

CHEMICAL PROFILE OF *SARGASSUM TENERRIMUM* - AN INDIAN BROWN SEAWEED: SOURCE OF POTENTIAL INGREDIENT IN FUNCTIONAL MEAT PRODUCTS

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ABSTRACT

Brown seaweeds contain exclusive and enriched source of health promoting bioactive metabolites especially dietary fibre, antioxidants, essential amino acids, short chain fatty acids and minerals that helps in regulating and improving the gut microbiota and gut health. Threonine was found to be the predominant essential amino acids in S. tenerrimum (2.14±0.67 g/100g). The presence of n-3 long chain PUFA and the low ratio of n6:n3 fatty acid in S. tenerrimum (0.28) suggests it to serve as an ideal ingredient in preparation of low fat meat products. The Na/K ratio being 0.25 in S. tenerrimum could be of interest from a nutritional point of view as a balanced Na/K ratio is important to control hypertension. The mineral profile of S. tenerrimum suggests its suitability in preparation of low salt meat foods. The results of chemical profile of S. tenerrimum identifies it to act as an ideal functional source of amino acids, fatty acids, minerals and anti-oxidants in meat products which will help to alleviate the ill effects of nutritional deficiencies.

Keywords: *Sargassum tenerrimum*, Amino acid, Fatty acid, Minerals

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INTRODUCTION

Seaweeds are the excellent source of dietary fibre, dietary protein, essential fatty acids, vitamins and minerals, carotenoids, polysaccharides, polyphenols (Bhaskar and Miyashita, 2005; Meenakshi *et al.*, 2011) and various types of antioxidant metabolites that counteracts the environmental stresses (Lesser, 2006). Brown seaweed serves to be the exclusive and enriched source of

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health promoting bioactive metabolites like fucoxanthin, sulfated polysaccharides, phenolic compounds, terpenoids and bromophenols (Gupta and Abu Ghannam, 2011) especially short chain fatty acids helps in regulating and improving the gut microbiota and gut health (You *et al.*, 2019).

Commonly, the marine algae protein contains all essential amino acids (EAA) of nutritional importance. The amino acid (AA) profile is important for evaluating the nutritional value of algae proteins, but the digestibility of those proteins is the primary factor of the availability of their AA. Studies have showed that the digestibility seems to be limited by the algal non-protein fraction (Galland-Irmouli *et al.*, 1999).

Although seaweeds are not a conventional source of energy (their total lipid content is low compared to other foods), their polyunsaturated fatty acid contents can be as high as those of terrestrial vegetables (Sanchez-Machado *et al.*, 2004). They constitute to be the important source of n-3 and n-6 PUFA which are essential for the development of structural lipids and elements of cell membrane and are beneficial for the prevention of cardiovascular diseases such as arteriosclerosis and other chronic diseases, such as diabetes, hyper tension, and autoimmune diseases (Murata *et al.*, 2002; Nestel *et al.*, 2002; Erkkila *et al.*, 2003).

Macro algae are a very good source of minerals due to its cell wall polysaccharides and protein containing anionic carboxyl, sulphate, and phosphate groups which serves

as excellent binding sites for metal retention (Rocha *et al.*, 2009). Seaweeds contain 10 to 20 times more minerals than the land-based plants (Murugaiyan and Sivakumar, 2008) and is high enough (8–40%) to fulfill the recommended daily intakes of essential macro elements and trace elements for human nutrition (Rupérez, 2002). Algal-based minerals are in a highly assimilable form as they are integrated into living tissues. Seaweeds are considered a very good source of calcium which carries high potential as a functional food owing to higher calcium concentration and easy assimilation when compared to the calcium of cow's milk, (Mendis and Kim, 2011). Iron deficiency is one of the most prevalent nutritional deficiencies worldwide and effects of zinc deficiency on morbidity, mortality, growth and development are well known.

Utilisation of seaweed is limited to pharmaceutical industry in the production of phycocolloids such as agar-agar, alginate and carrageenan and not utilized as part of regular diet or for health aspects. It is thus an emerging thrust that policymakers should pay much more attention to improve diet quality through food-based approaches or by supplementation to address severe deficiency. The current research on *S. tenerrimum* chemical profile reflects its potential source as a functional ingredient which can be utilized cost effectively to allieviate the ill effects of nutritional deficiencies. Seaweeds could find its suitability in products requiring high salt concentrations during processing, like emulsion meat products by which the Na intake could be reduced.

MATERIALS AND METHODS

S. tenerrimum was collected from the ocean of Gulf of Mannar (Mandapam coast) (Fig. 1), transported in refrigerated condition in thermocol containers, washed several times in fresh water to remove the salt and extraneous contaminants, shade dried for 2-4 days, dried in hot air oven at 40°C for 5-6 hours, powdered in mixer and sieved using muslin cloth to obtain a fine powder and packed in sterile polyethylene pouches and stored at -20°C for analysis. The amino acid profile of *S. tenerrimum* was analyzed using the standard amino acids at different concentrations, provided in the kit (Hewlett Packard). The amino acid analysis was done following the procedure of Bruckner *et al.* (1991) in High Performance Liquid Chromatography and a chromatogram was obtained.

Fatty acid profile of *S. tenerrimum* was analysed using gas chromatography (Chemito GC 8610, India) fitted with a SPTM-2380 Capillary GC Column (L×I.D. 30 m×0.25 mm, df 0.20 µm film thickness; Nitrogen as carrier gas; column temperature of -99°C to 450°C with an accuracy of +0.05°C) and a flame ionization detector. Fatty acid analysis was carried out following the procedure of Wang *et al.*, (2009).

Sodium and potassium were estimated by Flame photometric method (AOAC, 2016); calcium and magnesium by EDTA Titrimetric method (BIS IS 5949: 1990 (R 2010); iron, zinc, manganese and copper by Atomic Absorption Spectrometry (AOAC, 2016) and Iodine by Iodometric

titration method (BIS IS 7224:2006 (AMD 2010) using Na₂S₂O₃ were estimated in *S. tenerrimum* using the standard operating procedures describing the method for the determination of the mineral. All the minerals were expressed in mg/100g with the exception of iodine expressed in mg/1000g.

RESULTS AND DISCUSSION

Amino acid profile

The amino acid profile of *S. tenerrimum* is presented in Table 1. *Sargassum tenerrimum* possessed the essential amino acids such as threonine, histidine, valine, leucine, phenylalanine, isoleucine and methionine in the order of decline. The non essential amino acids were present in the declining order of tyrosine, aspartic acid, glycine, alanine, arginine, serine and glutamic acid.

High concentrations of arginine, aspartic acid and glutamic acid are found in many seaweed species (Fleurence, 1999). Threonine was found to be the predominant EAA in *S. tenerrimum* (2.14 ± 0.67 g/100g) which was higher than that reported in *S. polycystum* (2.60 ± 0.16 mg/g) by Matanjan *et al.* (2009). Tyrosine (12.49 ± 0.52 g/100g) was the predominant nonessential amino acid reported in *S. tenerrimum* followed by aspartic acid, glycine and alanine while phenyl alanine was reported to be the predominant amino acid in *S. polycystum* (30.42 ± 4.43 mg/g) by Matanjan *et al.* (2009) and aspartic acid was predominant in *Fucus spiralis* (Peinado *et al.*, 2014). Asp and Glu were the most abundantly

occurring amino acids in the 34 species of red and brown algae tested and the EAAs such as Met, Cys, and Lys are found to be at low concentrations in the brown seaweeds (Dawczynski, *et al.*, 2007). Yaich *et al.*, (2011) and Dawczynski *et al.*, (2007) found that aspartic acid and glutamic acid constituted, a substantial amount of the total amino acids (26%) for green and brown seaweeds. Glutamic (8.08 mg/g; 0.68-1.35 g/100g; 0.15–1.65 mg/g) and aspartic acids (4.47 mg/g; 0.5- 1.2g/100g; 0.05–3.09 mg/g) were in abundance among the edible brown seaweeds (Matanjan *et al.*, 2009; Cofrades *et al.*, 2010; Peinado *et al.*, 2014). Alanine content in *S. tenerrimum* (1.78 g/100g) is high than that reported in *H.elongata* and *U.pinnatifida* (Cofrades *et al.*, 2010) and *Laminaria digitata* (Peinado *et al.*, 2014).

Fatty Acid Profile

The fatty acid profile of *S. tenerrimum* is presented in Table 2. *Sargassum tenerrimum* contain oleic acid as the predominant unsaturated fatty acid and palmitic acid as the predominant saturated fatty acid. The seaweed reported to contain lower saturated fatty acid and beneficial n3 and n6 fatty acids with lower ratio of PUFA/ SFA.

S. tenerrimum recorded palmitic acid (C16:0) as the most abundant saturated fatty acid and oleic (C18:0) as the most abundant unsaturated fatty acid as recorded by Sanchez Machado *et al.* (2004); Ortiz *et al.* (2006); Dawczynski, *et al.* (2007); Matanjan *et al.* (2009); Cofrades *et al.* (2010); Kumar *et al.* (2011); Yaich *et al.* (2011); Silva *et al.*

(2013); Peinado *et al.* (2014); Chen *et al.* (2016) in brown seaweeds. Saturated fatty acid content in *S. tenerrimum* (25.86%) was less than that observed in *S. polycystum* (51.30%) by Matanjan *et al.* (2009) and in the Spanish edible brown seaweeds (39%) by Cofrades *et al.* (2010) but higher than other species of *Sargassum* (2.9-7.3%) (Silva *et al.*, 2013).

The ratio of n6:n3 fatty acid in *S. tenerrimum* (0.28) matches the ratio observed by Cofrades *et al.* (2010) in brown seaweeds (0.24-0.27) and is lower than that observed by Matanjan *et al.*, (2009) in *S. polycystum* (0.98) and other species of *Sargassum* (0.4-0.8) (Silva *et al.*, 2013). This information reinforces the potential application of some brown macroalgae as dietary sources of polyunsaturated fatty acids. PUFA/SFA in *S. tenerrimum* (1.3) is in accordance with the brown seaweeds (1.1-1.2) observed by Cofrades *et al.* (2010) and higher than the value observed by Matanjan *et al.* (2009) in *S. polycystum* (0.39) and in *Sargassum sp* (0.05-0.2) by Silva *et al.* (2013). High proportion of essential fatty acids particularly n-3 polyunsaturated fatty acids (PUFAs) and arachidonic acid (C20:4n-6) and eicosapentaenoic acid (C20:5n-3) were found to be the foremost PUFAs in the brown seaweeds (Dawczynski *et al.*, 2007; Kumar *et al.*, 2011; Chen *et al.*, 2016).

The variations in the lipid composition among different algal strains are influenced by factors such as temperature, characteristics, intensity of light, levels of minerals, nitrogen compounds, and the

period in the life cycle of the algae (Takagi *et al.*, 1985).

Mineral Profile

The mineral profile of *S. tenerrimum* was presented in Table 3. *Sargassum tenerrimum* present macro minerals in the order of calcium, potassium, magnesium and sodium and micro minerals such as iron, zinc, manganese and copper. Iodine was present in minimal quantity.

Macro minerals such as sodium (114.33 ± 5.96), potassium (454.00 ± 9.63), calcium (760.50 ± 5.54), magnesium (189.77 ± 3.00) content in *S. tenerrimum* (mg/100g) were lower than that reported by Matanjun *et al.*, (2009) in *S. polycystum* (1362.13 ± 0.00 ; 8371.23 ± 0.01 ; 3792.06 ± 0.51 ; 487.81 ± 0.24) and by Ruperez (2002) in three brown seaweeds ($3818 \pm 43-7064 \pm 166$; $4322 \pm 46-11$, 579 ± 128 ; $931 \pm 38-1005 \pm 5$; $659 \pm 6-1181 \pm 34$). High sodium ($2.33 \pm 0.13\%$) was reported in *S. wightii* (Kumar *et al.*, 2015). Relatively higher K content compared to Na was observed in *S. tenerrimum* as reported by Kumar *et al.* (2011). The potassium content of *S. tenerrimum* (454.00 ± 9.63 mg/100g) was found to be lower than that recorded in *S. wightii* (18.7 mg/g; 32-50 mg/g) (Murugaiyan and Sivakumar., 2008; Kumar *et al.*, 2015). High sodium content (281.33 ± 102.63) and low potassium content (233.0 ± 30) were reported in *S. tenerrimum*. The Na/K ratio being 0.25 in *S. tenerrimum* could be of interest from a nutritional point of view as a balanced Na/K ratio is important for people who take diuretics to control hypertension and suffer from excessive

secretion of potassium (Ruperez, 2002). *S. tenerrimum* can therefore help to provide balanced Na/K ratio diets in contrast to olives with Na/K ratio of 43.63 and sausages with 4.89 (Ortego-Calvo *et al.*, 1993). The low sodium/potassium ratio of <1.5 were reported in brown seaweeds (0.33-1.34) by Ruperez (2002), in *S. polycystum* (0.16) by Matanjun *et al.* (2009); in red and brown edible Spanish seaweeds (0.48- 0.83) by Cofrades *et al.* (2010); in *S. wightii* (0.37) by Kumar *et al.* (2015). Seaweed fibers have the ability to retain sodium, which would thus be excreted in the feces, thus have a beneficial effect on blood pressure.

Calcium levels presented in *S. tenerrimum* was in accordance with the brown seaweeds reported by Cofrades *et al.* (2010). Calcium levels of *S. tenerrimum* was also found to be lower than *S. wightii* (30 to 45mg/g) reported by Kumar *et al.* (2015). Seaweeds are excellent sources of calcium for growing children, for the prevention and treatment of osteoporosis and for pre and post-menopausal women. Mg content in *S. tenerrimum* (189.77 ± 3.00 mg/100g) is in lower range than that observed by Cofrades *et al.* (2010) in the brown seaweeds studied (800mg/100g) but higher than *S. wightii* ($1.09 \pm 0.11\%$) reported by Kumar *et al.* (2015). High values (9-12 mg/g) of magnesium were reported in other *Sargassum species* (McDermid and Stuercke, 2003; Chan *et al.*, 1997). Mg in seaweeds helps to maintain the electrical potential of nerve and muscle cells and regulates the heartbeat.

Among the microelements present in seaweeds, Fe was the most abundant

trace element compared to green and red algae. It was recorded as the predominant trace mineral as reported by Kumar *et al.* (2011) in brown seaweeds (19.88 to 44.72 mg/100g). Iron (31.68 ± 1.39 mg/100g) and iodine (<2 ppm) in *S. tenerrimum* was found to be lower than that reported in *S. polycystum* (68.21 ± 0.03 ; 7.66 ± 0.10). Iron content in *S. tenerrimum* (31.68 ± 1.39) was higher than the values reported by Ruperez (2002) in three brown seaweeds (3.29 ± 0.54 - 7.56 ± 1.13) in *S. tenerrimum* (4.38 ± 6.94); in *S. wightii* (0.43 ± 0.36) by Kumar *et al.* (2015); in brown seaweeds (1.81-13.28) by Cofrades *et al.* (2010). Iron content was lower in other species of Sargassum (0.02–0.07mg/g) reported by Matanjun *et al.* (2009).

Zinc content of *S. tenerrimum* (2.19 ± 0.52 mg/100g) matched the recorded value in *S. polycystum* (2.15 ± 0.00) reported by Matanjun *et al.* (2009); lower than that recorded in brown seaweeds (3.77-6.08) by Cofrades *et al.* (2010); in *S. wightii* (1.28-1.84%) by Kumar *et al.* (2015), but slightly higher than three brown seaweeds (1.74 ± 0.37) reported by Ruperez (2002). Copper content (0.86 ± 0.40 mg/100g) was found to be in agreement with earlier reports (Topcuoglu *et al.*, 2003; Tuzen *et al.*, 2009) but was higher than *S. polycystum* (0.03 ± 0.00) reported by Matanjun *et al.* (2009) and by Ruperaz (2002) in the brown seaweeds; by Kumar *et al.* (2015) in *S. wightii* (0.027-0.03%). Manganese content in *S. tenerrimum* (1.78 ± 0.29) matched some brown seaweeds (van Netten *et al.*, 2000; Rupérez, 2002; Tuzen *et al.*, 2009) but higher than *S. wightii* (0.43 ± 0.23) studied. Manganese is considered an essential

nutrient in view of its function as an enzyme activator and because it is part of various metallo enzymes. Mn in *S. tenerrimum* (1.78 mg/100g) is in accordance with the brown seaweeds (0.85-4.09; 4.75 mg/100g) reported by Cofrades *et al.* (2010) and Kumar *et al.* (2011).

Essential minerals and trace elements present in seaweeds are found to vary according to seaweed species, geographical place of harvest, oceanic residence time, wave exposure, seasonal, environmental and physiological factors, type of processing and method of mineralization (Mabeau and Fleurence., 1993; Ruperez, 2002).

In addition to the amino acid, fatty acid and mineral enrichment in *S. tenerrimum*, it serves to be a rich source of dietary fiber and antioxidant activity. Dietary fibre content of *S. tenerrimum* was in higher order of magnitude (68.07 ± 1.64) (Vasanthi, 2021) than terrestrial plants. Anti-oxidant potential is witnessed in the previous research where it proved to have significantly higher ($P < 0.05$) total antioxidant activity (51.95 ± 0.19 ; 40.87 ± 2.58), total phenol content (36.49 ± 0.01 ; 34.70 ± 0.16) and FRAP (13.35 ± 1.22 ; 4.02 ± 0.37) activity in aqueous extraction than the methanolic extraction (Vasanthi *et al.*, 2020) with the IC₅₀ (mg/ml) of DPPH radical scavenging activity being observed at lower concentration of 800µg/ml in *S. tenerrimum* aqueous extracts. The hydrogen donating ability of the secondary metabolites such as phenolic compounds is attributed to the effect of anti-oxidant activity of *S. tenerrimum* as observed by Senapati *et al.* (2016).

CONCLUSION

Some authors (Cofrades *et al.*, 2008; Lopez-Lopez *et al.*, 2009) have reported that the seaweeds could be utilized as non-meat ingredients in formulating healthier meat products by its potential to overcome technological difficulties associated with low-salt products, including problems concerning water and fat binding properties and texture. The chemical profile of *S. tenerrimum* seaweed reveals it to be an ideal functional source of amino acids, fatty acids, minerals and anti-oxidants required for

healthy life style. The mineral profile proves its suitability in low salt meat foods and also meets the minerals for health. Apart from the health benefit effects of dietary fiber in seaweed, its incorporation in meat products will also help to improve the product yield on enhancing the water binding capabilities. On the other hand, the antioxidants in seaweed can help to enhance the shelf life of meat products by scavenging the oxidizing free radical molecules. The chemical profile of *S. tenerrimum* thus suggests that the seaweed can serve as a functional ingredient in the development of functional meat products.



Fig 1. *Sargassum tenerrimum* collected from the Gulf of Mannar, Mandapam Coast, Tamil Nadu

Table 1. Mean±SD of the amino acid profile of *Sargassum tenerrimum*

Essential Amino Acids	(%)	Non Essential Amino Acids	(%)
Histidine	1.32±0.77	Aspartic acid	3.39±0.66
Threonine	2.14±0.67	Glutamic acid	0.02±0.01
Methionine	0.45±0.42	Serine	1.37±0.30
Valine	0.88±0.25	Arginine	1.57±0.49
Phenylalanine	0.83±0.47	Alanine	1.78±0.25
Isoleucine	0.52±0.19	Glycine	2.14±0.27
Leucine	0.85±0.46	Tyrosine	12.49±0.52
Lysine	0.88±0.41		

Table 2. Mean±SD of the fatty acid profile of *Sargassum tenerrimum*

Saturated Fatty Acids	(%)	UnSaturated Fatty Acids	(%)
Myristic Acid	2.61±0.42	Oleic Acid	36.37±6.90
Palmitic Acid	18.79±0.59	Palmitoleic Acid	3.68±0.78
Stearic Acid	3.97±2.53	Linoleic Acid	22.41±8.23
Arachidic Acid	0.07±0.02	Linolenic Acid	7.11±2.76
Behenic Acid	0.42±0.35	Eicosopentaenoic Acid	3.26±3.10
Others	0.34±0.47	Docosohexaenoic Acid	0.92±0.74
ΣSFA	25.86	Σ MUFA	40.05
SFA:USFA	1:1.92	Σ PUFA	33.70
PUFA / SFA	1.30	n6: n3	0.28

Table 3. Mean±SD of the mineral profile of *Sargassum tenerrimum*

Macro Minerals	(mg/100g)	Micro Minerals	(mg/100g)
Calcium (mg/100g)	760.50±5.54	Iron (mg/100g)	31.68±1.39
Magnesium (mg/100g)	189.77±3.00	Zinc (mg/100g)	2.19±0.52
Sodium (mg/100g)	114.33±5.96	Manganese (mg/100g)	1.78±0.29
Potassium (mg/100g)	454.00±9.63	Copper (mg/100g)	0.86±0.40
		Iodine (mg/1000g)	<2

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CONFLICT OF INTERESTS

We certify that there is no conflict of interest among the authors and the financial organization with respect to the materials discussed in the manuscript.

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