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Spatiotemporal Bioaccumulation of Lead, Cadmium, Zinc and Copper Metals in *Patella ferruginea* Collected in Two Algerian Coasts.

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ABSTRACT

The previous works carried out in 1994 showed the presence of metal pollution in the western coasts of the Mediterranean (Spain), leading to contamination of shellfish and fish by lead and cadmium. On the other hand, the Algerian coast showed the pollution in cadmium and zinc in mussels during year of 2007. This present work is devoted to the study of the quality of Mediterranean coastal waters of North-West of Algeria. So, it provides an evaluation of the quality of sea water in two close Algerian coasts, namely Honaine and Béni-Saf. The study was seasonally followed from the years of 2011 to 2012 with measuring the hydrological physico-chemical parameters (pH, temperature, turbidity and salinity), and using the mollusk (*patella ferruginea*) as an excellent quantitative bio-indicator of the metal contamination (Cd, Cu, Pb and Zn). Shellfish and seawater samples were picked in each season and in the three stations, characterized by sewage and industrial wastewaters, and by the recent desalination waste resulting from desalination unit placed in Honaine. Metal concentrations in seawater showed significant variations in function of stations and the harvest season, giving rise to the appearance of a pollution of Cd, Cu, Zn and Pb. In fact, the mollusk of the Honaine coast has presented higher levels of lead compared to that of Béni-Saf coast, exceeding the permissible standards.
Keywords: Patella ferruginea*,* Bio-indicator, Metal, mollusk, Honaine, Béni-Saf.

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INTRODUCTION

In the recent years, the dispersion of metal pollutants in Mediterranean areas (western Algeria) doesn't cease to increase. This situation is related to the wastewaters discharges of municipal, industrial and maritime traffic (oily waste) [1-2]. Currently, the protection of marine heritage constituted a challenge for the industrial and scientific communities for the economic and ecologic interests.

The Algerian coast regrouped a wide range of habitats where the marine biodiversity is considered among the highest in the Mediterranean pool. The fauna species of mollusk as: *patella ferruginea*, *panaessussp. and crasustreagasar* have the distinction of establishing the balance of the aquatic ecosystem. The patella (*gastropoda)*, this benthic specie colonized the mediolittoral upstairs plays a crucial role in the functioning of coastal ecosystems, and are a good indicator of environmental health. The role of patella in structuring communities of the rocky substrates is well documented [3].

In April 2002, there was a civil program for inspecting the quality of Mediterranean coasts (MEDPOL). In this program the researchers agreed the use of some fauna and flora species as quantitative bioindicators of metal traces. However, these species of wildlife such as mussels (*mitillusgalloprovincialis*, *mitillus sp*.) are located in the French, Spanish and Italian coasts for at least a few decades [4], also in the Moroccan coast [5] and the Tunisian coast [6]. In fact, other species as the *donaxtrunculus* and *mactracorallina* mussels including the African mussel (*pernaperna*, b*rachidontesvariabilis*) were used in the control of mercury, cadmium, lead, copper and zinc in the Egyptian coast [7-10]. While the flora species like the brown seaweed "*cystocera*" was used in the control of arsenic, chromium, cadmium and cobalt in Syrian coast. Deposition and bioaccumulation of heavy metals in fish and shellfish is very observable. This has been shown by many studies in different parts of the world [11-16]. In Algeria it's mostly *pernaperna* and *mitillusgalloprovincialis* who presented as quality control bioindicators [17]. In the present work we have considered the species that are abundant in our study area.

The *patella ferruginea* [18] is a protected mollusk species in the Mediterranean. Its distribution area was greatly reduced to the point that it exists only in Corsica, Sardinia, in south-eastern Spain and North Africa (Algeria, Tunisia and Morocco). The largest populations are found in Lavezzi Islands and Scandola [19]. With a diameter up to 8 cm, it is the largest Mediterranean *patella*. It is easily recognized by its size but also by its marked shell of scaly coast, which is also very thick and solid. She lives attached to rocks in the bottom of the mediolittoral upstairs, in the places beaten by the waves, just below *chthamalus* crustaceans (acorn barnacles) and slightly above the calcareous algae (*Lithophyllum lichenoides)* in a horizontal stripe very narrow (50-80 cm of wide). It moves to feed by rasping the algae along their way. Decimated by the fishermen (used as bait) and the pollution, this is probably the marine species the most endangered by the fast disappearing in the Mediterranean. In France, it is protected and prohibited of fishing since 26 November 1992. It is important to note that the *patella ferruginea* is considered, according to EEC as a species in danger because it is on the way of extinction [20].

The biotope is characterized by pollutants presence issues from agglomerations. On the Algerian coast, the researches on this species are rarest and are limited to the surveys. For this, we have undertaken an eco-biological study on the *patella*, at the north coast (Béni-Saf and Honaine), on its distribution and its exposure to heavy metal pollution.

The coastal region of Béni-Saf is located in the northwest coast of Algeria, 67 km from the Tlemcen city. Then, the coastal area of Honaine is halfway between Ghazaouet and Béni-Saf cities (Fig. 1). The Béni-Saf city, populated by 42,000 inhabitants [21], is marked by the beaches which attract a considerable flow of summer visitors. This harbor area was the second fishing harbor with an average production of 5000 tones/year [22], today demonstrated a real fall, to the point that some fish (*mullet, mirou*) and mollusks (*patella ferruginea*) have become rare. This area includes industrial activities (cement) and shipping. However, the area of Honaine (see Figure 1) populated by 5408 inhabitants [21] reveals urban and artisanal fishing activities, which is moderately developed compared to Béni-Saf city. Especially, this area has been equipped with a desalination unit, this will probably ask for any pollution against the saline discharges. The presence of toxic metals, from different discharges, manifested on the harbor coasts causing the degradation of the marine heritage [2].

The aim of this present work is to achieve a comparative study of the environmental quality of the two areas (Béni-Saf and Honaine) for determine the possible transfer of pollution between the areas taken into consideration, and raise the bioindicateur character of our specie. This will be conducted based on the spatiotemporal evolution of the quality of sea water and on the bioaccumulation of mollusk (*patella*) by toxic metals as: Pb, Cd, Cu, Zn, caused by the different discharges.

MATERIALS AND METHODS

The mollusk (*patella ferruginea*) was collected seasonally during 2011-2012 in the stations shown in Fig. 1. The choice of stations (S) is based on the presence of the mollusk, at the vicinity of discharges of sewage effluent (collectors; C), in order to evaluate the rate of the metallic pollutants bioaccumulation in the mollusk. The description and nature of sewage emissary are summarized as follows:

C1 (Béni-Saf): Collector receiving a mixture of urban and industrial wastewater (cement, marine traffic and fishing);
C2 (Honaine; beach): Collector receiving an urban wastewater from the Ouled Youcef city;
C3 (Honaine; harbor): Collector receiving water mixture (urban and artisanal fishing activities).

Sampling stations

The sampling stations are distributed as follows (Fig.1).

S1: Represents a point in the beach and near the harbor of Béni-Saf city, next to the C1 collector;
S2:Represents the beach of the Honaine, next to the C2 collector;
S3: Represents the harbor of Honaine, next to the C3 collector.

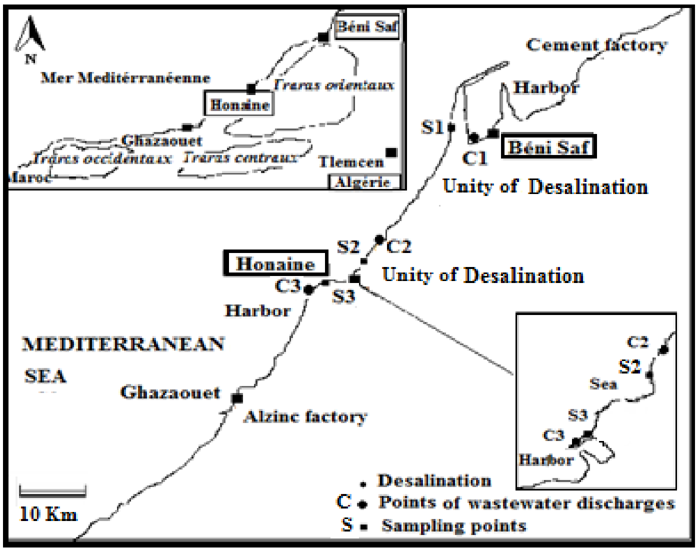

Figure 1: Localization of sampling sites of mollusks along the Béni-Saf and Honaine coasts

Collecting and sampling

The sea water and mollusk samples are carried out in three sampling points, distributed as follows:

A sampling point which borders between the harbor and the beach area of Béni-Saf (S1); Two sampling points in the Honaine area, in the vicinity of the beach (S2) and the other in the harbor (S3).

The sea water samples were taken during the year of 2011 to 2012, according to the protocol described previously [23].The physicochemical factors measures such as pH, electrical conductivity, air-water temperature and salinity, were made in situ.

Mollusk

Patella samples were snatched from the rock before being transported to the laboratory in ice box. The soft part of the flesh is pulled by a scalpel scissor and washed with ultra-filtered water (Millipore) to remove the external impurities, after the sample is maintained at 4°C. An amount of one gram of sample is taken after thawing. This quantity was dried in an oven at 110°C for 3 hours and then it is put into a muffle furnace for 15 minutes at 450°C, for a reduction in ashes. Then, the sample is moistened with a few drops of nitric acid (67% m/m) and back again in the muffle furnace at 350°C for a period of 90 mn. The obtained ashes were dissolved in 25 ml of nitric acid (1% w/w) and filtered through filter paper. In the end, the mineralization is conducted to assay for the quantification of metals by atomic absorption spectrophotometer.

Analytical instruments

The determination of metal contents of Pb, Cu, Cd and Zn was made by a graphite furnace atomic absorption spectrophotometer (PerkinElmer4110 type ZL, Graphite Furnace THGA). A conductimeter type TetraCon325 was used for conductivity and salinity measurements. While pH measurements were made by a pH-meter type ADWA1030, and the turbidity measurement by an apparatus of Hach2100.

Analytical performance

Detection limits

The limits of detection (LOD) were calculated relatively to the standard deviation, based on the arithmetic mean value. They are given in tables 1 and 2 for the sea water and the mollusk, respectively.

Table 1: Limits of detection (LOD) of sea water assays by atomic absorption spectrophotometry.

	Cd	Cu	Pb	Zn
LOD (ppm)	0.03	0.05	0.10	0.05

Table 2: Limits of detection (LOD) of mollusk assays by atomic absorption spectrophotometry.

	Cd	Cu	Pb	Zn
LOD (ppb)	0.05	8	0.5	2

Accuracy

Measurement accuracy was evaluated in function of the assay result of the certified sample (mollusk), obtained from the Department of Chemical Engineering and Environmental- Sevilla-Spain. Indeed, the error did not exceed five percent.

Statically treatment

The statistical treatment of the data is performed by using the Minitab program (trial version 16, 2012) to calculate the correlations between the analyzed metals and hydrological parameters.

RESULTS AND DISCUSSION

The seawater features of coastal towns; Béni-Saf and Honaine during the 2011-2012 campaign are given for the three stations in Table 3.

The physicochemical parameters of seawater, identified during the four seasons, indicate the status of the marine water quality in coastal areas of Béni-Saf and Honaine. The observed values indicate that the pH is neutral in the three stations both in wet and dry seasons. Indeed, the average pH value in both stations (S1 and S3) which receive the influence of urban and industrial discharges is 7.38. The latter is less than 8, thus indicating a slight acidification of the media. However this pH value remains in the Algerian standards recommended in the case of discharges in the receiving media [24].

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Table 3: Seawater features of coastal areas of Béni-Saf (S1) and Honaine (S2 and S3) during 2011-2012 campaign (limit values, mean ± standard deviation).

Stations	pH	Salinity (%)	Conductivity (µS/cm)	T_{water} (°C)	NTU* (mg/l)
S1	7.13-7.34 7.2±0.09	29.8-37.1 32.23±3.44	46.1-55.7 49.3±4.54	26-24.2 25.4±0.84	0.301-0.557 0.386±0.01
S2	7.25-8.11 7.54±0.33	25-37 29±0.47	50-55.7 51.9±2.68	24.3-29 25.86±2.21	0.203-0.854 0.42±0.025
S3	7.33-8.03 7.56±0.27	36.3-37.1 36.57±0.37	54.8-55.9 55.17±0.51	24.4-25 24.6±0.28	0.143-0.383 0.223±0.009

*NTU *(Nephelometric Turbidity Unit)*

The measurement of turbidity in the three study sites showed different values but they are less than 5. This is in accordance with the standards [25]. These results were predictable because the expertise is in agreement with surface waters which are clear.

The salinity of the three stations ranged from 29% to 36.57% during the annual campaign. It is in the standards that provide content between 35% and 39% [26].The salinity of closed or isolated seas may be different from that of the great oceans. The lowest average value is recorded at S2, which is located near the C2 collector (beach Honaire). The salinity can also vary during the year depending on the season. The conductivity of the sea water is subject to large variations depending on temperature and salinity. Our results of conductivity range from 49.3 to 55.17µs.cm^{-1}. The latter is found in the S3 station next to the collector C3 (wastewater from craft activities). However, these results are substantially in standards since the electrical conductivity is in the order of 40-53 µs.cm^{-1} at a temperature of 25 °C [27]. The temperature acts on the phenomenon of photosynthesis, which is responsible on the multiplication of the increase of our species (algae), whose role is very important in the solubility of salts and especially of the gases. On the other hand, it influences on the pH. The median value of 7 at 25 °C corresponds to a neutral solution [28]. It also permits the correction of the analysis parameters as conductivity values which are related to temperature.

Furthermore, by highlighting the water temperature contrasts on the media, it is possible to obtain information on the origin and water flow [29]. The temperatures of the sea water, in the three stations range between 24.60 °C and 25.86 °C. These ambient temperatures correspond to the climate prevailing at the time of sampling. They are in agreement with the Algerian discharge standards [24].

Metals contents in seawater

The heavy metals contents in seawater were calculated for the three stations, and presented in Table 4.

Table 4: Levels of metals (mg/l) in west coastal seawater of Béni-Saf (S1) and Honaine (S2 and S3); (limit values, mean ± standard deviation).

	Cd	Pb	Cu	Zn
S1	0.071-0,86 0.33±0.03	0.49-0,65 0.54±0.01	0.025-0,58 0.21±0.02	0-4.25 1.42±0.16
S2	0.022-0.25 0.098±0.01	0.016-0.69 0.24±0.02	0.058-0.083 0.066±0.01	0-0.35 0.12±0.01
S3	0.056-0.11 0.074±0.001	0.35-0.70 0.47±0.01	0.079-0.088 0.082±0.01	0-7.67 2.56±0.30

Cadmium

The observed seasonal concentrations reveal significant levels of cadmium in the three stations. The highest content was found in autumn, equal to 0.86 ppm, at the station S1 of the coastal area of Béni-Saf. The average annual value of cadmium was 0.33 ppm. This result is alarming as to the quality of the meadia, since cadmium content exceeds the standards of Mediterranean coastal areas, prescribed by FAO that provide levels of 0.01 ppm *10^{-3}[30]. This is due to urban and industrial waste (cement, shipping and fishing) that are discharged by the collector C1, which is located near the station S1.A similar result was found in the South Atlantic coast of Spain [31]. In fact, the levels of cadmium are found tolerable compared with discharge standards in the receiving media, defined by Algerian regulations and which shows the security levels at 3 mg/l [24].

Lead

The seasonal quantification of lead in seawater showed very high levels in three stations during the annual campaign. The highest value was observed in the station S3 (Honaine harbor), in spring. It was 0.7 ppm while the annual average content was of 0.54 ppm. The lead content is significantly higher than $0.03*10^{-3}$ppm which is the threshold requested by FAO [30-32], and at 0.5 ppm, requested by the Algerian regulations [24]. Regarding the annual change of lead only the station (S1) which exceeds the Algerian standards discharges into the receiving media. This allowed noting that the area of Béni-Saf (S1) is contaminated with lead. The hypothesis of contamination by lead due to the collector of discharges (C1) remains valid as previously. These results are in agreement with the previous work [31].

Copper

The copper seasonal levels are observed at values that exceed the standards of Mediterranean coastal waters (FAO), they are above the threshold value of 0.001 ppm [30-32], and this in the three stations. Indeed, an annual average value of 0.21 ppm is identified at the station S1. The copper pollution is probably due to the discharge of collector (C1). Previous work reported on the dispersed copper in the South Atlantic coast of Spain, showed similar findings [31] .On the other hand, the average annual copper remain below the Algerian standards on discharges in the receiving media, which reveals a limit value of 0.5 ppm [31].

Zinc

The zinc concentrations are high in the three stations during all seasons except summer. They go well beyond the threshold limit which is $2.5 * 10^{-3}$ppm, defined by FAO [30-32]. An important value equal to 7.67 ppm in zinc was found in autumn and at the station S3 (Honaine harbor). However, the annuel average content was of 2.56 ppm. The station S1 has presented a zinc peak in winter, where the concentration is 4.25 ppm. This latter reflects the pollution of the harbor area of Béni-Saf. This is due to the urban and industrial discharges from the collector C1. Thus, the contamination of the harbor site of Honaine area (S3), is due to the waste from collector C3, which probably receives the effluents from the zinc production unit (Al-Zinc) which is not far from our study site. This finding of pollution is similar to that given previously and which covers the seawaters of the Atlantic coast of the South of Spain[31]. Comparing with the Algerian standards, defining a threshold limit of 0.5 ppm, the pollution situation is still maintained [24].

In conclusion on the quality of our seawater, the gradient of annual accumulation of heavy metals in the three stations is as follows: Zn>Pb> Cd> Cu

Levels of heavy metals in the mollusks

The heavy metals content in the mollusks are calculated for the three stations, and presented in table 5.

Table 5: Levels of metals (mg/kg fresh weight) of the mollusk (*patella ferruginea*) collected from the west coast areas of Béni-Saf(S1) and Honaine (S2 and S3);(limit values, mean ± standard deviation).

	Cd	Pb	Cu	Zn
S1	0.091-0.38 0.19±0.01	0.001-0.59 0.20±0.02	0.3-2.16 0.92±0.07	18.01-82.63 39.55±2.53
S2	0.006-0.34 0.12±0.01	0.0012-2.06 0.69±0.08	0.025-0.58 0.21±0.02	22.15-131.8 58.7±4.30
S3	0.04-0.24 0.11±0.01	0.01-3.83 1.28±0.15	0.01-0.61 0.21±0.02	0.04-101.26 33.78±3.97

Cadmium

The seasonal content of cadmium bioaccumulated in the mollusk "*Patella ferruginea*" at the three stations are weak and showed an uniformity in concentrations level during the different seasons (see Fig. 2). The Highest content was found in autumn, equal to 0.38 mg/kg fresh weight and this at the station S1, near the collector C1 of the coastal area of Béni-Saf (table 5).The annual average content of cadmium in the flesh was 0, 19 mg/kg fresh weight. This result is in agreement with the work reported on the *patella caerulea*, where they found that this species bio-accumulates 0.45 mg/kg fresh weight of cadmium. So in this case, the bioaccumulation limits range from 0.15 to 0.75 mg/kg fresh weight of cadmium [1]. Other work of Boyden [33] showed that in general, the cadmium bioaccumulation in patella varied between 1.37 and 2.05.10^{-3}mg/kg fresh weight. The content of cadmium bioaccumuled by our species does not exceed the standards of Mediterranean coastal areas, prescribed by the world health organization (WHO) that provide levels of 2 mg/kg fresh weight [30-32]. However, this bioaccumulation of cadmium in the mollusk remains tolerable compared to the official standards of the European Communities (OSEC), which sets a threshold level of 1.0 mg/kg fresh weight in mollusks [34]. In our *patella ferruginea*, the bioaccumulated cadmium may be attributed to the receiving media which is contaminated by cadmium and to the individual's size of the species (maturity). This was shown by the work done on the bioaccumulation of cadmium in *patella vulgata* at the side of the France-England channel, which range from 3.6 to 5.3 mg/kg fresh weight over the species average size of 2 to 3 cm [35].

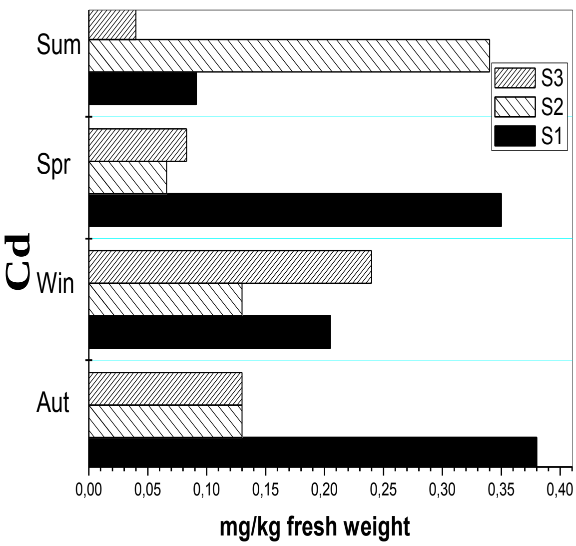

Figure 2: Seasonal concentrations of cadmium in mg/kg fresh weight in the three stations (S1-S3) in "*Patella ferruginea*", during the period (2011-2012).

Lead

The seasonal quantification of lead in the mollusk, showed very high levels in the stations S2 and S3 (table 5). Two peaks were observed in the station S3 (Honaine harbor) in spring and summer at 3.83 mg/kg and 2.51 mg / kg fresh weight, respectively (Fig. 3). These lead levels are much higher than the standard prescribed by the (WHO) which provides a level of 2 mg/kg fresh weight [34-36]. Although, the annual average content of lead which is of 1.28 mg/kg fresh weight, it remains within these standards. However, these lead levels exceed the official standards of the European Communities (OSEC), which sets a threshold of 1.0 mg/kg wet weight in mollusks [34].The previous work on the Spanish coast, Parstor et *al*. found that the mollusk *patella caerulea* has bioaccumulated 0.98mg /kg fresh weight [1]. So, they considered that as a bioindicator of lead pollution because it's in the range of bioaccumulation (0.67 to 1.29)[1].

The bioaccumulation of our studied specie (*patella ferruginea)* is in the range of lead concentrations, given by *patella caerulea* indicating that this marine animal is a bioindicator of lead pollution in the surveyed area including Honaine (S3). Boydena showed that the *patella* may bioaccumulate the trace amounts of lead to $0.75.10^{-3}$ mg/kg.mm depending on its size [33]. This finding can be attributed to the receiving media which was contaminated with lead in the harbor area (S3), and to the size of the individuals specie (maturity).The exchanges that occur between an aquatic organism and its environment accumulate in its flesh and they are produced through the integumentary or digestives epithelia gill, the absorbed elements pass into the internal environment where they are stored and concentrated in the organs or target tissues such as the digestive gland, muscle, gonad or the supporting tissues as in the sea urchin (*paracentrotuslividus)*[37].

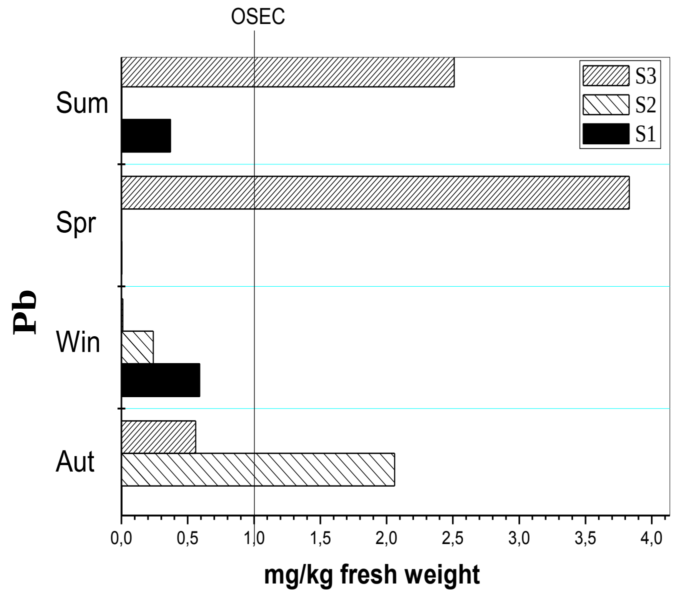

Figure 3: Seasonal concentrations of lead in mg/kg fresh weight in the three stations (S1-S3) in "*Patella ferruginea*", during the period (2011-2012).

Copper

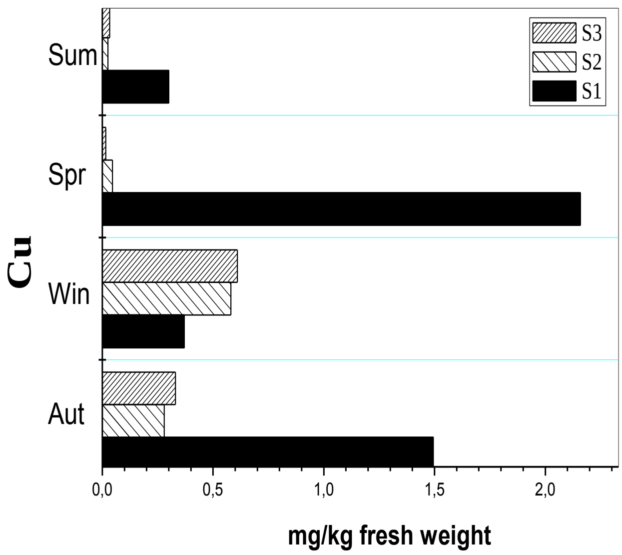

Figure 4: Seasonal concentrations of copper in mg/kg fresh weight in the three stations (S1-S3) in "*Patella ferruginea*", during the period (2011-2012).

The seasonal levels of copper, identified in *patella Ferruginea*" showed two peaks in the station S1 (Béni-Saf) in spring and autumn at 2.16 mg/kg and 1.49 mg/kg fresh weight, respectively (Fig.4). Thus, the annual average content of copper was of 0.92 mg/kg fresh weight (table5). The copper contents in the three stations are well lower to those of (WHO) standards which provides a threshold concentration of 30 mg/kg fresh weight [34,36]. These results do not really reflect the high copper concentrations in the receiving media. Our results are similar to those found by Bergasa et al. on the coasts of Canary Islands, which have shown that the species "*Patella Piperata*" has bioaccumulated 2.051 mg/kg fresh weight [38]. In another work on the Italian coasts, Cubadda et al. has identified a quantity of 1.67 mg of copper per kilogram of the fresh species of *patella caerulea* [39]. Although the bioaccumulated copper quantities by these different species, which are lower compared to standard, the authors suggest that these species are considered as cupric pollution biomarkers. So our species "*Patella ferruginea*" is too. This is discussed by Boyden which showed that the patella may bioaccumulate copper traces to $0.73.10^{-3}$ mg/kg.mm depending on its size [33]. In fact, copper is an trace-element, essential for the establishment of aquatic organisms, it is involved in metabolism or in specific physiological functions, it represents a component of the respiratory pigments of mollusks and crustaceans [40].

Zinc

The seasonal concentrations of zinc in the mollusk "*Patella ferruginea*" showed the lower values in all stations. Two peaks were observed in the stations S2 and S3 at 131 mg/kg fresh weight (autumn season) and 101 mg/kg fresh weight (spring season), respectively (Fig.5). This result can be attributed to the increased discharges of urban wastewater from the Ouled Youcef area, during the summertime. Also, the deposition of zinc during the dredging works and installation of the desalination unit (since 2008) could be behind such a zinc bioaccumulation by our mollusk. The results found do not reflect the quality of the receiving media which was polluted by the zinc. However, the annual average content bioaccumulated in zinc is 58.7 mg / kg fresh weight (table5). These results are not alarming, since the zinc content did not exceed the standards prescribed by (WHO) that provide levels of 1000 mg/kg fresh weight [34-36]. The previous work of Hamed et al., realized on the Egyptian coast using the mollusk "patella caerulea," have shown that the species has bioaccumulted 123.95 mg/kg fresh weight [41]. On the other side, Cubadda et al. have found that their mollusk "*patella caerulea*" has bioaccumulated only 3.9 mg/kg fresh weight, when inspecting the Italian coast [39]. In view of this, our specie "*Patella ferruginea*" is considered as bioindicator of zinc pollution especially in station S2 (Honaine beach). Indeed, Boyden showed that patella may bioaccumulate the traces of zinc from 0.84 to 0.93 mg /kg.mm depending on its size [33].

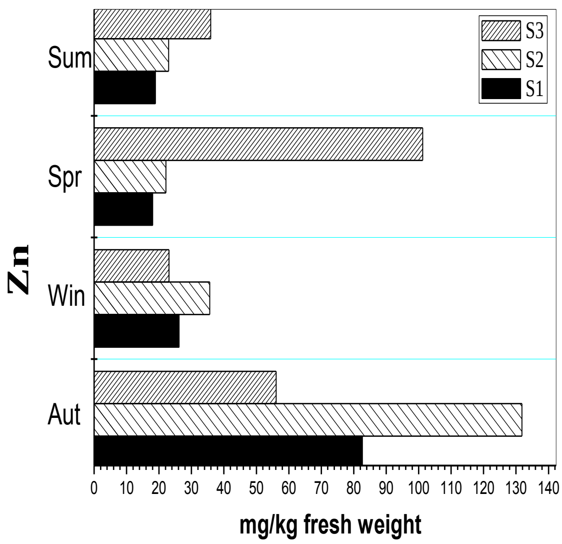

Figure 5: Seasonal concentrations of zinc in mg/kg fresh weight in the three stations (S1-S3) in "*Patella ferruginea*", during the period (2011-2012).

From the results, it is found that the most seasons affected by the cadmium and lead bioaccumulation in "*patella ferruginea*" are the spring and summer. As against, the copper is absorbed throughout the year much more in spring than autumn. In fact, the zinc bioaccumulation is as evident in autumn and spring than in winter and summer. This may be attributed to temporal changes in metal content based on the physicochemical characteristics of the seawater and to the reproductive cycle in "*patella ferruginea*", that develops in the spring and summer, and reaches their sexual maturity in autumn in order to reproduce [37].

Metal pollution index

The total metal content in our "*Patella ferruginea*", in the considered stations by this present study, was compared using the metal pollution index (MPI), calculated with the equation 1 [42]. The results are given in table 6.

$$PMI = (Cd * Pb * Cu * Zn)^{1/4} \quad (1)$$

Table 6: Metal pollution index in the three stations during the campaign (2011-2012)

Stations	PMI* (annual)	PMI^{Aut}	PMI^{Win}	PMI^{Spr}	PMI^{Sum}
S1	1.084	0.825	1.044	0.607	0.651
S2	1.005	1.770	0.892	0.111	0.217
S3	0.999	1.076	0.428	0.527	0.432

Although the species sites of "*Patella ferruginea*", the annual metal pollution index is practically identical and this in the three studied stations. The highest annual average index is observed in the Béni-Saf Station. This is attributed to industrial wastes resulting from harbor traffic and to urban wastes coming from the collector C1. The patella of Honaine harbor (S3) showed a low pollution index which reveals the modest activities developed during the years of our campaign.

The seasonal evaluation showed that the high metal pollution index has been identified at the beach of Honaine (S2), in autumn. This is due to the results of the intensification of urban wastewater during the summertime and to the consequences of the recent dredging activities related to installation of the desalination unit, which are behind the decantation phenomena in the following season. Indeed, the lowest seasonal pollution index was observed in the same station (S2), but in spring. It becomes more important (0.892) in winter because of the overland flow, giving rise to metal releases. Referring to previous work [43], the explored stations are not considered as polluted because the metal pollution index did not exceed two.

Based on our results of bioaccumulation, the deposition of heavy metals in our *patella ferruginea* confirms an annual bioaccumulation gradient in the three stations as follows: Zn>Pb> Cu> Cd. Thus, *patella ferruginea* seem be as a biomarker of the heavy metals especially in S2 and S3 stations. This may be related to nutritional practice and to the morphological and physiological effects of *patella ferruginea*. In the case of this specie, the lead concentrations have manifested instead of cadmium in the area of Honaine (S3). This result is the opposite of what was found by Nakhle working on patellidaes species [3].

Role of interactions

The response and survival of a metal depends on the other metals present in the environment, in relation to the potential role of the environmental factors. In fact, these positive or negative interactions exert an influence on the bioavailability of metals and hence on their bioaccumulation [44]. The study of metal interactions controls the difference between the metals in the mollusks samples which are function of size and origin [45].

Study of metal interactions

We have evaluated these interactions by the study of correlations between metals calculated in *patella ferruginea* in both coastal areas Béni-Saf and Honaine. The results are given in tables 7and 8 for Béni-Saf and Honaine, respectively.

Table 7: Correlation matrix between metals analyzed in the mollusk "*Patella ferruginea*", in the coast of Béni-Saf.

	Cd	Pb	Cu	Zn
Cd	1.000			
Pb	-0.781	1.000		
Cu	0.874	-0.896	1.000	
Zn	0.594	-0.465	0.241	1.000

Table 8: Correlation matrix between metals in the mollusk "*Patella ferruginea*", in the coast of Honaine.

	Cd	Pb	Cu	Zn
Cd	1.000			
Pb	-0.786	1.000		
Cu	0.449	-0.886	1,000	
Zn	-0.139	0.034	0.275	1.000

For the study area of Béni-Saf, the examination of correlation matrices showed that cadmium interacts negatively with lead and positively with copper, suggests a synergistic action between the metals. However, lead interacts negatively with the copper (Table 7). This result of interaction remains valid in the case of Honaine area (Table 8). While the interaction between cadmium and lead is negative. Indeed, a competition in bioaccumulation of lead and cadmium metals is observed in our *patella ferruginea*.

Study of interactions metal-media

To assess the impact of the quality of the receiving media on the bioaccumulation of the heavy metals in *patella ferruginea*, we also correlated the changes of metal concentrations in the flesh of the *patella ferruginea* with the parameters relating to the quality of seawaters, in the two study areas. And this, by using the overall data on all explored sites for tries to identify the potential influence of these parameters the metal loading. The results are given in tables 9 and 10 for Béni-Saf and Honaine, respectively.

Table 9: Matrix of correlations between metals in the mollusk"*Patella ferruginea*" and the water quality of the media, in the coast of Béni-Saf.

	Cd	Pb	Cu	Zn
pH	-0.571	0.955	-0.813	-0.258
T (°C)	-0.787	0.314	-0.656	-0.157
Salinity	-0.508	0.510	-0.215	-0.979
Conductivity	-0.034	0.328	0.107	-0.726
Turbidity	-0.034	-0.118	0.010	-0.629

The most significant correlation coefficients are observed for zinc in the two study areas. This is in agreement with the accumulation gradient of zinc in our mollusk. From table 8, the cadmium and zinc metals are negatively correlated with all studied parameters of the receiving media. For the case of lead, only its interaction with the turbidity is negative. Copper is correlated positively with conductivity and turbidity, and negatively with pH, temperature and salinity.

Table 10: Matrix of correlations between metals in the mollusk"*Patellaferruginea*" and the water quality of media, in the coast of Honaine.

	Cd	Pb	Cu	Zn
pH	0.674	-0.450	0.019	-0.824
T (°C)	0.440	0.173	-0.601	-0.478
Salinity	-0.147	-0.263	0.278	-0.668
Conductivity	-0.824	0.773	-0.419	0.610
Turbidity	0.280	-0.339	0.513	0.908

In the case of the area of Honaine (Table 9), we observed a good correlation between the zinc and turbidity where lead is positively correlated with conductivity. This last is negatively correlated with cadmium. Thus, copper is negatively correlated with temperature. These anti-correlations are interesting because they reflect the continental origin of the metals. The conductivity and turbidity factors play an important role for a potential pollution in cadmium, lead and zinc, in the coast of Honaine. These two parameters influence the quality of seawater and the availability of metals [44]. These results show clearly the bioaccumulation of metals in *patella ferruginea* which is manifested by a direct influence with the media where the mollusk lives [44].

CONCLUSION

The dispersion study of heavy metals as : Cd, Cu, Pb and Zn in seawater of the Béni-Saf and Honaine coasts showed that, theirs levels ranges in function of the sampling site and the season. The cadmium levels increase in autumn, winter and summer in the Béni-Saf station (S1), and in spring in the Honaine stations (S2 and S3). With the exception of copper, the highest heavy metals concentrations are detected in the receiving media, in winter and spring, at the station S3. The latter is characterized by discharges of domestic and industrial wastewater of collector C3. The accumulation of heavy metals in seawater is in the following order: Zn>Pb> Cd> Cu.

The observed bioaccumulation of heavy metals in the *patella ferruginea* in the Honaine station (S3) is higher than in the Béni-Saf station (S1). This is resulting from the contamination of mollusk by lead. Then the bioaccumulation gradient is as follows: Zn>Pb> Cu> Cd.

With increasing the urbanization and urban activities, the rate of heavy metals will increase in the time. So, the purification of the waste by a suitable ecological treatment system will be necessary if we want to keep the protection of our *patella ferruginea* and control the biotope and marine biodiversity, in the coast of BéniSaf and particularly Honaine.

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