▲, △) *T. deltoidalis* at different sediment metal concentrations. The relationships describing metal influx rate ( $I_s$ ) and metal concentrations in the dissolved phase were:  $I_s = 0.233[\text{Cd}]^{0.845}$  ( $r^2 = 0.94$ ) and  $I_s = 0.114[\text{Cu}]^{0.703}$  ( $r^2 = 0.86$ ) for *M. plumulosa*, and  $I_s = 0.008[\text{Cd}]^{1.09}$  ( $r^2 = 0.96$ ) and  $I_s = 0.0192[\text{Cu}]^{1.08}$  ( $r^2 = 1.00$ ) for *T. deltoidalis*. The calculated uptake rate constants ( $k_{u-s}$ ,  $\text{g g}^{-1} \text{ d}^{-1}$ ) from the dissolved phase were 0.115 (Cd) and 0.0226 (Cu) for *M. plumulosa*, and 0.0172 (Cd) and 0.0341 (Cu) for *T. deltoidalis*, calculated by linear regression of  $I_s$  and  $C_s$  without log-transformation

was primarily from excretion of ingested materials, rather than efflux of metals that had been assimilated into the organism tissues over longer time periods.

For *Melita plumulosa*, the excretion of non-assimilated material appeared to be virtually complete within 24 h, while for *Tellina deltoidalis*, the excretion of non-assimilated material appeared to be incomplete after the same time period (Fig. 5). The dissection of *T. deltoidalis* at the completion of the experiments indicated that ~14% of the metal uptake comprised of sediment trapped within the shell (not accumulated in tissues). The retention of this material within the shell may have contributed to the slow efflux of metals. The exposure duration (2 to 6 h for *M. plumulosa* and 4 to 8 h for *T. deltoidalis*) did not affect the percentage of metal retained by each organism.

The assimilation efficiency (AE) of *Melita plumulosa* was calculated as the percent of metal retained by the organism after 24 h of depuration. For *Tellina deltoidalis*, an adjustment factor was necessary to account for non-assimilated material that was expected to be egested after the 24 h depuration period. The AE of *T. deltoidalis* was calculated as  $0.75 \times$  the percent of metal retained by the organism after 24 h of depuration. This adjustment factor was based on previous studies of metal assimilation by clams that indicated excretion of ingested particles was complete after 3 d (Lee & Louma 1998, Griscom & Fisher 2002, Griscom et al. 2002).

Calculated metal AEs (% of ingested material, mean  $\pm$  SD) for sediments were  $38 \pm 10\%$  (Cd) and  $7.8 \pm 3\%$  (Cu) for *Melita plumulosa*, and  $28 \pm 3\%$  (Cd) and  $30 \pm 2\%$  (Cu) for *Tellina deltoidalis*. Calculated metal AEs for algae were  $56 \pm 4\%$  (Cd) and  $33 \pm 5\%$  (Cu) for *M.*

*plumulosa*, and  $73 \pm 3\%$  (Cd) and  $49 \pm 4\%$  (Cu) for *T. deltoidalis*. For both organisms, AEs from algae were greater than those from sediments. Efflux rate constants ( $k_{e-A}$ , % d<sup>-1</sup>, mean  $\pm$  SD), calculated following algae uptake experiments, were  $9.8 \pm 1$  (Cd) and  $16 \pm 4$  (Cu) for *M. plumulosa*, and  $5.0 \pm 2$  (Cd) and  $4.3 \pm 1$  (Cu) for *T. deltoidalis*.

### Influence of food type on metal assimilation

These experiments were undertaken to determine whether the presence of algae would affect the rate of metal uptake by *Melita plumulosa* from sediments. The accumulation of metals by *M. plumulosa* following exposure to Cd- or Cu-radiolabelled sediments in the absence and presence of algae, and Cd- or Cu-radiolabelled algae in the absence and presence of sediments is shown in Fig. 6. The rate of accumulation of both Cu and Cd from sediments was greater when algae were present as an additional food source. The presence of sediments increased the rate of accumulation of Cd from algae, but had little effect on the rate of accumulation of Cu. These observations indicate that *M. plumulosa* fed more rapidly, but not necessarily selectively, in the presence of algae. Stress to *M. plumulosa* that was attributable to the physical conditions in the test chambers was not expected to contribute to the differences in feeding rate, as the well conditions were very similar with and without algae. The redistribution of Cd, but not Cu, from algae to sediments may have resulted in an apparent increased rate of accumulation of Cd from algae in the presence of sediments.

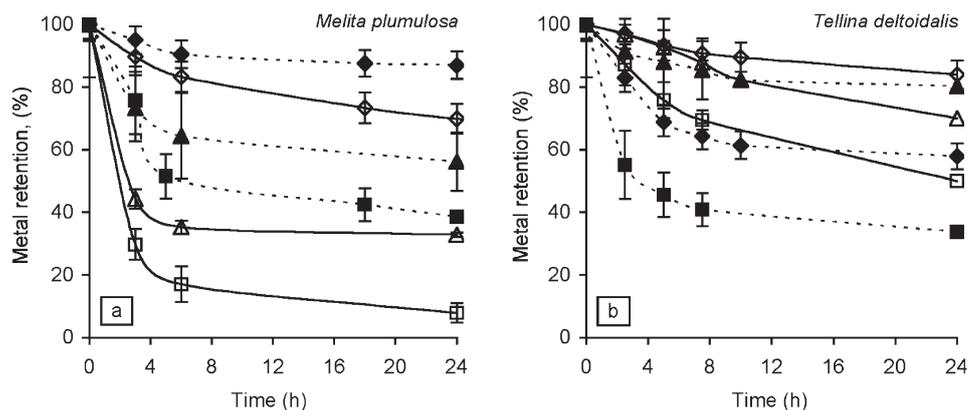


Fig. 5. *Melita plumulosa* and *Tellina deltoidalis*. Metal retention by (a) *M. plumulosa* and (b) *T. deltoidalis* following exposure to (◆, ■, ▲ ---) cadmium and (◇, □, △ —) copper from water (◆, ◇), sediment (■, □) and algae (▲, △) sources. Values are means of all experiments for waters (6 treatments), sediments (7) and algae (3), respectively where each treatment had 9 to 12 individuals per treatment. Error bars represent standard errors

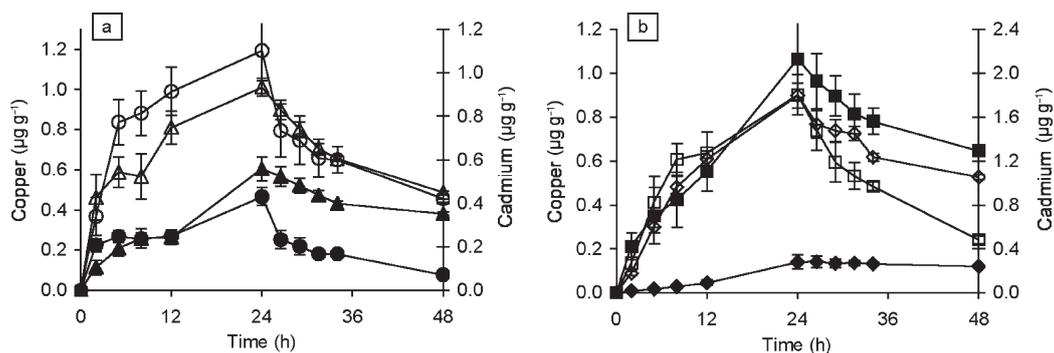


Fig. 6. *Melita plumulosa*. Metal accumulation by *M. plumulosa* following exposure to (a) Cd- (▲, △), or Cu- (●, ○) radiolabelled sediments and in the (●, ▲) absence and (○, △) presence of algae, and (b) Cd- (◆, ◇), or Cu- (■, □) radiolabelled algae and in the (◆, ■) absence and (◇, □) presence of sediments. Error bars represent standard errors for 12 replicates for each experiment

For these experiments, metal retention following the uptake period was calculated by the procedure used for calculating AEs; however, the uptake period was 24 h compared to 2–8 h for the AE determinations. The percent retention of metals from sediments increased in the presence of algae for Cu ( $17 \pm 15\%$  for sediments only and  $38 \pm 6\%$  for sediment and algae), but decreased for Cd ( $63 \pm 5\%$  and  $48 \pm 3\%$ , respectively). This may indicate that the presence of the algae food source stimulates the retention (assimilation) of Cu (essential element), while decreasing the retention of Cd (non-essential element). The percent retention of metals from algae decreased in the presence of sediments for both Cu ( $61 \pm 6\%$  for algae only and  $27 \pm 7\%$  for algae and sediments) and Cd ( $87 \pm 11\%$  and  $59 \pm 4\%$ , respectively). Redistribution of metals between water, digestive fluids, and algae and sediment is expected to affect the metal bioavailability and assimilation efficiency. The ingestion of both algae and sediments may affect metal assimilation by altering the availability, or residence time, of metals in the organisms' digestive tract.

## DISCUSSION

### Biokinetic model assumptions and limitations

All the parameters determined for constructing a biokinetic model of the Cd and Cu exposure pathways for *Melita plumulosa* and *Tellina deltoidalis* are shown in Table 1. Because the influx rate from sediments can be expressed as,  $I_S = AE_S \times IR \times C_S$ , the ingestion rate of the organisms was estimated from  $IR = I_S / (AE_S \times C_S)$ , using data from the metal uptake experiments for sediments with different metal concentrations. The calculated IRs appeared to be independent of metal concentration, the largest error coming from the AE determinations. IR values were  $\sim 0.2$  and  $\sim 0.1 \text{ g g}^{-1} \text{ d}^{-1}$  for *M. plumulosa* and *T. deltoidalis*, respectively. Wang et al. (1999) reported ingestion rates of 0.27 and  $0.42 \text{ g g}^{-1} \text{ d}^{-1}$  for the mussel *Mytilus edulis* and the copepod *Temora longicornis*, respectively.

During the water, sediment and algae exposure experiments, Cd and Cu concentrations in the wells declined by up to 7% with *Melita plumulosa* and by up to 40% with *Tellina deltoidalis* over the 24 h exposure

Table 1. *Melita plumulosa* and *Tellina deltoidalis*. Biokinetic model parameters when exposed to Cd and Cu from water, sediment and algae. Results are mean  $\pm$  standard deviation.  $k_{u-W}$  and  $k_{e-W}$  are the uptake and efflux rate constants from the dissolved phase, respectively;  $AE_S$  and  $AE_A$  are the assimilation efficiency from sediments and algae phases, respectively; IR is the organism's ingestion rate;  $k_{e-S}$  and  $k_{e-A}$  are the metal efflux rates following uptake from sediment and algae food phases, respectively

Parameter	$k_{u-W}$ ( $\text{l g}^{-1} \text{ d}^{-1}$ )	$k_{u-S}$ ( $\text{g g}^{-1} \text{ d}^{-1}$ )	$k_{e-W}$ ( $\% \text{ d}^{-1}$ )	$k_{e-S}$ ( $\% \text{ d}^{-1}$ )	$k_{e-A}$ ( $\% \text{ d}^{-1}$ )	$AE_S$ (%)	$AE_A$ (%)	IR ( $\text{g g}^{-1} \text{ d}^{-1}$ )
<i>M. plumulosa</i> , Cd	$0.028 \pm 0.03$	$0.12 \pm 0.02$	$6.0 \pm 0.6$	$19 \pm 3$	$9.8 \pm 1$	$22 \pm 10^a$	$56 \pm 4$	$0.25 \pm 0.15$
<i>M. plumulosa</i> , Cu	$0.12 \pm 0.02$	$0.023 \pm 0.02$	$16 \pm 0.8$	$31 \pm 4$	$16 \pm 4$	$7.8 \pm 3$	$33 \pm 5$	$0.15 \pm 0.05$
<i>T. deltoidalis</i> , Cd	$0.012 \pm 0.01$	$0.017 \pm 0.03$	$8.5 \pm 0.5$	$17 \pm 3$	$5.0 \pm 2$	$25 \pm 3^a$	$73 \pm 3$	$0.07 \pm 0.04$
<i>T. deltoidalis</i> , Cu	$0.19 \pm 0.01$	$0.034 \pm 0.02$	$11 \pm 1$	$20 \pm 4$	$4.3 \pm 1$	$30 \pm 2$	$49 \pm 4$	$0.10 \pm 0.03$

<sup>a</sup>Adjusted for contribution of Cd desorbed into waters during sediment exposures

period. The decline was attributed mostly to metal uptake by the organisms. While the decline with *M. plumulosa* was small and should not affect the developed model, the concentration declines with *T. deltoidalis* may have resulted in an underestimation of the metal accumulation rates. The use of metal uptake data from the 2 to 10 h exposure period reduced the effect of the declining exposure concentrations on the calculated uptake rates. Because of the large variation in the decline observed for each exposure period (e.g. 10 to 40% for algae), no adjustment was made for concentration decline that occurred during the exposure experiments, and nominal concentrations were used in the development of the biokinetic model.

During the 24 h exposure of *Melita plumulosa* and *Tellina deltoidalis* to metals from sediment and algal sources, the concentration of dissolved metals in the exposure wells may also have contributed to the measured metal accumulation. Measurements of dissolved Cu and Cd in the sediment and algae exposure wells indicated that while copper concentrations were low ( $K_{ds} \approx 10^5 \text{ l kg}^{-1}$ ), the dissolved cadmium concentrations were high ( $K_{ds} \approx 2 \text{ to } 9 \times 10^3 \text{ l kg}^{-1}$ ). According to preliminary biokinetic models, the copper in the water of the sediment and algae exposure wells would have contributed less than 1% of the total copper accumulated by either species. For cadmium experiments, the preliminary biokinetic models indicated that the exposure well water would have contributed ~30% of the total cadmium accumulated by *M. plumulosa* and ~10% of that by *T. deltoidalis* in sediment exposures, and ~5% for *M. plumulosa* and ~1% for *T. deltoidalis* in algae exposures.

There is no perfect way to separate uptake of the sediment-bound (or algae-bound) metal from that desorbed into the water. While for copper in sediment and algae exposures, and for cadmium in algae exposures, the contribution from water was small, an adjustment of the biokinetic model parameters is necessary to account for the 10 to 30% contribution of dissolved Cd in the sediment exposures. To correct for these contributions, the biokinetic model was used to calculate the amount of cadmium that was accumulated from the water in the sediment exposure wells. According to Eq. (2), metal accumulation from the sediment phase is represented by  $AE_S \times IR \times C_S/k_{e-S}$ , and the AE that was calculated with the assumption that the contribution of cadmium from water was negligible during sediment exposures will have been overestimated in the preliminary model. Adjustment of the AE for sediments from 38 to 22% for *Melita plumulosa*, and from 28 to 25% for *Tellina deltoidalis*, accounts for the overestimation associated with Cd desorbed into the water. The final model parameters are shown in Table 1. A larger adjustment was needed for *M. plumulosa* because the uptake rate constant of cadmium from the dissolved phase

( $k_{u-W}$ ,  $\text{l g}^{-1} \text{ d}^{-1}$ ) was larger for *M. plumulosa* (0.028) than for *T. deltoidalis* (0.012).

The efflux rates derived following exposure to water, sediment and algal metal sources are likely to be overestimated due to the relatively recent uptake of the metal ( $\leq 24 \text{ h}$ ) and the relatively short period over which metal efflux was observed. The use of a longer depuration period was prevented by the short half-life of copper and constraints on laboratory use. Longer exposure times would be expected to result in metals being bound more strongly and distributed among a wider range of tissue compartments, with metal efflux rates being slower than those derived in this study. The influx rate of metals to the organisms will be modified by organism behaviour (filtration versus ingestion), changes in metal partitioning (sediment type, sediment disturbance) and the assimilation efficiency of different ingested materials (sediment, algae, detritus). The net accumulation of metals by the organisms is likely to be affected to a greater extent by factors that alter influx rate than by the uncertainty associated with the long-term efflux rate.

Two models were developed to account for the accumulation of metals from sediment phases. The first model was developed in the same manner as the model for water, i.e. exposure of the organisms to a Cd and Cu concentration series in sediments, and allowed the calculation of an influx rate constant from the sediment phase,  $k_{u-S}$ . The second method involved determining the metals' assimilation efficiency (AE) following the ingestion of a known amount of sediment. Comparison between the 2 models gave a ratio (Model 1/Model 2) of 1.5 for Cd and Cu uptake by *Melita plumulosa* and Cu uptake by *Tellina deltoidalis*, but a ratio of 0.7 for Cd uptake by *T. deltoidalis*. The first model assumed that the metal accumulated during the exposure period had been assimilated by the organism. This may have overestimated metal uptake as the gut passage times of the organisms are not believed to be sufficiently short for this assumption to hold. The AE-based model for metal accumulation from sediment phases was used for the modelling calculations presented in the next section. This was consistent with the approach used for calculating metal accumulation from algae.

#### Effect of sediment properties on metal exposure pathways

It is well recognised that partitioning between sediments and pore waters is greatly affected by sediment properties (Besser et al. 2003, Simpson et al. 2004). In estuarine/marine environments, cadmium exhibits a greater solubility than copper due to strong complexation by chloride, and for oxic surface sediments,  $K_d$

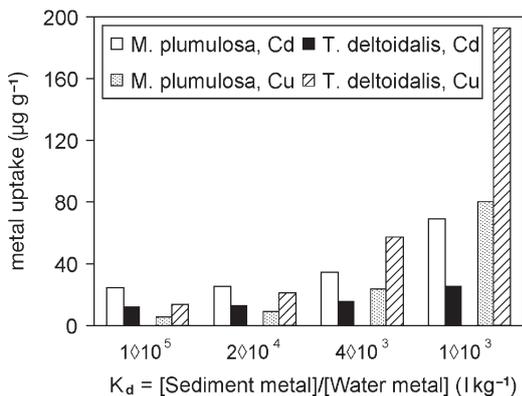


Fig. 7. *Melita plumulosa* and *Tellina deltoidalis*. Calculated accumulation of Cd and Cu in *M. plumulosa* and *T. deltoidalis* resident in sediments containing 100 µg g<sup>-1</sup> Cd and Cu sediment properties resulting in  $K_d$ s of 1 × 10<sup>5</sup>, 2 × 10<sup>4</sup>, 4 × 10<sup>3</sup> and 1 × 10<sup>3</sup> l kg<sup>-1</sup>

values are typically within an order of magnitude of 10<sup>3</sup> l kg<sup>-1</sup> for cadmium and 10<sup>4</sup> to 10<sup>5</sup> l kg<sup>-1</sup> for copper (Stumm & Morgan 1996). The distribution of metals between sediments and pore waters or overlying waters can be described by a partitioning coefficient,  $K_d = [\text{sediment}]/[\text{water}]$  (in l kg<sup>-1</sup>).  $K_d$  is influenced by a variety of factors, including (1) the speciation of the metals in the sediments (e.g. metal binding to acid volatile sulphide, particulate organic matter and iron, and manganese oxyhydroxides – influenced by redox potential,  $E_h$ ), (2) ions that compete for metal binding sites (e.g. H<sup>+</sup> [pH], Ca<sup>2+</sup>, Mg<sup>2+</sup> [hardness/salinity], Fe<sup>2+</sup>, Mn<sup>2+</sup> [suboxic sediments]) and (3) metal-complexing ligands (e.g. dissolved organic carbon, chloride). Disturbances to the sediments (by organisms or laboratory manipulation) will also affect partitioning. Mixing will cause changes in the oxidation state of previously redox-stratified sediment components (Fe(II)/Fe(OH)<sub>3</sub>/ FeS) and affect  $K_d$  through the subsequent reactions of these new phases (Simpson & Batley 2003, Simpson et al. 2004).

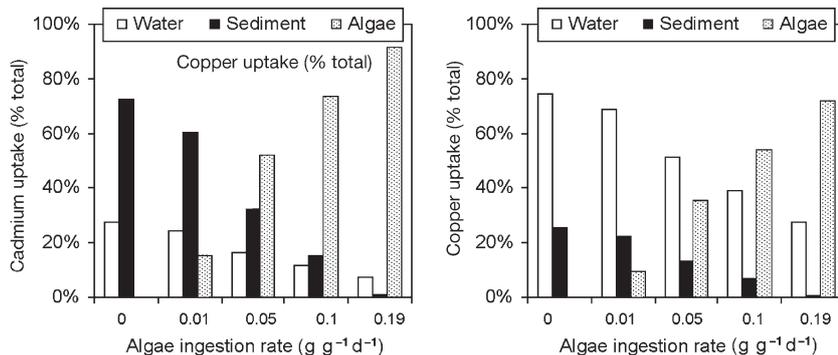


Fig. 8. *Melita plumulosa*. Effect of ingestion source on uptake pathways of Cd and Cu by *M. plumulosa* for a total ingestion rate of 0.2 g 'solid' g<sup>-1</sup> organism d<sup>-1</sup>, where the solid is either sediment or algae each with 100 µg g<sup>-1</sup> Cd or Cu. Dissolved Cd and Cu concentrations were 20 µg l<sup>-1</sup> ( $K_d = 5 \times 10^3$  l kg<sup>-1</sup>)

For sediments containing total metal concentrations of 100 µg g<sup>-1</sup> (Cd, Cu), sediment properties resulting in  $K_d$ s of 1 × 10<sup>5</sup>, 2 × 10<sup>4</sup>, 4 × 10<sup>3</sup> or 1 × 10<sup>3</sup> l kg<sup>-1</sup> would correspond to pore water metal concentrations of 1, 5, 25, or 100 µg l<sup>-1</sup> (Cd, Cu). Fig. 7 shows the steady-state body concentrations predicted from the biokinetic models for *Melita plumulosa* and *Tellina deltoidalis* resident in these sediments. For each species, the concentration of Cd or Cu accumulated from the sediment exposure pathway (41 and 13 µg g<sup>-1</sup> Cd, 5.0 and 12 µg g<sup>-1</sup> Cu for *M. plumulosa* and *T. deltoidalis*, respectively), is independent of  $K_d$ . However, because pore water metal concentrations increase as the  $K_d$  decreases, the amount of metal accumulated by the organisms also increases as the  $K_d$  decreases. The greatest effect of the changes in sediment properties is on the accumulation of Cu by *T. deltoidalis*, as this species has a high uptake rate constant for Cu from the dissolved phase ( $k_{u-w}$ ) compared to the corresponding efflux constant ( $k_{e-w}$ ). The accumulation of Cd by *T. deltoidalis* is least affected by sediment properties, due to the low uptake rate constant for Cd from the dissolved phase (Table 1).

With regard to the sensitivity of *Melita plumulosa* and *Tellina deltoidalis* to metal-contaminated sediments, if it is assumed that Cd and Cu accumulated from water and sediments cause the same effects in the organisms (Simpson & King 2005), then changes in sediment properties that affect metal partitioning would be expected to affect the toxicity of metals to the organisms, particularly the toxicity of copper to *T. deltoidalis*. Careful scrutiny should be given to metal partitioning in sediments when data are to be used to estimate toxicity thresholds or effects of concentrations, before such values are adopted in sediment quality guidelines. The use of poorly equilibrated metal-spiked sediments, that have unrealistically high pore water metal concentrations compared to naturally contaminated sediments, will greatly overestimate the sensitivity of the species to field-collected sediments with the same total metal concentration.

### Effect of food quality on metal exposure pathways

Experiments indicated that the presence of algae (as a food source) has a large effect on the feeding behaviour and metal uptake of *Melita plumulosa* (not investigated for *Tellina deltoidalis*). The effect that food quality (ingestion source) may have on uptake pathways of Cd and Cu by *M. plumulosa* is shown in Fig. 8. These calculations assumed that *M. plumulosa* has a total ingestion rate of  $0.2 \text{ g 'solid' g}^{-1} \text{ organism d}^{-1}$ . For both Cd and Cu, the assimilation efficiency of ingested materials was much greater for algae than for sediments. Consequently, the accumulation of the metals by *M. plumulosa* is predicted to increase from 33 to  $119 \mu\text{g g}^{-1}$  Cd and from 20 to  $54 \mu\text{g g}^{-1}$  Cu as the solid ingested changes from 100% sediment to 100% algae (each with  $100 \mu\text{g g}^{-1}$  Cd or Cu), respectively. The use of the biokinetic model for this purpose can only be considered as qualitative, due to assumptions regarding ingestion rates and assimilation efficiencies. Past studies have found that assimilation efficiency changes with food availability and quality (Wang & Wong 2003). An alternative scenario to that shown in Fig. 8 is sediment where the algae are not contaminated with metals. For this scenario, a shift from feeding exclusively on sediments to feeding predominantly on algae would result in a decrease in the accumulation of metals by *M. plumulosa*.

Assumptions about organism feeding behaviour (food choice and feeding rate) will probably result in the greatest uncertainty in predicting metal accumulation by sediment-dwelling organisms in natural environments. The deposition of clean surface sediments over contaminated deeper sediments has major implications for the assessment of contaminated sites. Many benthic organisms may create burrows into the deeper (contaminated) sediments with burrow linings that protect them from contaminant exposure. The feeding of these organisms on newly deposited surface materials may greatly reduce contaminant exposure and toxicity.

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