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FATTY ACIDS PROFILES OF RED SEAWEED, GRACILARIA MANILAENSIS

ABSTRACT

Seaweed was known to have good fatty acids content. Fatty acids are the densest dietary source of energy and play an important role in our diet. Thus, this research was carried out to determine the fatty acids (FAs) composition of edible red seaweed, Gracilaria manilaensis. The lipid content was extracted using petroleum ether. Fatty acids were converted to methyl esters by transmethylation of lipid sample by sodium methoxide before determined by GC-FID. The finding showed that, G. manilaensis consist 35 of FAs. The saturated fatty acids (SFAs) of G. manilaensis accounted for 25.47% of total FAs, monounsaturated fatty acids (MUFAs) for 35.70% and polyunsaturated fatty acids (PUFAs) for 42.18%. The analysis data reveal that G. manilaensis contained approximately 92% of total FAs as long-chain fatty acids (C15-C24). The noteworthy proportion of omega-9 isomers is unique characteristic of G. manilaensis. Furthermore, G. manilaensis contained almost all of the most important essential fatty acids (EFAs) like linoleic acid (LA, C18:2ω6), arachidonoic acid (AA, C20:4ω6), α-linolenic acid (ALA, C18:3ω3), eicosapentanoic acid (EPA, C20:5ω3) and docosahexanenoic acid (DHA, C22:6ω3). Dihomogamma linolenic acid (DGLA, C20:3ω6, 22.86%) was found to be the most abundant fatty acid in G. manilaensis. The ratio of omega-6/omega-3 is 2.70, not exceed the recommended values in the diet by the WHO (< 10).

Keywords: Fatty acids, Gracilaria manilaensis and red seaweed

INTRODUCTION

Seaweeds are known as a valuable sources of protein, minerals, dietary fibers, vitamins, essential amino acids [1-6]. Besides, they were also known for excellent source of bio-active compounds such as fatty acid compounds [7-9]. Fatty acids are hydrocarbon chains with carboxyl group at the head end and a methyl group at the tail end. The carbons may be connected by single or double bonds. Fatty acids are important for human and animal health because they are precursor in the biosynthesis of eicosanoids, which are important bioregulators in many cellular process [10]. It was also reported that the fatty acid of certain seaweeds have anticancer [11], anti-inflammatory [12-13], antibacterial [14] and antimicrobial [15].

Seaweed contains up to 2% of dry weight of lipids [2-3]. PUFAs can account up to half of the lipid content, with much of it occurring in the form of omega-3 and omega-6 which are normally found in the fish oil. Rhodophyta was reported to have higher proportion of omega-6 than omega-3 [6]. Lower value of omega-6/omega-3 is important to prevent inflammatory, cardiovascular and nervous system disorders [8].

The fatty acid content of seaweed varies greatly and demonstrated a dependence on such factors as season and environmental growth conditions. Assay of fatty acids in food are commonly carried by gas chromatography. It is usually either for crude extract of sample to be derivatized to form fatty acid methyl esters, or for one-step transesterification with methanolic HCl to be followed by extraction with an apolar solvent [16].

Nutritional, biochemical and physicochemical properties composition of Malaysia seaweeds have been attract great interest among researcher [17-21]. But to best of our knowledge, the study of fatty acids profile of G. manilaensis is poor. The purpose of this study was to determine the fatty acids profile of G. manilaensis.

2. MATERIALS AND METHODS

2.1 Seaweed collection and preparation

Fresh G. manilaensis was collected from commercial culture pond in Kuala Muda, Kedah. The sample was washed in seawater to remove epiphytes, sand and other extraneous matter and rinse with distilled water. G. manilaensis was put in ice box and directly send to laboratory for further analysis.

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2.2 Fatty acid contents

2.2.1 Lipid extraction

Total fat of G. manilaensis was determined according to AOAC [22] official method 991.36. Briefly, 3 g of fresh G. manilaensis was extracted with 40 ml petroleum ether in boiling position for 25 min and in rinsing position for 30 min and dried in oven (125ºC) for 30 min.

2.2.2. Fatty Acids Methyl Esters (FAMEs)

Fatty acids were converted to methyl esters by transmethylation of lipid sample by sodium methoxide. 1 ml of hexane was put in crude lipid of G. manilaensis. 1 ml of sodium methoxide solution was added and mixture was vortex vigorously for 10-20 seconds. Solution was allowed to stand for 10 minutes to separate out the clear solution of FAMEs. Upper layer contained FAMEs were collected and injected into GC for the analysis of FAs. GC operation: Injector temperature = 250ºC, detector temperature = 280ºC, capillary column (non-polar stationary phase BPX70) diameter = 0.25 mm, the split ratio = 1:50, carrier gas (nitrogen) = 1.0 ml/min, injection volume = 1.0 µl of FAMEs. FAMEs peaks were identified by comparison of their retention times with those of a standard mixture (PUFAs).

3. RESULTS AND DISCUSSION

In the present study, the proportion of FAs in G. manilaensis can be classified into 3 major parts, which are saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs). Seaweed has different fatty acids profile than terrestrial plants as they have higher proportion of saturated and unsaturated FAs [2]. Fatty acids composition in seaweed varied depend on the species, environmental growth conditions and seasonal period [8,23].

Even though, G. manilaensis have low content of lipid (0.175% DW w/w), but concentration of PUFAs were relatively high (42.18%). The analysis data reveal that G. manilaensis contained approximately 92% of total FAs as long-chain fatty acids (C15-C24). The proportion of short-chain (C4-C10) and middle-chain (C11-C14) FAs were generally low.

The concentration of SFAs are 25.42% of the total FA profile, which is slightly lower than the reported values for Gracilaria sp. range from 31.03% to 78.16% [6]. Interestingly, pentadecanoic acid (C15:0, 5.12%) is the most abundant SFAs in G. manilaensis and quite higher than the reported value for Gracilaria sp. (0.45% - 0.8%) [6]. G. manilaensis has moderate content of palmitic acid (4.45%). The value is relatively much lower than the reported value for the same genera although for different species – G. changii (26%) [17], G. edulis (84.60%) and G. folifera (81.28%) [7]. In contrast to what is described in the literature, palmitic acid present as the predominant SFAs in most seaweed – Porphyra sp. (30.8%) and Laminaria sp. (36%) [3], Caulerpa sp. (2.87% - 15.55%) [24]. John et al. [25] reported that the main content of SFAs is palmitic acid which is in green seaweed (23.9%), brown seaweed (27.9%) and red seaweed (33.8%). Kaneniwa et al. [26] also reported that C16:0 is the most abundant SFAs in 9 species algae from Japan. Chu et al., [18] reported that, seaweeds along coast of the Peninsular Malaysia to have C16:0 as predominant SFAs ranged from 51.2% to 84.4%. Low intake of SFAs and an increased PUFA-to-SFA ratio are associated with a lower risk of human coronary heart disease. In present study, this ratio was found to be 1.66, ≥0.4 and within nutritional guidelines that recommend ratio above 0.4 [27].

Among the MUFAs, omega-9 isomers were predominant and quite unique characteristics of G. manilaensis. The most Malaysian seaweeds showed the existence of omega-9 isomer. C18:1n9t is the predominant in G. changii [17], C18:1n9c is the predominant in C. lentillifera and S. polycystum [14] and C22:1n9 is the predominant in D. dichotoma [21]. In fact, G. manilaensis has showed the presence of MUFAs with chain length more than C21. These results were also found in Gracilaria sp [7], Caulerpa sp [24], Porphyra sp and Laminaria sp [3].

In the present study, the high amount of PUFAs (42.18%) were mostly contribute by omega-3 and omega-6 FAs. G. manilaensis contained almost all of the most important EFAs like LA, AA, ALA, EPA and DHA, except for adrenic acid. EPA is the most dominant EFAs with 8.26% followed by LA and trace amounts of DHA. Kumar et al. [6], reported that EPA and AA were the foremost PUFAs in rhodophyta and phaeophyta. EPA was reported to have potential to reduce the risk of heart disease, thrombosis and atherosclerosis [28].

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Even the content of DHA in G. manilaensis is low (0.40%), but it is still good because DHA is often not found in red seaweed or when present exist at low concentration [3,7,24,37]. Study by van Ginneken et al. [8] on macroalgae from north Atlantic and tropical seas, only S. natans was detected to have DHA proportion (16%). In contrast, Norziah and Ching [17] reported that G. changii to have high content of DHA (12.9%). EPA (8.26%) is the second highest EFAs in G. manilaensis and comparable with the other Malaysia seaweed [17,20-21]. High content of DHA and EPA is one of general characteristic of red seaweeds, which suggest that red seaweed may be the best source of this nutritional FA [8]. Apart of that, DHA and EPA are the important "fish" FAs, which is command found in fish oil. However, it is noteworthy that the original source of these long-chain PUFAs is not from the fish itself, but marine alga and phytoplankton which form their major dietary.

Apart from this, DGLA was the most abundant FAs (22.86%) in G. manilaensis. Interestingly, to best of our knowledge, this high level of DGLA was first reported on Gracilaria sp [6-7,17] and other seaweeds. This particular fatty acid has been reported to be the precursor for the synthesis of prostaglandin PGE1 [29] and a number of other related biologically substance [30].

The proportion of omega-6 was higher than omega-3. Similar results were found in other analyses of Gracilaria sp [6-7,17]. The ratio omega-6/omega-3 is 2.70, not exceed the recommended values in the diet by the WHO (< 10) [31]. This value is lower than G. cortica (12.35), G. dura (26.65), G. debilis (18.82) and G. fergusonii (18.65) [6]. Lower value of omega-6/omega-3 is important to prevent inflammatory, cardiovascular and nervous system disorders [8].

4. CONCLUSION

G. manilaensis has a good fatty acids profile with the present of the essential fatty acid like LA, AA, ALA, EPA and DHA. The ratio of omega-6/omega-3 is low, not exceeded the WHO (<10) recommended value. Thus, G. manilensis could be a potential food and feed resources for human.

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Figure 1: GC-FID chromatogram of fatty acids profile in G. manilaensis

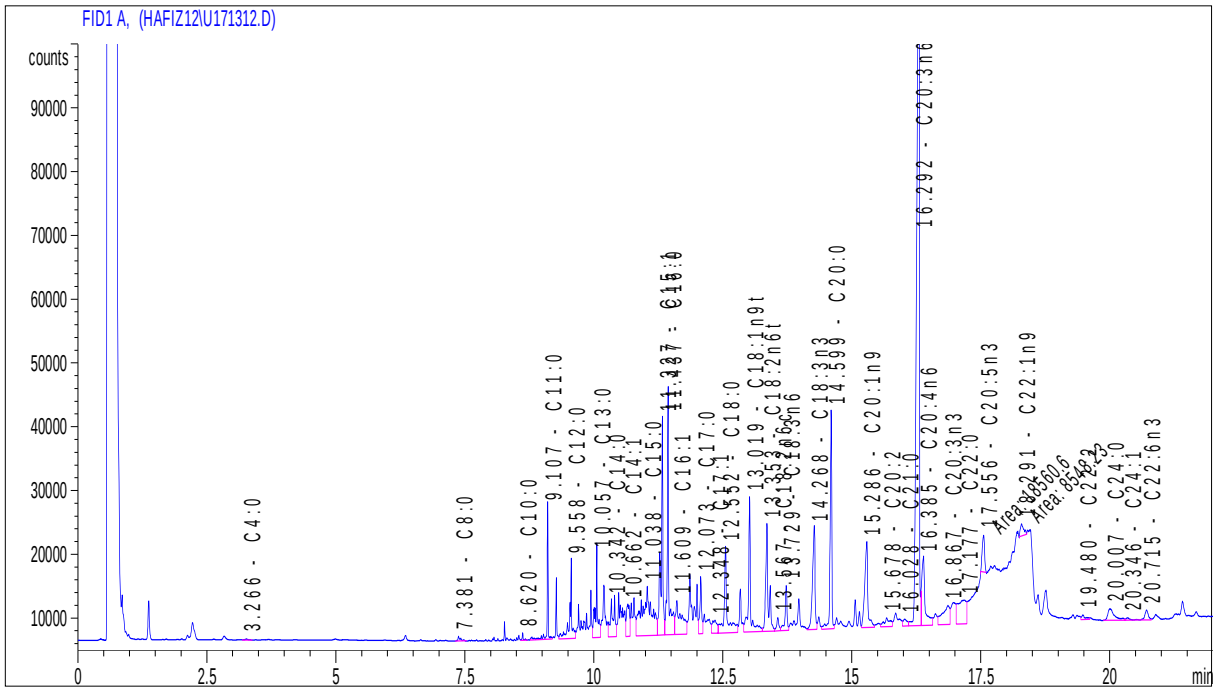

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Table 1: Fatty acid profiles of Gracilaria manilaensis (% of total FAs w/w) and some edible seaweeds (from Sabah, Malaysia)

Carbon no.	Fatty Acids	Gracilaria manilaensis[a]	Dictyota dichotama[21]	Sargassum granuliferum[21]
Saturated Fatty Acids				
C4:0	Butryic	0.711	NR	NR
C8:0	Caprylic	0.133	ND	ND
C10:0	Capric	0.089	1.2	ND
C11:0	Undeconoic	1.519	ND	ND
C12:0	Lauric	0.581	ND	ND
C13:0	Tridecanoic	1.926	ND	ND
C14:0	Myristic	1.606	3.2	4.6
C15:0	Pentadecanoic	5.214	0.5	0.4
C16:0	Palmitic	4.451	19.8	32.6
C17:0	Heptadecanoic	1.167	ND	ND
C18:0	Stearic	0.957	5.3	0.7
C20:0	Arachidic	0.231	1.6	0.6
C21:0	Henicosanoic	0.335	ND	ND
C22:0	Behenic	2.319	ND	0.6
C24:0	Lignoceric	0.881	ND	1.3
Monounsaturated Fatty Acids				
C14:1	Myristoleic	1.121	0.2	0.3
C15:1	Cis-10-Pentadecenoic	4.448	0.6	2.9
C16:1	Palmilatoeic	2.227	0.9	10.4
C17:1	Cis-10-Heptadecanoic	2.932	0.3	0.9
C18:1ω9t	Elaidic	3.169	2.6	13.6
C18:1ω9c	Oleic	ND	2.4	7.0
C20:1ω9c	Cis-11-Eicosenoic	3.042	NR	NR
C22:1ω9	Erucic	18.644	17.7	2.3
C24:1	Nervonic	0.114	1.5	ND
Polyunsaturated Fatty Acids				
C18:2ω6t	Linoleic acid (trans)	3.269	0.9	1.4
C18:2ω6c	Linoleic acid (cis)	0.341	2.3	7.0
C18:3ω6	ɣ- Linolenic acid	1.044	0.1	ND
C18:3ω3	α-Linolenic acid	0.908	5.4	ND
C20:2	Cis-11-14-Eicosenoic	0.611	0.9	0.2
C20:3ω6	Cis-11,8,14-Eicosatrienoic	22.856	3.7	ND
C20:4ω6	Arachidanoic acid	2.698	3.5	1.2
C20:3ω3	Cis-11,14,17-Eicosapentaenoic acid	1.593	8.4	1.6
C20:5ω3	Cis-5,8,11,14,17-Eicosapentaenoic	8.264	6.4	1.9
C22:2	Cis-13-16-Docisadienoic acid	0.192	2.9	ND
C22:6ω3	Cis-14,7,10,13,16,19-Docosahexaenoic acid	0.404	7.0	4.7
	SFA	25.42	31.6	40.6
	MUFA	35.70	26.9	41.4
	PUFA	42.18	41.6	18.0
	C20 PUFA	36.02		

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	C18 PUFA	5.56		
	ω9MUFA	24.86	22.7	22.9
	ω6PUFA	30.21	10.5	9.6
	ω3PUFA	11.17	27.2	8.2
	ω6/ω3 PUFA	2.70	0.39	1.17

a – mean of three determinations
NR – not recorded
ND – not detected
[21] – Bakar et. al. (2012)

Nor Salmi Abdullah[*1], Shamsul Muhamad[1], Ibrahim Che Omar[1] and Hasmah Abdullah[2]

[1] Faculty Agro-based Industry, University Malaysia Kelantan, Kampus Jeli, Locked Bag No. 100, 17600 Jeli, Kelantan, MALAYSIA.
[2]School of Health Sciences, Health Science, University Sains Malaysia, 16150 Kubang Kerian, Kelantan, MALAYSIA.

Corresponding author:
Nor Salmi Abdullah, M. Sc.*
Affiliation: Researcher, PhD Student
Research Field: Biotechnology Industry
University Malaysia Kelantan,
Kampus Jeli,
Locked Bag No. 100,
17600 Jeli,
Kelantan, MALAYSIA.