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Environmental Contamination and Toxicology

Heavy Metals in Bivalve Molluscs in the Huelva Estuary

M. López-Artíguez, M. L. Soria and M. Repetto

Instituto Nacional de Toxicologia, Aptdo. 864, 41080 Sevilla, Spain

Bivalve molluscs accumulate metals in their tissues in proportion to the degree of environmental contamination and can be used as indicators of marine metallic pollution (Goldberg et al, 1983; Elder et al, 1984). They are very appropriate as monitors in situ because they are sedentary, abundant, of relative longevity, large, easily collected and weighed (Hartley and Johnston, 1983). However, the concentration of metal in the molluscs depends not only on the level of the element in the environment but also on other factors: size, age, speed of growth, sex and reproductive conditions of the bivalve, season, salinity, chemical species and interaction with other pollutants (Philips, 1980).

On the Spanish south Atlantic shore there is a strip of coast, situated between the Huelva estuary and the point at Rota, which suffers heavy contamination by several metals; this is especially so in the Huelva estuary itself, probably due to the fact that there are great number of industries on the left bank and that the Tinto River can bring those metals as it flows through a mining area (Stenner et al, 1975). For years the accumulation of copper and zinc has been evident in bivalve molluscs in the estuary, the most sensitive species being the oyster (Crassostrea angulata) which turns a strong green colour. This contamination makes a purification process obligatory (Establier, 19691; Establier et al, 1974); in other studies iron, manganese and zinc were observed (Establier, 1972).

As a result of great mortality of fish in the estuary, coinciding with a strong acid pH in the water, perhaps as a consequence of industrial dumping, and in view of the concern of the health authorities we initiated a study of metallic contamination in the area, using molluscs as samplers.

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send reprint requests to Miguel López Artíguez at the above address

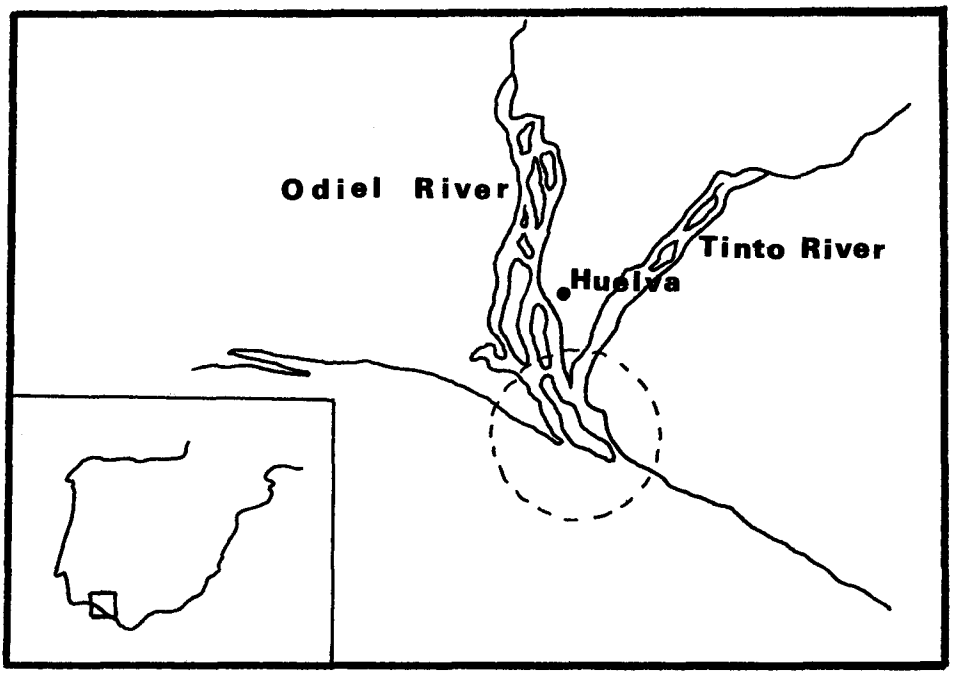

Figure 1. Huelva estuary. Bivalve molluscs sampling site.

MATERIALS AND METHODS

Three species of bivalve molluscs were used: clams (Tapes decussatus), oysters (Crassostrea angulata) and cockles (Cardium edule).

All these species were taken in the Huelva estuary (mouth of the rivers Tinto and Odiel) (Fig. 1) during the month of January, 1987.

The number of lots of each species was 4, 16 and 6 respectively.

The samples were prepared by cutting the adductor muscles and letting the intervalve water drain for ten minutes, then the edible part was separated from the valves and the tissues milled in a Moulinex blender to a fine paste.

The metals chosen for analysis were: arsenic, cadmium, copper, chromium, tin, mercury, lead and zinc. This latter was only determined in oysters:

For the assay of these metals, except arsenic and mercury, portions of about 15 g of each of the triturated samples were digested in a mixture of 15 mL concentrated nitric acid and 6 mL concentrated

sulphuric acid in a 250 mL Kjeldahl flask. The mixture was first shaken for about five minutes, at room temperature to avoid the formation of froth, then simmered for one or two hours and allowed to cool to room temperature. Finally 30% hydrogen peroxide solution was added; and the mixture heated to obtain a colourless solution. The different digestion liquids were made up to 100 mL with deionized water.

Arsenic and mercury assay required the appropriate SNP procedure for fatty tissues (Fedman, 1974) which consists of wet digestion with sulphuric, nitric and perchloric acids in a reaction flask fitted with an air condenser covered with asbestos and teflon.

The final solutions were analysed by atomic absorption spectrophotometry with flame (Cu, Zn), graphite furnace (Cd, Cr, Sn, and Pb), hydride system (As) and cold vapour (Hg), in a model 2380 Perkin-Elmer AA spectrometer equipped with an HGA 500 graphite furnace, an AS 40 Auto Sampler and an MHS-10 mercury hydride system.

For flame AAS an air-acetylene mixture was used. In electrothermic AAS the solutions were sequentially dried, mineralized and atomized.

Arsenic and mercury were analysed by the mercury hydride system, which uses a solution of sodium borohydride as a reducing agent, the arsenic hydride formed was carried in a current of nitrogen to a red hot quartz cell. In the case of mercury the process was similar but without heating the quartz tube.

All the material used in the analysis was kept in a 5% nitric acid solution all night, then treated with deionized water and dried.

All the reagents were of analysis reagent quality (Merck). Ultrapure water (Milli-Q water) was used.

Recuperation values for all the metals were over 90% except for tin which only reached 80%.

RESULTS AND DISCUSSION

The concentrations of trace metal residues in the samples of clams, cockles and oysters is presented in tables I, II and III, and the corresponding histograms in figures 2, 3 and 4.

The concentration of each metal varied greatly in the three types of bivalves.

The range of concentration of copper in clams was 2.19-

Results of the analysis of trace metals in molluscs in the Huelva estuary

Table 1. Clams

Elements	Range of concentration (ug/g)	Mean Value (ug/g)	% Relative Standard Deviation
As	3.63-4.82	4.00	13.86
Cd	0.48-1.30	0.85	41.77
Cu	2.12-3.99	2.89	26.28
Cr	0.71-0.94	0.87	11.15
Sn	2.07-9.07	6.09	49.79
Hg	0.78-1.18	0.98	19.90
Pb	0.15-0.25	0.20	19.31

4 Samples of clams

Table 2. Oysters

Elements	Range of concentration (ug/g)	Mean Value (ug/g)	% Relative Standard Deviation
As	1.18 - 2.07	1.51	19.11
Cd	0.69 - 4.07	1.98	45.50
Cu	26.37 -535.09	180.45	80.68
Cr	0.35 -0.44	0.41	8.12
Sn	0.49 -6.15	2.84	71.94
Hg	0.018-0.90	0.56	53.06
Pb	0.08 - 1.24	0.25	127.15
Zn	296.04 -651.16	465.74	35.22

16 Samples of Oysters

Table 3. Cockles

Elements	Range of concentration (ug/g)	Mean Value (ug/g)	% Relative Standard Deviation
As	1.31- 1.89	1.56	15.53
Cd	0.18- 0.86	0.41	53.27
Cu	2.32-10.90	6.42	51.98
Cr	0.88- 1.52	1.30	19.50
Sn	0.54- 9.67	4.58	69.89
Hg	0.02- 1.27	0.70	75.80
Pb	0.14- 1.53	0.59	102.49

6 Samples of Cockles

Note.- The concentrations of the elements refer to wet weight of the sample

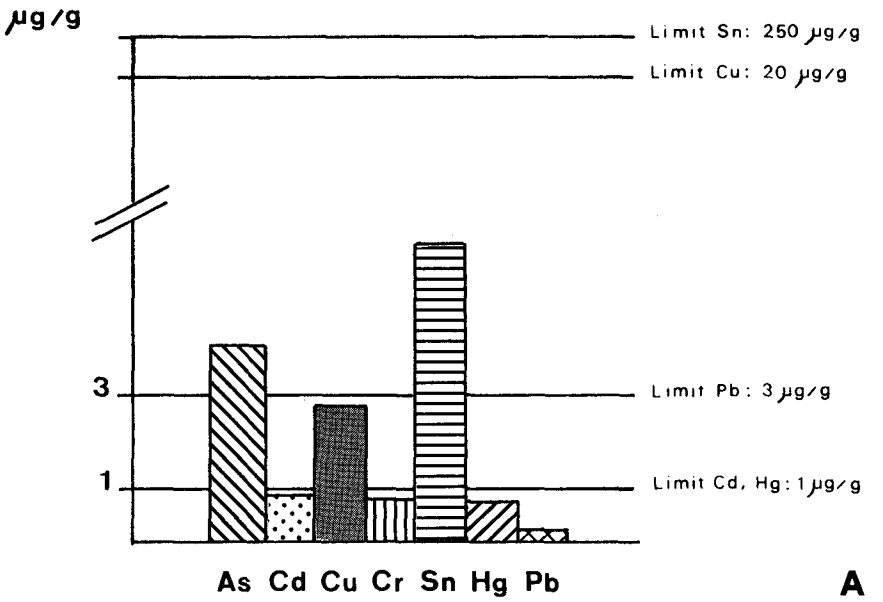

Figure 2.- Average concentration of the metals in clams.

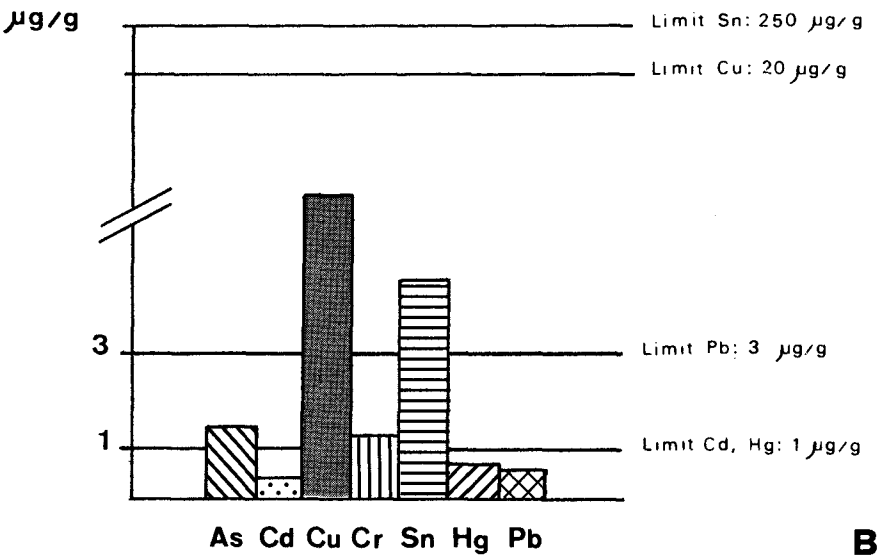

Figure 3.- Average concentration of the metals in cockles.

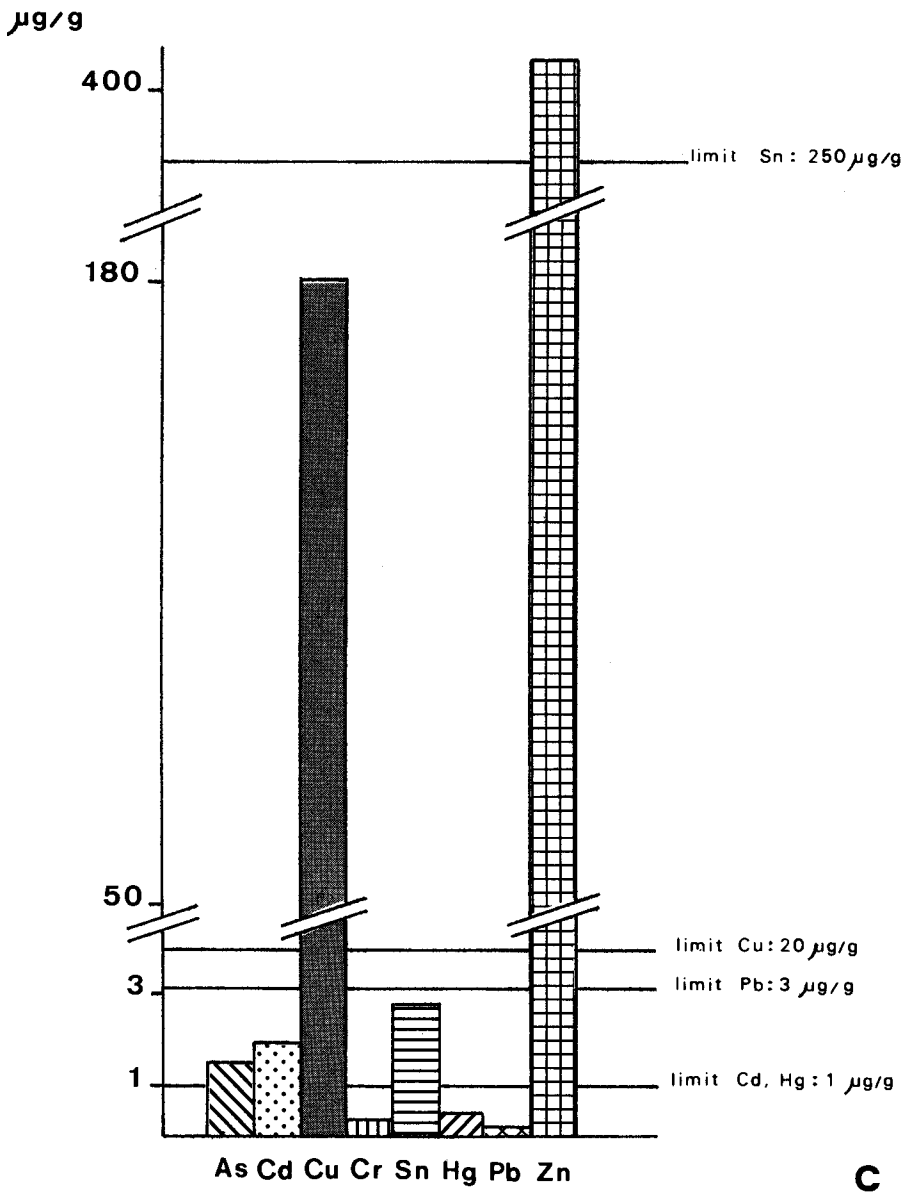

Figure 4.- Average concentration of the metals in oysters.

9.99 µg/g; similar values were obtained by other authors during the period 1976-1977 (Establier, 1987) in the area and in the three types of molluscs.

The high copper content in the oysters assayed could be explained both by the high copper contamination in this area and the great capacity of the oyster to accumulate this particular element.

The concentrations of mercury observed in clams varied between 0.78 and 1.18; in the oysters between 0.02 and 0.90 and in cockles between 0.02 and 1.27 µg/g. The values in clams were above the concentrations found previously in this species while those obtained in oysters and cockles were similar although odd specimens exceeded the average. These data confirmed that contamination with mercury was not very marked in this area.

The amount of cadmium in the three species were high compared with those found in the literature (Elder et al, 1984, Establier, R, 1975) reaching up to 50% above average in oysters, indicating that contamination with cadmium is high in the Huelva estuary.

The values for arsenic found in the three species were above the range (0.1-0.25 µg/g) observed by Elder (1984), however in English and in Portuguese oysters levels of up to 7.5 and 52.5 µg/g respectively have been found (Thorme et al, 1986)

Zinc was only assayed in oysters. The values were very similar to those obtained in the 1976-77 period on the coast of Cadiz and the Straits of Gibraltar and were below those found in the Huelva area in the same period (Establier, 1978).

In the literature revised the values of chromium in the oyster C. margaritacea on the Southafrican Coast fell between 0.1 and 0.9 µg/g (Watlerig et al, 1982); in the bivalves on the Huelva coast we observed concentrations ten times higher than this especially in clams and cockles.

The amount of lead turned out to be similar in clams and oysters while in cockles the value was duplicated. Our results were similar to those reported in the literature (Watlerig et al, 1982; Lytle et al, 1982).

The average concentrations of tin in the three species varied between 2.84 and 6.09 µg/g, well below the level permitted by Spanish Legislation, which is 250 µg/g.

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