Note

Detection of Fucoidan in Urine after Oral Intake of Traditional Japanese Seaweed, Okinawa mozuku (Cladosiphon okamuranus Tokida)

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(Received September 28, 2016)

Summary Seaweed has been considered an indigestible food. Fucoidan, which is found abundantly in seaweed, especially in Cladosiphon okamuranus (Okinawa mozuku), has a high molecular weight and has been long believed to be hardly absorbed in the human digestive system due to a lack of certain digestive enzymes. We previously reported that fucoidan can be detected in serum and urine after oral intake of purified fucoidan in humans and rats. However, it is unclear whether the fucoidan in mozuku can be absorbed after digestion of mozuku. Therefore, we attempted to detect fucoidan in urine before and after mozuku intake. We determined the fucoidan concentration in urine after oral intake of Okinawa mozuku and urinary fucoidan was detected in several volunteers. In conclusion, these results suggest that fucoidan in mozuku can be absorbed after ingestion of mozuku.

Key Words fucoidan, Cladosiphon okamuranus (Okinawa mozuku), urine, oral intake

According to the National Nutrition Survey in Japan, an adult Japanese eats about 14.3 g of seaweed daily (1, 2), and, other than Japanese, few people in the world eat seaweed as part of a traditional diet. Seaweed typically eaten in Japan includes konbu (Laminaria japonica), wakame (Undaria pinnatifida), hijiki (Hizikia fusiforme), nori (Porphyra tenera), and mozuku (Nemacystus decipiens). Since seaweed contains abundant indigestible dietary fiber, it has been considered an indigestible food. Mozuku is a type of brown seaweed of the genera Cladosiphon and Sphaerocysta, and several kinds of mozuku, such as ishimozuku, futomozuku, kuromo and Okinawa mozuku, grow naturally along the coast of Japan. Okinawa mozuku (Cladosiphon okamuranus Tokida) grows in a subtropical climate from the Amami Isles to the Yaeyama Isles, and only this mozuku has been cultivated commercially. Japanese people are familiar with most mozuku species and consume them as daily food (3). Mozuku, as well as konbu and wakame, contains approximately 1% fucoidan, based on wet weight (4).

Fucoidan is mainly composed of fucose and sulfate bases, each with a molecular weight typically of a magnitude of 1,000 kDa, and the molecular weight of fucoidan from Okinawa mozuku is 3,200 kDa (4, 5). Fucoidan is known to have various biological activities including antithrombotic (6), anti-inflammatory (7–9), antiviral (10, 11), anti-adhesive (12), and antitumor (13–15) activity. Fucoidan also plays a role in modulating host immune systems (16), arresting the cell cycle, and inducing apoptosis (17). It has been long believed that fucoidan, with a high molecular weight, is hardly absorbed in the human digestive system due to a lack of digestive enzymes for fucoidan. Intestinal absorption of fucoidan from Okinawa mozuku has been demonstrated in humans and rats (18, 19); however, it is unclear whether fucoidan in mozuku can be absorbed after digestion of mozuku.

We previously reported that the concentration of fucoidan in urine is significantly higher than in serum. Therefore, in order to elucidate the intestinal digestion of mozuku and the absorption of fucoidan, we examined the concentration of fucoidan in urine before and after oral intake of Okinawa mozuku in Japanese men and women.

Materials and Methods

Subjects. Forty-eight volunteers (34 males and 14 females with an average age of 45 y and an age range of 20–80 y) in Okinawa Prefecture, and 38 volunteers (17 males and 21 females with an average age of 23 y and an age range of 22–29 y) in Gunma Prefecture (with no seacoast), were enrolled in this study. Volunteers abstained from oral intake of seaweed and seaweed-derived products 24 h prior to this study. Some volunteers whose urinary fucoidan measured above 10 ng/mL before ingestion of mozuku were excluded.

Each volunteer ingested 100 g of Okinawa mozuku, containing 1 g of fucoidan, at 9:00 AM. Urine was collected 0, 3, 6, and 9 h after mozuku intake by spontaneous micturition at each point. In 4 men and 6 women out of 38 Gunma volunteers, blood samples were collected from the peripheral vein 0, 3, 6, and 9 h after mozuku ingestion, and serum was separated from whole blood by centrifugation.

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This study was carried out according to the Declaration of Helsinki and the study protocol was approved by the Ethics Committee of Gunma University (No. 11-11). Following an explanation of the study and its aims, all subjects gave informed consent.

Materials. Fucoidan powder was obtained from C. okamuranus Tokida (Okinawa mozuku) and Okinawa mozuku was obtained from South Product Co., Ltd. (Okinawa, Japan).

Quantification of fucoidan concentration in urine and serum. Fucoidan concentrations in urine and serum were determined using a sandwich ELISA method developed by our laboratory (3). The details of the method are described in supplementary information.

Statistical analysis. Differences in values for among several groups were analyzed using Kruskal Wallis analysis followed by the Steel-Dwass multiple comparison test using R version 3.4.1 for windows (R Development, Vienna, Austria). Statistical significance was defined as $p<0.05$.

Results

Urinary fucoidan concentrations before intake of mozuku showed an age-related difference in Okinawa volunteers between those in their 20 s and 30 s, although they did not eat any seaweed on the day prior to mozuku intake (Supplemental Online Materials, Fig. S1). Urinary fucoidan was detected (>10 ng/mL) in 14 out of 48 Okinawa volunteers, and a concentration of more than 20 ng/mL was found in 3 volunteers. In the Okinawa volunteers, urinary fucoidan was detected in subjects in their 40 s (57.1%), and the detection rate was similar among subjects in their 30 s, 50 s and 60 s. In addition, urinary fucoidan in Gunma volunteers was not detected, and a similar trend was seen in Okinawa volunteers in their 20 s (Supplemental Online Materials, Fig. S1).

In order to evaluate mozuku digestion and absorption in the intestinal tract, we determined the urinary concentration of fucoidan after oral intake of 100 g of mozuku. In the Okinawa volunteers, a urinary fucoidan concentration was detected in 1, 5, and 4 out of 34 subjects at 3 h, 6 h, and 9 h after mozuku intake, respectively. The highest fucoidan concentration was 37.2 ng/mL (Fig. 1).

Before mozuku intake, urinary fucoidan was not detected in the Gunma volunteers. After mozuku intake, urinary fucoidan in the Gunma volunteers was detected in 1, 7, and 17 subjects at 3 h, 6 h and 9 h, respectively, and, except for 1 subject, all had fucoidan concentrations of under 20 ng/mL (Supplemental Online Materials, Fig. S2).

In serum collected from 10 Gunma volunteers, fucoidan concentration was not detected before or after intake of Okinawa mozuku, although urinary fucoidan was detected in 4 out of the 10 volunteers after mozuku intake (data not shown).

Discussion

Brown seaweed has been considered an indigestible food, and absorption of its fiber is naturally impossible. The high molecular weight of fucoidan has also been believed to be unabsorbable because there are no digestive enzymes for fucoidan in human intestines. On the other hand, Irhimeh et al. first reported fucoidan absorption in human intestine after oral administration (20). We confirmed further details on the kinetics of fucoidan, intestinal absorption and urinary excretion in vivo and in vitro. The uptake of fucoidan through the intestinal tract seemed to be low, but was measurable by our ELISA method (18). The transport of fucoidan across Caco-2 cells was demonstrated in a dose-dependent manner (21,22) and reached a maximum after 1 h, followed by a rapid decrease. Immunohistochemical staining revealed that fucoidan accumulated in jejunal macrophages and hepatic Kupffer cells in fucoidan-treated rats (22). The molecular weight of fucoidan detected in serum was similar to that of the ingested fucoidan.

To the best of our knowledge, this is the first study to detect fucoidan in urine after mozuku intake. Although the detailed mechanism of mozuku digestion and fucoidan absorption is unclear and there is an age difference

Fig. 1. Time course of urinary fucoidan concentrations after mozuku intake in Okinawa male and female volunteers.
among volunteers, there were marked differences in urinary fucoidan concentration among individuals. It is possible that these differences relate to the difference in biological effect of fucoidan.

Our previous study showed that the fucoidan concentration after 1 g of purified fucoidan ingestion was elevated in the serum and urine of several volunteers. Fucoidan concentrations increased up to 100 ng/mL in the serum and up to 1,000 ng/mL in the urine (18). In this study, although only noted in Gunma volunteers, fucoidan was not detected in serum after *mizuku* ingestion, but was detected in the urine of several volunteer. In addition, the urinary fucoidan concentration was lower in those receiving 100 g of *mizuku* than in those receiving 1 g of purified fucoidan. These findings suggest that *mizuku* was incompletely digested by the gastrointestinal tract. Therefore, we assume that the fucoidan concentration in serum after *mizuku* intake was lower than the lower detection limit of our ELISA method.

In conclusion, we detected fucoidan in urine after oral intake of *Cladosiphon okamuranus (Okinawa mizuku)* in several Japanese volunteers. The fucoidan concentration was significantly higher after intake of Okinawa mizuku than before intake, although it is necessary to test this in age-matched volunteers and to adjust for creatinine levels. These results suggest that fucoidan in *mizuku* can be absorbed after digestion of *mizuku* and that there are marked differences in urinary fucoidan concentrations among individuals.

Acknowledgments

We gratefully acknowledge Dr. Hiroshi Mochida for supplying the anti-fucoidan serum, and Dr. Yoshiro Inunuma, South Product Co., Ltd., for administering fucoidan to the Okinawa volunteers. This work was supported by the Japanese Society of Clinical Chemistry Kanto Branch Project Research (No. 4).

Supporting Information

Supplemental Online Material is available on J-STAGE.

REFERENCES


