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Variations in the chemical composition of *Costaria costata* during harvest

Xin Wu · Gaoge Wang · Xiaoting Fu

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Abstract In this study, variations in the chemical composition of *Costaria costata* collected during 3 months of the harvest period were analyzed. Moisture (4.94–10.50 %), ash (29.25–38.19 %), protein (9.77–18.15 %), lipid (0.60–2.21 %), crude fiber (4.45–5.68 %), alginate (22.49–29.13 %), fucoxanthin (0.07–0.32 mg g^{-1}), polyphenol (1.579–4.796 mg g^{-1}) were analyzed from dried alga. Six mineral elements were analyzed and the most abundant were calcium (6.64–11.56 mg g^{-1}) and magnesium (7.02–7.92 mg g^{-1}). Analysis of fatty acid composition indicated that the polyunsaturated fatty acids palmitoleic acid and linoleic acid were abundant in May and June, whereas the saturated fatty acid palmitic acid was abundant in July. Amino acid composition was also analyzed and the most abundant amino acids were aspartic acid, glutamic acid, glycine, and alanine. The ratio of mannuronic acid to guluronic acid of alginate was 2.57, 2.17, and 1.66 in May, June, and July, respectively. The gel strength of alginate was 1,449.0, 1,935.0, and 980.5 g cm^{-1} in May, June, and July, respectively. The results of this study indicate that *C. costata* is an excellent resource that provides extensively applications in the industrial areas of chemicals, food, cosmetics, and pharmacy.

Keywords Monthly variation · Chemical composition · *Costaria costata* · Brown seaweed

Wu and G. Wang equally contributed to this work and should be considered joined first authors.

X. Wu · X. Fu (✉)
College of Food Science and Engineering, Ocean University of China, Qingdao 266003, China
e-mail: xiaotingfu@ouc.edu.cn

G. Wang
College of Marine Life Sciences, Ocean University of China, Qingdao 266003, China

Introduction

Edible seaweed can provide complete nutrition because it contains protein, fiber, minerals, and lipids (Marinho-Soriano et al. 2006). Historically, various seaweeds have been used as food by humans, especially in orient countries of China, Japan, and Korea, where these are included in the daily diet (Dawes 1998). In recent years, more countries in Europe, North America, and South America consume seaweeds as food (McHugh 2003; Marinho-Soriano et al. 2006).

The other important application of seaweed relates to the extraction of chemical compounds such as agar, alginate, carrageenan, and other biologically active compounds (Buchholz et al. 2012). Currently, human consumption of brown seaweed (66.5 %) is considerably higher than green seaweed (5 %) and red seaweed (33 %) (Dawes 1998).

The determination of the chemical composition of seaweed is highly essential in providing information on their potential applications (Denis et al. 2010). Numerous studies on seaweed have described changes in its nutritional composition based on the season, temperature, geography, and species (Ito and Hori 1989; Denis et al. 2010).

Costaria costata is an annual brown alga that grows from April and June, and matures in July. It is distributed in the north Pacific area, including the American coast from Alaska to California, as well as in Okhotsk, Japan Sea, northeast of Korean Peninsula, and northern Japan. Reports on the chemical composition of wild-type *C. costata* are limited; currently available descriptions include that of its polysaccharide (Imbs et al. 2009) and lipid (Gerasimenko et al. 2010) content. These studies have indicated that *C. costata* is a promising producer of raw materials for the food industry, cosmetic industry, and pharmacy. However, the output of the wild-type *C. costata* is not sufficient for seaweed industry and thus, its cultivation is important. In recent years, *C. costata* has been successfully cultivated in Rongcheng, China.

The aim of this study was to analyze the chemical composition of cultured *C. costata*, including moisture, ash, lipid, protein, crude fiber, minerals, alginate content and composition, fucoxanthin, and polyphenol, during the harvest period of May, June, and July, compared with those of the wild-type. This is the first report on the chemical composition of cultured *C. costata*, thus the results will serve as a foundation for its potential application in various industrial areas including that of chemicals, food, cosmetics, and pharmacy, as well as promote its cultivation.

Materials and methods

Sporeling nursery and field cultivation

Sporelings were obtained using the gametophyte clone hybridizing technique. Approximately 10 mL of female gametophyte clones, which were stored in our laboratory, were mixed and poured into 1-L beakers with discal palm ropes, which were then placed in an incubator (GXZ-280B JNYQ, Ningbo). The culturing condition was 13 °C temperature, 30 μmol photons m^{-2} s^{-1} irradiance, and a 12:12 h (L/D) photoperiod. The culture medium was sterilized sea water containing 200 μmol L^{-1} KNO_3 and 20 μmol L^{-1} KH_2PO_4, which was refreshed every week. Fifty days later, the juvenile sporelings were transferred to the open sea and cultivated at Rongcheng (37°9′N, 122°24′E), Shandong, China. Field cultivation was conducted by floating raft cultivation facilities in natural conditions.

Samples

The *C. costata* samples were collected from May to July in 2011 in Rongcheng. The samples were washed with tap water and distilled water. The samples were then dried at 60 °C and then ground into powder and stored at 4 °C until chemical analysis.

Proximate analysis

Moisture content was measured using a Mettler MJ33 moisture analyzer (Mettler Toledo). Total ash content was determined by weight loss using an initial weight of 5.0 g of materials that were placed in an oven at 550 °C (method 942.05; AOAC 1990). Crude protein was determined using the Kjeldahl method and was calculated by multiplying nitrogen content with 6.25 (method 976.05; AOAC 1990). Lipid was extracted using the Soxhlet extractor method (method 920.39; AOAC 1990). Crude fiber content was estimated by washing and weighing after boiling 5.0 g of algal samples for 30 min in 1.25 % H_2SO_4 and 1.25 % KOH successively (method 962.09; AOAC 1990). Four minerals — calcium, magnesium, iron, and zinc — were analyzed using an atomic absorption spectrophotometer (AA6800, Shimadu; method 965.09, AOAC). Amino acid composition was determined by HPLC (LC-20A, Shimadzu). Fatty acid composition was analyzed by using a gas chromatograph (6890 N, Agilent). Dry lipids were saponified using a 2 N solution of ethanolic sodium hydroxide, acidified, and then recovered by hexane extraction. Fatty acid methyl esters were obtained by transmethylation and then analyzed by gas chromatography (6890 N, Agilent; Denis et al. 2010).

Alginate content and composition

Approximately 5.0 g of a dried sample powder was soaked in 125 mL distilled water for 2 h, followed by 125 mL of 0.5 % methanol aqueous solution for 1 h. Alginate was then extracted using 0.5 % Na_2CO_3 at 70 °C and stirring for 3 h (Dai 2012); alginate content was measured using the carbazole colorimetric method (Li 2011).

The ratio of mannuronic acid and guluronic acid content (M/G) was estimated by ^1H NMR. Alginate samples were dissolved in water, and 0.1 N of HCL was added to adjust the pH to 3.0. Samples were hydrolyzed in boiling water for 1 h, dialyzed (exclusion limit, M=14,000) against deionized water for 48 h, freeze-dried, and dissolved in deuterium oxide 99.96 atom %D for GG, MM, and GM block determination.

Gel strength of the alginate

Alginate gels were prepared by mixing 30 mL of a 1 % alginate solution with distilled water and pouring the mixture into a dialysis tube. The tubes were immersed in a 10 % calcium chloride solution overnight to allow gel formation. The membranes were discarded to collect the gels, which were then cut into two cylinders. Gel strength was measured using a TMS-PRO Texture Analyzer. The average of the measurements of the two gels was computed (Camacho and Hernández-Carmona 2012).

Fucoxanthin

Fucoxanthin was isolated by double extraction using 50 mL 80 % ethanol at 40 °C for 1 h. The liquids were diluted to a suitable volume. The absorbance was determined at a wavelength of 445 nm using a 722 Visible Spectrophotometer. Fucoxanthin content was calculated by using the following formula (Liu et al. 2010):

$$\text{Fucoxanthin content}\left(\text{mg g}^{-1}\right) = \frac{A_{445} \times n \times V \times 1000}{A \times m \times 100};$$

in which A_{445} is the absorbance at 445 nm, n is the dilution factor, V is the volume of the extracted liquid (mL), A=1,600, and m is the mass of sample (g).

Phenolic compounds

Phenolic compounds were extracted in distilled water (2 g of algal powder in 50 mL distilled water) by shaking in the dark at 30 °C for 24 h. The extracts were centrifuged and the phenolic compounds determined using the Folin–Denis method. Phloroglucinol was used as the reference standard (Yan 1996).

Statistical analysis

Measurements were made in triplicate. All mean values were analyzed by one-way ANOVA (SPSS V17.0). Data were expressed as the mean ± standard deviation (SD). Group means were considered to be significantly different at $P<0.05$.

Results

Proximate analysis

The significant monthly variation in the chemical composition of *C. costata* is shown in Table 1 ($P<0.05$). Moisture, protein, lipid, and phenolic compound levels increased during the three harvest months. Moisture varied by 5.56 %, from 4.94 % (May) to 10.50 % (July). Protein varied by 8.38 %, from 9.77 % (May) to 18.15 % (July). Lipid varied by 1.81 %, from 0.40 % (May) to 2.21 % (July). Phenolic content varied from 1.579 mg g^{-1} (May) to 4.796 mg g^{-1} (July). Ash content in young tissues was higher than that in mature tissues; it was highest in May and lowest in July. Fucoxanthin content in June was significantly ($P<0.05$) lower than those of 0.31 mg g^{-1} in May and 0.32 mg g^{-1} in July.

Table 1 Monthly variation in the chemical composition of *Costaria costata*

	May	June	July
Moisture (% dw)	4.94±0.12^{b}	5.11±0.15^{b}	10.50±0.01^{a}
Ash (% dw)	38.19±0.06^{a}	35.08±0.28^{b}	29.25±0.04^{c}
Proteins (% dw)	9.77±0.11^{b}	11.20±0.33^{b}	18.15±0.96^{a}
Lipids (% dw)	0.40±0.00^{c}	0.60±0.00^{b}	2.21±0.01^{a}
Crude fibers (% dw)	4.45±0.09^{c}	4.97±0.03^{b}	5.68±0.04^{a}
Fucoxanthin (mg g^{-1} dw)	0.31±0.01^{a}	0.07±0.008^{b}	0.32±0.024^{a}
Phenolic compounds (mg g^{-1} dw)	1.579±0.138^{c}	1.914±0.183^{b}	4.796±0.107^{a}

Values are means ± standard (n=3). Mean values in the same row followed by different letters differ significantly ($P<0.05$)

Mineral elements

The monthly change in mineral elements in *C. costata* is shown in Table 2. All values were expressed in mg g^{-1} (dry weight). High monthly changes were found for the four minerals analyzed. Calcium and magnesium were identified as the major minerals of this alga. The contents of calcium, magnesium, and zinc increased from May to July, whereas the level of iron decreased. Calcium concentration varied ($P<0.05$) from 6.64 mg g^{-1} (May) to 11.56 mg g^{-1} (July). The concentration of magnesium was not significantly different during harvest ($P>0.05$). Zinc concentration did not significantly vary during May and June, but then changed ($P<0.05$) to 0.192 mg g^{-1} in July. Iron concentration decreased from 1.77 mg g^{-1} (May) to 0.74 mg g^{-1} (July) ($P<0.05$).

Amino-acid composition

The monthly variation in the total amino-acid (TAA) composition of *C. costata* is shown in Table 3. Significant differences in the concentration of amino acids were observed during the 3 months ($P<0.05$). Alanine (12.03–18.50 %) was the most abundant amino acid. The second-most abundant group consisted of aspartic acid (8.78–12.31 %), glutamic acid (9.78–11.77 %), glycine (10.43–12.59 %), leucine (6.73–8.00 %), and proline (4.81–7.79 %). The third group included valine (5.52–6.03 %), threonine (4.98–6.86 %), serine (5.38–6.44 %), lysine (3.56–4.39 %), arginine (3.57–4.18 %), isoleucine (3.36–3.85 %), and phenylalanine (3.48–3.79 %). The last group consisted of tyrosine (1.34–2.38 %), methionine (1.14–2.06 %), and histidine (0.96–1.46 %).

Fatty acid composition

The monthly variation in the fatty composition of *C. costata* is shown in Table 4. Significant differences in the concentration of fatty acids during 3 months were observed ($P<0.05$). The concentration of palmitoleic acid (C16:1 n-9; 29.72–29.72)

Table 2 Monthly changes in mineral elements of *Costaria costata* (mg g^{-1} dw)

	Calcium	Magnesium	Iron	Zinc
May	6.64±0.53^{c}	7.18±0.16^{b}	1.77±0.14^{a}	0.03±0.00^{b}
June	9.87±0.12^{b}	7.02±0.24^{c}	1.51±0.05^{b}	0.03±0.00^{b}
July	11.56±0.09^{a}	7.92±0.18^{a}	0.74±0.07^{c}	0.19±0.01^{a}

Values are means ± standard (n=3). Mean values in the same row followed by different letters differ significantly ($P<0.05$)

Table 3 Monthly variations in the amino acid composition of *Costaria costata* (%)

Amino acid	May	June	July
Alanine	18.50±0.01[a]	18.25±0.02[b]	12.03±0.00[c]
Aspartic acid	8.78±0.00[c]	10.29±0.00[b]	12.31±0.02[a]
Glutamic acid	9.78±0.00[c]	10.58±0.01[b]	11.77±0.00[a]
Glycine	10.43±0.02[c]	11.98±0.02[b]	12.59±0.02[a]
Leucine	8.00±0.00[a]	7.02±0.00[b]	6.73±0.01[c]
Proline	7.79±0.01[a]	6.55±0.00[b]	4.81±0.00[c]
Valine	6.03±0.00[a]	5.52±0.00[c]	5.81±0.00[b]
Threonie	4.98±0.01[c]	5.95±0.00[b]	6.86±0.00[a]
Serine	5.38±0.00[c]	6.44±0.01[a]	5.56±0.01[b]
Lysine	4.21±0.00[b]	3.56±0.00[c]	4.39±0.00[a]
Arginine	3.83±0.01[b]	3.57±0.00[c]	4.18±0.01[a]
Isoleucine	3.85±0.00[a]	3.36±0.00[c]	3.52±0.01[b]
Phenylalanine	3.79±0.01[a]	3.48±0.01[c]	3.54±0.00[b]
Tyrosine	1.70±0.00[b]	1.34±0.00[c]	2.38±0.00[a]
Methionine	1.75±0.00[b]	1.14±0.00[c]	2.06±0.00[a]
Histidine	1.20±0.00[b]	0.96±0.00[c]	1.46±0.00[a]

Values are means (%, protein)±stand (n=3). Different superscripts indicate significant differences among 3 months. Data in bold indicate essential amino acids for humans

Table 4 Monthly variations in the fatty acid composition of *Costaria costata* (%)

Fatty acid	May	June	July
C14:0	7.94±0.01[b]	7.80±0.01[a]	5.74±0.00[c]
C14:1 n-5	0.38±0.00[b]	0.45±0.00[a]	ND
C15:0	0.46±0.00[a]	0.47±0.00[a]	ND
C16:0	ND	ND	23.97±0.03
C16:1 n-9	29.75±0.01[a]	29.72±0.01[b]	1.00±0.00[c]
C17:0	1.34±0.00[a]	0.27±0.00[c]	0.92±0.00[b]
C18:0	ND	ND	3.63±0.00
C18:1 n-9	ND	4.67±0.00[b]	15.42±0.03[a]
C18:2 n-6	28.42±0.03[a]	23.78±0.04[b]	6.62±0.02[c]
C18:3 n-6	0.83±0.00[c]	0.89±0.00[b]	1.20±0.00[a]
C18:3 n-3	1.63±0.00[b]	1.62±0.00[b]	1.69±0.00[a]
C20:1 n-9	3.80±0.00[b]	2.28±0.00[c]	3.97±0.01[a]
C20:3 n-6	7.82±0.00[b]	7.76±0.00[c]	11.41±0.02[a]
C23:0	0.63±0.00[c]	0.68±0.00[b]	4.25±0.00[a]
MUFA	36.98	42.94	24.22
PUFA	49.45	40.84	27.75
SFA	13.57	16.22	48.03

Values are means (%, FA)±stand (n=3). Different superscripts indicate significant differences among 3 months

ND not detected

and linoleic acid (C18:2 n-6; 23.78–23.42) were highest in May and June, followed by myristic acid (C14:0; 7.80–7.94) and C20:3 n-6 (7.76–7.82). The concentration of palmitic acid (C16:0; 23.97), C18:1 n-9 (15.42), and C20:3 n-6 (11.41) were high in July, followed by myristic acid (C14:0; 5.74) and linoleic acid (C18:2 n-6; 6.62). Polyunsaturated fatty acid (PUFA) and monounsaturated fatty acid (MUFA) levels varied from 51.97 % (July) to 86.43 % (May) of the total fatty acid, and that of saturated fatty acid (SFA) varied from 13.57 % (May) to 48.03 % (July) of the total fatty acid. PUFA content was highest in May (49.45 %), and then was 40.84 % in June and 27.75 % in July. MUFA content was highest in June (42.94 %), followed by 36.98 % in May and 24.22 % in July. SFA content was highest in July (48.03 %), followed by 16.22 % in June and 13.57 % in May.

Alginate content and M/G ratio

Alginate content varied from 22.49 % to 29.13 % (P<0.05) and was highest in June (Table 5). Analysis of ^{1}H NMR spectra (data not shown) and Grasdelen calculations (Zheng et al. 1992; Grasdalen 1983) showed that the M/G ratio was 2.57 in May, 2.17 in June, and 1.66 in July.

Alginate gel strength

The gel strength of alginate derived from *C. costata* significantly changed (P<0.05) from May to July (Table 5). Gel strength was highest in June (1,935.0 g cm^{-1}) and lowest in July (980.5 g cm^{-1}).

Discussion

The comparison of the chemical composition in *C. costata* with that of other seaweeds is summarized in Table 6. The moisture content of *C. costata* was lower than another brown alga, *Eisenia arborea* (Hernández-Carmona et al. 2009), especially in the juvenile tissues. The moisture content of *Capsosiphon fulvescens* varied by 3.9 % (Sun et al. 2012), whereas that of *Enteromorpha* spp. varied by 2.3 % (Augilera-Morales et al. 2005), which was lower than that observed in *C. costata* (5.56 %). Low moisture level can prevent microorganism grow to assure the quality of other components and

Table 5 Monthly changes of the alginate content and gel strength of the alginate from *Costaria costata*

	May	June	July
Alginate[1] (% dw)	22.90±1.93[b]	29.13±1.28[a]	22.49±0.78[b]
Gel strength[2] (g cm^{-1})	1449.0±154.1[b]	1935.0±141.4[a]	980.5±21.9[c]

Values are means ± standard deviation ([1] n=3; [2] n=2). Mean values in the same row followed by different letters differ significantly (p<0.05)

Table 6 Comparison of chemical composition in *Costaria costata* with other seaweeds

Group	Species	(%)	references
Moisture			
G	*Capsosiphon fulvescens*	4.4–8.3	Sun et al. 2012
G	*Enteromorpha* spp.	6.70–9.00	Augilera-Morales et al. 2005
B	*Eisenia arborea*	9.90–10.52	Hernández-Carmona et al. 2009
B	*Costaria costata*	4.49–10.50	This study
Ash			
R	*Grateloupia turuturu*	18.5	Denis et al. 2010
R	*Gracilaria cervicornis*	10.5	Marinho-Soriano et al. 2006
G	*Enteromorpha* spp.	32.64–36.38	Augilera-Morales et al. 2005
B	*Sargassum horneri*	21.8–36.7	Murakami et al. 2011
B	*Sargassum vulgare*	19.4	Marinho-Soriano et al. 2006
B	*Costaria costata*	29.25–38.19	This study
Protein			
R	*Grateloupia turuturu*	22.9	Denis et al. 2010
R	*Catenella repens*	2.78–16.03	Banerjee et al. 2009
R	*Gracilaria cervicornis*	19.7	Marinho-Soriano et al. 2006
G	*Enteromorpha* spp.	9.45–14.10	Augilera-Morales et al. 2005
B	*Sargassum horneri*	5.64–12.8	Murakami et al. 2011
B	*Sargassum vulgare*	13.6	Marinho-Soriano et al. 2006
B	*Costaria costata*	9.77–18.15	This study
Lipids			
R	*Grateloupia turuturu*	2.6	Denis et al. 2010
R	*Gracilaria cervicornis*	0.43	Marinho-Soriano et al. 2006
R	*Catenella repens*	0.17–0.24	Banerjee et al. 2009
G	*Enteromorpha* spp.	2.24–2.27	Augilera-Morales et al. 2005
B	*Sargassum horneri*	0.52–1.15	Murakami et al. 2011
B	*Sargassum vulgare*	0.49	Marinho-Soriano et al. 2006
B	*Costaria costata*	0.57–0.71	Gerasimenko et al. 2010
B	*Costaria costata*	0.40–2.21	This study
Fiber			
R	*Gracilaria cervicornis*	5.65	Marinho-Soriano et al. 2006
B	*Sargassum vulgare*	7.74	Marinho-Soriano et al. 2006
B	*Costaria costata*	4.45–5.68	This study

G green alga, *R* red alga, *B* brown alga

allow longer storage times (Hernández-Carmona et al. 2009). Previous reports have shown that the ash content in brown algae was generally high, whereas that in red alga was low (Hernández-Carmona et al. 2009). Compared with *Eisenia arborea* (19.25–29.32 %; Hernández-Carmona et al. 2009), *Sargassum horneri* (27.9–31.0 %; Murakami et al. 2011),

Table 7 Composition of minerals in *Costaria costata* with other brown seaweeds (mg g^{-1} dw)

seaweeds	Calcium	Magnesium	Iron	Zinc	reference
Eisenia arborea	11.731	16.156	6.153	0.01	Hernández-Carmona et al. 2009
Fucus vesiculosus	1.2–5.4	5.1	0.28–0.32	0.28	Truus et al. 2004
Sargassum horneri	10.3–14.7	12.1–19.8	–	0.03–0.05	Murakami et al. 2011
Scytosiphon lomentaria	1.673	1.172	1.546	0.0810	Zhang et al. 2011
Saccharina (*Laminaria japonica*)	2.64	1.05	0.549	0.0250	Zhang et al. 2011
Costaria costata	6.637–11.562	7.022–7.923	0.735–1.774	0.031–0.192	This study

– Unknown

Table 8 Composition of content and M/G ration of alginate extracted from *Costaria costata* with other species

Seaweeds	Content (% dw)	M/G	Reference
Saccharina (*Laminaria*) *japonica*	14.2–22.5	2.26–3.95	Chen et al. 2009
Turbinaria conoides	11.0–38.0	0.57–0.60	Jothisaraswathi et al. 2006
Sargassum polycystum	17.1–27.6	0.58–0.76	Saraswathi et al. 2003
Sargassum. oligocystum Mont	18.9–20.5	0.49–0.62	Davis et al. 2004
Macrocystis pyrifera	–	1.63	Murillo-Álvarez and Hernández-Carmona 2007
Eisenia arborea	–	1.08	Murillo-Álvarez and Hernández-Carmona 2007
Sargassum sinicola	–	0.56	Murillo-Álvarez and Hernández-Carmona 2007
Sargassum mangarevense	9.3	1.42	Zubia et al. 2008
Costaria costata	22.49–29.13	1.66–2.57	This study

– Unknown

Scytosiphon lomentaria (30.63 %; Zhang et al. 2011), and *Saccharina* (*Laminaria) japonica* (20.0 %; Zhang et al. 2011), the ash content of *C. costata* (29.25–38.19 %) was at a high level. The protein content of *C. costata* (9.77–18.15) was higher than those reported for *Laminaria japonica* (8.7 %; Zhang et al. 2011), *Undaria pinnatifida* (13.8 %; Zhang et al. 2011), and *Catenella repens* (2.78–16.03 %; Banerjee et al. 2009) and lower than *Scytosiphon lomentaria* (19.71 %; Zhang et al. 2011). The protein content of *C. costata* increased with growth, which agreed with the findings of *Sargassum horneri*, which was 8.8 % in immature algae and 10.7 % in mature ones (Murakami et al. 2011). The highest lipid content was 0.71 % in wild-type *C. costata* (Gerasimenko et al. 2010), whereas this was 2.21 % in cultured alga in our current study. The lipid content of cultured *C. costata* markedly varied from 0.40 % to 2.21 % as the algae matured. The results indicate that the lipid content of cultured *C. costata* was higher than that of the wild-type (0.57–0.71 %; Gerasimenko et al. 2010); therefore, adult alga are better sources for lipid isolation. The crude fiber content of cultured *C. costata* was lower than that of *Scytosiphon lomentaria* (21.58 %; Zhang et al. 2011), *Undaria pinnatifida* (12.2 %; Zhang et al. 2011), *Sargassum vulgare* (4.8–10.5 %; Marinho-Soriano et al. 2006), and *Eisenia arborea* (4.32–6.44 %; Hernández-Carmona et al. 2009). Low fiber has been strongly associated with high nutrition (Hernández-Carmona et al. 2009); thus, *C. costata* can be considered as a highly nutritious resource. Fucoxanthin is a major carotenoid present in the chloroplasts of brown algae (Nomura et al. 2012). The levels of fucoxanthin in *Laminaria japonica* (0.556 mg g^{-1}; Liu et al. 2010), *Sargassum horneri* (1.35–3.23 mg g^{-1}; Nomura et al. 2012), and *Cystoseira hakodatensis* (0.63–4.14 mg g^{-1}; Nomura et al. 2012) were higher than that of *C. costata* (0.07–0.32 mg g^{-1}). Several methods (ethanol and distilled water extraction at different temperatures) have been used to extract phenolic compounds and the results showed no significant differences (data not shown). The phenolic compounds were extracted with distilled water to replace the use of an organic solution, so that the phenolic compounds could be safely and conveniently used in the food industry. Compared with other Chinese common brown alga, i.e., *Saccharina* (*Laminaria*) *japonica* (0.3 %), *Undaria pinnatifida* (0.1 %), *Sargassum thunbergii* (0.2 %), and *Sargassum pallidum* (0.7 %) (Yan 1996), *C. costata* (0.16–0.48 %) contained intermediate concentrations of phenolic compounds.

The comparison of minerals in *C. costata* with those in other seaweeds is summarized in Table 7. The mineral content of *C. costata* was similar to that described in previous reports. Calcium and magnesium were the most abundant elements, whereas iron and zinc were the minor elements. The calcium content of *C. costata* was noticeably higher than *Scytosiphon lomentaria* (1.673 mg g^{-1}; Zhang et al. 2011), *Saccharina* (*Laminaria) japonica* (2.64 mg g^{-1}; Zhang et al. 2011), and *Fucus vesiculosus* (1.2–5.4 mg g^{-1}; Truus et al. 2004). Therefore, *C. costata* can be a potential source for mineral supplements for animals and humans.

According to the amino acid composition presented in Table 3, the levels of alanine (12.03–18.50 %), aspartic acid (8.78–12.31 %), glutamic acid (9.78–11.77 %), and glutamic acid (10.43–12.59 %) were high, which were similar with those reported for other alga (Zhang et al. 2011; Hernández-Carmona et al. 2009; Sun et al. 2012). Other researchers have also reported that the levels of histidine and methionine were low in this algal species. Glutamic acid and glycine and alanine are responsible for the sweet flavor in some seaweed such as nori (Hernández-Carmona et al. 2009), and thus this may also be true for *C. costata*. Essential amino acids are depicted in bold characters in Table 3. The levels of essential amino acids were 32.61 % and 32.91 % in May and July, respectively, which were higher than the recommended value

of the FAO/WHO (32.3 %) (FAO/WHO 1973). The composition of amino acids indicated that *C. costata* was nutritious and delicious.

The highest unsaturated fatty acid content (86.43 %) was observed in May and the lowest (51.97 %) was observed in July. It has been previously reported that during maturation, the unsaturated fatty acid content of wild *C. costata* collected from the Sea of Japan decreased from 82.5 to 80.4 % (Gerasimenko et al. 2010). Thus the content of unsaturated fatty acid was highest in cultured *C. costata* during the May harvest. n-6 PUFAs and their derivatives are important in various biological systems including that involving the immune response, thrombosis, and the brain (Nomura et al. 2012). The results of this study indicated that *C. costata* could be a useful resource for unsaturated fatty acids. The results also indicated that the composition of fatty acids varied among different growth periods. Linoleic acid (C18:2 n-6) and palmitoleic acid (C16:1 n-9) were mainly present in May and June, whereas oleic acid (C18:1 n-9) and palmitic acid (C16:0) were mainly in July.

The comparison of content and M/G ratios of alginates extracted from *C. costata* and other species is presented in Table 8. The results indicate that the alginate content of *C. costata* (22.49–29.13 %) was higher than that of *Saccharina* (*Laminaria) japonica* (14.2–22.5 %; Chen et al. 2009), which is known as the most important resource for alginate production in the world. Thus, *C. costata* could be developed as a new raw material for the production of alginate. The M/G ratio of alginate from *C. costata* varied from 1.66 to 2.57, which was much lower than that in *Laminaria japonica*, but higher than that of other three algae such as *Turbinaria conoides* (Jothisaraswathi et al. 2006), *Sargassum polycystum* (Saraswathi et al. 2003), and *Sargassum oligocystum* (Davis et al. 2004). The M/G ratio of *C. costata* decreased during algal growth. Similar results have been previously observed in *Turbinaria conoides* (Jothisaraswathi et al. 2006). Alginic acids from young tissues mainly consist of mannuronic acid, whereas older tissues accumulate alginic acids, with a higher content of guluronic acid. Alginates that contain high levels of polymannuronic blocks impart antitumor effects, and that with high levels of polyguluronic blocks showed biosorption properties (Imbs et al. 2009). The M/G ratio and the distribution of alginate chains strongly influence gel characteristics (McHugh 1987). It has been previously reported that the alginate gel with more guluronic acid was firm, whereas that with more mannuronic acid was elastic (Imbs et al. 2009). The gel strength of alginate from *C. costata* (980.5 to 1,935.0 g cm^{-1}) was higher than that of *Sargassum cymosum* (709.7 g cm^{-1}) and *Sargassum* sp. (866.0 g cm^{-1}) (Camacho and Hernández-Carmona 2012). The gel strength of alginate from *C. costata* suggested that this could serve as a useful resource for the food industry. Alginates from different harvest periods exhibited various physical and biological activities and thus could be applied in a wide range of industries.

In conclusion, the chemical composition of *C. costata* extensively varied during harvest. This research improved our knowledge of the nutritional value of this alga. The cultured *C. costata* can be recommended as a nutritious food, based on its low lipid and crude fiber contents, as well as its high levels of minerals, essential amino acids, and PUFA. Moreover, this alga might be of great interest to the polysaccharide industry, because the cultured *C. costata* can be developed into a new resource for the production of alginate, which offers superior gel strength and a different M/G ratio compared to the traditional material from *Saccharina japonica*.

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