Estimation and comparison of effective compounds in two algae species identified in Qeshm Island (Persian Gulf)

Etemadian Y.¹; Shabanpour B.^{1*}; Ramzanpour Z.²; Shaviklo A.R.³; Kordjazi M.¹

Received: July 2017

Accepted: September 2017

Abstract

Estimation and comparison of effective compounds (mineral contents, amino acids compositions, fatty acids distributions, protein, fat and total fiber contents) in two algae species Sirophysalis trinodis and Polycladia myrica collected from Qeshm Island were done. The samples washed up with tap water and then were dried in an oven at 40 °C for 3 days. The dried algae samples were pulverized and packed in airtight plastic bags. The results showed that the marine macro-algae included low lipid contents about 0.03±0.01 % and 0.33±0.10% dry sample weight and proved to be a rich source of fatty acids (Palmitic and Oleic acid) and minerals. The protein content of these brown algae was lower than 15%. All essential amino acids were detected in the algae species, featured uniquely high concentrations of amino acids when compared to ground plants. Also, results showed that the chlorophyll amount dependent on solvent type and cell wall structure of algae. Hence, in this study, among the three solvents of acetone, diethyl ether and methanol, acetone showed the best performance for the extraction of chlorophyll in both algae S. trinodis and P. myrica. In general, the aim of this study was to determine the valuable compounds of two abundant species of brown algae in the south of the country and introduce them to food units.

Keywords: Brown algae, Effective compounds, Qeshm Island, Persian Gulf

¹⁻Faculty of Fisheries Science, Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Iran.

²⁻International Sturgeon Research Institute, Agricultural Research Education and Extension Organization (AREEO), Rasht, Iran.

³⁻Animal Science Research Institute of Iran, Agricultural Research, Education and Extension Organization (AREEO), Karaj, Iran.

^{*}Corresponding author's Email: b_shabanpour@yahoo.com

Introduction

The oceans and seas cover 31% of the Earth's surface and are the largest reservoir on Earth. Of marine products, algae is a valuable food and mineral resource (Akhtar and Sultan, 2002), as far as being known as healthy food. In general, macro-algae can be classified as red algae (Rhodophyta), brown algae (Phaeophyta) or green algae depending (Chlorophyta), on their nutrient and chemical composition (Dawczynski et al., 2007). Red and brown algae are mainly used as human food sources. So that, the different species consumed present a great nutritional value as source of proteins, carbohydrates, minerals and vitamins (Marinho-Soriano et al., 2006). On the other hand, variable values of protein in different types of algae can be explained by variations of seasons and environmental conditions such as temperature, salinity and nutrients (Dadollahi Sohrab et al., 2012). The protein content of brown algae species, e.g., Laminaria japonica or Undaria pinnatifida is relatively low with 7-16% dry weight (DW). In contrast, red algae, e.g., Palmaria palmata and Porphyra tenera contain 21-47 g protein 100g⁻¹ DW (Ruperez and Saura-Calixto, 2001). The protein in algae contains all essential amino acids is available throughout the year although with seasonal variations. Also, the fat content of marine macro-algae has been found in many brown algae varieties and has high concentrations of oleic al., acid (Dawczynski et 2007: Matanjun et al., 2009; Silva et al., 2013; Ragonese et al., 2014). The types and abundance of carbohydrates vary strongly between algae species. The typical carbohydrates in brown algae varieties consist of fucoidan, laminaran, and cellulose. alginates mannitol (Kraan et al., 2012). Also, brown algae mainly fibers are cellulose and insoluble alginates. The typical algae carbohydrates are not digestible by the human gastrointestinal tract and therefore, they are dietary fibers. The consumption of this dietary fiber has been proven to be health-promoting and it benefits are well documented in the scientific literature (Guidel-Urbano and Goni, 2002). Hence, in countries such as China, Japan, and Korea, they have been commonly utilized in human alimentation (Lahaye, 1991). In Iran algae are found in the southern shores of the country, especially on in the coast of Chabahar (Gulf of Oman) and Qeshm Island (Persian Gulf). During a research conducted by Sohrabipour and Rabiei (2017), 347 algae species were identified, including 79 green algae species, 80 brown algae species and 167 red algae species. The most important families of brown algae were Dictyotaceae and Sargassaceae. Both brown algae S. trinodis and P. myrica selected in this study are in the group of Sargassaceae family. From the point of economic value, these species have high production capacity and can be used to create a variety of business fields with proper commercial With exploitation. all these interpretations, algae differ greatly in the quality, color, consistency, and nutrient content. Therefore, the present investigation estimates and compares

the effective compounds of two available brown algae in the south of Qeshm Island.

Materials and method

Sample collection and preparation

Two types of brown macro-algae *Sirophysalis trinodis* and *Polycladia myrica* (Fig. 1) were collected from the middle and lower parts of the tidal area on the rocky bed surface of Qeshm Island (the southern coast of the Persian

Gulf) in the winter of 2015 in February and March months. They were washed up with tap water to remove salt and epiphytes. Then they were dried for 3 days in an oven at 40 °C (The temperature was no higher than 60 °C) and pulverized to obtain uniform particles (0.5 mm). The pulverized algae samples were stored in airtight plastic bags and were put up in a desiccator prior design of to experiments.



Sirophysalis trinodis

Polycladia myrica

Figure 1: Appearance of two species of brown algae collected from Qeshm Island (Persian Gulf).

Proximate analyses

Protein, moisture, ash, crude lipids, carbohydrate (reducing and non-reducing sugar), crude fiber and nitrogen-free extract contents were estimated by the method of AOAC, (2005).

Chlorophyll extraction process

The extraction process of chlorophylls was done using the method of Jeffrey and Humphrey (1975) and Dere *et al.* (1998) with a slight modification. Briefly, 1 g sample with 50 ml of three solvents (acetone 100%, diethyl ether 95% and methanol 96%) was separately

homogenized by a homogenizer (IKA T2 Digital, Ultraturrax, Germany) at 15000 rpm for 2 min at room temperature. Then, the mixture was filtered through two layer cheese cloths and centrifuged at 2500 rpm for 10 min. The supernatant was separated. The absorbance was measured at 400-700 nm using UV-Vis spectrophotometer (Biochrom Ltd, Cambridge CB4OFJ England). The calculation of brown algae chlorophyll ($\mu g g^{-1}$ of fresh weight) was given by the following formula: (C_a=Chlorophyll $C_{b}=$ a. Chlorophyll b, C_{x+c} =Total carotene) (Dere et al., 1998).

Acetone	$\begin{split} C_a &= 11.75 \ A_{662} - 2.350 \ A_{645} \\ C_b &= 18.61 \ A_{645} - 3.960 \ A_{662} \\ C_{x+c} &= 1000 \ A_{470} - 2.270 \ C_a - 81.4 \ C_b / 227 \end{split}$
Diethyl ether	$\begin{split} C_a &= 10.05 \ A_{662} - 0.766 \ A_{644} \\ C_b &= 16.37 \ A_{644} - 3.140 \ A_{662} \\ C_{x+c} &= 1000 \ A_{470} - 1.280 \ C_a - 56.7 \ C_b / \ 230 \end{split}$
Methanol	$\begin{split} C_a &= 15.65 \ A_{666} - 7.340 \ A_{653} \\ C_b &= 27.05 \ A_{653} - 11.21 \ A_{666} \\ C_{x+c} &= 1000 \ A_{470} - 2.860 \ C_a - 129.2 \ C_b / \ 245 \end{split}$

Assessment of mineral content

The mineral content was measured by atomic absorption spectrophotometer (Perkin Elmer Analyst 800, USA). In remove order to the organic constituents present in S. trinodis and P. myrica powders, 10 ml of HCl was added to it. Then after 5 min, 10 ml of concentrated HNO₃ was added. The samples were incubated for 5 min. After that, 10 ml of concentrated HCl was added to it, again. The solutions were evaporated by heating. The residue was dissolved in 10 ml of concentrated HCl. The solutions were filtrated and measured by atomic absorption spectrophotometer. The results were expressed as ppm and percentage (Ruperez, 2002).

Assessment of fatty acids composition

Fatty acids composition was assessed according to the protocol of Yayli *et al.* (2001). Briefly, 0.075 g of *S. trinodis* and *P. myrica* powders were dissolved in 1 ml of toluene and 2 ml of 1% H_2SO_4 (in methanol). The esters were extracted twice with 5 ml of hexane. The organic layer was separated and washed up with 4 ml of 2% KHCO₃. The mixture was dried using anhydrous Na_2SO_4 and filtered. The organic solvent was removed and fatty acid methyl ester (FAME) was subjected to gas chromatography [6890 N system, GC Agilent Technologies]. The initial temperature was 70°C, and then temperature increased to 250°C. The injection temperature was 220°C. Helium was applied as the carrier gas at the flow rate of 1 μ l min⁻¹.

Estimation of proline

Proline contents of S. trinodis and P. determined mvrica algae were according to the method of Bates (1973). The acid – ninhydrin solution was prepared by adding 1.25 g of ninhydrin in 30 ml of glacial acetic acid and 20 ml of 6 M phosphoric acid. The mixture was agitated until dissolved and stored at 4°C (It is stable for 24h). About 0.5 g of S. trinodis and P. myrica powders were separately homogenized in 10 ml of 3% aqueous sulfosalicylic acid and filtered with Whatman No. 1 filter paper. 2 ml of the filtered extract was mixed with 2 ml of acid ninhydrin and 2 ml of glacial acetic acid. This mixture was kept in an incubator at 100°C for 60 min. The reaction was terminated by placing the tubes in ice bath. The mixture was extracted with 4 ml toluene and mixed vigorously for 15-20 seconds. The chromophore containing toluene was collected and warmed at room temperature. Then, the absorbance was measured at 520 nm using UV-Vis spectrophotometer (Biochrom Ltd, Cambridge CB4OFJ England). The toluene and proline were used as blank and standard. respectively.

 $\label{eq:proline (mol/g)} \mbox{Proline (\mu mol/g)} = \frac{[\mbox{proline (\mu g/ml)} \times \mbox{toluene (ml)} + 115.5 \ (\mbox{\mu g/\mu mol)}]}{[\mbox{sample (g)} + 5]}$

Amino acids analysis

Amino acids profile of S. trinodis and was *P. myrica* algae determined according to Gratzfeld-Huesgen (1999). Briefly, 2 g of S. trinodis and P. myrica powders were merged with phosphate buffer (pH 7) and centrifuged at 3000 rpm for 20 min at $4^{\circ C}$. The supernatant was collected and proteins were precipitated by adding 10% TCA. The protein pellet was suspended in NaOH (1 N) and hydrolyzed by incubating the solution with HCl (6 N) in boiling water bath for 24 h. Then. the supernatant was collected bv centrifuging the sample at 3500 rpm for 15 min. The supernatant was filtered and neutralized with NaOH (1N). The filtered supernatant was diluted to 1:100 (v/v) with milliQ water and subjected to reversed phase HPLC analysis (HP-1101 Agilent Technologies with UV) and Fluorescent detectors (Agilent Technologies, Palo Alto, USA). Amino acids profile of S. trinodis and P. myrica algae was expressed as mg of amino acid per gram of protein.

Statistical analysis

Statistical analysis was performed using Analysis of Variance (ANOVA). The LSD and Duncan tests (p=0.05) was used to determine any significance of differences between specific means (Sigma Stat, 21 version, 2012, USA). All determinations were performed in triplicates, and the data are expressed in terms of mean±(SD (standard deviation)).

Results

Proximate compositions of two brown algae have been shown in Table 1. Chlorophyll a and x+c contents in two brown macro-algae species of S. trinodis and P. myrica were determined (Table 2). The highest chlorophyll a content was observed in S. trinodis species. In terms of chlorophyll x+c (the total carotene) contents, the significant difference was observed between the species (p < 0.05). The highest content of the total carotene was also showed in S. trinodis. Regarding to chlorophyll b, there was no absorption because of the absence of chlorophyll b in brown algae, so it was neglected. On the other hand, results indicated that there was a significant relationship solvents used between in the especially chlorophyll extraction, between the acetone and others. Generally, it has been found that the use of different solvents in order to extract chlorophyll will show different results. The reason could be because of the various structures of the cell walls and the plants type. These results were with Wellburn (1994)consistent studies. However, there are very little results in relation to the brown algae chlorophyll. The results of mineral contents (Table 3) revealed that S. trinodis and P. myrica contain high amount of calcium (p < 0.05). Also, the amount of iron was 0.19±0.02% and $0.32\pm0.01\%$ in S. trinodis and P. respectively (*p*<0.05). mvrica. Magnesium content was 1.04±0.04% and 1.48±0.06% in S. trinodis and P. *myrica*, respectively (*p*<0.05).

powder.		
Proximate compositions	S. trinodis	P. myrica
Crude protein	10.39 [*] ±0.14% DW	9.57±0.06% DW
Moisture	13.42 [*] ±0.15% DW	7.26±0.76% DW
Ash	33.16±2.45% DW	33.62±0.61% DW
Crude lipid	0.03±0.01% DW	$0.33^{*}\pm0.10\%$ DW
Sugar	36.53±8.95% DW	34.33±10.92% DW
Crude fiber	$5.88^{*} \pm 0.14\%$ DW	3.90±0.36% DW
Nitrogen-free extract	37.12±2.56%DW	45.32 [*] ±3.40% DW

Table 1: Proximate compositions of brown algae Sirophysalis trinodis and Polycladia myrica powder.

*Star sign in each row indicate significant differences (p < 0.05).

Table 2: Chlorophyll contents of two brown algae Sirophysalis trinodis and Polycladia myrica powder using different solvents (μg g⁻¹ fresh weight).

		Solvents		
		Acetone	Diethyl ether	Methanol
	Ca	5.41 ± 0.94^{ab}	3.35±0.93 ^{cd}	4.25 ± 1.71^{bc}
S. trinodis	C_b	-	-	-
	C_{x+c}	$2.40{\pm}0.26^{a}$	2.11 ± 0.55^{ab}	1.46 ± 0.54^{bcd}
	C_a	4.46±0.65 ^{abc}	2.69±0.36 ^{de}	3.42±0.41 ^{cd}
P. myrica	C_b	-	-	-
	C_{x+c}	1.93±0.13 ^{abc}	1.46 ± 0.26^{bcd}	1.01 ± 0.22^{de}

 C_a =Chlorophyll a, C_b =Chlorophyll b, C_{x+c} =Total carotene;

Lowercase letters in each row indicate significant differences (p < 0.05).

 Table 3: Mineral compositions determined by atomic absorption spectrophotometer in brown algae

 Sirophysalis trinodis and Polycladia myrica.

Name of the element	Observed co	ncentration
	S. trinodis	P. myrica
Calcium	4.58±0.44 %	$10.28^{*}\pm0.39$ %
Magnesium	1.04±0.04 %	$1.48^{*}\pm0.06$ %
Iron	0.19±0.02 %	$0.32^{*}\pm0.01$ %
Manganese	62.67±9.81 ppm	$147.00^{*} \pm 4.00 \text{ ppm}$
Copper	6.33±1.53 ppm	9.67 [*] ±5.51 ppm
Zinc	15.33±0.58 ppm	19.67 [*] ±3.79 ppm

*Star sign in each row indicate significant differences (p<0.05).

Manganese in *S. trinodis* and *P. myrica* was 62.67 ± 9.81 ppm and 147.00 ± 4.00 ppm, respectively (p<0.05). Apart from these major elements, *S. trinodis* and *P. myrica* were found to possess trace elements like copper and zinc in the concentrations of 6.33 ± 1.53 ppm (copper), 15.33 ± 0.58 ppm (zinc) and 9.67 ± 5.51 ppm (copper), 19.67 ± 3.79 ppm (zinc), respectively (p<0.05). The results of fatty acids (Table 4) showed that there was a mixture of both

saturated and unsaturated fatty acids. The saturated fatty acids in *S. trinodis* and *P. myrica* were myristic acid, palmitic acid, stearic acid and arachidic acid. The unsaturated fatty acids were included oleic acid, α -linoleic acid and α -linolenic. Investigation of amino acids profile of *S. trinodis* and *P. myrica* revealed that both seaweeds contain all the essential amino acids. Amino acids such as Alanine, Serine, Methionine, Glycine, Aspartic acid, Isoleucine, Leucine, Lysine, Threonine, Glutamic acid, Tyrosine, Valine, Phenyl alanine and Arginine have been found in *S. trinodis* and *P. myrica*. The value of amino acids in *S. trinodis* was more than *P. myrica*. There was a significant difference between amino acids of these brown algae, statistically (p<0.05). Four amino acids histidine, glutamine, ornithine and citruline were absent in *S. trinodis* and *P. myrica* (Table 5). The proline content of *S. trinodis and P. myrica* was observed $11.32\pm0.64 \mu mol g^{-1}$ and $5.64\pm0.49 \mu mol g^{-1}$ DW, respectively. There was a significant difference between the proline content of samples, statistically (*p*<0.05).

 Table 4: Fatty acids composition of brown algae Sirophysalis trinodis and Polycladia myrica powder.

Fatty acids	S. trinodis (%w/w)	P. myrica (%w/w)
Myristic acid (14:0)	$3.45^{*}\pm0.01$	2.99±0.01
Palmitic acid (16:0)	33.60±0.01	35.67±0.01
Stearic acid (18:0)	$2.66^* \pm 0.01$	1.72 ± 0.01
Arachidic acid (20:0)	1.59 ± 0.01	$5.14^{*}\pm0.01$
Palmitoleic acid (16:1)	4.43 ± 0.01	4.03±0.01
Oleic acid (18:1)	$24.27^{*}\pm0.01$	15.71±0.01
α -Linoleic acid (18:2)	7.76 ± 0.01	7.35±0.01
α -Linolenic acid (18:3)	$4.42^{*}\pm0.01$	3.69±0.01

*Star sign in each row indicate significant differences (p<0.05).

Table 5: Amino acids composition of Sirophysalis trinodis and Polycladia myrica powder.

	I I I I I I I	J
Amino acids	S. trinodis (mg/g of protein)	P. myrica (mg/g of protein)
Alanine	$3.84^* \pm 0.01$	2.99±0.01
Serine	3.71±0.01	3.11±0.01
Methionine	1.16 ± 0.01	0.91±0.01
Glycine	3.63 ± 0.01	3.13±0.01
Aspartic acid	$4.47^{*}\pm0.01$	3.05±0.01
Isoleucine	1.57 ± 0.01	0.97±0.01
Leucine	$2.15^* \pm 0.01$	1.26 ± 0.01
Lysine	3.39 ± 0.01	3.16±0.01
Threonine	3.12 ±0.01	2.64 ± 0.01
Glutamic acid	$4.61^* \pm 0.01$	2.90±0.01
Tyrosine	0.55 ±0.01	0.42 ± 0.01
Valine	1.96 ± 0.01	1.36 ± 0.01
Phenyl alanine	$1.23^{*}\pm0.01$	0.58 ± 0.01
Arginine	$3.18^* \pm 0.01$	2.61±0.01
Proline	$11.32^* \pm 0.64 \ \mu mol \ g^{-1}$	$5.64\pm0.49 \ \mu mol \ g^{-1}$

*Star sign in each row indicate significant differences (p < 0.05).

Discussion

Proximate compositions of two brown algae selected in this study were varied. The reason for this could be due to differences in species, geographic area, seasons or environmental conditions (Dawczynski *et al.*, 2007). Generally, marine plants undergo physical and chemical parameters change in different seasons, which may be affected by these factors, metabolic responses (Photosynthesis and growth rate). On the other hand, their nutritional value may change due to their changes in different seasons (Ordu na-Rojas et al., 2002). The crude protein content was respectively 10.39±0.14% and 9.57±0.06% DW for S. trinodis and P. mvrica (*p*<0.05). According to Fleurence (1999) results, most brown algae have protein contents lower than 15% (DW), except for Undaria pinnatifida. Gómez et al. (2010) have also reported higher protein contents (10.9 to 25.7% DW) in Himanthalia elongate, Bifurcaria bifurcata, and Laminaria saccharina. So, the protein content of algae varieties was varies greatly and demonstrates a dependence factors such as season on and environmental growth conditions (Dawczynski et al. 2007). The moisture content in both species was statistically significant (p < 0.05). The lowest value of moisture-has been found for P. myrica and the highest has been found for S. trinodis. Ash content was high and ranged about 33.16±2.45% DW for *S. trinodis* and 33.62±0.61% DW for *P*. myrica. Similar values of ash content have been reported by other authors for brown algae (Ruperez and Saura-Calixto, 2001). The high ash content is a general feature for algae and it is much higher than other terrestrial vegetables (Ruperez, 2002). The high ash content in algae invariably indicates the presence of appreciable amount of diverse minerals. However, ash contents are varied between species. geographical locations and seasons (Gómez et al. 2010). Lipids are the minor components of algae. In the present study, total lipid content was 0.03±0.01% and 0.33±0.10% DW for S. trinodis and P. myrica respectively. On

the other hand, at least 90% of carbohydrates in nature are in the form of polysaccharides. Carbohydrates usually make up more than 70% of the caloric value of the human diet. Reducing non-reducing and sugar were 36.53±8.95% and contents 34.33±10.92% DW. On the other hand, nitrogen-free extract of samples were 37.12±2.56% and 45.32±3.40% for S. trinodis and P. myrica, respectively wide (*p*<0.05). The variation of carbohydrate contents in different species of brown algae might be due to the influence of different factors such as temperature, salinity and sunlight intensity. The crude fiber contents of S. trinodis Ρ. myrica were and 5.88±0.14% 3.90±0.36% and DW. Dawczynski et al. (2007)tested nutritious compounds of 34 species of red and brown algae and reported that both types of algae have a low fat content but rich in dietary fiber. Cofrades et al. (2010), studying the nutritional and antioxidant properties of various types of red and brown algae, reported that these algae have a significant potential as nutritional ingredients such as fiber, minerals, and rare elements, proteins and lipids, and also have high antioxidant capacity. Chlorophyll is an important source of nitrogen storage and a predominant constituent in green plants and algae with positive effects on oxidation. inflammation and wound healing (Inanc, 2011). Also, chlorophyll can act as a powerful antioxidant (Hsu et al., 2013) against carcinogens and protects body from the lipid peroxidation of low density lipoprotein (Sakthivel and Devi,

2015). Although there is the chlorophyll a in all algae groups, in the present study it was observed that the amount of chlorophyll a in different species depends on the type of solvent have been used to extract them. For example, other researchers also found that the type of solvents used in the pigment extraction have an important effect, so that in the studies on Scenedesmus and Selenastrum quadricauda capricornutum microalgae and solvents such as ethanol, methanol and acetone, the best solvent was ethanol (Sartory and Grobbelaar, 1984; Ritchie, 2006). For safety reasons, efficient extraction of chlorophylls and the convenience of being able to use polystyrene cuvettes, for the algorithms ethanol are recommended for routine assays of chlorophylls. Overall in addition to the solvent type, other findings from many researches indicate that some factors such as environmental conditions, the light, the water depth and the variety of the cell wall structures can affect the amount of chlorophyll (Rossa et al. 2002; Sultan et al., 2016). Therefore, the use of suitable methods and materials for breaking up cell walls will be useful for chlorophyll extraction. The total carotene content is various in algae groups and it was proved that more carotene pigments belonging to Phaeophyta group. In these samples studied, the carotene content in S. trinodis was observed higher compare to the other brown algae. There was an inverse relationship between chlorophyll a and x+c (Table 2). In fact, this results indicated that when chlorophyll a decreases, the carotene increases and the algae turns to yellow. In some other studies it has been found that methanol algorithms would be convenient for assays associated with work HPLC for extraction of chlorophyll (Ritchie, 2006). But it should be noted that the selection of the method and solvent in connection with pigments extraction is useful. For example, although methanol is a good solvent in some extraction methods, but it is a toxic solvent. In this project, acetone was the best solvent for S. trinodis and P. myrica species for chlorophyll extraction. With regards to all above, these tests still need to be explored further in the future, because there is high demand for use of natural pigments of algae in food products, particularly dairy and beverages (Christaki et al., 2013) as food additives and also in cosmetics. As a result, this study shows that sea algae can be a promising natural resource for chlorophyll also. chlorophyll and extraction with different solvents can help to better use in food-drug industry as potential functional ingredients.

Other nutritional compositions of algae such as mineral and fatty acids make algae a nutritive, low-energy food which represents an important food alternative. Regards to the mineral contents, algae may serve as a food supplement in daily intakes of some minerals and trace elements. Essential minerals and trace elements for human nutrition are the major constituents of algae which range from 8-40% (Ruperez, 2002). Algae are a rich source of minerals when compared to land plants. In fact, the cell wall

polysaccharides, sulfates. anionic carboxyl and phosphate groups act as binding sites for metal retention. The results of mineral contents (Table 3) revealed that S. trinodis and P. myrica contain high amount of calcium. Calcium has an important role in maintaining bone strength. So, eating material this is essential. The recommended dietary allowance (RDA) for calcium is $1.000-1.200 \text{ mg day}^{-1}$ for adults (Oregon State University, 2017). The amount of iron was 0.19±0.02% and 0.32±0.01% in S. trinodis and P. myrica respectivley. Iron involved to makes red blood cells and provides oxygen. Much of our iron requirement is met through recycling iron from senescent red blood cells. The RDA for iron is 8 mg/day for men and postmenopausal women, 18 mg day⁻¹ for premenopausal women and 27 mg day⁻¹ for pregnant women (Oregon State University, 2017). The amount of magnesium was $1.04 \pm 0.04\%$ and $1.48\pm0.06\%$ in S. trinodis and P. myrica, respectively. The low level of magnesium contributes to the heavy metals deposition in the brain that precedes Alzheimer's disease. Thus magnesium is essential in regulating central nervous system (Murck, 2002). The Linus Pauling Institute supports the latest RDA for magnesium intake (400-420 mg day⁻¹ for men and 310-320 mg dav⁻¹ for women). Following the Linus Pauling Institute recommendation to take a daily multivitamin mineral⁻¹ supplement may ensure an intake of at least 100 mg of magnesium day⁻¹ (Oregon State University, 2017). Algae can be interesting candidates to explore

manganese sources. A study in healthy postmenopausal women found that a supplement containing manganese (5 mg day⁻¹), copper (2.5 mg day⁻¹), and zinc (15 mg day⁻¹) are necessary for body (Oregon State University, 2017). Generally, the results showed that both families from these brown algae can be useful for human and animals nutrition. Algae when compared to terrestrial vegetables possess beneficial effects cardio-protective, like antimitotic, anticancer, antiviral and anti-mutagenic activities. So, the fatty acids are an parameter important for human nutrition (Yaich et al., 2011). The results of fatty acids (Table 4) showed that there was a mixture of both saturated and unsaturated fatty acids. Also, S. trinodis algae contain lower palmitic acid and arachidic acid and more amounts of myristic acid, stearic acid, oleic acid, α -linoleic acid and α linolenic acid. In total, S. trinodis was contained 41.30% (w/w) saturated fatty acids and 40.88% (w/w) unsaturated fatty acids. P. myrica was contained 45.52% (w/w) saturated fatty acids and 30.78% (w/w) unsaturated fatty acids. Dawczynski et al. (2007) reported that three edible brown algae Laminaria sp., Undaria pinnatifida and Hizikia fusiformehas had suitable levels of saturated and unsaturated fatty acids. For example, the level of unsaturated fattv acid (a-Linoleic acid) of Laminaria sp. was 5.48% w/w, that it was low compared with S. trinodis and P. myrica in this study. The results of fatty acids analysis showed that S. trinodis and P. myrica had an adequate amount of the saturated and unsaturated

fatty acids, which can be influenced on health of body. In particular, oleic acid and α -linolenic acid might help in lowering the blood cholesterol, act as an antioxidant, repair the damaged cells and improve the heart function. In addition to the major nutrient elements, several studies have indicated that proline is mainly accumulated in salinetolerant halophilic plants, which elicits common stress response like phenolic biosynthesis and stimulation of antioxidant and hydroxyl radical scavenging activities (Sakthivel and Devi, 2015). Therefore, evaluating the proline content of the algae will be an added advantage in the face of therapeutics. In this study, the proline content of S. trinodis and P. myrica was observed 11.32 ± 0.64 µmol g⁻¹ and 5.64 \pm 0.49 µmol g⁻¹ DW, respectively. There was a significant difference between samples, statistically (Table 5). Investigation of amino acids profile of S. trinodis and P. myrica revealed that both algae contain all the essential amino acids. There was a significant difference between amino acids of these brown algae, statistically (Table 6). Overall, the value of amino acids in S. trinodis was more than P. myrica. In genral, the purpose of this study was to determine and compare effective compounds in two algal (S. trinodis and P. myrica) species identified in Qeshm Island. So that, their results can gives some new insight for the nutraceutical industry. According to the data of this study, S. trinodis species are presented as the best algae in terms of nutritional value.

Acknowledgements

The support provided by Gorgan University of Agricultural Sciences and Natural Resources (Gorgan, Iran), Marine Biotechnology Center (Qeshm Island, the Persian Gulf) is truly acknowledged.

References

- Akhtar, P. and Sultana, V., 2002. Biochemical studies of some sea weed species from Karachi coast. *Zoological Survey*, 14, 1-4.
- AOAC., 2005. Official methods of analysis. Washington DC. USA, Horowitz. 2200pp.
- Bates, L., 1973. Rapid determination of free proline for water-stress studies. *Journal of Plant Soil*, 39, 205-207.
- Dadollahi Sohrab, A., Geravand Karimi, M. and Emadabadi, A., 2012. Investigation of seasonal changes in distribution and biomass content of major algae in tidal shores of Bushehr Province (North Coast of Persian Gulf). *Oceanography*, 9, 17-26.
- Dawczynski, Ch., Schubert, R. and Jahreis, G., 2007. Amino acids, fatty acids, and dietary fiber in edible seaweed products. *Journal of Food Chemistry*, 103, 891-899.
- Christaki, E., Bonos, E., Giannenas,
 I. and Florou-Paneri, P., 2013.
 Functional properties of carotenoids originating from algae. *Journal of the Science of Food and Agriculture*, 93, 5–11.
- Cofrades, S., Lopez-Lopez, I., Bravo, L., Ruiz-Capillas, C., Bastida, S., Larrea, M.T. and Jimenez-Colmenero, F., 2010. Nutritional

and antioxidant properties of different brown and red Spanish edible seaweeds. *Food Science and Technology International*, 10, 1-10.

- Dere, S., Gunes, T. and Sivaci, R., 1998. Spectrophotometric Determination of Chlorophyll - A, B and Total Carotenoid Contents of Some Algae Species Using Different Solvents. *Turkish Journal of Botany*, 22, 13-17.
- Fleurence, J., 1999. Seaweed proteins: biochemical, nutritional aspects and potential uses. *Trends in Food Science and Technology*, 10, 25-28.
- Gratzfeld-Huesgen, A., 1999. Sensitive and reliable amino acids analysis in protein hydrolisates using the Agilent 1100 Series HPLC. Technical Note by Agilent Technologies. Publication Number. pp. 5968–5658E.
- Gómez, E., Jiménez, A. and Rupérez, P., 2010. Dietary fiber and physicochemical properties of several edible seaweeds from the northwestern Spanish coast. *Journal of Food Research International*, 43, 2289-2294.
- Guidel-Urbano, M. and Goni, I., 2002. Effect of edible seaweeds (*Undaria pinnitafida* and *Porphyra tenera*) on the metabolic activities of intestinal microflora in rats. *Nutrition Research*, 22, 323–331.
- Hsu, C.Y., Chao, P.Y., Hu, S.P. and Yang, C.M., 2013. The antioxidant and free radical scavenging activities of chlorophylls and pheophytins. *Journal of Food and Nutrition Sciences*, 4, 1-8.

- Inanc, A.L., 2011. Chlorophyll: Structural properties, health benefits and its occurrence in virgin olive oils. *Journal of Akademik Guda*, 9, 26-32.
- Jeffrey, S.W. and Humphrey, G.F., 1975. New spectrophotometric equations for determining chlorophylls a, b, C₁ and C₂ in higher plants, algae and natural phytoplankton. *Journal of Biochemie physiol Pflanzen*, 167, 191-194.
- Kraan,S.,2012.AlgalPolysaccharides, Novel Applicationsand Outlook.Chapter 22,licenseeInTech.pp.489-532.DOI:org/10.5772/51572.
- Lahaye, M., 1991. Marine algae as sources of fibers: Determination of soluble and insoluble dietary fiber contents in some, sea vegetables. *Journal of Science and Food Agriculture*, 54, 587–594.
- Marinho-Soriano, E., Fonseca, P.C., Carneiro, M.A.A. and Moreira, W.S.C., 2006. Seasonal variation in the chemical composition of two tropical seaweeds. *Bioresource Technology*, 97, 2402–2406.
- Matanjun, P., Mohamed, S., Mustapha, N.M. and Muhammad, K., 2009. Nutrient content of tropical edible seaweeds, *Eucheuma cottonii*, *Caulerpa lentillifera* and *Sargassum polycystum. Journal of Apply Phycology*, 21, 75–80.
- Murck, H., 2002. Magnesium and affective disorders. *Nutritional Neuroscience: An International Journal on Nutrition, Diet and Nervous System*, 5, 375-389.

- Ordu^{na-Rojas, J., Robledo D. and} Dawes C.J., 2002. Studies on the tropical agarophyte Gracilaria Journal cornea. of Agardh (Rhodophyta, Gracilariales) from Seasonal Yucat'an, Mexico. Ι. **Physiological Biochemical** and Responses; Botanical Marine, 45, 453-458.
- Oregon State University. 2017. Linus Pauling Institute, Micronutrient Information Center. Corvallis, Oregon.

http://lpi.oregonstate.edu/mic/minerals.

- Ragonese, C., Tedone, L., Beccaria, М., **G.**, Cichello, Torre, F. Cacciola, F., Dugo, P. and 2014. Mondello, L., Characterisation of lipid fraction of marine macroalgae by means of chromatography techniques coupled to mass spectrometry. Journal of Food Chemistry, 145, 932-940.
- Ritchie, R.J., 2006. Consistent sets of spectrophotometric chlorophyll equations for acetone, methanol and ethanol solvents. *Photosynthesis Research*, 89, 27-41.
- Rossa, M.M., Oliveira, M.C., Okamoto, O.K., Lopes, P.F. and ecoleicolo, P., 2002. Effect of visible light on super oxidase dismutase SOD activity in the red Alga *Gracilariopsis* tenuifrons (Gracilarials, Rhodophyta). Jornal of Applied Phycology, 14, 151-157.
- Ruperez, P. and Saura-Calixto, F., 2001. Dietary fibre and physicochemical properties of edible Spanish seaweeds. *European Food Research Technology*, 212, 349–354.

- Ruperez, P., 2002. Mineral content of edible marine seaweeds. *Journal of Food Chemistry*, 79, 23-26.
- Sakthivel, R. and Devi, P.K., 2015. Evaluation of physicochemical properties, proximate and nutritional composition of *Gracilaria edulis* collected from Palk Bay. *Journal of Food Chemistry*, 174, 68-74.
- Sartory, D.P. and Grobbelaas, J.U., 1984. Extraction of chlorophyll a from freshwater phytoplankton for spectrophotometric analysis. *Journal of Hydrobiologia*, 114, 177-187.
- Silva, G., Pereira, R.B., Valentão, P.,
 Andrade, P.B. and Sousa, C., 2013.
 Distinct fatty acid profile of ten
 brown macroalgae. *Revista Brasileira de Farmacognosia Brazilian Journal of Pharmacognosy*, 23(4), 608-613.
- Sohrabipour, J. and Rabiei, R., 2017. Algal vegetation in southern coastline of Iran. *Iran Nature*, 1, 62-68.
- Sultan, H.T., Noroozi, M. and Amozegar, M.A., 2016. Determination of chlorophylls a, b, total carotenoids and antioxidant activity of four green algae isolated from the coast of Golestan of the Caspian Sea. Journal of Cellular Biotechnology – Molecular, 6, 31-36.
- Wellburn, A.R., 1994. The spectral determination of chlorophylls a and b, as well as total carotenoids, using various solvents with spectrophotometers of different resolution. *Journal of Plant Physiology*, 144, 307-313.

- Yaich, H., Garna, H., Besbes, S.,
 Paquot, M., Blecker, C. and Attia,
 H., 2011. Chemical composition and functional properties of *Ulva lactuca* seaweed collected in Tunisia. *Journal of Food Chemistry*, 128, 895-901.
- Yayli, N., Kiran, Z., Seymen, H. and Genc, H., 2001. Characterization of lipids and fatty acid methyl ester contents in leaves and roots of Crocus vallicola. *Turkey Journal of Chemistry*, 25, 391-395.