

Heavy metal distribution in marine sediments from the southwest coast of Spain

José Morillo *, José Usero, Ignacio Gracia

*Department of Chemical and Environmental Engineering, University of Seville,
Camino de los Descubrimientos s/n, 41092 Seville, Spain*

Received 18 February 2003; received in revised form 29 September 2003; accepted 12 October 2003

Abstract

The latest version (1999) of the BCR-sequential extraction technique was used to determine the distribution of metals (Cu, Zn, Cd, Pb, Fe, Ni, Cr, and Mn) with major sedimentary phases (acid-soluble, reducible, oxidisable and residual) in samples from the southwest coast of Spain. The total metal content was also determined. The results showed that an extensive area along the coast (35 km long), near the joint mouth of the Tinto and Odiel Rivers, contains sediments with high concentrations of metals, with maximums of 649 mg/kg of Zn, 336 mg/kg of Cu, 197 mg/kg of Pb and 2.5 mg/kg of Cd. The values found for Cr, Ni and Mn are comparable to those in unpolluted areas. Based on the chemical distribution of metals, we found that Zn is the most mobile (i.e., it can pass easily into the water under changing environmental conditions). This metal showed the highest percentages in the acid-soluble fraction (the most labile), especially in the central coastal area, where the samples contained over 50% of this element associated with this fraction. This area close to the Tinto and Odiel river mouths also shows a significant increase in the mobility of Cd and Cu. In both cases the amount present in the residual fraction is lower, and the acid-soluble fraction is increased for Cd and the oxidisable fraction for Cu. However, the highest percentages of Fe, Cr and Ni are found in the residual fraction (84%, 89% and 75%, respectively), which implies that these metals are strongly bound to the sediments.
© 2003 Elsevier Ltd. All rights reserved.

Keywords: Metal pollution; Sediment; Metal distribution; Sequential extraction; Spain; Rio Tinto

1. Introduction

The 145 km long Atlantic coast of southwestern Spain lies between the mouths of two large rivers: the Guadiana, at its most western point, and the Guadalquivir, at its most eastern part. The other rivers that drain into this coastal area are the Carreras, the Piedras, the Tinto and the Odiel. These last two rivers join and drain together, forming the Huelva estuary (see Fig. 1). The Odiel River also flows into the Punta Umbría estuary.

The dominant winds in this area are south-westerly, which means that the surf comes into the coastline obliquely, generating a current parallel to the coastline (from left to right); this is known as the longshore drift and it has a high capacity for carrying matter.

The main sources of pollution of this coast are sewage from the coastal towns, industrial dumps and pollution from the rivers. With regard to this last source, most of the contaminants come from the Tinto and Odiel Rivers, both of which pass through the Iberian Pyrite Belt, a region long known for its mining activities (Leblanc et al., 2000). These rivers are acidic (the pH occasionally falls to 2 or 3) and contain large amounts of metals from erosion and mining (Nelson and Lamothe, 1993). Furthermore, sewage from the city of Huelva and

* Corresponding author. Tel.: +34-954-487-276.

E-mail address: jmorillo@esi.us.es (J. Morillo).

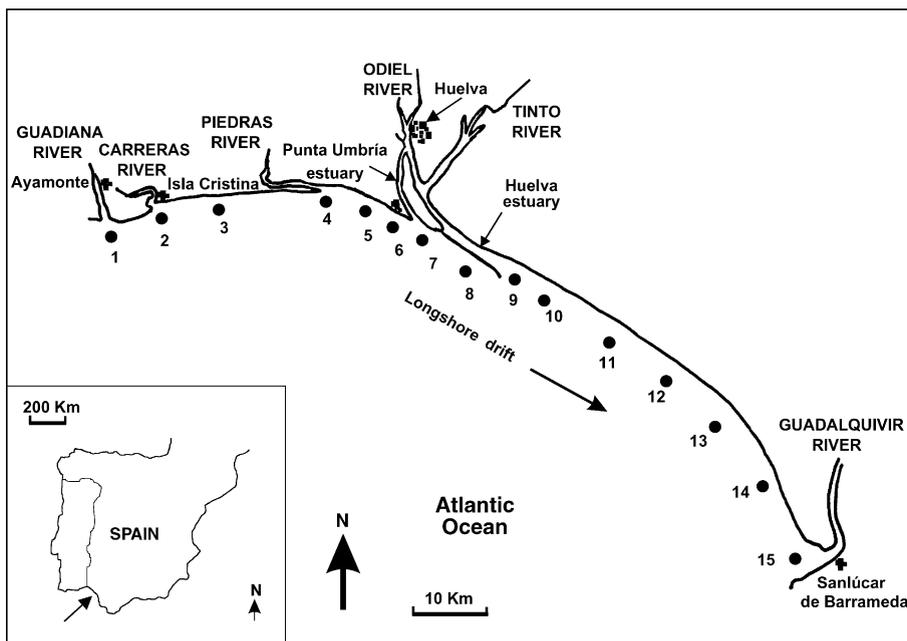


Fig. 1. Map of the area studied showing the location of the sampling points.

waste from industrial estates in the area with numerous chemical plants are discharged into the Huelva estuary. The amount of metal pollution from the other rivers that drain into the coastal area is considerably less (Nelson and Lamothe, 1993; Van Geen et al., 1997).

Information on total concentrations of metals alone is not sufficient to assess the environmental impact of polluted sediments because heavy metals are present in different chemical forms in sediments (easily exchangeable ions, metal carbonates, oxides, sulphides, organo-metallic compounds, ions in crystal lattices of minerals, etc.), which determine their mobilisation capacity and bioavailability (Lopez-Sanchez et al., 1996; Weisz et al., 2000; Kuang-Chung et al., 2001).

Several methods for determining the different forms of metals in sediments are described in the scientific literature (Kersten and Förstner, 1991; Lopez-Sanchez et al., 1993; Das et al., 1995). The most widely used methods are based on sequential extraction procedures whereby several reagents are used consecutively to extract operationally defined phases from the sediment in a sequence.

The purposes of this study of sediments from southwestern coast of Spain are: (a) to determine the metal pollution levels in sediments on this coast, which is an area where large amounts of contaminated waste from mining and industrial activities are discharged, and (b) to determine the forms or phases in which heavy metals are associated with the sediments, which will tell us about the bioavailability and mobility of these metals in the sediments—i.e., their ability to pass into water

when changes occur in environmental conditions (pH, sediment redox potential, etc.).

2. Material and methods

Fifteen sampling points were distributed throughout the area covered by this study at a distance of approximately 500 m from the coastline. Fig. 1 shows the location of the sampling points in the vicinity of the mouths of all the rivers (points 1, 2, 4, 7, 9 and 15). The greatest density of sampling points is around the Huelva estuary, where the mouths of the Tinto and Odiel Rivers are located.

Undisturbed surface sediment samples were collected with a grab sampler. Only grabs that showed no evidence of leakage or surface disturbance were retained; the top 3 cm were then removed and transferred to a cooler.

In the laboratory the samples were kept at 4 °C until processing and analysis. The metal analyses were performed in the <63- μm fraction, because the metals are usually associated with small grains (Dassenakis et al., 2003). This fraction is also easily homogenised for better reproducibility in metal measurements (Förstner and Salomons, 1988). Use of this fraction in pollution studies is recommended by several authors (Salomons and Förstner, 1984; Rauret et al., 1988; Thomas et al., 1994). All the sieving and sequential extraction procedures were performed in a glove box purged with nitrogen (Lopez-Sanchez et al., 1996).

The total metal content was determined by digesting the samples (0.5 g) with a mixture of concentrated HNO_3 (4 ml) and HClO_4 (2 ml) in a microwave oven at 600 W for 1 h (Kingston and Jassie, 1988). CEM 3010 high-pressure digestion bombs (consisting of a body made of a specific microwave-transparent polymer with a Teflon cup and cover) were used for sample digestion. These bombs are designed specifically for microwave heating, are chemically inert and combine the advantages of closed high-pressure (13.8 bar) and high-temperature digestion.

The chemical distribution of metals was determined by means of the sequential extraction scheme proposed by the European Union's Standards, Measurements and Testing program (SM&T, formerly BCR). A detailed description of the protocol can be found elsewhere (Rauret et al., 1999). This procedure, which has been improved as a result of extensive collaborative research studies (Rauret et al., 1999; Sahuquillo et al., 1999) consists of three successive extractions that allow us to associate the metals with one of the following phases:

Acid-soluble phase (fraction 1): This was obtained by adding 40 ml of acetic acid (0.11 mol l^{-1}) to a 1-g sample in a 100-ml centrifuge tube. After 16 h of shaking, the supernatant was separated from the solid residue by centrifugation. The residue was washed with 20 ml of distilled water, shaken for 15 min and centrifuged for 20 min. This phase is made up of exchangeable metals and others bound to carbonates that are able to pass easily into the water column, for example, when the pH drops. It is the fraction with the most labile bond to the sediment and, therefore, the most dangerous for the environment.

Reducible phase (fraction 2): This phase was removed by adding 40 ml of hydroxylamine hydrochloride (0.5 mol l^{-1} , acidified with HNO_3 0.05 mol l^{-1}) to the residue of the first step and proceeding in a similar fashion to the first step. This phase consists of metals bound to iron and manganese oxides that may be released if the sediment changes from the oxic to the anoxic state, which may be caused, for example, by the activity of microorganisms present in the sediments.

Oxidisable phase (fraction 3): This phase was removed with 10 ml of H_2O_2 (8.8 mol l^{-1} , acidified to pH 2–3 with HNO_3) added to the residue of the second step. After hot digestion for 1 h at 85°C and reduction of the liquid to a small volume, another 10 ml of H_2O_2 were added and the step repeated (1 h of digestion at 85°C and reducing the liquid to a small amount). The cool, moist residue was topped up with 50 ml of ammonium acetate (1.0 mol l^{-1} , acidified to pH 2 with HNO_3) and the procedures described for the previous steps were repeated. This phase shows the amount of metal bound to organic matter and sulphides that can be released under oxidising conditions. Such conditions can occur, for example, if the sediment is resuspended (by dredging,

currents, flooding, tides, etc.) and the sediment particles come into contact with oxygen-rich water.

Furthermore, a fourth phase—residual or inert (fraction 4)—was determined; this is the difference between the total metal content and the sum of the contents in the three previous phases. The metals found primarily in this fraction are those that are associated with minerals, forming part of their crystalline structure and which, as a result, are unlikely to be released from sediments.

The analysis of metals in the solutions obtained following sample digestion and sequential extraction was carried out by atomic absorption spectrophotometry (AAS) using a double-beam Perkin–Elmer 2380 with deuterium background correction and, in some cases (low concentrations), by graphite furnace atomic absorption spectrometry (GFAAS) using a Perkin–Elmer 4110 ZL with Zeeman background correction.

All reagents were Merck analytical grade or extra-pure quality. Standard working solutions of the elements analysed were prepared from the corresponding 1000 mg/l Merck titrisol solutions using the extractants employed in each step of the sequential extraction procedure (Lachica and Barahona, 1993) as matrices. All standard reagent solutions were stored in polyethylene bottles.

The accuracy of the analytical procedures for total metal determinations was checked using CRM 320 (sediment reference material). Replicate analysis of this CRM showed good accuracy, with recovery rates for metals between 89% and 102%. A standard reference material (CRM 601) was used to verify the accuracy of the sequential extraction method. The recovery rates for heavy metals in the standard reference material were between 85% and 105%. In order to determine the precision of the analytical processes, three samples were analysed in triplicate. The average values of the variation coefficients obtained (in general, less than 10%) can be considered satisfactory for environmental analysis.

The total metal concentrations underwent principal component analysis (a particular type of factor analysis) followed by varimax rotation, using the Biomedical Computer Program BMDP4M described by Dixon (1992). The aim of the technique is to reduce the number of variables to a smaller set of orthogonal factors making it easier to interpret by displaying the correlations existing among the original variables (Stella et al., 2002).

3. Results and discussion

3.1. Total metal content

As Fig. 2 shows, the total concentrations of Cd, Cu, Pb and Zn in the sediments from the southwestern coast

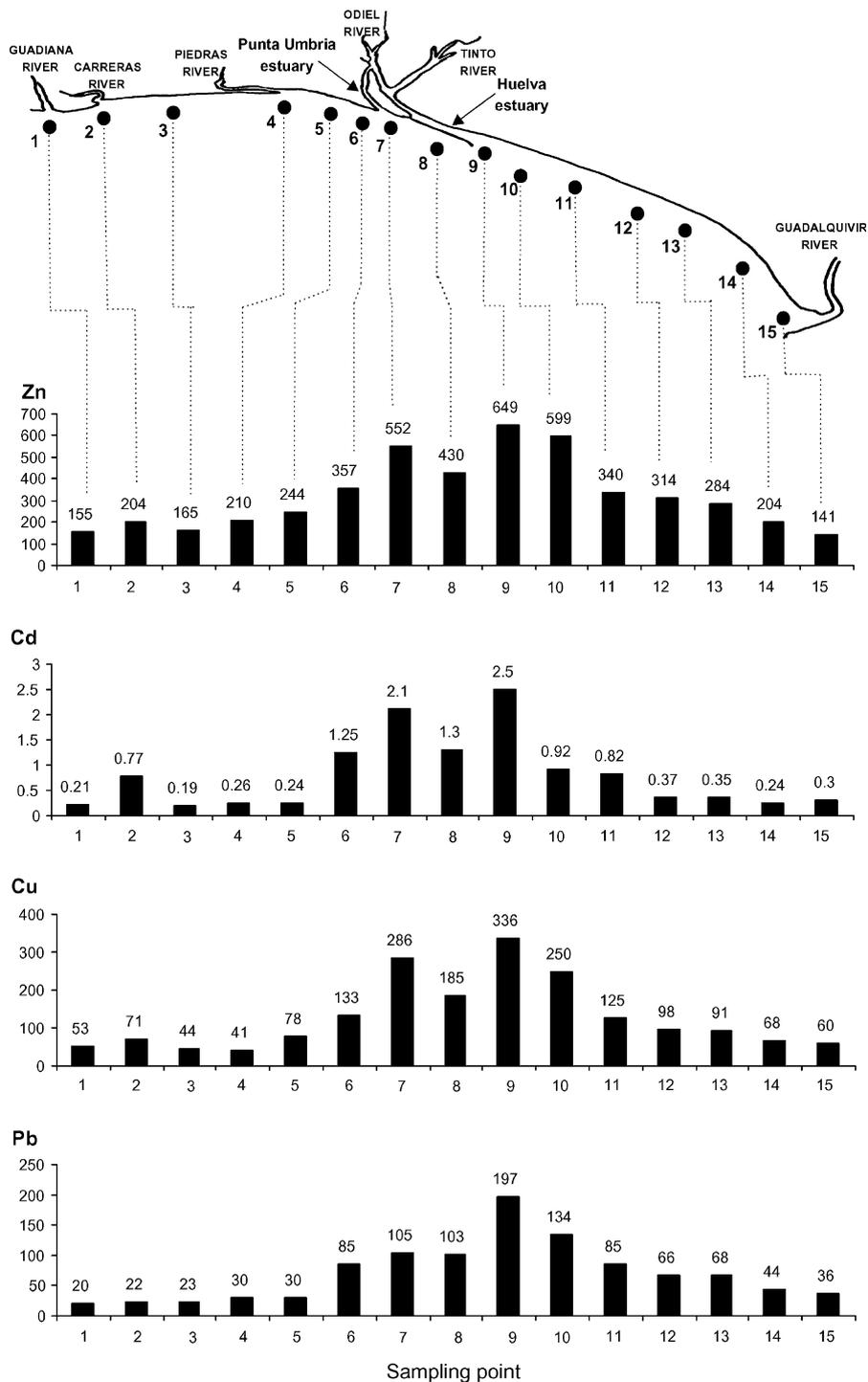


Fig. 2. Spatial distribution of Zn, Cd, Cu and Pb concentrations (mg/kg, dry mass).

of Spain have a similar spatial evolution characterised by the highest levels at points 6–10, which are located in the central area of the coast near the joint mouth of the

Tinto and Odiel Rivers (the Huelva estuary). The highest values are found at points 9 and 10 in the case of Zn and Pb (the points closest to the Huelva estuary) and the

second-highest values are from point 7 (the point closest to the Punta Umbría estuary). This implies that the inputs from the Tinto and Odiel Rivers are the main source of pollution for these metals along the coast. This is not surprising, considering that both rivers contain large amounts of dissolved and particulate metals from erosion and mining activity (Nelson and Lamothe, 1993; Palanques et al., 1995; Braungardt et al., 2003).

Fig. 2 also shows that, as a result of the longshore drift (the left-to-right current that flows along the coast), the content of these metals is higher at points 11–15 (located to the right of the central area) than at points 1–5 (located to the left). Furthermore, in the samples collected at points 11–15, there is a less-pronounced decrease in the concentrations of these metals because the longshore drift carries metals from the Tinto and Odiel Rivers toward these points. These results concur with those from the study by Elbaz-Poulichet et al. (2001), which demonstrated that the coastal waters carry dissolved metals from the Tinto and Odiel Rivers a distance of more than 200 km.

In general, we can state that the concentration levels for Cu, Cd, Pb, and Zn on this coast are similar to the average values found by Usero et al. (2000) on the southern coast of Spain, except at points 6–10, previously mentioned, which reach levels comparable to those found in marine sediments in polluted areas (Savvides et al., 1995; Lopez-Sanchez et al., 1996; Benamar et al., 1999).

Fig. 3 shows that Fe has the highest concentrations between points 6 and 10, although the differences are less pronounced than for the metals mentioned previously. The rest of the metals studied (Cr, Ni and Mn) behave very differently and show no significant increase in concentration in the central area. The highest values occur in sample 1, taken in the vicinity of the mouth of the Guadiana (36, 82 and 576 mg/kg of Ni, Cr and Mn, respectively) and in sample 15, taken near the mouth of the Guadalquivir River (50, 92 and 569 mg/kg of Ni, Cr and Mn, respectively).

The total metal concentration data underwent principal component analysis. Only components with Eigen values greater than 1 after rotation were included. The two principal components account for 84% of the total variance. Most of the commonalities are greater than 0.7, suggesting that the extraction of the two principal components is reasonable for this study. To illustrate the contribution of the different variables and samples to individual principal components, the distribution of the loads and object scores in the PC1–PC2 (principal component 1–principal component 2) axis is shown in Figs. 4 and 5. As Fig. 4 shows, the first component (most important), which accounts for 53% of the variance among the metals, is characterised by high positive contributions (loads) for Cd, Pb, Cu and Zn. 30.7% of the variance is accounted for by the second component,

which presents high positive loads for Cr, Mn and Ni. In Fig. 5, we see that the samples displaying positive values for the first principal component (6–11) come from the central coastal area near the mouth of the Tinto and Odiel Rivers, leading us to surmise that the main source of Cd, Pb, Cu and Zn in sediments from the southwest coast of Spain is input from these rivers. Samples 1 and 15 stand out because they present the highest values for the second principal component. These samples, as mentioned before, show the highest concentrations of Cr, Ni and Mn because of inputs from the Guadiana (sample 1) and Guadalquivir (sample 15) Rivers.

3.2. Metal distribution

Of the elements studied, Zn has the highest mobility, since it presents the highest content in fraction 1 (the most labile). This is particularly marked in the central coastal area, where the Zn associated with the acid-soluble fraction reaches levels of more than 50% of the total (see Fig. 6). The distribution found for Zn is not unusual; high percentages of total Zn have been found associated with more labile fractions in other studies of polluted sediments (Suriya and Branica, 1995; Lopez-Sanchez et al., 1996). Samples 2 and 15, taken in the vicinity of the Carreras and Guadalquivir river mouths, respectively, show a phase distribution for this element that is very different from the rest. The Zn in sample 2 is found in a high percentage in the oxidisable fraction (43%), probably due to the input of organic matter from urban sewage from the town of Isla Cristina, located close to the mouth of the Carreras River. In sample 15 the Zn is accumulated almost entirely in the residual fraction (91%), which indicates that Zn mobility in this area is very low.

The distribution of Cd among the four fractions varies considerably along the coast. In the western area (samples 1–5) about 70% of the total Cd is found in the residual fraction and between 15% and 25% in the acid-soluble fraction. From this we infer that Cd mobility in this area is low. In the central area (samples 6–10, taken around the Tinto and Odiel river mouths) there is a sharp increase in this metal's mobility. It is in this area where the Cd content in the residual fraction drops considerably and increases in the other fractions, especially in the acid-soluble fraction, where it is as high as 56% in sample 9. Finally, in the eastern area (samples 12–15), the mobility of Cd is midway between that of the other two areas and about 50% is associated with the residual fraction.

Among the metals studied, Cu is the one found with the greatest proportion in the oxidisable fraction (coinciding with organic and sulphur compounds). Cu can easily form complexes with organic matter due to the high stability constant of organic Cu compounds. This agrees with the results of many studies (Usero et al.,

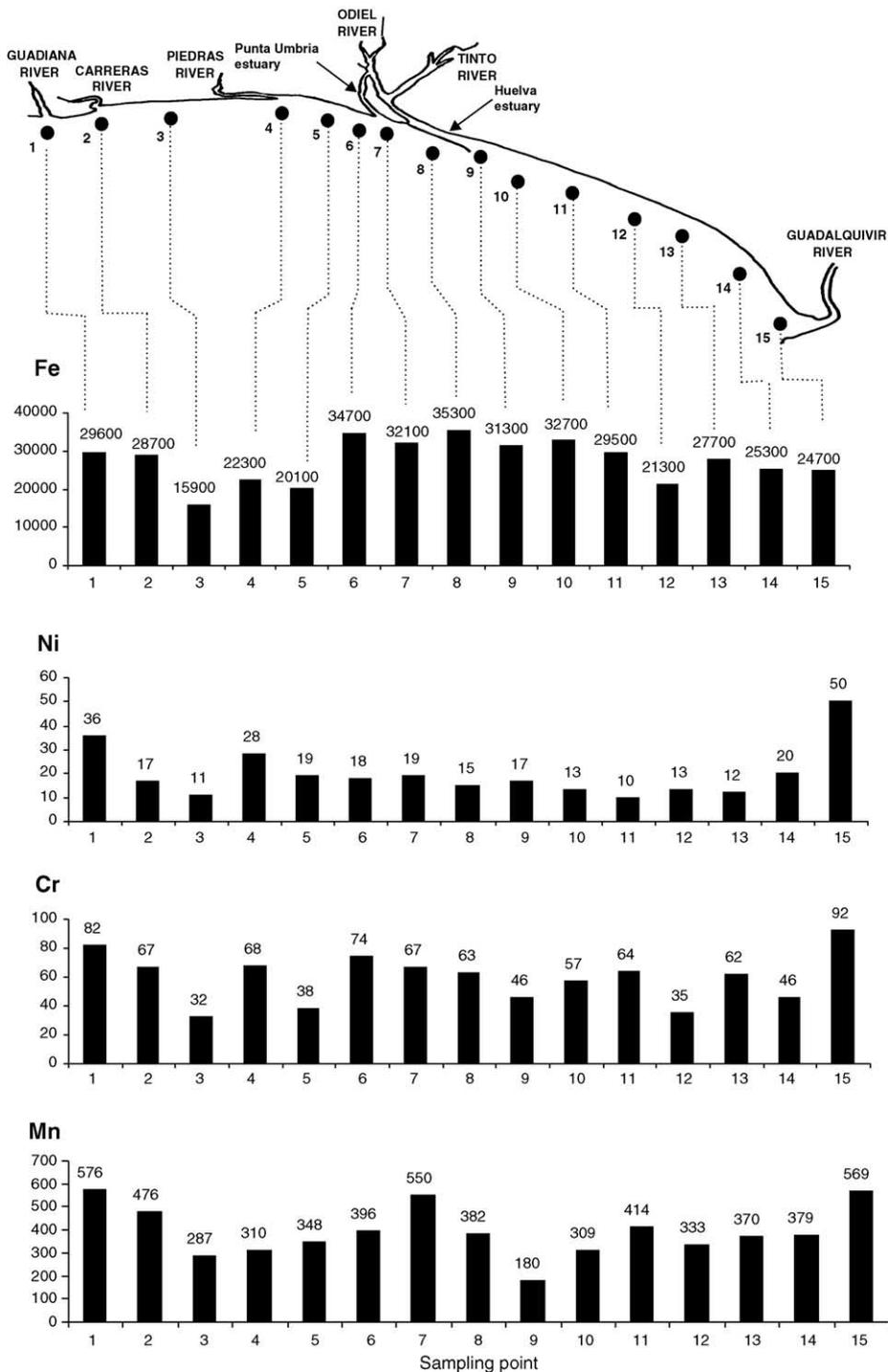


Fig. 3. Spatial distribution of Fe, Ni, Cr and Mn concentrations (mg/kg, dry mass).

1998; Zhou et al., 1998; Ramos et al., 1999), which state that a high proportion of Cu in the sediments may form part of the organic matter. For this metal also we note an increased percentage in the first three fractions of the

samples taken in the central coastal area, especially in the third (oxidisable) fraction. Likewise, sample 2 shows a high percentage in the oxidisable fraction; we should bear in mind that this sample comes from the vicinity

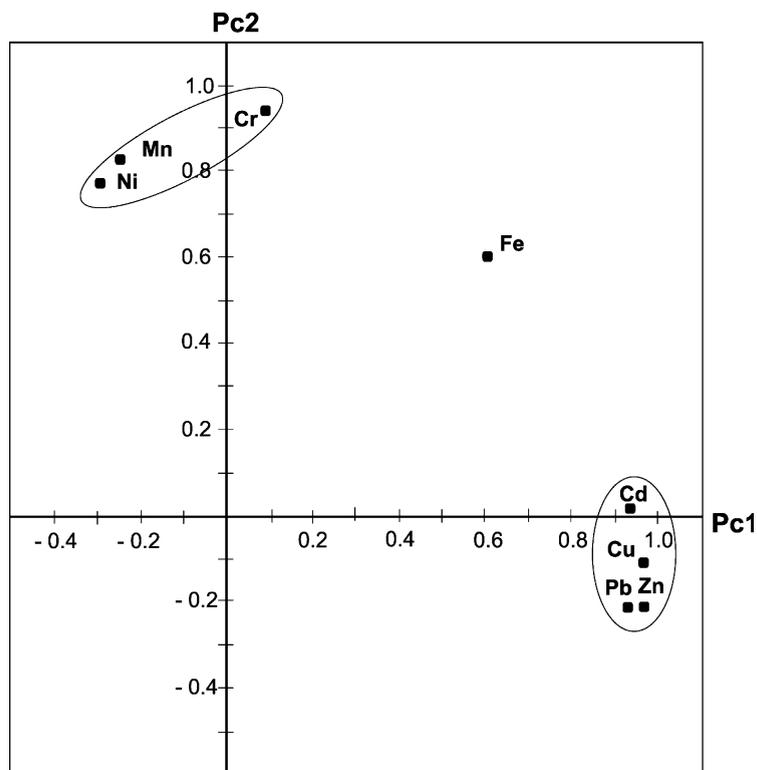


Fig. 4. Plots of the rotated principal component loads. The loads of the variables for principal component one are plotted against those for principal component two.

of the Carreras River mouth, which is a contributor of organic matter from sewage coming from Isla Cristina.

Most of the Pb accumulates in the residual fraction, which accounts for around 60% of the total. This is followed by the reducible fraction, which presents values of around 25% in most of the samples. This is consistent with results obtained by several authors (Hudson-Edwards et al., 1996; Dawson and Macklin, 1998) who found that Fe and Mn hydrous oxides are important scavengers of Pb in sediments. In sediments from this coast it is mainly the Fe oxides that scavenge Pb in the reducible fraction. A significant correlation ($p < 0.01$) was found between the Fe and Pb contents in the reducible fraction, whereas this is not so for Mn and Pb (reducible Mn shows a low correlation with reducible Pb). The presence of Pb in the oxidizable fraction is small, and practically negligible in the acid-soluble fraction. Based on these results, we can conclude that the mobility of Pb is low and that it basically depends on the dissolution of the reducible fraction. This is important, given that the concentrations of Pb in the sediments of the central area reach high levels, and Pb is a very toxic element for aquatic organisms and fish (Routh and Ikramuddin, 1996). It is worth noting that

for this metal, as opposed to the two previous ones, there is no increase in mobility in the central coastal area. This can be explained by the fact that a large part of the sediment-bound Pb in the central area consists of insoluble mineral particles rich in Pb (e.g., plumbogonite) that are transported by the Tinto and Odiel Rivers down to the mouths in the central area, without having undergone any weathering in transit (Hudson-Edwards et al., 1999; Leblanc et al., 2000). Other studies carried out in the Tinto and Odiel Rivers also show low mobility for Pb (Perez et al., 1991; Van Geen et al., 1997; Morillo et al., 2002).

Fe is found mainly in the residual fraction, with percentages around 90 along the coast except for the central area where it is around 75% (see Fig. 7). The rest of the Fe is distributed between the reducible and oxidizable fractions. The great accumulation of Fe in the residual fraction is probably due to its being an element whose origin is fundamentally natural (it is one of the most common elements in the Earth's crust).

Ni and Cr show similar behaviour. They are found mainly in the residual fraction in all the coastal samples and the content of these two metals in this fraction shows a significant correlation ($p < 0.01$). These results are in agreement with other sediment studies

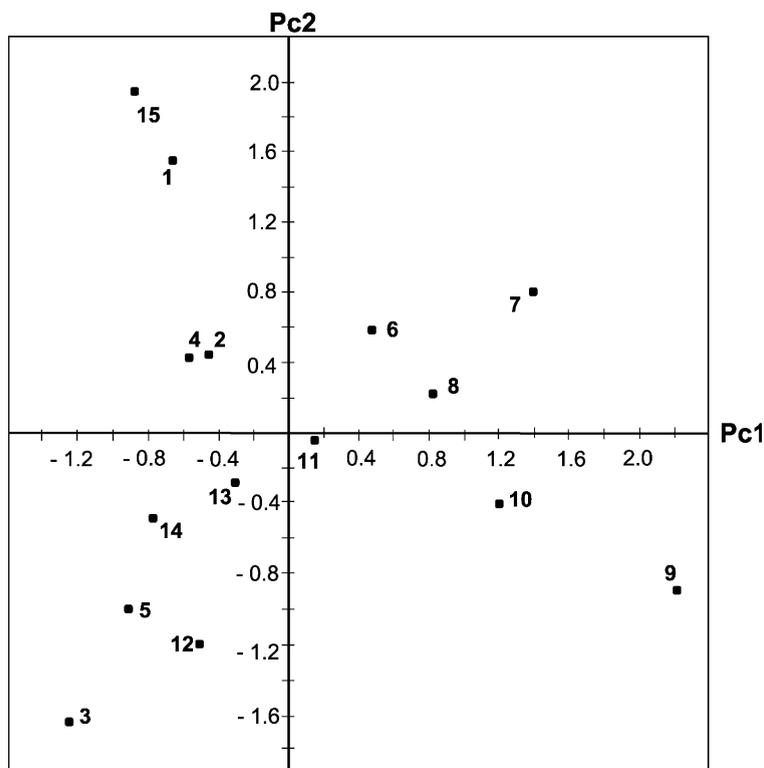


Fig. 5. Plots of object sample scores in space spanned by principal component one and principal component two.

(Lopez-Sanchez et al., 1996; Jones and Turki, 1997; KARBASSI, 1998) in which large amounts of Ni and Cr were found in the residual fraction. Thus, given that these elements have low mobility and are found in small concentrations in the sediments studied, it is unlikely that the sediments of this coast are an important source of Ni and Cr for its waters.

For Mn we find no variations worthy of mention in its distribution along the coast. It is associated mainly with the residual fraction, with an average of 57%, and with an average of 25% in the acid-soluble fraction. This distribution is similar to that found by Marin and Giresse (2001) in sediments from the Gulf of Lions in the Mediterranean Sea (France).

4. Conclusions

In the central area of Spain's southwestern coast, near the joint mouth of the Tinto and Odiel Rivers, there are high concentrations of Cd, Cu, Pb and Zn, which demonstrates the importance of the contribution of the Tinto and Odiel Rivers to the contamination of the sediments along the coast. We note also that the eastern coastal area is affected more than the western area by contributions from these rivers, which has its explana-

tion in the longshore drift (the left-to-right current that flows along the coast). We can also conclude from our study that the influence of the other rivers is considerably less. Only in the case of Cr, Ni and Mn is there an increase in their content in the mouths of the Guadiana and Guadalquivir Rivers, although less than for the metals previously discussed.

This study provides valuable information on the potential mobility of heavy metals in marine sediments. Zn is the metal that presents the greatest mobility. Large amounts of this element are present in the acid-soluble fraction, especially in the central coastal area, and it can therefore be expected that the exchange of this element between sediment and the water column will take place easily. The distribution of Cd among the different phases varies widely along the coast and shows the lowest mobility in the eastern area, intermediate mobility in the western area and the highest in the central area. There is a striking increase in the percentage found in the acid-soluble fraction in this last area, similarly to that observed for Zn. Cu is the element showing the highest proportion in the oxidisable fraction, especially in the central coastal area, highlighting the important role played by organic matter, which is responsible for the accumulation of Cu from the Tinto and Odiel Rivers in sediments. In spite of the high concentrations of Pb (a

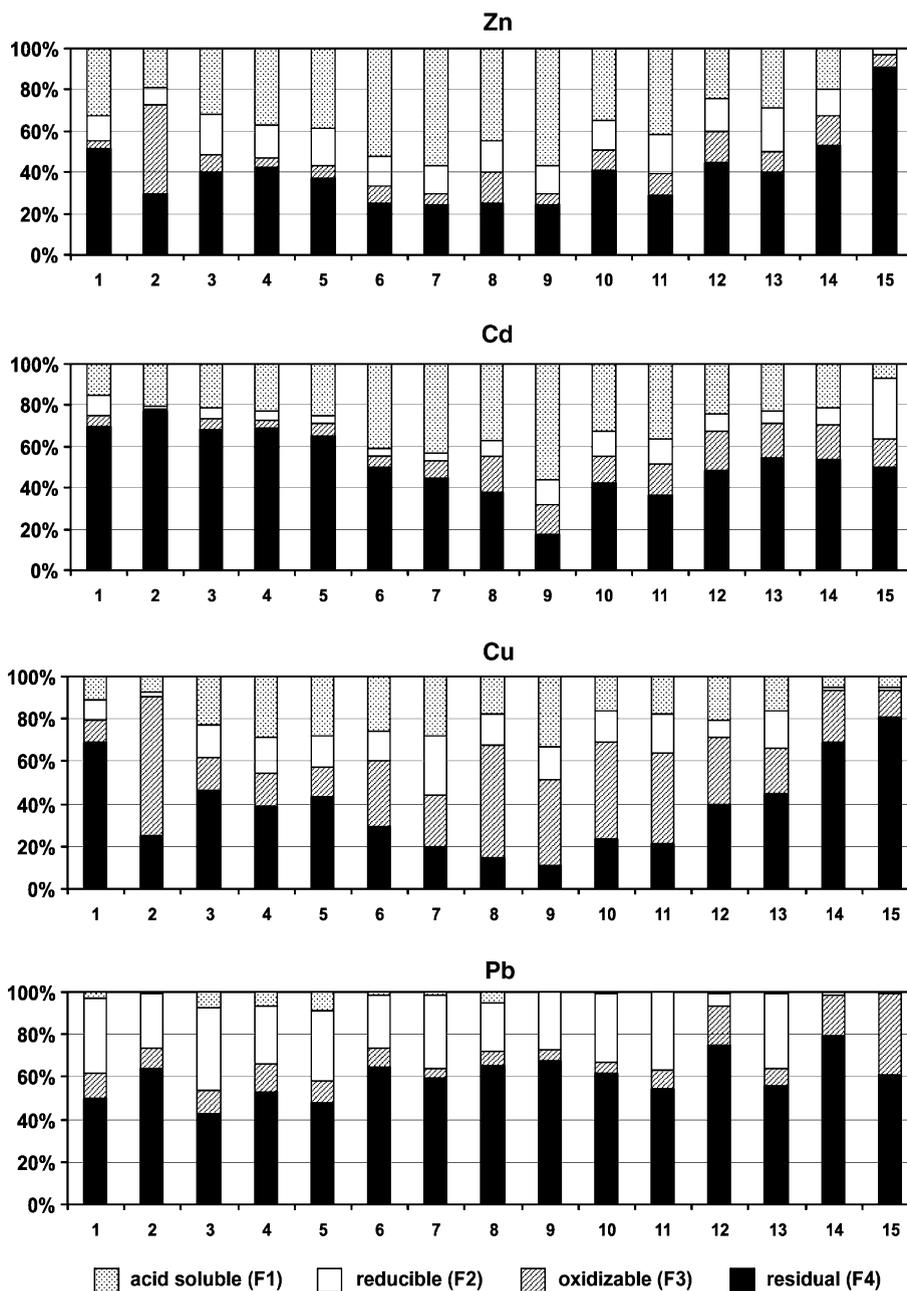


Fig. 6. Zn, Cd, Cu and Pb distribution in the different phases. The percentage of metal removed in each step of the extraction procedure is represented as a bar chart.

highly toxic metal) in the sediments of the central coastal area, it is not likely that this element is a major hazard for the aquatic environment since it is found mainly in the residual fraction. Mn shows intermediate mobility because, although it accumulates mainly in the residual fraction, there is a large amount in the acid-soluble fraction. The elements with the least mobility are Fe, Ni

and Cr since they reach the greatest percentages in the residual fraction.

The metals studied can be ranked as follows according to the percentage of each in the acid-soluble fraction (the most mobile and bioavailable):

$$\text{Zn} > \text{Mn} > \text{Cd} > \text{Cu} > \text{Ni} > \text{Pb} > \text{Fe} > \text{Cr}$$

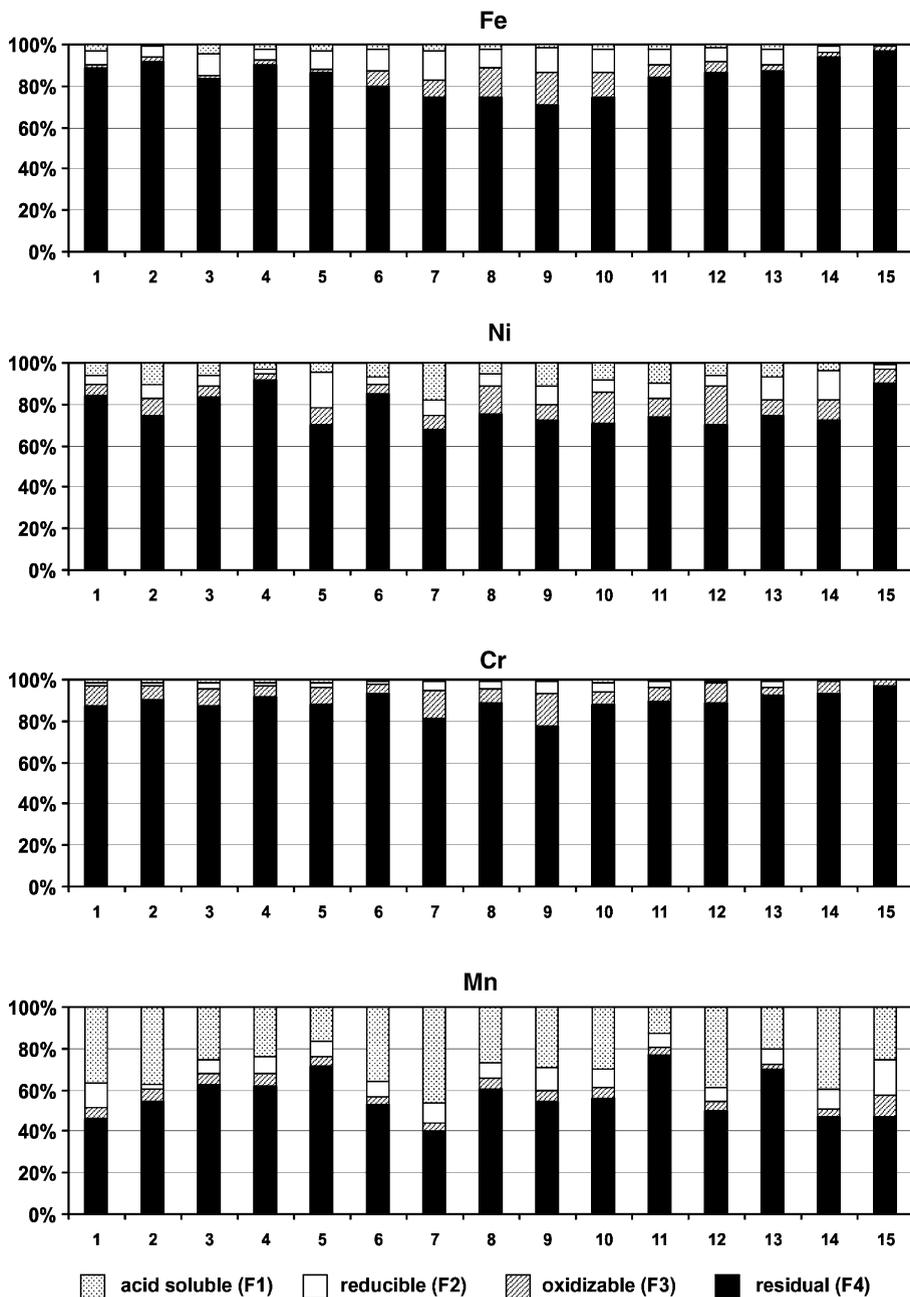


Fig. 7. Fe, Ni, Cr and Mn distribution in the different phases. The percentage of metal removed in each step of the extraction procedure is represented as a bar chart.

Acknowledgements

This research was supported by the Environmental Council of the Junta de Andalucía (the Andalusian Governing Body), Spain.

References

Benamar, M.A., Toumert, I., Tobbeche, S., Tchanchane, A., Chalabi, A., 1999. Assessment of the state of pollution by heavy metals in the surficial sediments of Algiers Bay. *Appl. Radiat. Isotopes* 50, 975–980.

- Braungardt, C.B., Achterberg, E.P., Elbaz-Poulichet, F., Morley, N.H., 2003. Metal geochemistry in a mine-polluted estuarine system in Spain. *Appl. Geochem.* 18, 1757–1771.
- Das, A.K., Chakraborty, R., Cervera, M.L., de la Guardia, M., 1995. Metal speciation in solid matrices. *Talanta* 42, 1007–1030.
- Dassenakis, M., Andrianos, H., Depiazi, G., Konstantas, A., Karabela, M., Sakellari, A., Scoullou, M., 2003. The use of various methods for the study of metal pollution in marine sediments, the case of Euvoikos Gulf, Greece. *Appl. Geochem.* 18, 781–794.
- Dawson, E.J., Macklin, M.G., 1998. Speciation of heavy metals in floodplain and flood sediments: a reconnaissance survey of the Aire Valley, West Yorkshire, Great Britain. *Environ. Geochem. Health.* 20, 67–76.
- Dixon, W.J., 1992. *BMDP Statistical Software Manual*. University of California Press, Berkeley. 201–225.
- Elbaz-Poulichet, F., Morley, N.H., Beckers, J.M., Nomerange, P., 2001. Metal fluxes through the Strait of Gibraltar: the influence of the Tinto and Odiel rivers (SW Spain). *Mar. Chem.* 73, 193–213.
- Förstner, U., Salomons, W., 1988. Trace metals analysis on polluted sediments. I. Assessment of sources and intensities. *Environ. Technol. Lett.* 1, 494–505.
- Hudson-Edwards, K.A., Schell, C., Macklin, M.G., 1999. Mineralogy and geochemistry of alluvium contaminated by metal mining in the Rio Tinto area, southwest Spain. *Appl. Geochem.* 14, 1015–1030.
- Hudson-Edwards, K.A., Macklin, M.G., Curtis, C.D., Vaughan, D., 1996. Mineralogical forms, processes of formation and distribution of Pb, Zn, Cd and Cu in the Tyne catchment basin, Northern Pennines, North East England. *Environ. Sci. Technol.* 30, 72–80.
- Jones, B., Turki, A., 1997. Distribution and speciation of heavy metals in surficial sediments from the Tees estuary, north-east England. *Mar. Pollut. Bull.* 34 (10), 768–779.
- Karbassi, A.R., 1998. Geochemistry of Ni, Zn, Cu, Pb, Co, Cd, V, Mn, Fe, Al and Ca in sediments of North Western part of the Persian Gulf. *Int. J. Environ. Stud.* 54, 205–212.
- Kersten, M., Förstner, U., 1991. Speciation of trace elements in sediments. In: Batley, G.E. (Ed.), *Trace Element Speciation: Analytical Methods and Problems*. CRC Press, Boca-Ratón, FL, pp. 245–317.
- Kingston, H.M., Jassie, L.B., 1988. Introduction to microwave sample preparation. In: Kingston, H.M., Jassie, L.B. (Eds.), *ACS Professional Reference Book*, American Chemical Society, Washington DC, 263 pp.
- Kuang-Chung, Y., Li-Jyur, T., Shih-Hsiuns, C., Shien-Tsong, H., 2001. Correlation analyses binding behavior of heavy metals with sediment matrices. *Water Res.* 35 (10), 2417–2428.
- Lachica, M., Barahona, E., 1993. The determination of trace elements by flame atomic absorption spectrometry: effects of the composition of standard solution matrices. *Int. J. Environ. An. Ch.* 51, 219–221.
- Leblanc, M., Morales, J.A., Borrego, J., Elbaz-Poulichet, F., 2000. 4500 year-old mining pollution in southwestern Spain: long-term implications for modern mining pollution. *Econ. Geol.* 95, 655–662.
- Lopez-Sanchez, J.F., Rubio, R., Rauret, G., 1993. Comparison of two sequential extraction procedures for trace metal partitioning in sediments. *Int. J. Environ. An. Ch.* 51, 113–121.
- Lopez-Sanchez, J.F., Rubio, R., Samitier, C., Rauret, G., 1996. Trace metal partitioning in marine sediments and sludges deposited off the coast of Barcelona (Spain). *Water Res.* 30 (1), 153–159.
- Marin, B., Giresse, P., 2001. Particulate manganese and iron in recent sediments of the Gulf of Lions continental margin (north-western Mediterranean Sea): deposition and diagenetic process. *Mar. Geol.* 172, 147–165.
- Morillo, J., Usero, J., Gracia, I., 2002. Heavy metal fractionation in sediments from the Tinto river (Spain). *Int. J. Environ. An. Ch.* 82 (4), 245–257.
- Nelson, C.H., Lamothe, P.J., 1993. Heavy metal anomalies in the Tinto and Odiel river and estuary system, Spain. *Estuaries* 16 (3A), 496–511.
- Palanques, A., Diaz, J.I., Farran, M., 1995. Contamination of heavy metals in the suspended and surface sediment of the Gulf of Cadiz Spain: the role of sources, currents, pathways and sinks. *Oceanolog. Acta* 18 (4), 469–477.
- Perez, M., Usero, J., Gracia, I., Cabrera, F., 1991. Trace metals in sediments from the “Ria de Huelva”. *Toxicol. Environ. Chem.* 31–32, 275–283.
- Ramos, L., González, M.J., Hernández, L.M., 1999. Sequential extraction of copper, lead, cadmium, and zinc in sediments from Ebro river (Spain): relationship with levels detected in earthworms. *Bull. Environ. Contam. Toxicol.* 62, 301–308.
- Rauret, G., Rubio, R., López-Sánchez, J., Casassas, E., 1988. Determination and speciation of copper and lead in sediments of a mediterranean river (river Tenes, Catalonia, Spain). *Water Res.* 22, 449–455.
- Rauret, G. et al., 1999. Improvement of the BCR three step sequential extraction procedure prior to the certification of new sediment and soil reference materials. *J. Environ. Monitor.* 1, 57–61.
- Routh, J., Ikramuddin, M., 1996. Trace-element geochemistry of Onion Creek near Van Stone lead–zinc mine (Washington, USA). *Chem. Geol.* 133, 211–224.
- Sahuquillo, A. et al., 1999. Use of a certified reference material for extractable trace metals to assess sources of uncertainty in the BCR three-stage sequential extraction procedure. *Anal. Chim. Acta* 382, 317–327.
- Salomons, V., Förstner, U., 1984. *Metals in the Hydrocycle*. Springer-Verlag, Heidelberg.
- Savvides, C., Papadopoulos, K.J., Loizidou, M., 1995. Sea sediments contaminated with heavy metals: metal speciation and removal. *Water Sci. Technol.* 32 (9–10), 65–73.
- Stella, A., Piccardo, M.T., Coradeghini, R., Redaelli, A., Lanteri, S., Armanino, C., Valerio, F., 2002. Principal component analysis application in polycyclic aromatic hydrocarbons “mussel watch” analyses for source identification. *Anal. Chim. Acta* 461, 201–213.
- Suriya, B., Branica, M., 1995. Distribution of Cd, Pb, Cu and Zn in carbonate sediments from the Krka river estuary obtained by sequential extraction. *Sci. Total Environ.* 170, 101–118.
- Thomas, R.P. et al., 1994. Three-stage sequential extraction procedure for the determination of metals in river sediments. *Anal. Chim. Acta* 286, 423–429.

- Usero, J., Gamero, M., Morillo, J., Gracia, I., 1998. Comparative study of three sequential extraction procedures for metals in marine sediments. *Environ. Int.* 24 (4), 487–496.
- Usero, J., Gracia, I., Leal, A., Fraidiás, J., 2000. Calidad de las Aguas y Sedimentos del Litoral de Andalucía, Plan de Policía (1995–1998), Junta de Andalucía, Consejería de Medioambiente, Sevilla.
- Van Geen, A., Adkins, J.F., Boyle, E.A., Nelson, C.H., Palanques, A., 1997. A 120 year record of widespread contamination from mining of the Iberian pyrite belt. *Geology* 25 (4), 291–294.
- Weisz, M., Polyák, K., Hlavay, J., 2000. Fractionation of elements in sediment samples collected in rivers and harbors at lake Balaton and its catchment area. *Microchem. J.* 67, 207–217.
- Zhou, H.Y., Cheung, R.Y.H., Chan, K.M., Wong, M.H., 1998. Metal concentrations in sediments and *tilapia* collected from inland waters of Hong Kong. *Water Res.* 32 (11), 3331–3340.