ORIGINAL

Dietary zinc intake and its effects on zinc nutrition in healthy Japanese living in the central area of Japan

Nobuko Sarukura[1], Miho Kogirima[2], Shinji Takai[3], Yoshiaki Kitamura[4], Bukasa Kalubi[4], Shigeru Yamamoto[5] and Noriaki Takeda[4]

[1]*Project for the National Health and Nutrition Survey, Nutritional Epidemiology Program, National Institute of Health and Nutrition, Tokyo, Japan ;* [2]*Department of Food Science and Nutrition, Doshisha Women's College, Kyoto, Japan ;* [3]*Department of Pharmacology, Osaka Medical School, Osaka, Japan ;* [4]*Department of Otolaryngology and Communicative Neuroscience, Institute of Health Bioscience, the University of Tokushima Graduate School, Tokushima, Japan ; and* [5]*Department of International Nutrition, Graduate School of Humanities and Sciences, Ochanomizu University, Tokyo, Japan*

Abstract : In the present study, we first examined the dietary zinc intake from food groups in 109 healthy Japanese (24-82 years old, 45 male and 64 female) by means of the 72-h recall method. We then used the ratio of apo/holo-activities of angiotensin converting enzyme (ACE ratio) that is a more sensitive index of zinc nutrition than zinc concentration in the serum and examined the correlation between their zinc intake and ACE ratio. Dietary zinc intake in healthy Japanese was maximal from rice and rice products. There were significant inverse correlations between the ACE ratio and dietary zinc intake from rice and rice products and shellfish, and a significant positive correlation between ACE ratio and dietary zinc intake from other beans and bean processed foods. On the other hand, there were no significant correlations between serum zinc concentrations and dietary zinc intake from any food group except processed fish. These findings suggested that rice is a major source of dietary zinc intake in healthy Japanese. It is also suggested that shellfish also has a major impact on zinc nutrition, although dietary zinc intake from this source is minimal. Since beans contain phytic acid, which inhibits the absorption of dietary zinc, it is suggested that intake of beans causes impairment of zinc nutrition. J. Med. Invest. 58 : 203-209, August, 2011

Keywords **:** *zinc nutrition, dietary zinc intake, zinc concentration in serum, angiotensin converting enzyme*

INTRODUCTION

Zinc is an essential trace element playing a role in several physiological functions in both humans and animals. In fact, zinc deficiency has been associated with growth disturbance (1-3), impairment of special senses including vision, taste and smell (4, 5), anorexia (6.7), dermatitis (8), sexual dysfunction as well as fetal and pregnancy complications (9-12). Especially, a major clinical manifestation of zinc deficiency is taste impairment, as shown by double-blind, placebo-controlled studies that have shown the efficacy of zinc supplementation in the taste recovery in patients with hypogeusia and low serum zinc concentration (13, 14).

But, in another double-blind, placebo-controlled

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Address correspondence and reprint requests to Noriaki, Takeda, M.D., Ph.D., Department of Otolaryngology and Communicative Neuroscience, Institute of Health Bioscience, the University of Tokushima Graduate School, 3-18-15 Kuramoto, Tokushima 770-8503, Japan and Fax : +81-88-633-7170.

study, Yoshida *et al.* reported the therapeutic effect of zinc picolinate in patients with idiopathic taste impairment with normal zinc levels in the serum (15). Therefore, it was suggested that zinc deficiency is a predominant factor underlying hypogeusia even when zinc concentrations are within normal ranges in the serum. It was also suggested that serum zinc concentration that has been widely used to assess zinc nutrition (16) is not a reliable indicator of the zinc nutritional status, probably because most zinc is associated with albumin and α-microglobulin in the blood, of which levels are influenced by stress and invasion, with the remaining less than 5% being the free form of zinc (17).

Recently, we developed a new test for the assessment of zinc deficiency taking the ratio of apo/holo-activities of angiotensin converting enzyme (ACE), a zinc dependent enzyme, as an index, suggesting that the ratio of apo/holo-ACE activities (ACE ratio) is a more sensitive index of zinc nutrition than measuring zinc concentration in the serum (18-20). In the present study, we first examined the daily zinc intake from each food group in healthy Japanese living in the central area of Japan (Nagano prefecture) by means of the 72-h recall method. We then used ACE ratio and zinc concentration in the serum as indices and examined the correlations between dietary zinc intake and zinc nutrition in healthy Japanese.

MATERIALS AND METHDS

Subjects

The present study included 109 healthy Japanese (24-82 years old, 45 male and 64 female) who participated in an annual mass health examination program of the Kita-mimaki area of Tomi city in Nagano prefecture (the central area of Japan) (21) in 2005. Their mean body height and weight was 158.6±7.8 cm and 58.9±9.0 kg, respectively. This study was performed in accordance with the declaration of Helsinki. The purpose and procedures of the study were explained to all subjects before obtaining informed consent. Dietary survey and blood sampling were performed in each subject in the morning.

Dietary zinc intake

Dietary survey was performed using the 72-h recall method (22). Dietitians interviewed each subject to identify foods and beverages ingested in the last three days. Subjects' dietary zinc intakes by food group were then calculated with the Standard Tables of Food Composition in Japan (5th revised and enlarged edition) (23). Foods recorded in the survey were grouped into 33 groups : rice and rice products, flour and flour products, other cereals, potato and potato products, other starch, sugar and sweetener, soybean and soybean processed foods, other beans and bean processed foods, seeds and nuts, green and yellow vegetables, other vegetables, vegetable juice, pickles, fruit, jam, fruit juice, mushrooms, seaweed, fish, processed fish, shellfish, beef, pork and other meat, poultry, organ meat, other meat, eggs, milk and dairy products, oils and fats, sweets, alcohol, other beverages, seasonings and spices.

Zinc concentration

Zinc concentration in the serum was measured by means of atomic absorption spectrometry (24) by SRL Co., Ltd. (Tokyo, Japan). Normal ranges of zinc concentrations in the serum are from 65 to 110 μg/dl.

ACE ratio

ACE activity in the serum was measured using a synthetic substrate, hippuryl-His-Leu (HHL), specifically designed for ACE (Peptide Institute Inc. Osaka, Japan). Twenty five μl of serum were incubated for 30 min at 37°C with 5 mmol/l HHL in 100 μl of 100 mmol/l phosphate buffer, pH 8.3, containing 600 mmol/l NaCl. The reaction was terminated by addition of 375 μl of 3% metaphosphoric acid, and then the mixture was centrifuged at 10,000 rpm for 10 min at 4°C. The supernatant was applied to a reverse-phase column (4 mm i.d. × 250 mm ; IRICA Instrument, Kyoto, Japan), which had been equilibrated with 10 mmol/l KH_2PO_4 and CH_3OH (1 : 1, pH3.0), and eluted with the same solution at a rate of 0.5 ml/min. Hippuric acid was detected by ultraviolet absorbance at 218 nm. One unit of ACE activity was defined as the amount of enzyme that cleaved 1 μmol hippuric acid/min (25).

Since ACE is a zinc-metallo enzyme, holo-ACE with zinc shows full ACE activity. After measuring ACE activity in the serum, ACE activity was then measured by the addition of zinc (150 μM in phosphate buffer at pH 8.3) to the serum *in vitro*. The increase of activity over the initial holo-ACE activity was determined as that of apo-ACE in the serum (26). The ratio of apo/holo-ACE activities was calculated as follows : ACE ratio (%)=apo-ACE activity/holo-ACE activity × 100 (15-17).

Statistical analysis

The Pearson correlation analysis was used to evaluate the relationships among groups (Statistical Package for Social Science, version 16.0, Japan Inc., Tokyo, Japan). Results were considered statistically significant at *p*-values < 0.05.

RESULTS

The average total dietary zinc intake was 9.49± 3.11 mg/day (mean± S.D.) in 109 healthy Japanese (Table 1). Among food groups, subjects' dietary zinc intake was maximal from rice and rice products at 1.97± 0.90 mg/day, followed by milk and dairy products, soybeans and soybean processed foods, beef,

Table 1 Dietary zinc intake from food groups in healthy Japanese.

Food group	Dietary zinc intake (mg/day, mean ± S.D., n = 109)
Rice and rice products	1.97± 0.90
Milk and dairy products	0.91± 0.55
Soybean and soybean processed foods	0.83± 0.55
Beef, pork and other meat	0.81± 0.66
Fish	0.77± 0.71
Other vegetables	0.76± 0.53
Eggs	0.47± 0.28
Green and yellow vegetables	0.46± 0.33
Flour and Flour products	0.39± 0.30
Processed fish	0.36± 0.72
Other beverages	0.22± 0.29
Fruit	0.20± 0.19
Seasonings	0.20± 0.09
Sweets	0.16± 0.22
Potato and potato products	0.13± 0.11
Seeds and nuts	0.13± 0.19
Poultry	0.12± 0.22
Mushrooms	0.08± 0.08
Shellfish	0.07± 0.17
Other cereals	0.07± 0.16
Seaweed	0.06± 0.05
Other beans and bean processed foods	0.06± 0.17
Organ meats	0.04± 0.24
Alcohol	0.02± 0.05
Vegetable juice	0.01± 0.03
Spices	0.01± 0.01
Other meat	0.00± 0.04
Pickles	0.00± 0.01
Oils and fats	0.00± 0.00
Sugar and sweeteners	0.00± 0.01
Fruit Juice	0.00± 0.01
Jam	0.00± 0.00
Other starch	0.00± 0.00
Total	9.49± 3.11

pork and other meat, and fish. Although dietary zinc intake from flour and flour products was 0.39± 0.30 mg/day, the dietary zinc intake from rice and rice products correlated inversely with that from flour and flour products (R= -0.46, $p < 0.001$).

There were significant inverse correlations between the ACE ratio and dietary zinc intakes from rice and rice products (R= -0.190, $p < 0.05$), shellfish (R= -0.207, $p < 0.05$) and other starches (R= -0.268, $P < 0.01$) in healthy Japanese (Table 2). On the other hand, there were significant positive correlations between the ACE ratio and dietary zinc intakes from other beans and bean processed foods (R=0.333, $p < 0.001$) and poultry (R=0.265, $p < 0.01$). However, there was only a significant positive correlation between zinc concentration in the serum and dietary zinc intake from processed fish (R=0.192, $p < 0.05$) in healthy Japanese (Table 2).

There was an significant inverse correlation between the ACE ratio and zinc concentrations in

Table 2 Correlation between dietary zinc intake from food groups and ACE ratio or zinc concentration in the serum of healthy Japanese.

Food group	R	
	ACE ratio	Serum zinc concentration
Rice and rice products	-0.190*	0.093
Milk and dairy products	0.075	-0.086
Soybean and soybean processed foods	-0.001	0.022
Beef, pork and other meat	0.014	0.052
Fish	0.060	-0.011
Other vegetables	0.107	0.024
Eggs	-0.078	0.065
Green and yellow vegetables	0.141	-0.147
Flour and Flour products	0.156	-0.083
Processed fish	-0.095	0.192*
Other beverages	0.030	0.002
Fruit	0.107	-0.003
Seasonings	-0.127	0.057
Sweets	0.184	-0.144
Potato and potato products	0.074	-0.015
Seeds and nuts	0.079	0.047
Poultry	0.265**	-0.168
Mushrooms	0.023	0.007
Shellfish	-0.207*	0.161
Other cereals	-0.029	-0.050
Seaweed	-0.143	0.167
Other beans and bean processed foods	0.333***	-0.158
Organ meats	0.008	-0.004
Alcohol	0.119	-0.106
Vegetable juice	-0.122	0.091
Spices	-0.029	-0.011
Other meat	0.104	-0.132
Pickles	0.092	-0.080
Oils and fats	0.046	-0.074
Sugar and sweeteners	0.021	0.006
Fruit Juice	-0.075	0.057
Jam	0.062	-0.093
Other starch	-0.268**	0.045
Total	0.047	0.036

*$p < 0.05$, **$p < 0.01$, ***$p < 0.001$

the serum in healthy Japanese (R= -0.549, p< 0.001) (Fig. 1). But, there were no correlations between dietary zinc intake and the ACE ratio or zinc concentrations in the serum (Figs. 2 and 3).

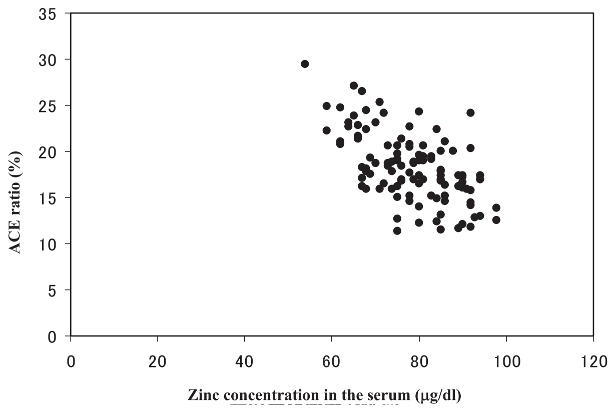

Fig. 1 : Inverse correlation between ACE ratio and zinc concentration in the serum in healthy Japanese living in Nagano prefecture (R= -0.549, p< 0.001).

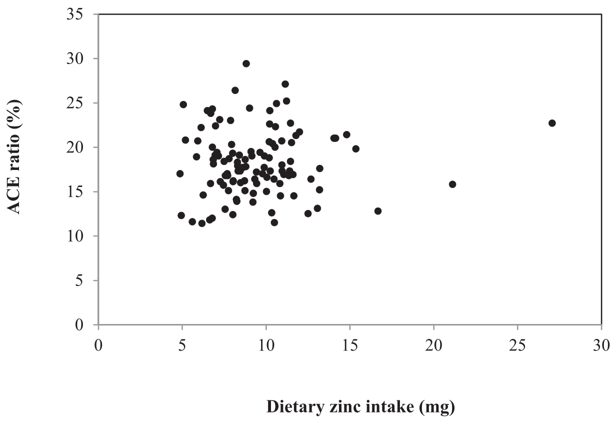

Fig. 2 : No correlation between dietary zinc intake and ACE ratio in healthy Japanese Nagano prefecture (R=0.047).

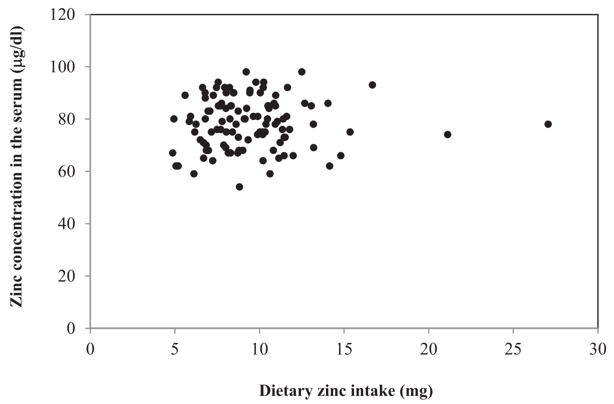

Fig. 3 : No correlation between dietary zinc intake and zinc concentration in the serum in healthy Japanese living in Nagano prefecture (R=0.036).

The average total dietary intake of energy, protein, fat and carbohydrate in 109 healthy Japanese were 1914± 448 kcal/day, 79.4± 20.8 g/day, 48.0± 15.3 g/day and 277.9± 75.9 g/day, respectively.

DISCUSSION

In the present study, the average total dietary zinc intake in healthy Japanese living in Nagano prefecture was 9.49± 3.11 mg/day, which is slightly more than the averaged Japanese value of dietary zinc intake (8.33 mg/day) from the National Nutrition Survey in Japan conducted by the Ministry of Health, Labor and Welfare, 2005 (27). Their dietary zinc intake was maximal from rice and rice products at 1.97± 0.90 mg/day, which was followed by milk and dairy products, soybeans and soybean processed foods, beef, pork and other meat, and fish. Although the concentration of zinc in rice is not so high (0.6 mg/100 g of well-milled rice from the Standard Tables of Food Composition in Japan) (23), it is suggested that rice and rice products are a major source of dietary zinc intake, probably because most Japanese eat large amounts of rice as a staple food. On the other hand, dietary zinc intake from flour and flour products, which are a minor source of zinc for most Japanese, was 0.39± 0.30 mg/day. However, the dietary zinc intake from rice and rice products correlated inversely with that from flour and flour products. This finding suggested that flour and flour products are another major source of dietary zinc intake in some Japanese who eat bread as a staple food.

There were inverse correlations between the ACE ratio and dietary zinc intake from rice and its products, and from shellfish. Since healthy Japanese receive zinc maximally from rice as a staple food, it is suggested that rice has a major impact on zinc nutrition as evaluated by ACE ratio, in addition to its being a major source of dietary zinc intake. On the other hand, dietary zinc intake from shellfish was minimal (0.07± 0.17 mg/day). However, the inverse correlation between the ACE ratio and dietary zinc intake from shellfish suggested that shellfish also has a major impact on zinc nutrition in healthy Japanese, probably because shellfish contains high concentrations of zinc (for example, 13.2 mg/100 g in oyster from Standard Tables of Food Composition in Japan) (23).

Although dietary zinc intake from other beans and bean processed foods was 0.06± 0.17 mg/day, there was a positive correlation between the ACE ratio and dietary zinc intake from other beans and bean processed foods. Generally, healthy Japanese consume a considerable amount of beans, which contains a large amount of phytic acid (for example, 0.61-2.38 mg/100 g of kidney beans) (27, 28). Since phytic acid has been shown to inhibit the absorption of dietary zinc from the bowel (29), it is suggested that intake of beans causes impairment of zinc nutrition in healthy Japanese.

Although there was a significant inverse correlation between the ACE ratio and zinc concentration in the serum, there were no significant correlations between zinc concentration in the serum and dietary zinc intake from any food groups except processed fish. These findings were in line with our previous conclusion that the ACE ratio is a more sensitive indicator than the concentration of zinc in the serum in evaluating dietary zinc nutrition (18-20).

Although number of subjects tested in the present study was limited and their food intake may vary from day/season to day/season, their dietary intake of energy, protein fat and carbohydrate were in line with those reported from the National Nutrition Survey in Japan conducted by the Ministry of Health, Labor and Welfare, 2005 (27).

In conclusion, in the present study, the dairy zinc intake from food groups and the effects of food groups on zinc nutrition using the ACE ratio as an index were examined in healthy Japanese living in Nagano prefecture. It was suggested that rice and rice products are a major source of dietary zinc intake in healthy Japanese, probably because most Japanese eat large amounts of rice, which contains low concentrations of zinc, as a staple food. It was also suggested that shellfish, which contains high concentration of zinc, also has a major impact on zinc nutrition, although its dietary zinc intake was minimal. Since beans generally contain a large amount of phytic acid, which inhibits the absorption of dietary zinc from the bowel, it is suggested that intake of beans causes impairment of zinc nutrition.

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