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Bioaccumulation of some metallic elements in the gills, the muscle and the liver of the eel (*Anguilla anguilla* L.) and the cabot mullet (*Mugil cephalus* L.) of the estuary of Oued Sebou (Morocco)

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Abstract
Chemical pollution of estuaries often affects the different compartments of this ecosystem. Pollutants are in water, sediment and biocenosis. Anthropogenic activity, responsible for this pollution, continues to grow. Thus, the assessment of the degree of this pollution has become a necessity.
In the present work, we have evaluated the content of four metallic elements (Fe, Zn, Cu, Pb) in the sediment of one of the most polluted estuaries of Morocco: the estuary of Sebou. At the same time we estimated the contents of these metallic elements in the muscle of two fish harvested in the same estuary: *Anguilla anguilla* L. and *Mugil cephalus* L.
The results showed that, for the four metal elements studied, the concentrations noted in the sediment vary from one season to another. Similarly, organ tissues of the species studied show concentrations of all metals evaluated. These contents differ according to the metals and according to seasons. These are the gills that bioaccumulate more metal followed by the liver and most often it is the muscle that bioaccumulates metal month. In addition, the degree of bioconcentration of iron and zinc in the tissues studied does not differ significantly among species. However, the concentration of copper seems to be higher in *Anguilla anguilla* and that of lead seems to be higher in *Mugil cephalus*.
The classification of the concentrations of the metals studied ranked in descending order of the average concentrations is the same as that noted in the sediment: Fe> Zn> Pb> Cu.

Keywords: fish, metallic pollution, sediment, estuary, sebou, Morocco

1. Introduction

Aquatic ecosystems are exposed to pollution from human activity that continues to grow. Among the pollutants likely to accumulate in water, sediment and biocenosis of continental and marine waters heavy metals. By passing directly into the body of beings exposed to the pollutant or indirectly through the trophic chain [1, 2], these metals are potentially capable of being disrupters of animal life. This double contamination by heavy metals could be the case of fish [3]. Therefore, consuming fish contaminated with heavy metals, or bacteria, can have serious consequences on the health of the consumer [4]. Thus, monitoring the living environment of these fish, including aquatic ecosystems, is necessity.

In addition, two environmental sites in the urban area of the city of Kenitra (Morocco), the estuary of Sebou and Lake Fouarate, are known by the importance and diversity of their pollution [5, 6]. In fact, for *Gumbusia houlbrouki* (Poisson), samples taken in the Sebou estuary and Fouarate Lake and analyzed showed a bioaccumulation of heavy metals in the flesh of this fish [7]. Similarly, Lepomis gibbosus L. "(Fish) sampled in Fouarate Lake showed bioaccumulation of certain heavy metals in various tissues of its organs [8]. For *Anguilla aguilla* in Morocco, the population of the calendar years is clearly declining since the 1980s. The modifications are multiple and complex, notably the degradation of their habitats, the overfishing, the pathogens, the climate change (obstacles to their anadromous migrations and catadromous, river developments) [8]. Levels of metal contamination of Loukkos eels are dangerous for the human consumption of the muscle chair of this fish [8].

In this study, we will focus on the assessment of the degree of contamination by heavy metals in the waters of the Oued Sebou estuary and the gills, muscle and liver of two fishes: *Anguilla anguilla* L. and *Mugil cephalus* L.

Material and Methods

1. Studied fish

The European eel is a teleost fish that belongs to the order Anguilliformes, family Anguillidae represented by a single genus, the genus Anguilla [9]. The species Anguilla anguilla or "European eel" has a very wide geographical distribution, moreover, it is able to adapt to various freshwater and brackish environments. The originality of the eel is based on its particular life cycle with a larval marine phase (Leptocephalus and Civelle transparent), a sea-continent transition period (Civelle) and a long continental stay (yellow and silver eel).

According to El Hilali (2007) [10], the minimum length is 10.35 cm observed in February while the longest eel (35.75 cm) was captured in January. Monthly average sizes show seasonal fluctuations with significantly lower values in the summer period. The average size of the entire population is 17.48 ± 4.59 cm. On the other hand, linear growth modeling
Cosmopolitan in the coastal waters of the tropical, subtropical

and temperate zones of all the seas, *Mugil cephalus* is a coastal species that can go up estuaries and rivers. It lives on sandy or muddy bottoms, often less than 10 m deep and feeds on zooplankton algae, benthic organisms and small fish [11].

2. Middle and Study stations

The estuary of Sebou is subjected, at the level of the city of Kenitra, to a multiple pollution provoked by industrial activities (pumping of water and rejection of toxic effluents), agricultural (pumping of water and infiltration of the nitrogenous derivatives of fertilizer) and domestic (wastewater).

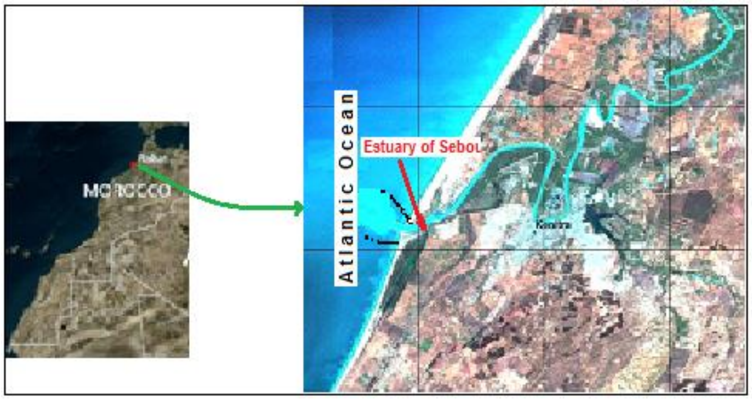

Fig 1: Sebou estuary (LANDSAT 7ETM photograph P202R036 of April 10, 2001 - excerpt (Source: Sogreah, 2011)).

3. Metallic elements studied and method of study

To evaluate the degree of metallic pollution of the Sebou estuary, we measured the concentration of four metallic elements in the water and sediment of the estuary namely Pb, Cu, Fe and Zn The analysis is seasonal and was done for one year. The metal elements (Pb, Cu, Fe and Zn) are measured by ICP (Inductively Coupled Plasma). The same analysis was performed in four gill, muscle and liver samples from both fish.

Result and Discussion

The variations of the contents of the metallic elements in the sediment, gills, muscles and liver of *Anguilla anguilla* and *Mugil cephalus* are grouped in Tables I and II:

Table 1: Seasonal contents (in mg/kg) of heavy metals detected in sediments

Season	Fe	Zn	Cu	Pb
Fall	26531	109,22	20,9	10,05
Winter	18622	69,67	19,84	8,21
Spring	17934	114,05	18,93	11,03
Summer	22327	112,61	17,02	12,41
Average	21353	101,38	19,17	10,42

Table 2: Seasonal contents (in mg / kg fresh weight) of heavy metals detected in different tissues of *Anguilla anguilla* and *Mugil cephalus*.

Season	Organ	Fe	Zn	Cu	Pb	Classification of concentrations
Anguilla anguilla						
Full	Liver	1109	198.2	13.99	1.26	Fe> Zn>Cu >Pb
	Muscles	1017	127.6	14.09	0.28	Fe> Zn>Cu >Pb
	Gills	4752	102.51	27.82	2.29	Fe> Zn>Cu >Pb
Winter	Liver	1779	119.22	1.77	1.09	Fe> Zn>Cu >Pb
	Muscles	3014	54	9.19	0.79	Fe> Zn>Cu >Pb
	Gills	2246	86.69	16.81	1.99	Fe> Zn>Cu >Pb
Spring	Liver	2682	381	13.5	1.18	Fe> Zn>Cu >Pb
	Muscles	1143	175.5	8.45	1.35	Fe> Zn>Cu >Pb
	Gills	1662	162	11.32	1.02	Fe> Zn>Cu >Pb
Summer	Liver	2281	203.9	25.62	1.93	Fe> Zn>Cu >Pb
	Muscles	1139	169	13.23	1.17	Fe> Zn>Cu >Pb
	Gills	46853	162.36	17.32	2.12	Fe> Zn>Cu >Pb
Average in the liver		1962.7	250.58	13.92	1.34	Fe> Zn>Cu >Pb
Average in the muscle		1578	131.52	11.21	0.89	Fe> Zn>Cu >Pb
Mean in the in the gills		13878	128.39	18.33	1.85	Fe> Zn>Cu >Pb
Classification of organs according to their degree of bioconcentration of metal		Gills > liver > Muscle	liver > Muscle±Gills	Gills >> liver±Muscle	Gills > liver > Muscle	———
Mugil cephalus						
Full	Liver	1321	250.14	14.16	7.56	Fe >Zn>Cu>Pb
	Muscles	1231	209	1.65	2.03	Fe >Zn>Pb>Cu

	Gills	981	89.3	4.1	15.2	Fe >Zn>Pb>Cu
Winter	Liver	1168	134	13.21	9.16	Fe >Zn>Cu> Pb
	Muscles	1202	90.9	1.6	6.01	Fe >Zn>Pb>Cu
	Gills	1321	167.1	3.8	17.21	Fe >Zn>Pb>Cu
Spring	Liver	1514.8	154.9	2.91	5.95	Fe >Zn>Pb>Cu
	Muscles	1080	65	7.1	1.04	Fe >Zn> Cu>Pb
	Gills	1402.5	294	12.12	9.53	Fe >Zn> Cu> Pb
Summer	Liver	2147	178.32	5.4	10.93	Fe >Zn>Pb>Cu
	Muscles	910.04	132	2.72	1.02	Fe >Zn> Cu>Pb
	Gills	2294	431	9.91	9.09	Fe >Zn> Cu>Pb
Average in the liver		1787.7	169.34	8.92	8.52	Fe >Zn> Cu>Pb
Average in the muscle		1105.76	124.2	3.23	2.52	Fe >Zn> Cu>Pb
Mean in the in the gills		1499.61	245.35	7.48	12.50	Fe >Zn> Pb>Cu
Classification of organs according to their degree of bioconcentration of metal		liver > Gills> Muscle	Gills > liver > Muscle	liver > Gills > Muscle	Gills > liver >> Muscle	———

1. Concentration of metals in the sediment

For the iron, the concentrations recorded in autumn (26531 mg/kg) and in summer (22327 mg/kg) exceed those recorded in winters (18622 mg/kg) or in spring (17934 mg/kg). The annual average is 21353 mg/kg. For the zinc, with the exception of the winter where the concentration was 69.67 mg/kg, other season values are around 110 mg/kg and the mean value is 101.38 mg/kg. For the copper, the seasonal values noted are not very different and are around 19 mg/kg and the average value is 19.17 mg/kg. For the lead, the winter value (8.21 mg/kg) is the lowest, the seasonal values are not very variable, the values of other seasons are close to 11 mg/kg and the average value is 10.42 mg/kg.

In addition, the ranking of the concentrations of the metals studied ranked in descending order of the average concentrations is Fe> Zn> Cu> Pb.

2. Concentration of metals in the tissues of *Anguilla anguilla*

- The gills

For the iron, the concentration observed in summer (46853 mg/kg) is highest, that noted in spring (1662 mg/kg) is the lowest and those of winter (2246 mg/kg) and the fall (4752 mg/kg) are intermediate. The seasonal average is 13898 mg/kg. For the zinc, the seasonal mean concentration is 128.39 mg/kg but the concentration varies with the seasons. The highest value (162.36 mg/kg) is summer; the lowest (86.69 mg/kg) is winter. The concentrations recorded in autumn (102.51 mg/kg) and in the spring (162 mg/kg) are intermediate. For the copper, the highest value (27.82 mg/kg) is autumnal and the lowest value (11.32 mg/kg) is spring. Summer (17.32 mg/kg) and winter (16.81 mg/kg) are intermediate and very similar. The seasonal average is 18.33 mg/kg. For the lead, the concentrations are low. Autumnal (2.29 mg/kg) and summer (2.12 mg/kg) are the highest, the spring (1.02 mg/kg) is the lowest and the winter is 1.99 mg/kg). The seasonal average is 1.85 mg/kg.

In addition, the classification of the concentrations of the metals studied ranked in descending order of the average concentrations is the same as that noted in the sediment: Fe> Zn> Cu> Pb.

- The muscle

For the iron, relative to other concentrations in other seasons, the concentration noted in winter was the highest. The concentrations noted in spring (1143 mg/kg) and in autumn (1017 mg/kg) and in summer (1139 mg/kg) are numerically similar. The all-season average is 1578 mg/kg. For the zinc, the seasonal average of the concentrations is 131.52 mg/kg but the concentration varies with the seasons. The highest value is found in the spring (175.5 mg/kg) and the lowest value is winter (54 mg/kg). The concentrations that were noted in summer (169 mg/kg) and autumn (127.6 mg/kg) are intermediate. For the copper, the highest value (14.09 mg/kg) is autumnal, the summer value (13.23 mg/kg) is also high; those of winter and spring are 9.19 mg/kg and 8.45 mg/kg, respectively. The seasonal average is 11.21 mg/kg. For the lead, the spring concentration (1.35 mg/kg) is highest and the autumn concentration (0.28 mg/kg) is the lowest. Winter (0.79 mg/kg) and summer (1.17 mg/kg) are intermediate and the seasonal average is 0.89 mg/kg.

In addition, the classification of the concentrations of the metals studied ranked in descending order of the average concentrations is the same as that noted in the sediment: Fe> Zn> Cu> Pb..

- Liver

For the iron, the concentrations noted in spring (2682 mg/kg) and in summer (2281 mg/kg) are higher than those noted in winter (1779 mg / kg) and in autumn (1109 mg/kg). The all-season average is 1962.7 mg/kg. For the zinc, the seasonal mean concentration is 250.58 mg/kg but the concentration varies with the seasons. The spring value (381 mg/kg) is the highest, followed by the summer value (203.9 mg/kg). Winter and spring are 119.22 mg/kg and 198.2 mg/kg, respectively. For the copper, the highest value (25.62 mg/kg) is summer; those winter and spring are respectively 1.77 mg/kg and 13.5 mg/kg, and fall is 13.99 mg/kg. The seasonal average is 13.92 mg/kg. For the lead, the seasonal concentrations are very similar: the winter concentration was 1.09 mg/kg, the spring one 1.18 mg/kg, the summer one 1.93 mg/kg and the autumn one 1.26 mg/kg. The seasonal average is 1.34 mg/kg.

In addition, the classification of the concentrations of the metals studied ranked in descending order of the average concentrations is the same as that noted in the sediment: Fe> Zn> Cu> Pb.

3. Concentration of metals in the tissues of *Mugil cephalus*

- The gills

For the iron, the concentration noted in summer (2294 mg/kg) is the highest and that recorded in autumn (981 mg/kg) is the lowest. The concentrations noted in spring (1402.5 mg/kg) in winter (1321 mg/kg) are intermediate and the seasonal average was 1499.61 mg/kg. For zinc, the seasonal mean concentration is 245.35 mg/kg but the concentration varies with the seasons. The highest value (431 mg/kg) was summer, the spring value (294 mg/kg) was also high, the winter value was 167.1 mg/kg and the autumn value was 89.3 mg/kg. weaker. For the copper, the highest value (12.12 mg/kg) was spring; the lowest (3.8 mg/kg) was winter. Summer (9.91 mg/kg) is also high, and fall was 4.1 mg/kg. The seasonal average is 7.48 mg/kg. For the lead, the highest concentration (17.21 mg/kg) was winter followed by autumn (15.2 mg/kg). Spring (9.53 mg/kg) and summer (9.09 mg/kg) were intermediate and the mean seasonal concentration was 8.52 mg/kg.

In addition, the classification of the concentrations of the metals studied ranked in descending order of the average concentrations is the same as that noted in the sediment: Fe> Zn> Cu> Pb.

- The muscle

For iron, the concentrations that have been noted do not vary much with the seasons. In fact, the spring (1080 mg/kg), autumn (1231 mg/kg) winter (1202 mg/kg) and summer (910.04 mg/kg) values are numerically very high. The all-season average is 1105.76 mg/kg. For zinc, the seasonal mean concentration is 124.2 mg/kg but the concentration varies with the seasons. The highest value (209 mg/kg) is autumnal, the summer value (132 mg/kg) is also high; those winter and spring are 90.9 mg / kg and 65 mg/kg respectively. For copper, the highest value (7.1 mg/kg) was spring and the lowest (1.65 mg/kg) and (1.65 mg/kg) were respectively autumn and winter. The summer population was 2.72 mg/kg and the seasonal average is 3.23 mg/kg. For lead, it is in winter that the highest concentration (6.01 mg/kg) was noted. The lowest concentrations were numerically very similar and the spring concentration was 1.04 mg/kg and the summer concentration was 1.02 mg/kg. The fall has been 2.03 mg/kg and the seasonal average is 2.52 mg/kg. In addition, the classification of the concentrations of the metals studied ranked in descending order of the average concentrations is the same as that noted in the sediment: Fe> Zn> Cu> Pb.

- Liver

For the iron, the summer concentration (2147 mg/kg) was the highest. This was successively followed by that of spring (1514.8 mg/kg), autumn (1321 mg/kg) and winter (1168 mg/kg). The all-season average is 1787 mg/kg. For the zinc, the fall concentration (250.14 mg/kg) was highest, followed by the summer concentration (178.32 mg/kg), the spring concentration 154.9 mg/kg and the winter concentration 134 mg/kg. The seasonal average of the concentrations is 169.34 mg/kg. For the copper, the highest concentrations were recorded in autumn (14.16 mg/kg) and in winter (13.21 mg/kg); the lowest (2.92 mg/kg) was noted in the spring. The summer concentration (5.4 mg/kg) is intermediate. For the lead, the concentrations recorded in summer (10.93 mg/kg) or in winter (9.10 mg/l) were the highest. Those spring (5.95 mg/kg) or autumn (7.56 mg/kg) are lower. The seasonal average is (8.52 mg/kg).

It should be noted that, for the origin of this pollution of certain living beings of the estuary of Sebou, this estuary is connected to one of the most polluted rivers of Morocco. The origin of pollution is very diverse [12, 13]. In fact, in addition to pollutants that are drained from the watershed of the Sebou River, wastewater rich in toxic substances of urban or industrial origin is dumped, without prior treatment, into the river or directly into the estuary. Among these pollutants are heavy metals. In the estuary, these metallic elements are found in water and in sediment and can even through the food chain be deposited in the body of certain living beings of the medium [14]. But, as our results and the results of other previous works show, it is in the sediment that the quantity of the heavy metals is more important.

In addition, the results (Tabs I and II) showed that in sediment and in water, the available quantity of heavy metals differs. This difference in concentration could be the result of the difference in the degree of water solubility of these metals, the action of certain physicochemical conditions which favor the adsorption of metals on the solid particles of the medium, the action of the phenomenon release of existing metallic elements from water and sediment [15-17]. As reported by Liu *et al*. (2010) [18], through feeding, breathing or dermal metal elements can pollute many animal species by accumulating at different levels of concentration in the tissues of their bodies. According to Khamar *et al*., (2002) [19], the amount of bioaccumulation depends on many factors including the metal element concerned, the degree of availability of this metal and the action of certain physicochemical parameters of the medium. The stage of development of bioaccumulator fish could also be a factor. Wariaghli (2013) [20] reported that in the Loukkos estuary (Morocco), large eels (50 to 60 cm), that is, the oldest, are more contaminated with toxic values, of the order 0.58 μg/g fray weight (f.w.) Exceeding the standard set by WHO (0.5 μg/g spawning pea (m.p.) for human consumption). The same author pointed out that the average concentration of mercury in the muscle of sebou and Loukkos eels is high, of the order of 0.47 μg/g (f.w.) (Loukkos) and 0.46 μg/g f.w. (Sebou).

A noter que d'après Wariaghli (2013) [20], chez *Anguilla anguilla*, l'analyse des éléments métalliques révèle une contamination tissulaire généralisée avec des concentrations plus élevées de certains éléments métalliques par rapport aux autres. Le facteur « saison » est un facteur de variation de concentration du métal dans le corps de l'être biaccumulateur: la variation saisonnière de la concentration des métaux dans les différents organes étudiés pourrait être due aux changements des conditions physico-chimiques du milieu et au degré du stade de développement du poisson analysé. Pour l'influence des conditions physico-chimiques sur le degré d'absorption d'un métal par l'organisme de certaines espèces du milieu, Kourradi *et al.*, (2001) [21] ont signalé que chez *Solen marginatus* (Mollusque), le changement saisonnier des conditions physico-chimiques du milieu influence la quantité de certains éléments métalliques bioconcentrés dans la chair. De même, pour plusieurs mollusques, les teneurs accumulées du zinc, du plomb et du cuivre sont inversement proportionnelles à celle du degré de la salinité du biotope [22]. Ces mêmes constatations ont été mentionnées chez d'autres Mollusques bivalves tels chez Pecten et Chlamys [23]. Chez *Venerupis decussata*, dans Piedras River estuary (southwest Spain), une variation saisonnière sinusoïdale significative de Cu et de Zn a été observée par Ruiz-Azcona *et al*. (2013) [24]. Ainsi, ces derniers résultats pourraient expliquer les variations saisonnières des teneurs métalliques que nous avons noté dans les tissu de *Anguilla anguilla* et *Mugil cephamus*.

Par ailleurs, la contamination métallique étudiée pourrait être une cause de stresse des espèces étudiés. En effet, les résultats d'un travail effectué par Guimarães *et al*. (2009) [25] ont noté que le développement des anguilles dans les estuaires pollués des rivières Lima et Douro (Portugal) interfère avec des fonctions physiologiques déterminantes pour leur survie et leur performance. La contamination d'espèces fortement contaminées par des métaux lourds pourrait nuire aussi à la santé du consommateur.

Conclusion

For both species, the tissues of the organs studied showed concentrations of the various studied metals. The degree of this concentration differs according to the metals and according to the seasons. In addition, the degree of bioconcentration of iron and zinc in the tissues studied does not differ significantly among species. However, the concentration of copper seems to be higher in *Anguilla anguilla* and that of lead seems to be higher in *Mugil cephalus*. Similarly, it is the gills that bioaccumulate more metal followed by the liver. Most often it is the muscle that bioaccumulates metal month.

The classification of the concentrations of the metals studied ranked in descending order of the average concentrations is the same as that noted in the sediment: Fe> Zn> Pb> Cu.

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