SEQUENTIAL EXTRACTION AND ANALYSIS BY ELECTROTHERMAL ATOMIC ABSORPTION SPECTROMETRY OF CADMIUM AND LEAD IN MARINE SEDIMENTS

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ABSTRACT: Study of sediments can provide valuable data to assess the degree of sea water pollution. Herein, we investigate two toxic elements; cadmium and lead, Their analysis requires a highly-sensitive analytical method. ETAAS, with a thermal program adapted to the nature of the sample, has been used. The determination of the total amount of metals does not provide data to indicate the risk of toxicity, or even to understand the various geochemical processes. For these reasons, chemical fractionating (or speciation) is now widely used. It is important to determinate the sediment fractions with which the metals are associated. In this study, a comparison of three and five-step sequential extraction schemes has been established. For ETAAS, the use of calibration curves and thermal programs for each fraction is necessary to give satisfactory results. The procedures of analysis is applied to Pb and Cd in two reference sediments with certified total Pb and Cd content and to different sediments from ports of the Gulf of Tunis. The results show that the three step sequential extraction gives better recoveries .

Key words: Sequential extraction, ETAAS, lead, cadmium, marine sediments

RESUME: L'étude des sédiments marins donne une information pertinente sur l'état de pollution de l'eau de mer en un point donné. Parmi les contaminants nous avons retenu pour notre étude deux éléments métalliques: plomb et cadmium. Leur analyse nécessite une recherche du protocole opératoire de traitement de l'échantillon le plus appropriée et une méthode analytique de grande sensibilité. Ainsi, nous avons utilisé la Spectrométrie d'Absorption Atomique Electrothermique (SAAE) avec un programme thermique adapté à la nature de l'échantillon. Le dosage des métaux totaux ne donne que des renseignements partiels sur leur degré de toxicité, ainsi on a utilisé la méthode de fractionnement chimique ou d'extraction séquentielle. Au cours de ce travail, nous avons comparé les résultats de la méthode d'extraction à 5 étapes avec celle à 3 étapes. Pour la SAAE nous avons optimisé un programme thermique et une courbe d'étalonnage spécifique pour chaque fraction. La procédure d'analyse et d'optimisation du protocole de minéralisation du plomb et du cadmium est appliquée sur des sédiments de référence standard et sur des échantillons de sédiments prélevés de quelques stations proches de différents ports du Golfe de Tunis. Les résultats montrent que l'extraction séquentielle à 3 étapes donne un meilleur recouvrement.

Mots clés : extraction séquentielle, SAAE, plomb, cadmium, sédiments marins

INTRODUCTION

The eco-toxicity and mobility of metals in the environment depend strongly on their specific chemical configuration or kind of binding rather than on the total element contents. Therefore, it is very important to study the distribution of metals in the different sediment phases. The sequential extraction technique is used for the assessment of the different forms of trace metals (e.g. mobile, bioavailable, carbonate-bound.). This method of selective extraction highlights the difference in geochemical behaviour of trace metals like Cu, Zn, Pb and Cd, according to their natural or anthropogenic origin [1]. Tessier et al [2] proposed a five step sequential extraction of toxic metals in sediments. This method does not allow a good recovery. A better method was proposed by a group of European experts in order to improve the extraction efficiency in heavily polluted sediments. which is a three-step sequential extraction, of Cd, Cr, Cu, Ni, Pb and Zn in sediments and soils [3-6]. These authors use the same thermal program for the analysis of each fraction.

In this study we used five and three sequential extractions, the metal content in each fraction was mainly determined by ETAAS, and a thermal program was used to analyse trace metals in diverse fractions. We established the best instrumental conditions to avoid the matrix effects because the data depend on experimental conditions [7,8]. An optimization of furnace program and calibration curve was realized for each fraction because of the matrix effect.

The validation conditions for Pb and Cd determination in the extracts are applied to study the Pb and Cd partitioning in two sediments for which the total Pb and Cd content is certified. Finally, samples from ports of the Gulf of Tunis are also studied.

EXPERIMENTAL

Apparatus

- An electrothermal atomic absorption spectrometer « Varian Spectr AA 10 Plus » equipped with a graphite tube atomizer « GTA 96 » was used. Pyrotically coated tubes were used for the measurement of Cd and Pb. As background correction a deuterium lamp was automatically selected. Pb and Cd hollow cathod lamps were used as radiation sources. The measurements were performed at a wavelengh of 283.3 nm for Pb and 228.8 nm for Cd and peak area measurements were employed.

Argon was used as carrier gas (3 bars). The slit was 0.5 nm.

- For digestion a microwave digestion system « MDS-2000 » which consists of a combination of temperature and pressure with time and power control was used.

Reagents

Standard stock solutions of 1000 mg/l of $Cd(NO_3)_2$ and $Pb(NO_3)_2$ (Merck, pro-analysis) in 0.5 mol/l HNO_3 diluted working solutions were prepared daily with the corresponding final medium for each of the steps of the sequential extraction procedure.

The reagents used for the preparation of the extracting agents are :NaOAc Prolabo, $NH_2OH.HCl$ Fluka pro-analysis, CH_3COOH Merck suprapur, H_2O_2 30% (V/V) MerckCH_3COONH_4 Merck, pro-analysis, HNO_3 , HF, $HClO_4$ Merck suprapur.

Sediment samples

The reference materials used for the application of the method of sequential extraction and the optimization of the analytical method are SDM-2/TM and BCSS-1.

SDM-2/TM (International Atomic Energy Agency Analytical Quality Control Service Laboratory Seibersdorf Vienna Austria) were collected using a large box corer, in July 1987 in the Mediterranean sea at a depth of 1240 meters. This sediment was deep-frozen on board the ship immediately after collection, then, it was freeze-dried in the laboratory, ground, passed through a 150 µm seive and homogenized for 10 days [9]. This material was produced in cooperation with the Regional Seas Prograae of UNEP. A sediment for BCSS-1 (National Research Council of Canada Marine Analytical Chemistry Standards Program Division of Chemistry Canada) was collected in the Baie des Chaleurs, Gulf of St.Laurence. The sediments were freeze-dried, screened to pass through a 125 µm Screen, blended, bottled, and radiation-sterilized [10]. (table I)

Table I: *Concentration of Pb and Cd in reference materials*

Standard reference	**[Cd]** mg/Kg	**[Pb]** mg/Kg
SDM-2/TM	0.113 ± 0.041	22.8 ± 5.5
BCSS-1	0.25 ± 0.04	22.7 ± 3.4

The sequential extraction scheme was also applied to four samples taken from the Gulf of Tunis. These points are sampled by our services in INSTM, within the framework of marine pollution assessment in different ports of the Gulf of Tunis (Rades and Goulette). (figure 1)

station 1: reference station in the sea far from pollution

station 2 and 3: situated in port of Goulette known by their heavy marine trafic

station 4: situated in port of Rades near the industrial wastes zone from Megrine and Djebel-djloud

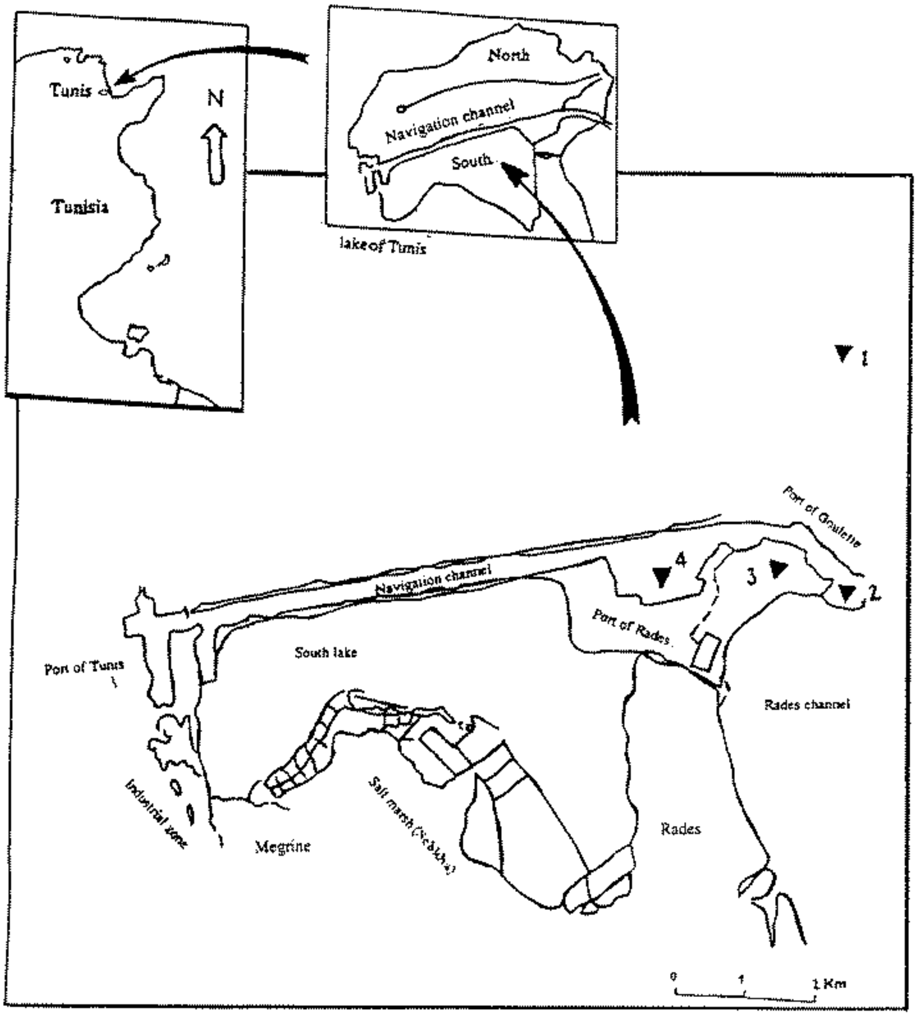

Figure 1: Map of sampling sites

Sequential extraction procedure

Two protocols were used: the first protocol has been described in the proceedings of the workshop on the sequential extraction of trace metals in soils and sediments held in Sitges [3] (figure 2), the second protocol was proposed by Tessier et al [2] (figure 3).

Procedure for total metal determination [11]

0.3 g of the two standard materials (SDM-2/TM and BCSS-1) was digested in a closed Teflon vessel for 1 hour with a mixture of HNO_3, HF (1:2) in a microwave according to the following program :

step 1	30mn	35% power
step 2	20mn	50% power
step 3	10mn	100% power

After cooling and evaporating to dryness at 100°C on a hot plate, mixtures of $HClO_4$ and HF (1:2) and (1:4) were added alternatively and then evaporated each time to dryness at 150°C. Finally 1 ml of concentrate HNO_3 was added to the treated sample and the wall of the container was rinsed with milli-Q water. The digestion vessel was closed tightly, placed in microwave oven and digested at 35% power for 20mn. The residue was filtered then the volume was made up to 50 ml with 2% nitric acid in a volumetric flask.

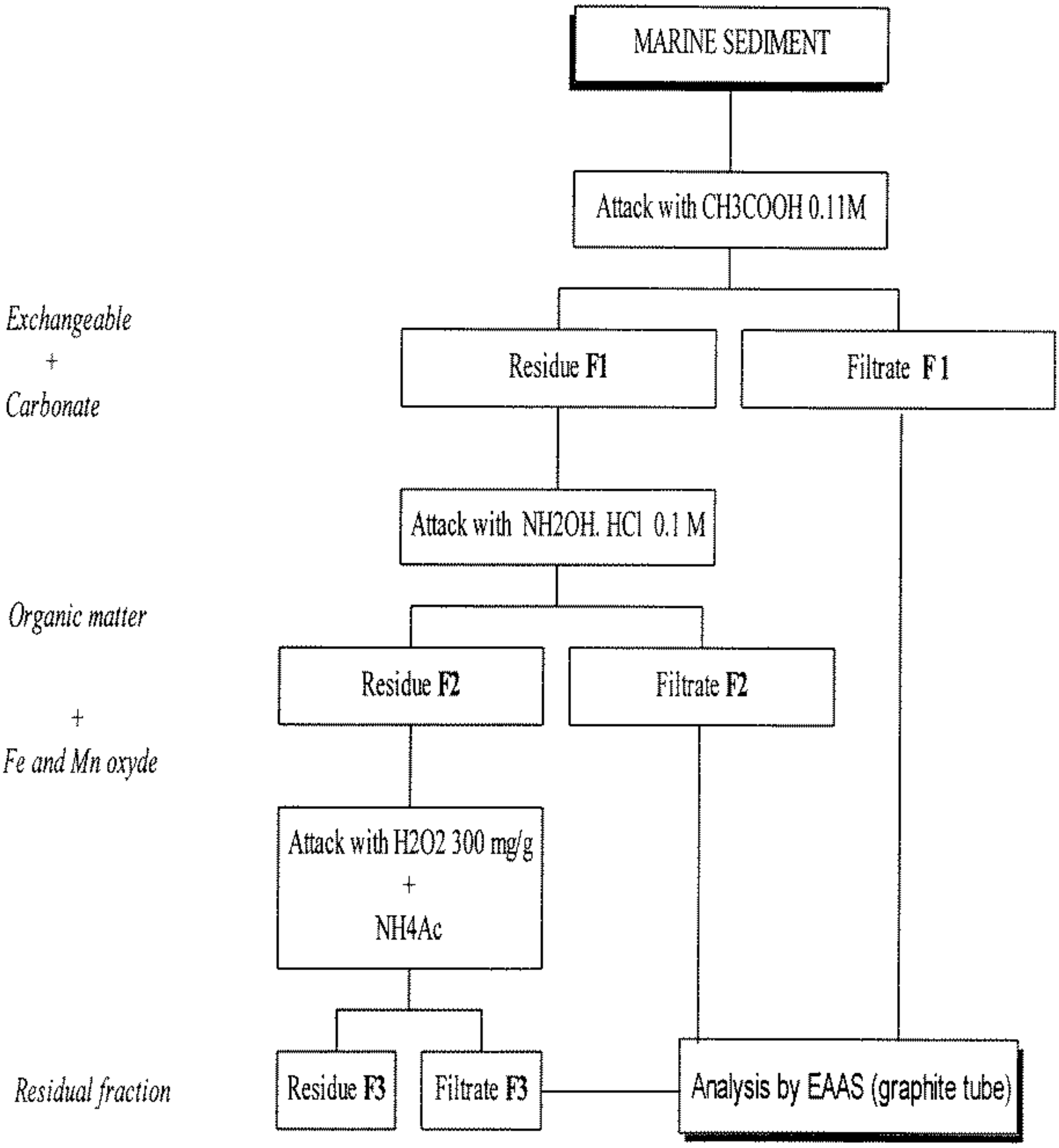

Figure 2:*Digramme of 3 step sequential extractions*

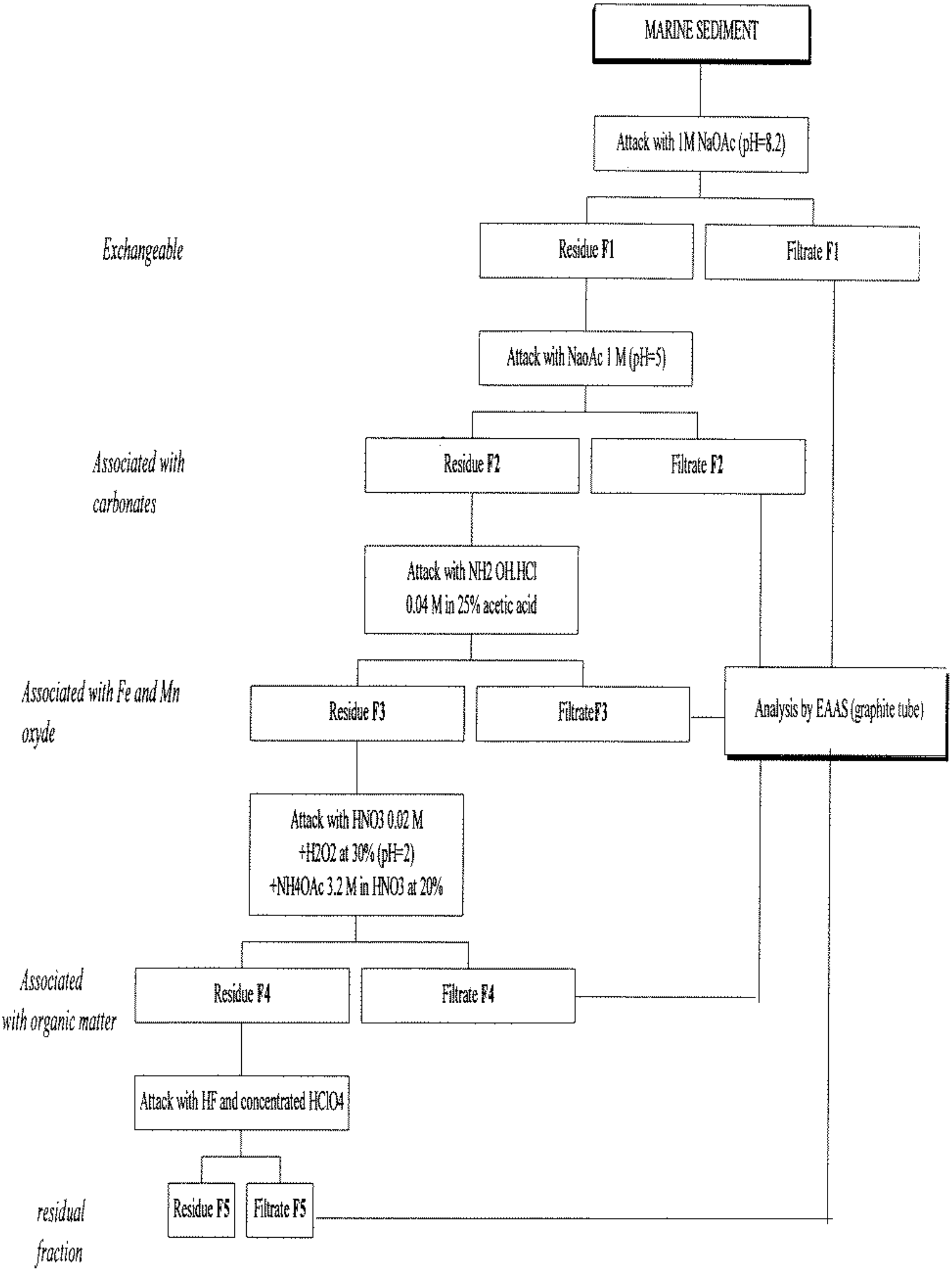

Figure3: *Digramme of 5 step sequential extractions*

RESULTS AND DISCUSSION

Optimisation of the instrumental conditions : *thermal program and calibration curves*

Decomposition and atomisation temperature require a particular study. Figure 4 represents optimisation of the decomposition temperature of Pb with atomisation temperature of 2000°C. A decrease or a reduction of absorbance for a decomposition temperature higher than 450°C is noted. Figure 5 summarises the optimisation protocol of lead atomisation temperature for a decomposition temperature of 450°C. This optimisation protocol was realized for all fractions in the case of five and three step sequential extractions. For total Pb and Cd, the results obtained are shown in table II. Temperatures change with the nature of fractions. The program used is detailed in table III.

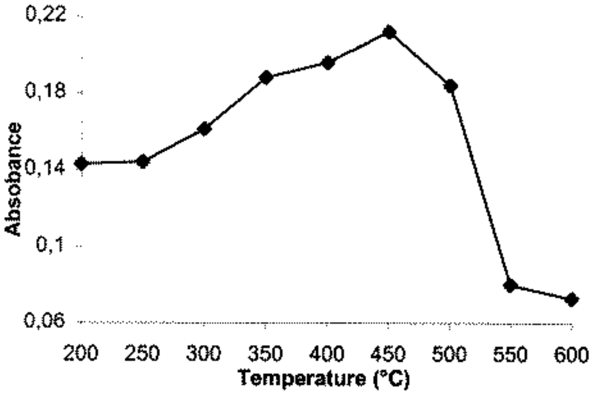

Figure 4: *Determination of decomposition temperature of lead at constent atomisation temperature (2000°C)*

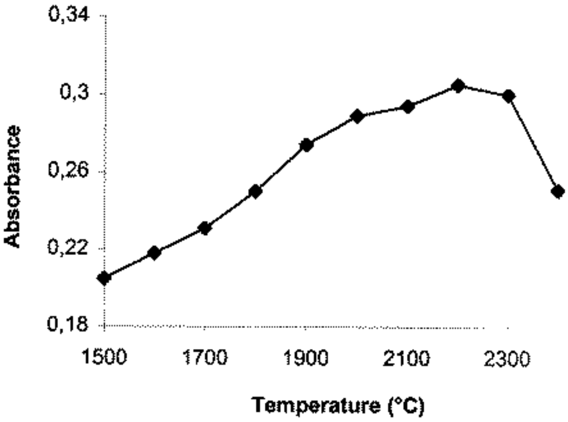

Figure 5: *Determination of atomization temperature of lead at constent decomposition temperature (450°C)*

Table II : *Optimization of decomposition and atomization temperatures*

			A 5 steps extraction					A 3 steps extraction		
Temperature	Element	Total metal	F15	F25	F35	F45	F55	F13	F23	F33
Ashing temperature (°C)	Pb	450	550	350	400	450	450	450	450	600
	Cd	400	250	400	400	400	400	200	450	450
Atomization temperature (°C)	Pb	2200	1650	1900	2100	2000	2200	1900	1900	2000
	Cd	2000	1900	1700	1700	1600 (**)	2000	1900	1400 (*)	1900

(*) Pallier from 1400 to 2000°C and (**) Pallier from 1600 to 1900°C

Table III : *Graphite furnace program*

Step	T(°C)	Ramp time (s)	Hold time (s)
Drying	120	12	15
Ashing	see table 2	10	12
Atomizing*	see table 2	3	5
cleaning	2400	1	3

* Stopped flow of inert gas

Calibration graphs

The matrix effect in different fractions in three and five step sequential extractions is shown in fig 6 and 7 respectively. Therefore calibration curve for each type of extraction is required.

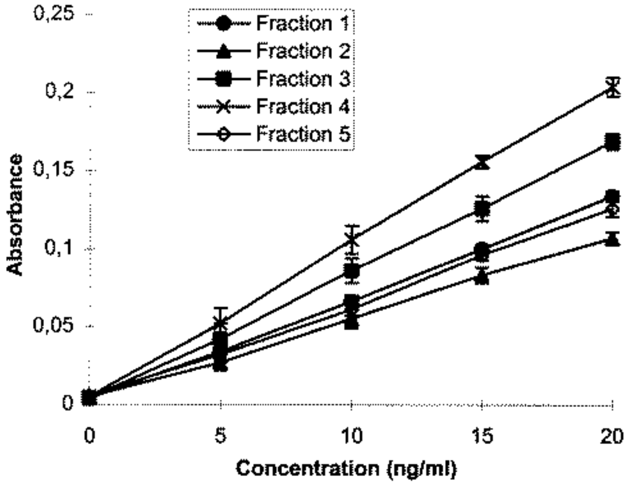

Figure 6 : *Comparison of lead calibration curves in each fractions of a 5 step sequential extraction*

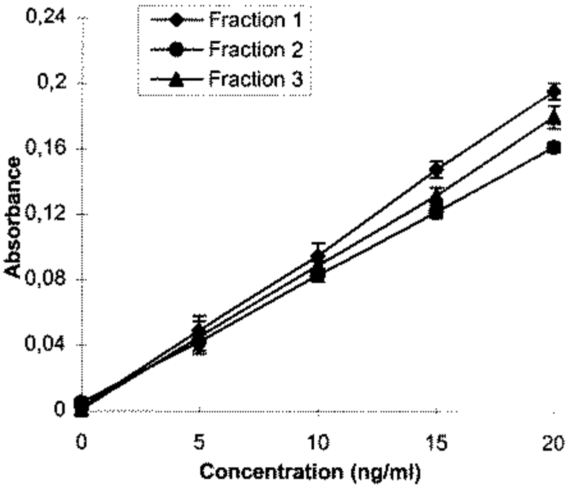

Figure 7 : *Comparison of lead calibration curves in each fractions of a 3 step sequential extraction*

The slope values obtained with calibration graphs for each extracts are different. They are shown in fig 6 and 7. There is a significant difference between calibrations for any of the extracts.

Application of the method

- *Application to sediments with certified total lead and cadmium content*

Two reference materials for total Pb and Cd content were analysed by the proposed procedure : SDM-2/TM and BCSS-1. These reference materials concentrations were given with 95 % confidence intervals. In this study, the lead and cadmium concentration in the extracts were calculated at 95 % confidence interval.

The 95 % confidence interval was calculated as follow :

$$T = s \times t\,(P,f) \quad [12]$$

T: dispersion domain; s: standard deviation; t: standard error of the difference; P:probability; f : degrees of freedom

The sequential extraction procedures were applied to the sediments using ETAAS. The extracts were also measured as shown in table IV and fig 8.

Table IV : *Lead and cadmium Concentration (µg/g) in 5 and 3 step sequential extractions of reference materials*

		a 5 step extraction						a 3 step extraction				
Ref standard	Element	F15	F25	F35	F45	F55	Σ	F13	F23	F33	Σ	T_{Ref}
SDM-2/TM	Pb	0.26 ± 0.02	5.0 ± 1.5	4.2 ± 0.2	3.9 ± 0.5	2.0 ± 0.2	15.4 ± 0.5	0.60 ± 0.05	6.40 ± 0.05	15.6 ± 1.0	22.6 ± 0.5	22.8 ± 5.5
	Cd	0.012 ± 0.002	0.032 ± 0.005	0.02 ± 0.002	0.013 ± 0.002	0.013 ± 0.002	0.09 ± 0.004	0.034 ± 0.005	0.06 ± 0.001	0.04 ± 0.005	0.134 ± 0.01	0.11 ± 0.04
BCSS-1	Pb	0.4 ± 0.05	3.2 ± 1	2 ± 0.2	6.2 ± 0.1	3.5 ± 0.2	15.3 ± 0.5	1 ± 0.05	5.5 ± 0.5	19 ± 2	25.5 ± 1	22.7 ± 3.4
	Cd	0.023 ± 0.005	0.036 ± 0.005	0.046 ± 0.01	0.063 ± 0.01	0.018 ± 0.005	0.186 ± 0.01	0.52 ± 0.006	0.06 ± 0.01	0.1 ± 0.02	0.21 ± 0.01	0.25 ± 0.04

Fig 8 shows that the three step sequential extraction presents the best recovery of lead and cadmium and required less products and time than the five step sequential extraction. Thus, the three step sequential extraction was used to analyse samples from ports.

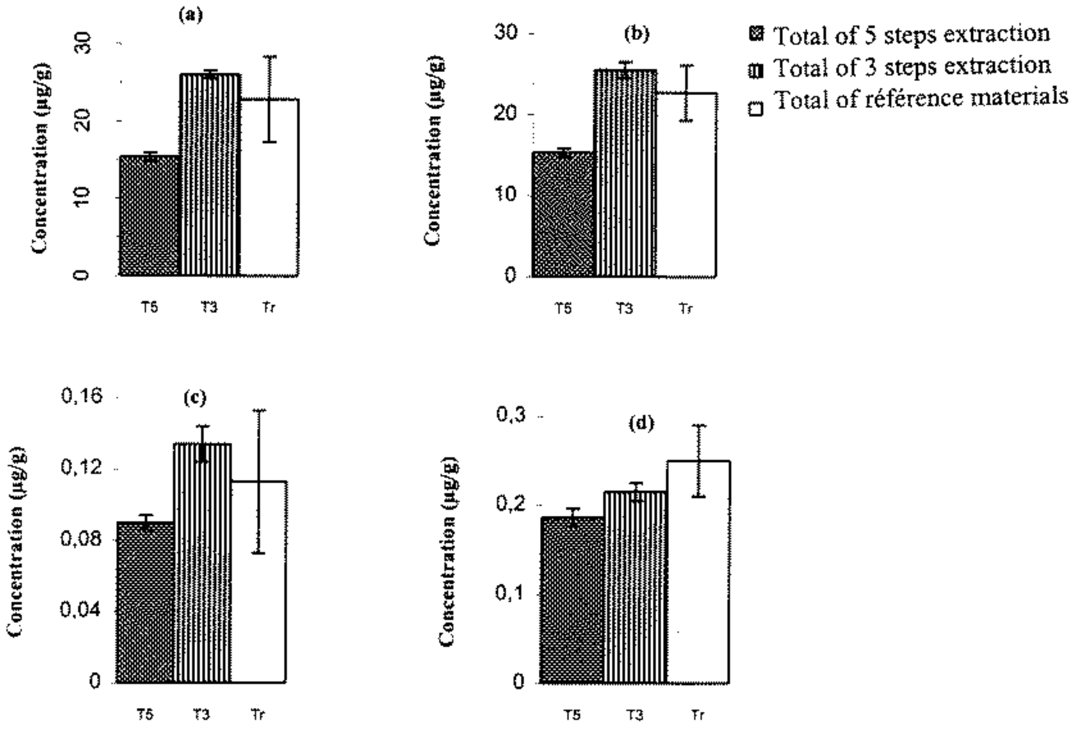

Figure 8 : *concentration of total lead and cadmium respectively in 5, 3 sequential extraction and total in reference materials : (a) Pb in SDM-2TM, (b) Pb in BCSS-1, (c)Cd in SDM-2TM, (d) Cd in BCSS-1*

- Application to the port samples from (Gulf of Tunis)

The sequential extraction procedures was also applied to four samples from several ports of the Gulf of Tunis where the determination of Pb and Cd content is important due to the presence of industrial and marine traffic in this zone. Table V shows the partitioning obtained for these samples and the values are means of three determinations.

Table V : *Application of 3step sequential extraction : determination of lead and cadmium concentration (µg/g) in sediment from ports of Rades and Goulette.*

		a 3 step sequential extraction				
Stations	Element	F13	F23	F33	∑	Total metal
Station 1	Pb	0.2±0.05	0.28±0.03	14.3±1.5	14.8±0.5	15.2±1
	Cd	0.18±0.004	0.45±0.01	1.8±0.09	2.5±0.05	2.6±0.2
Station 2	Pb	1.5±0.5	1.6±0.1	51.5±3	54.6±1.2	56±4
	Cd	0.4±0.02	0.27±0.02	1.95±0.1	2.7±0.05	2.7±0.1
Station 3	Pb	0.5±0.04	0.5±0.04	16±1.5	17±0.5	18±1
	Cd	0.2±0.006	0.8±0.09	1.6±0.1	2.6±0.07	2.7±0.1
Station 4	Pb	0.5±0.05	0.7±0.1	27±1	28.2±0.5	30±1.5
	Cd	0.5±0.008	0.3±0.03	2.1±0.1	2.9±0.1	3.28±0.5

The results of the sample from station 1 may be considered as a reference because this sediment was collected in a relatively unpolluted area of the zone studied. Goulette port station contains the higher concentration of lead

The results indicate that the main part of the extractable metal (sum of steps 1, 2 and 3) is associated with step 3, whereas Pb and Cd content in steps 1 and 2 is negligible.

CONCLUSION

ETAAS requires a thermal program for each fraction to analyse three and five steps sequential extraction. The use of the three step sequential extraction procedures for Pb and Cd gave a better recovery in this study.

Fraction 1 and 2 contain the lower concentration of cadmium and lead whereas fraction 3 has the bulk of these elements ratio more than 90% for lead and more than 70% for cadmium in all treated samples. These results indicates that cadmium and lead are weakly exchangeable in marine sediment samples. In these samples, metallic elements are relatively mobile indicating a relatively low toxicity in the medium.

As for total metals, all stations contain a total concentration of Cd and Pb lower than the maximum levels tolerated by International Standards except for station 2 and 4 which have a relatively high level of lead. These norms are respectively 19 mg/kg for lead and in the range of 0.2 - 5 mg/Kg for cadmium.[13]

These results indicate that sediment contaminations were relatively recent since residual fractions are comparatively low. According to these results, industrial wastes in these ports of the Gulf of Tunis are not important.

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