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TRACE METAL CONCENTRATIONS IN SUSPENDED PARTICLES, SEDIMENTS AND CLAMS *(RUDITAPES PHILIPPINARUM)* FROM JIAOZHOU BAY OF CHINA

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Abstract. Suspended particulate matter (SPM), sediments and clams were collected at three sites in Jiaozhou Bay to assess the magnitude of trace metal pollution in the area. Metal concentrations in SPM (Cu: 40.11–203; Zn: 118–447; Pb: 50.1–132; Cd: 0.55–4.39; Cr: 147.6–288; Mn: 762 − 1670 μg/g), sediments (Cu: 17.64–34.26; Zn: 80.79–110; Pb: 24.57–49.59; Cd: 0.099–0.324; Cr: 41.6–88.1; Mn: 343 − 520μg/g) and bivalves (Cu: 6.41–19.76; Zn: 35.5–85.5; Pb: 0.31–1.01; Cd: 0.51–0.67; Mn: 27.45−67.6 μg/g) are comparable to those reported for other moderately polluted world environments. SPM showed a less clear pattern. Metal concentrations in sediments displayed a clear geographical trend with values increasing with proximity to major urban centers. The clams (on dry weight) showed a complex pattern due to the variability introduced by age-related factors. Cd showed an apparent reverse industrial trend with higher concentrations in clams collected at distant stations. Zn, Pb and Mn showed no clear geographical pattern, whereas Cu increased in the clams collected in the most industrialized area. In addition, the bioaccumulation factors (BAF) were calculated. The result indicated that the studied *Ruditapes philippinarum* in Jiaozhou Bay possessed different bioaccumulation capacities for Cd, Zn, Cu, Pb and Mn, and Cd, Zn had a relatively high assimilation of those metals from sediment particles. A significant relationship with clam age was observed for Zn (positive) and Cu (negative) suggesting different physiological requirements for both metals with age. Trace metal concentrations measured in the tissue of the investigated clam were in the range considered safe by the WHO for human use.

Keywords: trace metals, clams, SPM, sediment, Jiaozhou Bay

1. Introduction

Marine bivalves, including clams, mussels and oysters, can be employed as bio-indicators to assess the marine environment, since they can accumulate (concentrate) trace elements and other substances (LaBrecque *et al.*, 2004). In addition to using bivalve molluscs as bio-indicator organisms of coastal contamination, it is also recognized that they can be important links for contaminants between sediments and higher organisms, including man, that consume them. Hence, information on the contaminant concentrations in their tissues is potentially useful in considering toxicological and public health implications of estuarine contamination (Cheggour *et al.*, 2005). Jiaozhou Bay is an important area in terms of marine products. This semi-closed zone receives a large input of contaminations from the effluents of inhabitants of Qingdao City, tourism activities, aquaculture activities, Qingdao Port activities. Previous studies demonstrated the seasonal variation of some trace metals in the clam collected from the heavily industrialized area (Liu *et al.*, 1983), but no comprehensive study has been undertaken to examine the magnitude of trace metal pollution in the area. The main aim of this work was to determine the total contents of Cd, Cu, Pb, Cr, Mn and Zn in clam (on dry weight), SPM and sediment collected from sampling points where had high densities of the clam *Ruditapes philippinarum* (Han *et al.*, 2004).

2. Materials and Methods

2.1. Sampling

Samples of suspended particulate matter (SPM), sediments and clams were collected in November 2004, at three sites along the northern part of Jiaozhou Bay(Figure 1). Water samples were collected manually with polyethylene bottles. The particles were recovered by filtration through 0.45 μg Millipore membrane filters immediately. After filtration, suspended sediments were preserved at 0–4°C before analysis. Sediment samples were collected manually with a plastic spatula so as to give a representative composite of the area inhabited by the clams (109.27units/m^2). Sediments were stored in polyethylene containers and transported to the laboratory. Clams were collected manually, and the exterior (shells) of the clams were well rinsed with seawater to eliminate marine sediments before being transferred to the bottles filled with distilled deionized water (LaBrecque *et al.*, 2004). After the bottles were closed, they were placed in an iced cooler, for transportation to the laboratory.

2.2. Chemical analyses

In the laboratory, suspended sediments were dried at 105°C in a clean oven. Filters for trace metals analyses were completely digested with a mixture of HF–HNO_3–$HClO_4$ in close Teflon systems (Agemian and Chau, 1975). The wet sediments were thawed and oven-dried at 105°C for 24h, grounded in agate mortar, sieved (160 mesh) and prepared for the analysis elements following the same procedure used for SPM. The levels of LOI in the sediments were determined from loss of ignition of dried sediments heated to 550°C for 2h (Borg and Jonsson 1996). The grain size composition was measured by the method of wet sieving.

Before analysis, the clams were sorted by age into 4 age classes (<1 year, 1 year. 2 years and 3 years of age). After a few days in the refrigerator the clams opened by themselves. The complete soft tissue inside the shells was removed and washed

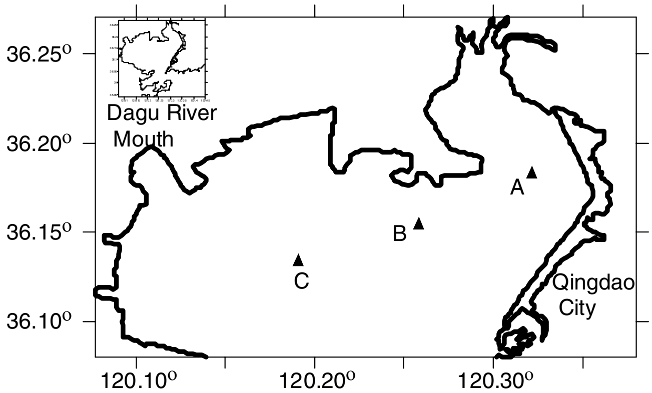

Figure 1. Map of the study area and the sampling stations.

with distilled deionized water to remove marine sediment and other foreign material, then oven-dried to constant weight at 105°C for 3h. The dried soft clam tissue was ground into a fine powder with an agate mortar and pestle and well mixed to prepare a representative sample. Samples were completely mineralized with a mixture of concentrated sulphuric and nitric acids and hydrogen peroxide (Bilos *et al.*, 1998). Duplicate digestions were performed for each sample. Metal analyses of sediment and clam dry tissues were carried out by atomic absorption spectrophotometry (AAS) using either air–acetylene flame or graphite furnace instrumentation.

All procedures including the treatment of sample collection and analyses were carefully undertaken in order to reduce potential contamination. The labware was carefully cleaned (detergent, tap water, 10% nitric acid and deionized water) and the accuracy and precision of heavy metal determinations were monitored through repeated analyses of reference sediments material from North Sea substation of State Ocean Administration, coded (GSBZ50012–88). The accuracy of the most determination was within 5% of the reported values, with the exception of Cd, which was higher about by 11.4% than the reported values.

3. Results and Discussion

3.1. TRACE METAL CONCENTRATIONS IN SUSPENDED MATTER AND SEDIMENTS

Table I presents the trace metal concentrations for the Jiaozhou Bay SPM and sediment samples. In SPM, the concentrations averaged 98.2 μg/g for Cu, 232 μg/g for Zn, 79.7 μg/g for Pb, 2.06 μg/g for Cd, 206 μg/g for Cr and 1311 μg/g for Mn. In sediments the concentrations were significantly lower (25.8, 90.8, 34.8, 0.20, 65.3 and 433 μg/g for Cu, Zn, Pb, Cd, Cr and Mn, respectively).

Trace metal concentrations in Jiaozhou Bay are compared to those reported for other world areas inTable II. Particulate Cu values in Jiaozhou Bay are comparable

TABLE I

The concentration of heavy metals (μg/g) and bulk parameters in suspended particulate matter (SPM) and sediment from the Jiaozhou Bay

Site		Cu	Zn	Pb	Cd	Cr	Mn	%LOI	%Clay	%Silt	%Sand
A	SPM	203	447	50.1	4.39	182	762				
	Sediment	34.26	110	49.59	0.099	88.1	520	2.39	26.6	68.9	4.6
B	SPM	44.11	131	57.1	0.55	148	1500				
	Sediment	25.43	81.73	30.33	0.179	66.1	436	2.13	19.5	65.9	14.6
C	SPM	47.6	118	132	1.23	288	1670				
	Sediment	17.64	80.79	24.57	0.324	41.6	343	1.94	7.2	29.0	63.8
Mean	SPM	98.2	232	79.7	2.06	206	1311				
	Sediment	25.8	90.8	34.8	0.20	65.3	433				
Standard	Actual values	28.2	60.1	23.6	0.031	78.48	1111				
Material	Certified values	28.1	61.5	24.0	0.035	79.9	1063				

to those published for the Changjiang Estuary and Hangzhou Bay. The lowest Cu level is found in the Hangzhou Bay. SPM Zn values in Jiaozhou Bay are lower than those published for Homebush Bay of the Port Jackson Estuary in Australia, while Cr values are higher than those of Homebush Bay. The Cd contents of Jiaozhou Bay SPM are relatively high; the highest value (4.39 μg/g) is four times of the highest concentration recorded for Changjiang Estuary. Particulate Pb values in Jiaozhou Bay are comparable to those published for the German Bight, whereas in Changjiang Estuary, Hangzhou Bay, SPM Pb levels are lower. Mn levels in Jiaozhou Bay SPM are comparable to the German Bight, reflecting the well-established major natural origin (mineral particles) of this element. SPM trace metal data thus indicate that the Jiaozhou Bay is a seriously polluted environment. These comparisons, however, should be considered with care due to the usually high variability often displayed by SPM. Water dynamics, including the influence of tides and water discharge, and anthropogenic activities could have dramatic effects over the particulate chemical composition determining strong temporal and spatial changes (Bilos *et al.*, 1998). For this reason sediment, which integrates the signals over longer periods, is a better compartment to do a comparative evaluation of the magnitude of the anthropogenic heavy metal impact (Nguyen *et al.*, 2005).

Trace metal levels in Jiaozhou Bay sediments are similar to those reported for the Hangzhou Bay, Changjiang Estuary, Güllük Bay, Texas Estuaries, Izmir Bay and lower than those published for the Scheldt Estuary and Boston Harbor. Although trace metal concentrations in sediments are influenced by grain size, the levels of selected elements in Jiaozhou Bay sediments are lower than those published for 'sandy silt' sediments (21% sand, 68% silt, 11% clay) in Río de la Plata Estuary (Bilos *et al.*, 1998). This suggests a moderate degree of trace metal pollution in the Jiaozhou Bay. In summary, based on trace metal concentrations in SPM and surface sediments, the Jiaozhou Bay seems to be a moderately polluted system exhibiting

TABLE II
Trace metal concentrations in Jiaozhou Bay suspended particulate matter and sediments compared with other world areas

	SPM (μg/g)						
Site	Cu	Zn	Pb	Cd	Cr	Mn	References
Jiaozhou Bay	40.11–203	118–447	50.1–132	0.55–4.39	147.6–288	762–1670	This study
Changjiang Estuary	44.8		44.3	1.15			Che *et al.* (2003)
Changjiang Estuary	61.4		29.2	0.39			Che *et al.* (2003)
Hangzhou Bay	31.8		33.3	0.70			Che *et al.* (2003)
Hangzhou Bay	46.5		22.3	0.12			Che *et al.* (2003)
Homebush Bay	44.7	600.6	180.9		85.0		Hatje *et al.* (2001)
German Bight			131			1327	Hinrichs et L., (2002)
	Sediments (μg/g)						
Jiaozhou Bay	17.64–34.26	80.79–110	24.57-49.59	0.099–0.324	41.6-88.1	343–520	This study
Hangzhou Bay	24.1		22.6	0.09			Che *et al.* (2003)
Changjiang Estuary	21.2		21.7	0.12			Che *et al.* (2003)
Güllük Bay	25	81	20	0.56		(2005)	Dalman *et al.*
Texas Estuaries	1–20.5	40–295	8–150	0.2–45.5	3.5–132		Sharma *et al.* (1999)
Scheldt Estuaries	1–2600	9–1500	4–455	0.1–20.2	7–205		Zwolsman *et al.* (1996)
Izmir Bay	13–49	45–114	23–52				Balcı and Türkoğlu (1993)
Boston Harbor	67	118	86		131		Bothner *et al.* (1998)

trace metal concentrations in the range of other urbanized and industrialized areas of the world.

3.2. TRACE METAL CONCENTRATIONS IN CLAMS (ON DRY WEIGHT)

Table III summarizes trace metal concentrations in clams from Jiaozhou Bay. The most abundant trace metals were Zn, Mn and Cu, followed in much lower concentrations by Pb and Cd, while Cr was undetectable levels in all clam samples (<0.01mg/kg). InTable IV trace metal concentrations of clams from different environments are compared. Cu concentrations in Jiaozhou Bay *Ruditapes*

TABLE III
Trace metal concentrations (μg/g) in clams from Jiaozhou Bay

Site	Age	Cu	Zn	Pb	Cd	Mn
A	1 year	19.76 ± 2.39	85.5 ± 3.0	0.60 ± 0.12	0.64 ± 0.037	46.5 ± 3.67
	2 years	17.96 ± 2.15	79.45 ± 7.77	0.53 ± 0.07	0.64 ± 0.12	47.17 ± 8.03
	3 years	18.79	80.1	0.60	0.51	48.0
B	<1 year	6.41	35.5	0.80	0.68	38.2
	1 year	11.72 ± 2.22	44.03 ± 6.58	0.53 ± 0.15	0.63 ± 0.16	58.87 ± 0.55
	2 years	13.79	36.36	1.01	0.58	67.7
	3 years	12.65	74.9	0.89	0.59	51.5
C	2 years	10.33	44.17	0.34	0.67	33.4
	3 years	7.18 ± 2.89	57.75 ± 10.54	0.31 ± 0.066	0.67 ± 0.19	27.45 ± 2.05

philippinarum agree with those previously reported for *Ruditapes philippinarum* in Jiaozhou Bay (Liu *et al.*, 1983). The other bivalves that show low Cu levels are *Circentia callipyga* from Qatar, *Ruditapes philippinarum* from Bohai Sea and *Vesicomya gigas* from the Gulf of Colifornia. *Corbicula fluminea* from Río de la Plata and *Corbicula fluminea* from Shatt al-Arab River show high Cu levels. Zn is a metal that shows marked differences among the clam species. Our values are much higher than that reported for *Ruditapes philippinarum* from Bohai Sea, and much lower than those reported for *Scrobicularia plana* from Moroccan estuaries, *Vesicomya gigas* from the Gulf of Colifornia, *Corbicula fluminea* from Río de la Plata and *Cardium* from Atlantic coast of Spain and Portugal. Pb concentrations in Jiaozhou Bay clams are comparable to those reported for other organisms with the exception of *Ruditapes philippinarum* from Bohai Sea and *Cardium* from Atlantic coast. Our levels of Cd in Jiaozhou Bay clams are much lower than *Vesicomya gigas* from the highly polluted Gulf of Colifornia, *Corbicula fluminea* from Shatt al-Arab River and *Cardium* from Atlantic coast. Mn shows uniform contents among species from different environments.

The bioaccumulation factors (BAF) calculated according to the following formula (Chen, 1999): BAF = Metal concentration (mg kg^{-1} dry wt.) in whole organism/Metal concentration (mg kg^{-1} dry wt.) in sediment(Table V), suggest that clams accumulate Cd (BAF = 2.07-6.03; mean: 3.85), Zn (BAF = 0.58–0.74; mean: 0.65) and Cu (BAF = 0.44–0.55; mean: 0.50), whereas the accumulation of Pb and Mn is 1 order of magnitude lower (BAF = 0.012 – 0.027 for Pb and 0.07 – 0.12 for Mn). The above result shows that metal concentrations in tissues of *Ruditapes philippinarum* from Jiaozhou Bay were lower than those in surrounding sediments with the exception of Cd. In this study the assimilation of these metals from these sediments by clams was not measured, so it is not possible to interpret unambiguously relationships of trace metal concentrations in *Ruditapes philippinarum* tissues with those in the sediments. But the correlations of sediment-bound metals and the metals burden found in clams could suggest some information. The correlations

TABLE IV
Trace metal concentrations (μg/g) in dried soft tissue of clams from different environments

(location) Species	Cu	Zn	Pb	Cd	Mn	References
Ruditapes philippinarum (Jiaozhou Bay)	6.41–19.76	35.5–85.5	0.31–1.01	0.51–0.67	27.45–67.6	This study
Ruditapes philippinarum (Jiaozhou Bay)	0.67-23.6	8.8-120.5	0.33-1.66	0.21-2.33		Liu *et al.* (1983)
Scrobicularia plana (Moroccan Atlantic estuaries)	16.2-20	142-222		0.19-1.64	14.6-39.7	Cheggour *et al.* (2005)
Ticela mactroidea (Edo.Miranda Venezuela)	11-49	55-166	$< 1.5 - 4.9$	$< 1 - 1.9$		LaBrecque *et al.* (2004)
Circentia callipyga (Ras Al Nouf, Qatar)	8.35	69.1	1.45	1.17	17.7	Mora *et al.* (2004)
Ruditapes philippinarum (Bohai Sea)	1.28-4.37	9.95-20.06	0.13-0.33	0.14-0.63		Liang *et al.* (2004)
Vesicomya gigas (Gulf of California	8.26 ± 5.8	844.8 ± 1042	2.89 ± 2.5	115.2 ± 196	18 ± 17	Ruelas-Inzunzaa *et al.* (2003)
Corbicula fluminea (Río de la Plata Argentina)	28-89	118-316		0.5-1.9	15-81	Bilos *et al.* (1998)
Corbicula fluminea (Shatt al-Arab River Iraq)	40-1065	31-83		2.2-70	1.8-3.0	Abaychi and Mustafa (1988)
Cardium (Atlantic coast of Spain and Portugal)	15	210	32	7.5		Stenner and Nickless (1975)

TABLE V
(The bioaccumulation factors (BAF) calculated on a dry weight basis relative to sediments

Site	Cu	Zn	Pb	Cd	Mn
A	0.55	0.74	0.012	6.03	0.09
B	0.44	0.58	0.027	3.46	0.12
C	0.51	0.63	0.013	2.07	0.07
Mean	0.50	0.65	0.017	3.85	0.09

BAF = Metal concentration (mg kg^{-1} dry wt.) in whole organism/Metal concentration (mg kg^{-1} dry wt.) in sediment.

between sedimentary and biological compartments were strongly positive for Cd ($r = 0.997$; $p < 0.01$) and Zn ($r = 0.993$; $p < 0.1$), which could indicate a relatively high assimilation of those metals from sediment particles. In contrast, the correlations of Cu, Pb and Mn concentrations between clam and sediment were not significant.

The above results indicated that the studied *Ruditapes philippinarum* in Jiaozhou Bay possessed different bioaccumulation capacities for Cd, Zn, Cu, Pb and Mn, The concentrations of Cd, Zn and Pb in R. philippinarum from the three sites studies did not exceed the maximum permissible levels (MPLs) of 2 μg/g Cd, 2.0 μg/g Pb and 100 μg/g Zn according to the codes of WHO (1982). But Cu contents of most samples exceeded the MPLs (10 μg/g) set up by WHO (1982).

3.3. Geographical trends of trace metal concentrations and compositions

SPM trace metal concentrations show a less clear geographical trend (Table 1); Zn show a strong anthropogenic influence as indicate the increasing trend observed from C (rural area) to A which is the most industrialized and populated region of the bay. Pb, Cr and Mn values show an opposite trend with Zn, reflecting their major natural origin. In case of Cu and Cd, their highest contents were obtained at station A, whereas the lowest were found at station B. In the sediments, the anthropogenic pattern is more obvious. All trace metals except Cd show an apparently same industrial trend (values increasing with proximity to major urban centers at C-B-A). In bivalves, the geographical pattern is complicated by the variability introduced by age-related factors. Cd showed an apparently reverse trend with industrial impact (values increasing from A→B→C), others (Zn, Pb, Mn) showed no clear pattern, while Cu was the only element to show an increasing trend from C→B→A, where A is the most industrialized region with the highest SPM and sediment trace metal concentrations.

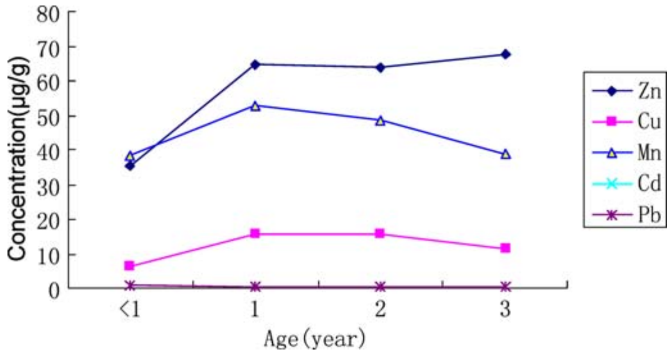

Figure 2. Heavy metal concentrations in *Ruditapes philippinarum* related to clam age.

3.4. Age-related trends of trace metals in clams

Figure 2 presents the heavy metal concentrations in *Ruditapes philippinarum* from Jiaozhou Bay related to clam age, which indicates that some trace elements show a clear relationship with clam age. Relationships between heavy metal concentrations Cu, Cd, Zn, Pb and Mn in *Ruditapes philippinarum* and the clam ages (< 1, 1, 2 and 3 years) are classified into two types: 1. Type A, in which element accumulation in clam is increasing with age. 2. Type B showed a decreasing trend with age. This classification was obtained according to the regression lines: $Y = AX+b$ (Rashed, 2001) where Y is the element concentration (μg/g) in clam and X is the clam age (year). The results showed that in case of type A, Zn, Pb and Cd showed increasing tendency with clam age (Zn $= 65.91 + 0.026$ age; $r = 0.992$, $p < 0.01$; Pb $= 0.56 + 0.002$ age; $r = 0.978$, $p < 0.01$; Cd $= 0.62 + 0.00055$ age; $r = 0.944$, $p < 0.01$). In case of type B, concentrations of Cu and Mn decreased with clam age (Cu $= 14.43–0.078$ age; $r = 0.898$, $p < 0.01$; Mn $= 46.81–0.084$ age; $r = 0.59$, $p < 0.01$). Phillips (1977) found that the several variables such as season, type of tissue, sex or feeding habits affect the relationships between the age, tissue weight or size of bivalves and metal concentrations. In Río de la Plata of Argentina, Cu levels in clams show an important increase with increasing bivalve size (age), whereas Zn follows a significant opposite trend. Considering that Cu and Zn are both essential metals involved in several enzymatic systems, which thus display partial or total regulation in bivalves (Kraak *et al.*, 1994), the clear opposite concentration-age trends observed in Jiaozhou Bay clams strongly suggest different physiological requirements with age. In case of Cd, the concentration-age relationship is similar to that of clams in Río de la Plata. Bivalves, as other aquatic organisms, do not regulate Cd and usually accumulate this element (Tessier, 1994). Our results might suggest that *Ruditapes philippinarum* could not regulate the body concentration of Cd to constant level as other species.

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