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A novel method for producing softened edible seaweed kombu

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ABSTRACT

Kombu is difficult to soften by conventional cooking. We developed a novel softened kombu, while maintaining its original appearance, for elderly people with chewing or swallowing difficulties. Kombu treated with enzyme and phosphate buffer (EP) resembled conventionally-cooked kombu (C) in appearance, and was soft enough to be mashed completely using the tongue and upper jaw. The firmness of EP was 32 kPa, which was 1/30th that of C, and the stickiness of EP was lower than that of kombu treated only with phosphate buffer (P). The calcium content of EP decreased to 70% of that of C. The cell wall of EP was distorted compared with that of C. Scanning electron microscopy showed that the cell wall of EP was a mesh surface composed of microfibers and void spaces, unlike the dense structure of C and the spongy structure of P. These results suggest that the softening was caused by removing calcium from alginate in kombu using phosphate buffer and distorting the cell wall. The reduction in stickiness was considered to come from the change in the cell wall microstructure by proteolysis that prevented viscous sodium alginate from flowing out.

1. Introduction

Edible seaweed is an especially popular food among elderly people in Japan. The rate of Japanese elderly people aged 60–79 years old who eat seaweed more than once a month is reportedly 98.5% (Iso, Date, Noda, Yoshimura, & Tamakoshi, 2005). Kombu, which generally signifies the edible seaweed belonging to the Laminariaceae family of brown algae (Phaeophyceae), has been used most often in Japanese seaweed dishes (Nakama & Nakata, 1999). In nutritional aspect, kombu is abundant in dietary fiber, accounting for approximately 30–40 g/100 g the dry weight of the alga body (MacArtain, Gill, Brooks, Campbell, & Rowland, 2007; Suzuki, Ohsugi, Yoshie, Shirai, & Hirano, 1996; Yoshie, 2001). Kombu, in particular, includes a large amount of soluble dietary fiber that is rich in functionality, such as alginate and fucoidan. Alginate reportedly has anti-hypercholesterolemic and anti-hypertensive activity (Ren, Noda, Amano, Nishino, & Nishizawa, 1994; Tsuji et al., 1968; Tsuji, Tsuji, & Suzuki, 1978) and activity in promoting pancreatic function (Ikegami et al., 1984; Ikegami, Hosoda, Tosen, Umeki, & Yamada, 2006). Fucoidan reportedly has anti-tumor and anti-angiogenic activity (Gupta & Abu-Ghannam, 2011; Koyanagi, Tanigawa, Nakagawa, Soeda, & Shimeno, 2003; Maruyama, Tamanuchi, Hashimoto, & Nakano, 2003), anti-viral activity (Ponce, Pujol, Damonte, Flores, & Stortz, 2003), and anti-coagulant and anti-thrombotic activity (Berteau & Mulloy, 2003; Millet et al., 1999; Mourão & Pereira, 1999). Kombu contains a wide diversity and large quantity of minerals, such as calcium, potassium, iron, and iodine (MacArtain et al., 2007).

Although kombu is familiar and has been eaten daily in Japan, elderly people with chewing or swallowing difficulties tend to avoid it because the elastic firmness of kombu requires that the people consuming it bite off and chew the kombu with their teeth. To provide ingredients that are difficult to eat like kombu, these foods are minced or ground into a paste. However, the minced or paste food substance does not look appetizing due to the large difference from the original appearance, which leads to the decrease of appetite, pleasure to eat and quality of life in the elderly (Cichero et al., 2013; Miyake, Ishii, Adachi, & Ikeda, 2009; Miyake, 2010; Nagai, Suzuki, Shibata, & Matsumoto, 1994). For this reason, the development of a method that makes kombu, which has high palatability, nutrition and functionality, soft enough to mash with the tongue, while maintaining its original appearance, is required for elderly people with chewing or swallowing difficulties. To date, there are few reports regarding softened kombu that has maintained its original appearance. Dried kombu was reportedly softened by boiling in cooking solutions; however, there are no descriptions stating that the softening process resulted in achieving

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both an increase in softness while maintaining the appearance at the same time (Okuda & Nakagawa, 1987).

The purposes of this study are to produce kombu that was soft enough to mash completely and smoothly with the tip of the tongue and upper jaw for the consumption by elderly people with chewing or swallowing difficulties, while maintaining the original appearance, and to analyze the texture and microstructure of the cell wall.

2. Materials & methods

2.1. Sample

A conventionally-cooked kombu was produced according to the following method. Dried kombu, *Saccharina japonica* (*Laminaria japonica*), which is harvested in Hokkaido, was purchased from Takase Bussan Co., Ltd., Tokyo, Japan. The dried kombu material was soaked for 1 h in water at 20 °C, was subsequently taken out of the water, cut into approximately 4 cm squares, was heated with steam at 100 °C for 20 min, and was cooled down for 1 h at 4 °C in a refrigerator. This conventionally-cooked kombu was labeled as the "C" sample.

In order to produce a softened kombu with low stickiness, a following softening treatment was performed. The dried kombu material was soaked for 1 h in water at 20 °C, was subsequently taken out of the water, cut into approximately 4 cm squares, was heated with steam at 100 °C for 20 min, and was cooled down for 1 h at 4 °C in a refrigerator as well as C. This cooked kombu was immersed in an enzyme solution that included 1 g/100 g protease (Yakult Pharmaceutical Industry Co., Ltd, Tokyo, Japan) with 0.3 mol/L sodium phosphate buffer (pH 8.0) for 15 h at 4 °C, was taken out of the solution, and heated with steam at 85 °C for 5 min to deactivate the enzyme. This sample, which was treated with enzyme and phosphate buffer, was labeled as the "EP" sample.

The sample which was treated with only sodium phosphate buffer without protease was produced according to the following method. The dried kombu material was soaked for 1 h in water at 20 °C, was subsequently taken out of the water, cut into approximately 4 cm squares, was heated with steam at 100 °C for 20 min, and was cooled down for 1 h at 4 °C in a refrigerator as well as C. This cooked kombu was immersed in 0.3 mol/L sodium phosphate buffer (pH 8.0) for 15 h at 4 °C, was taken out of the solution, and heated with steam at 85 °C for 5 min as well as EP. This sample, which was treated only with phosphate buffer, was labeled as the "P" sample.

C, EP, and P were frozen at −20 °C. These samples were thawed and used in the following assessments: sensorial, textural, chemical, and microstructural. Three individual batches of kombu were produced and the samples were provided for respective experiments without mixing batches in this study. We confirmed that there was no difference among batches in preliminary tests.

2.2. Sensory evaluation

The sensory attributes of each sample, such as appearance, taste, firmness, and stickiness, were evaluated with scoring method by six skilled panelists (five males and one female, 27–35 years old) (Inoue, 2012). The panelists graded the test samples on a scale from 1 to 5 according to the following definitions. A score of 5 was defined as "being able to recognize the sample as kombu clearly at a glance," "having a similar taste to kombu itself," "being soft enough to mash completely with the tip of the tongue and upper jaw without using teeth or gums," "not being sticky at all during chewing or swallowing," and a score of 1 represented the opposite meaning. The high score of a sensory attribute meant that the sample had a high possibility to suit foods for elderly people with chewing or swallowing difficulties in terms of the sensory attribute. The samples were presented to each panelist at the same time and were evaluated in a random order. All samples were coded with random numbers. The panelists were provided water to clean their mouth in in-between tastings. The sensory evaluation was performed once.

2.3. Puncture test

The firmness of each sample was measured using a creep meter (RE2-33005B; Yamaden Co., Ltd., Tokyo, Japan) in accordance with a modified method for puncture tests for films because kombu is very thin and its thickness is approximately 1–2 mm (Landová et al., 2014). The measurement was performed with a columnar plunger (1.5 mm in diameter) at a rate of 10 mm/s until the plunger pierced the sample, and maximum stress was defined as firmness. The measurement was performed ten times.

2.4. Probe tack test

The stickiness of each sample was examined using the creep meter (RE2-33005B; Yamaden Co., Ltd.) in accordance with a modified method for probe tack tests for pressure sensitive adhesives (Zosel, 1989). The sample was pressed on a plastic plunger (20 mm in diameter) until the sample was distorted to 66.7% of its original height in order to be adhered to the plunger, after which measurements were taken as the plunger was lifted off the sample at a rate of 10 mm/s. The total work needed from the start of the measurement to the complete separation of the plunger and the sample was defined as stickiness. The measurement was performed six times.

2.5. Analysis of calcium content

Calcium content was determined using microwave digestion and inductively coupled plasma optical emission spectrometry (ICP-OES) in accordance with the analysis methods for nutrition information in the Nutrition Labeling Standards (Ministry of Health, Labour and Welfare of Japan, 1999). Microwave digestion was performed using Multiwave 3000 (PerkinElmer, Inc., Waltham, USA), and ICP-OES was performed using the Perkin Elmer Model Optima 5300 DV spectrometer (Perkin Elmer, Inc.) at a wavelength of 318 nm. The measurement was performed three times.

2.6. Light microscopy

The specimens were fixed in 0.28 mol/L glutaraldehyde in 0.1 mol/L sodium phosphate buffer for 2 h. The samples were then washed with 0.1 mol/L sodium phosphate buffer, post-fixed in 1 g/100 mL OsO_4 buffered with 0.1 mol/L sodium phosphate buffer for 2 h, dehydrated in a graded series of ethanol and embedded in Epon 812. Semi-thin sections were cut at 1 μm using Ultract S microtome (Reichert, Wien, Austria). The sections were stained with 0.5 g/100 mL toluidine blue buffered with 0.1 mol/L sodium phosphate buffer for 2 min at 85 °C, and washed with ultrapure water (Ichinose, Tagami, Muneta, Mukohyama, & Sekiya, 2003). A cross section of each sample was observed using light microscopy (VHX-1000; Keyence Corporation, Osaka, Japan).

2.7. Scanning electron microscopy

A microstructure of the surface of the epidermis of each sample was observed using scanning electron microscopy (SEM). The specimens were fixed in 0.28 mol/L glutaraldehyde in 0.1 mol/L