Essential and toxic elements in seaweeds for human consumption

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Abstract

Essential elements (K, Ca, P, S, Cl, Mn, Fe, Cu, Zn, Ni, Br, I) and nonessential or toxic elements (Al, Ti, Si, Rb, Sr, As, Cd, Sn, and Pb) were determined by Energy Dispersive Polarized X-Rays Fluorescence Spectrometry in 14 seaweeds purchased in local specialty stores in Italy and consumed by humans. The differences in elements between the algae species reached up to 2-4 orders of magnitude. *Lithothamnium calcareum* showed the highest levels of Ca, Al, Si, Fe and Ti. *Palmaria palmata* showed the highest concentrations of K, Rb and Cl. The highest content of S was in *Chondrus crispus*. *Laminaria digitata* contained the highest concentrations of total As, Cd, Sn, Br and I. The highest concentration of Zn was in *Chlorella Pyrenoidosa*. *Ulva lactuca* displayed the highest levels of Cu, Ni, Mn and Pb. Iodine levels ranged from 3.4 in *Chlorella Pyrenoidosa* to 7316 mg/kg_{dry} in *Laminaria digitata*. The nutrimental importance of essential elements was assessed using nutritional requirements. The results showed that the consumption of algae might serve as an important source of the essential elements. Health risk due to the toxic elements present in seaweed was estimated using risk estimators. Total As, Cd and Pb concentrations ranged from <1 to 67.6, to 7.2 and to 6.7 mg/kg$_{dry}$ respectively; therefore, their contribution to total elemental intake does not appear to pose any threat to the consumers, but the concentrations of these elements should be controlled to protect the consumer against potential adverse health risks.

Keywords: Nutraceutical, seaweeds, essential elements, toxic elements, EDPXRF

INTRODUCTION

Marine algae or seaweeds are well known seafood most popular in Asian countries, particularly in Japan. In Europe and in many occidental countries, the use of seaweeds for cosmetic and medical purposes has recently increased, due to the growing success of natural remedies (Darcy-Vrillon, 1993; Dawczynski et al., 2007; Rodenas de la Rocha et al., 2009; Smith et al., 2010). Further many cosmetic and pharmaceutical products contain seaweed polysaccharides namely agars and alginates etc. (Khan et al., 2015). Moreover as seaweeds are richer in proteins, amino acids and mineral elements than other usual edible plants, these are recommended as food supplements to increase intake of trace elements essential for human functions (Burtin., 2003; Rodenas de la Rocha et al., 2009). However, it should be noted that the trace elements in a suitable range level are beneficial, but that excess quantities result in detrimental effects (Hsu et al., 2001; Aggett, 2010; Stern, 2010). Moreover, seaweeds are also an important source of exposure to non essential or toxic elements. The contamination of the ecosystem with elements such as arsenic (As), cadmium (Cd), mercury (Hg), lead (Pb), and antimony (Sb), which are ranked among hazardous substances of high priority (ATSDR, 2005), is a serious problem, threatening the habitat and the health of wild animals and humans (Nacano et al, 2014; De Almeida Lopes et al., 2015). These elements are natural components of the Earth's crust and biological systems. Their concentrations increased in the ecosystem during the last decades due to industrial and other anthropogenic activities (Meli et al., 2015; Roselli et al., 2015). These activities enhanced the release of chemical effluents into the environment, resulting in damage to aquatic organisms.

The capacity of seaweeds to accumulate metals depends on the bioavailability of metals in the surrounding water which is dependent upon environmental parameters of the sampling sites as salinity, pH, light and nutrients as well as on uptake capacity of algae which relies upon structural differences among the algae. Uptake takes place in two ways: 1) by a surface reaction through electrostatic attraction to negative sites (process independent of factors influencing metabolism); 2) a transport of metals across the cell membrane into the cytoplasm. This latter type of uptake is more dependent upon metabolic processes and it is subject to variations of temperature, light and age of the plant (Sanchez- Rodriguez et al., 2001).

The impact of toxic elements like Pb, Cd and As on human health is of great interest as these elements may exert adverse effects even at low levels when ingested over a long period of time. Cd exerts an adverse effect on immune system, kidney, liver, lungs, reproductive organs, and bones (Ginsberg, 2012; Huang et al., 2009). Lead intoxication produces damage to the nervous and immune systems and may also result in dysfunction of the renal tubules, liver, and the cardiovascular system (Tsuchiva et al., 1986; Garcia-Leston et al., 2012; Counter et al., 2009). Children are particularly at risk from Pb ingestion both before and after birth (Buchanan et al., 2011; Counter at al., 2009). Arsenic is a problematic element for humans producing carcinogenesis of liver, kidney, bladder and skin (Tsai et al., 1998; Bernstam and Nriagu, 2000). It is well known that the speciation of As plays an important role in determining As-induced toxicity to humans (Zavala et al., 2008).

The EU Regulation for heavy metals in foodstuffs (EC, 2011) does not consider algae except for Pb. Efforts are currently being made by a Committee of Experts within the European Commission to decide whether general advise or a special regulation should be issued (Besada et al., 2009). France was the first European country to set up regulations on the use of seaweeds for human consumption as non traditional foods.

Although surveys to determine the levels of essential and non essential elements in seaweeds have been conducted in many countries especially in Asian countries (Hou and Yan, 1998; Hsu et al., 2001; Dawczynski et al., 2007; Khan et al., 2015) and taking into account that seaweeds are more and more important in modern Western diet (Phaneuf et al., 1999; Almela et al., 2002), it is important to know the concentration of essential and toxic elements in edible seaweeds and subsequently their potential effects on population health.

In the present study, 14 algae products purchased in local specialty stores in Italy and consumed by humans are taken into account. No data are available about the consumption of edible seaweeds in Italy, but this country is no different from other Western countries that show an increase in the algae consumption in recent decades. The purposes of this work were to provide information on the concentration of essential and non essential or toxic elements in these algae products, to compare their metal content with the tolerable intake level. In addition, by analysing the inorganic elemental content, the properties of algae food could also be characterized. The elements taken into account are 21 subdivided in essential, major (K, Ca, P, S, Cl) and trace (Mn, Fe, Cu, Zn, Ni, Br, I,) and non essential or toxic (Al, Ti, Si, Rb, Sr, As, Cd, Sn, and Pb).

MATERIALS AND METHODS

Samples and sample pretreatment

Analyses were carried out on 14 edible seaweeds samples purchased in local specialty stores in Italy. Seaweeds belonged to 4 groups of algae (Table 1): red (5 samples), brown (5 samples), green (3 samples) and blue-green (1 sample). Seaweeds, in Italy, are rarely sold fresh as all seaweeds analysed are purchased in a dried form. All samples were analysed within the effective usage period according to the expiration date shown on the packages. A sample of 0.1 kg of each seaweed was weighed individually, dried in an oven at 105 °C for 24 hr, until constant weight was achieved. Then samples were weighed again and homogenized. The nutrimental importance of essential elements was assessed on the basis of nutritional requirements (EFSA, 2006). Health risk due to the toxic elements present in seaweeds was estimated using risk estimators. (EFSA, 2006; JECFA, 2006, WHO, 2011a and 2011b)

Analytical method

Elemental determinations were carried out by Energy Dispersive Polarised X-Rays Fluorescence Spectrometry (EDPXRF). This is a simultaneous, reliable, sensitive, quantitative multielemental and non-destructive technique, suitable for routine analysis due to minimal sample preparation. This technique was previously used successfully for characterization of different complex matrices (Desideri et al., 2011, Stephens and Calder, 2004). Each sample was prepared by mixing it with Wachs-C 80004005 Mikropulver, a paraffin wax that helps to reduce the sample to tablet though pressure. The determinations were conducted with a Spectro-X-LAB2000 (SN DK 949196) Energy Dispersive Polarised X-Ray Fluorescence Spectrometry (EDPXRF).

The quality of data was ensured by calibrating the instrument with the following certified reference materials (CRM): MURST-ISS-A1 Marine sediment, GBW07310 Stream sediment, GBW08303 Farmland Soil, LGC6138 Soil, SRM 12-3-12 Sludge, STD 12-1-12 Fly ash, BCR

CRM 144R Sludge, CCRM LKSD1 Lake sediment, CCRM PACS-2 Marine sediment, NIST SRM 2709 Agricultural soil, NIST SRM 2711 Montana soil, NIST SRM 1633b Fly ash, NIST SRM 1575 Pine needles. The averaged analytical standard errors observed with respect to the reported certified materials were below 10% for As and Cd, and below 7% for the other measured elements (Apitz et al., 2009).

The detection limits, in mg/kg$_{dry.,}$ were: 120 for Al, 70 for Si, 6 for Mn, 1 for As, Rb, Cd, Sn, Pb Cu, Zn, Ni, Br, and I.

Statistical analyses

For every element the arithmetical mean concentration and relevant standard deviation, median, minimum and maximum values are reported. For the concentrations below the detection limit, the Minimum Detection Concentration (MDC) was considered to calculate the mean. Statistical analysis using Student's *t*-test was carried out. The criterion for significance was set at $P < 0.05$. Pearson coefficients (r) were also calculated in order to highlight the correlations between the metals content.

RESULTS

Element concentration

Tables 2 and 3 show, for each seaweed sample, the measured concentrations (mg/kg$_{dry}$) of the elements taken into account with the relevant standard deviation of repeated measures (3), minimum and maximum concentration, arithmetical mean and median.

Element concentrations varied according to the seaweed species and differences reached 2-4 orders of magnitude. Ca, Al, Si, Fe and Ti levels in *Lithothamnium calcareum* (red alga), a typical calcareous algae, were significantly higher than those found in other red algae and than those in brown and green algae; Ca was as high as 30% dry wt..

Palmaria palmata or dulse (red alga) showed the highest concentrations of K, Rb and Cl. The highest content of S was in *Chondrus crispus* or Irish mos (red alga). *Laminaria digitata* or kombu (brown alga) contained the highest concentrations of total As, Cd, Sn, Br and I. The highest concentration of Zn was in *Chlorella Pyrenoidosa* (green alga). *Ulva lactuca* or sea lettuce (green alga) displayed the highest levels of Cu, Ni, Mn and Pb.

Iodine levels varied greatly depending on the species of algae ranging from 3.4 in *Chlorella Pyrenoidosa* (green alga) to 7316 mg/kg$_{dry}$ in *Laminaria digitata* (brown alga). In the 4 types of seaweeds, brown algae presented the highest enrichment of I, followed by red algae. However, I levels in green and in green-blue algae were low.

The levels of Pb and Cd were not markedly different between the 4 types of seaweeds. In contrast a significant difference was found for As levels in brown algae compared to other seaweeds. With regard to essential elements, a significant difference was found for Cu in green algae compared to other seaweeds.

Pearson coefficients (r) were presented in Table 4. An optimum positive correlation ($r > 0.92$) occurred between Al *vs* Ca, *vs* Ti, *vs* Fe; Fe *vs* Si, *vs* Ti, *v*s Ca; Ca *vs* Ti; K *vs* Rb; Sn *vs* I. Positive correlations ($0.9 > r > 0.8$) occurred between: Sr *vs* Al, *vs* Ca, *vs* Ti; Si *vs* Al, *vs* Ca, *vs* Ti; Cl *vs* Rb, Mn *vs* Ni, Pb *vs* Br, Cd *vs* Sn and *vs* I. Similarly positive correlations ($0.8 > r > 0.7$) occurred between: Mn *vs* Cu, Sr *vs* Fe, As *vs* Sn and *vs* I, Br *vs* Rb.

The determination of many inorganic elements in various algae was reported by several investigators (Table 5). However, because of the differences in the treatment of materials and analytical procedures it is not possible to adequately compare findings with certainty. Further, Besada et al. (2009)) attributed seasonal variations to differences in metal concentrations in algae. Indeed, Pack et al (2014) demonstrated that rising temperatures in seawater enhanced Hg absorption by loaches suggesting that climate warming may increase risk of adverse effects due greater accumulation of the heavy metal in aquatic organisms.

Beneficial health effects resulting from seaweeds consumption

As far as the essential elements, as K, Ca, Cl and P (major) and Cu, Zn, Mn, Fe and I (trace) are concerned in order to assess beneficial health effects due to the consumption of algae, it was essential to know the amount of alga consumed. Some seaweeds are consumed in greater quantities than others. Some seaweeds are ingested in soups and stews and therefore up to 10 g dry weight, or more, might be consumed per serving. Others seaweeds are used as ingredients in salads and might be used in single gram quantities (Smith et al., 2010). Therefore, in our investigation, two approaches were considered as suggested by Phaneuf et al (1999): the average daily consumption of 3.3 g (dray wt) and the maximum amount of algae in a single service 12.5 g (dray wt). Table 6 shows for some essential element the mean and maximum concentration, daily intake (mg/day) from a 3.3-12.5 g daily seaweed consumption and comparison with nutritional requirements. The Population Reference Intake (PRI) and Adequate Intake (AI) (EFSA, 2006) were used in this study to evaluate the benefits from seaweed to the consuming population.

Among major elements, algae consumption of 3.3-12.5 g/day resulted in a daily intake of K and Cl (calculated from the mean concentrations) of 84-318 and 114-433 mg/day, which represents the 2.1-8.1, 4.9-18.8% of AI respectively. An intake of 11.7-44.4 and 90.8-343 mg/day were calculated for P and Ca respectively which represent 1.7-6.3 and 9.1-34.3 % of PRI.

Among trace elements, algae consumption of 3.3-12.5 g/day accounted for a daily intake (calculated from the mean concentrations) of 4.1-15.5, 0.11-0.41, 0.027-0.10, and 0.29-1.1

mg/day of Fe, Zn, Cu and Mn respectively which represent the 41.1-155, 0.91-3.4, 3.0-11.2 and 10.6-40.4% respectively of AI (Cu) or of PRI (Fe, Zn and Mn).

Risk characterization

The consumption of seaweed in Western countries showed an increase in recent years. Seaweeds, in fact, are part of diet due to their high content of protein, fatty acids, vitamins and minerals (Burtin, 2003). Nevertheless, their contaminant content is regulated in various ways by different legislation around the world: some countries have regulatory limits for heavy metals in algae food products such as France that authorized a specific list of seaweeds for human consumption and specified upper limits for the content of Pb, Cd, Sn, Hg, As and I (Burtin, 2003). At present, Italy or the European Union has not enforced specific legislation on elements in seaweed. In this study, the Provisional Tolerable Weekly or Monthly Intakes (PTWI and PTWM) recommended by Joint FAO/WHO Expert Committee on Food Additives (JECFA, 2006; WHO, 2011a; 2011b) referring to an average adult body weight of 70 kg and Tolerable Upper Intake Level (UL) recommended by European Food Safety Authority (EFSA, 2006) were used for the elements analysed to determine any potential toxicity from seaweeds to consumers.

The Joint FAO/WHO Expert Committee on Food Additives has designated for Al from all sources a PTWI of 70 mg (10 mg/day, 1 mg/day body weight per week) (JECFA, 2006). For inorganic As a PTWI of 1.05 mg (0.15 mg/day, 0.015 mg/kg body weight per week) was indicated (WHO, 2011a). The Joint FAO/WHO Expert Committee on Food Additives established for Cd for all sources a PTMI of 1.75 mg (0.058 mg/day, 0.025 mg/kg body weight per month) (WHO, 2011b), and for Pb a PTWI of 1.75 mg (0.25 mg/day, 0.025 mg/kg body weight per week). European Food Safety Authority (EFSA, 2006) has designated a UL of 2500, 25, 5 and 0.6 mg for Ca, Zn; Cu and I respectively.

Table 6 shows for every element the mean and maximum concentration, the daily dose for a 3.3-12.5 g daily seaweeds consumption and comparison with the risk estimations.

For toxic elements, a 3.3-12.5 g daily of *Ulva lactuca* consumption with the highest Pb level (6.7 mg/kg$_{dry}$) corresponds to a daily intake of 0.022-0.084 mg of this metal. This intake only represents approximately 8.8-33.7% of tolerable daily dose (0.25 mg) for Pb. The European Commission recommended 3.0 ppm Pb concentration in fresh seaweeds as maximum permissible limit (EC, 2011). Since the dry weight of algae is approximately 10-16% of fresh weight, a value of 3 ppm corresponds to a concentration range of 17-30 ppm dry weight. It is noteworthy that the contents of Pb in algae analyzed were all below this range.

For the tested sample (*Laminaria digitata*) with the highest Cd (7.2 mg/kg$_{dry}$), a 3.3-12.5 g daily seaweeds consumption corresponds to a daily intake of 0.024-0.090 mg of this metal. This intake alone represents approximately 40-150% of tolerable daily dose for Cd. For the tested sample (*Lithothamnium calcareum*) with the highest Al (8750 mg/kg$_{dry}$), an 3.3-12.5 g daily seaweeds consumption corresponds to a daily intake of 28.9-109 mg. This intake alone represents approximately 289-1094% of tolerable daily dose for Al. A 3.3-12.5 g daily consumption of *Laminaria digitata* corresponds to a daily intake of 0.22-0.85 mg of total As. This intake alone represents approximately 147-566% of tolerable daily dose for inorganic As.

With regard to the essential elements, the comparison between their intake and the relevant UL reported by EFSA (EFSA, 2006) shows that a 3.3-12.5 g daily consumption of *Lithothamnium calcareum* with the highest Ca level (303 g/kg$_{dry}$) corresponds to a daily intake of 1.00-3.78 g of this metal. This intake alone represents approximately 40-151% of tolerable daily dose (2.5 g) for Ca. A 3.3-12.5 g daily of *Ulva lactuca* consumption with the highest Cu (23.1 mg/kg$_{dry}$) corresponds to a daily intake of 0.076-0.29 mg of this metal. This intake only represents approximately 1.6-5.8% of tolerable daily dose (5 mg) for Cu. A 3.3-12.5 g daily consumption of *Chlorella Pyrenoidosa* with the highest Zn concentration (69.2 mg/kg$_{dry}$)

corresponds to a daily intake of 0.228-0.865 mg of this metal. This intake alone represents approximately 0.92-3.5% of tolerable daily dose (25 mg) for Zn. A 3.3-12.5 g daily consumption of *Laminaria digitata* with the highest I level (7316 mg/kg$_{dry}$) corresponds to a daily intake of 24.1-91.4 mg of this metal. This intake alone represents approximately 4,017-15,233% of tolerable daily dose (0.6 mg) for I (EC SCF, 2002).

DISCUSSION

This risk assessment shows that the intakes of Cd and Ca (for 12.5 g daily seaweeds consumption) and Al, As, I (for either 3.3 and 12.5 g daily seaweeds consumption) were higher than the reference dose limits. This should not be a cause for concern as the scenario selected for this study was extremely conservative: in fact, this scenario was based on: 1) daily consumption of 12.5 g of algae, which is a large amount for a single meal and 2) the maximal elemental concentration in seaweeds analyzed. In addition, depending upon methods of preparation, such a soaking, boiling, heating, marinating, or roasting the amounts of the various compounds in the seaweed material and in the medium in which it is prepared varies substantially (Mouritsen et al., 2013)

With regard to Al, Ca and Cd the relevant intakes are higher than the reference dose limits only in consumption of algae with the maximum concentration (*Lithotamnium calcareum* for Al and Ca and *Laminaria digitata* for Cd). When taking into account the mean levels of Al, Ca and Cd, the intakes of these metals are lower than the reference dose limits.

Thus ingestion of these species of seaweeds at concentrations below the maximal threshold limit would reduce the intake of Al, Ca and Cd. As far as arsenic is concerned, it was assumed that the species of metal found in seaweeds was inorganic As. It should be noted that the As measured was total As and not inorganic As; and thus the estimations showed are overestimated because, as reported in literature (Phaneuf et al., 1999; Almela et al., 2002; Besada et al., 2009), As is found mostly in the nontoxic organic compounds (i.e. arsenobetaine, arsenocholine). In order to assess the real risk of As-mediated toxicity, chemical composition and biochemical properties of the individual As species are important (Bernstam and Nriagu, 2000).

The situation of I is more problematic as the UL was exceeded for mean and maximal concentration. The response of the humans to an oversupply of I is specific to each individual and depends upon to previous and current I status (Dawczynski et al., 2007). Various investigators demonstrated the correlation between an high I consumption and some diseases as hypo and hyperthyroidism, goiter, thyroiditis while I deficiency leads to hypothyroidism (Pennington, 1990; Phaneuf et al., 1999). A regular consumption of seaweeds, particularly of *Laminaria sp.* and in general algae with a iodine concentration >45 $mgkg^{-1}_{dry}$, may result in excess I levels with consequent thyroid dysfunctions. Our results obtained found that 10 of the 14 analysed algae products exceeded this value. For regular consumers, it would be preferable to consume species with low I concentrations, such as *Chlorella pyrenoidosa* or *Spirulina platensis*.

Evidence indicates that individuals with known or potential thyroid complications should be cautioned with respect to dietary seaweed intake.

CONCLUSIONS

Although the algae samples analysed in this study are not completely devoid of contaminants, the heavy metals intake from algae was well below the recommended dose. However, it is advisable to check the content of Pb, Cd and inorganic As to protect the consumer against potential health risks. In the case of I, it is essential to know the thyroid tissue function status as certain seaweed species contain high quantities of this element. Therefore, continuous surveillance of contaminants in edible and commercialized seaweeds is important for consumer protection. Moreover, there is a need for an European regulation on maximal concentration of metals in these seaweeds.

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Table 1

Classes, species and commercial name of fourteen edible seaweeds

Color	Classes	Species	Commercial name
Red	*Rhodophyceae*	*Gelidium spp-r*	Agar
		Chondrus crispus	Irish mos
		Lithothamnium calcareum	
		Palmaria palmata	Dulse
		Phorphyra umbelicalis	Nori
Brown	*Phaeophyceae*	*Aschophyllum nodosus*	
		Fucus vescicolosus	Fucus
		Himanthalia elongata	Seaweed spaghetti
		Laminaria Digitata	Kombu
		Undaria pinnatifida	Wakame
Green	*Chlorophyceae*	*Chlorella Pyreinodosa*	
		Ulva/Enteromorpha sp	Green nori
		Ulva lactuca	Sea lattuce
Blue-green	*Cyanophyceae or Cyanobacteria*	*Spirulina Platensis*	

Table 2

Concentration (mg/kg$_{dry}$) of essential (major and trace) elements in algae samples

Sample	K	±	Ca	±	P	±	S	±	Cl	±	Mn	±	Ni	±	Cu	±	Zn	±	Br	±	I	±
Gelidium spp-r	153	15.3	1555	78	198	20	3449	345	3012	301	<6.0		1.9	0.1	1.8	0.1	3.8	0.3	103	10.3	88.9	8.9
Chondrus crispus	15365	1536	10913	546	2849	285	44571	4457	7457	746	43.2	3.0	5.9	0.4	3.0	0.2	66.8	4.7	856	85.6	604	60
Lithothamnium calcareum	1404	140	303025	15151	585	59	1581	158	3544	354	175.2	12	6.5	0.5	14.1	1.0	14.8	1.0	40.6	4	34.3	3.4
Palmaria palmata	95614	9561	861	43	2694	269	9882	988	119333	11933	17.4	1.2	4.2	0.3	4.4	0.3	26.2	1.8	673	67	220	22
Phorphyra umbelicalis	20614	2061	945	47	5023	502	19020	1902	41558	4156	31.4	2.2	2.3	0.2	13.6	1.0	44.9	3.1	101	10	41.8	4.2
Aschophyllum nodosus	22211	2221	10173	509	1403	140	17521	1752	19141	1914	42.2	3.0	2.0	0.1	3.6	0.3	28.7	2.0	347	35	811	81
Fucus vescicolosus	23803	2380	9353	468	1443	144	20529	2053	36760	3676	71.6	5.0	3.0	0.2	4.7	0.3	38.3	2.7	522	52	655	65
Himanthalia elongata	61546	6155	4994	250	1189	119	15866	1587	78767	7877	21.1	1.5	3.2	0.2	3.8	0.3	32.6	2.3	352	35	122	12
Laminaria Digitata	50516	5052	22765	1138	2196	220	10982	1098	72605	7260	34.2	2.4	2.3	0.2	5.2	0.4	11.0	0.8	1032	103	7316	732
Undaria pinnatifida	3303	330	10698	535	2872	287	8994	899	78347	7835	19.4	1.4	4.2	0.3	6.7	0.5	46.5	3.3	394	39	191	19
Chlorella Pyreinodosa	8509	851	1995	100	9655	965	6407	641	1986	199	60.9	4.3	2.1	0.2	17.5	1.2	69.2	4.8	2.1	0.2	3.4	0.3
Ulva Enteromorpha	25743	2574	2348	117	6555	656	15447	1545	5233	523	31.7	2.2	<1		10.1	0.7	31.7	2.2	63.3	6.3	28.1	2.8
Ulva lactuca	12480	1248	4800	240	3412	341	42716	4272	12457	1246	636.6	45	12	0.8	23.1	1.6	34.6	2.4	446	45	63.7	6.4
Spirulina Platensis	15418	1542	679	34	9657	966	6879	688	4431	443	30.0	2.1	1.3	0.1	2.2	0.2	13.1	0.9	3.8	0.4	4.8	0.5
Minimum	153		679		198		1581		1986		<6.0		<1		1.8		3.8		2.1		3.4	
Maximum	95614		303025		9657		44571		119333		636.6		12		23.1		69.2		1032		7316	
Mean	25477		27507		3552		15989		34616		87.2		3.9		8.1		33.0		353		727	
Standard deviation	26685		79535		3088		13052		38035		163.4		2.9		6.5		19.5		329		1915	
Median	18016		4897		2771		13214		15799		33.0		3.0		5.0		32.1		349		106	

Table 3

Concentration (mg/kg) of non-essential or toxic elements determined in algae samples

Sample	**Al**	±	**As**	±	**Rb**	±	**Sr**	±	**Cd**	±	**Sn**	±	**Pb**	±	**Ti**	±	**Si**	±
Gelidium spp-r	<120		<1		<1		4.9	0.5	<1.0		1.4	0.2	1.1	0.1	35.9	2.5	1612	113
Chondrus crispus	<120		3.8	0.8	13.7	1.4	83.2	8.3	1.0	0.2	3.4	0.5	5.1	0.4	53.8	3.8	868	61
Lithothamnium calcareum	8750	875	2.3	0.5	3.7	0.4	2095	209.5	<1.0		1.0	0.1	1.8	0.1	1364	95.5	22309	1562
Palmaria palmata	<120		10.1	2.0	43.8	4.4	71.0	7.1	<1.0		1.1	0.2	4.4	0.3	5.3	0.4	<70	
Phorphyra umbelicalis	<120		26.2	5.2	4.8	0.5	25.1	2.5	3.5	0.5	<1.0		2.2	0.2	33.9	2.4	432	30
Aschophyllum nodosus	<120		36.6	7.3	12.4	1.2	598	59.8	1.1	0.2	4.7	0.7	2.5	0.2	53.6	3.8	512	36
Fucus vescicolosus	<120		34.3	6.9	13.9	1.4	537	53.7	1.1	0.2	4.1	0.6	4.0	0.3	55.1	3.9	<70	
Himanthalia elongata	<120		35.3	7.1	26.3	2.6	560	56.0	<1.0		<1.0		3.0	0.2	3.5	0.2	<70	
Laminaria Digitata	<120		67.6	13.5	26.9	2.7	760	76.0	7.2	1.1	34.0	5.1	6.5	0.5	69.8	4.9	<70	
Undaria pinnatifida	<120		31.6	6.3	6.8	0.7	883	88.3	1.3	0.2	1.7	0.3	4.6	0.3	88.4	6.2	<70	
Chlorella Pyreinodosa	<120		1.3	0.3	3.8	0.4	13.9	1.4	<1.0		1.0	0.1	3.1	0.2	25.0	1.7	12690	888
Ulva Enteromorpha	<120		15.4	3.1	4.1	0.4	26.8	2.7	3.5	0.5	<1.0		1.3	0.1	1.7	0.1	1644	115
Ulva lactuca	<120		2.8	0.6	13.7	1.4	62.6	6.3	<1.0		1.5	0.2	6.7	0.5	156	10.9	8316	582
Spirulina Platensis	<120		1.1	0.2	1.8	0.2	27.6	2.8	<1.0		<1.0		<1		16.3	1.1	1998	140
Minimum	<120		<1.0		<1		4.9		<1		<1		<1		1.7		<70.0	
Maximum	8750		67.6		43.8		2095		7.2		34.0		6.7		1364		22309	
Mean	736		19.3		12.6		411		1.8		4.1		3.4		140		3624	
Standard deviation	2307		20.0		11.8		579		1.8		8.7		1.9		355		6531	
Median	<120		12.7		12.4		77.1		1.0		1.2		3.0		44.8		690	

Table 5

Comparison between some elements contents found in this study (mg/kg) and those reported by other authors (Phaneuf et al., 1999 [a]Dawczynski, 2007, [b]Hou and Yan, 1998, [c] Caliceti et al., 2002) in the seaweeds commercialized in different world's regions.

Element	Palmaria palmata	Porphyra umbelicalis	Fucus vescicolosus	Laminaria sp	Undaria pinnatifida	Aschilophyllum nodosus	Ulva lactuca	Enteromorpha
Fe	1009 (203)	724-590[c] (597)	427-231[c] (383.4)	308 (764)	171[c] (588)	171 (354)	2486-1033[c] (2007)	7037 (190)
Cu	8.1 (4.4)	10.3-9[c](13.6)	4.1-8[c] (4.7)	2.8 (5.2)	2[c] (6.7)	4.2 (3.6)	19.2-13[c](23.1)	22.7 (10.1)
Zn	31.2 (26.2)	30.3-48[c] (45)	32.6-110[c] (38.3)	31.2 (11)	97[c] (46.5)	35.6 (28.7)	33.3-64[c](34.6)	38.2 (31.7)
Mn	35.8 (17.4)	33.2 (31.4)	158 (71.6)	9.7 (34.2)		19.1(42.2)	409 (637)	156 (31.7)
Ni		11[c] (2.3)	4.6[c](3.0)		1.2[c] (4.2)		2.6 [c] (12)	
As	9.4-7.6[b] (10.1)	19-13[c] (26.2)	31.6-128[b]-40[c](34.3)	60.1-53[b] (67.6)	32[b]-70[c] (31.6)	23 (36.6)	6-7[c] (2.8)	7.2-2.3[b] (15.4)
Pb	0.84-1.1[b] (4.4)	0.67-2.7[c] (2.2)	0.44-0.63[b]-1.6[c] (4.0)	0.40, <0.05[b]-2.7[c] (6.5)	<0.05[b]-2.2[c](4.6)	0.16 (2.5)	1.6-7.3[c] (6.7)	3.2-1.33[b] (1.3)
Cd	0.32-0.70[b] (1.0)	0.29-0.1[c] (3.5)	1.4-1.45[b]-0.6[c](1.1)	1.53-0.30[b] (7.2)	1.5[b]-0.1[c] (1.3)	0.46 (1.1)	0.22-0.2[c](1.0)	0.28-0.03[b](3.5)
I	173 (220)	31.7 (41.8)	226 (654.7)	763(7613)		482 (811)	136 (63.7)	22.7 (28.1)
K		27700[a] -32000[b] (20615)		102000[a] (50516)	4800[a] (3303)		50400[b] (12480)	8500[b] (25743)
Ca		3300[a] --2400[b](945)		7420[a] (22765)	8990[a] (10690)		4900[b] (4800)	7800[b] (2300)
P		5380[a](5023)		2660[a](2197)	3620[a](2872)			
Cl		34900[b](41558)					117800[b](12457)	81600[b](5233)

Table 6 Element mean and maximum concentration (mg/kg), its daily dose (mg/day) from algae consumption and comparison with daily dose risk estimators (PTWI= Provisional Tolerable Weekly Intake, PTMI= Provisional Tolerable Monthly Intake; UL= Tolerable Upper Intake Level) for a 70 kg man (EFSA, 2006; JECFA, 2006, WHO, 2011a, 2011b) and with nutritional requirements (PRI= Population reference intake; AI= adequate intake) (EFSA, 2006)

Element	Seaweed with a maximum concentration	Mean-max concentration	Daily dose for 3.3 g consumption	Daily dose for 12.5 g consumption	Daily dose from risk estimators	Daily nutritional requirements
Al	Lithothamnium calcareum	736-8750	2.43-28.87	9.2-109.4	10 (from PTWI)	
total As	Laminaria digitata	19.3-67.6	0.064-0.22	0.241-0.85	0.15 (inorganic) (from PTWI)	
Pb	Ulva lactuca	3.4-6.7	0.011-0.022	0.042-0.084	0.250 (from PTWI)	
Cd	Laminaria digitata	1.8-7.2	0.006-0.024	0.022-0.09	0.058 (from PTMI)	
Zn	Chlorella pyrenoidosa	33-69.2	0.109-0.228	0.412-0.865	25 (from UL)	12 (PRI)
Cu	Ulva lactuca	8.1-23.1	0.027-0.076	0.101-0.29	5 (from UL)	0.9 (AI)
I	Laminaria digitata	727-7316	2.40-24.1	9.09-91.4	0.6 (from UL)	0.15 (AI)
Fe	Lithothamnium calcareum	1242-9190	4.11-30.3	15.5-115		10 (PRI)
Mn	Ulva lactuca	87.2-637	0.288-2.10	1.09-7.96		2.7 (PRI)
K	Palmaria palmata	25477-95614	84.1-315	318-1195		3900 (AI)
Ca	Lithothamnium calcareum	27507-303025	90.8-1000	343-3787	2500 (from UL)	1000 (PRI)
P	Spirulina Platensis	3552-9657	11.7-31.87	44.4-121		700 (PRI)
Cl	Palmaria palmata	34616-119333	114-394	433-1491		2300 (AI)