



## Seasonal changes in the fatty acid profile of *Cystoseira crinita* Duby, 1830, distributed on the Sinop Peninsula Coast of the Black Sea

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### Article History

Received: 18.02.2023

Accepted: 05.07.2023

Published: 20.12.2023

### Research Article

**Abstract** – This study aimed to determine the fatty acids profile and seasonal change in *Cystoseira crinita* Duby, 1830 from the Sinop Peninsula coasts. The fatty acids profile was analyzed by GC/MS and their seasonal variation was studied. Along the sampling, it was possible to identify 37 different fatty acids in *C. crinita*, from C4 to C22. It was determined that palmitic acid was the most abundant fatty acid in all seasons, and further, the season which provided the highest contents of SFA, PUFA, and MUFA was winter. As a matter of fact, in our study, it was determined that the highest PUFA values ranged from 40.63% in winter to 32.23% in summer. It has been determined that the MUFA value varies between 25.88% in winter and 30.79% in summer, and the SFA value varies between 33.50% in winter and 35.98% in summer. In this study, the PUFA/SFA ratio of *C. crinita* was determined to change between 1.01% - 1.21% from winter to summer. In addition, the total  $\omega$ -6/ $\omega$ -3 PUFA ratio was found to be greater than 1 and ranged from 1.61 (winter) to 2.07 (summer). The atherogenicity and thrombogenicity index and h/H ratio were calculated from the fatty acid profiles of *C. crinita*, and the AI index was determined to change from 0.71 (winter) to 0.74 (autumn), TI index was 0.44 (winter) to 0.58 (in summer). The h/H ratio of 1.71 (summer) to 2.00 (winter) was calculated. These results of our study showed that the seasons have a significant effect on the fatty acid profile and the fatty acids in *C. crinita* may have important contributions to human nutrition. For this reasons, it is thought that it is extremely important to reveal the nutritional content of different seaweed species that spread in the seas of Turkey and to observe the seasonal changes in their contents.

**Keywords** – Atherogenicity index, Black Sea, fatty acid, macroalgae, thrombogenicity index

## 1. Introduction

As marine primary producers, macroalgae are among the rich sources of lipids for the growth and reproduction of marine organisms (Ivanova, Stancheva, & Petrova, 2013; Schram, Kobelt, Dethier, & Galloway, 2018). Fatty acids (especially PUFA), which are transferred from macroalgae to fish and even humans through the food chain in marine ecosystems, are among the important nutrients (Sijtsma & de Swaaf, 2004; Filimonova, Goncalves, Marques, Trochc, & Goncalves, 2016; Caf, Özdemir, Yılmaz, Durucan, & Ak, 2019). However, it is stated that alternative sources may be needed as a source of PUFA due to the uncertainty in future fish stocks, and it is suggested that seaweeds can be used as a new source of PUFA (Dawczynski et al., 2007; Polat & Ozoguz, 2013; Vizetto-Duarte et al., 2015; Belattmania et al., 2018).

Macroalgae, particularly brown, are a part of the main diet of many countries (Dawczynski, Schubert, & Jahreis, 2007; Miyashita et al., 2012; Muradian, Vaiserman, Min, & Fraifeld, 2015), and different ratios of polysaccharides, antioxidants, vitamins, minerals, proteins, and lipids that contain (Schmid et al., 2018; Nunes, Valente, Ferraz, Barreto, & Carvalho, 2020; Al-Adilah et al., 2021). In addition, it has been suggested that brown seaweeds have great amounts of PUFA, which are not present in land plants (Kumari, Bijo, Mantri,

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Reddy, & Jha, 2013; Polat & Ozogul, 2013; Schmid et al., 2018; Al-Adilah et al., 2021). Seaweeds have contained a significant amount of essential fatty acids, despite the low lipid concentration (Rocha et al., 2021), they can be used as an alternative to a marine lipid resource due to brown seaweeds abundance among coastal algae (Airanthi et al., 2011).

Although macroalgae are very diverse in their amount of nutrients (Dawczynski et al., 2007), it is reported that their contents are affected by geographical situation, sampling terms, environment, season, temperature, salinity, and light intensity (Nelson, Phleger, & Nichols, 2002; Polat & Ozoğuz, 2013; Silva, Pereira, Valentao, Andrade, & Sousa, 2013). Therefore, it is thought that it is important to determine the seasonal changes in the nutrient content in order to expand the use of brown seaweed as a total source of SFA (saturated fatty acid), MUFA (monounsaturated fatty acid) and PUFA (polyunsaturated fatty acid). On the other hand, there is little study about the substance of fatty acids of macroalgae in Turkey (Polat & Ozogul, 2008; Yazıcı et al., 2008; Caf, Yılmaz, Durucan, & Özdemir, 2015; Caf et al., 2019; Aras & Sayın, 2020). *Cystoseira crinita* from the order Fucales (Ochrophyta, Phaeophyceae) is one of the most common species of the Sinop coast (Karacuha & Ersoy-Karacuha, 2013), but no information with these species on seasonal variations in fatty acid content has been published.

The aim of this study that to define the seasonal changes in the fatty acid profile of *C. crinita*, which is widely distributed in the coastal waters of Sinop province.

## 2. Materials and Methods

Samples were collected seasonally in 2014 by scuba diving from various rocky substrates at 0 and 1 m depths of the Sinop Peninsula coast (Figure 1). The sample was then transferred to the laboratory, separated from foreign materials such as stone and sand, and dried on blotting paper after washing with distilled water. The dried samples were pounded into powder and stored at  $-20^{\circ}\text{C}$ . 0.5 grams of each sample were used for fatty acid analysis. For taxonomic classification of the species, [www.algaebase.org](http://www.algaebase.org) was consulted.

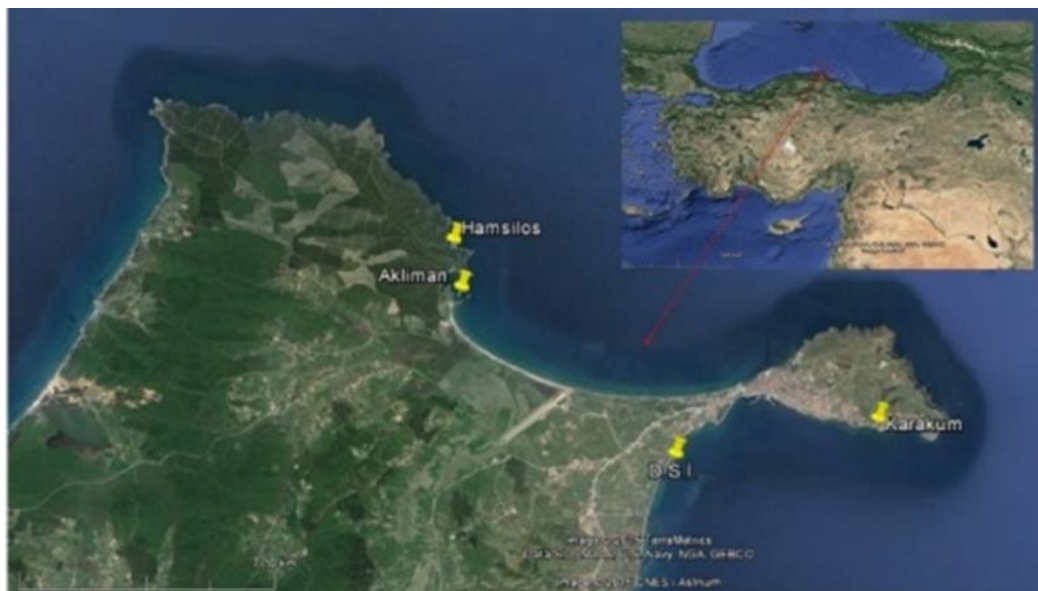


Figure 1. Sampling area (Google Maps).

### 2.1. Lipid extraction

Lipid extracts were prepared according to Bligh & Dyer (1959), using a mixture of chloroform and methanol. For this process, lipid samples were derivatized into methyl esters in a gas chromatography device (Thermo Scientific Trace 1310). In this process, 0.25 g of the extracted oil was dissolved by adding 4 ml heptane, after that 0.4 ml 2 N KOH was added. This mixture was then vortexed for 2 minutes followed by centrifugation at 5000 rpm for 5 minutes. After centrifugation, 1.5-2 ml of clear heptane phase was taken and transferred to glass tubes and GC/MS analysis was performed. Samples were injected into the device with the help of the autosampler (Autosampler AI 1310).

## 2.2. Fatty acid profile

The extracted algal lipid was then transesterified to fatty acid methyl esters (FAME) according to Ichihara et al. (1996). Samples were analyzed GC/MS gas chromatography-mass spectrometry (Thermo Scientific ISQ LT model). A 60 m long Trace Gold TG-WaxMS capillary column (Thermo Scientific code: 26088-1540) with an inner diameter of 0.25  $\mu\text{m}$  and a film thickness of 0.25  $\mu\text{m}$  was used for this analysis. The temperature of the injection block for the analysis was set to 240°C. In addition, the column temperature was programmed to remain constant at 100 °C for 3 minutes, and thereafter increase to 240 °C in increments of 4 °C/min. A separation ratio of 1:20 was applied using constant-flow (1 ml/min) helium gas as the carrier gas. Fatty acids were identified by comparing the standard FAME (fatty acid methyl ester) mixture of 37 components according to their arrival time. All analyses of fatty acids were performed in triplicate per biological sample during the sampling period. The results were given as mean $\pm$ standard deviation.

## 2.3. Lipid quality

Ulbricht & Southgate (1991) method to health lipid indices (AI and TI) to determine the nutritional and lipid quality of *C. crinita* was used.

The formula for calculating the Index of Atherogenicity (AI) is:

$$AI = \frac{C12:0 + (4 \times C14:0) + C16:0}{\sum(w-3 \text{ PUFA}) + \sum(w-6) \text{ PUFA} + \sum \text{MUFA}} \quad (2.1)$$

The formula for calculating the index of thrombogenicity (IT) is:

$$TI = \frac{C14:0 + C16:0 + C18:0}{(0.5 \times \sum \text{MUFA}) + (0.5 \times \sum w-6 \text{ PUFA}) + (3 \times \sum w-3 \text{ PUFA}) + (w-3/w-6)} \quad (2.2)$$

where PUFA and MUFA indicate monounsaturated and polyunsaturated FA.

The hypocholesterolemic/hypercholesterolemic (h/H) index is calculated to Santos-Silva, Bessa, & Santos-Silva (2002) method. The formula for calculating the Index of h/H is:

$$HH = \frac{(C18:1 + \sum \text{PUFA})}{(C14:0 + C16:0)} \quad (2.3)$$

## 2.4. Statistical analysis

Differences between seasonal average were evaluated using one-way ANOVA, followed by Tukey test ( $P < 0.05$ ). The contribution (percentage) of each fatty acid was taken into account during the calculations. Principal component analysis (PCA) was performed for sampling periods and fatty acid major classes (MUFA, SFA, and PUFA). All calculations were made with SPSS version 22.0.

## 3. Results and Discussion

In this study, the fatty acid profiles of *C. crinita* were analyzed by GC/MS, seasonally. It identified 37 different fatty acids from *C. crinita*, and are given in Table 1. It was determined that there were statistically significant differences in the fatty acid profile of *C. crinita* between seasons (Table 1). In this study, C16:0 (palmitic acid; from SFA) was determined to be the major fatty acid, followed by C18:1 $\omega$ 9c (elaidic acid; from MUFA), C20:4 $\omega$ 6 (arachidonic acid: ARA; from PUFA), C18:2 $\omega$ 6c (linoleic acid: LIN; from PUFA), C18:3 $\omega$ 3 ( $\alpha$ -linolenic acid: ALA; from PUFA), C20:5 $\omega$ 3c (eicosapentaenoic acid: EPA from PUFA), palmitoleic acid (C16:1; from MUFA), and oleic acid (C18:1 $\omega$ 9t; from MUFA), respectively. Palmitic acid was the dominant fatty acid during the study, with values ranging from 25.89 $\pm$ 0.99% total in summer to 24.32 $\pm$ 0.19% in winter. Similar to our finding, palmitic acid is reported to be the primary saturated fatty acid in several seaweeds (Kamenarska et al., 2002; Ivanova et al., 2013). Elaidic acid and oleic acid were the most abundant MUFAs in our study. Similarly, oleic acid is reported by Vizetto-Duarte et al. (2015) as one of the main fatty acids in brown seaweed, too. Besides, Kamenarska et al. (2002) reported that the main fatty acid for *C. crinita* from Eastern Mediterranean was palmitic acid, and then oleic and myristic acids. On the other hand, we determined that the total of PUFA was highest, followed by SFA and MUFA, respectively. (Table 1). PUFA was the most abundant component with 37.24% seasonal mean of total fatty acid. The seasonal mean of MUFA was 27.79%, and SFAs were 34.46%. Although this study results are concordant with the literatures

(Kumari et al., 2013), it has been found to differ from studies with *C. crinita*, which has the highest SFA values (Ivanova et al., 2013; Bouafif, Messaoud, Boussaid, & Langar, 2018) (Table 2).

Table 1

Seasonal change of fatty acid profile of *C. crinita* (percentage of the total FAME) with the significant differences between the seasons with one-way ANOVA and the Tukey test ( $p < 0.05$ ), and the result of indexes. (n=3).

Fatty acid (%)	SEASONS					F	Sig.
	Spring	Summer	Autumn	Winter			
C4:0	0.17±0.00 <sup>a</sup>	0.14±0.02 <sup>b</sup>	0.07±0.01 <sup>c</sup>	0.08±0.00 <sup>c</sup>	86.500	0.000	
C6:0	0.04±0.01 <sup>a</sup>	0.02±0.00 <sup>b</sup>	0.03±0.00 <sup>ab</sup>	0.02±0.01 <sup>b</sup>	9.833	0.005	
C8:0	0.05±0.00 <sup>a</sup>	0.02±0.01 <sup>b</sup>	0.04±0.01 <sup>a</sup>	0.01±0.01 <sup>b</sup>	35.000	0.000	
C10:0	0.02±0.01 <sup>a</sup>	0.01±0.01 <sup>a</sup>	0.02±0.01 <sup>a</sup>	0.01±0.00 <sup>a</sup>	1.222	0.363	
C11:0	0.02±0.00 <sup>b</sup>	0.01±0.01 <sup>c</sup>	0.04±0.00 <sup>a</sup>	0.01±0.01 <sup>c</sup>	44.667	0.000	
C12:0	0.03±0.00 <sup>ab</sup>	0.04±0.00 <sup>a</sup>	0.04±0.01 <sup>a</sup>	0.02±0.01 <sup>b</sup>	9.833	0.005	
C13:0	0.03±0.01 <sup>a</sup>	0.02±0.00 <sup>a</sup>	0.02±0.00 <sup>a</sup>	0.02±0.00 <sup>a</sup>	4.000	0.052	
C14:0	6.68±0.08 <sup>a</sup>	6.20±0.13 <sup>b</sup>	5.96±0.02 <sup>c</sup>	5.75±0.04 <sup>d</sup>	74.644	0.000	
C15:0	0.40±0.00 <sup>ab</sup>	0.39±0.01 <sup>b</sup>	0.40±0.01 <sup>ab</sup>	0.42±0.01 <sup>a</sup>	6.000	0.019	
C16:0	24.88±0.09 <sup>ab</sup>	25.89±0.99 <sup>a</sup>	24.64±0.07 <sup>ab</sup>	24.32±0.19 <sup>b</sup>	5.363	0.026	
C17:0	0.08±0.02 <sup>a</sup>	0.07±0.02 <sup>a</sup>	0.07±0.00 <sup>a</sup>	0.06±0.01 <sup>a</sup>	0.556	0.659	
C18:0	2.14±0.02 <sup>b</sup>	2.11±0.06 <sup>b</sup>	2.26±0.03 <sup>a</sup>	1.63±0.03 <sup>c</sup>	159.615	0.000	
C20:0	0.01±0.01 <sup>a</sup>	0.02±0.01 <sup>a</sup>	0.01±0.01 <sup>a</sup>	0.01±0.01 <sup>a</sup>	0.501	0.693	
C21:0	0.06±0.01 <sup>b</sup>	0.11±0.02 <sup>a</sup>	0.07±0.00 <sup>b</sup>	0.06±0.01 <sup>b</sup>	17.212	0.001	
C22:0	0.40±0.01 <sup>c</sup>	0.40±0.04 <sup>c</sup>	0.54±0.01 <sup>a</sup>	0.48±0.01 <sup>b</sup>	25.661	0.000	
C23:0	0.06±0.03 <sup>a</sup>	0.05±0.04 <sup>a</sup>	0.04±0.00 <sup>a</sup>	0.05±0.02 <sup>a</sup>	0.410	0.750	
C24:0	0.56±0.03 <sup>a</sup>	0.49±0.16 <sup>a</sup>	0.54±0.04 <sup>a</sup>	0.54±0.02 <sup>a</sup>	0.399	0.758	
<b>ΣSFA</b>	<b>35.61±0.11<sup>ab</sup></b>	<b>35.98±0.76<sup>a</sup></b>	<b>34.77±0.10<sup>b</sup></b>	<b>33.50±0.28<sup>c</sup></b>	<b>21.634</b>	<b>0.000</b>	
C14:1	0.34±0.01 <sup>a</sup>	0.33±0.02 <sup>a</sup>	0.22±0.00 <sup>b</sup>	0.17±0.00 <sup>c</sup>	188.385	0.000	
C15:1c	0.03±0.01 <sup>b</sup>	0.08±0.02 <sup>a</sup>	0.03±0.01 <sup>b</sup>	0.02±0.00 <sup>b</sup>	31.222	0.000	
C16:1	3.10±0.02 <sup>b</sup>	3.83±0.06 <sup>a</sup>	2.76±0.03 <sup>c</sup>	2.25±0.02 <sup>d</sup>	1033.804	0.000	
C17:1c	0.29±0.02 <sup>a</sup>	0.22±0.00 <sup>c</sup>	0.25±0.01 <sup>b</sup>	0.26±0.00 <sup>b</sup>	34.667	0.000	
C18:1ω9c	20.25±0.02 <sup>b</sup>	21.70±0.43 <sup>a</sup>	19.06±0.12 <sup>c</sup>	19.43±0.05 <sup>c</sup>	83.362	0.000	
C18:1ω9t	2.07±0.04 <sup>a</sup>	2.08±0.05 <sup>a</sup>	1.48±0.05 <sup>b</sup>	1.23±0.06 <sup>c</sup>	222.849	0.000	
C18:2ω6t	0.08±0.07 <sup>b</sup>	0.22±0.02 <sup>a</sup>	0.26±0.00 <sup>a</sup>	0.11±0.00 <sup>b</sup>	18.509	0.001	
C20:1c	0.13±0.01 <sup>c</sup>	0.32±0.03 <sup>a</sup>	0.19±0.02 <sup>b</sup>	0.12±0.02 <sup>c</sup>	89.213	0.000	
C22:1ω9	1.84±0.02 <sup>b</sup>	1.88±0.15 <sup>b</sup>	1.87±0.01 <sup>b</sup>	2.20±0.02 <sup>a</sup>	14.583	0.001	
C24:1	0.16±0.04 <sup>a</sup>	0.12±0.05 <sup>a</sup>	0.10±0.02 <sup>a</sup>	0.10±0.02 <sup>a</sup>	1.911	0.206	
<b>ΣMUFA</b>	<b>28.28±0.12<sup>b</sup></b>	<b>30.79±0.22<sup>a</sup></b>	<b>26.22±0.18<sup>c</sup></b>	<b>25.88±0.10<sup>c</sup></b>	<b>592.929</b>	<b>0.000</b>	
C18:2ω6c	7.95±0.02 <sup>b</sup>	7.36±0.19 <sup>c</sup>	8.94±0.06 <sup>a</sup>	7.90±0.02 <sup>b</sup>	132.422	0.000	
C18:3ω3	7.92±0.11 <sup>b</sup>	6.75±0.24 <sup>c</sup>	8.66±0.11 <sup>a</sup>	9.01±0.03 <sup>a</sup>	146.735	0.000	
C18:3ω6	0.16±0.03 <sup>c</sup>	0.57±0.03 <sup>a</sup>	0.26±0.01 <sup>b</sup>	0.14±0.01 <sup>c</sup>	275.033	0.000	
C20:2ω6c	0.48±0.03 <sup>b</sup>	0.64±0.08 <sup>a</sup>	0.48±0.00 <sup>b</sup>	0.50±0.02 <sup>b</sup>	11.441	0.003	
C20:3ω3c	0.01±0.01 <sup>a</sup>	-	-	0.02±0.01 <sup>a</sup>	0.250	0.643	
C20:3ω6c	1.39±0.02 <sup>c</sup>	1.44±0.15 <sup>c</sup>	2.26±0.05 <sup>a</sup>	2.01±0.01 <sup>b</sup>	89.229	0.000	
C20:4ω6	12.64±0.06 <sup>c</sup>	12.45±0.15 <sup>c</sup>	14.07±0.06 <sup>b</sup>	14.63±0.09 <sup>a</sup>	379.144	0.000	
C20:5ω3c	5.44±0.04 <sup>b</sup>	3.80±0.21 <sup>d</sup>	4.15±0.02 <sup>c</sup>	6.32±0.04 <sup>a</sup>	332.140	0.000	
C22:2c	0.06±0.03 <sup>a</sup>	0.11±0.05 <sup>a</sup>	0.06±0.02 <sup>a</sup>	0.05±0.02 <sup>a</sup>	2.700	0.116	
C22:6ω3c	0.05±0.01 <sup>b</sup>	0.11±0.05 <sup>ab</sup>	0.15±0.01 <sup>a</sup>	0.06±0.02 <sup>b</sup>	9.632	0.005	
<b>ΣPUFA</b>	<b>36.11±0.02<sup>c</sup></b>	<b>33.23±0.96<sup>d</sup></b>	<b>39.01±0.08<sup>b</sup></b>	<b>40.63±0.16<sup>a</sup></b>	<b>133.911</b>	<b>0.000</b>	
<b>ΣPUFAs/ΣSFAs</b>	<b>1.01±0.00<sup>c</sup></b>	<b>0.92±0.05<sup>d</sup></b>	<b>1.12±0.00<sup>b</sup></b>	<b>1.21±0.01<sup>a</sup></b>	<b>81.871</b>	<b>0.000</b>	
<b>ω-6</b>	<b>22.22±0.05<sup>c</sup></b>	<b>21.04±0.52<sup>d</sup></b>	<b>25.78±0.08<sup>a</sup></b>	<b>24.79±0.11<sup>b</sup></b>	<b>142.421</b>	<b>0.000</b>	
<b>ω-3</b>	<b>13.42±0.13<sup>b</sup></b>	<b>10.66±0.41<sup>c</sup></b>	<b>12.95±0.12<sup>b</sup></b>	<b>15.40±0.05<sup>a</sup></b>	<b>225.794</b>	<b>0.000</b>	
<b>ω-6/ω-3</b>	<b>1.65±0.02<sup>c</sup></b>	<b>2.07±0.03<sup>a</sup></b>	<b>1.99±0.03<sup>b</sup></b>	<b>1.61±0.01<sup>d</sup></b>	<b>337.026</b>	<b>0.000</b>	
<b>AI</b>	<b>0.80</b>	<b>0.79</b>	<b>0.74</b>	<b>0.71</b>	<b>81.871</b>	<b>0.000</b>	
<b>TI</b>	<b>0.51</b>	<b>0.58</b>	<b>0.50</b>	<b>0.44</b>	<b>142.421</b>	<b>0.000</b>	
<b>h/H</b>	<b>1.79</b>	<b>1.71</b>	<b>1.90</b>	<b>2.00</b>	<b>225.794</b>	<b>0.000</b>	

Different letters of inline are significantly different.

The PUFA/SFA proportion, which is widely used in determining the nutritional quality of foods, in this present study, was determined to change between 1.01%-1.21% winter to summer (Table 1). In addition, the total ω-6/ω-3 PUFAs ratio of *C. crinita* was found to be greater than 1 and ranged from 1.61 (winter) to 2.07 (summer) (Table 1). The ω6/ω3 PUFAs ratio also seemed to present seasonal variations. The amounts of the ω-3 PUFA ranged between 10.66±0.41% (in summer) and 15.40±0.05% (in winter), while the ω-6 PUFAs were verified, between 21.04±0.52% (in summer) and 24.79±0.11% (in winter) (Table 1, Figure 2). According to the WHO guideline, ω-6/ω-3 ratio must be lower than the 10 in the diet (Jayasinghe, Jinadasa, & Chinthaka, 2018). In our study, the ratios of the ω-6/ω-3 values of *C. crinita* are lower than the recommended level. Besides, it was

found that the ratio between PUFA/SFA was higher than one in all seasons except summer (0.92). In addition, *C. crinita* showed a PUFA/SFA ratio higher than 1 (Table 1). The PUFA/SFA ratio, which is the determinant of nutritive lipid quality in foods, was above 0.4 recommended by Wood (2004) in this study. Therefore, we can say that the PUFA/SFA ratio obtained from the studied samples are extremely important.

Table 2

The fatty acid, PUFA/SFA,  $\omega 6/\omega 3$ , AI, and TI index of *C. crinita* in comparison with the current study.

	Blacksea (Ivanova et al., 2013)	Mediterranean (Bouafif et al., 2018)	Present study
SFA	65.40	40.64	34.46
MUFA	11.88	27.71	27.79
PUFA	22.72	30.35	37.24
PUFA/SFA	0.35	0.75	1.07
$\omega 6/\omega 3$	1.01	4.79	1.82
IA	-	0.99	0.76
TI	-	0.94	0.51

In this study, the contents of PUFA were high (approximately 40.63 % in winter to 32.23% in summer). The contents of MUFA were low (25.88% in winter to 30.79% in summer), while SFA ranged from 33.50% in winter to 35.98 % in summer (Table 1, Figure 2). When the prominent fatty acids in the all- season were examined, it was determined that palmitic acid belonging to SFA was in the first place, followed by elaidic acid belonging to MUFA and arachidonic acid belonging to PUFA, respectively.

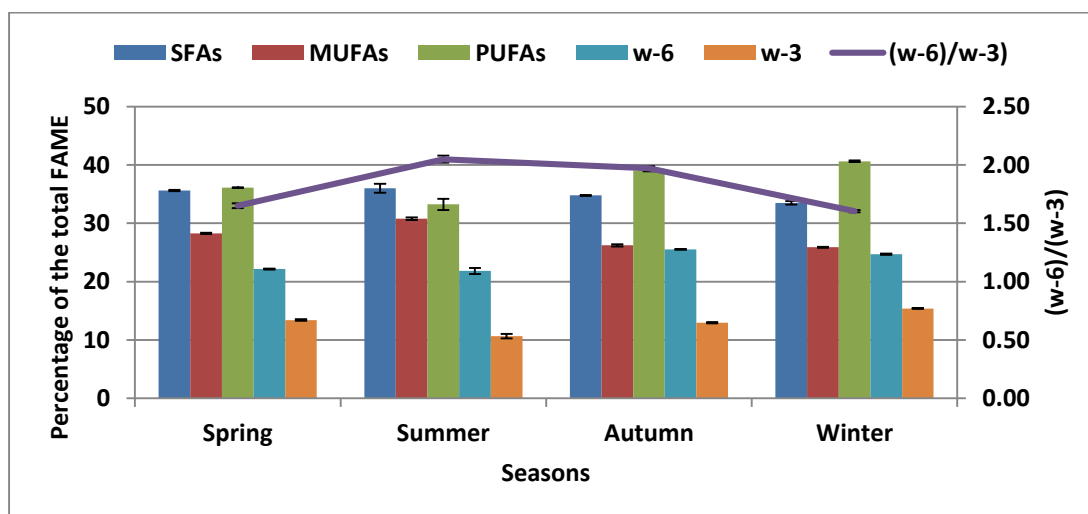


Figure 2. Comparison of the PUFA, SFA, MUFA,  $\omega$ -6,  $\omega$ -3, and  $\omega$ -6/ $\omega$ -3 ratio of *C. crinita*. (n = 3).

The fatty acid compositions changed from 33.50±0.28–35.98±0.76% saturated, to 25.88±0.10–30.79±0.22% monounsaturated and 32.23±0.96–40.63±0.16% polyunsaturated (PUFA) fatty acids (Figure 2). MUFA which predominates in this study was found to contain either 14 or 24 carbons (Table 1). Elaidic acid was the most potent MUFA with 21.70±0.43% of total FAME, especially in the summer. On the other hand, arachidonic acid (ARA) was the major PUFA (Table 1, Figure 3). Rajapakse and Kim (2011) reported that seaweeds are a good sources of PUFA that can be used for health, and Miyashita et al. (2012) declared that PUFA is obtained from especially brown seaweeds. In this study, the season with higher PUFA, SFA, and MUFA contents in *C. crinita* was winter. The results of our research showed lower SFA, but higher MUFA and PUFA, compared with findings from other studies with *C. crinita* (Ivanova et al., 2013; Bouafif et al., 2018) (Table 2). Similarly, Nelson, Phleger, & Nichols (2002) reported that the macroalgal total lipid ratio increased from winter to spring but decreased in summer. The different results in our study than the others may be because of the different environmental conditions of the habitats.

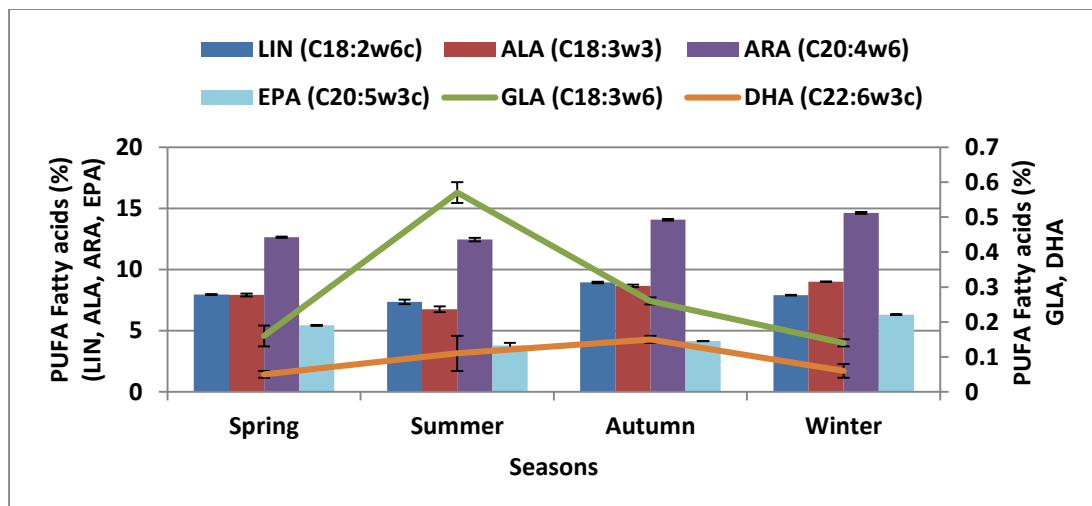


Figure 3. Comparison of the most abundant fatty acids of PUFA of *C. crinita*

The nutritional indexes of *C. crinita* were calculated from the fatty acid ingredients, and are given in Table 1 and Figure 4. It is stated that fatty acid found in macroalgae is extremely important in human nutrition,  $\omega 6/\omega 3$  PUFA ratio is important for health, and also AI and TI ratios should be less than one (Kumari. et al., 2013; Hamid et al., 2015; Schmid et al., 2018; Moreira et al., 2021). Studies have reported that high IA and IT values may worsen nutritional quality for human health (Ulbricht & Southgate, 1991; Bouafif et al., 2018). In this study, it was determined that the thrombogenic index (TI) values ranged from 0.44 (winter) to 0.58 (summer) and the values of the atherogenic index (AI) ranged between 0.71 (winter) and 0.74 (autumn) (Table 1, Figure 4). These values obtained in our study were found to be lower than the AI (0.99) and TI (80.94) values reported by Bouafif et al. (2018) (Table 2). In our study, *C. crinita* was determined to be a good source of  $\omega$ -6 and  $\omega$ -3, particularly arachidonic acid and  $\alpha$ -linolenic acid, respectively.

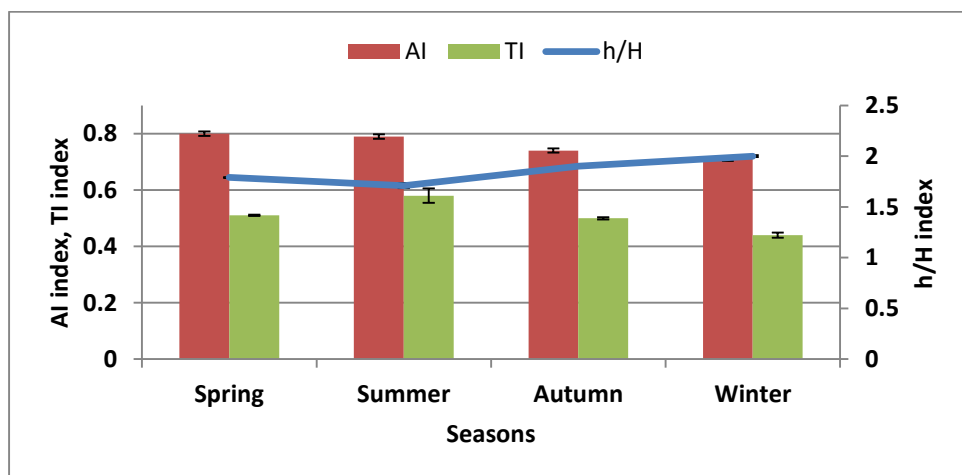


Figure 4. Results of Indexes according to the seasons.

In this study, PCA was calculated over the 7 fatty acids with the highest content to evaluate the relationship of fatty acids between seasons. Data were analyzed using direct oblimin rotation with Kaiser normalization for each component with eigenvalues greater than 1. The other fatty acids were removed from this analysis due to their low ratios, therefore promoting a more reliable the analysis. PCA explained 90.46% of the variables, PC1–77.83%; PC2–12.63% (Figure 5). The fact that total PUFA-LIN and total MUFA-SFA-EPA are in opposite positions on the plot indicate that they have opposite correlation.

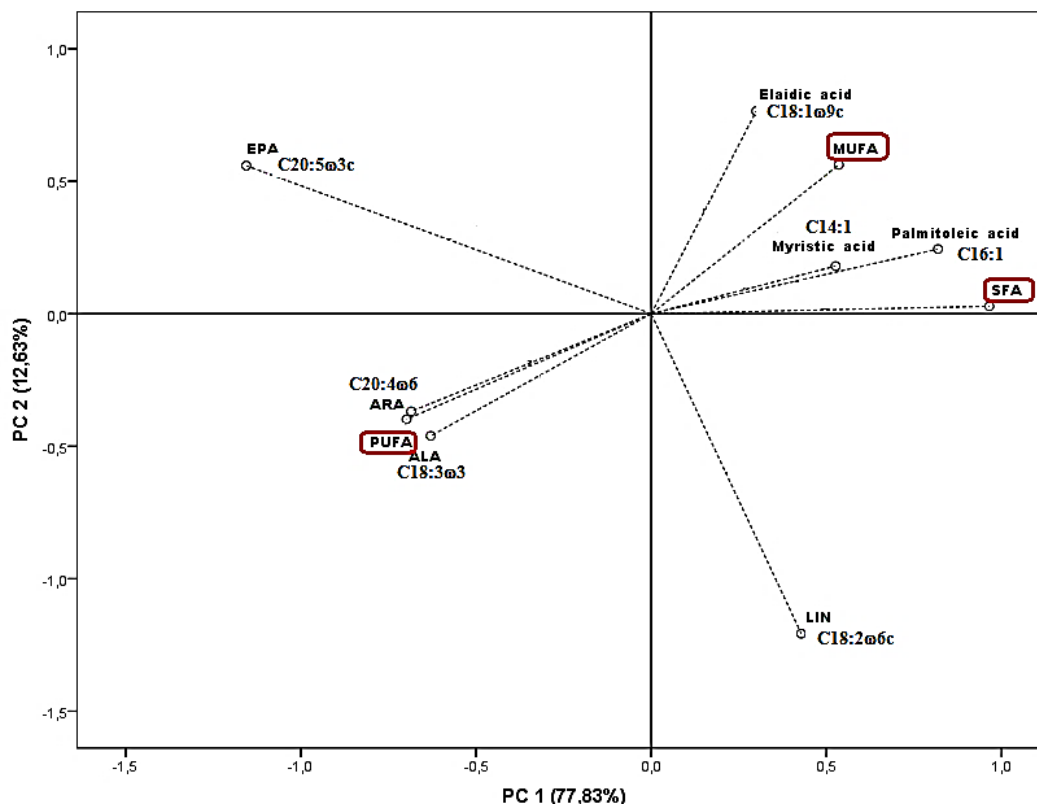


Figure 5. Results of loading plot of multivariate analysis (PCA).

#### 4. Conclusion

In conclusion, our findings showed that *C. crinata* has the highest PUFA content compared to SFA and MUFA. It was determined that among the fatty acids obtained in all seasons from this study, palmitic acid belonging to SFA was in the first place, followed by elaidic acid belonging to MUFA and arachidonic acid belonging to PUFA, respectively. The results of our research showed lower SFA, but higher MUFA and PUFA, compared with findings from other studies with *C. crinita*. Our research results also show that *C. crinita* can be a good source especially for arachidonic acid from  $\omega$ -6 and  $\alpha$ -linolenic acid from  $\omega$ -3. Moreover, in our study,  $\omega$ -6/ $\omega$ -3 PUFA ratio, which is important for health, was found at the recommended level (<10). In addition, PUFA/SFA ratio, which is the determinant of nutritional lipid quality in foods, and AI and TI ratios were again within the recommended values in *C. crinita*. These results suggest that the fatty acids in *C. crinita* may play an important role in human nutrition. In addition, our results demonstrated that the seasons have a significant effect on the fatty acid profile of the studied seaweed. As a matter of fact, in our study, it was determined that the highest PUFA values ranged from 40.63% in winter to 32.23% in summer. It has been determined that the MUFA value varies between 25.88% in winter and 30.79% in summer, and the SFA value varies between 33.50% in winter and 35.98% in summer. For this reasons, it is thought that it is extremely important to reveal the nutritional content of different seaweed species that spread in the seas of Turkey and to observe the seasonal changes in their contents.

#### Acknowledgment

Authors thank to Sinop University, Scientific and Technological Research Application and Research Center (SUBİTAM) for the analysis by gas chromatography-mass spectrometry (GC-MS).

#### Author Contributions

Ali Karaçuha: Collected data, planned the analysis and wrote the article

Gökhan Yıldız: Collected data, performed the analysis.

Melek Ersoy Karaçuha: Performed statistical analysis and wrote the article.

## Conflicts of Interest

The authors non declare conflict of interest.

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