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Investigation of Seasonal Variations in Biochemical Composition of some Red Algae Distributed in the Strait of Çanakkale (Dardanelles), Turkey

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ABSTRACT

The utilization of marine resources has become very important nowadays due to the rapid increase of world population and raw material requirements. The food industry is facing new challenges in food supply for human consumption, seeking new resources as alternatives to terrestrial foods. Marine based food resources are promising candidates for this new challenge. As a protein source for human consumption, terrestrial animal production alone may not be sufficient as a food supply for the world population. Nowadays, studies on the utilization of marine algae as a food resource for human consumption have become an important topic in human nutrition. As an important source of protein, marine algae are not only used as a food for human but also effectively used in a variety of fields from fertilizers to industrial products. In the present study, temporal and spatial changes of the chemical compositions of some red algae distributed in the Strait of Çanakkale (Dardanelles) were investigated. The biochemical analyses (lipid, ash, protein analyses) were carried out seasonally (fall, winter, spring and summer) in duplicate. Significant differences were recorded in results obtained for the species collected in relation to the seasons and stations. In the present study % lipid levels are at minimum stage. The highest lipid level (3.13 ± 0.98%) is for G. acicularis taksonunda from Eceabat in autumn. The miniumum lipid level (0.36 ± 0.72%) is for C. rubrum from Gelibolu in winter. The highest protein level is for C. ciliatum in spring in Lapseki (24.96 ± 0.23%). The minimum protein level for all species is 2.54 ± 0.76% for J. rubens from İntepe in winter.

Keywords: Rhodophyta, Chemical components, Dardanelles, Seasonal variation

INTRODUCTION

Seaweed is considered as a significant source of many nutritional factors such as proteins, vitamins, and minerals. The huge diversity of seaweed species is the reason for its different chemical composition. In fact, the great extent of secondary seaweed metabolites is formed as an ecological response. Seaweeds are water-living organisms, which are exposed to ultraviolet radiation and should have effective protection from the effect of free radicals. Seaweed polyphenols are formed also as defense mechanism against herbivores and to reinforce seaweed tissue against wave exposure. Bioactivity of diverse secondary metabolites and other compounds extracted from different seaweed species is an important topic of numerous scientific studies. Seaweed contribution to prevention of different serious diseases including cancer and cardiovascular diseases has been confirmed. The antioxidant activity of different seaweed extract and possibility of its utilization as effective protective agents against harmful effects of free radicals have been studied extensively.

Studies on chemical composition of algae began in the 1900's and there are numerous studies in this field today. In earlier studies the possible utilization of algae as a food source has been reported with special reference to their high protein contents comparable to the terrestrial product [1-3].

In many countries the utilization of algae is increasing with the outcomes of new studies and reports on their nutritional composition and advantages as a functional food source. The production, marketing and consumption of algae have shown a significant increase in countries such as China, Japan, Korea and France [4].

Furthermore, the high levels of protein, vitamins, amino acids, minerals and the low level of fat in algae has brought

this food source to a higher rank after fish as an health food product for human consumption [5]. Marine based food resources are considered as important alternatives for the food supply of the increasing world population for the future [6].

Marine algae are considered as important resources in marine environment; hence various studies in terms of their utilization have been conducted for many years. The industrial utilization of algae began with soda and iodine production and continued with the production of organic materials such as alginate, carrageen or carrageenan [7].

In the far east countries especially in China, Japan and Korea, great majority of algae are consumes because of their valuable nutrients [8]. For instance in Japan, capitation of algae per year is 1.6 kg [9].

The economic inputs and social benefits in new employments in algae production especially in Asian countries has attracted other countries with marine costal zones. As an important food source with high level of protein contents, and their potential use in various fields attracts scientists and recently there is an increase of the utilization of using algae in the industry.

In this study materials are 10 different macro algae from Rhodophyta collected. Samplings made seasonally at seven localities. The aim of the present study is to determine the essential ingredients of some red algae.

Hence the results of the present study will give us important data for determining the best season and the location to benefit from algae for optimum yield in the Çanakkale Strait.

MATERIALS AND METHODS

In this study materials are 10 different macro algae from Rhodophyta. These materials are *Ceramium ciliatum var. ciliatum* (J. Ellis) Ducluzeau, *Ceramium rubrum var. rubrum* C. Agardh, *Corallina officinalis Schnetter & U. Richter*, *Jania rubens* (Linnaeus) J. V. Lamoroux *var. rubens*, *Gelidium spinosum* (S. G. Gmellin) P. C. Silva, *Gracilaria gracilis* (Stackhouse) Steentoft, Irvine & Farnham *var. gracilis*, *G. bursa-pastoris* (Gmelin) Silva, *Gigartina acicularis* (Roth) Fredericq, *Chondria dasphylla* (Woodward) C. Agardh, *Phyllophora crispa* (Hudson) *Dixon f. crispa*. Samplings made seasonally at seven localities (Gelibolu, Eceabat, Havuzlar, İntepe, Yapıldak, Çanakkale and Lapseki) along Çanakkale Strait (40º02'-40º30' N, 26º10'-26º45'E) between September 2007 and June 2008 (Figure 1).

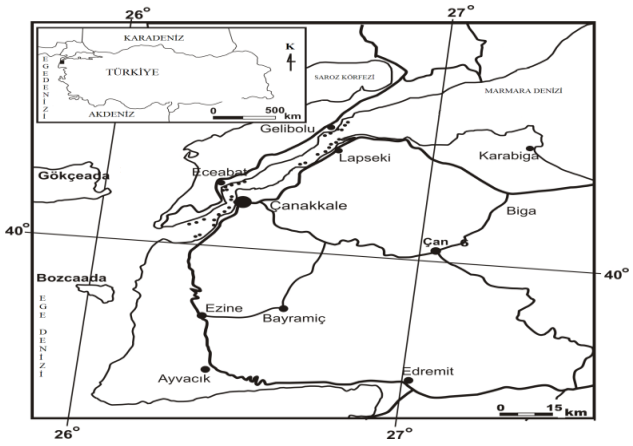

Figure 1: Location of sampling sites in the Çanakkale Strait (Dardanelles).

Collected samples were separated from epiphytes and carefully washed with tap water. Examples was allowed to dry naturally between 7-10 days. Afterwards the samples were dried in a fume hood at 70ºC to a constant weight. The dried samples were powdered using a rotatory grinder. These samples were then used for nutritional analyses such as crude protein, crude lipid and crude ash contents. Lipid analyses were conducted according to Morales et al., [10] protein analyses were carried out according to the Kjeldahl method and ash were analyzed in duplicates according to AOAC [11].

Dried materials were calculated by standard methods in duplicates. The tares weight of porcelain crucibles were measured. 0.5 g material was put in porcelain crucibles at 525ºC for 12 hours. Then the porcelain crucibles were scaled by assay balance. The amount of ash was measured by the formula below.

Crude ash amount (%)=(tr-tf) / m × 100 (tr: recent rhythm, ti: first rhythm, m: sample weight)

Lipid analyses were carried out with the method of Folch[12]. 0.5 g material was measured and put in volumetric flask. Then 10 ml 2:1 methanol- chloroform solution was added. The samples were standed up for 24 hours at room temperature. Samples were leached and put in evaporator at 60ºC. Than volumetric flask was put in stove at 103ºC for 2 hours. Volumetric flasks were scaled by assay balance. The amount of lipid was measured by the formula below.

Crude lipid amount (%)={(tr-tv)/m} × 100 (m=sample weight, tv=first weight of volumetric flask, tr=recent weight of volumetric flask and the weight of lipid)

Protein analyses were carried out with the method of Kjeldahl [11]. 0.5 g material was measured and put in Kjeldahl tube. One piece of Kjeldahl tablet was put in each Kjeldahl tube as catalyzer. Then 15 ml 96% H_2SO_4 was added in each tube. Afterwards each tube was put in wet decomposition for 2 hours. After 2 hours samples were put to distillation. At the end of the distillation samples were standardized with HCl. The amount of protein was measured by the formula below.

$$\text{Crude protein amount (\%)} = \frac{(tt - tk)14.007 \times 6.25}{\text{m}} \times 100$$

(tt: amount used in titration, tk: amount used in titration of blank sample, m: sample weight)

Nitrogen free extracts were calculated with deduction of nutritional fractions from hundred. The amount of NFE was calculated by the formula below.

Nitrogen free extracts (NFE-%)=100 – (protein amount+ash amount+lipid amonut)

RESULTS AND DISCUSSION

The true value of nitrogen-to-protein conversion factor should be determined for each seaweed genus from the total nitrogen content based on amino acid composition and the distribution of nitrogen in protein and in other nonprotein nitrogen compounds [13-16]. In different genera of green, brown, and red seaweed the values of nitrogen-to-protein conversion factor have been provided. The average value of the nitrogen-to-protein conversion factor is 5.13 for green algae, 5.38 for brown algae and 4.92 for red algae [17]. In this study 10 different species were collected from different locations from Çanakkale Strait in different seasons. Numeric datas are shown in Table 1. The chemical composition of each samples are different from each other for each location and season (Table 1). Also the annual avarage results in chemical composition of some samples were shown in Figures 2-7.

The results for *G. bursa pastoris* are shown in Table 1. According to the results, in autumn in Gelibolu 10.18 ± 0.92% protein, 1.59 ± 0.54% lipid, 56.09 ± 0.22% ash and 32.14 ± 0.98% nitrogen free extract. (NFE), in Yapıldak 7.86 ± 0.28% protein, 1.39 ± 0.20% lipid, 41.78 ± 0.73% ash and 48.97 ± 0.18% NFE were recorded. *G. bursa pastoris* was colleceted in winter from Havuzlar. The results for this station; 15.9 ± 0.61% protein, 1.87 ± 0.66% lipid, 31.28 ± 0.93% ash and 50.95 ± 0.28% NFE. In summer in Lapseki 9.56 ± 0.18% protein, 2.15 ± 0.42% lipid, 45.18 ± 0.48% ash and 43.11 ± 0.59% NFE were recorded.

G. spinosum was collected from Gelibolu in autumn. These results were; 14.34 ± 0.64% protein, 2.08 ± 0.78% lipid, 35.55 ± 0.14% ash and 48.03 ± 0.64% NFE. The results for *G. acicularis* are shown in Table 1. In autumn in Gelibolu 11.48 ± 0.45% protein, 2.07 ± 0.86 % lipid, 33.34 ± 0.32% ash and 53.11 ± 0.54% NFE, in Eceabat 12.46 ± 0.72% protein, 3.13 ± 0.98% lipid, 39.99 ± 0.64% ash and 44.42 ± 0.42% NFE were recorded. In autumn in Çanakkale 13.3 ± 0.39% protein, 2.34 ± 0.17% lipid, 48.1 ± 0.22% ash and 36.26 ± 0.76% NFE, in Lâpseki 13 ± 0.88% protein, 1.85 ± 0.62%l ipid, 46.01 ± 0.54% ash and 39.14 ± 0.58% NFE were investigated. *C. rubrum* was collected from Gelibolu in winter. 22.72 ± 0.66% protein, 0.36 ± 0.72% lipid, 37.05 ± 0.74% ash and 39.87 ± 0.42% NFE were recorded. For *C. officinalis* in winter from Eceabat 4.01 ± 0.33% protein, 2.49 ± 0.66% lipid, 76.45 ± 0.52% ash and 17.05 ± 0.58% NFE were investigated. In the same season in Lapseki 6.18 ± 0.77% protein, 2.01 ± 0.74% lipid, 76.93 ± 0.88% ash and 14.88 ± 0.64% were recorded. Species were collected from Gelibolu in spring. The results for this location are 6.05 ± 0.64% protein, 2.32 ± 0.76% lipid, 74.04 ± 0.14% ash and 17.59 ± 0.65% NFE. *J. rubens* was collected in summer from Gelibolu. The amount of protein is 5.77 ± 0.88%, the amount of lipid is 1.13 ± 0.34%, the amount of ash is 72.99 ± 0.66% and the amount of NFE is 20.11 ± 0.77%. In winter in İntepe 2.54 ± 0.76% protein, 1.93 ± 0.24% lipid, 78.68 ± 0.92% ash and 16.85 ± 0.54% NFE.

The results for *C. ciliatum* were shown in Table 1. From Eceabat in spring 15.26 ± 0.84% protein, 1.63 ± 0.12% lipid, 44.04 ± 0.76% ash and 39.07 ± 0.22% NFE, in Lapseki 24.96 ± 0.23% protein, 1.04 ± 0.12% lipid, 46.87 ± 0.61% ash and 27.13 ± 0.64% NFE, in İntepe 9.93 ± 0.38% protein, 0.83 ± 0.16% lipid, 44.88 ± 0.86% ash and 44.36 ± 0.24%

Table 1: Seasonal changes in chemical composition of samples in different locations.

	SEASONS	**AUTUMN**				**WINTER**				**SPRING SPRING**						**SUMMER**			
	TAXON	**% Protein**	**% lipid**	**% ash**	**NFE**	**% Protein**	**% lipid**	**% ash**	**NFE**	% Protein	% lipid	% ash	% lipid	% ash	NFE	% Protein	% lipid	% ash	NFE
GELİBOLU	*G. bursa pastoris*	10.18 ± 0.92	1.59 ± 0.54	56.09 ± 0.22	32.14 ± 0.98														
	G. spinosum	14.34 ± 0.64	2.08 ± 0.78	35.55 ± 0.14	48.03 ± 0.64														
	G. acicularis	11.48 ± 0.45	2.07 ± 0.86	33.34 ± 0.32	53.11 ± 0.54														
	C. rubrum					22.72 ± 0.66	0.36 ± 0.12	37.05 ± 0.74	39.87 ± 0.42										
	C. officinalis									6.05 ± 0.64	2.32 ± 0.76	74.04 ± 0,14	17,59 ± 0,65	74,04 ± 0,14	17,59 ± 0,65				
	J. rubens											72.99 ± 0.66				5,77 ± 0,88	1,13 ± 0,34	72,99 ± 0,66	20,11 ± 0,77
ECEABAT	*G. acicularis*	12.46 ± 0.72	3.13 ± 0.98	39.99 ± 0.64	44.42 ± 0.42														
	C. officinalis					4.01 ± 0.33	2.49 ± 0.66	76.45 ± 52	17.05 ± 0.58										
	C. ciliatum									15.26 ± 0.84	1.63 ± 0.12	44,04 ± 0,76	39,07 ± 0,22	44,04 ± 0,76	39,07 ± 0,22				
HAVUZLAR	*G. gracilis*					15.9 ± 0.14	1.95 ± 0.54	31.79 ± 0.22	50.36 ± 0.76										
	C. ciliatum					22.57 ± 0.33	1.47 ± 0.77	47.71 ± 0.19	28.25 ± 0.14										
	G. bursa pastoris					15.9 ± 0.61	1.87 ± 0.66	31.28 ± 0.93	50.95 ± 0.28										

Continued in next page

İNTEPE	*C. ciliatum*					8.03 ± 0.78	1.05 ± 0.76	65.09 ± 0.14	25.83 ± 0.48	9.93 ± 0.38	0.83 ± 0.16	44,88 ± 0,86	44,36 ± 0,24	44,88 ± 0,86	44,36 ± 0,24					
	J. rubens					2.54 ± 0.76	1.93 ± 0.24	78.68 ± 0.92	16.85 ± 0.54											
	C. dasphylla	7.64 ± 0.64	0.6 ± 0.12	48.54 ± 0.14	43.22 ± 0.52															
ÇANAKKALE	*G. acicularis*	13.3 ± 0.39	2.34 ± 0.17	48.1 ± 0.22	36.26 ± 0.76															
	P. crispa									13.34 ± 0.86	1.69 ± 0.54	49,67 ± 0,98	35,3 ± 0,78	49,67 ± 0,98	35,3 ± 0,78					
YAPILDAK	*G. gracilis*					10.61 ± 0.14	0.93 ± 0.10	45.72 ± 0.78	42.74 ± 0.88											
	G. bursa pastoris	7.86 ± 0.28	1.39 ± 0.20	41.78 ± 0.73	48.97 ± 0.18															
LAPSEKİ	*G. acicularis*	13 ± 0.88	1.85 ± 0.62	46.01 ± 0.54	39.14 ± 0.58															
	C. ciliatum									24.96 ± 0.23	1.04 ± 0.12	46,87 ± 0,61	27,13 ± 0,64	46,87 ± 0,61	27,13 ± 0,64					
	C. officinalis					6.18 ± 0.77	2.01 ± 0.74	76.93 ± 0.88	14.88 ± 0.64											
	G. bursa pastoris															9,56 ± 0,18	2,15 ± 0,42	45,18 ± 0,48	43,11 ± 0,59	

NFE were recorded. In winter in Havuzlar 22.57 ± 0.33% protein, 1.47 ± 0.77% lipid, 47.71 ± 0.19% ash and 28.25 ± 0.14% NFE, in İntepe 8.03 ± 0.78% protein, 1.05 ± 0.76% lipid, 65.09 ± 0.14% ash and 25.83 ± 0.48% NFE were investigated. The chemical composition of samples are different from each other for each location and each season as shown in Table 1.

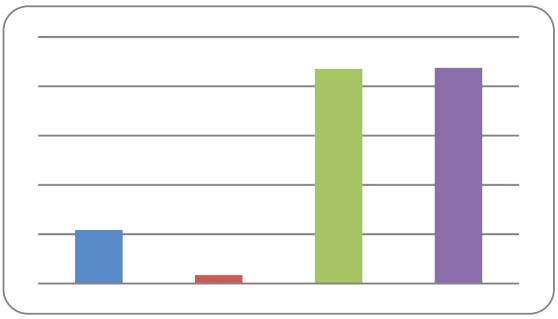

Figure 2: Annual average values for *G. bursa pastoris*

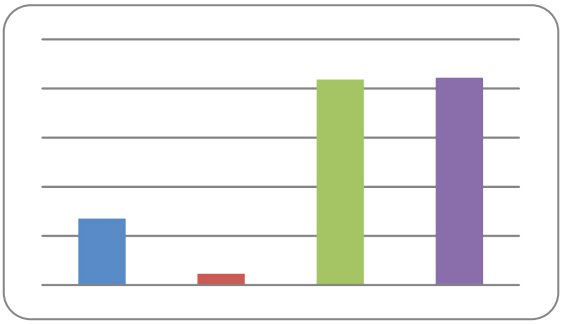

Figure 3: Annual average values for *G. acicularis*

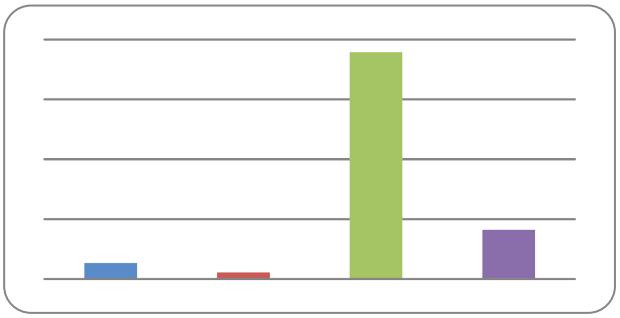

Figure 4: Annual average values for *C. officinalis*

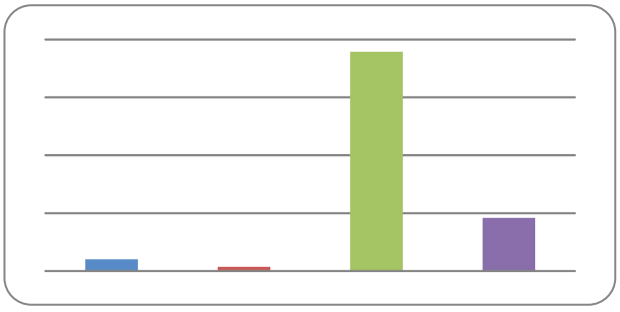

Figure 5: Annual average values for *J. rubens*

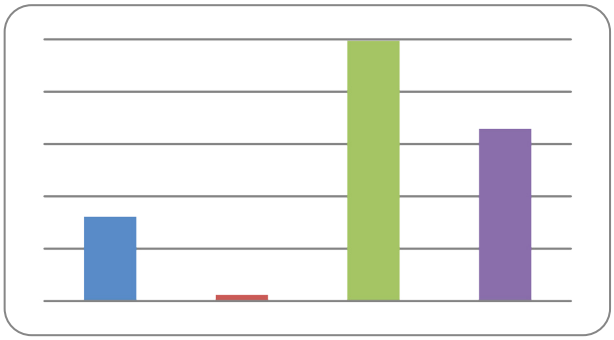

Figure 6: Annual average values for *C. ciliatum*

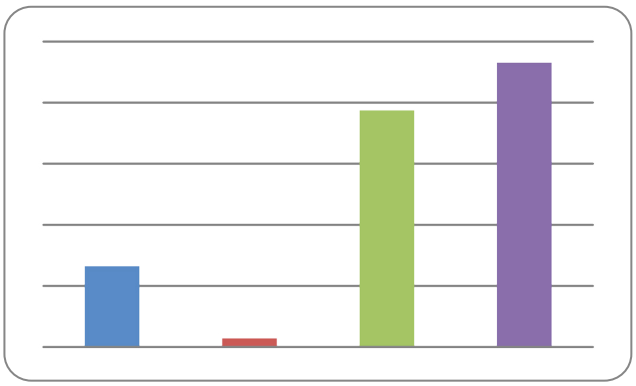

Figure 7: Annual average values for *G. gacilis*

C. dasphylla was collected from İntepe in autumn. The results are 7.64 ± 0.64% protein, 0.6 ± 0.12% lipid, 48.54 ± 0.14% ash and 43.22 ± 0.52% NFE. *P. crispa taksonu* was collected from Çanakkale in spring. The results are 13.34 ± 0.86% protein, 1.69 ± 0.54% lipid, 49.67 ± 0.98% ash and 35.3 ± 0.78% NFE. The results for *G. gracilis taksonu* in winter from Havuzlar are 15.9 ± 0.14% protein, 1.95 ± 0.54% lipid, 31.79 ± 0.22% ash and 50.36 ± 0.76% NFE. In the same season in Yapıldak 10.61 ± 0.14% protein, 0.93 ± 0.10% lipid, 45.72 ± 0.78% ash and 42.74 ± 0.88% NFE were recorded.

In the present study 10 different species from Rhodophyta were investigated for about their seasonal chemical variation in the Çanakkale Strait. The protein levels of different species changed for each location and season. percentage ash and lipid contents of the species are similar to each other. The important values of species can be put it this way below. The highest protein level is for *C. ciliatum* in spring in Lapseki (24.96 ± 0.23%). In turn the protein level of *C. rubrum* in Gelibolu in winter is 22.72 ± 0.66%, for *G. bursa pastoris* in winter in Havuzlar is 15.9 ± 0.61% and for *G. gracilis* in winter in Havuzlar is 15.9 ± 0.14%. The protein level of the same species is close to each other. The minimum protein level for all species is 2.54 ± 0.76% for *J. rubens* from İntepe in winter. The protein levels of species are at good level and similar to the results determined before. The protein levels of Ochrophyta in summer are at low stage (7-16 gr/100), the protein levels in summer for Rhodophyta is higher (21-40 gr/100) [18,19] investigated the chemical composition of *Gracilaria cervicornis* (Turner) *J. agardh* and *S. vulgare* C. Agardh. The protein level is determined between 15.97 - 23.05%. The highest protein level is for *G. cervicornis*. Rhodophyta and Chlorophyta species have higher protein than other species. The chemical composition of seaweed provides their high nutritional value contributing to human nutrients – such as proteins with all essential amino acids, minerals and vitamins. In addition, they consist of bioactive secondary metabolites and many different compounds with health benefits [20-25]. For this reason many researcher uphold Rhodophyta and Chlorophyta species should be used as protein resource [26]. McDermid and Stuerckke [27] investigated protein, lipid, carbonhydrates, ash, mineral and vitamine contents of 22 macro algae. They determined high level protein for *Halymenia formosa* Harvey ex Kützing and *Porphyra vietnamensis* T. Tanka and PhamHoang Ho. Many of the species contain less than 5% raw lipid. The lipid level of Algae is less than other marine products but the fatty acid levels of Algae are higher than other marine products [28]. In the present study percentage lipid levels are at minimum stage. The highest lipid level (3.13 ± 0.98%) is for *G. acicularis taksonunda* from Eceabat in autumn. The miniumum lipid level (0.36 ± 0.72%) is for *C. rubrum* from Gelibolu in winter.

CONCLUSION

As an important protein source with a wide range of utilization, in recent years algae have attracted researchers more and more. However, knowledge on the seasonal variations in their chemical composition and their availability in different locations are important information for sustainable utilization of algae as high value protein supply for human consumption. Further studies are encouraged on the utilization and production of algae as raw materials or food supplements in different fields. In this study, *C. ciliatum* and *G. spinosum* were identified as algae that can be considered as food supplements with high protein content.

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