Cadmium and Lead in a Trophic Marine Chain

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Heavy metals participate in complex biogeochemical cycles whose final destination (except on a geological scale) is the bottom of oceans and lakes. This implies that to reach the bottom the metals of the sediment have to travel through the water column. The main source of heavy metals in sea-water is particulate matter both air and water. Different organisms can bioaccumulate heavy metals and eventually reach toxic levels. This happens when their normal levels are exceeded because of anthropic activities thus exposing to the human population who consume seafood to a potential danger. Metal concentration in seafood is determinated by age, feeding habits, and by the trophic level to which the respective species belong to.

Heavy metal transfer from one trophic level to a higher one in marine environments have been described already (McDonald and Sprague, 1988; Blomqvist et al., 1987). However, there is a lack of knowledge about heavy metals dynamics in relation to their accumulation in living organisms and their access to the marine environment of the Arauco Gulf. The present report attempts to study the behaviour of metals in a benthonic-demersal trophic chain which originates in the organic matter of the sea bottom sediments where these metals, due to hypoxic conditions ("oxygen minimum") during almost all part of the year, may accumulate. This organic matter serves as nourishment to *Pleuroncodes monodon* ("squat lobster") which, in turn, is an important food source for *Genypterus maculatus* ("black ling") . Both of these species are abundant in this area and are economically important as food supply for the population, both in their fresh and frozen forms. Our working hypothesis was that sediments in the northern part of the Arauco Gulf become a temporary reserve for the potential or actual pollutants generated upriver by anthropic activities in the Biobio basin. As an open system it receives, in the northern sector of the gulf, all the contributions of the basin, mainly in the area of the Biobio canyon and in the immediate continental platform.

MATERIALS AND METHODS

Cadmium and Pb measurements were performed using a Perkin Elmer instrument furnished with flame (FAAS, for sediments and organism determination) and graphite furnace attachments and an AS 70 autosampler (GFAAS, for TSM). Cd was measured at 228.8 nm with a limit of detection of 0.2 µg/L, for GFAAS, and 8 µg/L, for FAAS. Pb was measured at 283.1 nm with a limit of detection of 10 µg/L, for GFAAS, and 64 µg/L, for FAAS. All measures were carried out in

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Table 1. Laboratory determination of certified standard (NRCC)

Standard National Research Council Canada (NRCC)	Metal	Certified μg/g	Laboratory μg/g
MARINE SEDIMENTS			
BCSS-1	Cd	0.25 ± 0.04	0.5 ± 0.1
	Pb	22.7 ± 3.4	20.0 ± 7
MESS-1	Pb	34.0 ± 6.1	38 ± 5
PACS-1	Cd	2.38 ± 0.20	2.5 ± 0.1
	Pb	404 ± 20	355 ± 60
LOBSTER HEPATOPANCREAS (CRUSTACEA) TORT-1	Cd	26.3 ± 2.1	24.2 ± 2.1
	Pb	1.04 ± 2	2 ± 2
SPRING DOGFISH LIVER (SHARK) DOLT-1	Cd	4.18 ± 0.28	4.6 ± 0.6
	Pb	1.36 ± 0.29	n.d. (*)
SPRING DOGFISH MUSCLE (SHARK) DORM-1	Cd	0.086 ± 0.012	n.d.
	Pb	0.4 ± 0.12	0.4 ± 0.1

(*) n.d. : non detected.

reproducibility of the procedures for metal determinations in biological materials, a test of measures was carried out. Using different samples a 5.4% coefficient of variation was obtained for the Cd (n=7; 3.750.2 μg/g)and of 20% for the Pb (n=7; 2±0.4 μg/g). All glass and plastic material used were precleaned (UNEP 1984).

The areas studied were: i) Arauco Gulf between Punta Cullinto (36°47'S) and Punta Lavapié (37°09'S, 73°lO'W); ii) Cobquecura, the reference area for the *Pleuroncodes monodon,* at 36°07'S and 72°57'W and iii) Lebu, the reference area for the *Genypterus maculatus,* at 36°55'S and 73°39'W.

Samples of water containing suspended particulate matter were collected, using 5 L van Dorn bottles, between 4-5 m above the bottom at the same sampling sites of sediments. Temperature and dissolved oxygen were determined on water samples. On board of the vessel the water samples were filtered through a 0.45 mm cellulose acetate membrane filter, the residue was dried at 40°C until a constant weight was obtained. The material filtered was dried at 30°C during 24 h, then treated in a polycarbonate jar with 4 mL of 30% HNO_3 plus 4 mL bidistilled water, kept in an oven at 30°C for 3 h, and then levelled up to 10 mL in 10% HNO_3.

Samples of sediments were collected at a 100 m depth using a 0.1 m^2 Smith-McIntyre dredge (UNEP 1984). Twenty four samples were obtained (1992-1993), kept at -80°C and then dried at 40°C up to a constant weight. The particle size of sediments was determined by sieving a wet sample through a 4 phi (0.0625 mm) sieve. Organic matter was determined with $H2O_2$ 150 vol, and muffle combustion (Mook and Hoskin 1982). Chlorophyll and phaeophytin were quantified (Golterman *et al.* 1978).

For total attack, to determinate metal content in sediments, a sequential extraction procedure with 1) HNO_3, 2) HF, and 3) $HClO_4$ (González 1994). For partial attack, to estimate metal bioavailability (Louma 1990), a sequential extraction procedure to obtain four fractions (Giordano *et al.* 1992 modified Gonzalez 1994): 1)Exchangeable (sodium acetate 1M, pH 4.98); 2)Reducible (NH_2OH 0.004 M in acetic acid 25%); 3)Bonding to organic matter and sulphur (HNO_3 0.8 N); 4)Residual (residue obtained).

Samples of adults of *G. maculatus* (standard length 49.4 ± 5.8 cm), were obtained in the Arauco gulf near the mouth of the Biobio River (105 individuals), and in Lebu (13 individuals). A pilot sample (9 specimens) was used to calculate the minimum number of individuals (44 specimens)that would be representative of the total population (Keith *et al.* 1983). All the specimens were dissected by surgical steel knife obtaining liver, kidneys, gills, stomachs content and muscles (below pectoral fins), lyophilised and stored at -80°C. Ten individuals of 10 cm of cephalothorax length were used to calculate minimum sample size (20 individuals). Sampling of "squat lobster" was collected using a trawl net at 100 to 150 m depth in Cobquecura, and in Arauco Gulf. Cobquecura is not affected by human activity of the Biobío Basin and Arauco Gulf. Moisture content of tissues was calculated. Between 0.5-l g of this lyophilised material was then soaked during 8 h in HNO_3, in a refrigerated system, and then heated during 1 h at 40°C, filtered through a MFS N° 11 filter paper and the filtrate made up to volume (20 mL) with Milli-Q bidistilled water.

The data obtained was statistically analysed using LOTUS 123 generated tables and SYSTAT programs (System of Statistics, Wilkinson 1981) and a NTSYS (Numerical Taxonomy and Systematics, Rohlf 1982). Each compartment (particulate matter (TSM), sediment, biota) was analysed as a function of the date and geographical location of the sampling station.

RESULTS AND DISCUSSION

The oceanographic and meteorological conditions allowed sampling at around 120 m depth during February, April and May (in 1992-1993). Water temperature as well as dissolved oxygen were measured, and TSM in waters above the bottom was isolated and its Cd and Pb contents determined (Table 2).

An analysis of conglomerates was used to detect possible relationships between heavy metals and particulate matter concentrations. Using a basic standard matrix of 3 variables: Cd, Pb, TSM for 17 stations, they were grouped in a dendrogram based on the similarity coefficients obtained by algorithm "UPGMA" and binding average method. Differences observed were statistically significant (*P* 0.95, Fig.2) using a multivariate variance analysis (SYSTAT software) (Wilkinson 1988).

The area sampled contains a high percentage of fine grained materials in the bottom sediments: mud (>90%) and shows reducing properties and presence of *Thioploca* sp. Organic matter, as determined by H_2O_2 oxidation averages 2.03%+1.19 (c.v. 58.9%; and ranges 4.8:0.44-5.24), whereas by the combustion method (50°-500°C) the contents were much higher (Fig. 1). Pigments in sediments averaged 57.6 ±41.3 µg/g of dry weight, 78% of them as phaeophytins. From February to May increased from 10 to 87 µg/g. The mean concentrations of Cd and Pb are 4.6±2.3 µg/g and 19±6 µg/g repectively.

The so-called exchangeable fraction comprises the metal adsorbed in charged surfaces (i.e. humic acids). A change in the normal composition of the sea water would therefore affect metal adsorption or desorption. About 43% of the Cd present is found in this fraction. In the case of Pb, this fraction comprise only about 2% of the total Pb content and therefore would be of low importance.

Fe and Mn oxides (reducible form fraction) which are *excellent* sequestering agents, will not retain the metal under anoxic conditions, since at low redox the oxides are unstable. The reducible Cd fraction is about 16.3% of the total metal present. No Pb was found in this form.

Sulphide and organically bound fraction includes metals covalently bound to organic matter and to sulphide which are not bioavailable

ORGANIC MATTER CONTENT

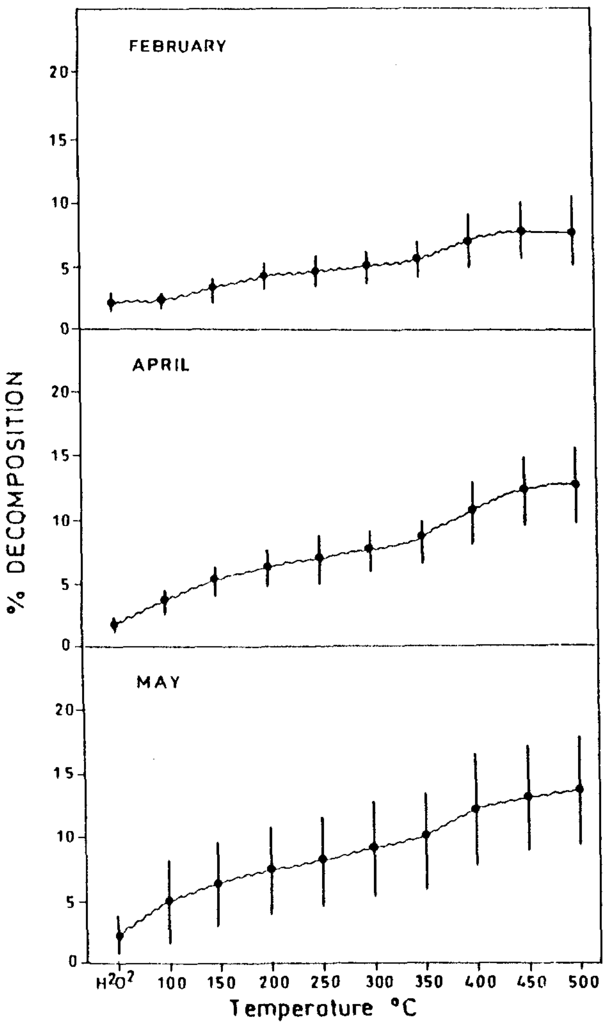

Figure 1. Organic matter content in sediment samples in Arauco Gulf. The first point is H_2O_2.

at low redox potentials. Only 10.7% of Cd is found in this fraction.The mean of lead organically bound is about 25.9% average but, it varies widely. In February 44% of total Pb was in this form whereas in April and May only about 20%.

There is a change in anoxic conditions from February to May when dissolved oxygen in the water column above sediment increases solubilizes sulphide bound metal. Thus indicating that Pb and Cd are bound to sulphide more than to organic matter. The residual fraction (non bioavailable fraction) is of 30.1% for Cd and 72.1% for Pb.

A factorial analysis of principal components (PAC) allows the grouping of stations. The first component explained 35.18% of the total variables and showed a good correlation with organic matter content. The second component explained 13.35% and was related to mud content.

A discriminate analysis of the heavy metal variable, was in agreement with the same distribution of variables (Fig. 2, Fig. 3).

Cadmium and Pb content in "squat lobster" and in different organs of "black ling" are shown in Table 3. As it can be seen, the metal content in the Lebu "black ling" control specimens was higher than the one found in the Gulf of Arauco specimens.

In Arauco Gulf the organic matter content in sediments is the main pneryy source for the benthodemersal fauna of the Gulf of Arauco. It is thus the starting point for metal transfer analysis studies.

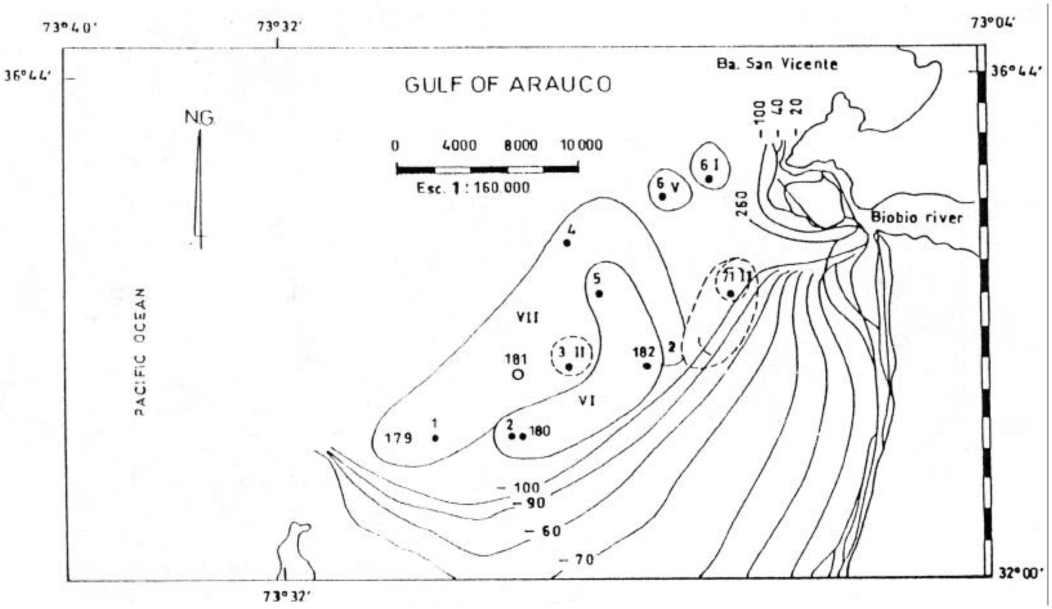

Figure 2. Grouping of suspended particulate matter (TSM) sampling stations, using as metric distance Euclidian distance and UPGMA as binding.

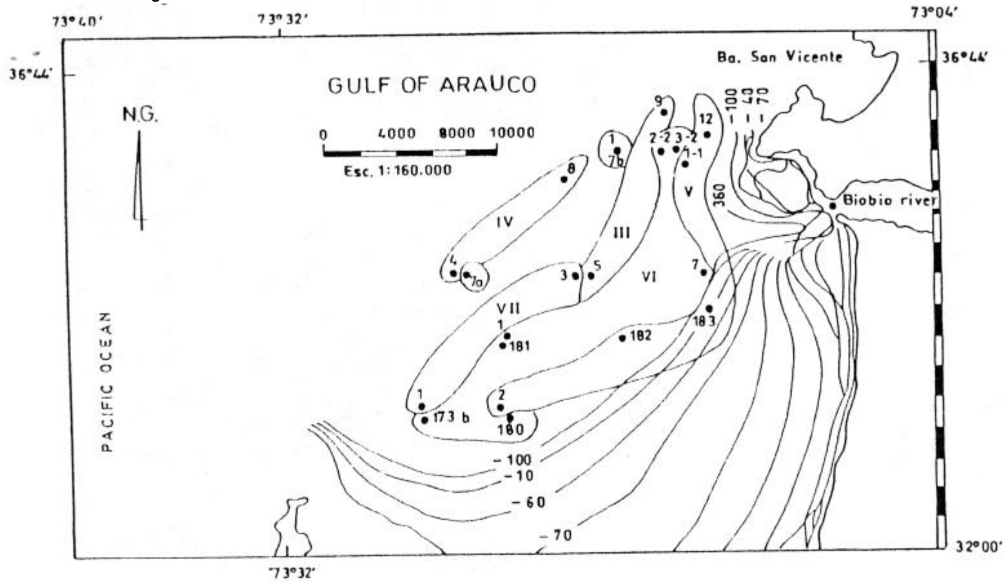

Figure 3. Grouping of sediment sampling stations, using as metric distance Euclidian distance and UPGMA as binding; according to their main axes (axes F_1 and F_2, PCA).

Table 2. Monthly average of Temperature, Oxygen and TSM and Cd and Pb of TSM.

MONTH	DEPTH m	TEMPERATURE ° C	OXIGEN mL/L	TSM mg/L	Cd in TSM µg/g	Pb in TSM µg/g
FEBRUARY	136	11.2	------	7.4	0.05	0.10
APRIL	119	11.9	0.65	2.3	0.01	0.03
MAY	122	-------	3.60	3.5	0.05	<0.01

Table 3. Average content of Cd and Pb in biological matrixes(ppm dry weight)

MATRIX	CADMIUM ARAUCO	CADMIUM CONTROL	LEAD ARAUCO	LEAD CONTROL
P.monodon ("squat lobster")TOTAL	2.3	2.5	1	11
G. maculatus ("black ling")TOTAL	1.1		2	
G. maculatus LIVER	1.0	1.7	2	5
G. maculatus BRANCHES	0.8	0.4	1	8
G. maculatus KIDNEYS	1.6	1.4	5	3
G. maculatus STOMACS CONTENTS	1.7	3.6	2	2
G. maculatus MUSCLES	0.6	0.3	1	5

This means that metal bioavailability has to be defined. For the purpose of this study, the exchangeable, reducible and organically bound fractions of the metal were considered to be the bioavailable forms, because of an organism might incorporate these forms through its digestive tract while feeding. In other words, total minus residual fraction equals bioavailable metal.

The amount of Cd and Pb found in "squat lobster" (*P. monodon*) and in "black ling" (*G. maculatus)* (Table 3) are in the same range as those described for other species (Alliot and Frenet-Piron 1990, Harding and Goyette 1989) living in the muddy beds of a restricted bay. *P. monodon,* like all crustaceans, feeds on detritus through the resuspension of sediments followed by suction and filtration (Gallardo et al. 1993; Allen et al. 1990). In this form "squat lobster" forms the first link in the sediment based trophic chain.

The second level refers to the concentration factor calculated by the ratio of "black ling" metal content/ "squat lobster" metal content. It was found to be 0.50 for Cd and 1.57 for Pb. The ratio of total metal in "black ling" vs. total metal in sediment was 0.23 for Cd and 0.12 for Pb. Cadmium can be intake through the gills but undoubtedly the stomach content is more important.

Enrichment factors of Cd and Pb in the gulf sediments as related to their concentration in the earth surface crust where found to be 12.5 for Cd and 1.0 for Pb. This means that Cd and Pb in the Gulf of Arauco sediments have a natural origin (Trucco et al. 1990). Cd was found in a range of concentration of 0-20 µg/g in the sediment of bays in the north of Chile and therefore no metal pollution as related to Cd and Pb occurs in the south central region adjacent to the Gulf of Arauco Biobio canyon.

According to the data obtained in this work, the northern part of the Gulf of Arauco acts as a trap for Cd and Pb accumulating these metals with the help of hydrodynamic processes and tranferred to biota.

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REFERENCES

Allen JRL, Rae JE, Zanin PE (1990) Metal speciation (Cu, Zn, Pb) and organic matter in an oxic salt marsh Severn estuary, southwest Britain. Mar Pollut Bull 21:574-580.

Alliot A, Frenet Piron M (1990) Relationship between metal in sea water and accumulation in shrimp. Mar Pollut Bull 21:30-33.

Blomqvist S, Frank A, Petersson LR (1987) Metals in liver and kidney tissues of *C.alpina* and *C. ferruginea* staging at Baltic Sea. Mar Ecol Progr Ser 35:1-13.

Gallardo VA, Enriquez S, Roa R, Acuña A, Cañete I, Baltazar M (1993) Biología del langostino Colorado: *P. monodon. In* F. Faranda and O. Parra, eds. Monografías EULA. Chile. 2:67-109.

Giordano R, Musmeci L, Ciaralli L, Vernilo I, Chirico M, Piccioni A, Costantini S (1992) Total contents sequential extractions of Hg, Cd and Pb in coastal sediments. Mar Pollut Bull 24:350-357.

Golterman HL, Clymo RS, Ohnstad MA (1978) Methods for physical and chemical analysis of freshwater. Blackwell Sci. Publ. 213 pp.

Gonzalez F (1994) Metales pesados en la cadena trófica, materia orgánica en sedimento-langostino -congrio del Golfo de Arauco, Chile. Tesis. U. de Concepción. Chile. 167 pp.

Harding L, Goyette D (1989) Metals in northeast Pacific coastal sediments and fish, shrimp, and prawn tissues. Mar Pollut Bull 20:187-189.

Keith LH, Libby RA, Crummett W, Taylor JK, Deegan Jr J (1983) Principles of environmental analysis. Anal Chem 55:2210-2218.

Louma SN (1990) Processes affecting metal concentrations in estuarine and coastal marine sediments. In: Heavy metals in the marine environment. Furness, R.W. and Rainbow, P.S., eds. CRC Press Inc., Florida. USA. 51-66.

McDonald CR, Sprague JB (1988) Cd in marine invertebrates and artic cod in the Canadian Artic. Mar Ecol Prog Ser 47:17-30.

Mook D, Hoskin C (1982) Organic determination by ignition: caution advised. Estuarine, Coastal and Shelf Science 15:697-699.

Rohlf FJ (1982) NT-SYS, numerical taxonomy system of multivariate statistical program. Tech.Rep. SUNY Stony Brook, N.Y. 213 pp.

Trucco RG, Inda J, Fernández ML (1990) Heavy metal concentration in sediments from Tongoy and La Herradura Bays, Coquimbo, Chile. Mar Pollut Bull 21:229-232.

UNEP (1984) Sampling of marine organisms and preparation for trace metal analysis. Ref. Methods for Marine Pollution Studies N° 7.

WHO (1989) Lead. IPCS. Environmental Criteria 85.106 pp.

Wilkinson L (1988) SYSTAT: The system for statistics. Evanston, II. 369 pp.