



Asian Journal of **Biochemistry**

ISSN 1815-9923



Academic
Journals Inc.

www.academicjournals.com

Biochemical Composition and Fatty Acid Profile of the Green Alga *Ulva reticulata*

Annian Shanmugam and Chendur Palpandi
Center of Advanced Study in Marine Biology, Annamalai University,
Parangipettai-608 502, India

Abstract: The total lipid, protein, carbohydrate, water and fatty acid contents of seaweed *U. reticulata* belonging to the class of Chlorophyceae that had been collected from Vellar estuary, southeast coast of India. Water contents of the *U. reticulata* was found as 75.33%. Carbohydrate, protein and lipid contents have been estimated as 50.248, 8.484, 19.98 and 1.7% of the dry sample correspondingly. The analysis of fatty acids by gas chromatography revealed the presence of myristic acid, palmitic acid, heptadecenoic acid, oleic acid and linoleic acid. Among the fatty acids, palmitic acid was predominant in all the fatty acids studied.

Key words: Biochemical composition, saturated fatty acids, monounsaturated fatty acids, polyunsaturated fatty acids, *Ulva reticulata*, vellar estuary

INTRODUCTION

Seaweeds have been used since ancient times as food, fodder and fertilizer and as source of medicinal drugs. Today seaweeds are the raw material for industrial production of agar, carrageenan and alginates (Barbara and Cremades, 1993), but their consumption as food in Asian countries are still going on (Mishra *et al.*, 1993). They are nutritionally valuable as fresh or dried vegetables, or as ingredients in a wide variety of prepared foods (Robledo and Pelegrin, 1997). In particular, certain edible seaweeds contain significant quantities of lipids, protein, vitamins and minerals (Norziah and Ching, 2000; Sanchez-Machado *et al.*, 2002; Wong and Cheung, 2000), although nutrient contents vary with species, geographical location, season and temperature (Dawes *et al.*, 1993; Kaehler and Kennish, 1996).

The study of natural products and the chemical constituents occurring in algae is known as phycochemistry. Usually the natural products are the secondary metabolites. The metabolic behaviour of lipids is largely determined by the structure of the component fatty acids. Generally, fatty acids occurring in algal lipids are straight chain, even carbon number molecules and usually contain one or more double bonds. Much work has been done fatty acid composition of microalgae and seaweeds (Marolia *et al.*, 1982; Takagi *et al.*, 1985; Wood, 1988; Liekenjie, 1989). However, information on the fatty acids of seaweeds collected from Parangipettai coast is lacking. The objective of this study was to determine the biochemical composition and fatty acid profile of the green alga *U. reticulata* originating from Vellar estuary, Parangipettai coast.

MATERIALS AND METHODS

Seaweed was collected from Vellar estuary, during low tide. Samples has been cleaned from epiphytes, epifauna, pebbles and other molluscan shells and washed several times in fresh water, blotted and weighed.

Corresponding Author: Chendur Palpandi, Center of Advanced Study in Marine Biology, Annamalai University, Parangipettai-608 502, India Tel: 04144-243223 Fax: +914144243555

The fresh samples were dried at 60°C for 24 h (in hot air oven) and the dried powder was sieved to get fine powder. Total protein was estimated by following the method described by Raymont *et al.* (1964), total carbohydrate using phenol sulphuric method (Dubois *et al.*, 1956) and total lipid was extracted by Folch *et al.* (1956) using chloroform: methanol mixture. The moisture content was estimated by subtracting the dry weight from the known wet weight of the sample. Protein, carbohydrate and lipid content of the species of *U. reticulata* were analyzed and expressed in percentage. An extraction procedure for fatty acid was followed as described by Bligh and Dyer (1959). Identification and quantification of fatty acids were done using Agilent Technologies 6890 N, Network GC system.

RESULTS AND DISCUSSION

Lipids, proteins and carbohydrates are the most important biochemical components in algal biomass. Lipids are rich in -C = O- bonds, providing much more energy in oxidation processes than other biological compounds. Mainly owing to their high reduction levels, they constitute a convenient storage material for living organisms. In algae, the lipids are widely distributed, specially in several resistance stages (Miller, 1962).

Carbohydrate is the most important component for metabolism and it supplies the energy needed for respiration and other metabolic processes. In the present experiment the carbohydrate content of *U. reticulata* was found as 50.248% (Fig. 1). The findings of the present experiment coincide well with that of carbohydrate contents reported by the earlier studies like Reeta and Kulandaivelu (1999). They found that the carbohydrate contents were 48.4% of *Gracilaria* spp. in dry weight basis. Hossain *et al.* (2003) reported that the carbohydrate contents were 19.93 and 20.81% in sample 1 and 2 from *Sargassum horneri*. Reeta (1993) has studied the carbohydrate content varied from 6.65 in March to 15.18 in January in *S. wightii*. Haroon (2000) investigated the carbohydrate contents were 54.71±8.17% of DW in *Enteromorpha* spp. Sasikumar (2000) studied the two species of green algae *Enteromorpha intestinalis* and *C. linum*, the carbohydrate contents were 20.4 to 54.6%. The highest average total carbohydrate contents from *U. rigida* (Chlorophyta) was 63.04±29.15 kg⁻¹ D.W (6.3%) (Dere *et al.*, 2003). Whereas in the present investigation the carbohydrate contents was reported as 50.248% which is higher than that of *S. wightii* and lower than that of the others such as *Enteromorpha* spp., *C. linum* and *U. rigida*. Lipids and carbohydrate contents should obviously be very variable depending upon the state of nutrition of cells (Ricketts, 1966).

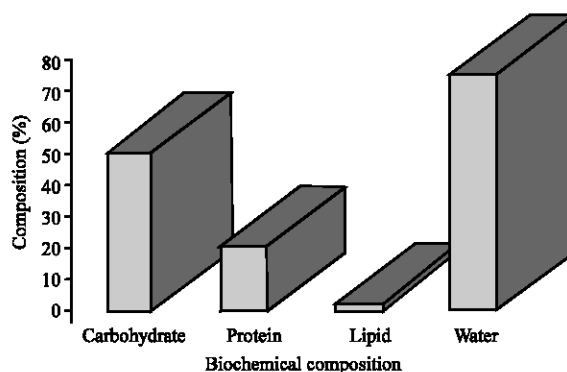


Fig. 1: Biochemical composition of green alga *U. reticulata*

Protein are having crucial functions in all the biological processes. Their activities can be described by enzymatic catalysis, transport and storage, mechanical sustentation, growth and cellular differentiation control (Stryer, 1988). In the present study the protein content was estimated as 19.98% in *U. reticulata* (Fig. 1). The findings of the present experiment coincide well with the protein contents reported by Sasikumar (2000) who has reported the varying protein content between 5.2 and 18.6% in *Enteromorpha intestinalis* and *C. linum*. Dere *et al.* (2003) observed the maximum protein content (0.94 to 31.03%) in some of the Rhodophyta and some green seaweeds belonging to the genus *Ulva*, the content was ranging between 2.9 and 28.1%. Burtin (2003) investigated the higher protein contents in green and red seaweeds (on an average of 10-30% of the dry weight). Hossain *et al.* (2003) reported the protein content of 22.38 and 21.96% of dry sample 1 and 2 from *S. hornei*. Roslin (2003) calculated the protein content in Chlorophyceae and found varying varied from 1.5 to 24.8%. In Phaeophyceae, the maximum protein content was shown by *S. ilicifolium* (28.2%) and *S. wightii* (28.2%). In the present study the protein contents were higher than that of the earlier studies like Haroon (2000) i.e., the protein content of *Enteromorpha* spp. showed the minimum of 9.42±4.62% and maximum of 20.60±5.00% on dry weight basis.

In general the total lipid content was always found less than 4% (Herbreteau *et al.*, 1997) in almost all the seaweeds studied so far. Falling in the same line the lipid content in *U. reticulata* was also reported as 1.7% (Fig. 1). This general trend is further supported by the findings of Reeta (1993) in *S. wightii* (0.159 to 1.551%), Reeta and Kulandaivelu (1994) in *Gracilaria* spp. (0.78 to 3.97%) and Sasikumar (2000) in *Enteromorpha intestinalis* and *C. linum* (1.2 to 8.6%). Hossain *et al.* (2003) also reported 1.38 and 1.96% of Glycolipid in samples 1 and 2 of *S. horneri*.

Water is biologically significant as an essential metabolite i.e., it participates in the chemical reactions of metabolism. In particular it is used as a source of hydrogen ion in photosynthesis and is used in hydrolysis reactions. In the present study the moisture contents were 75.33% in *U. reticulata* (Fig. 1). This result is in conformity with the observation of Hossain *et al.* (2003) where the moisture contents were found as 86.94% in sample -1 and 87.0% in sample-2 from *S. horneri*.

Lipid represent only 1-5% of algal dry matter and show an interesting polyunsaturated fatty acid composition particularly omega 3 and omega 6 acids which play a role in the prevention of cardiovascular diseases, osteoarthritis and diabetes. The green algae show interesting levels of alpha linolenic acid (ω 3 C 18:3) (Burtin, 2003).

Although seaweeds are not a conventional source of energy (their total lipid content is low), their polyunsaturated fatty acids contents are high when compared to terrestrial vegetable (Darcy-Vrillon, 1993). In the present study saturated fatty acids were dominating (70.01%) the other groups of fatty acids among the total fatty acids and the individual contribution of 16:0 and 14:0 was found as 50.76% and 11.77%, respectively (Table 1). These results are in conforming with the previous reports

Table 1: Fatty acid profile (% of total fatty acids) from *U. reticulata*

Fatty acids	Composition (%)
Saturated fatty acids	
14:0	11.77
16:0	50.76
17:0	2.59
18:0	4.89
Monounsaturated fatty acids	
16:1 ω 7c	5.30
17:1	6.77
Polyunsaturated fatty acids	
16:2 ω 6c	3.42
18:2 ω 6c	4.54
Unknown	9.96
Total	100.00

(Venkatesalu *et al.*, 2003a, b, 2004; Vasanthi *et al.*, 2003). The highest relative percentage of palmitic acid (85.35%) was recorded in *Gellidium micropterum* (Venkatesalu *et al.*, 2004). The single most abundant fatty acid C16:0 (which in *Porphyra* spp. accounted for 63.19% of all fatty acid) and (Sanchez-Machado *et al.*, 2004). Palmitic acid was reported as a major fatty acid common to *Ergrezia menziesii*, *Chondracanthus canaliculatus* and *Ulva lobata* (Nelson *et al.*, 2002). The highest relative percentage of myristic acid (7.86) was recorded in *Acanthophora spicifera* and the highest relative percentage of pantedecanoic acid (1.29) was recorded in *Padina tetrastromatica* (Venkatesalu *et al.* 2004). In the present study the myristic acid was recorded in 11.77% and pantedecanoic acid was not recorded.

The monounsaturated fatty acids (MUFA) were the next most common fatty acids (12.07%) with the higher levels of mostly 18:1 ω 7c (6.77%). The findings of the present experiment coincide well with the earlier study by Dawczynski (2006) the concentrations of oleic acid were at high levels in *Porphyra* spp. From China and *Laminaria* spp. and accounted for more than 20% of total FAME but such a high content was only found in single samples. Furthermore, seaweed varieties tested has low levels of palmitoleic acid (16:1n-7) and the concentrations of this FA ranged from 0.1 to 3.6% of total FAME.

The polyunsaturated fatty acids occupying the third position by contributing 7.96% of total fatty acids that are mainly represented by 18:2 ω 6c (4.54%). In the previous study by Hossian *et al.* (2003) reported that the saturated fatty acids 16:0 and 22:0 and monounsaturated fatty acid 18:1 were the major fatty acids for simple lipids.

In India, development of novel foods such as functional foods could be a new possibility for the use of this alga *U. reticulata*, especially for the protein-rich species, in human nutrition. In general, the lipid contents of all edible seaweeds were low and high levels of polyunsaturated fatty acids of the omega-3 and omega-6 families. This investigation of edible macroalga *U. reticulata* verified the presence of several health-promoting and beneficial nutrients, such as EAAs, important FAs and highly proteins and low levels of lipids. Further experimentation should be conducted with the samples of all seasons through a year for getting biochemical variation in a year.

ACKNOWLEDGMENT

Authors are thankful to the Director, CAS in Marine Biology and authorities of Annamalai University for providing with necessary facilities. The authors are also thankful to the Ministry of Environment and Forests, New Delhi for the financial assistance.

REFERENCES

- Bligh, E.G. and W.J. Dyer, 1959. A rapid method for total lipid extraction and purification. *Can. J. Biochem. Physiol.*, 37: 911-917.
- Burtin, P., 2003. Nutritional value of seaweeds. *Ejeafche*, 2: 498-503.
- Darcy-Vrillon, B., 1993. Nutritional aspects of the developing use of marine macroalgae for the human food industry. *Int. J. Food Sci. Nutr.*, 44: 23-35.
- Dawczynski, C., R. Schubert and G. Jahreis, 2006. Amino acids, fatty acids and dietary fibre in edible seaweed products. *Food Chem.*, 103: 891-899.
- Dawes, C.J., C. Kovach and M. Fridlander, 1993. Exposure of *Gracilaria* to various environmental conditions II. The effect on fatty acid composition. *Botanica Marina*, 36: 289-296.
- Dere, S., N. Dalkiran, D. Karacaoglu, G. Yildiz and E. Dere, 2003. The determination of total protein, total soluble carbohydrate and pigment contents of some macroalgae collected from Gemlik-Karacaali (Bursa) and Erdek-Ormanli (Balikesir) in the Sea of Marmara, Turkey. *Oceanologia*, 45: 453-471.

- Dubois, M., J. Gillas, R. Hamilton, A. Rebus and F. Smith, 1956. Colorimetric method for determination of sugars. Anal. Chem., 28: 350-356.
- Folch, J.M., G.H. Lees and S. Stanely, 1956. A simple method for the isolation and purification of total lipids from animal tissues. J. Biol. Chem., 226: 497-509.
- Haroon, A.M., 2000. The biochemical composition of *Enteromorpha* spp. from the Gulf of Gdansk coast on the southern Baltic Sea. Oceanologia, 42: 19-28.
- Herbretau, F., L.J.M. Coiffard, A. Derrien and Y. De Roeck-Holtzhauer, 1997. The fatty acid composition of five species of macroalgae. Botanica Marina, 40: 25-27.
- Hossain, Z., H. Kurihara and K. Takahashi, 2003. Biochemical composition and lipid compositional properties of the brown alga *Sargassum horneri*. Pak. J. Biol. Sci., 6: 1497-1500.
- Kaehler, S. and R. Kennish, 1996. Summer and winter comparisons in the nutritional value of marine macroalgae from Hong Kong. Botanica Marina, 39: 11-17.
- Liekenjie, M.S.F., 1989. Fatty acids and glycerides. Nat. Prod. Rep., 6: 231-261.
- Marolia, V.J., S. Joshi and H.H. Mathur, 1982. Fatty acid composition of neutral lipids from some red algae. Indian J. Mar. Sci., 11: 102-103.
- Miller, J.P.A., 1962. Fats Steroids. In: Physiology and Biochemistry of Algae. Lewin, R.A. (Ed.), Academic Press, New York, pp: 92.
- Mishra, V.K., F. Temelli, P.F. Ooraikul and J.S. Shacklock Craigie, 1993. Lipids of the red alga *Palmaria palmate*. Botanica Marina, 36: 169-174.
- Nelson, M.M., C.F. Phleger and P.D. Nichols, 2002. Seasonal lipid composition in macroalgae of the North Eastern Pacific Ocean. Bot. Mar., 45: 58-65.
- Norziah, M.H. and Y. Ching Ch, 2000. Nutritional composition of edible seaweed *Gracilaria changgi*. Food Chem., 68: 69-76.
- Raymont, J.E.G., A. Austine and E. Lingfold, 1964. Biochemical studies on marine zooplankton. 1. The biochemical composition of *Neonysis interger*. J. Cons. Int. Explor. Mer., 28: 354-563.
- Reeta, J., 1993. Seasonal variation in biochemical constituents of *Sargassum wightii* (Grevillie) with reference to yield in alginic acid content. Seaweed Res. Utiln., 16: 13-16.
- Reeta, J. and G. Kulandaivelu, 1999. Seasonal variation in the biochemical constituents of *Gracilaria* spp. with reference to growth. Indian J. Marine Sci., 28: 464-466.
- Ricketts, T.R., 1966. On the chemical composition of some unicellular algae. Phytochemistry, 5: 67-76.
- Robledo, D. and Y.F. Pelegrin, 1997. Chemical and mineral composition of six potentially edible seaweed species of Yucatan. Botanica Marina, 40: 301-306.
- Roslin, S., 2003. Seasonal variations in the protein content of marine algae in relation to environmental parameters in Arockiapuram coast. Seaweed Res. Utiln., 25: 77-86.
- Sanchez-Machado, D.I., J. Lopez-Hernandez and P. Paseiro-Losada, 2002. High-performance liquid chromatographic determination of α -tocopherol in macroalgae. J. Chromatogr. A., 976: 277-284.
- Sanchez-Machado, D.I., J. Lopez-Cervantes, J. Lopez-Hernandez and P. Paseiro-Losada, 2004. Fatty acids, total lipid, protein and ash contents of processed edible seaweeds. Food Chem., 85: 439-444.
- Sasikumar, K., 2000. Studies on biochemical composition and heavy metal accumulation in seaweeds in the Vellar and Uppanar estuaries, southeast coast of India. M.Phil Thesis, Annamalai University, India.
- Stryer, L., 1988. Biochemistry. 3rd Edn., Stanford University, pp: 939.
- Takagi, T., M. Asahi and Y. Itabashi, 1985. Fatty acid composition of twelve algae from Japanese waters. Yukagaki, 34: 1008-1012.

- Vasanthi, V.H., G.V. Rajamanickam and A. Saraswathy, 2003. Fatty acid profile of some marine macroalgae in and around Rameswaram coastal waters. *Seaweed Res. Utilin.*, 25: 123-126.
- Venkatesalu, V., P. Sundaramoorthy, M. Anantharaj and M. Chandrasekaran, 2003a. Fatty acid composition of some marine macroalgae. *Seaweed Res. Utilin.*, 25: 95-98.
- Venkatesalu, V., P. Sundaramoorthy, M. Anantharaj and M. Chandrasekaran, 2003b. Fatty acid composition of some Rhodophycean marine macroalgae. *Phykos*, 41: 59-62.
- Venkatesalu, V., P. Sundaramoorthy, M. Anantharaj, M. Gopalakrishnan and M. Chandrasekaran, 2004. Studies on the fatty acid composition of marine algae of Rameshwaram coast. *Seaweed Res. Utilin.*, 26: 83-86.
- Wong, K.H. and P.C.K. Cheung, 2000. Nutritional evaluation of some subtropical red and green seaweeds. Part I-Proximate composition, amino acid profiles and some physico-chemical properties. *Food Chem.*, 71: 475-482.
- Wood, B.J.B., 1988. Fatty Acids and Lipids in Algae. In: *Microbial Lipids*. Ratledge, C. and S.G. Wilkinson (Eds.), Vol. 1, Academic Press, New York, pp: 807-867.