

## Physical Characteristics and Antioxidative Capacity of Major Seaweeds

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### Abstract

Seaweeds is a rich sources of dietary fibers exerting a number of physiological properties. However, the reported dietary fiber contents of seaweeds are not consistent and vary widely. Also, a limited number of studies on the biological effects of specific seaweeds have been reported. In this study, water-holding capacity, viscosity and antioxidative activity of major dietary seaweeds were measured to assess their physiological effects. Results showed that total dietary fiber contents ranged from 28 to 51% of dried weight, and large proportions of dietary fiber were insoluble fibers. Water-holding capacity was highest in sea mustard being 1310%, while laver, sea tangle, and green laver exhibited 943, 854, and 816%, respectively. The viscosity of seaweed samples was 20 to 40 cP in sea mustard and sea tangle, while laver and green laver possessed much lower values. All seaweed samples revealed a weak, albeit significant electron donating ability. Also, lipid peroxidation was reduced by 7 to 18%. However, there was no difference in antioxidative activity among seaweeds and sample concentrations used. These results imply that most commonly used seaweeds possibly exert parts of their physiological effects through their water-holding, gel-forming, and/or antioxidative activities.

**Key words:** seaweeds, viscosity, antioxidative capacity, dietary fibers

### INTRODUCTION

One of the most recent research interests in the area of nutrition is to elucidate physiological functions of non-nutritive compounds present in plant foods. Since possible roles of dietary fibers in the prevention of digestive, metabolic and cardiovascular diseases were suggested in the late 1970s, many other physiologically active compounds have been identified.

Physiological activities of functional food components are related to their chemical structures which determine the physical properties including solubility and viscosity (1). Recent research results indicated that soluble dietary fibers form gel matrix in the intestine delaying nutrient absorption, including glucose absorption (2). Also, they produce short chain fatty acids by microbial fermentation to reduce colon pH and inhibit the formation of several toxic compounds (3).

Seaweeds are widely used food items in Korea and Japan. Most frequently used seaweeds include sea mustard, sea tangle, laver, green laver, gulf weed and seaweed fusiforme. They are rich sources of dietary fibers and minerals. Major polysaccharides present in seaweeds include alginic acid, fucoidan, laminaran, carrageenan, and porphyran (4). Due to high contents of these undigestible or partly digestible carbohydrates, seaweeds are known to possess considerable water-holding capacity, viscosity, metal-binding capacity, and bile acid-binding capacity (5,6). Also, antioxidative activity of several compounds present in seaweeds has been suggested (7). These fundamental physiological functions are directly or indirectly related to the occurrence of major chronic diseases. However, few studies have been performed to examine their physical

and biological properties and study results are not consistent. Therefore, the purpose of this study was to evaluate physiological importance of most frequently used seaweeds by examining viscosity, water-holding capacity, and antioxidative activity.

### MATERIALS AND METHODS

#### Sample preparation and general analysis

Dried sea mustard (*Undaria pinnatifida*), sea tangle (*Laminaria japonica*), laver (*Porphyra tenera*), and green laver (*Enteromorpha compressa*) were purchased from a local agricultural cooperative union market. All seaweeds were grown in Wan-do, Korea. Samples were ground with a grinder (FM680, Hanil) at the speed of 13,000 rpm and passed through a 20-mesh screen. Protein, lipid and ash contents were determined by macro-kjeldahl method, soxhlet extraction method, and wet-dry ashing, respectively. Moisture content was measured by oven-drying method. Carbohydrate content was determined by extracting above values from the total sample weight. Total, insoluble and soluble dietary fiber contents were analyzed based on the method by Lee et al. (8).

#### Measurement of water-holding capacity and viscosity

Water-holding capacities of samples were measured based on the method of Deshpande et al. (9). One g of prepared seaweed sample and 30 ml of distilled water were mixed and left for 1 hr followed by centrifugation at 8000 × g for 20 min. After removing the supernatant, the tube was left upside down for 15 minutes and weighed.

For the measurement of viscosity, 100 ml of distilled water

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were added to 20 g of dried and powdered samples and extracted for 8 hr at 70°C. Sample solutions were centrifuged at 2,100×g and the supernatants were freeze-dried. One g of freeze-dried sample was mixed with 30 ml of distilled water, vortexed for 1 hr and left at room temperature for one hour. Viscosity was measured using a Brookfield DV-III rheometer.

#### Measurement of antioxidative capacity

Electron-donating ability (EDA) was measured based on the method of Blies (10) which measures EDA of charcoal-decolorized seaweed samples and ascorbic acid to DPPH (1,1-diphenyl-2-picryl-hydrazyl). Briefly, 20 mg of DPPH was dissolved in 150 ml of ethanol, and sample solutions were prepared to give concentrations of 0.05 to 1.0 mg/ml. A half ml of each sample solution was added to vials containing 0.5 ml of DPPH solution, and mixed thoroughly for 5 sec. After 30 min of incubation at room temperature, absorbance was measured at 570 nm. Absorbance of the vial containing each compound was compared to that of the vial containing DPPH only. Percent EDA was calculated using the following formula.

$$\text{EDA (\%)} = [1 - (\text{sample absorbance} / \text{control absorbance})] \times 100$$

Inhibitory action of lipid peroxidation of samples was measured by the decreased formation of thiobarbituric acid reactive substances using linoleic acid as the substrate. Linoleic acid (0.1% w/v) was dispersed in sodium lauryl sulfate solution (0.8% w/v). To an aliquot (0.8 ml) of above linoleic acid solution, 0.1 ml EDTA (0.1 mM) and 0.1 ml of sample solution were added, Ultraviolet lamp (40 W) was illuminated at a distance of 30 cm for 90 min. A half ml of trichloroacetic acid (0.44 M) and 0.5 ml of 0.8% 2-thiobarbituric acid were mixed and incubated for 15 min at 100°C. The absorbance was measured at 532 nm.

## RESULTS AND DISCUSSION

### General composition of seaweeds

Carbohydrate, protein, lipid, and ash contents of seaweed samples are shown in Table 1. Carbohydrate of seaweeds ranged from 46 to 57% of dried samples. Green laver and laver contained 32 and 37% protein, respectively. However, sea mustard and sea tangle contained only 18% and 8% protein. Lipid contents of seaweed samples were less than 5% in all samples used. Ash contents ranged from 8% to 32%. These values correlate well with the values in the food composition table (11). Sea tangle was especially a rich source of minerals

**Table 1.** Composition of seaweeds (% , dry basis)

	Carbohydrate	Protein	Lipid	Ash
Sea mustard	52.3 <sup>1)</sup>	18.5	2.1	26.9
Green laver	46.1	32.4	4.2	17.1
Laver	52.3	37.8	1.3	8.4
Sea tangle	57.3	8.1	1.9	32.5

<sup>1)</sup>Mean of two separate experiments

while protein content is low, however laver contained a large amount of protein with less minerals.

The total, insoluble, and soluble dietary fiber contents of seaweed samples are shown in Table 2. Total dietary fiber contents of seaweed samples ranged from 28% to 51% on a dry basis, and sea mustard was the richest source of total dietary fiber. All seaweed samples except sea mustard, contained limited amounts of soluble dietary fiber. Depending on the methods used and experimental conditions in each laboratory, huge variations in dietary fiber contents have existed. Recently, a modified enzymatic-gravimetric method, which were used in this study, was chosen as the most reliable method to determine total, insoluble and soluble dietary fiber contents (8). The neutral detergent fiber method was not adequate for dietary fiber determination of seaweeds, because seaweeds contained indigestible polysaccharides soluble in neutral detergent (12). There is very little information available on the distribution of soluble and insoluble dietary fibers in seaweeds. Total dietary fiber contents of seaweeds used in the present study are very similar to those reported by Hwang (13) and Kim (14). However, soluble fiber contents of sea tangle and laver were less than the value reported by Hwang (13) in which soluble dietary fiber content was calculated by subtracting insoluble dietary fiber determined from total dietary fiber. In the present study, however, soluble and insoluble dietary fibers were separately determined, and resulting soluble dietary fiber contents were lower than the previous report. Lahaye (15) determined the total, soluble and insoluble fibers in major seaweeds using the method of Prosky et al. (16). The soluble fiber contents were ranged from 17 to 59%, while the contents of insoluble fibers were only from 5 to 16%. On the other hand, when the analysis was performed by Suzuki et al. (6) using the same method with similar varieties of seaweeds, the soluble dietary fiber contents were between 7 to 25%. Therefore, more detailed information on analytical methods for different food samples, especially seaweeds are required.

### Water-holding capacity and viscosity

All seaweed samples used in this study had high water-holding capacity and it was in the order of sea mustard > laver > sea tangle = green laver being 1310%, 943%, 854%, and 816% (Table 3). Sea mustard bound significantly higher amount of water compared to other seaweed samples. When the viscosity of water-soluble fraction of seaweed samples was measured, sea tangle showed a maximum value (39.7 cP) followed by sea mustard (23.4 cP). However, the viscosities of green

**Table 2.** Contents of dietary fiber (%)

	Dietary fiber		
	Total	Insoluble	Soluble
Sea mustard	47.0(51.0) <sup>1)</sup> *	36.3(39.4)	10.6(11.5)
Green laver	25.9(28.5)	25.7(28.2)	0.2(0.2)
Laver	32.3(34.9)	29.7(32.0)	2.7(2.9)
Sea tangle	28.5(30.8)	25.4(27.5)	3.0(3.3)

<sup>1)</sup>Mean of two separate experiments

\*Values in parentheses are based on dry weight

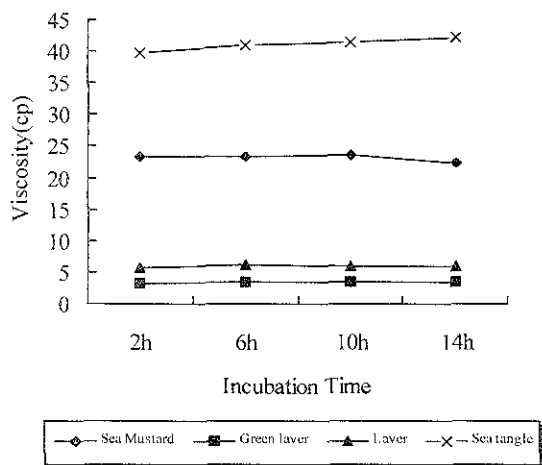
**Table 3.** Water-holding capacity (WHC) of seaweeds

	WBC (%)
Sea Mustard	1310 ± 13 <sup>ad</sup>
Green laver	816 ± 41 <sup>c</sup>
Laver	943 ± 8 <sup>b</sup>
Sea tangle	854 ± 11 <sup>c</sup>

<sup>d</sup>Means with different letters (a,b,c) within a column are significantly different from each other at  $p < 0.05$  as determined by Duncan's multiple range test.

laver and laver were considerably lower than those of the other two seaweeds (Fig. 1). There was no change in the viscosity of samples as the incubation time increases.

Water can be bound by the forces of adsorption to food solids. Also, some enters into colloidal gels when soluble fractions such as pectin and gums are present. Water-holding capacity depends mostly on the chemical nature of the material. Suzuki et al. (5) investigated the water-holding capacity of the major seaweeds, and the setting volumes were around 50% which is much lower than the values from the present study. This may be due to the differences in the measurement method, especially the speed of centrifugation which was  $14,000 \times g$  in a study by Suzuki et al. Stephen and Cummings (17) stated that no value could be obtained by the centrifugation method for most of the gel-forming polysaccharides because they did not centrifuge down. Therefore, seaweeds containing large

**Fig. 1.** Changes of viscosity in seaweeds at different incubation time.**Table 4.** Electron donating abilities (EDA) of seaweeds to 1-diphenyl-2-picrylhydrazyl (DPPH) radicals

conc. (mg/ml)	EDA (%) <sup>1)</sup>				
	Sea mustard	Green laver	Laver	Sea tangle	L-Ascorbic acid
0			0 <sup>z3)</sup>		
0.1	4.9 ± 5.8 <sup>yzb2)</sup>	4.1 ± 3.7 <sup>yb</sup>	3.4 ± 2.2 <sup>yb</sup>	2.9 ± 2.4 <sup>yb</sup>	95.6 ± 0.3 <sup>ya</sup>
0.5	5.2 ± 5.7 <sup>yb</sup>	4.4 ± 1.9 <sup>xyb</sup>	5.5 ± 3.3 <sup>xyb</sup>	4.8 ± 5.7 <sup>xb</sup>	95.9 ± 0.2 <sup>xa</sup>
1.0	3.9 ± 5.6 <sup>yzc</sup>	6.9 ± 2.8 <sup>xbc</sup>	8.5 ± 4.8 <sup>xb</sup>	3.9 ± 5.6 <sup>wbc</sup>	96.0 ± 0.3 <sup>xa</sup>

<sup>1)</sup>EDA (%) =  $[1 - (\text{sample Abs.} / \text{control Abs.})] \times 100$

<sup>2)</sup>Means with different letters (a,b,c) within a row are significantly different from each other at  $p < 0.01$  as determined by Duncan's multiple range test.

<sup>3)</sup>Means with different letters (w,x,y,z) within a column are significantly different from each other at  $p < 0.01$  as determined by Duncan's multiple range test.

amount of these polysaccharides may require proper centrifugation speed to measure water-holding. These authors suggested that water uptake was related to uronic acid content ( $r=0.87$ ) and increased by a smaller particle size in the food materials. Seaweeds were known to have many different types of polysaccharides containing uronic acid and sulfate. Also, water-holding capacity is known to be influenced by the starch content of plant materials (18).

The viscosity of food materials is an important factor in determining physiological actions in the gastrointestinal tract. In this study, seaweeds were incubated at room temperature for 2 to 14 hr considering the transit time of the foods. Major compounds giving viscosity to seaweeds are known to be alginic acid and fucoidans. Lee et al. (19) reported that the viscosity of 1% water-soluble alginic acid extracted from sea mustard and sea tangle were 147 cP and 74 cP, respectively and alginic acid contents were 7% in sea mustard, and 4% in sea tangle. However, in this study, the viscosity of water-soluble fractions of seaweeds was lower than that of the pure alginic acid solution, indicating that alginic acids incorporated as parts of plant structure do not fully exert its gel forming ability. Also, gel formation seems to occur in a short time period.

#### Antioxidative activities of seaweeds

Seaweeds used in this study showed weak, however, significant antioxidative activities. EDA of seaweeds were between 3.4 to 8.5% when 0.1 to 1.0 mg/ml of samples were tested (Table 4), while ascorbic acid at a same concentration range possessed more than 95% EDA. There was a tendency of dose-dependent increases in EDA except for sea mustard although EDA of seaweed samples at same concentrations were not significantly different from each other. When the capacity of inhibiting lipid peroxidation was measured, seaweed samples showed 6.7 to 17.7% inhibition, however, there was no dose-dependent response (Table 5). At a same concentration range, ascorbic acid revealed 65.1 to 83.2% inhibition.

There is a limited number of studies on the antioxidative activity of seaweeds. Kaneda and Ando (20) separated phospholipids possessing a strong antioxidative property. Fujimoto and Kaneda (21) found 5-bromo-3,4-hydroxybenzaldehyde from red algae. Recently, Park et al. (22) separated two compounds with molecular weights of 181 and 238 from sea

**Table 5.** Inhibition(%) of lipid peroxidation by U.V. radiation of seaweeds

Conc. (mg/ml)	Inhibition(%) <sup>1)</sup>				
	Sea mustard	Green laver	laver	Sea tangle	L-Ascorbic acid
0			0 <sup>2)</sup>		
0.1	10.6 ± 3.3 <sup>yb2)</sup>	11.4 ± 1.9 <sup>yb</sup>	10.9 ± 8.7 <sup>yzb</sup>	11.8 ± 4.9 <sup>yb</sup>	65.1 ± 6.4 <sup>xa</sup>
0.5	6.7 ± 4.2 <sup>xb</sup>	9.9 ± 4.8 <sup>yb</sup>	17.7 ± 10.5 <sup>yb</sup>	8.2 ± 4.9 <sup>xb</sup>	81.1 ± 3.3 <sup>ya</sup>
1.0	6.3 ± 3.7 <sup>xc</sup>	10.6 ± 4.0 <sup>ybc</sup>	10.5 ± 8.0 <sup>yzbc</sup>	12.9 ± 5.1 <sup>yb</sup>	83.2 ± 3.1 <sup>ya</sup>

<sup>1)</sup>Inhibition (%) = [1 - (sample Abs./control Abs.)] × 100

<sup>2)</sup>Means with different letters (a,b,c) within a row are significantly different from each other at p < 0.01 as determined by Duncan's multiple range test.

<sup>3)</sup>Means with different letters (w,x,y,z) within a column are significantly different from each other at p < 0.01 as determined by Duncan's multiple range.

mustard, sea tangle, laver, and green laver. Extracts from these seaweed showed similar antioxidative activity with BHT, a synthetic antioxidant. Since these seaweeds are used as a whole, not as extracted forms in our diet, it may also be important to evaluate their physiological properties. In the present study, electron-donating ability and inhibition of lipid peroxidation of freeze-dried samples of four major seaweeds were measured, and the results showed significant antioxidative activities although they were significantly weaker antioxidants than ascorbic acid. Seaweeds used in this study were slightly more efficient materials to inhibit lipid peroxidation than to donate electrons implying that they act efficiently in lipophilic systems. Considering the proportion of ascorbic acid in foods, the antioxidative activity of whole seaweeds may have important implications.

Results from this study suggest that dietary seaweeds are rich sources of dietary fiber possessing important physical properties and antioxidative abilities. Since each seaweed revealed characteristic viscosity and water-holding capacities which are not directly proportional to their total dietary fiber contents, further studies on individual fiber and/or other components responsible for their functionalities will expand utilization of seaweeds as physiologically active food items.

### ACKNOWLEDGEMENTS

This work was supported by a research grant from The Center for Industrial Technology in Seowon University, 1997.

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(Received July 14, 1999)