Production and Use of Marine Algae in Japan

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Abstract

About 200 kinds of marine algae grow in the ocean surrounding Japan. Moreover, a large amount of marine algae is cultivated domestically for use as foods and industrial materials. The production of marine algae was 639,210 t, amounting to \(\frac{1}{2}\)164,642 million in 1998. The marine algae used as foods consist of about 50 species, and the production was about 172,000 t in 1996. Japanese have eaten marine algae as a source of supply of minerals and vitamins. However, the dietary intake of marine algae is about 1/5 compared with that of fish. Recently, it has been suggested that the increase in the incidence of adult diseases in Japan, such as diabetes, hypertension, hyperlipidemia, etc. was caused by the decrease in the dietary intake of fish, shellfish and marine algae. The nutritional role of marine algae was examined and it was recognized that marine algae contribute to the prevention and treatment of various diseases. We also analyzed the effect of *Undaria pinnatifida* (Wakame) on the decrease of the concentration of serum and liver triacylglycerol in rats.

Discipline: Fisheries

Additional key words: seaweed, Undaria pinnatifida (Wakame), functional food materials,

health food

Introduction

Since Japan is a small country surrounded by the sea, the relations between the Japanese and the sea have always been very close since olden times, including the supply of food from the sea. Fish, shellfish, and marine algae are valuable foods for the Japanese people. Especially, marine algae play an important role in the maintenance of health as a source of supply of vitamins and minerals.

Since various kinds and large amounts of marine algae are produced in the sea around Japan, marine algae are used as foods and also as industrial products in Japan. Therefore, the production and consumption of marine algae in Japan are the highest in the world.

Moreover, physiologically active substances have been extracted from various marine algae and their physiological properties have been studied. The development of an assay and separation methods for useful elements from marine algae should enhance the value of marine algae in the future. Marine algae may become a valuable resource worldwide as a source of supply of nutrients for the maintenance of health and extraction of various elements for industries in future.

Historical background

Japan is surrounded by the North Pacific Ocean, East China Sea, Sea of Japan and Sea of Okhotsk. The Japan current (Kuroshio), a warm ocean current, flows northward, while the cold Okhotsk current originates in polar waters and flows southward along Hokkaido. Since most of the coasts are rocky, they provide a firm growth base for the algae. The variety of marine environments results in a variety of algal species. In Japan, marine green algae consist of about 200 species including *Enteromorpha* sp., *Ulva* sp., *Monostroma* sp., brown algae consist of about 270 species including *Laminaria* sp., *Undaria* sp., *Nemacystus* sp., *Hizikia* sp., *Eizenia* sp., *Ecklonia* sp., and red algae consist of about 670 species including *Porphyra* sp., *Gelidium* sp., *Gracilaria* sp.¹⁰⁾.

Japan islands have been inhabited for more than 100,000 years and algae were identified in kitchen midden, suggesting that they were used as foods, salt and iodine resources. After the Nara period, the law books "Taihou-rituryou (in 701 AD)" and "Engi-siki (in 927 AD)" indicated that processed marine algae were paid as

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Fig. 1. Undaria pinnatifida (Wakame)

The part of Wakame used for food is the sterile leaf of the asexual generation and wakame is marketed in a dried or salted state for long preservation.

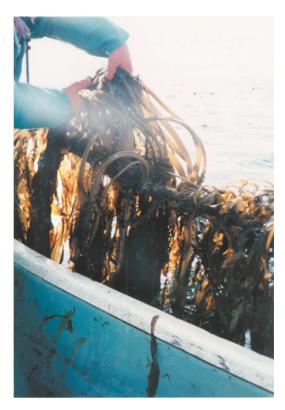


Fig. 2. Cultured Wakame at Shiogama bay in Miyagi Prefecture



Fig. 3. Dried or salted Wakame is softened by flushing the salt with water and is eaten in seafood salad with vegetables or as an ingredient of miso soup in Japan

This picture illustrates typical traditional Japanese eating habit, namely the consumption of slices of raw fish (Sashimi), seaweed salad and noodles.

Table 1. Supply and demand of fisheries products in Japan

 $(1 \times 10^{3} t)$

		Year					
		1993	1994	1995	1996	1997	
Fishes	Domestic production	8,477	8,013	7,325	6,768	6,743	
	Import	4,718	4,788	5,635	6,755	5,921	
	Total	13,195	12,801	12,960	13,523	12,664	
Marine algae	Domestic production	158	139	155	144	135	
	Import	60	62	70	70	68	
	Total	218	201	225	214	203	
	Export	5	3	2	2	2	
	Domestic use	212	198	223	212	201	
	Food	178	168	190	181	172	
	materials	34	30	33	31	29	

taxes. The fact that many shrines used algae as offerings indicates that algae were very important products from olden times in Japan²).

Main edible algae in Japan are Nori (*Porphyra* sp.), Wakame (*Undaria* sp.) Konbu (*Laminaria* sp.) and Hiziki (*Hizikia* sp.)(Fig. 1). Research about edible algae in the Japan sea coastal region in 1999 revealed that other algae were used as domestic foods.

Production

Though many kinds of marine algae were produced in Japan, based on available statistical data, 6 species (*Laminaria*, *Undaria*, *Hizikia*, *Gelidium*, *Porphyra*, *Nemacystus*) and others were identified. In 1998, production from natural resources was 116,614 t, amounting to \(\frac{\pmathcal{2}}{2}5,208\) million, and that from culture was 523,426 t, amounting to \(\frac{\pmathcal{2}}{1}39,434\) million\(^{1}\). Main algae include cultured *Porphyra*, cultured *Undaria* (Fig. 2), natural *Laminaria* and cultured *Nemacystus*. *Nemacystus* production has increased very recently with the development of culture in Okinawa island.

Comparison of the production in 1966 and 1998 shows that natural resources decreased to about half for every species and in every region, and the production of cultured algae increased by about 2 times. After 1993, the production of natural and cultured algae remained sta-

ble, except for the increase in the production of cultured *Nemacystus*.

Production of cultured algae could increase along with studies on the beneficial functions of algal foods.

Utilization as foods

Japan has over 1,000 algal species, with more than 50 edible species. Main algae used as foods consist of green algae, brown algae and red algae. In 1997, domestic production amounted to 135,000 t and imports to 68,000 t, with only 2,000 t as exports. For domestic use, 172,000 t were used as foods and 29,000 t as materials for the alginic, agar and other industries (Table 1). Annual intake of marine products in Japan has been slightly increasing from 1993 to 1997. Fish intake was 69.6 kg and algal intake was 1.4 kg in 1997 (Table 2).

Marine algae are used traditionally as foods that are boiled and seasoned, as soup, as "Yaki-Nori" itself, as ingredient of minced fish paste products, and recently as salad (Fig. 3).

Market size of main algae (*Porphyra*, *Undaria*, *Laminaria and Hizikia*) reaches ¥5,540 billion (Table 3). Main products are simply dried for convenient preservation. Though Japanese like to eat raw fish, they use dried or seasoned marine algae. Marine algae can be rapidly dried and the nutrient content remains stable at room tem-

Table 2. Intake of marine products in Japan

	Year (kg/person)							
	1993	1994	1995	1996	1997			
Fishes	66.4	67.8	71.0	71.0	69.6			
Marine algae	1.4	1.3	1.5	1.4	1.4			

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Table 3. Market size of main marine algae in Japan (¥ Billion)

		`	
Poryp	hyra		
	Yaki-Nori ^{a)}	1,429	
	Seasoning	523	
	Tsukudani ^{b)}	310	
	Others	228	
Undar	ria		
	Boiled salted Wakame	195	
	Cut-Wakame ^{c)}	140	
	Dried	28	
	Raw materials	127	
Lamin	aria		
	Dried	1,500	
	Tsukudani	630	
	Others	300	
Hizikid	a		
	Me-Hiziki ^{d)}	80	
	Naga-Hiziki ^{e)}	40	
	Seasoning	10	

- a): Slightly roasted Nori sheet,
- b): Food cooked in a mixture of soy sauce and sugar,
- c): Small pieced and dried product,
- d): Dried soft young part,
- e): Dried hard old part.

perature, while the volume can be easily reduced for cooking use.

Nutritional elements and physiological function of marine algae

The moisture content of marine algae is very high, accounting for more than 70% of raw material. However, marine algae, which are used as foods, are sold in a salted or dried state for long preservation, with a moisture con-

tent ranging from 7.0 to 20% (Table 4). Though marine algae contain nutritional elements such as proteins, lipids, carbohydrates, vitamins and minerals as in the case of other plants, the content of these elements varies depending on the season and the area of production. Table 4 shows the nutrient composition of 40 kinds of marine algae and vegetables based on the data of Standard Tables of Food Composition in Japan⁸. The protein content of marine algae varies with the species, being about 40% in *Porphyra atropurpurea* (Amanori), and about 15% in Wakame.

The first limiting amino acid in most of the proteins of marine algae is lysine, and the content of histidine is lower than that of sardine or beef (Table 5). The nutritional value of proteins referred to as "amino acid score" is evaluated based on the composition of essential amino acids. In general, the amino acid score of the proteins included in cereals and vegetables is lower than that of the proteins of animal origin. The amino acid score of the proteins included in marine algae ranged from 60 to 100, a value higher than that of the proteins in cereals and vegetables. The amino acid score of proteins which was 91 in Amanori and 100 in *Undaria pinnatifida* (Wakame) was almost the same as that of animal foods.

Taurine which is an amino acid leading to the decrease of the concentration of cholesterol in the serum and liver also exerts an anti-hypertension effect, and prevention effect of vascular diseases, chronic hepatitis and diabetes⁵. Taurine was included in *Laminaria saccharina* (Konbu) and *Porphyra tenera* (Asakusanori) at a level of about 400 mg/100 g (dried weight) as in the case of lobster, crab, shellfish and squid.

The content of lipids of marine algae is lower than that of other marine organisms. For example, the content

Table 4. Chemical composition of marine algae and vegetables⁸⁾

(/100g)Moisture Protein Lipid Carbohydrate Fiber Ash (g) (g) (g) (g) (g) (g) Porphyra complex 11.1 38.8 1.9 39.5 1.8 6.9 (Amanori) Enteromorpha intestinalis 7.3 18.1 0.3 53.9 6.3 14.1 (Aonori) Undaria pinnatifida 13.0 15.0 3.2 35.3 2.7 30.8 (Wakame) Hizikia fusiformis 13.6 10.6 1.3 47.0 9.2 18.3 (Hiziki) Cabbage 92.4 1.4 0.1 4.9 0.6 0.6 Soybean 12.5 40.3 21.7 27.1 5.7 5.0 Wheat 13.5 10.5 3.0 69.3 2.1 1.6

The values refer to the analyzed data of the products which are available in the market.

Amino acid scorea) First limiting amino acid 1973 FAO/WHO Porphyra complex (Amanori) Lysine 91 Laminaria saccharina (Konbu) Lysine 82 Hizikia fusiformis (Hiziki) Lysine 62 Undaria pinnatifida (Wakame) 100 Short neck shellfish Valine 81 Sardine 100 Squid Valine 71 Beef 100 Milk 100 Rice 65 Lysine

Table 5. First limiting amino acids and amino acid score in the proteins from various food materials⁸⁾

of lipids of Analipus japonicus (Matsumo) and Amanori was 4.9 g/100 g and 2-3 g/100 g, respectively, and that of other marine algae was less than 1 g/100 g. The lipids of marine algae consist of sterols, triacylglycerols, diacylglycerols, monoacylglycerols and phospholipids, while phospholipids are the main lipids of marine algae. The fatty acids, composing the lipids of marine algae contain n-3 polyunsaturated fatty acids such as α-linolenic acid and eicosapentaenoic acid (EPA) and other n-3 polyunsaturated fatty acids such as 18:4n-3 which are not included in other organisms (Table 6). It was reported that these n-3 polyunsaturated fatty acids exert an antiarteriosclerosis effect, anti hypertension effect, antiinflammation effect and immunoregulation effect³⁾. Moreover, it was suggested that 18:4n-3 affects the immune system of humans⁴⁾. Marine algae are an important source of supply of n-3 polyunsaturated fatty acids

for the maintenance of health.

The kinds and the contents of sterols in marine algae vary with the species of marine algae. Green algae contain 28-isofucocholesterol, cholesterol, 24-methylene-cholesterol and β -sitosterol, while brown algae contain fucosterol, cholesterol, and brassicasterol. It was reported that plant sterols such as β -sitosterol and fucosterol lead to the decrease of the concentration of cholesterol in the serum in experimental animals and humans 9 .

Marine algae are characterized by the presence of polysaccharides, such as frame polysaccharides, mucopolysaccharides and storage polysaccharides. The frame polysaccharides of marine algae mainly consist of cellulose. Green algae contain sulfuric acid polysaccharides, brown algae alginic acid, fucoidan and sargassan and red algae agar-agar and porphyran as mucopolysaccharides located in the intercellular spaces. Starch and laminaran

Table 6. Fatty acid composition of lipids in marine algae (%)8)

	16:0	18:0	18:1	18:2 n-6	18:3 n-3	18:4	20:4 n-3	20:4 n-6	20:5 n-3	22:6 n-3
Porphyra complex (Amanori)	23.3	0.7	3.1	1.8	0.2	0.2	0.8	4.6	54.2	_
Laminaria saccharina (Konbu)	22.5	2.3	27.9	7.9	3.9	3.9	0.5	10.9	5.4	_
Hizikia fusfiormis (Hiziki)	30.1	1.1	10.3	5.1	8.0	4.5	1.0	13.5	7.2	_
Undaria pinnatifida (Wakame)	11.2	1.1	5.0	6.1	11.8	29.5	0.8	12.2	15.1	_
Sardine	19.0	3.3	13.0	2.6	2.6	3.5	1.0	0.9	13.0	10.7
Beef (Japanese cattle rib loin, total edible)	28.6	10.9	45.0	1.9	_	_	_	_	_	_
Soybean	11.6	3.2	21.3	52.0	10.9	_	_	_	_	_
Rapeseed	4.0	1.7	58.6	21.8	10.8	_	_	_	_	-

a): Amino acid score was calculated from amino acid requirements by FAO in 1973.

	Frame polysaccharides	Mucopolysaccharides	Storage polysaccharides
Green algae	Cellulose	Sulfated	Amylose
	β-1,3-Xylan	glucuronoxylorhamnan	Amylopectin
	β-1,4-Mannan	Sulfated xyloarabinogalactan	
		Sulfated glucuronoxylorhamn ogalactan	
Brown algae	Cellulose Hemicellulose	Alginic acid Fucoidan	Laminaran
Red algae	Cellulose Hemicellulose β-1,3-Xylan β-1,4-Mannan	Agar-agar Carrageenan Porphyran	Starch

Table 7. Polysaccharides in marine algae

are present in marine algae as storage polysaccharides (Table 7).

Various industries related to food processing, pharmaceuticals, fodder and cosmetics use alginic acid extracted from Konbu and Wakame. Moreover, it was reported that alginic acid leads to a decrease in the concentration of cholesterol and exerts an anti hypertension effect, prevention effect of absorption of toxic chemical substances, and plays a major role as dietary fiber for the maintenance of human health. Agar-agar is a typical and traditional food material in Japan and it is used as a material for cooking and Japanese-style confectionery. In addition, agar-agar is used for the manufacture of capsules for medical applications and as a medium for cell culture, etc. It was reported that agar-agar leads to a decrease in the concentration of blood glucose and exerts an anti-aggregation effect of red blood cells and ultraviolet rays absorption effect. Recently, studies carried out on the physiological effects of fucoidan, the mucopolysaccharide of Wakame and Konbu, have indicated that fucoidan exerted an anti-blood coagulation effect and anticancerous effect¹¹⁾.

The amount of dietary fibers not digested by the human digestive tract in marine algae which was determined by the Prosky-AOAC method, was higher than that of other food materials, and the content of dietary fibers in marine algae was 81.29% (g/100 g dry weight) for Tengusa, 54.94% for *Hizikia fusiformis* (Hiziki), 38.62% for *Enteromorpha intestinalis* (Aonori), 57.90% for Wakame, 29.70% for Amanori and 28.58% for Konbu⁶). The dietary fibers included in marine algae are classified into 2 types, namely into insoluble dietary fibers such as cellulose, mannans and xylan, and watersoluble dietary fibers such as agar-agar, alginic acid, funoran, laminaran and porphyran, depending on the solubility in water. These dietary fibers were found to be effective in the prevention of obesity, hypercholester-

olemia, large intestine cancer and diabetes.

Marine algae are used as foods for the supply of minerals. Especially, Hiziki contains 1,400 mg minerals / 100 g dried matter, the highest recorded value in marine algae. In addition, the kinds of minerals included in marine algae vary with the species. Phosphorus, iron, sodium and potassium were present in Aonori, Hiziki, Wakame and Konbu, respectively (Table 8).

The development of analytical methods for vitamins indicated that marine algae contained a large amount of vitamins. The amount of vitamins in Asakusanori is higher than that in other marine algae. Amanori and Aonori contain almost all kinds of vitamins such as vitamin B family and vitamin C (Table 8). Moreover, Asakusanori, Amanori, and Wakame contain a large amount of β-carotene, which is converted into vitamin A in the algal body. The content of vitamin C in algae varies depending on the state of marine algal products. Since vitamin C is decomposed under dry conditions and/ or during the processing of marine algae, vitamin C content in dried or processed marine algal products is lower than that in raw marine algae. Though it was reported that the content of vitamin D was very low in marine algae, it was confirmed that Aonori and Fucus vesiculosus (Hibamata) contain ergosterol, i.e. provitamin D, and vitamin D₃, respectively. Moreover, brown algae contain a large amount of vitamin E, which shows an anti-oxidative property, compared with green algae and red algae.

In general, the contents of vitamins of the foods are compared with those of spinach, which contains a large amount of vitamins. Table 8 shows a comparison of the contents of vitamins in marine algae and vegetables based on the data of Standard Tables of Food Composition in Japan⁸⁾. The content of all types of vitamins in the Amanori was higher than that of spinach, especially, that of vitamin A was about 3 times the content in carrot.

Marine algae, which contain a large amount of bene-

Table 8. Mineral contents in marine algae⁸⁾

	Minerals (mg/g)				Vitamins (/100 g)				
-	Na	K	Ca	P	Fe	A(IU)	B ₁ (mg)	B ₂ (mg)	C(mg)
Enteromorpha compressa (Aonori)	530	3,200	840	740	32.0	12,000	0.56	1.90	40.0
Undaria Pinnatifida (Wakame)	6,100	5,500	960	400	7.0	1,800	0.30	1.15	15.0
Hizikia fusiformis (Hiziki)	1,400	4,400	1,400	100	55.0	310	0.01	0.14	0
Laminaria saccharina (Konbu)	2,800	6,100	710	200	3.9	560	0.48	0.37	25.0
Porphyra complex (Amanori)	120	2,100	390	580	12.0	14,000	1.15	3.40	100.0
Tomato	2	230	9	18	0.3	220	0.05	0.03	20.0
Spinach	21	740	55	60	3.7	1,700	0.13	0.23	65.0
Carrot	26	400	39	36	0.8	4,100	0.07	0.05	6.0
Orange (Valencia)	1	190	20	20	0.1	42	0.01	0.03	40.0

The values refer to the analyzed data of the products which are available in the market.

ficial elements for the maintenance of health have been used as foods by human beings. Konbu from Hokkaido was used to treat diseases of the thyroid gland about 800 BC in China. Moreover, it was reported that the consumption of marine algae was large in Okinawa Prefecture where people are known to live longer. Recently, beneficial elements for the maintenance of human homeostasis have been extracted from marine algae and many researchers have examined their physiological

effects.

Nishizawa classified the physiologically active substances of marine algae into 2 types based on the difference in the mechanisms as follows: 1) the non-absorbed high-molecular materials like dietary fibers; 2) low-molecular materials, which are absorbed and affect the maintenance of human homeostasis directly. Among the physiologically active substances, antibiotics, anti-tumor elements and anti-ulcer elements were detected in marine

Table 9. Functional substances in marine algae

Substance	Function	Origin
Proteins		
Lectin	Blood cell aggregation	Gracilaria verrucosa (Ogonori)
Lymphocyte differentiation		Codium fragile (Miru)
Aromatic amino acids	Ultraviolet rays absorption	Tichocarpus crinitus (Karekigusa)
Lipids		
Fatty acids and its derivatives		
Acrylic acid	Antibiotics	Enteromorpha intestinalis (Bouaonori)
Polysaccharides	Anti-tumor action	Laminaria saccharina (Konbu)
Acidic polysaccharides	Anti-tumor action	Undaria pinnatifida (Wakame)
Fucoidan	Anti- arteriosclerosis	Eisenia biyclis (Arame)
	Anti- cancer	
Alginic acid	Anti- cancer	Undaria pinnatifida (Wakame)
Others		
Nucleic acid	Ultraviolet rays absorption	Chondrus ocellatus (Tsunomata)
Porphyosin	Anti- ulcer	Porphyra tenera (Asakusanori)
Vercoyosin	Anti- ulcer	Gracilaria verrucosa (Ogonori)
Tannin	Antibiotics	Ulva lactaca (Aosa)
		Entermorpha compressa (Aonori)
Terpenoid	Antibiotics	Dictyota dichotoma (Amizikusa)
Halogen	Antibiotics	Rhodomela subfusa (Itofuzimatsu)
Extracts	Anti- oxidation	Laurencia okamurai (Mitsudesoso)

algae. Preservation of marine algae over a long period of time without oxidation of the lipids has been reported, though marine algae contain a large amount of polyunsaturated fatty acids. This characteristic indicated that marine algae contain anti-oxidation substances. Nakamura isolated 5 anti-oxidation substances from *Phaeophyceae eisenia* (Arame), which delays the autooxidation of methyl α -linolenic acid, by using silicic acid chromatography. In addition, a polyphenol, which exerts an anti-oxidation effect, had been extracted from *Phaeophyceae ecklonia* (Kajime). The physiologically active substances included in marine algae are listed in Table 9.

Mechanism whereby Japanese eating habits including the consumption of marine products contribute to human health

Marine algae have been used as foods which contain valuable nutrients in Japan. When the Japanese eating habits prevailed in the 1960s, the taste and texture of Nori, Konbu and Wakame foods were improved to meet the requirements of Japanese people for tasty food. In the 1970s, since the incidence rate of lethal diseases such as arteriosclerosis, coronary heart disease, thrombosis and apoplexy increased, it was expected that some foods could exert effects on these diseases. Recently, in the case of marine algae, it has been suggested that dietary fibers, minerals, and low-molecular substances may play important roles in the maintenance of health.

It was reported that the change in food habits from the Japanese type which is characterized by the consumption of rice, vegetables and marine products to the European and American types characterized by the consumption of meat and dairy products has led to an increase in the incidence of various adult diseases. The Japanese eating habits are characterized by the consumption of various food materials such as cereals, vegetables, fish, shellfish, marine algae and meats which are cooked by various techniques such as steaming, roasting, broiling and frying. Therefore, properties of functional substances in food materials may be enhanced or weakened by the combination of various food materials, and the chemical changes which functional substances undergo during the processing and the cooking steps. Therefore, it is necessary to analyze the influence of Japanese type eating habits on human health scientifically.

Recently, the presence of elements which enable to maintain homeostasis in organisms has been reported in Wakame. However, since there were few reports on the metabolism of lipids other than cholesterol, although Wakame is considered to contain elements which regulate the lipid metabolism, we examined the effect of Wakame on the lipid metabolism to determine whether Wakame could be used as food to prevent hyperlipidemia⁷⁾.

Rats were fed diets to which dry Wakame powder had been added to the basic AIN-76 diet (control diet) at levels of 0.5, 1, 2, 5, and 10%, and the concentration of the lipids in the serum and liver was compared with that of rats fed control diets. In this experiment, there were no differences in the food intake and the body weight gain of rats between each group. The concentration of serum triacylglycerol in rats fed Wakame diets was significantly lower than that of rats fed the control diet, depending on the amount of dry Wakame powder added, and a significant decrease appeared when more than 2% of Wakame powder was added to the control diet. Furthermore, dietary Wakame also significantly decreased the concentration of rat liver triacylglycerol depending on the amount of dry Wakame powder added to the control diet.

Table 10.	Concentration of lipid components in the serum and liver of rats fed a diet containing different
	proportions of Wakame

		Dietary Wakame powder (g/100 g diet)							
	0	0.5	1.0	2.0	5.0	10.0			
Serum			(mm	ol/L)					
Triacylglycerol	3.3 ± 0.5^a	2.8 ± 0.5^a	2.7 ± 0.2^a	$2.4\pm0.3^{\rm b}$	$2.3\pm0.2^{\text{b}}$	$1.7 \pm 0.1^{\circ}$			
Cholesterol	3.9 ± 0.3	3.9 ± 0.3	3.6 ± 0.5	3.9 ± 0.3	3.6 ± 0.3	3.9 ± 0.3			
Phospholipids	6.7 ± 0.6	6.4 ± 0.6	6.7 ± 0.3	7.0 ± 0.9	6.7 ± 0.3	6.7 ± 0.3			
Liver			(µm	ol/g)					
Triacylglycerol	58.8 ± 7.9^a	57.0 ± 4.7^a	45.6 ± 4.3^{b}	43.3 ± 2.5^{b}	32.3 ± 1.2^{c}	$26.9 \pm 2.8^{\circ}$			
Cholesterol	5.2 ± 0.3^a	5.4 ± 0.3^a	5.4 ± 0.3^a	5.4 ± 0.3^a	4.7 ± 0.3^a	2.1 ± 0.3^{1}			
Phospholipids	68.9 ± 4.3	61.0 ± 3.3	67.4 ± 4.0	70.2 ± 6.4	67.1 ± 3.0	65.9 ± 3.3			

a,b,c,d): Different superscripts in each row indicate significant differences (P<0.05) between dietary treatment groups. Values are mean \pm SEM for 7 rats.

Table 11. Concentration of β-hydroxybutyrate in serum and activities of enzymes involved in fatty acid oxidation in the liver of rats fed diets containing different proportions of Wakame

	Dietary Wakame powder (g/100 g diet)					
	0	5.0	10.0			
β-Hydroxybutyrate (μmol/L serum)	116.9 ± 6.3^{b}	152.3 ± 9.3^a	162.9 ± 7.2^{a}			
Enzyme activities (nmol/(min.mg protein)))					
Carnitine palmitoyltransferase	$9.8\pm0.5^{\rm b}$	$10.4\pm0.5^{\text{b}}$	12.5 ± 0.6^a			
Acyl-CoA dehydrogenase	31.6 ± 3.7^{c}	46.2 ± 6.5^{b}	71.9 ± 4.5^{a}			
Acyl-CoA oxidase	$3.3\pm0.3^{\text{b}}$	4.2 ± 0.8^a	4.4 ± 0.4^a			
3-Hydroxyacyl-CoA dehydrogenase	346.9 ± 35.8	416.8 ± 43.9	459.0 ± 56.0			
Enoy-CoA hydratase	$1,868.8 \pm 207.0^{b}$	$3,006.6 \pm 330.5^a$	$3,476.0 \pm 346.7^{a}$			
2,4-Dienoyl-CoA reductase	$7.5\pm0.2^{\mathrm{b}}$	9.6 ± 0.7^{a}	11.2 ± 0.7^a			

a,b,c): Different superscripts in each row indicate significant differences (P<0.05) between dietary treatment groups.

Values are mean \pm SEM for 7 rats.

The addition of more than 1% of dry Wakame powder significantly decreased the concentration of rat liver triacylglycerol (Table 10).

Thus, dietary Wakame led to a remarkable decrease in the concentration of triacylglycerol in the rat serum and liver depending on the amount added. As for the mechanism(s) involved, since it is known that the synthesis and oxidation of fatty acids by the liver control the concentration of triacylglycerol in the rat serum and liver, the decrease in the concentration of triacylglycerol in the serum and liver by dietary Wakame was ascribed to the change in the ability of synthesis and oxidation of fatty acids in the liver. Therefore, the enzyme activities involved in the synthesis or the oxidation of fatty acids in the liver of rats fed the Wakame diet were determined to elucidate the mechanism of the decrease in the concentration of serum and liver triacylglycerol by dietary

Wakame.

We measured the activities of the enzymes involved in fatty acid synthesis in the liver from rats fed dietary Wakame. The activity of glucose-6-phosphate dehydrogenase increased by dietary Wakame, while, the activities of the other enzymes, fatty acid synthetase and malic enzyme, were not different between the dietary groups. On the other hand, the activities of acyl-CoA dehydrogenase, acyl-CoA oxidase, and carnitine palmitoyltransferase, involved in fatty acid oxidation, significantly increased by dietary Wakame (Table 11).

These results indicated that the decrease in the concentration of triacylglycerol in the serum and liver by dietary Wakame was due to the promotion of fatty acid oxidation in rat liver. Since the increase in the concentration of triacylglycerol in the serum and liver leads to the increase of the incidence of arteriosclerosis, Wakame

Table 12. Composition of the experimental diets (g/100 g diet)

Ingredients	Control ^{a)}	10% Wakame	5% Fish oil	5% Fish oil 10% Wakame
Casein	20.0	19.65	20.0	19.65
Rapeseed oil	5.0	4.92	_	_
Fish oil ^{b)}			5.0	4.92
Cellulose	5.0	4.92	5.0	4.92
Potato starch	15.0	15.0	15.0	15.0
AIN-vitamin mixture	1.0	1.0	1.0	1.0
AIN-mineral mixture	3.5	3.5	3.5	3.5
DL-Methionine	0.3	0.3	0.3	0.3
Choline bitartrate	0.2	0.2	0.2	0.2
Wakame powder	0	10.0	_	10.0
Sucrose	50.0	40.51	50.0	40.51

a): AIN-76 basic diet.

b): Fish oil used in this experiment contains 25% of eicosapentaenoic acid (EPA).

	Dietary groups						
	Control	10% Wakame	5% Fish oil	5% Fish oil 10% Wakame			
Serum		(mm	ol/L)				
Triacylglycerol	2.8 ± 0.2^a	1.0 ± 0.1^{b}	$1.2\pm0.1^{\scriptscriptstyle b}$	$0.6\pm0.1^{\circ}$			
Cholesterol	2.4 ± 0.3^a	2.8 ± 0.2^a	1.6 ± 0.1^{b}	1.7 ± 0.2^{b}			
Phospholipids	2.7 ± 0.4^a	$1.8\pm0.2^{\text{b}}$	$1.1\pm0.1^{\rm c}$	$1.8\pm0.2^{\rm b}$			
Liver		(µm	ol/g)				
Triacylglycerol	55.1 ± 7.9^a	$32.7\pm4.7^{\text{b}}$	$32.8\pm4.3^{\text{b}}$	12.3 ± 2.5^{c}			
Cholesterol	4.1 ± 0.3	4.1 ± 0.4	4.1 ± 0.3	4.1 ± 0.4			
Phospholipids	40.4 ± 3.3	38.1 ± 4.3	41.3 ± 3.8	42.2 ± 5.1			

Table 13. Concentration of lipid components in the serum and liver of rats fed diets containing fish oil and/or Wakame

a,b,c,d): Different superscripts in each row indicate significant differences (P<0.05) between dietary treatment groups.

Values are mean±SEM for 7 rats.

may play a role in the prevention and treatment of arteriosclerosis. In addition, since the decrease in the concentration of triacylglycerol in the serum and liver by dietary Wakame is associated with the promotion of oxidation of fatty acids in the liver, Wakame may become a food material for the prevention of obesity.

It was reported that fish oil rich in n-3 polyunsaturated fatty acids such as EPA and docosahexaenoic acid (DHA) decreases the concentration of lipids in the serum and liver, and that these fatty acids are effective in the prevention and treatment of arteriosclerosis. Along with the decrease in the concentration of triacylglycerol by dietary Wakame, fish oil also affected the synthesis and oxidation of fatty acids in the liver.

In rats fed the diets shown in Table 12 for 3 weeks, the concentration of serum and liver lipids was determined. It was found that fish oil diet and Wakame diet decreased the concentration of triacylglycerol in the serum and liver significantly compared with the control diet. In addition, when rats were fed Wakame and fish oil at the same time, the decrease in the concentration of triacylglycerol in the serum and liver became even more conspicuous (Table 13). The mechanism(s) involved is being currently examined. These results suggest that the simultaneous consumption of fish (fish oil) and Wakame may lead to a decrease of the concentration of triacylglycerol in the serum and liver, for example, the combination of baked or fried fish and miso soup with Wakame is beneficial.

Thus, it appears that the combination of food materials enhances the functions of the foods.

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