Evaluation of the metallic contamination impact on ’Mytilus Edulis’ mussel at the level of the mouth of Sebou’s estuary, Marocco

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EVALUATION OF THE METALLIC CONTAMINATION IMPACT ON '*MYTILUS EDULIS*' MUSSEL AT THE LEVEL OF THE MOUTH OF SEBOU'S ESTUARY, MAROCCO

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Abstract: The understanding of the bioaccumulation phenomenon of heavy metals in the living substance is of an extreme complexity. This is due to the interpenetration of several parameters which influence this bioaccumulation such as: the physico-chemical characteristics of the study area, the chemical properties of the contaminant and the biologic factors of the organism. This work constitutes, using *Mytilus edulis* as bioindicator, a contribution to study the metal contamination at different points of the mouth of Sebou's estuary. The samples were collected during the spring and the summer of the year 2005. The obtained results reveal a contamination of the study area by all the measured metals especially by lead, cadmium and chromium. This study allowed also discerning the existence of a variation in the accumulation of heavy metals according to *Mytilus edulis* size, as it was reported in the literature. Indeed, it showed that the small mussels accumulate heavy metals more than the big, mainly for manganese, chromium and cadmium. Seven trace metals (Mn, Cu, Cr, Pb, Co, Fe and Cd) were detected in the four studied organs (hepatopancreas, gills, gonads and muscle); the variations was least or more significant according to the organ. So, we conclude that the bioaccumulation does not make in a strictly selective way.

Keywords: Mytilus edulis; Trace metals; Bioaccumulation; Bioindicator.

1. INTRODUCTION

Oued Sebou draining basin is situated in the North West of Morocco and collects waters from the southern slopes of the Rif range and from the north western slopes of the middle Atlas. It lies between the 33rd and 35th north parallels, and being in the north western part of Africa has Mediterranean type weather. Oued Sebou is a main water body in Morocco. It waters serves important towns (Sidi Slimane, Sidi Kacem, Bel Ksiri, Kenitra, etc...) and receives, without any treatment, waste waters from many industrial plants (sugar mills, paper mills, petrol refinery, canneries, etc.). The excessive and uncontrolled use of pesticides also contributes to the degradation of Sebou water quality.

Thus, the study of pollution of the estuary and its effects on the ecosystem has become the major concern of several researches on ecotoxicology with an increased attention accorded to the analysis of biological materials (Bouachrine, 1996; Bouachrine et al., 1998; Benkirane et al., 2001; El Bouhali, 2001; Benkirane, 2002). Indeed, considering the difficulty of the collection of valid samples for trace metal analysis in water as well as the low spatial and temporal representativeness of these, the use of the quantitative bioindicators of the chemical contamination attracts more and more attention in ecotoxicology.

It is in this context that we have elaborated this work to estimate the degree of the metal contamination at the level of Sebou's mouth and this by dosage of seven trace metals (Cu, Mn, Fe, Co, Cr, Pb and Cd) in mussels (*Mytilus edulis*). In fact, Mytilus edulis have been found to be suitable indicator species for trace metals (Goldberg, 1975; Phillips, 1976; Phillips, 1977; Brown & Luoma, 1995; Jumshamn & Grahl-Nielsen, 1996). It is capable to accumulate trace metals such as cadmium, mercury and lead to a larger extent than for example fish and algae (Jumshamn & Grahl-Nielsen, 1996; Riget et al., 1996; Airas et al., 2004). It has wide geographical distribution and tolerance range for different salinities and temperatures. In addition, it has sufficient size, sessile life form and is robust in laboratory conditions.

2. METHODS AND MATERIALS

2.1 Sampling Sites

The sampling of mussels was elaborated at four stations belonging to the mouth of the Sebou estuary (Fig. 1):

- S1: Monegasque; near the discharges of the "Monegasque" factory for the canning of fishes;
- S2: Camping; very important domestic discharges especially in summer;
- S3: the extremity of the mouth; important fishing activity;
- S4: Mehdia beach.

These four stations were chosen for their socio-economic interest (main zones for harvesting mussels by fishermen). The mussel samples were collected at low tide. They were placed in plastic flasks filled with water of each station, then immediately transported to the laboratory.

2.2 Sample Preparation

The shells were carefully opened so as not to damage tissues. Four organs were taken from each mussel: gills, gonads, muscle and hepatopancreas for organotropisme study, a sample of measure corresponds to ten units of the same organ.

The samples were conserved in sterilized petri dishes, weighed and stored at a temperature of -18°.

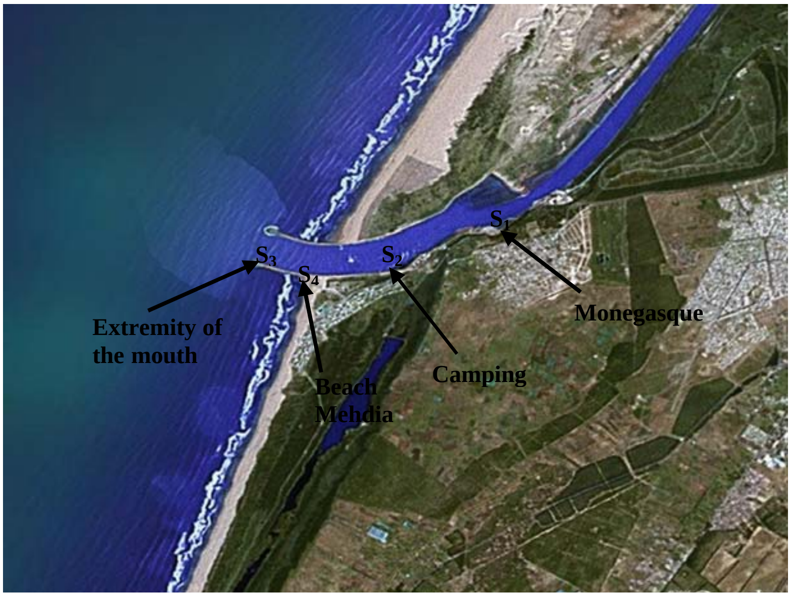

Figure 1: Approximate locations of sampling sites at the level of Sebou’s mouth (Image Source: Google Earth).

The procedure adopted for the preparation of mussels was as bellow:

- Lyophilization during 48 hours under vacuum and a temperature of -50°C, and under a pressure of about 4000 bar;
- Grinding of each sample in a porcelain mortar until the obtaining of a fine powder;
- Recovery of 0.5 g of broyats in Teflon bombs;
- Addition of 3 ml of concentrated HNO3 (69°C) in each bomb to solubilise the metals and 1 ml of H2O2 to oxidize the organic matter;
- Digestion in a microwave oven at high pressure and high temperature.

Analysis of final solutions for all trace elements detected by Atomic Absorption Spectroscopy in graphite furnace. For each series of analysis, a negative control reagent was prepared in the same way as the samples, except that no sample has been added in the flask of digestion. The results are expressed in µg/g of dry weight (ppm).

3. RESULTS AND DISCUSSION

3.1 Preferential Distribution of Heavy Metals in the Studied Tissues

In table 1, we present the concentrations of the heavy metals (Fe, Mn, Cu, Co, Cr, Cd and Pb) analyzed in the four studied organs.

Table 1: Average heavy metal concentrations (in μg/g) in the four studied organs.

	Gills	Hepato-pancreas	Muscle	Gonads
Fe	15.19	15.69	12.81	10.34
Mn	73.17	31.41	47.26	63.31
Cu	51.36	63.88	52.23	49.81
Co	11.44	33.36	11.66	11.05
Cr	88.56	77.80	51.85	24.42
Cd	7.44	12.86	11.59	13.13
Pb	16.20	20.28	21.52	31.43

According to table 1, all the measured metals are detected in the four studied organs with significant but variable concentrations according to the accumulator organ. So, from this study it appears that the gonads concentrate more Mn, Cu, Pb and Cr; the gills accumulate preferentially Cr, Mn and Cu; the hepatopancreas is the preferred location for almost all the studied metals; and finally the muscle concentrates more Cu, Mn, Cr and Pb.

Access to the internal compartments of the body and the processes of accumulation in tissues are extremely complex and depend on several factors including the properties of the receiving tissue (vascularization, membrane properties, chemical composition, active processes, etc.) (Maury & Engrand, 1986); the nature of the contaminant (molecule size, chemical speciation, etc.) (Bowen, 1966) and the intra and extracellular environment (temperature, pH, etc.).

Contrary to fishes, the organs of the mussel are all exposed to chemical pollutants suspended in the water; direct contamination could have the same significance as the trophic contamination which could explain the wide repartition of metals for all the studied organs.

According to the bibliography, bivalves accumulate Cd mainly in hepatopancreas and in kidney in the form of deposits in lysosomes (Serafin et al., 2002), and accumulate Cu mainly in hepatopancreas, gonads and gills (Adami et al., 2002) what does not are in contradiction with our results. Indeed, in molluscs, the blood contains a respiratory pigment based on copper, the hemocyanine, which explains its distribution in these richly vascularized organs.

Figure 2 shows the variations of heavy metals contents in the various studied organs. Cr shows an accumulation significantly low in gonads compared to other organs. Indeed these greasy organs have more affinity to divalent elements (Simkiss, 1983), which is not the case for Cr.

Fe shows the least concentrations in all the studied organs, which is not the case for fishes where Fe, constituent of hemoglobin, is very concentrated. Indeed, in bivalves, Fe is involved in the tanning processes of the shell, foot and byssogene gland (George et al., 1976; Kesavan et al., 2010).

The hepatopancreas plays a vital role in achieving the metabolism phenomena, and then by crossing the branchial or intestinal walls, metals reach the bloodstream to join this organ, which can explain the importance of the accumulation of the most metals measured in this organ of target metabolism.

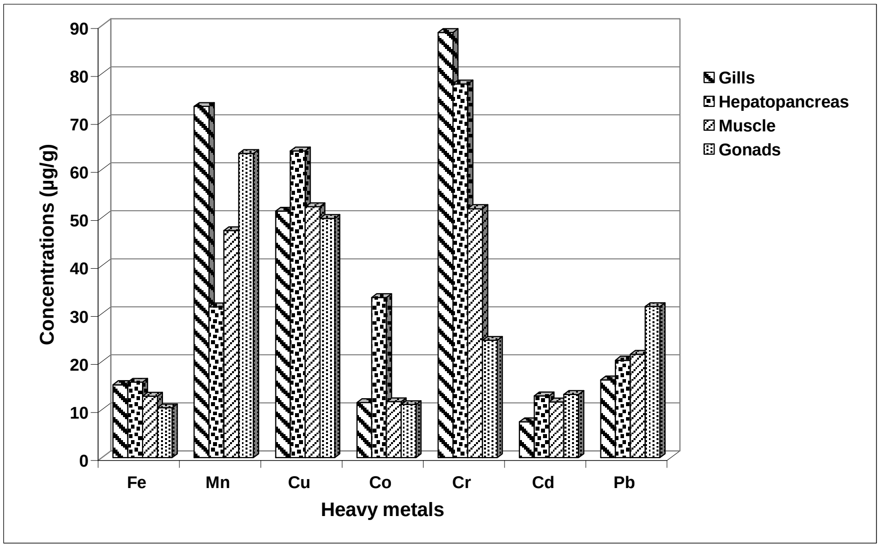

Figure 2: Contents of heavy metals in the various studied organs.

3.2 Variation of Average Contents of Heavy Metals According to the Size

Figure 3 shows the concentrations of heavy metals measured and detected both in big and small mussels.

The global observation of results concerning the influence of size factor on the accumulation of the studied heavy metals (Fe, Cu, Mn, Co, Cr, Pb and Cd) in *Mytilus edulis* shows a tendency of concentration in mussels with small size; these results are in concordance with those of the bibliography.

Indeed, many studies have highlighted the relationship metal/size in the mussel. Cossa and al. (1980) showed net relationship between the concentrations of different metal contaminants (Cd, Cu, Fe, Mn, Ni, Zn) in the mussel *Mytilus edulis* and its weight; the small mussels present high concentrations than those measured in large mussels.

Morono and al. (2001) then Morono and al. (2003) showed that the quantities of metals concentrated in mussels result from their net accumulation (incorporation - excretion) which itself is a function of the size and weight.

According to Casas (2005), the metal concentration arises from the quotient of two components: the total metal content in the organism and its weight. Therefore, the changes in the weight of the bioindicator tissues can significantly affect the trace metal concentrations by diluting or simply by concentrating the total mass of the metal. So, the increase of the weight would cause a dilution of trace metals in the organism, which could explain concentrations the high concentrations in small mussels.

3.3 Variation between Marine and Estuarine Environment

This study showed that all the measured metals (except the cadmium) revealed in the maritime environment average contents greater or equal to those founded in the estuary environment. This can be explained by the fact that the sea is the final reservoir of all the river draining so the pollutant rates will increase; or may be the human activities (swimming, surfing, zodiac) that start at the Mehdia beach (Kenitra, Morocco) in spring and raise in summer, are behind this noticed increase.

Another hypothesis can be considered to explain this result is the increase of salinity: according to Amiard (1989) and Kesavan et al. (2010), natural phenomena of solubilization occurring in the presence of salt water, imply that metals become more available for alive organisms, the forms ionized with chlorides and hydroxides must mainly be considered.

The cadmium bizarrely appears to escape this hypothesis and to be more available in estuary environment than in marine environment. Indeed, studies led by Borchardt (1983 and 1985) on the bioaccumulation of cadmium in mussels showed the preponderance of the dissolved way with regard to the particulate way knowing that in the seawater the chloride ions complex strongly the cadmium(Baric & Branica, 1967). However, on the other hand, the study led by Bouquegneau and al. (1983) showed that the Murex trunculus mollusc presents a very high contents of cadmium in a bay compared to those collected at sea.

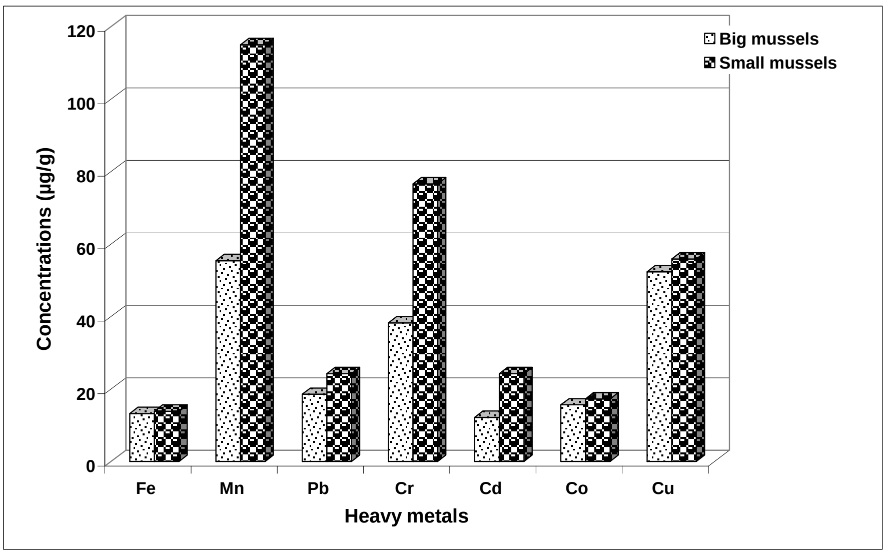

Figure 3: Heavy metal contents according to Mytilus edulis size.

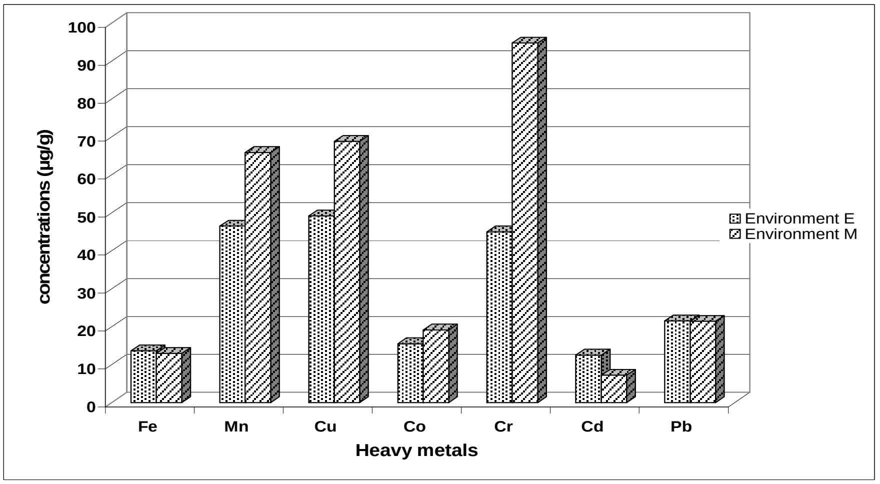

Figure 4: Variations of heavy metal contents between marine and estuarine environment.

4. CONCLUSION

Understanding the phenomenon of bioaccumulation of heavy metals in living substance is of extremely complex. This is due to the interpenetration of several parameters which influence the bioaccumulation namely in general: the physico-chemical characteristics of the environment, the chemical properties of the contaminant and the biologic factors of the organism.

In this work, we have tried to contribute to the study of metal contamination in a sensitive point of Sebou's estuary which is the mouth using *Mytilus edulis* as bioindicator. Obtained results reveal a contamination of the studied area by all the measured metals mainly by the not essential metals: lead, cadmium and chromium.

The comparison of trace metal contents in the estuary environment (Stations 1, 2 et3) and the marine environment (Station 4) revealed a wide availability of Mn, Cu, Co and Cr in this marine environment which may be related to the degree of salinity in the sea.

Also, this study has confirmed the existence of a variation of accumulation of heavy metals according to the size of Mytilus edulis. It showed that the small mussels accumulate more heavy metals than the big, mainly for manganese, chromium and cadmium.

The seven studied trace metals (Mn, Cu, Cr, Pb, Co, Fe and Cd) were detected in the four studied organs (hepatopancreas, gills, gonads and muscle) with variations more or less marked according to the organ: gonads concentrate more Pb, Cd, Cu and Mn; gills accumulate preferentially Cr, Mn, Cu and Fe; hepatopancreas is the preferred site for most metals namely Co, Cu, Fe, Cr and Cd; finally muscle concentrates more Cu and Cd. So, we can deduce that the bioaccumulation is not made in a strictly selective way.

In conclusion, the results of this study show the persistent degradation of the Sebou estuary as a result of industrial, agricultural and domestic discharges, which would increase if no strategy of protection of this environment by concerned authorities, is developed and implemented. So, to reduce the contamination of Sebou estuary, the implementation and the application of regulations on management of releases are needed, as well as the creation of pilot projects of depollution.

5. ACKNOWLEDGMENTS

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