

# Metal (Fe, Zn, Cu, Pb and Cd) concentration patterns in components of a macrophyte-based coastal lagoon ecosystem

Theodora Boubonari · Theodoros Kevrekidis · Paraskevi Malea

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**Abstract** Information on the metal biological fate in macrophyte-based coastal lagoons is provided; this information can contribute to the assessment of the environmental effects of metal pollution and to the development of predictive models for rational management of coastal lagoons. Iron, Zn, Cu, Pb and Cd concentrations in the dominant invertebrate and fish species of Monolimni Lagoon, Mediterranean Sea, as well as in potential major sources for metal accumulation in these animals (water, sediments, angiosperms, seaweeds), were measured. Principal Component Analysis (PCA) was conducted using metal concentrations in invertebrates and fishes. All five metal concentrations loaded significantly on the first PCA axis; however, Zn and Cu loadings were less significant than Cd and even less than Fe and Pb ones. The samples of deposit-feeding invertebrates were separated from those of the rest of the organisms (browsing, herbivorous and carnivorous invertebrates, carnivorous gobies and muscle tissues of detritivorous mullets)

along the first PCA axis. Deposit-feeding invertebrates displayed the highest Fe and Pb contents, and in general, the highest or comparatively high Cd, Zn and Cu ones. Carnivorous gobies showed comparatively high Zn contents and carnivorous shrimps the highest Cu ones, while muscle tissues of detritivorous mullets had low metal loads. In addition, there was no essential increase in metal concentrations corresponding to the increasing trophic level (autotrophs, to herbivores, to carnivores). Our findings suggest that (a) the variability in Fe, Pb and Cd contents in invertebrates and gobies depends at least to some extent on interspecific differences in feeding habits—deposit feeders accumulated the highest metal amounts probably due to high rates of uptake from sediments, (b) the variability in Zn and Cu concentrations in these organisms depends also on other interspecific differences apart from those in feeding habits, (c) metal accumulation in mullet muscle tissues does not depend markedly on feeding habits and (d) the trophic transfer of macrophyte-bound metals to the coastal lagoon food web is of relatively minor importance.

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T. Boubonari · T. Kevrekidis (✉)  
Laboratory of Environmental Research and Education,  
Democritus University of Thrace, Nea Hili,  
68100 Alexandroupolis, Greece  
e-mail: tkebreki@eled.duth.gr

P. Malea  
Department of Botany, School of Biology, University of  
Thessaloniki, P.O. Box 109, 54124 Thessaloniki, Greece

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## Introduction

Coastal lagoons constitute a common environment around the world. Three types of coastal lagoons are

distinguished according to the dominance of autotrophic populations: the phytoplankton-based lagoons, the macrophyte- and macroalgal-based ones and the algal mat-based systems (Knoppers, 1994). Coastal lagoons are highly productive and ideal systems for aquaculture projects, but frequently are heavily impacted by anthropogenic inputs and human activities (Kjerfve, 1994). Many coastal lagoons receive amounts of contaminants from industrial, domestic and agricultural sources; among the many anthropogenic contaminants, heavy metals are of greatest environmental concern (Lacerda, 1994). Heavy metals are extremely persistent in aquatic environments and tend to bioaccumulate; they are toxic to organisms above threshold availability and at elevated concentrations can adversely affect the structure and function of biotic communities (Kennish, 1997).

Although anthropogenic input of heavy metals to lagoons is a potentially serious problem, scientific knowledge concerning the accumulation of heavy metals in coastal lagoon and their effects on biota is limited. Past efforts have focused on metal accumulation in one or a few species of the same taxonomic group, particularly in bivalves (e.g. Niencheski et al., 2001; Pérez et al., 2001; Caliceti et al., 2002; Otc here et al., 2003); in several cases, metal concentrations in the lagoon environment, mainly in sediments, have been simultaneously assessed in addition to those in organisms (e.g. Marmolejo-Rivas & Páez-Osuna, 1990; Fernandes et al., 1994; Cheggour et al., 2001). A few studies have determined metal concentrations in the environment and in some macrophyte and macroinvertebrate or macroinvertebrate and fish species (Capone et al., 1983; Sfriso et al., 1995; Storelli & Marcotrigiano, 2001). In addition, the concentrations of heavy metals in abiotic and several biotic components of the lagoon ecosystem, such as the dominant macrophyte, macroinvertebrate and fish species, and even more metal transfer processes between ecosystem components have not been studied in detail. A better understanding of this subject could contribute to the assessment of the environmental effects of metal pollution and the development of predictive models for rational management of coastal lagoons.

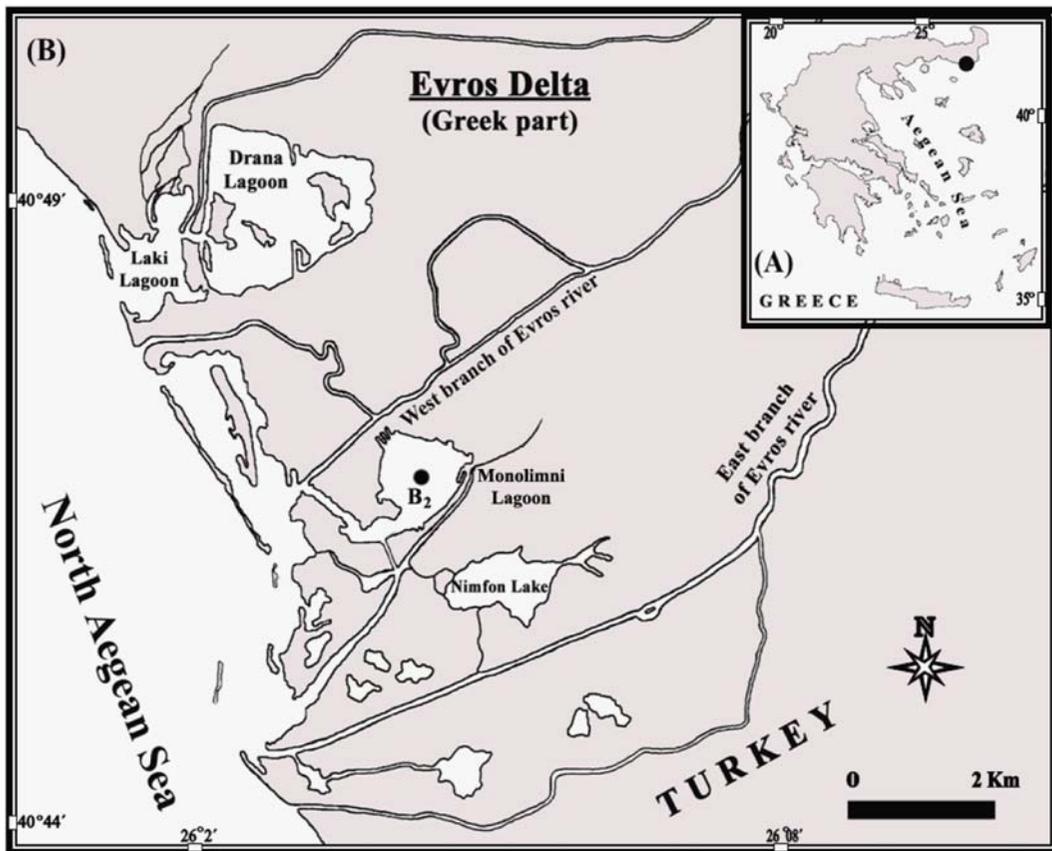
The main goal of this study is to provide information on the metal biological fate in macrophyte-based coastal lagoons. The patterns of Fe, Zn, Cu, Pb and Cd concentrations in the dominant macrozoobenthic and

fish species of Monolimni Lagoon (Evros River Delta, Northern Aegean Sea) were determined and compared; metal concentrations in potential major sources for metal accumulation in these organisms (water column, sediments, angiosperm and dominant macroalgae species) were additionally determined. Our data could be used as a basis for speculation on metal transfer processes. Provided that (a) several marine and estuarine organisms are poor regulators of heavy metals, mainly of the non-essential ones, and thereby, accumulate them almost in proportion to metal concentrations in the surrounding medium (Amiard et al., 1987; Clark, 2002), (b) bottom sediments in estuaries and coastal lagoons contain several times the concentration of heavy metals found in overlying waters (Lacerda, 1994; Kennish, 1997) and (c) sediment is a major route of metal uptake by several aquatic animals, mainly by deposit feeders (Zingde et al., 1976; Wang & Fisher, 1999), we predict that the variability in most metals' concentrations in coastal lagoon macroinvertebrate and fish species largely depends on interspecific differences in ecology and feeding habits, deposit feeders accumulating high metal amounts. We mainly explore whether field data support this hypothesis.

## Materials and methods

### Study area

Monolimni (or Paloukia) Lagoon (Evros River Delta, Northern Aegean Sea) occupying an area of about 112 ha and communicating with the coastal section of the Evros River Delta through a 15-m wide opening (Fig. 1) is a typical, relatively enclosed macrophyte-based Mediterranean coastal lagoon. The maximum depth at its main part, the innermost section, is about 60 cm; salinity near the bottom varied between 0.3 and 5.7 psu, and the sediment was very fine sand during February 1998 to February 1999 (Kevrekidis, 2004). This part of the lagoon harboured a perennial population of the cosmopolitan submerged angiosperm *Ruppia maritima* L., and occasionally, macroalgae, mainly the green seaweed *Ulva rigida* C. Agardh (Malea et al., 2004). The mudsnail *Ventrosia maritima* (Milaschewitch, 1916), the bivalve *Abra segmentum* (Recluz, 1843), the amphipods *Corophium orientale* Schellenberg, 1928 and *Gammarus aequicauda*



**Fig. 1** (A) Geographical location of the study site. (B) Map of the Evros River Delta showing the sampling station (B<sub>2</sub>)

(Martynov, 1931) and the polychaete *Hediste diversicolor* (O. F. Müller, 1776) dominated the macrozoobenthic assemblage; the shrimp *Crangon crangon* (Linnaeus, 1758) and the goby *Knipowitschia caucasica* Kawrajsky, in Berg, 1916 were the dominant species among epibenthic decapods and small fishes (Kevrekidis, 2004). The main exploitation pattern occurring in this lagoon is the extensive culture of mullets, particularly of *Mugil cephalus* Linnaeus, 1758 and *Liza aurata* Risso, 1810 (Kevrekidis, 2004). Amounts of heavy metals, mostly transported by Evros River, reached the coastal section of its delta during May 1998 to May 1999 (Boubonari et al., 2008) and possibly Monolimni Lagoon.

#### Sample collection

Samples were collected seasonally (May, August and October 1998, and January and May 1999) at the central part of the innermost section of Monolimni

Lagoon (site B<sub>2</sub>, 40°46' N, 26°03' E) (Fig. 1). Late spring and summer and mid-autumn and winter were chosen as the most representative periods of the annual cycle of several biotic components (see Kevrekidis, 2004; Malea et al., 2004). Each time, at least three replicate samples of the following components were collected: (a) of water using acid-washed plastic vessels, (b) of surface sediments using an 8-cm diameter plastic corer penetrating to a depth of 10 cm, (c) of macroalgae using a 25 × 25 cm plastic frame and (d) of *R. maritima* and of macrozoobenthos using a 20-cm diameter plastic corer penetrating to a depth of 20 cm; in addition, two replicate samples of epibenthic decapods and small fish using a special nylon net (with a 40 × 40 cm opening) pulled on the sediment for a distance of 10 m were collected. Macrozoobenthos samples were sieved through a 1-mm plastic screen and all biological samples were washed in lagoon water. Replicate samples were pooled and placed in plastic vessels. Mulletts were also

collected using a special nylon net. All procedures were performed wearing plastic gloves. Samples were transported to the laboratory in the same day of capture.

#### Sample pretreatment and analysis

Lagoon water samples were acidified (1.5 ml l<sup>-1</sup> conc. HNO<sub>3</sub>) and filtered through an acid-washed glass fibre (0.45 µm). *Ruppia maritima* and the dominant seaweed species (*U. rigida*) were washed in bidistilled water; *R. maritima* leaves were separated from the rest of the plant fractions. After a 36-h purging period in habitat water, individuals of the dominant macrozoobenthic species (*H. diversicolor*, *A. segmentum*, *V. maritima*, *C. orientale* and *G. aequicauda*) and of the dominant epibenthic decapod and small fish ones (*C. crangon*, *K. caucasica*) (at least 30 individuals of each species per sample) were rinsed in bidistilled water and the shell of bivalves was removed. Three to ten individuals comprised a sample of each of the dominant mullet species (*M. cephalus*, *L. aurata*). They were washed in bidistilled water and measured for total length (length ranged from 27.2 to 44.5 cm for *M. cephalus* and from 24.5 to 33.4 cm for *L. aurata*); a portion of the muscle tissue was taken from the left side of each fish between the pelvic and dorsal fins. All samples were frozen (-20°C) until analysis.

After being dried to a constant weight (80°C), sediment samples were sieved through a nylon net with mesh size of 63 µm; the <63 µm sediment fraction (silt and clay) was used in the following procedures. Biological samples were dried to a constant weight (80°C) and ground in an agate mill; the powdered muscle tissue of each mullet specimen, at least three subsamples of each of the other powdered biotic ecosystem components samples (a total of nine components) and at least three subsamples of each of the powdered sediment samples were digested in Teflon vessels for 12 h with HNO<sub>3</sub>/HClO<sub>4</sub> (4/1). The above procedures have been frequently applied in previous studies (see Boubonari et al., 2008).

The total Fe and Zn concentrations in sediments and in biota and the dissolved Fe and Zn concentrations in the lagoon water were measured with flame Atomic Absorption Spectrophotometry (AAS Perkin-Elmer 2100); the limits of detection for the analysis of

Fe and Zn in sediments and biota were 500 ng g<sup>-1</sup> dry wt and 50 ng g<sup>-1</sup> dry wt, respectively, and in water, 5 ng ml<sup>-1</sup> and 0.5 ng ml<sup>-1</sup>, respectively. The total Cu, Pb and Cd concentrations in sediments and in biota and the dissolved Cu, Pb and Cd concentrations in the lagoon water were measured using graphite furnace AAS (Perkin-Elmer 2100) with Deuterium background correction; the limits of detection for the analysis of Cu, Pb and Cd in sediments and biota were 2 ng g<sup>-1</sup> dry wt, 5 ng g<sup>-1</sup> dry wt and 0.3 ng g<sup>-1</sup> dry wt, respectively, and in water 0.02 ng ml<sup>-1</sup>, 0.05 ng ml<sup>-1</sup> and 0.003 ng ml<sup>-1</sup>, respectively. Pro-analysis grade reagents (Merck) were used and reagent blank was run concurrently. The accuracy of the technique was checked with the analysis of the standard reference materials of sea lettuce, cod muscle and mussel tissue (*Ulva lactuca* n. 279, cod muscle n. 422, mussel tissue n. 278R, Community Bureau of Reference). The precision of the analysis was calculated by the coefficient of variation (CV %), which was calculated by measuring at least three independent subsamples of each sample; CV of the biological samples varied between 1.2 and 16.3% for Fe, between 1.1 and 15.2% for Zn, between 0.5 and 8.6% for Cu, between 1.5 and 15.7% for Pb and between 0 and 19.2% for Cd.

#### Data analysis

Mean values (±standard error) of the metal concentrations of the seasonal samples of each ecosystem component were calculated.

A PCA was conducted using the metal concentrations of the seasonal samples of macroinvertebrate and fish species. The usefulness of PCA was assessed using Kaiser–Meyer–Olkin's measure of sampling adequacy. KMO ranges from 0 to 1, and should be well above 0.5 if variables are very interdependent (Benejam et al., 2008). Variables were normalized, and the PCA was performed using the *princomp* function of the package *stats* of the program 'R' (R Development Core Team, 2006).

The number of principal components for which eigenvalues accounted for a significant amount of total variance was identified by use of an objective randomization procedure based on the eigenvalues (Peres-Neto et al., 2005). The randomization protocol was conducted as follows. We randomized the values within variables in the data matrix and conducted a PCA on the

reshuffled data matrix. Then, we repeated the above procedure a total of 999 times. The  $P$  value for each axis and the observed eigenvalue was estimated as follows: (number of random eigenvalues for axis  $k$  equal to or larger than the observed + 1)/1000.

The contribution of individual variables to significant principal components was assessed using the bootstrapped eigenvector method (Peres-Neto et al., 2003). This method re-samples entire rows from the original data with replacements and computes ordinations for each bootstrapped sample. One thousand bootstrap samples were drawn, and a PCA was performed on each of them. Variables with 95% confidence intervals around their loadings that did not incorporate zero were considered to contribute significantly to the relevant component. For the computations, we used R code snippets provided by Pedro Peres-Neto.

## Results

### Metal concentrations in water, sediments and phytobenthos

Mean ( $\pm$ standard error) Fe, Zn, Cu, Pb and Cd concentrations in the water were  $407 (\pm 76) \mu\text{g l}^{-1}$ ,  $48.4 (\pm 10.4) \mu\text{g l}^{-1}$ ,  $10.0 (\pm 2.4) \mu\text{g l}^{-1}$ ,  $25.1 (\pm 2.9) \mu\text{g l}^{-1}$  and  $0.1 (\pm 0.1) \mu\text{g l}^{-1}$ , respectively. Most metals' concentrations in sediments were relatively higher than those in the plants (Fig. 2). As for the most metals analyzed, *R. maritima* leaves had relatively higher concentrations than *U. rigida* (Fig. 2).

### Metal concentration patterns in invertebrates and fishes

Iron and Pb concentrations in macroinvertebrate and fish species were comparatively lower than those in sediments; in contrast, almost all animals showed higher Cd levels than sediments (Fig. 2). Among the investigated macroinvertebrates and gobies, the deposit-feeding bivalve *A. segmentum* and the deposit-feeding polychaete *H. diversicolor* showed the highest Fe and Pb contents, followed by the selective deposit-feeding amphipod *C. orientale*, while the carnivorous shrimp *C. crangon* and the carnivorous goby *K. caucasica* displayed the lowest ones (Fig. 2). Soft tissues of the deposit-feeding

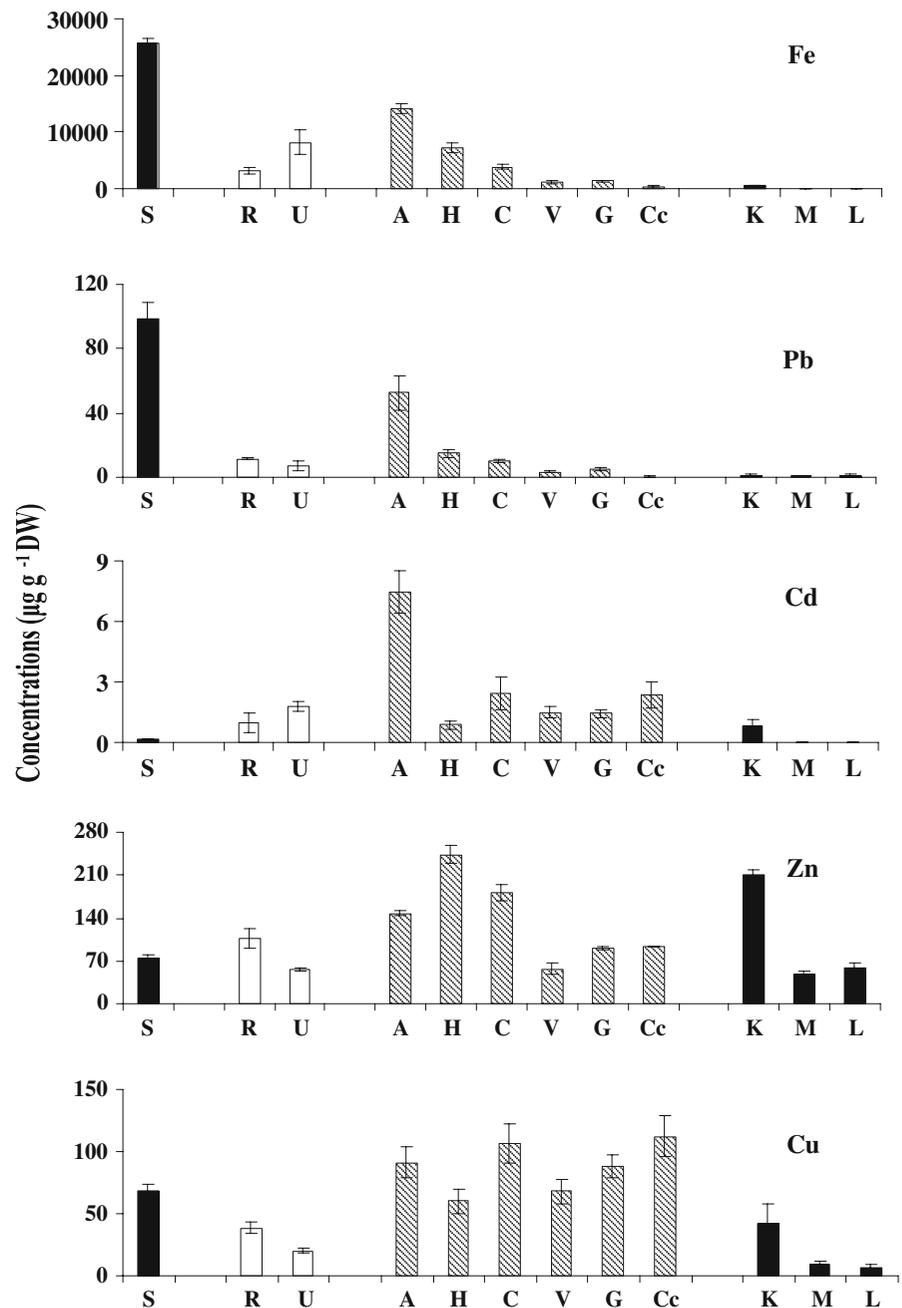
*A. segmentum* also had the highest Cd signatures; cadmium body concentrations showed relatively little interspecific variation in the rest of the organisms (Fig. 2). The deposit-feeding nereid polychaete *H. diversicolor* contained the highest Zn loads, followed by the carnivorous goby *K. caucasica*, the selective deposit-feeding amphipod *C. orientale* and the deposit-feeding bivalve *A. segmentum*, while the carnivorous shrimp *C. crangon* showed the highest Cu contents closely followed by the selective deposit-feeding amphipod *C. orientale*, the deposit-feeding bivalve *A. segmentum* and almost all the rest species (Fig. 2).

Muscle tissues of the detritivorous mullets *M. cephalus* and *L. aurata* showed generally lower metal contents than the macroinvertebrates and the carnivorous goby *K. caucasica* (Fig. 2); especially, Cd concentrations in mullet muscles were below detectable limit.

The KMO index had a value of 0.661 showing the usefulness of the PCA conducted using the metal concentrations in macroinvertebrate and fish species. The randomization procedure based on eigenvalues showed the presence of only one significant axis ( $P < 0.05$ ) that accounts for 61.5% of the variability in the original data. According to the bootstrapped eigenvector method, all five variables were found to load significantly on the first axis ( $P < 0.05$ ), having negative weights (Table 1); however, the Zn and Cu loadings were not significant at the  $P < 0.01$  level and the Cd one at the  $P < 0.001$  level (Table 1). The samples of deposit-feeding invertebrates (*A. segmentum*, *H. diversicolor*) and selective deposit-feeding ones (*C. orientale*) showing negative scores on the first principal component were separated from those of the rest of the organisms, namely of browsing invertebrates (*V. maritima*), herbivorous invertebrates (*G. aequicauda*), carnivorous invertebrates (*C. crangon*), carnivorous gobies (*K. caucasica*) and detritivorous mullets, which almost all showed positive scores on the first principal component (Table 2). In particular, the samples of muscle tissues of the detritivorous mullets *M. cephalus* and *L. aurata* showed the highest positive scores on the first principal component (Table 2).

*Gammarus aequicauda* generally showed relatively low metal concentrations compared to the other investigated macroinvertebrate species (Fig. 2). In addition, the concentrations of most metals

**Fig. 2** Metal concentrations in sediments (*black column*), macrophyte species (*open columns*), macroinvertebrate species (*shaded columns*) and fish species (*black columns*) in Monolimni Lagoon; each column represents the mean value, whiskers represent the standard error. S: sediments; R: *Ruppia maritima* leaves; U: *Ulva rigida*; A: *Abra segmentum* (soft tissues); H: *Hediste diversicolor*; C: *Corophium orientale*; V: *Ventrosia maritima*; G: *Gammarus aequicauda*; Cc: *Crangon crangon*; K: *Knipowitschia caucasica*; M: *Mugil cephalus* (muscle tissues); L: *Liza aurata* (muscle tissues)



analyzed in this herbivorous amphipod (Fe:  $1,404 \mu\text{g g}^{-1}$ ; Pb:  $5.2 \mu\text{g g}^{-1}$ ; Cd:  $1.4 \mu\text{g g}^{-1}$ ; Zn:  $91.9 \mu\text{g g}^{-1}$ ; Cu:  $88.2 \mu\text{g g}^{-1}$ ) were lower than or comparable to mean ( $\pm$ SE) metal concentrations in the dominant autotroph species (*R. maritima*, *U. rigida*) (Fe:  $5,626.7 \pm 2,497 \mu\text{g g}^{-1}$ ; Pb:  $9.2 \pm 2.3 \mu\text{g g}^{-1}$ ;

$\text{g}^{-1}$ ; Cd:  $1.4 \pm 0.4 \mu\text{g g}^{-1}$ ; Zn:  $81.4 \pm 25.7 \mu\text{g g}^{-1}$ ; Cu:  $29.5 \pm 9.3 \mu\text{g g}^{-1}$ ) and higher than or comparable to those in the dominant carnivores (*C. crangon*, *K. caucasica*) (Fe:  $479.8 \pm 53.2 \mu\text{g g}^{-1}$ ; Pb:  $1.0 \pm 0.5 \mu\text{g g}^{-1}$ ; Cd:  $1.6 \pm 0.8 \mu\text{g g}^{-1}$ ; Zn:  $152.2 \pm 59.2 \mu\text{g g}^{-1}$ ; Cu:  $77.4 \pm 34.8 \mu\text{g g}^{-1}$ ).

**Table 1** Loadings on the first principal component and the associated probabilities for the bootstrapped eigenvector

Variable	Loading	<i>P</i>
Fe	−0.530	<0.001
Pb	−0.516	<0.001
Zn	−0.265	<0.05
Cu	−0.341	<0.05
Cd	−0.517	<0.01

**Table 2** The scores of the macroinvertebrate and fish samples on the first principal component

Sample	Score	Sample	Score
A	−4.650	K	0.448
A	−4.457	V	0.498
A	−4.287	G	0.573
A	−2.623	V	0.703
C	−1.693	V	0.765
H	−1.254	Cc	0.791
H	−0.754	V	0.897
C	−0.625	K	0.914
C	−0.462	V	0.948
H	−0.373	K	1.045
C	−0.161	M	1.718
Cc	−0.128	L	1.729
Cc	0.051	L	1.767
G	0.274	M	1.797
K	0.283	M	1.806
G	0.308	L	1.842
G	0.438	M	1.872

For abbreviations see Fig. 2 caption

## Discussion

Mean dissolved Cu concentrations in the study area and to a lesser extent Zn and Pb ones exceeded the European Environmental Quality Standards (EQSs) of waterborne contamination in estuaries, whereas those of Fe and Cd did not [ $<5.0 \mu\text{g l}^{-1}$ ,  $<40 \mu\text{g l}^{-1}$ ,  $<25 \mu\text{g l}^{-1}$ ,  $<1.0 \text{mg l}^{-1}$  and  $<5.0 \mu\text{g l}^{-1}$ , respectively, according to McLusky & Elliott (2004)], suggesting that amounts of heavy metals, most probably transported by Evros River, reached Monolimni Lagoon. In addition, mean Cu and Pb levels in surface sediments exceeded the baseline levels in estuarine sediments, while those of Zn and Cd did not [c.  $10 \mu\text{g g}^{-1}$  DW,  $25 \mu\text{g g}^{-1}$  DW,  $<100 \mu\text{g g}^{-1}$

DW and  $0.2 \mu\text{g g}^{-1}$  DW, respectively, according to Kennish (1997)]. The leaves of the submerged angiosperm *R. maritima* generally showed a higher metal accumulation capacity than the green seaweed *U. rigida*, which is considered as an efficient bioindicator of metals (Boubonari et al., 2008). Having a high metal accumulation capacity and potentially attaining a large biomass mainly during the growing season (Bendoricchio et al., 1994; Malea et al., 2004), both *R. maritima* and *U. rigida* may serve as major pools of metals in coastal lagoons.

The patterns of metal concentrations in the dominant macrozoobenthic and fish species of Monolimni Lagoon can be utilized for the assessment of the effects of metal pollution on coastal lagoon biota and for the development of predictive models for rational management of coastal lagoons. Interspecific differences in ecology, feeding habits, morphology, accumulation strategy and physiological requirements for the essential elements can generally explain the observed variability in metal concentrations in macroinvertebrate and fish species (Blackmore, 2001). Metal uptake by aquatic animals occurs directly from water across permeable body surfaces and from food and imbibed water and sediment; the relative importance of each route varies between organisms and between metals for the same organism (Bat et al., 1998; Blackmore, 2001).

The patterns of metal concentrations in the investigated macroinvertebrates and the results of the PCA suggest that the variability in all the five metals' concentrations but mainly in Fe and Pb ones in macroinvertebrates of the coastal lagoon ecosystem depends at least to some extent on their interspecific differences in ecology and feeding habits. The bivalve *A. segmentum* and the polychaete *H. diversicolor*, which are infaunal and at least primarily deposit-feeding species, i.e. animals which ingest their substrate (Gouvis & Koukouras, 1993; Scaps, 2002), showed the highest Fe and Pb amounts, followed by the amphipod *C. orientale* which is a selective deposit feeder (McLusky & Elliott, 2004), probably due to high rates of uptake from sediments, where both these metals are substantially accumulated. Wang & Fisher (1999) suggested that  $>98\%$  of the metals in a surface deposit-feeding polychaete are accumulated from ingested sediments largely because of high ingestion rates of sedimentary particles and low uptake from solution. All the other species,

namely the mudsnail *V. maritima*, which feeds mainly on diatoms growing on the surface of sediment particles (McLusky & Elliott, 2004), the epifaunal amphipod *G. aequicauda*, which primarily feeds on macroalgae and on micro-organisms attached on vascular plant leaves (Casagrande et al., 2006) and the mobile epibenthic decapod *C. crangon*, which preys on benthic macroinvertebrates (Möller et al., 1985), most probably ingest relatively small or mainly accidentally only inconsiderable amounts of sediment particles (Fialkowski et al., 2003).

The deposit-feeding *A. segmentum* also accumulated the highest Cd contents, possibly due to high rates of uptake from surface sediments. The relatively little interspecific variability observed in body Cd concentrations among most of the investigated macroinvertebrates may mainly have arisen from the low Cd concentrations. The contents of this element, being a non-essential one, vary between macroinvertebrates mainly because of differences in routes and rates of uptake (Blackmore, 2001).

The finding that the investigated macroinvertebrates were not grouped strictly according to their feeding habits, as for Zn and mainly Cu concentrations, as well as the PCA results suggest that the variability in these metals concentrations in macroinvertebrates of the coastal lagoon ecosystem depends also on other interspecific differences apart from those in ecology and feeding habits; interspecific differences in morphology, accumulation strategy and physiological requirements for these essential elements may also contribute to this variability (Blackmore, 2001). Several marine invertebrates such as the shrimp *C. crangon*, the polychaete *H. diversicolor*, the bivalve *Scrobicularia plana* and amphipod species regulate their internal Zn concentrations to approximately constant levels, which probably reflect their requirements for this trace metal (Amiard et al., 1987; Bordin et al., 1996; Abdennour, 1997); as for *H. diversicolor*, a large proportion of the total body burden of Zn is found in the jaws (Bryan & Gibbs, 1980). The behaviour of marine invertebrates towards Cu displays cases of full regulation, regulation above certain levels or continual accumulation (Amiard et al., 1987; Bordin et al., 1996); for instance, *C. crangon* regulates total body Cu concentrations to an approximately constant level, which probably reflects the requirements of both copper-associated enzymes and the copper-bearing respiratory pigment haemocyanin

(White & Rainbow, 1985; Amiard et al., 1987; Abdennour, 1997), while *H. diversicolor* accumulates Cu in proportion to its concentrations in the surrounding medium (Amiard et al., 1987; Berndts et al., 1998).

The observation that *K. caucasica* showed low concentrations of most metals analyzed compared to most macroinvertebrate species could be, at least partially, explained by its ecology and feeding habits. This goby feeds mainly on macroinvertebrates (e.g. Kevrekidis et al., 1990) and probably ingests accidentally only inconsiderable amounts of sediment particles. Taking into account that for many metals there is a lack of consensus about the importance of dietary uptake by fish (Reinfelder et al., 1998), water is possibly the major source of metal accumulation in *K. caucasica*, potentially explaining the low concentrations in the whole body of this goby. On the other hand, food has been found to be more important than water as a source of Zn for gobies (Renfro et al., 1974 in Bryan, 1979); thereby, the comparatively high Zn concentrations found in *K. caucasica* were possibly accumulated from its food sources (polychaetes, amphipods and bivalves), which also had high Zn loads.

The observations that (a) only *G. aequicauda* among the dominant macroinvertebrate and fish species in Monolimni Lagoon feeds on plants (mainly on seaweeds), (b) this herbivorous amphipod generally contained relatively low metal loads and (c) there was no essential increase in metal concentrations corresponding to the increasing trophic level (autotrophs, to herbivore, to carnivores) suggest that the trophic transfer of seaweed and mainly of angiosperm-bound metals to the coastal lagoon food web is of relatively minor importance. Thereby, both submerged angiosperms and seaweeds, potentially constituting major pools of metals in coastal lagoons, may act mainly as filters of heavy metal pollution in these systems reducing metal availability to the rest of the organisms.

The observation that muscle tissues of mullets showed comparatively low metal contents in contrast to the deposit-feeding macroinvertebrates suggest that metal accumulation in mullet muscle tissues does not depend markedly on feeding habits. Mulletts, being mainly detritus feeders, probably take up appreciable quantities of metals from sediment particles; they feed by sucking up the surface layer of sediments, transferring the sediment particles into the digestive system along with the food (Zingde et al., 1976). It is possible

that the concentrations of the elements analyzed are accumulated as a function of environmental exposure in certain organs of mullets, such as liver and kidney, and are regulated to low levels in their muscles (Reinfelder et al., 1998; Kalay et al., 1999). Metal concentrations in muscle tissues of both mullet species, which are a human food source, were generally comparable to those previously reported for these species from other coastal locations (e.g. Panayotidis & Florou, 1994; Kalay et al., 1999) and below maximum permitted concentrations for human consumption (Barwick & Maher, 2003). In spite of being below the maximum tolerable concentrations for human consumption, care is necessary when stressing that consumption of mullets will not threaten human health. As said, mullet's organs such as kidney and liver may contain high concentrations of heavy metals. In addition, given the high concentrations of heavy metals in sediment and water (usually exceeding the upper limit of the European Environmental Quality Standards), constant monitoring is necessary.

In conclusion, our results suggest that the variability in iron, lead and cadmium concentrations in macroinvertebrate and goby species of the macrophyte-based coastal lagoon ecosystem depends at least to some extent on interspecific differences in ecology and feeding habits, thus substantially supporting our initial hypothesis; deposit feeders accumulated the highest metal amounts probably due to high rates of uptake from surface sediments. These results also suggest that the variability in zinc and copper concentrations in these components of the coastal lagoon ecosystem depends also on other interspecific differences apart from those in ecology and feeding habits, as well as that metal accumulation in mullet muscle tissues does not depend markedly on mullet feeding habits. Our data additionally suggest that the trophic transfer of macrophyte-bound metals to the coastal lagoon food web is of relatively minor importance. Further studies should concentrate in more detail on the multiple factors potentially affecting metal transfer between coastal lagoon ecosystem components.

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