Assessement of pesticides and heavy metals contents in edible seaweeds from El Jadida region coastal zone, Morocco and their safety for human consumption

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Abstract. The aim of the study was to assess the distribution of pesticides and heavy metals contents in edible seaweeds species in El Jadida region coastal zone, and to evaluate the threshold limit values of seaweeds intake without risks to human health. Eleven seaweed species (*Laminaria ochroleuca*, *Sargassum vulgare*, *Fucus spiralis*, *Ulva lactuca*, *Gelidium spinosum*, *Gelidium corneum*, *Gelidium pulchellum*, *Chondracanthus acicularis*, *Gracilaria* sp., *Gracilaria multipartita*, and *Hypnea musciformis*) were collected seasonally from the coast of the studied area from spring 2022 to winter 2023, and were analyzed for pesticides 175 organochlorine, 42 organophosphorus, and 21 organohalogens, and for heavy metals (HVMs) contents (As, Pb, Cd, and Cr). Pesticides were determined using LC-MS-MS method, and heavy metals contents (As, Pb, Cd, and Cr) were analyzed by ICP-MES method. The contents of all pesticides were under the limit of quantification ($LQ \leq 0.010$ µg g^{-1} dry weight (DW)) of seaweeds, so under the limit of toxicity risk. HVMs contents in the seaweed samples from the studied zone were found to follow a bioaccumulation pattern: As > Cd > Pb >Cr. Except for the autumn sample of the Rhodophyceae species *C. acicularis*, which showed very high contents for all the four HVMs, the rest of species mean contents ranged from 2.45 to 10.28 µg g^{-1} DW, 0.09 to 5.34 µg g^{-1} DW, 0.48 to 2.9 µg g^{-1} DW, and 0.22 to 0.81 µg g^{-1} DW, respectively for As, Pb, Cd and Cr. Seasonal variation was highly significant for As, Pb and Cr ($p < 0.005$), and HVMs contents were high in the most period of the study, higher than the toxicity threshold limit values, mainly in the spring and autumn, and dropped down to very low level in the winter. The highest amounts of As were recorded in the Phaeophyceae, *Sargassum,* and *Laminaria* in the spring, *F. spiralis* in the spring, summer and autumn, and *C. acicularis* in the autumn with values ranging from 11 to 27.7 µg g^{-1} DW. Pb contents were very high in the autumn and very low in the spring and winter. Except the very high Pb content which was detected in *C. acicularis* (24.79 µg g^{-1} DW, the contents ranged from 0.67 to 1.45 µg g^{-1} DW in the autumn. The highest Cd values were recorded in *F. spiralis, U. lactuca* and *G. corneum* (4.8 to 6.9 µg g^{-1} DW). The highest Cr values were in *U. lactuca* (2.08 µg g^{-1} DW) and *G. corneum* (1.59 µg g^{-1} DW).

Key Words: bioaccumulation, edible seaweeds, El Jadida region coastal zone, heavy metals, human health, intake threshold limit, pesticides.

Introduction. Seaweeds have been consumed as feed in Asia during centuries ago, and only occasionally in other parts of the world (Nisizawa et al 1987). Seaweeds are beneficial to humans and animal nutrition (Caliceti et al 2002). Numerous species are edible and have been utilized in both industry and agriculture (Gomez-Ordonez 2012; Jimenez-Escrig et al 2012; Khan et al 2015; Al-Homaidan et al 2021), and as pharmaceutical and cosmetic products, as well as in the food, feed and as food additives due to their distinguished chemical composition rich in bioactive compounds of high nutritional and dietetic value (Peñalver et al 2020; Skrzypczyk et al 2023).

Seaweeds are a very important source of bioactive substances of high value such as protein, fatty acids and PUFA, polysaccharids, fibers, minerals, polyphenols (Chen et al 2021). Thus, they are considered of high economic interest, and seaweed industry is inceasingly thriving, relying on the exploitation of wild resources and aquaculture. The production was 35.1 millions tonnes in 2020 (FAO 2020). Nevertheless, this industry faces the main challenges of the exposure of seaweed resources to contamination with a wide range of hazardous substances, especially pesticides and heavy metals (HVM) that occur in high levels in seawater as a harmful impact of anthropogenic activities on coastal marine environment (Miltra et al 2021). Seaweeds have a large capacity of bioaccumulation of these contaminants (Sanchez-Rodriguez et al 2001), which poses a big issue regarding human health risks within consumption.

The use of pesticides has increased significantly in many countries in the world, because of their low cost and wide application in industry, agriculture, and sanitation. Their intensive use in agriculture specifically makes food consumption as a main route of human exposure to organochlorines pesticides (OCP) (Wells et al 2017; Kumar & Sharma 2021). It also constitute the main route of marine wildlife contamination including seaweeds. The pesticide residue concentrations in edible seaweed species may pose serious human health risks (Rodrigues et al 2018).

OCP are ubiquitous in the environment and may continue to pose health threat to both wildlife and human (Guo et al 2007). Another threat is heavy metal pollution in aquatic systems, which has become a global environmental pollution problem (Lü et al 2018). They are usually present at low concentration in aquatic ecosystem but deposits of anthropogenic origin have raised the HVM concentration, creating environmental problems in coastal zones, lakes and rivers (Kamala-Kannan et al 2007). Given the toxicity, persistence, and non-degradable nature of HVMs in the environment, HVM contamination represents one of the greatest ecological risks for marine ecosystems (Pekey 2006). Due to their ability of adaptation to a large range of environmental conditions, seaweeds are highly abundant in different ecosystems (Rajfur et al 2010) Due to their high capability of HVM bioaccumulation from water, even in low contentrations, seaweeds are amongst the most suitable organisms as bioindicators for studies of HVM contamination in aquatic ecosystems (Wallenstein et al 2009; Agarwal et al 2022). HVM concentrations in algae are strongly dependent on the environmental parameters of their habitat (salinity, temperature, pH, light, nutrient concentrations, oxygen, etc.) (Zbikowski et al 2006). However, the environmental characteristics of the water in which algae grow (salinity, turbidity, nutrient content and HVM contamination) largely determine the mineral content they can absorb (Riget et al 1997; Vasconcelos & Leal 2001; Lozano et al 2003; Marinho-Soriano et al 2006; Riekie et al 2006). So they have a huge potential as a tool for environmental bioremediation, as bioindicators and for pollution bioremediation by monitoring HVM pollution on many coasts and for wastewater treatment in controlled environments (Agarwal et al 2022). The capacity of algae to accumulate contaminants as metals depends on a variety of factors, the two most relevant ones are being the bioavailability of metals in the surrounding water and the uptake capacity of the algae (Sanchez-Rodriguez et al 2001).

Seaweeds are known to be organisms of great importance. But, their ability to bioaccumulate HVM, and a wide kind of toxic chemicals make them of a high risk for human consumption (Løvdal & Skipnes 2022). Nevertheless, seaweeds could be consumed carefully in a way that their supply in HVM could be kept less than the permissible intake rate of different heavy metals These limits of intake are: Cd ≤ 0.35 µg/kg bw/day, Pb ≤ 0.5 µg/kg bw/day, and As ≤ 0.3 to 8 µg/kg bw/day (EFSA 2009). The permissible consumption rate (PCR) of Cr was estimated in 1986 to be equal or less than 0.3 to 8 µg/kg bw/day (US EPA 1980). A seaweed daily consumption rate of 5 to 10 g DW per person (Bw of 85 kg) could be safe and sufficient to supply the body needs in essential micronutrients, including Cr (Skrzypczyk et al 2023).

In Morocco, El Jadida region coastline is a highly productive zone, where a very important upwelling occurs along the coastline making it one of the most important wildstock of seaweeds, especially agarophytes, and Phaeophyceae, and many edible species that could be exploided by the locals as a source of food and revenues. The present study investigates the state of contamination of some edible seaweeds with pesticides and some most toxic heavy metals (HVMs) as arsenic (As), lead (Pb), cadmium (Cd) and chromium (Cr), and the potential of their use in human consumption with minimum risk for human health hazard.

Material and Method

Description of the study site. The coastline of Sidi Bouzid (coordinates 33°12'30”-33°14'30” N latitude and 8°32'50.58" W longitude) is part of El Jadida maritime circumscription that extends on 150 km. It is located 5 km far from the city of El Jadida and 11.3 km far from the commune of Moulay Abdellah (Figure 1). This coastal region is highly subject of the influence of a coastal upwelling that makes this zone highly rich in nutrients and highly productive. The choice of the site was justified by the high productivity and diversity of edible seaweeds, the high frequency of visits by seaweeds fishers, and ease of access to the seabed and harvesting.

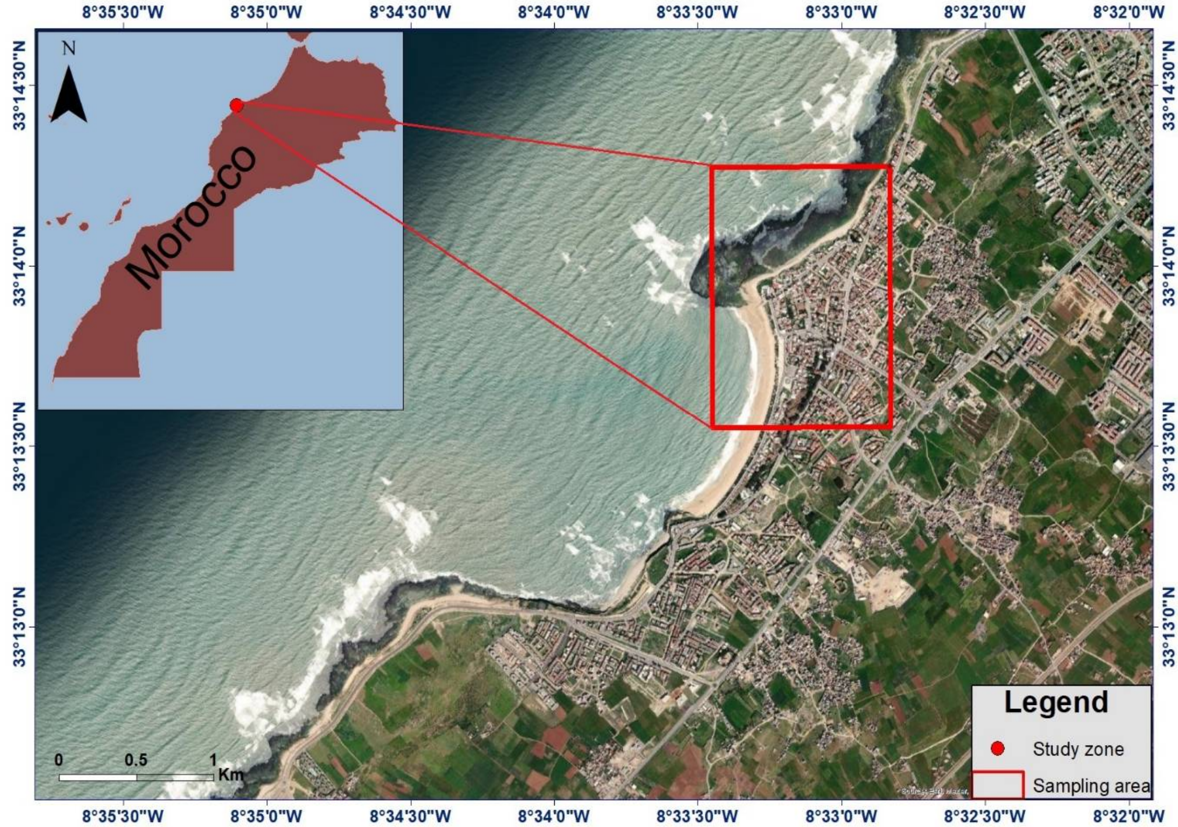

Figure 1. Map showing location of sampling site (Sidi Bouzid coast - El Jadida region).

Targeted species sampling. Selected species from the study area were surveyed for their contents in pesticides, namely 175 organochlorine, 42 organophosphorus and 21 organohalogens, and for some most toxic HVMs (As, Pb, Cd and Cr). Targeted species were selected relaying on the following criteria: edible species, and which were more abundant in the region. Seaweed samples belonging to the three main Phycophyta groups namely were: Phaeophycea (*Fucus spiralis* (Fu-spi), *Laminaria ochroleuca* (La-oc), *Sargassum vulgare* (Sa-vu)), Rhodophyceae (*Gelidium spinulosum* (Ge-spi), *Gelidium corneum* (Ge-co), *Gelidium pulchellum* (Ge-pu), *Gracilaria* sp. (Gr-sp.), *Gracilaria multipartita* (Gr-mu), *Hypnea musciformis* (Hy-mu), *Chondracanthus acicularis* (Ch-ac) and Chlorophycea (*Ulva lactuca* (Ul-la)).

Samples of the 11 taxa of seaweeds were harvested each time during four visits of Sidi Bouzid seabed. Approximately 1 kg of wet biomass of each target species was harvested by hand from the intertidal zone and the higher part of the infralittoral zone during spring tides, at low tide, from April 2022 to January 2023. In the case of *Laminaria ochroleuca*, due to conservation awareness, only very fresh drifted fronds were used as samples.

The samplings dates were 13 April, 19 July, 21 September and 4 January which were matched with the four seasons of a year: spring, summer, autumn, and winter. Some species were not found regularly at every seabed visit, and were sampled one time namely, *C. acicularis* and *G. multipartita* or twice, namely, *G. spinulosum and H. musciformis*. The samples were washed in situ with seawater, stored in food plastic bags, and transported to the laboratory in an isotherm box at low temperature, where they were cleaned from epiphytes, and washed with deionized water to remove sand and salt. Then, they were stored in the freezer at very low temperature until treatment.

***Seaweeds samples pretreatment and analysis**.* Seaweed samples of the 11 target species harvested in 4 seasons were pretreated and analyzed for pesticides and HVMs contents. Seaweeds were analyzed for 175 organochlorines, 42 organophosphorus, 21 organoalogens, arsenic (As), cadmium (Cd), lead (Pb) and chromium (Cr). All samples analyses were performed in triplicates and data quality was assessed.

***Seaweed samples pre-treatment**.* Seaweed samples were prepared as follows: 1 kg of wet biomass of each species was dried in an oven at a temperature of 50°C until constant weight and then was crashed using a food blinder, and homogenized in mortar.

***Pesticides residues analysis method**.* Extraction of pesticides from the seaweeds, using QuECHERS (NF EN 15662) extraction method was adopted. Pesticides were extracted with acetonitrile after the addition of magnesium sulphate, sodium chloride and buffer citrate salts. The organic phase was purified using a solid phase extraction (SPE) consisting of secondary primary amine (PSA) and magnesium sulfate and activated carbon in order to remove interfering substances and water residual.

About 2 g of dried samples were weighed in triplicate, put in a flask, and mixed separately with 8 mL of distilled water and 10 mL of acetonitrile and vortexed for 1 min. Then the following salts were added: 4 g of magnesium sulfate, 1 g sodium chloride, 1 g trisodium citrate dehydrate and the disodium hydrogen citrate sesquihydrate, and the whole components were vortexed for 1 min and centrifuged at 3500 rpm for 5 min. The extracted aliquot was transferred to a centrifuge tube containing 25 mg of PSA and 150 mg of magnesium sulphate and 15 mg of activated carbon. The purification was made using amino-adsorbent, a 6 mL of aliquot of acetonitrile phase obtained after the extraction in a tube containing 150 mg PSA, 900 mg of magnesium sulfate and 15 mL of activated carbon vortexed for 30 s and centrifuged at 3500 rpm for 5 min (LAB GTA 26). The analyses were performed with liquid chromatography tandem mass spectrometry LC-MS-MS (Agilent).

***Heavy metals analysis method**.* For each seaweed sample, a 1 g of dried biomass was weighed in triplicate, and was transferred into digestion flasks, and 2 mL of nitric acid was added to the sample. Then, digestion flasks were kept on a hotplate set to a temperature of 100°C for 2 h. The remaining digested solution was transferred to 30 mL flasks and diluted to a volume of 25 mL with distilled water. Then metal concentrations in the samples were measured using inductively coupled plasma-mass spectrometry ICP-MS, of Ultima Expert brand, of the Independent Institution for Exports Control and Coordination MOROCCO FOODEX, Larache, Morocco.

***Calculation method of the permissible consumption limit of the seaweed species harvested from El Jadida region coastline**.* The human permissible consumption limit of the sampled seaweed species (SW-PCL) was established for each species sample, and calculation was based on their HVM contents (HVM-C) (expressed in µg/g DW of seaweed), and the Permissible Consumption Limit of different HVM (HVM-PCL) expressed in µg per kg of body weight (bw) per day. The HVM-PCL is: Cd ≤ 0.36 µg/kg bw/day, Pb ≤ 0.5 µg/kg bw/day, and As ≤ 0.3-8 µg/kg bw/day (EFSA 2009). SW-PCL was not calculated for Cr due to the absence of a reliable HVM-PCL value issued by any health care organization. The calculation equation is: SW-PCL = HVM-PCL x $(HVM - C)^{-1}$.

Statistical analysis. The data obtained were subject to statistical analysis using Excel and two-way analysis of variance (ANOVA), Pearson correlations in order to test different HVMs correlations under R studio version 3.6.2. To determine any significant differences in the HVM contents in the seaweeds related to species and seasons. Correlations between different metals were established. Two variables are considered highly dependent and the correlation is significant when "r" exceeds 0.50, and p value < 0.05.

Results

Pesticides contents in the seaweed species. As the first step, the present study determined pesticides residues (175 organochlorines, 42 organophosphorus, 21 organoalogens) in seaweed samples: Phaeophycea (*F. spiralis*, *L. ochroleuca*, *S. vulgare*), Rhodophyceae (*G. spinulosum*, *G. corneum*, *G. pulchellum*, *Gracilaria* sp., *G. multipartita*, *H. musciformis*, *C. acicularis*) and Chlorophycea (*U. lactuca*).

All seaweeds samples contents in organochlorines, organophosphorus, and organoalogens pesticides were under the limit of quantification (LQ = 0.010 μg g^{-1}). All samples content complied with the European standards according to regulation EC 396/2005 ANNEX II and IIIB without taking into account the uncertainty associated with the results.

Heavy metals mean contents in the seaweeds during the study period. The results of the global mean contents and standard deviations (SD) of the HVMs As, Pb, Cd and Cr in the seaweed biomass (regardless to species and seasons) during the study period are displayed in Table 1. The seaweeds HVM contents showed the following pattern of distribution: As > Cd > Pb > Cr. The mean contents were: As (6.36±7.1 μg g^{-1} dry weight (DW), Cd (1.67±1.8 μg g^{-1} DW), Pb (1.08±4.3 μg g^{-1} DW), and Cr (073±1.07 μg g^{-1} DW). Standard deviations had high dispersion ranges, highlighting the important fluctuations of the species and seasonal contents during the study period.

Table 1

Summary of main statistics and two-way ANOVA HVM analyses results (95% confidence)

	As	*Pb*	*Cd*	*Cr*
Mean	6.362	1.08338	1.66818	0.7307
SD	7.09005	4.345266	1.848946	1.076946
Max	28.143	24.79750	6.89000	5.9350
Min	0.115	0	0.00975	0.0100

The Pearson correlation coefficient between the seaweeds HVM contents (Table 2) showed that As was slightly correlated with both Pb and Cr (R = 0.5), while Pb was highly and positively correlated with Cr (R = 0.9). This means that the seaweeds As contents changed slightly in the same way as Pb and Cr, and Pb and Cr contents changes occurred in the same way; Cd contents were not correlated with any of the other HVM (R < 0.3), showing a great independence of distribution from the rest of HVM.

Table 2

The Pearson correlation coefficient between seaweeds heavy metals contents

Metals	*As*	*Pb*	*Cd*	*Cr*
As	--	0.5419251*	0.2229715	0.5222139*
Pb		--	0.0216797	0.8873163*
Cd			--	0.04169148
Cr				--

Statistically significant correlations (r-value < -0.50 or > 0.50) are marked with asterisks (*).

The results of As, Pb, Cd and Cr mean contents distribution in 9 target seaweed species, their standard deviations, and patterns of distribution in species biomass during the study period are displayed in Figure 2. Species sampled one time (*C. acicularis* and *G. multipartita* are represented in Table 3. Except for *C. acicularis* sample, which showed very high contents for all the four HVMs (27.7, 24.8, 1.51 and 5.9 µg g^{-1} DW, respectively), the other species mean contents ranged from 2.45 to 10.28 µg g^{-1} DW, 0.09 to 5.34 µg g^{-1} DW, 0.48 to 2.9 µg g^{-1} DW, and 0.22 to 0.81 µg g^{-1} DW, respectively for As, Pb, Cd and Cr. The results showed significant contents in HVM in most of the studied species biomass, and at varying levels and depending on the species and on the kind of metal. HVM standard deviations values were very high for all species, showing a high variation.

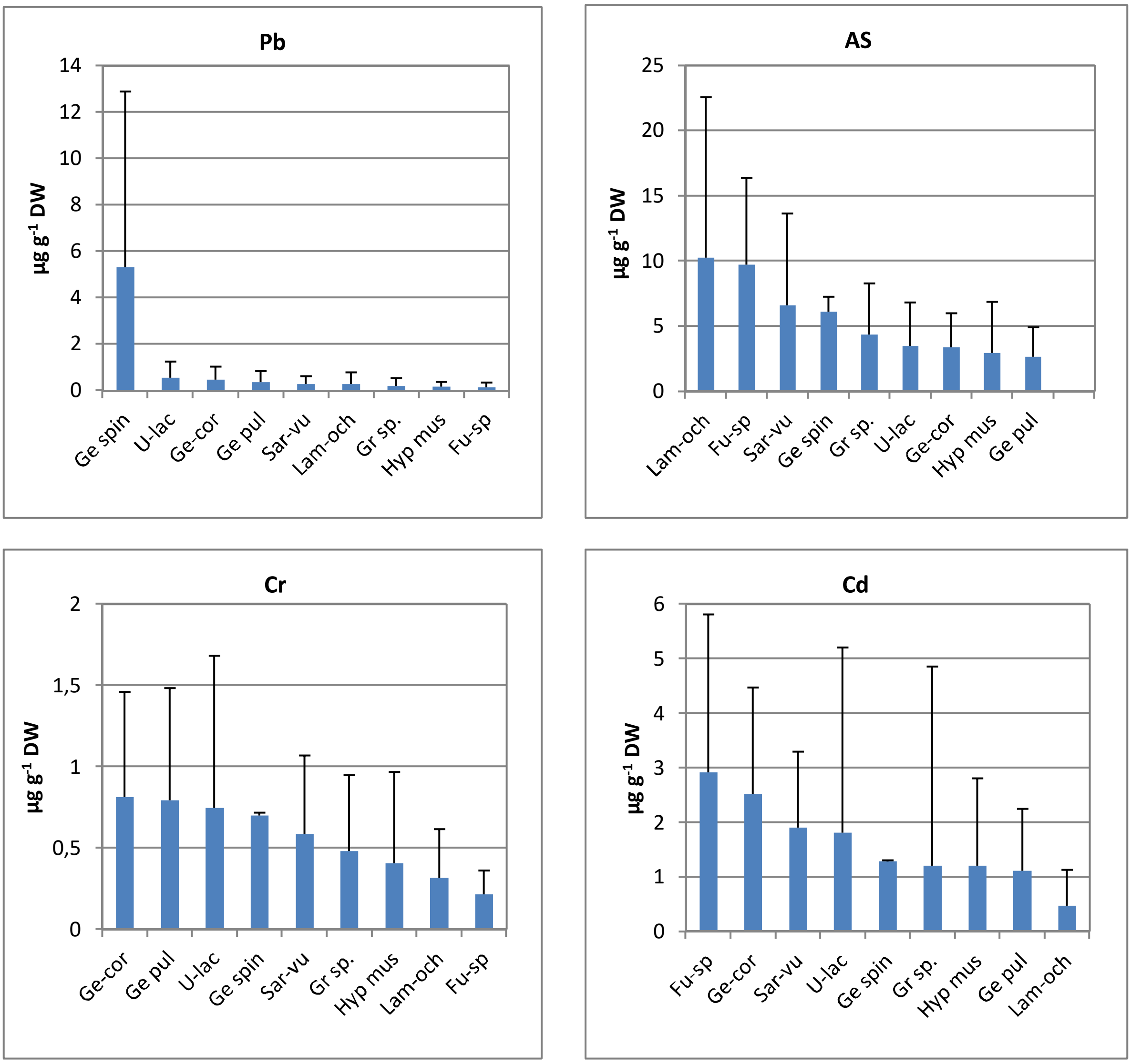

Figure 2. HVM mean contents (in µg.g^{-1} DW) (mean±standard deviations (SD) and patterns of distribution among seaweeds species in El Jadida region coastal zone during the study period.

Table 3

Heavy metals contents in seaweed species found and sampled one time during the study period (in µg g^{-1} DW of seaweed)

Species	*Season*	*As*	*Pb*	*Cd*	*Cr*
Gracilaria multipartita	Winter	2.45	0.09	1.33	0.70
Chondracanthus acicularis	Autum	27.68	24.80	15.08	5.94

Arsenic. Phaeophyceae species showed the highest mean contents in As, 10.3, 9.72, and 6.62 μg g^{-1} DW, respectively for *L. ochroleuca*, *F. spiralis* and *S. vulgare*. Rhodophyceae mean contents were in the range of 2.45 to 6.12 μg g^{-1} DW, and the Chlorophyceae species *U. lactuca* mean content was 3.52 μg g^{-1} DW.

Lead. Except for *G. spinulosum*, which showed a very high content in Pb (5.34 μg g^{-1} DW), all the remaining species had mean contents less than 0.6 μg g^{-1} DW.

Cadmium. Cadmium mean contents mostly ranged from 1.8 to 2.9 μg g^{-1} DW in *U. lactuca*, *S. vulgare, G. corneum* and *F. spiralis*, with the highest value recorded in *Fucus spiralis*, while they were more stable between 1.12 and 1.33 μg g^{-1} DW for the remaining species, with the lowest value noticed in *L. ochroleuca*.

Chromium. Chromium mean contents ranged from 0.6 to 0.8 μg g^{-1} DW in *Gelidium* species, *G. multipartita* and *U. lactuca* and were less than 0.5 μg g^{-1} DW for *Gracilaria* sp., *H. musciformis*, *L. ochroleuca* and *F. spiralis*.

As, Pb, Cd and Cr seaweed species mean contents distribution patterns during the study period. According to aforementioned results, the As, Pb, Cd and Cr contents in seaweed species mean contents distribution during the study period occurred according to following patterns, thus, highlighting the most bioaccumulative species for each of the target HVM:

- for As: *C. acicularis* > *L. ochroleuca* > *F. spiralis* > *S. vulgare* > *G. spinulosum* > *Gracilaria* sp. > *U. lactuca* > *G. corneum* > *H. musciformis* > *G. pulchelum* > *G. multipartita*;
- for Pb: *C. acicularis* > *G. spinulosum* > *U. lactuca* > *G. corneum* > *G. pulchelum* > *S. vulgare* > *L. ochroleuca* > *Gracilaria* sp. > *H. musciformis* > *F. spiralis* > *G. multipartita*;
- for Cd: *C. acicularis* > *F. spiralis* > *G. corneum* > *S. vulgare* > *U. lactuca* > *G. multipartita* > *G. spinulosum* > *Gracilaria* sp. > *H. musciformis* > *G. pulchelum* > *L. ochroleuca*;
- for Cr: *C. acicularis* > *G. corneum* > *G. pulchelum* > *U. lactuca* > *G. spinulosum* > *G. multipartita* > *S. vulgare* > *Gracilaria* sp. > *H. musciformis* > *L. ochroleuca* > *F. spiralis*.

***Seasonal changes in heavy metals contents in seaweed species*.** Results of HVM contents seasonal changes in seaweed species biomass are shown in Figure 3. For the sampled one time seaweeds species (*C. acicularis* in the autumn, and *G. multipartite* in winter), HVM values are shown in Table 3.

Arsenic. Arsenic contents in the seaweeds knew many changes depending of the species and seasons (Figure 3). They were in the range of 3.92 to 28.14 μg g^{-1} DW in the spring, 1.36 to 10.58 μg g^{-1} DW in the summer, 2.57 to 27.67 μg g^{-1} DW in the autumn, and went down to very low values (0.11 to 0.49 μg g^{-1} DW) for all species in winter. They were mostly higher than the toxicity threshold limit of 3 μg g^{-1} DW recommended by (AFSSA 2009) in most species (Figure 3, Table 3).

The highest values were recorded for the Phaeophyceae (*L. ochroleuca*, *S. vulgare*, and *F. spiralis*) in the spring (12 to 28.14 μg g^{-1} DW), with a pattern of contents distribution as*: L. ochroleuca* > *S. vulgare* > *F. spiralis*, and in the summer (7.2 to 10.6 μg g^{-1} DW), with a pattern of distribution as: *F. spiralis* > *L. ochroleuca* > S. *vulgare*, and in the autumn for *F. spiralis* (15.8 μg g^{-1} DW), while the contents of *L. ochroleuca* and *S. vulgare* in the autumn went down to 4.9 and 2.6 μg g^{-1} DW respectively (Figure 3).

For Rhodophyceae, *G. corneum* contents were in the range of 3.2 to 3.92 μg g^{-1} DW in the spring and summer and increased to 6.4 μg g^{-1} DW in the autumn*. G. pulchellum* contents were low for the three seasons, in the range of 3.6 to 4.3 μg g^{-1} DW, while *Gracilaria* sp. contents were higher in the spring and autumn, in the range of 5.22 to 7.7 μg g^{-1} DW (Figure 3). The autumn sample of the species *C. acicularis* recorded the highest value in As content (27.7 μg g^{-1} DW) (Table 3).

The Chlorophyceae *U. lactuca* contents decreased from 5.6 to 1.37 μg g^{-1} DW from spring to summer and increased again to 7 μg g^{-1} DW in the autumn.

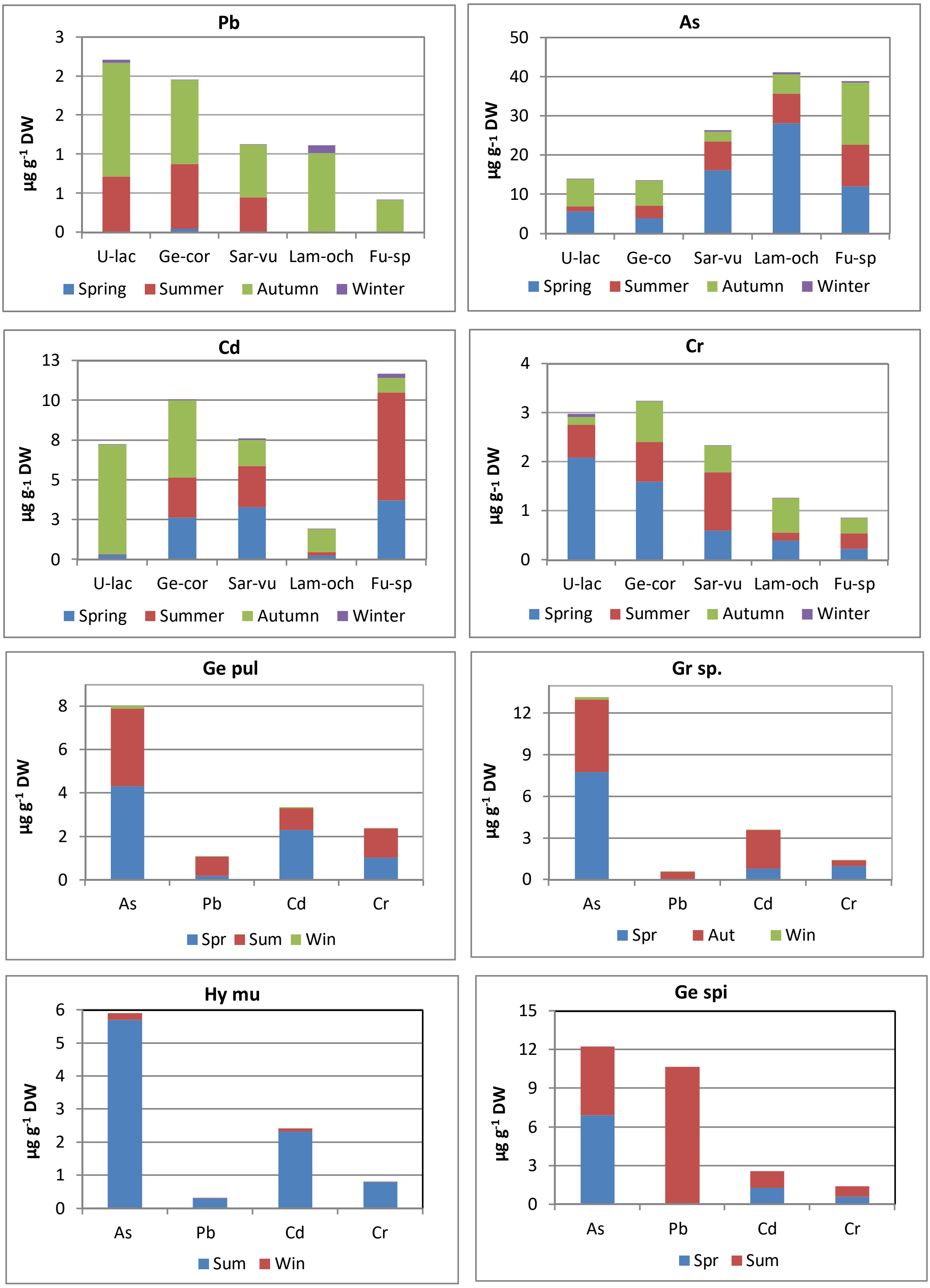

Figure 3. Seasonal changes in the seaweed species HVM contents (in µg g^{-1} DW) in the coastal zone of El Jadida region (Spr: spring, Sum: summer, Aut: autumn, Win: winter).

According to the ANOVA analysis results (Table 4), there is no significant effect of species on As contents distribution ($p > 0.05$), while there was a highly significant effect of seasons ($p < 0.005$). The Pheophyceae species and *C. acicularis* seemed to be the most bioaccumulating macroalgae of As. The spring, and autumn seasons seemed to be the most suitable for this metal bioaccumulation, while the winter seemed to be the season within which the seaweeds can eliminate this toxic metal, allowing the quality of their biomass to be conforme with the standards of safety regarding this element (< 2 µg g^{-1} DW).

Table 4

Summary of main statistics and two-way ANOVA HVM analyses results (95% confidence)

	As	*Pb*	*Cd*	*Cr*
		2 way ANOVA (p values)		
Species	0.052372	≅ 0*	0.8606	0.00000478*
Season	0.005072*	0.00001271*	0.1101	0.008408*

Note: *statically significant differences ($p < 0.05$).

Lead. Except the highest lead content which was detected in *C. acicularis* autumn sample (24.79 µg g^{-1} DW), followed by *G. spinulosum* (11 µg g^{-1} DW), the contents were under the threshold limit of toxicity of of 3 µg g^{-1} DW recommended by (European Commission 07/2008), and ranged from undetectable to 0.17 µg g^{-1} DW in spring, 0.02 to 1.067 µg g^{-1} DW in summer, 0.09 to 1.45 µg g^{-1} DW in autumn, and were the lowest (0.025 to 0.1 µg g^{-1} DW) in winter. In the spring, lead was undetectable in most species, except for *G. corneum* and *G. pulchellum* (0.05 and 0.18 µg g^{-1} DW respectively) (Figure 3, Table 3). In the summer, the highest contents values, ranging from 0.72 to 1.1 µg g^{-1} DW were recorded according to the pattern: *G. spinulosum* > *G. pulchellum* > *G. corneum* > *U. lactuca*. *S. vulgare* contents were in the range of 0.45 µg g^{-1} DW. Lead was undetectable in *F. spiralis* and *L. ochroleuca* biomass in the summer (Figure 3). In the autumn, the highest values (1.013 to 1.45 µg g^{-1} DW) were detected according to the following pattern: *U. lactuca* > *G. corneum* > *L. ochroleuca*. In the same season, the contents were lower (0.415 to 0.67 µg g^{-1} DW) for the following species: *S. vulgare* > *Gracilaria* sp. > *F. spiralis*. In the winter, except for *L. ochroleuca* (0.1 µg g^{-1} DW), the lead contents went down to 0.04 to 0.01 µg g^{-1} DW and according to the pattern: *U. lactuca* > *Gracilaria* sp. > *G. pulchellum*, and to 0.003 to 0.008 µg g^{-1} DW, according to the pattern: *F. spiralis* > *S. vulgare* > *G. corneum* (Figure 3). The ANOVA analysis results (Table 4) showed there was very high significant effect of species and seasons on lead distribution ($p < 0.005$). The summer and autumn were the seasons during which occurred a slight bioaccumulation, while it was non-significant during spring and winter.

Cadmium. Except winter season, most of the species had very high contents in cadmium in comparison with the toxicity limit value of 3 µg g^{-1} DW (AFSSA 2009) and ranged from 3.71 to 0.23 µg g^{-1} DW) in spring, 6.78 to 0.02 µg g^{-1} DW in summer, 6.89 to 0.91 µg g^{-1} DW in autumn, with a peak of 15.1 µg g^{-1} DW in the *C. acicularis* autumn sample, and 0.27 to 0.009 µg g^{-1} DW in winter. Spring and summer contents were low and ranged from 1 to 3.3 µg g^{-1} DW for *G. pulchellum*, *G. corneum*, and *S. vulgare*, while they rose up from 3.7 to 6.8 µg g^{-1} DW for *F. spiralis*, and remained very low for *U. lactuca*, *Gracilaria* sp. and *L. ochroleuca* (0.03 to 0.8 µg g^{-1} DW). The autumn cadmium contents in *U. lactuca* and *G. corneum* were higher (6.9 and 4.8 µg g^{-1} DW respectively), while the ones of *S. vulgare*, *G. multipartita, L. ochroleuca* and *F. spiralis* were lower and remained in the range of 0.9 to 1.7 µg g^{-1} DW. Winter contents went down to the range of 0.01 to 0.08 µg g^{-1} DW for most species, except for *F. spiralis* (0.3 µg g^{-1} DW) (Figure 3, Table 3). The ANOVA test didn't give any significant effect of species or seasons ($p > 0.05$). (Table 4).

Chromium. The contents of total chromium ranged from 0.22 to 2.08 µg g^{-1} DW in spring, 0.16 to 5.93 µg g^{-1} DW in summer, 0.01 to 5.9 µg g^{-1} DW in autumn, 0.16 to 1.33 µg g^{-1} DW in winter (Figure 3). In the spring, the contents were higher and ranged from 0.98 to 2.1 µg g^{-1} DW according to the pattern *U. lactuca* > *G. corneum* > *G. pulchellum* > *Gracilaia* sp. In the summer, *G. corneum* and *U. lactuca* contents went down to 0.8 µg g^{-1} DW to undetectable values, while *G. pulchellum* maintained a close content as in the spring, and *S. vulgare* content increased from 0.6 µg g^{-1} DW in the spring to 1.2 µg g^{-1} DW in the summer. The contents remained lower (0.16 to 0.4 µg g^{-1} DW) during the spring and summer for *L. ochroleuca* and *F. spiralis.* In the autumn, *C. acicularis* showed the highest content value of 5.9 μg g^{-1} DW, while the rest of species had contents in the range of 0.6 to 0.83 μg g^{-1} DW for *G. corneum* > *L. ochroleuca* > *S. vulgare*, and in the range of 0.16 to 0.42 μg g^{-1} DW for *Gracilaria* sp. > *F. spiralis* and *U. lactuca. U. lactuca* and *S. vulgare* are in decrease in contamination from spring to autumn, *G. corneum* and *F. spiralis* remained stable after the decrease recorded in summer, while *L. ochroleuca* contents increased from 0.16 to 0.7 μg g^{-1} DW from the summer to the autumn. The contents went down to values less than 0.07 μg g^{-1} DW in the winter for all winter sampled species (Figure 3, Table 3). The ANOVA analysis results (Table 4) showed there was very high significant effect of species and seasons on chromium bioaccumulation ($p < 0.005$). The spring and summer were the seasons during which occurred bioaccumulation, followed in a less degree by autumn season, while this bioaccumulation was non-significant during winter.

Discussion

Risks of seaweeds contamination with pesticides*.* The distribution of pesticides in the environment varies according to their chemical structure, physical properties, formulation type, application method, the climate and agricultural conditions (Topçu Sulak 2012). Pesticides are also applied directly to water to control unwanted algae and invertebrates (Kaya 2007). All these pollutants arrive to the marine environment and are a source of contamination of all the bodies of the ecosystem (water column, sediments, fauna and flora). Most of pesticides are used in agriculture. Lindane, also known as γ-HCH (hexachlorocyclohexane) or γ-BHC (benzene hexachloride), is widely used in agriculture as insecticide (ATSDR 2005), Aldrin is used in agriculture to treat seed and soil to control worms, beetles and termites, and is banned in most of the countries since 1990, it is a highly carcinogenic substance banned for 20 years due to its persistence and toxicity (Sundhar et al 2021). Endrin exists as endrin aldehyde or endrin ketone in the environment, mainly in bottom sediments of water bodies (HHS 1996), it is noncarcinogenic to humans; however, it affects the central nervous system (IARC 1987). The accumulation of HCH residues was found to be more related with the fruiting season of the seaweeds (Sundhar et al 2019). It is a potential occupational carcinogen with IDLH (immediately dangerous to life or health) value of 25 mg m-3 in humans (Baskin 1975). The use of endosulfan is restricted in agriculture due to its acute toxicity. Methoxychlor causes reproductive toxicity in humans but not classified as a carcinogen (Cummings 1997), but in others research it has no carcinogenic effect on humans but causes liver damage in animals (Kim & Lee 2017).

The results showed that all the seaweeds species had pesticides contents very low and less than the quantification limit (LQ) < 0.010 μg g^{-1} DW so, widely under the limit of toxicity for all seasons. So no risk of pesticides toxicity could be related to the consumption of the seaweeds of El Jadida region coastal zone.

Risks of contamination with heavy metals*.* Previous studies had demonstrated the high ability of seaweeds to accumulate HVMs in their tissue, so many researchers had focused in studying the use of these seaweeds ability to bind and accumulate metals (Vasquez & Guerra 1996). In this way, they can be considered as good indicators of micropollution in the marine environment (Weis et al 2004). Seasonal variations of HVMs in macroalgae have been noted by many authors (Rönnberg et al 1990; Catsiki et al 1991) while others have reported that there was no variation (Haug et al 1974; Shiber 1980).

In the present study, the results showed significant bioaccumulation of HVM in all species at varying levels, depending on the species and seasons, and also on the nature of the HVM. Comparison of seaweeds HVM contents with those naturally found in surface waters (seas and rivers) can confirm the bioaccumulation of HVMs by these seaweeds in natural environments (Wedepohl 1991). The distribution of HVM in the seaweeds is highly dependent of the one in seawater, and which is highly dependent of the anthropogenic inputs and of the biogeochemical cycle in seawater of these metals, which changes according to seasonal variations. This could be demonstrated in the case of the present study within the seasonal changes of arsenium, lead and chromium contents in the seaweed that occurred during the study period, with the highest values in spring and autumn and the lowest in the winter. The variation of upwelling activity that occurs in the study area is also a very important factor that could influence the seaweeds HVM distribution among seasons in the studied area.

***Arsenic**.* According to Neff (1997), arsenic occurs in the seawater and seabed sediment under mineral forms (arsenate (As V) more dominant in oxygenated productive marine ecosystems seawaters, and oxidized sediments primarily associated with iron oxyhydroxides, and arsenite (As III)), more toxic and potentially carcinogenic, representing 10 to 20% of total arsenic in seawater. In reducing marine sediments, arsenate is reduced to arsenite and is associated primarily with sulfide minerals (Neff 1997). Natural concentrations of arsenic in coastal marine and ocean waters are always much higher than the US EPA (1980) human health (fish consumption) water quality criterion for total arsenic in seawater (Neff 1997), the average concentration of total arsenic in the ocean is about 1.7 μg L^{-1}, about two orders of magnitude higher than the value of 0.0175 μg L^{-1} US Environmental Protection Agency's human health criterion for fish consumption (US EPA 1980). The concentration of total arsenic in clean coastal waters is 1 to 3 mg L^{-1}, with a mean of about 1.7 mg L^{-1} (Andreae 1979; Andreae & Andreae 1989; Li 1991), a concentration about 100 times higher than the US EPA human health water quality criterion (fish consumption) value (0.0175 mg L^{-1}) (Neff 1997). The uncontaminated marine sediments contain 5 to 40 μg g^{-1} DW total arsenic (Neff 1997). Marine algae accumulate arsenate from seawater, reduce it to arsenite, and then oxidize the arsenite to a large number of organoarsenic compounds (Neff 1997). The algae release arsenite, methylarsonic acid, and dimethylarsinic acid to seawater. For the most part study period (except in winter), the seaweeds had shown high arsenic contents, much higher than 3 mg kg^{-1} DW recommended by AFSSA (2009) and especially brown seaweeds, which are widely known to have a high capacity to accumulate arsenic from seawater, their bioaccumulation capacity was estimated to be hundred times higher than the one of land plants (Ito & Hori 1989). The lowest values of arsenic where found in *U. lactuca* and *G. corneum.* These results show that arsenic contents were highly variable between species, and the concentrations were higher in brown algae than in green and red ones (Phillips 1990). Nevertheless, this difference was not significant ($p > 0.05$), while it was obvious that season has a strong effect on arsenic distribution in seaweed tissues ($p < 0.025$). This could be likely related to concentration of dissolved arsenic in surface waters in the continental shelf, which varies seasonally due to natural cycling of arsenate between sediments and the overlying water column (Byrd 1988), which behavior resembles that of phosphate (Maher 1984), which is known to undergo seasonal remineralization and mobilization, with peak remobilization often in late summer (Hopkinson Jr. 1987). Comparison of seaweeds HVM contents with a previous study in the same area conducted by Caliceti et al (2002) showed rather a wider range in arsenic contents (0-360 μg g^{-1} DW, versus 0.12 to 27.7 μg g^{-1} DW in the present study). They found the highest values in the brown seaweed *Cystoseira barbata*, and higher values in *L. ochroleuca* (28.14 μg g^{-1} DW). For *U. lactuca*, *F. spiralis* and *Gracilaria* sp., the concentrations in the present study were lower than those reported in Caliceti et al (2002), Miao et al (2014) for *S. vulgare*, and de la Rocha et al (2009) for *L. ochroleuca*, and higher than those reported by Rezzoum et al (2016) for *F. spiralis* and *L. ochroleuca.* The high arsenic levels recorded in the seaweeds are likely related to the presence of a high level in arsenic in the seawater of the study region.

Lead. Lead is a non-essential and no biodegradable metal that occurs in the environment and mostly forming complex compounds with other metals as copper, selenium, and zinc (Botté et al 2022). In marine environment, lead comes from the terrestrial areas and could have many perturbation effects on invertebrate’s lifecycle (Botté et al 2022). Seaweeds lead contents in the present study were lower than the limit of toxicity of 5 μg g^{-1} DW (Mabeau & Fleurence 1993), 3 μg g^{-1} DW (European Commission (07/2008), and lower than recorded in the study of Caliceti et al (2002) for *U. lactuca*, *Gracilaria* sp., and

F. spiralis, where they reported higher lead mean concentrations values of 7.3±6.4 μg g^{-1} DW, 6.9±6.2 μg g^{-1} DW, and 1.6±0.6 μg g^{-1} DW respectively. Comparison with research works results in other world regions for *U. lactuca* showed high variation in the contents, which remained higher than the present study results, namely 0.67 to 5.77 mg kg^{-1} in Sicily (Bonanno et al 2020), 1.44 to 2.20 μg g^{-1} DW in Suez Gulf, Agaba Gulf and Suez Canal in Egypt (Mourad & Abd El-Azim 2019), 2.64±3.45 μg g^{-1} DW for Senegalese coast (Diop et al 2016), while in other regions the contents were lower than in the present study (0.55±0.99 μg g^{-1} DW, 0.02 mg kg^{-1} and 0.02 mg kg^{-1}, respectively in Tartous (Syria) (Al Masri et al 2003), Thermaikos and Crete (Greece) (Sawidis et al 2001).

Cadmium. The ocean cadmium concentration is less than 0.5 μg L^{-1}, in seawaters; cadmium occurs at different levels, higher in upwelling surface waters and continental shelf surface waters (50 to 70 μg L^{-1}), due to the release of cadmium from the bottom sediment of the seabed (Mart & Nürnberg 1986), and much higher in coastal areas impacted by anthropogenic pollution. Cadmium is a non essential and a highly toxic HVM. In the studied area, cadmium contents were much higher than the limit of toxicity value of 3 μg g^{-1} (European Commission 07/2008), in *U. lactuca* and *G. corneum*, in the autumn, and *F. spiralis* in the summer highlighting a very important contamination with this element at these periods. While these contents remained lower than the toxicity limit for the rest of seasons and for the rest of species. This variation could be explained by the capability of the three first species to accumulate cadmium that occurs in abundance in seawater (Kaimoussi 2002), concomitant with a higher activity of the upwelling activity that occurs in this zone, where the rise of deep waters rich in HVMs, including cadmium (Sidoumou et al 1992; Romeo et al 1993), make the surface waters very rich in trace elements. Cadmium levels in *U. lactuca* were higher than those reported by Bonanno et al (2020) in the coast of Sicily in Italy, Mourad & Abd El Azim (2019) in Suez Gulf, Aqaba Gulf and Suez Canal in Egypt, Valdes et al (2018) in Valparaiso in Chile, and lower than those reported by Al-Masri et al (2003) in Tartous in Syria (11.0 μg g^{-1} DW). Cadmium contents in *Gracilaria* sp. and *F. spiralis* were higher than those reported by Caliceti et al (2002). *Gelidium* spp. contents were higher than those found by Besada et al (2009). *L. ochroleuca* contents were lower than those found by Besada et al (2009).

Chromium. Chromium is considered an important micronutrient in animal and human nutrition (Skrzypczyk et al 2023), the reduced form Cr(III) is one of the essential elements and the oxidized form Cr(VI) is toxic and carcinogenic at high doses (Chiffoleau 1994). The biogeochemistry depends on the oxic level of the marine environment: in the fully dissolved phase in the form of Cr(III) in anoxic environments and in the Cr(VI) form in well-oxygenated ocean waters (Chiffoleau 1994). The concentrations of chromium in seaweeds of El Jadida had previously been determined by Caliceti et al (2002), Bonnano et al (2020) and Mourad & Abd El-Azim (2019). Many studies agree that *U. lactuca* has numerous features that make it one of the best bioindicators of metal pollution in marine environment (Areco et al 2021). The concentrations of chromium in *U. lactuca* were lower than those reported by Bonnano et al (2020) in coast of Sicily in Italy, Chakraborty et al (2014) in golf of Kutch in India, and in Tartous in Syria (Al Masri et al 2003), but higher than those determined by Mourad & Abd-El-Azim (2019) in Egypt and Diop et al (2016) in Senegalese coast. For *Gracilaria* sp. the concentrations are higher in El Jadida region in comparison with Caliceti et al (2002), but for *F. spiralis* the concentrations are lower than those reported by Caliceti et al (2002).

Permissible consumption limit of seaweeds from the coastal zone of El Jadida region. Seaweeds are considered as a high quality food for human health enhancement if consumed in a reasonable way. The evaluation of Monteiro et al (2019) on human health risk assessment of seaweed consumption was performed based on a daily serving of different species of seaweeds of 5 g of DW per body weight (bw) per day and showed that the exposure to lead and cadmium was respectively 0.0796 and 0.0387 μg/kg bw/day for *F. spiralis*, 0.0214 and 0.0568 μg/kg bw/day for the Laminariale *Saccharina latissima*, and 0.0065 and 0.0031 μg/kg bw/day for *U. lactuca*.

Carefully HVM consumption in very low quantities could prevent toxicity risks. Permissible comsumption limit of HVM (expressed in µg/kg body weight (bw) per day) was established by the EFSA (2009) as: Cd ≤ 0.36 µg/kg bw/day, Pb ≤ 0.5 µg/kg bw/day, and As ≤ 0.3 µg/kg bw/day. Cd limit was lowered to 0.35 µg/kg bw/day by ANSSES (2020). The PCL of Cr was estimated to be equal or less than 0.3 µg/kg bw/day (US EPA 1980). Nowaday, no PCR was established for Cr, which is considered as a valuable micronutrient. A seaweed daily consumption rate of 5 to 10 g DW per person (of bw of 85 kg) could be safe and sufficient to supply the body needs in essential micronutrients, including Cr (Skrzypczyk et al 2023). ANSES (2020) lowered Cd PCL to 0.35 µg/kg bw/day following investigations on the impact of Cd PCL of 0.36 µg/kg bw/day on human health.

Herein, an estimation of the permissible consumption limit (SW-PCL) of the seaweeds from El Jadida region coastlinet was calculated using the note below Table 5, and relying on the aforementioned HVM – PCL (EFSA 2010). For cadmium, the value of 0.35 µg/kg bw/day (ANSSES (2020) was used. Until now, there is no defined and reliable threshold of chromium consumption by health authorities. So, SW-PCR was not calculated for it. Calculations were based on the highest value of HVM contents within the study period to avoid any health risk. The mean body weight used was 75 kg.

Table 5

HVM contents in seaweeds species and seaweed permissible consumption limit (SW-PCL)

Species	*HVM contents ($\mu g\ g^{-1}$ DW)*	*SW-PLC (g DW/kg bw/day)*	*SW-PCL (DW individual $^{-1}$ day^{-1})*
		Arsenic	
Ul-la	6.95	0.050	3.8
Gr-sp	7.78	0.045	3.4
Ge-co	6.35	0.055	4.1
Ge-pu	4.33	0.081	6.1
Ge-spi	6.91	0.051	3.8
Sa-vu	16.21	0.022	1.6
La-oc	28.14	0.012	0.9
Fu-spi	15.84	0.022	1.7
Gr-mu	2.44	0.143	10.8
Ch-ac	27.67	0.013	0.9
Hy-mu	5.70	0.061	4.6
		Lead	
Ul-la	1.14	0.439	32.9
Gr-sp	0.56	0.893	67.0
Ge-co	1.06	0.472	35.4
Ge-pu	0.90	0.556	41.7
Ge-spi	1.06	0.472	35.4
Sa-vu	0.45	1.111	83.3
La-oc	1.01	0.495	37.1
Fu-spi	0.41	1.220	91.5
Gr-mu	0.09	5.556	416.7
Ch-ac	24.79	0.020	1.5
Hy-mu	0.31	1.613	121.0
		Cadmium	
Ul-la	6.69	0.045	3.4
Gr-sp	2.80	0.107	8.0
Ge-co	4.83	0.062	4.7
Ge-pu	2.29	0.131	9.8
Ge-spi	1.29	0.233	17.4
Sa-vu	3.29	0.091	6.8
La-oc	1.43	0.210	15.7
Fu-spi	6.78	0.044	3.3
Gr-mu	1.33	0.226	16.9
Ch-ac	1.50	0.200	15.0
Hy-mu	2.33	0.129	9.7

HVM PCL used for calculation is as follows: Cd ≤ 0.35 µg/kg bw/day, Pb ≤ 0.5 µg/kg bw/day, and As ≤ 0.3 µg/kg bw/day, bw: individual body weight: 75 kg.

The limitations of seaweeds consumption is essentially due to cadmium and arsenic high levels, while lead contents remain almost very low during the whole period. The PCL of seaweeds due to arsenic contents changes mainly from 3.4 to 3.8 g DW/individual/day for most species, except for the three Pheophyceae and *C. acicularis* (< 1.8 g DW/individual/day).

The PCL of seaweeds due to cadmium contents is higher than 8 g DW/individu/day, while it remains in the range of 3 to 4.7 g DW/individual/day for *U. lactuca*, *G. corneum*, and 6.8 g DW/individual/day for *S. vulgare*.

The PCL of seaweeds due to cadmium contents is very high for most species (> 33 g DW/individual/day), except for *C. acicularis* (1.5 g DW/individual/day), while it stays in the range of 3 to 4.7 g DW/individual/day for *U. lactuca*, and *G. corneum*, and 6.8 g DW/individual/day for *S. vulgare*.

The most relevant seaweed PCL should be the lowest value of the three calculated PCL. For example, according to Table 5, the seaweed PCL should be 10.8 g DW/individual/day for *G. multipartita*, 3.4, 3.4, 3.8, 4.1, 4.6, and 6.1 g DW/individual/day, respectively for *U. lactuca*, *Gracilaria* sp., *G. spinulosum*, *G. corneum*, *H. musciformis*, and *G. pulchellum*, and very low, less than 1.8 g DW/individual/day for the three Pheophyceae and *C. acicularis*. These values remain lower than the seaweeds PCL used worldwide in Asia (8.5 g/day in South Korea), 5.2 in China (Chen et al 2018), and 1.4 in Japan (Murai et al 2021).

Conclusions. Heavy metals in the seaweeds of the El Jadida region coastal zone have been found in a wide range of concentrations, mostly higher than the toxicity threshold limit values, and showed a great difference between species and seasons. Pesticides remained under the detectable limit during the whole period of the study. Thus, it was possible to conclude that there was no risk of contamination of seaweeds with pesticides, while the risk was more important due to heavy metals, especially cadmium and arsenic. The presence of heavy metals in the seaweed of the coast of El Jadida is still relatively lower than that found in other regions of the world.

Although the present study suggested that human exposure to arsenic, cadmium, chromium and lead through seaweed consumption was low if the permissible consumption limit is respected, toxic metals should be surveyed in the seaweeds and seawater using analysis methods of the highest performances and precision, and species with high bioaccumulation potential be used as bioindicators of toxicity.

Conflict of interest. The authors declare that there is no conflict of interest.

References

AFSSA, 2009 Avis relatif à la teneur maximale en arsenic inorganique recommandée pour les algues laminaires et aux modalités de consommation de ces algues compte tenu de leur teneur élevée en iode. SA. 2007-0007. [in French]

Agarwal S., Albeshr M. F., Mahboob S., Atique U., Pramanick P., Mitra A., 2022 Bioaccumulation factor (BAF) of heavy metals in green seaweed to assess the phytoremediation potential. Journal of King Saud University – Science 34:102078.

Al-Homaidan A. A., Al-Ghanayem A. A., Al-Qahtani H. S., Al-Abbad A. F., Alabdullatif J. A., Alwakeel S. S., Ameen F., 2021 Effect of sampling time on the heavy metal concentrations of brown algae: a bioindicator study on the Arabian Gulf coast. Chemosphere 263:127998.

Al-Masri M. S., Mamish S., Budier Y., 2003 Radionuclides and trace metals in eastern Mediterranean Sea algae. Journal of Environmental Radioactivity 67(2):157-168.

Andreae M. O., 1979 Arsenic speciation in seawater and interstitial waters: the influence of biological-chemical interactions on the chemistry of a trace element. Limnology and Oceanography 24(3):440-452.

Andreae M. O., Andreae T. W., 1989 Dissolved arsenic species in the Schelde estuary and watershed, Belgium. Estuarine, Coastal and Shelf Science 29(5):421-433.

ANSES, 2020 Opinion of the French Agency for Food, Environmental and Occupational Health & Safety on "maximum cadmium levels for seaweed intended for human consumption". ANSES Opinion Request, 57 pp.

Areco M. M., Salomone V. N., dos Santos Afonso M., 2021 *Ulva lactuca*: a bioindicator for anthropogenic contamination and its environmental remediation capacity. Marine Environmental Research 171:105468.

ATSDR, 2005 Toxicological profile for hexachlorocyclohexane. U.S. Department of Health and Human Services, Agency for Toxic Substances and Disease Registry. Public Health Service, Atlanta, GA, pp. 263-398.

Baskin A. D. (ed), 1975 Handling guide for potentially hazardous materials. Material Management and Safety, Inc., Niles, Illinois.

Besada V., Andrade J. M., Schultze F., González J. J., 2009 Heavy metals in edible seaweeds commercialised for human consumption. Journal of Marine Systems 75(1-2):305-313.

Bonanno G., Veneziano V., Piccione V., 2020 The alga *Ulva lactuca* (Ulvaceae, Chlorophyta) as a bioindicator of trace element contamination along the coast of Sicily, Italy. The Science of the Total Environment 699:134329.

Botté A., Seguin C., Nahrgang J., Zaidi M., Guery J., Leignel V., 2022 Lead in the marine environment: concentrations and effects on invertebrates. Ecotoxicology 31(2):194-207.

Byrd J. T., 1988 The seasonal cycle of arsenic in estuarine and nearshore waters of the South Atlantic Bight. Marine Chemistry 25(4):383-394.

Caliceti M., Argese E., Sfriso A., Pavoni B., 2002 Heavy metal contamination in the seaweeds of the Venice lagoon. Chemosphere 47(4):443-454.

Catsiki V. A., Papathanassiou E., Bei F., 1991 Heavy metal levels in characteristic benthic flora and fauna in the central Aegean Sea. Marine Pollution Bulletin 22(11):566-569.

Chakraborty S., Bhattacharya T., Singh G., Maity J. P., 2014 Benthic macroalgae as biological indicators of heavy metal pollution in the marine environments: a biomonitoring approach for pollution assessment. Ecotoxicology and Environmental Safety 100:61-68.

Chen L., Liu R., He X., Pei S., Li D., 2021 Effects of brown seaweed polyphenols, a class of phlorotannins, on metabolic disorders via regulation of fat function. Food and Function 12(6):2378-2388.

Chen Q., Pan X. D., Huang B. F. Han J. L., 2018 Distribution of metals and metalloids in dried seaweeds and health risk to population in southeastern China. Scientific Reports 8(1):3578.

Chiffoleau J. F., 1994 [Chromium in marine environment]. In: Repères océans (1240-1153), No. 8, Ifremer, 49 pp. [in French]

Commission Regulation (EC) 629/2008 amending Regulation (EC) 1881/2006 setting maximum levels for certain contaminants in foodstuffs. Official Journal of the European Union L173:6-9.

Cummings A. M., 1997 Methoxychlor as a model for environmental estrogens. Critical Reviews in Toxicology 27(4):367-379.

de la Rocha S. R., Sanchez-Muniz F. J., Gomez-Juaristi M., Larrea Marin M. T., 2009 Trace elements determination in edible seaweeds by an optimized and validated ICP-MS method. Journal of Food Composition and Analysis 22(4):330-336.

Diop M., Howsam M., Diop C., Goossens J. F., Diouf A., Amara R., 2016 Assessment of trace element contamination and bioaccumulation in algae (*Ulva lactuca*), mussels (*Perna perna*), shrimp (*Penaeus kerathurus*), and fish (*Mugil cephalus*, *Saratherondon melanotheron*) along the Senegalese coast. Marine Pollution Bulletin 103:339-343.

EFSA (European Food Safety Authority), 2009 Scientific opinion of the panel on contaminants in food chain. EFSA Journal 980:1-139.

EFSA (European Food Safety Authority), 2010 Scientific opinion on lead in food. EFSA Journal 8(4):1570.

FAO, 2022 The state of world fisheries and aquaculture 2022. Towards blue transformation. FAO, Rome, Italy, 266 pp.

Gomez-Ordonez E., 2012 Evaluacion nutricional y propiedades biologicas de algas marinas comestibles. Estudios *in vitro* e *in vivo*. Thesis doctoral, Universidad Complutense de Madrid, 218 pp. [in Spanish]

Guo J. Y., Zeng E. Y., Wu F. C., Meng X. Z., Mai X. B., Luo X. J., 2007 Organochlorine pesticides in seafood products from southern China and health risk assessment. Environmental Toxicology and Chemistry 26(6):1109-1115.

Haug A., Melsom S., Omang S., 1974 Estimation of heavy metal pollution in two Norwegian fjord areas by analysis of the brown algae *Ascophyllum nodosum*. Environmental Pollution 7(3):179-192.

HHS, 1996 Annual update of the poverty guidelines. A notice by the Health and Human Services Department. Federal Register 61(43):8286-8288.

Hopkinson Jr. C. S., 1987 Nutrient regeneration in shallow-water sediments of the estuarine plume region of the nearshore Georgia Bight, U.S.A. Marine Biology 94:127-142.

IARC, 1987 Silica and some silicates. IARC monographs on the evaluation of carcinogenic risk of chemicals to humans. Volume 42. International Agency for Research on Cancer, Lyon, France, 121 pp.

Ito K., Hori K., 1989 Seaweed: chemical composition and potential food uses. Food Reviews International 5(1):101-144.

Jimenez-Escrig A., Gomez-Ordonez E., Ruperez P., 2012 Brown and red seaweeds as potential sources of antioxidant nutraceuticals. Journal of Applied Phycology 24: 1123-1132.

Kaimoussi A., 2002 Etat de la pollution métallique au niveau du littoral de la région d’El Jadida: etude comparative entre l’année 94/95 et l’année 98/99. Thèse d’Etat, Université Chouaib-Doukkali, Faculté des Sciences, El Jadida, Maroc, 175 pp.

Kamala-Kannan S., Batvari B. P. D., Lee K. J., Kannan N., Krishnamoorthy R., Shanthi K., Jayaprakash M., 2007 Assessment of heavy metals (Cd, Cr and Pb) in water, sediment and seaweed (*Ulva lactuca*) in the Pulicat Lake, South East India. Chemosphere 71(7):1233-1240.

Kaya H., 2007 Atikhisar Barajı ve Sarıçay’da pestisit ve evsel kirliliğin araştırılması. M.A. Thesis, Department of Seafood, Çanakkale Onsekiz Mart University, 106 pp. [in Turkish]

Khan N., Ryu K. Y., Choi J. Y., Nho E. Y., Habte G., Choi H., Kim M. H., Park K. S., Kim K. S., 2015 Determination of toxic heavy metals and speciation of arsenic in seaweeds from South Korea. Food Chemistry 169:464-470.

Kim H. S., Lee B. M., 2017 Mutagenicity and carcinogenicity: human reproductive cancer and risk factors. In: Reproductive and developmental toxicology. 2nd edition. Gupta R. C. (ed), Academic Press, Cambridge, Massachusetts, USA, pp. 1123-1138.

Kumar M. S., Sharma S. A., 2021 Toxicological effects of marine seaweeds: a cautious insight for human consumption. Critical Reviews in Food Science and Nutrition 61(3):500-521.

Li Y. H., 1991 Distribution patterns of the elements in the ocean: a synthesis. Geochimica et Cosmochimica Acta 55(11):3223-3240.

Lozano G., Hardisson A., Gutierrez A. J., Lafuente M. A., 2003 Lead and cadmium levels in coastal benthic algae (seaweeds) of Tenerife, Canary Islands. Environment International 28(7):627-631.

Løvdal T., Skipnes D., 2022 Assessment of food quality and safety of cultivated macroalgae. Foods 11(1):83.

Lü F., Dind G., Liu W., Zhan D., Wu H., Guo W., 2018 Comparative study of responses in the brown algae *Sargassum thunbergii* to zinc and cadmium stress. Journal of Oceanology and Limnology 36:933-941.

Mabeau S., Fleurence J., 1993 Seaweed in food products: biochemical and nutritional aspects. Trends in Food Science and Technology 4(4):103-107.

Maher W. A., 1984 Mode of occurrence and speciation of arsenic in some pelagic and estuarine sediments. Chemical Geology 47(3-4):333-345.

Marinho-Soriano E., Fonseca P. C., Carneiro M. A. A., Moreira W. S. C., 2006 Seasonal variation in the chemical composition of two tropical seaweeds. Bioresource Technology 97(18):2402-2406.

Mart L., Nürnberg H. W., 1986 The distribution of cadmium in the sea. In: Cadmium in the environment. Mislin H., Ravera O. (eds). Experientia Supplementum, vol. 50, Birkhäuser Basel, pp. 28-40.

Miao L., Yan W., Zhong L., Xu W., 2014 Effect of heavy metals (Cu, Pb, and As) on the ultrastructure of *Sargassum pallidum* in Daya Bay, China. Environmental Monitoring and Assessment 186(1):87-95.

Mitra A., Zaman S., 2021 Estuarine acidification: exploring the situation of mangrove dominated Indian Sundarban estuaries. Springer Cham, 402 pp.

Monteiro M. S., Sloth J., Holdt S., Hansen M., 2019 Analysis and risk assessment of seaweed. EFSA Journal 17(2):e170915.

Mourad F. A., Abd El-Azim H., 2019 Use of green alga *Ulva lactuca* (L.) as an indicator to heavy metal pollution at intertidal waters in Suez Gulf, Aqaba Gulf and Suez Canal, Egypt. Egyptian Journal of Aquatic Biology and Fisheries 23(4):437-449.

Murai U., Yamagishi K., Kishida R., Iso H., 2021 Impact of seaweed intake on health. European Journal of Clinical Nutrition 75(6):877-889.

Neff J. M., 1997 Ecotoxicology of arsenic in the marine environment. Environmental Toxicology and Chemistry 16(5):917-927.

Nisizawa K., Noda H., Kikuchi R., Watanabe T., 1987 The main seaweed foods in Japan. Hydrobiologia 151:5-29.

Pekey H., 2006 The distribution and sources of heavy metals in Izmit Bay surface sediments affected by a polluted stream. Marine Pollution Bulletin 52(10):1197-1208.

Peñalver R., Lorenzo J. M., Ros G., Amarowicz R., Pateiro M., Nieto G., 2020 Seaweeds as a functional ingredient for a healthy diet. Marine Drugs 18(6):301.

Phillips D. J. H., 1990 Arsenic in aquatic organisms: a review, emphasizing chemical speciation. Aquatic Toxicology 16(3):151-186.

Rajfur M., Klos A., Waclawek M., 2010 Sorption properties of algae *Spirogyra* sp. and their use for determination of heavy metal ions concentrations in surface water. Bioelectrochemistry 80(1):81-86.

Regulation of the European Parliament and of the Council, 2005 Maximum residue levels of pesticides in or on food and feed of plant and animal origin and amending Council Directive 91/414/EEC with EPA Relevance. 1766 pp.

Rezzoum N., Mouradi A., Givernaud T., Merzouk N., Bennasser L., 2016 Variations saisonnières des concentrations en quelques oligo-éléments et métaux lourds chez *Fucus spiralis* (Linnaeus, 1753) et *Laminaria ochroleuca* (Bachelot de la Pylaie, 1824) du littoral de la ville d'El Jadida, Maroc. Afrique Science 12(3):293-305.

Riekie G. J., Williams P. N., Raab A., Meharg A. A., 2006 The potential for kelp manufacture to lead to arsenic pollution of remote Scottish islands. Chemosphere 65(2):332-342.

Riget F., Johansen P., Asmund G., 1997 Baseline levels and natural variability of elements in three seaweed species from West Greenland. Marine Pollution Bulletin 34(3):171-176.

Rodrigues E. T., Alpendurada M. F., Ramos F., Pardal M. A., 2018 Environmental and human health risk indicators for agricultural pesticides in estuaries. Ecotoxicology and Environmental Safety 150:224-231.

Romeo M., Gnassia-Barelli M., Sidoumou Z., Nguyen P., 1993 Variations spatio-temporelles des concentrations en métaux traces chez le mollusque *Donax rugosus* de la côte Mauritanienne. Bulletin d'Ecologie 24(2-4):83-86.

Rönnberg O., Adjers K., Ruokolahti C., Bondestam M., 1990 *Fucus vesiculosus* as an indicator of heavy metal availability in a fish farm recipient in the Northern Baltic Sea. Marine Pollution Bulletin 21(8):388-392.

Sanchez-Rodriguez I., Huerta-Diaz M. A., Choumiline E., Holguin-Quinones O., Zertuche-Gonzalez J. A., 2001 Elemental concentrations in different species of seaweeds from Loreto Bay, Baja California Sur, Mexico: implications for the geochemical control of metals in algal tissue. Environmental Pollution 114(2):145-160.

Sawidis T., Brown M. T., Zachariadis G., Sratis I., 2001 Trace metal concentrations in marine macroalgae from different biotopes in the Aegean Sea. Environment International 27(1):43-47.

Shiber J. G., 1980 Trace metals with seasonal considerations in coastal algae and molluscs from Beirut, Lebanon. Hydrobiologia 69:147-162.

Sidoumou Z., Roméo M., Gnassia-Barelli M., Nguyen P., Caruba R., 1992 Détermination de la qualité des eaux du littoral Mauritanien par la mesure des métaux traces chez les mollusques *Donax rugosus* et *Venus verrucosa*. Hydroécologie Appliquée 4(2): 33-41.

Skrzypczyk V. M., Callahan D. L., Francis D. S., Bellgrove A., 2023 Australian brown seaweeds as a source of essential dietary minerals. Journal of Applied Phycology https://doi.org/10.1007/s10811-023-03074-0.

Sundhar S., Shakila R. J., Jeyasekaran G., Shalini R., Arisekar U., Jenishma J. S., 2019 Safety assessment of edible red seaweeds *Gracilaria* and *Gelidiella* of Gulf of Mannar in terms of OCP residual contamination. Environmental Nanotechnology, Monitoring and Management 12:100245.

Sundhar S., Jeyashakila R., Jeyasekaran G., Aanand S., Shalini R., 2021 Assessment of organochlorine pesticides in different seaweed species of Thoothukudi coast, Tamil Nadu, South India. Indian Journal of Fisheries 68(2):148-153.

Topçu Sulak M., 2012 Pestisitlerin taşınım süreçleri ve çevresel etkileri. I. Tarım Sağlığı ve Güvenliği Sempozyumu. 6-7 April 2012, Şanlıurfa. [in Turkish]

U.S. Environmental Protection Agency, 1980 Ambient water quality criteria for toxaphene. Office of Water Regulations and Standards. Criteria & Standards Division, Washington, DC, 119 pp.

Valdes F. A., Lobos M. G., Díaz P., Saez C. A., 2018 Metal assessment and cellular accumulation dynamics in the green macroalga *Ulva lactuca*. Journal of Applied Phycology 30:663-671.

Vasconcelos M. T. S. D., Leal M. F. C., 2001 Seasonal variability in the kinetics of Cu, Pb, Cd and Hg accumulation by macroalgae. Marine Chemistry 74(1):65-85.

Vasquez J. A., Guerra N., 1996 The use of seaweeds as bioindicators of natural and anthropogenic contaminants in northen Chile*.* Hydrobiologia 326:327-333.

Wallenstein F. M., Couto R. P., Amaral A. S., Wilkinson M., Neto A. I., Rodríguez A. S., 2009 Baseline metal concentrations in marine algae from Sao Miguel (Azores) under different ecological conditions - urban proximity and shallow water hydrothermal activity. Marine Pollution Bulletin 58(3):438-443.

Wedepohl K. H., 1991 The composition of the upper Earth's crust and the natural cycles of selected metals. In: Metals and their compounds in the environment. Merian E. (ed), Verlag Chemie, Weinheim, pp. 3-17.

Weis J. S., Skurnick J., Weis P., 2004 Studies of a contaminated brackish marsh in the Hackensack Meadowlands of North eastern New Jersey: benthic communities and metal contamination. Marine Pollution Bulletin 49(11-12):1025-1035.

Wells M. L., Potin P., Craigie J. S., Raven J. A., Merchant S. S., Helliwell K. E., Smith A. G., Camire M. E., Brawley S. H., 2017 Algae as nutritional and functional food sources: revisiting our understanding. Journal of Applied Phycology 29(2):949-982.

Zbikowski R., Szefer P., Latala A., 2006 Distribution and relationships between selected chemical elements in green alga *Enteromorpha* sp. from the southern Baltic. Environmental Pollution 143(3):435-448.

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