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Fatty Acid Compositions of Nineteen Species of Marine Algae Mainly Obtained from the Yamaguchi Prefecture Coast

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The fatty acid compositions of five species of Chlorophyta, six species of Phaeophyta and eight species of Rhodophyta mainly obtained from the Yamaguchi prefecture coast were examined by capillary column gas chromatographic analysis. All of the samples contained of 16:0 more than 10% of the total fatty acid. The major fatty acids other than 16:0 were 16:3n-3, 16:4n-3, 18:2n-6, 18:4n-3 and 22:5n-3 in Chlorophyta, 14:0, 16:1n-7, 18:1n-9, 18:1n-7, 18:2n-6, 18:3n-3, 20:4n-6 and 20:5n-3 in Phaeophyta, and 14:0, 16:1n-7, 18:1n-9, 18:1n-7, 20:4n-6 and 20:5n-3 in Rhodophyta.

The carbon number of the major polyunsaturated fatty acids of Chlorophyta were $C_{16}$and C_{18}, C_{18} and C_{20} in Phaeophyta, and C_{20} in Rhodophyta. The algae examined in this study revealed characteristic fatty acids feature in the class level.

In the gas chromatographic analysis of the fatty acids of *Codium fragile* and *Laurencia okamurae*, a high content of unknown components was detected. These unknown components comprised 10% (*C. fragile*) and 16% (*L. okamurae*) of the total fatty acids.

1 Introduction

There are many marine algae flourished in Japanese waters. Some of them are used as food or industrial materials, but many species of marine algae have not been available for industrial use. These unavailable marine algae are expected for new fisheries resources, and it is important to know the chemical components and contents of the marine algae for utilization.

On the other hand, it has become apparent that various marine algae can produce useful fatty acids such as arachidonic acid (20:4n-6) and icosapentaenoic acid (20:5n-3)[1−3]. Especially, it is known that *Gracilaria aciatica* (Japanese name, "Ogonori") contains 20:4n-6,[1−2] and produces large amount of prostaglandin E_2 from 20:4n-6 by some stimuli such as cutting or soaking in fresh water[4−5]. New biologically active lipid resources like these marine algae have been developed in many laboratories. There are several studies on the lipids of marine algae[6−10]. We examined detail fatty acid compositions of nineteen species of marine algae mainly obtained from coast of Yamaguchi prefecture by the capillary column gas chromatography (GC) for the utilization of algae and development of new lipid resources.

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2 Materials and Methods

The samples of algae used are shown in Table 1. Marine algae were identified by professor Hitoshi Kito and Noboru Murase, National Fisheries University. The contents of water were determined by drying method at 105℃ until reached constant weight. The total lipids of the algae were extracted according to the procedure of Bligh and Dyer[11]. The lipids were converted to fatty acid methyl esters according to the procedure of Prevot and Mordret[12], and furthermore, the free fatty acids in the lipids were methylated by diazomethane. The methyl esters obtained were purified by a thin layer chromatography (TLC) with Kiesel gel 60G plate of 0.5mm thickness by developing with n-hexane/ether (85:15, v/v). The capillary column GC of the methyl esters was conducted with a Shimadzu GC14A instrument (Shimadzu Seisakusho Co.), with a flame ionization detector on a fused-silica capillary column coated with Silar 5CP (50m x 0.32mm i.d.). The carrier gas was N_2. The column, and the injector and detector temperatures were 210℃ and 230℃, respectively. Each peak area percentages were calculated with Shimadzu integrator C-R2A. Each peak on the gas chromatogram was identified on the basis of the agreement of relative retention time with those of authentic specimens. The log plot procedures and systematic separation factor procedures[13] were used concurrently for the identification.

3 Results and discussion

The fatty acid compositions from nineteen species of marine algae are shown in Table 2 – 4. Saturated fatty acids occupied 21-54% in total fatty acids. The ratio of total monoenoic fatty acids in total fatty acids were 4-20% in Chlorophyta and Rhodophyta, and 22-36% in Phaeophyta, respectively, and those of polyunsaturated fatty acids were 30-70% in Chlorophyta, 28-37% in Phaeophyta, and 27-66% in Rhodophyta, respectively.

All the samples contained 16:0 as one of the major fatty acids and the ratio in total fatty acids was more than 10%. The major fatty acids other than 16:0 were 16:3n-3, 16:4n-3, 18:1n-9, 18:1n-7, 18:2n-6, 18:3n-3, 18:4n-3 and 22:5n-3 in Chlorophyta (Table 2), 14:0, 16:1n-7, 18:2n-6, 20:4n-6 and 20:5n-3 in Phaeophta (Table 3), and Rhodophyta were 14:0, 16:1n-7, 18:1n-9, 18:1n-7, 20:4n-6 and 20:5n-3 (Table 4), respectively. The total amounts of major fatty acids, described above, were up to 70% of the total fatty acids in all samples.

Carbon number of major polyunsaturated fatty

Table 1. Species of marine algae and contents of water and total lipids

Class	Japanese Name	Scientific Name	Date	Place	Water contents(%)	Total Lipid Contents(%) *
Chlorophyta	Anaaosa	*Ulva pertusa*	31, Jul., 1996	Yoshimo, Yamaguchi	76.1	4.0
	Usubaaonori	*Enteromorpha linza*	21, May, 1996	Kawatana, Yamaguchi	83.8	8.5
	Hitoegusa	*Monostroma nitidum*	21, May, 1996	Kogushi, Yamaguchi	74.1	1.7
	Fusaiwaduta	*Caulerpa okamurae*	31, Jul., 1996	Yoshimo, Yamaguchi	95.8	4.9
	Miru	*Codium fragile*	27, May, 1996	Yoshimo, Yamaguchi	94.5	5.1
Phaeophyta	Kurome	*Ecklonia kurome*	21, Apr., 1996	Kiwado, Yamaguchi	80.7	2.8
	Arame	*Eisenia bicyclis*	30, May, 1996	Yoshimo, Yamaguchi	83.3	2.7
	Wakame	*Undaria pinnatifida*	21, May, 1996	Kogushi, Yamaguchi	93.1	4.7
	Hijiki	*Hizikia fusiformis*	21, May, 1996	Kogushi, Yamaguchi	88.1	3.7
	Habanori	*Petalonia binghamiae*	3, Jun., 1996	Kawatana, Yamaguchi	80.9	4.6
	A species of Hibamata	*Ascophyllum nodosum*	25, Sep., 1996	Nahant, Mass., U.S.A.	40.4	3.4
Rhodophyta	Susabinori	*Porphyra yezoensis*	12, Dec., 1995	Yanagawa, Fukuoka	77.7	13.8
	Ogonori	*Gracilaria asiatica*	21, May, 1996	Kawatana, Yamaguchi	84.7	4.6
	Tsukumonori	*Nemalion multifidum*	3, Jun., 1996	Kawatana, Yamaguchi	93.4	3.3
	Tsunomata	*Chondrus occellatus*	28, May, 1996	Yoshimo, Yamaguchi	76.5	1.2
	Fukurofunori	*Gloiopeltis furcata*	3, Jun., 1996	Kawatana, Yamaguchi	84.5	1.9
	Makusa	*Gelidium elegans*	3, Jul., 1996	Kawatana, Yamaguchi	72.4	2.9
	Mitsudesozo	*Laurencia okamurae*	21, May, 1996	Yoshimo, Yamaguchi	90.7	9.0
	A species of Itogusa	*Polysiphonia ianosa*	25, Sep., 1996	Nahant, Mass., U.S.A.	71.5	6.5

* Total lipid contents were presented in the percentage ot dry matter.

Table 2. Fatty acid composition of the lipids of Chlorophyta (%)

Fatty acid * 1	RRT * 2	ECL * 3	*U. pertusa* (Anaaosa)	*E. linza* (Usubaaonori)	*M. nitidum* (Hitoegusa)	*C. okamurae* (Fusaiwaduta)	*C. fragile* (Miru)
14:0	0.288	14.00	0.3	0.8	2.2	2.6	1.6
16:0	0.547	16.00	34.1	30.7	17.2	26.3	34.6
16:1n-7	0.618	16.40	1.7	2.5	1.6	4.3	2.5
16:2n-9	0.708	16.85	1.2	0.7	0.1	6.0	1.4
Unknown	0.806	17.30	—*4	—	—	—	9.8
16:3n-3	0.866	17.52	1.2	0.9	2.9	11.4	—
16:4n-3	0.931	17.76	4.5	13.8	14.9	—	—
18:0	1.000	18.00	0.2	0.3	1.5	0.6	1.2
18:1n-9	1.119	18.37	3.3	0.3	1.2	1.5	12.5
18:1n-7	1.143	18.44	12.5	7.6	0.7	1.9	
18:2n-6	1.323	18.92	17.4	6.0	2.6	16.2	5.1
18:3n-6	1.457	19.23	1.0	1.1	0.1	1.0	1.9
18:3n-3	1.645	19.63	12.4	17.7	26.7	14.3	15.4
18:4n-3	1.802	19.93	2.7	9.3	14.6	0.5	1.3
20:4n-6	2.824	21.41	2.3	0.9	0.1	3.8	2.3
20:4n-3	3.233	21.86	0.1	0.7	1.2	0.3	0.2
22:0	3.372	22.00	0.4	0.7	1.2	0.1	2.6
20:5n-3	3.435	22.06	0.6	1.8	1.4	2.9	1.8
24:0	6.117	23.97	-	-	0.6	1.0	1.4
22:5n-3	6.425	24.13	0.8	1.8	5.4	0.4	0.3
Others			3.2	2.6	3.7	5.0	4.1
Sat.			35.0	32.4	22.8	30.7	41.3
Mono.			17.5	10.4	3.6	7.7	15.0
Poly			44.3	54.6	70.0	56.6	29.8
Unknown			—	—	—	—	9.8

*1 Sat.: Saturated fatty acids, Mono.: Monoenoic fatty acids, Poly.: Polyunsaturated fatty acids.
*2 Relative retension time.
*3 Equivalent chain length.
*4 Not detected.

acids of Chlorophyta were C_{16} and C_{18}, Phaeophyta were C_{18} and C_{20}, and Rhodophyta were C_{20}. These features of fatty acids composition among the phyta were similar to the results of previous papers reported on the fatty acids in 10 species of Chlorophyta, 25 species of Phaeophyta and 22 speceis of Rhodophyta, that collected in Australian waters, Scotland coasts and Japanese waters [1-3, 6-10]. Whereas C16:3 n-3 and C16:4 n-3 were not detected in fatty acids of *Codium fragile* (Chlorophyta) in present study. In the present study, unknown peaks were detected in the gas chromatograms of *C. fragile*, *Ascophyllum nodosum* and *Laurencia okamurae*. The component percentages in the unknown peaks comprised 10% (*C. fragile*), 4% (*A. nodosum*) and 16% (*L. okamurae*) to their total fatty acids. The structural analysis of the component has not been done. But it is presumed that the structure of these components are *trans* monoenoic, odd-chain or branched-chain fatty acids from the retention data of gas chromatograms. The existence of *trans* 16:1n-13 in *G. asiatica* was described Araki *et al.* [2].

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Table 3. Fatty acid composition of the lipids of Phaeophyta (%)

Fatty acid *1	RRT *2	ECL *3	*E. kurome* (Kurome)	*E. bicyclis* (Arame)	*U. pinnatifida* (Wakame)	*H. fusiformis* (Hijiki)	*P. binghamiae* (Habanori)	*A. nodosum* (A species of Hibamata)
12:0	0.151	12.00	4.0	0.3	0.0*5	0.5	0.0	0.0
Unknown	0.254	13.61	1.5	0.4	—*4	0.3	—	—
Unknown	0.271	13.91	—	—	—	1.2	—	—
14:0	0.288	14.00	5.9	7.0	5.6	3.0	8.5	9.3
16:0	0.547	16.00	24.4	23.1	36.0	33.5	30.3	11.1
16:1n-7	0.618	16.40	7.9	10.3	1.2	5.3	1.1	1.5
18:0	1.000	18.00	0.7	0.7	3.3	0.5	1.0	0.5
18:1n-9	1.119	18.37	13.9	13.5	21.6	8.1	21.1	—
18:1n-7	1.143	18.44	—	0.6	—	—	1.3	34.0
18:2n-6	1.323	18.92	6.0	5.1	5.9	3.6	5.8	7.1
18:3n-6	1.457	19.23	1.2	0.8	0.8	0.3	0.3	0.3
18:3n-3	1.645	19.63	2.3	3.4	1.6	6.9	2.8	2.8
18:4n-3	1.802	19.93	3.0	4.7	2.3	2.2	2.3	2.4
20:0	1.840	20.00	2.2	1.7	1.4	0.3	1.8	0.2
20:1n-9	2.048	20.35	0.2	0.4	—	2.6	0.1	0.2
20:1n-7	2.169	20.54	—	—	—	1.4	—	—
20:2n-6	2.415	20.90	0.3	0.2	0.2	0.2	0.3	1.8
Unknown	2.584	21.12	—	0.1	—	0.1	0.1	3.8
20:3n-6	2.671	21.23	0.9	0.4	1.2	0.5	0.5	0.5
20:4n-6	2.824	21.41	16.0	17.5	14.0	13.8	9.1	12.8
20:4,5,11,14,17	3.139	21.76	—	—	—	0.1	—	1.3
20:5n-3	3.435	22.06	3.5	4.5	3.8	5.7	7.0	6.8
22:1n-9	3.680	22.28	—	—	—	4.0	—	0.3
22:5n-3	6.425	24.13	—	—	—	—	—	1.1
Others			6.1	5.3	1.0	5.8	6.4	2.1
Sat.			37.3	32.8	46.2	37.8	41.7	21.1
Mono.			22.0	24.8	22.9	21.5	23.6	36.0
Poly.			33.2	36.6	29.9	33.3	28.1	37.0
Unknown			1.5	0.5	—	1.6	0.1	3.8

*1 Sat.:Saturated fatty acids, Mono.: Monoenoic acids, Poly.: Polyunsaturated fatty acids.
*2 Relative retension time.
*3 Equivalent chain length.
*4 Not detected.
*5 <0.05%

Table 4. Fatty acid composition of the lipids of Rhodophyta (%)

Fatty acid *1	RRT *2	ECL *3	*P. yezoensis* (Susabinori)	*G. asiatica* (Ogonori)	*N. multifidum* (Tsukumonori)	*C. occellatus* (Tsunomata)	*G. furcata* (Fukuro funori)	*G. elegans* (makusa)	*L. okamurae* (Mitsudesozo)	*P. ianosa* (A species of Itogusa)
12:0	0.151	12.00	0.1	0.2	1.0	0.1	0.1	0.2	0.1	0.1
iso−14:0	0.241	13.52	—*4	0.0*5	—	—	0.0	—	2.2	0.0
14:0	0.288	14.00	0.1	5.2	3.7	5.7	2.8	4.3	7.3	3.3
Unknown	0.422	15.23	—	—	—	—	—	—	2.5	—
16:0	0.547	16.00	22.0	40.0	35.1	46.9	26.1	40.9	30.8	32.1
16:1n−7	0.618	16.40	3.6	0.7	1.7	1.2	1.2	1.1	1.4	8.3
17:0	0.753	17.00	—	—	0.0	0.1	0.0	0.1	0.2	6.4
Unknown	0.774	17.16	—	—	—	—	—	—	6.9	—
18:0	1.000	18.00	0.5	0.7	0.6	1.1	2.1	1.0	0.4	0.5
18:1n−9	1.119	18.37	1.7	4.3	5.3	12.6	16.1	6.3	5.9	2.2
18:1n−7	1.143	18.44	0.4	1.0	1.9	—	2.4	—	2.5	6.7
18:2n−6	1.323	18.92	1.6	0.8	0.7	0.7	1.3	0.5	0.9	4.0
18:3n−6	1.457	19.23	0.4	0.6	0.2	0.5	0.4	0.2	0.5	0.6
Unknown	1.543	19.42	—	—	0.0	—	—	—	6.9	—
18:3n−3	1.645	19.63	0.1	0.0	0.2	0.1	0.3	0.1	0.1	1.2
18:4n−3	1.802	19.93	0.3	—	0.1	0.2	0.1	0.1	0.1	1.1
20:1n−9	2.048	20.35	2.9	—	—	0.0	0.1	—	—	—
20:2n−6	2.415	20.90	1.0	0.2	0.1	—	—	—	0.1	—
20:3n−6	2.671	21.23	1.9	3.8	1.3	0.6	1.3	0.3	0.7	0.6
20:4n−6	2.824	21.41	3.4	41.2	5.7	17.7	11.8	20.8	6.9	9.2
20:5n−3	3.435	22.06	57.5	0.1	37.2	11.6	30.5	20.4	17.9	20.7
Others			2.4	1.4	5.1	1.1	3.5	3.9	5.7	3.1
Sat.			22.8	46.0	40.5	53.8	31.0	46.4	41.0	42.4
Mono.			8.6	6.0	8.9	13.9	19.8	7.4	9.8	17.2
Poly.			66.2	46.7	45.5	31.2	45.8	42.3	27.1	37.4
Unknown			—	—	0.0	—	—	—	16.4	—

*1 Sat.: Saturated fatty acids, Mono.: Monoenoic fatty acids, Poly.: Polyunsaturated fatty acids.
*2 Relative retension time.
*3 Equivalent chain length.
*4 Not detected.
*5 <0.05%

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主に山口県の沿岸から採取した海藻類の脂肪酸組成

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主に山口県沿岸で採取した19種の海藻類の脂肪酸組成をキャピラリーカラムガスクロマトグラフィーによって明らかにした。本研究で試料として用いた全ての海藻類は10%以上の16:0を含んでいた。また16:0以外の脂肪酸では，緑藻類では，16:3n-3，16:4n-3，18:1n-9，18:1n-7，18:2n-6，18:3n-3，18:4n-3，22:5n-3，褐藻類では14:0，16:1n-7，18:2n-6，20:4n-6，20:5n-3，紅藻類では14:0，16:1n-7，18:1n-9，18:1n-7，20:4n-6，20:5n-3がそれぞれ主要な脂肪酸であった。高度不飽和脂肪酸は，緑藻類では炭素数16及び18のもの，褐藻類では炭素数18及び20のもの，紅藻類では炭素数20のものが多かった。*Codium fragile* 及び *Laurencia okamurae* のガスクロマトグラフィー分析において，全脂肪酸中それぞれ約10%及び約16%もの未同定成分が検出された。