

Effects of Cooking Methods on the Retention of Vitamin B₆ in Foods, and the Approximate Cooking Loss in Daily Meals

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Data were obtained that might be applicable to estimate the cooking loss of vitamin B₆ (B₆) in daily meals against the actual B₆ intake. Based on the result of our dietary survey of 19 healthy women, the amount of B₆ in meals after cooking measured by HPLC was approximately 13% of that before cooking (calculated by the standard food composition table). To further ascertain the actual B₆ loss, the meals from the dietary records were reproduced, and the result obtained from measuring B₆ in these meals before and after cooking also proved to be approximately 13%. Most of the animal foods in the daily meals studied were cooked by dry heating, resulting in a relatively low level of B₆ loss, whereas many plant foods were cooked by wet heating. The cooking loss was greatest when foods were boiled, followed by deep-frying and sauteeing in that order. If the water used for boiling, which can be expected to contain dissolved B₆, was taken with the ingredients, however, boiling was found to result in relatively little B₆ loss.

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INTRODUCTION

Loss and retention of vitamins in food during cooking have been reported elsewhere in the literature. Much of the experimental data pertaining to cooking loss has been of limited use, however, because they reflect only controlled laboratory conditions, rather than conditions likely to be encountered outside a laboratory setting. Although information about cooking loss obtained from a case study on an individual food increases the overall basic knowledge of nutrition, it is of limited applicability, and simple averaging of such data does not necessarily result in data of any practical use. For example, studies on the cooking loss of B₁^{1,2)} and B₂^{2,3)} have produced results which differed between the individual foods and meals. Moreover, the relationship between this loss and daily intake of vitamins from meals cannot be properly assessed from these basic data. Daily meals usually consist of several dishes cooked by various methods, and the dish combinations are almost habitual. Furthermore, calculation and analysis methods are also important^{4,5)} for a nutrient analysis. One particularly useful method for calculating the vitamin content of a meal, from the standpoint of nutrition control, is to multiply the total content in all ingredients by the approximate loss

during cooking.

Vitamin B₆ (B₆) was not included in Recommended Dietary Allowances (RDAs) for the Japanese until their 6th revision,⁶⁾ and as a result, there has been insufficient study of B₆ intake in Japan. In western countries, earlier reports on B₆ intake based on dietary surveys have dealt with food sources or health effects related to the quantity of B₆ intake, and most studies of cooking loss were case studies. There have been few reports on the relationship between the cooking loss in daily meals and dietary habit. Moreover major studies of B₆ retention (%) in cooked foods have generally relied on a micro-bioassay to determine the total B₆ content, an approach which does not provide information about differences in the bioavailability of vitamers.^{7,8)} We have previously investigated the relationship between the dietary B₆ intake and serum B₆ level,⁹⁾ and the retention of B₆ in cooked foods as measured by HPLC,¹⁰⁾ but we did not report on B₆ retention in daily meals in relation to the food source and cooking method.

The purpose of this investigation is to determine the approximate cooking loss in daily meals and to collect other useful data pertaining to B₆ intake. We first studied the actual B₆ intake from daily meals by the duplicate meal method, using HPLC, dietary

recording, and measurement of serum B₆ levels, in healthy women. We then conducted an experimental examination of the retention of B₆ in several foods (all of which are good food sources of B₆ and are often consumed), using cooking methods commonly used by respondents to the survey. Finally, we assessed the approximate cooking losses in daily meals by preparing reproduced meals based on random samples from the dietary records.

MATERIALS AND METHODS

B₆ intake using the dietary survey, analysis of duplicate meals and B₆ serum levels

1. Subjects and experimental design

Nineteen healthy female students aged 22–30 years were recruited at Kagawa Nutrition University. They were not consuming any nutrient supplements and were trained in keeping dietary records. To determine the actual B₆ intake, we compiled dietary records over 3 consecutive days (by the dietary weighting method) and collected each subject's duplicate meals. The morning after the third day of dietary recording and duplicate meal collection, fasting blood samples were obtained and total B₆ in the serum was determined. This study was approved by the Human Studies Committee of Kagawa Nutrition University, and written informed consent was obtained from each subject.

2. Analysis of dietary records

The values for the intake of B₆ and other nutrients were calculated from the dietary records, based on a Japanese standard food table.¹¹⁾

3. Analysis of duplicate meals

The B₆ content in the duplicate meal was measured by HPLC. Collected samples were stored at –45°C until needed for analysis. Total samples from each person for a single day were combined and homogenized. B₆ was extracted with 1 × cold HClO₄ according to the method of Toukairin-Oda *et al.*¹²⁾ The B₆ vitamers—pyridoxine (PN), pyridoxal (PL), pyridoxamine (PM), pyridoxal 5'-phosphate (PLP) and pyridoxamine 5'-phosphate (PMP)—were determined according to the method of Edwards *et al.*,¹³⁾ using a post-column reagent containing Na₂S₂O₅. A TSK-gel ODA-80TsQA column (4.6 mm ID × 250 mm, Tosoh, Japan) was used. The separation was performed at room temperature (25°C), using a mobile phase consisting of 0.075 M KH₂PO₄–0.075 M NaClO₄–0.05% N(CH₂CH₂OH)₃–1.5% CH₃CN (pH 3.38) at a flow rate of 1.0 ml/min. The post-column reagent was 0.25 M K₂HPO₄–5.3 mM Na₂S₂O₅ (pH 11.7)

at a flow rate of 1.0 ml/min. The fluorescence spectrophotometer was set at an emission wavelength of 325 nm and an excitation wavelength of 400 nm. Pyridoxine 5'-phosphate (PNP) was determined according to the method of Sierra and Vidal-Valverde,¹⁴⁾ using acid phosphatase (from potato). Pyridoxine 5'-β-glucoside (PNG) was determined according to the method of Gregory and Sartain,¹⁵⁾ using β-D-glucosidase (from almonds). The total B₆ content is expressed as the PN equivalent calculated by converting each vitamer—PN, PL, PM, PLP, PMP, PNP and PNG—into PN. The standard vitamers of B₆ (PN, PL, PM, PLP and PMP), acid phosphatase (from potato) and β-D-glucosidase (from almonds) were obtained from Sigma Chemical Co. (St. Louis, Mo., U.S.A.).

4. Total B₆ content in the serum

The B₆ content in the serum was determined according to the method of Edwards *et al.*¹³⁾

Experimental examination of B₆ retention in selected cooked foods

1. Sample and cooking methods

The sample foods selected were good sources of B₆ and often consumed (according to dietary records) by our subjects, as well as by the Japanese in general. The selected foods were purchased from supermarkets in Saitama Prefecture. Samples were prepared as shown in Table 1. The cooking methods were in two main categories according to heating: dry heating methods (sauteing, grilling and deep-frying) and wet heating method (boiling). These methods were applied to each food in accordance with their cooking properties. Cooking equipment included an electric hot plate (220 ± 20°C) for sauteing, a convection oven (280 ± 20°C) for grilling, and an electric cooker and stainless steel pan (18 cm ID) for deep-frying in 1 l of salad oil (170 ± 10°C). The equipment used for deep-frying was also used for the wet heating method, with distilled water in place of salad oil. The volumes of water used for wet heating are shown in Table 1. An electric rice cooker (Toshiba RCK-10CMT) was used to cook rice.

The heating time for the meat and fish was based on the internal temperature of a sample, because adequate endpoints of heating were designated as the internal temperatures for meat samples given in the guidelines from AMSA¹⁶⁾ and those for fish given by our pre-experiment. The internal temperature of a sample was monitored by a copper-constantan thermocouple placed at the geometric center of the sample. The temperature rose from 20°C to the

Effects of Cooking Methods on the Retention of Vitamin B₆ in Foods, and the Approximate Cooking Loss in Daily Meals

Table 1. Sample preparations and cooking methods

| Food item | Sample size and weight per unit | Method, heating to internal temperature and heating time |
|-------------------------------------------------|------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------|
| Beef sirloin (separable lean) | 5×5×1.5 cm 50 g/piece | saute to 60°C (rare), 70°C (medium), 80°C (well-done) deep-fry to 80°C (approximately 4 min) boil for 10 min |
| Beef round (separable lean) | slice: 5×10×0.2 cm 10 g/piece | saute for 10 s boil for 5 s and 30 min |
| | cube: 3×3×3 cm, 25 g/piece | boil for 60 min |
| Pork loin (separable lean) | 4×4×1 cm 40 g/piece | saute to 85°C (approximately 3 min) deep-fry to 85°C (approximately 2 min) boil to 85°C (approximately 3 min) |
| Pork ham (separable lean) | slice: 4×10×0.2 cm 10 g/piece | saute for 50 s boil for 5 s and 20 min |
| | cube: 3×3×2 cm 25 g/piece | deep-fry to 85°C (approximately 5 min) boil for 30 min |
| Chicken breast Chicken thigh (fresh only) | 3×3×2 cm 30 g/piece | saute to 85°C (approximately 7 min) deep-fry to 85°C (approximately 4 min) boil for 20 min |
| Salmon | 5×10×1.5 cm 60 g/piece | grill and saute to 80°C (approximately 7 min) deep-fry to 80°C (approximately 8 min) boil to 80°C (approximately 8 min) |
| Horse mackerel | 90 g/piece (without head & entrails) | grill to 80°C (approximately 7 min) deep-fry to 80°C (approximately 6 min) boil to 80°C (approximately 11 min) |
| Chicken egg | M size (60 g, 50 g net) 3 pieces (100 g net)/time | scramble for 2 min boil in shell for 13 min |
| Polished rice | 500 g + distilled water 750 g/time | cook for 45 min |
| Soybean | 150 g in 600 g of distilled water soaked for 15 h (5°C) | boil for 90 min |
| Broccoli | cut into small pieces, 20 g/piece 200 g/l, 0.5% NaCl, time | boil for 4 min |
| Carrot | 1 cm round slice, 20 g/slice 300 g/500 g of distilled water, time | boil for 20 min |
| Potato | 5 mm cube×5 cm length 150 g/time | deep-fry for 5 min |
| | divided into 8-12 pieces, 20 g/piece 350 g/500 g of distilled water, time | boil for 20 min |

temperature designated in Table 1. Foods other than meat and fish were heated according to the times and conditions shown in Table 1. Deep-fried foods were

coated with a small amount of flour. Polished rice was washed with distilled water and then soaked in distilled water at 20°C for 1 h before cooking. The rice

was washed in double the weight of water added to the rice, this mixture being stirred 10 times in 10 s, before the water was renewed and this process repeated five times, according to the method of Kainuma *et al.*¹⁷⁾

2. Analysis of B₆ in the food samples

Each raw and cooked food sample which had been cooled to room temperature (25°C) was ground with a food processor. The B₆ vitamers were then extracted from the food sample, the method of extraction and determination of B₆ vitamers being the same as that used for the duplicate meal analysis already described. As with the duplicate meal analysis, the total B₆ content is expressed as the PN equivalent.

3. Calculation of B₆ retention

The retention ratio of B₆ was calculated according to the method of Murphy *et al.*¹⁸⁾

Determination of the B₆ retention ratio in the reproduced meals

Meals from five subjects (5 days) randomly selected from the dietary records were reproduced in order to measure the B₆ content before and after cooking. The B₆ content was measured for each subject (day) by using the method already described.

RESULTS

B₆ intake using the dietary survey, analysis of duplicate meals and B₆ serum levels

1. Determination of the B₆ intake from the dietary records

The average B₆ intake calculated from the dietary records was 1.06 ± 0.19 mg/day (mean \pm SD) and the average protein intake was 56.9 ± 8.5 g/day. The B₆ intake per gram of protein intake was thus 0.020 ± 0.003 mg/g of protein. Most of the subjects therefore met or exceeded the requirement for reference B₆,⁶⁾ the nutrient adequacy ratio (an index of adequacy for a nutrient based on the corresponding RDA⁶⁾ value for that nutrient) for B₆ being 88.3%. As Kim *et al.*¹⁹⁾ have pointed out, imposition of the tasks for recording the food intake and collecting duplicate food samples would probably have reduced the usual food intake.

In our analysis of the relationship between food groups and B₆ intake, the main sources of B₆ were vegetables (21.0%), meats (15.0%), fishes (13.8%), cereals (11.8%) and fruits (10.9%). The cooking methods used in the preparation of foods from the various groups are shown in Table 2. The majority of foods were cooked by heating before intake, the exceptions being dairy products and fruits. Meat, with

the exception of processed foods, was heated. Most of the animal foods were cooked by a dry heating method (Table 2) which resulted in less B₆ loss than that by wet heating (boiling). Most plant foods were eaten raw or were cooked by wet heating (Table 2) which resulted in an outflow of B₆ into the cooking water. Thus, not only the food source but also the heating method was important factors affecting the dietary B₆ intake.

2. Determination of the B₆ intake from the by duplicate meals

The average B₆ intake from the duplicate meal analysis measured by HPLC was 0.91 ± 0.25 mg/day (PN equivalent). The ratio of this value to the value calculated from the dietary records was $86.7 \pm 10.8\%$. Significant correlation was found between the measured value and the calculated value ($r=0.899$, $p<0.01$).

PNG, which has relatively low bioavailability, accounted for 15.8% of the total B₆ intake, a significant correlation being observed between PNG and the total B₆ intake ($r=0.704$, $p<0.01$).

3. Assessment of the B₆ serum level

Total serum B₆ was 11.1 ± 3.3 ng/ml; thus, nobody was observed to have a B₆ deficiency. It is considered that the intake of B₆ was never less than 0.010–0.015 mg/g of protein,²⁰⁾ at which level there is no deficiency. Significant correlation was observed between the intake of B₆ (mg/g of protein) determined by the duplicate meal method and the B₆ level in the serum ($r=0.499$, $p<0.05$).

Experimental examination of B₆ retention in selected cooked foods

1. Retention ratio of B₆ in cooked foods

The retention of B₆ in beef steak decreased with rising internal temperature of the meat as the steak was heated from rare to well-done (Table 3). The retention ratio of B₆ was similar for different types of meat (Table 3), with the exception of rare and medium steaks which both had a relatively high ratio. With identical internal temperature end points of heating, B₆ retention in pork was lower when it was boiled than when it was sauteed. Although B₆ retention was observed to be relatively low for sliced meat boiled for a long time, the shape was not observed to be an important factor with a short heating time. B₆ retention was higher for cube-shaped meat when deep-fried (Table 3). The B₆ retention ratio in scrambled egg and egg boiled in the shell was similar, and it is assumed that the presence of the eggshell offset the effect on retention of the longer

Effects of Cooking Methods on the Retention of Vitamin B₆ in Foods, and the Approximate Cooking Loss in Daily Meals

Table 2. Frequency of using cooking methods per food group from dietary records of three days

(n=19)

| Food group | Cooking method | Actual numbers from records | | | Frequency per person (mean ± SD) |
|----------------------|-------------------------------|-----------------------------|-------------------------|-----------------------|-------------------------------------|
| | | Total ^a | Individual ^b | (Ratio%) ^c | |
| Milks | uncooked ^d | 75 | 75 | (100.0) | 3.8 ± 2.5 |
| Eggs | uncooked | 36 | 1 | (2.8) | 0.1 ± 0.2 |
| | indirect heating ^e | | 21 | (58.3) | 1.1 ± 0.9 |
| | boiled in shell | | 8 | (22.2) | 0.4 ± 0.6 |
| | cooked in soup | | 6 | (16.7) | 0.3 ± 0.7 |
| Fishes | uncooked | 57 | 25 | (43.9) | 1.3 ± 1.5 |
| | grilling | | 16 | (28.1) | 0.8 ± 1.1 |
| | deep-frying | | 6 | (10.5) | 0.3 ± 0.7 |
| | boiling | | 10 | (17.5) | 0.5 ± 0.6 |
| Meats | uncooked | 73 | 7 | (9.6) | 0.4 ± 0.7 |
| | indirect heating | | 35 | (47.9) | 1.8 ± 1.4 |
| | deep-frying | | 8 | (11.0) | 0.4 ± 0.6 |
| | boiling | | 23 | (31.5) | 1.2 ± 1.1 |
| Pulses | uncooked | 32 | 16 | (50.0) | 0.8 ± 1.1 |
| | boiling | | 16 | (50.0) | 0.8 ± 0.9 |
| Vegetables | uncooked | 159 | 70 | (44.0) | 3.5 ± 1.8 |
| | indirect heating | | 17 | (10.7) | 0.9 ± 0.8 |
| | boiling | | 72 | (45.3) | 2.2 ± 1.8 |
| Potatoes | uncooked | 29 | 1 | (3.4) | 0.1 ± 0.2 |
| | indirect heating | | 1 | (3.4) | 0.1 ± 0.2 |
| | deep-frying | | 8 | (27.6) | 0.4 ± 0.8 |
| | steaming | | 4 | (13.8) | 0.2 ± 0.4 |
| | boiling | | 15 | (51.7) | 0.8 ± 0.9 |
| Fruits | uncooked | 63 | 63 | (100.0) | 3.2 ± 2.7 |
| Cereals ^f | bread | 173 | 56 | (32.4) | 2.8 ± 1.5 |
| | rice | | 88 | (50.9) | 4.4 ± 1.7 |
| | noodles | | 23 | (13.3) | 1.2 ± 0.7 |
| | others | | 6 | (3.5) | 0.3 ± 0.7 |

^aTotal number of dishes prepared from each food group. ^bNumber of dishes prepared by each cooking method. ^cUsage ratio of cooking methods for each food group (%) = no. of times used/total × 100. ^dRaw or uncooked foods (fresh salad, sashimi, processed foods and fermented foods). ^eSauteeing, grilling and stir-frying. ^fIndividual cereal foods are listed, because there are standard cooking methods normally used with specific cereal foods.

heating time. As for plant foods, B₆ retention in boiled broccoli was 41.7%, in spite of the short boiling time, because of its relatively large surface area.

2. Effects of cooking method and B₆ vitamin on the retention ratio of B₆

The B₆ retention ratios of the meat samples were

similar or lower when they were deep-fried than when they were sauteed or grilled (Table 3). The B₆ retention ratio was higher for meat than for fish when both were cooked by dry heating, but a precise reason for this could not be obtained. In order to better understand the causes of B₆ loss, the B₆

Table 3. Retention ratio of vitamin B₆ in different foods after cooking(%₀)

| Food item | Cooking method | | |
|--------------------|----------------------|-----------------------|------------------------|
| | Sauteeing & Grilling | Deep-frying | Boiling |
| Beef sirloin | rare | 95.2±5.6 ^a | |
| | medium | 86.3±3.6 ^a | 67.8±6.0 ^c |
| | well-done | 79.1±1.4 ^b | 43.2±3.3 ^d |
| Pork loin | | 71.2±2.5 ^a | 66.0±1.2 ^b |
| Chicken breast | | 73.3±8.8 ^a | 62.6±5.3 ^{ab} |
| Chicken thigh | | 77.2±2.5 ^a | 72.7±4.2 ^a |
| Beef round (slice) | | | short time |
| | | | long time |
| (cube) | | | |
| | | | |
| Pork ham (slice) | | | short time |
| | | | long time |
| (cube) | | | |
| | | | |
| Salmon | | | |
| Horse mackerel | | | |
| Chicken egg | | | |
| Polished rice | | | |
| Soybean | | | |
| Broccoli | | | |
| Carrot | | | |
| Potato | | | |

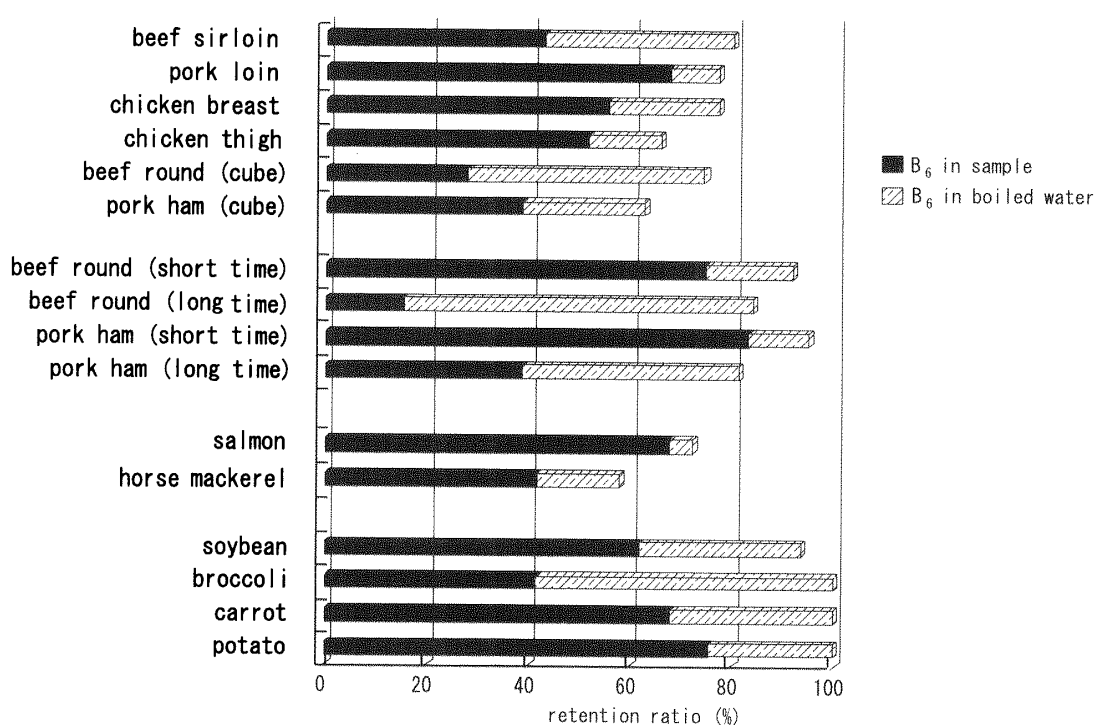
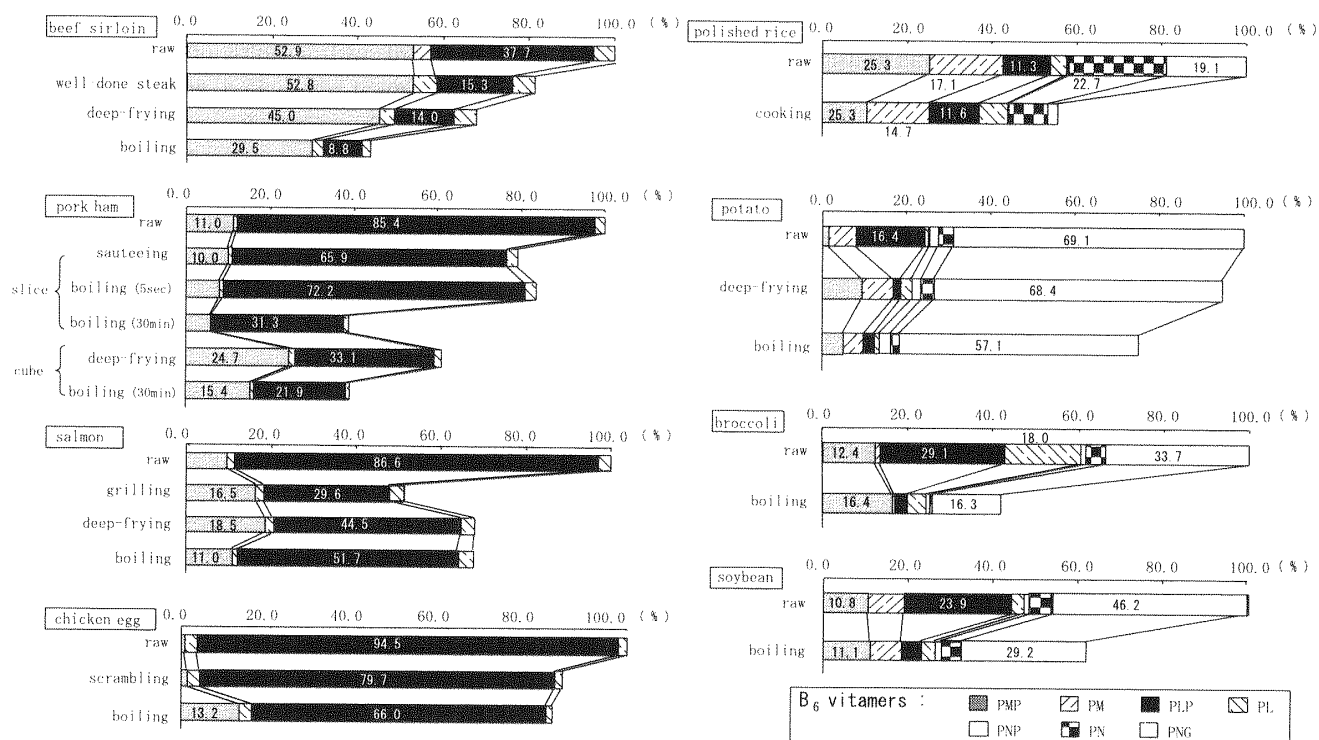
Mean value±SD ($n=3$). Retention ratio of B₆ (%₀)=(B₆ content/g in food after cooking × weight of food after cooking)/(B₆ content/g in food before cooking × weight of food before cooking) × 100. ^{a,b,c} Significant differences between the different letters (using different cooking methods for the same food).

content in water used to boil the food was analyzed. When the amount of B₆ present in the water used for boiling was added to the amount remaining in the cooked food, the B₆ retention ratio of a boiled food sample increased (Fig. 1) to almost 100% for the plant foods, so it was concluded that the loss of B₆ from these foods was not due to thermal degradation. In contrast, after adding the amount of B₆ present in the water used for boiling, the B₆ retention ratio for boiled animal foods was approximately 70%, a result which suggests that the B₆ loss with boiled animal food was due to reasons other than migration. There was a variation in the vitamer stability of B₆ among the food samples. We theorize that the composition of B₆ vitamers might have been a factor in the variation of B₆ retention that we observed among different foods. The composition ratios of B₆ vitamers in a food were altered by cooking (Fig. 2). The predominant vitamer in animal foods (with the exception of beef)

was PLP, whereas the predominant vitamers in plant foods were PN and PNG. Similar patterns of vitamer composition were seen in chicken meat and pork, horse mackerel and salmon, and carrots and potatoes (data not shown). Of all the vitamers, PLP was the one which showed the greatest decrease in composition ratio after cooking, whereas the composition ratio of PMP tended to increase with cooking. The PNG composition ratio varied among the plant foods; for example, it was 20% in rice and 50% in potatoes.

Determination of B₆ retention ratios in the reproduced meals

No significant difference between the pre-cooking B₆ content measured by HPLC and that calculated from the food tables was apparent in the reproduced meals, as shown in Table 4. The retention ratio of B₆ in a mixed diet prepared by using various methods of cooking was approximately 85.5% (compared with the measured value) and 88.8% (compared with the

Effects of Cooking Methods on the Retention of Vitamin B₆ in Foods, and the Approximate Cooking Loss in Daily MealsFig. 1. Retention ratios of B₆ in various foods cooked by wet heatingFig. 2. Changes in the total vitamin B₆ content and vitamers composition in food prepared by various cooking methods

The total B₆ content (PN equivalent) of the raw material is defined as 100%. Values of 10% or more are included. PMP, pyridoxamine 5'-phosphate; PM, pyridoxamine; PLP, pyridoxal 5'-phosphate; PL, pyridoxal; PNP, pyridoxine 5'-phosphate; PN, pyridoxine; PNG, pyridoxine 5'-β-glucoside.

Table 4. Comparison of the daily B₆ content in a mixed diet before and after cooking

| (n=5) | | |
|---------------------------------|-----------------------------------|---------------------------------------|
| Content determination procedure | B ₆ content (mg/day) | Retention ratio of B ₆ (%) |
| Measured value (HPLC) | before cooking (all ingredients) | 1.05 ± 0.37 |
| | after cooking (all cooked dishes) | 0.91 ± 0.36 |
| Calculated value | before cooking (food table) | 1.01 ± 0.33 |
| | | |

Mean ± SD.

calculated value). It is concluded that the cooking loss of B₆ was approximately 13% in daily meals.

DISCUSSION

The results of our dietary survey and measurement of the serum B₆ level indicate that the daily B₆ intake by most of our subjects (all of whom were healthy women) met or exceeded the requirement for B₆,⁴¹ and that none of the subjects suffered from a B₆ deficiency. Their B₆ intake level was a little lower than that obtained by the American NHANES II survey,²¹ in which the B₆ intake was found to be 1.14 ± 0.01 mg/day in American women. In the US study, the main foods eaten were beef steak, alcoholic beverages, fried potatoes and fortified cereals, whereas in our study, the main foods eaten were raw or grilled fish, sauteed meat, fresh or boiled vegetables and cooked rice (Table 2). The results we obtained for the frequency of consuming rice and bread over 3 days (9 meals) are similar to those reported by Hatae *et al.*²² in Japan. While many foods in the US study were prepared or cooked food by dry heating, the subjects in our study cooked food by wet heating more often than by dry heating, due to the different dietary habits. The Japanese diet has been evaluated as being well-balanced, with a proper level of cereal consumption and a good degree of diversity of food products, resulting in a balanced intake of protein, fats, and carbohydrates. In our study, the calculated PFC calorie ratios were nearly optimal:⁴² protein, 14.8%; fat, 28.3%; carbohydrate, 55.9%, although fat tended to be a little high. Protein from animal foods accounted for 50.5% of the total protein intake. These results are partly due to the level of variety in the Japanese diet. The important factors in B₆ loss were not only the food source but also the cooking method

used. Wet heating has the disadvantage of losing B₆ into the water used for boiling. Plant foods were more frequently cooked by wet heating than were animal foods, because of the cooking properties of these foods (for example, rice and soybean). Wet heating only resulted in a low B₆ retention ratio when the water used for boiling (containing lost B₆) was not consumed.

Our experimental data for B₆ retention with cooked foods are consistent with results of earlier studies involving sauteed meat,²³⁽²⁴⁾ boiled soybeans²⁵⁽²⁶⁾ and boiled vegetables.²⁷ The 50% B₆ retention ratio of cooked rice was the direct result of washing.¹⁰¹ Our results suggest that differences in the B₆ retention between animal foods and plant foods adjusted for the loss of B₆ into the water used for boiling may have been due to different vitamers compositions. B₆ was more stable in plant foods than in animal foods, as indicated by the high PN and PNG contents in plant foods and the high PLP and PL contents in animal foods. Reductive bonding of PL and PLP to the lysyl epsilon amino groups of proteins and peptides has been demonstrated to occur during thermal processing,²⁸ and the ratio of B₆ compounds bound to protein in cooked chicken breast meat has been reported to be 20%.²⁹ It has also been demonstrated that PN was more stable than PL or PM, and that PLP was less stable than PL.³⁰ B₆ retention is likely to be affected by the stability in composition of the various vitamers. Increases in the composition ratio of PMP and PM after cooking were also observed, a finding consistent with those of Ang *et al.*,³¹ and decreases in the composition ratios of PLP and PL were simultaneously observed. This phenomenon has also been reported by Gregory *et al.*,³² who found that the extent of dephosphorylation was less than that of transamina-

Effects of Cooking Methods on the Retention of Vitamin B₆ in Foods, and the Approximate Cooking Loss in Daily Meals

tion. Changes in the ratio of PNG due to heating were not clearly observed in our previous study.¹⁰⁾ The relatively low bioavailability of PNG is not considered an important factor in daily meals, because it has been observed (in our previous study,¹⁰⁾ and by Andon *et al.*³²⁾ and Nakano *et al.*³³⁾) that serum PLP was not affected by the PNG intake, even when PNG accounted for approximately 15–16% of total B₆ in daily meals.

The effect of salt (a frequently used seasoning) on B₆ retention was also studied. Although salt has a coagulating effect on protein, it was not observed to affect B₆ retention when it was used in cooking at a concentration of 1% (data not shown). This observation is consistent with that of Selman.²⁷⁾

The ratio of the average B₆ intake obtained from the duplicate meals (measured by HPLC) to that calculated from the dietary records was 86.7%. Most B₆ values in the Japanese standard food tables¹¹⁾ are calculated for raw foods. The difference between measured and calculated values, the apparent cooking loss (approximately 13%), was obtained in the first step of this investigation. This value for cooking loss is in reasonably close agreement with the B₆ retention ratios arrived at experimentally from the reproduced duplicate meals. The experimental results confirm that the cooking loss of B₆ was approximately 13% in home-cooked daily meals which included 40% of non-heated foods such as fresh and processed types.¹⁰⁾

It is difficult to calculate B₆ in a meal when not consulting the data for some cooked foods or cooking methods in the food composition table,¹¹⁾ because analysis data for cooked foods and cooking methods in this table are limited. When the nutrient values for raw foods in the table¹¹⁾ are used for the calculation, the loss during cooking is not considered. We propose the application of the cooking loss to the calculation of B₆ in meals as a simple and convenient method to estimate the actual B₆ intake.

Some measures which can be used to lessen the cooking loss of B₆ are (1) eating boiled foods with soup made from the water used for boiling; (2) avoiding a long heating time, particularly with animal foods; (3) using preparation methods which result in less B₆ loss (eating raw foods, and using dry heating with a short heating time).

CONCLUSION

The approximate cooking loss of B₆ in daily meals consisting of various dishes was found to be 13%.

The experimental examination shows the cooking loss to be the greatest from boiling (especially boiling meat for a long time), this being followed by deep-frying and sauteeing. The use of dry heating for the preparation of most of the animal foods in this study resulted in a relatively high B₆ retention ratio. Wet heating was found to result in a high retention ratio when the water used for boiling was consumed together with the boiled food.

REFERENCES

- 1) Kimura, M., Itokawa, Y., and Fujiwara, M.: Cooking Losses of Thiamin in Food and Its Nutritional Significance, *J. Nutr. Sci. Vitaminol.*, **36**, S17–S24 (1990)
- 2) Murata, K.: Cooking and Processing of Vitamins (in Japanese), *Shin Vitamingaku*, The Vitamin Society of Japan, Kyoto, 548–553 (1969)
- 3) Ishibashi, G., Mutou, K., Yamanouchi, Y., Takizawa, K., and Ishibashi, S.: Effect of Large- and Small-Scale Cooking on Losses of Nutrients (in Japanese), *Rinshou Eiyou (Jpn. J. Clin. Nutr.)*, **70**, 509–513 (1987)
- 4) Lena, I., and Bergstrom, M.: Different Techniques of Food Preparation and Cooking: Implication for Dietary Surveys, *Proc. Nutr. Soc.*, **55**, 671–678 (1996)
- 5) Power, P.M., and Hoover, L.W.: Calculating the Nutrient Composition of Recipes with Computers, *J. Am. Diet. Assoc.*, **89**, 224–231 (1989)
- 6) Society for the Information of Health and Nutrients: *Recommended Dietary Allowances for the Japanese, 6th revision, Dietary Reference Intakes*, Dai-ichi Shuppan, Tokyo, 101–103 (1999)
- 7) Leklem, J.E.: Vitamin B₆ Bioavailability and Its Application to Human Nutrition, *Food Technol.*, **October**, 194–196 (1988)
- 8) Gregory III, J.F.: Bioavailability of Vitamin B₆ from Plant Foods, *Am. J. Clin. Nutr.*, **46**, 717–719 (1989)
- 9) Shibata, K., Hiraoka, M., and Yasuda, K.: Study on Dietary Vitamin B₆ Intake in Female Students by Duplicate Meal Method, *Vitamins (Japan)*, **73**, 459–464 (1999)
- 10) Shibata, K., Yasuhara, Y., and Yasuda, K.: Study on Dietary Vitamin B₆ Intake in Female Students by Duplicate Meal Method (Part II), *Vitamins (Japan)*, **74**, 423–433 (2000)
- 11) Resources Council, Science and Technology Agency, Japan: *Standard Tables of Food Composition in Japan, Fifth Revised Edition* (2000)
- 12) Toukairin-Oda, T., Sakamoto, E., Hirose, N., Mori, M., and Tsuge, H.: Determination of Vitamin B₆ Derivatives in Foods and Biological Materials by Reversed-Phase HPLC, *J. Nutr. Sci. Vitaminol.*, **35**, 171–180 (1989)
- 13) Edwards, P., Liu, P.K.S., and Rose, G.A.: A Simple

- Liquid-Chromatographic Method for Measuring Vitamin B₆ Compounds in Plasma, *Clin. Chem.*, **35**, 241-245 (1989)
- 14) Sierra, L., and Vidal-Valverde, C.: A Simple Method to Determine Free and Glycosylated Vitamin B₆ in Legumes, *J. Liq. Chrom. & Rel. Technol.*, **20**, 957-969 (1997)
 - 15) Gregory III, J.F., and Sartain, D.B.: Improved Chromatographic Determination of Free and Glycosylated Forms of Vitamin B₆ in Foods, *J. Agric. Food Chem.*, **39**, 899-905 (1991)
 - 16) Cross, H.R., Bernholdt, H.F., Dikeman, M.E., Greene, B.E., Moody, W.G., Staggs, R., and West, R.L.: *Guidelines for Cookery and Sensory Evaluation of Meat*, American Meat Science Association in Cooperation with The National Live Stock & Meat Board, Illinois, 1-24 (1978)
 - 17) Kainuma, Y., Nagao, K., Hatae, K., and Shimada, A.: Effect of the Washing Methods of Rice on Sensory Palatability of the Cooked Rice (in Japanese), *Jpn. Sci. Cookery*, **23**, 419-423 (1990)
 - 18) Murphy, E.W., Criner, P.E., and Gray, B.C.: Comparisons of Methods for Calculating Retentions of Nutrients in Cooked Foods, *J. Agric. Food Chem.*, **23**, 1153-1157 (1975)
 - 19) Kim, W.W., Mertz, W., Judd, J.T., Marshall, M.W., Kelsay, J.L., and Parther, E.S.: Effect of Making Duplicate Food Collections on Nutrient Intakes Calculated from Diet Records, *Am. J. Clin. Nutr.*, **40**, 1333-1337 (1984)
 - 20) Miller, L.T., and Linkswiler, H.: Effect of Intake on the Development of Abnormal Tryptophan Metabolism by Men during Vitamin B₆ Depletion, *J. Nutr.*, **93**, 53-59 (1967)
 - 21) Kant, A.K., and Block, G.: Dietary Vitamin B₆ Intake and Food Sources in the US Population: NHANES II, 1976-1980, *Am. J. Clin. Nutr.*, **52**, 707-716 (1990)
 - 22) Hatae, K., Ikeda, N., Ayabe, S., Nagao, K., and Shimada, A.: Survey of Japanese Dietary Life by Questionnaire (Part 2), *Nihon Chourikagaku Gakkaishi (J. Cookery Sci. Jpn.)*, **24**, 317-321 (1991)
 - 23) Yang, J., Sulaeman, A., Setiawan, B., Atughonu, A., Giraud, D., Hamouz, F.L., and Driskell, J.A.: Sensory and Nutritive Qualities of Pork Strips Prepared by Three Household Cooking Techniques, *J. Food Quality*, **17**, 33-40 (1994)
 - 24) Batenhorst, J.H., Sun, J., Giraud, D.W., Young, L.Y., Hamouz, F.H., and Driskell, J.A.: Retention of Nutrients in Pork Roasts Prepared by Variation in Cookery Methods, *J. Muscle Food*, **6**, 359-368 (1995)
 - 25) Soetrisno, U., Holmes, Z.A., and Miller, L.T.: Effect of Heating Time of Soybean on Vitamin B₆ and Folic Acid Retention, Trypsin Inhibitor Activity, and Microstructure Changes, *J. Food Sci.*, **47**, 530-537 (1982)
 - 26) Augustin, J., Beck, C.B., Kalbfleish, G., and Kagel, L.C.: Variation in the Vitamin and Mineral Content of Raw and Cooking Commercial Phaseolus Vulgaris Classes, *Food Technol.*, **March**, 75-76 (1981)
 - 27) Selman, J.D.: Vitamin Retention during Blanching of Vegetable, *Food Chem.*, **49**, 137-147 (1994)
 - 28) Gregory, J.F., and Kirk, J.R.: Interaction of Pyridoxal and Pyridoxal Phosphate with Peptides in a Model Food System during Thermal Processing, *J. Food Sci.*, **42**, 1554-1557 (1977)
 - 29) Gregory III, J.F., Ink, S.L., and Sartain, D.B.: Degradation and Binding to Food Proteins of Vitamin B₆ Compounds during Thermal Processing, *J. Food Sci.*, **51**, 1345-1351 (1986)
 - 30) Gregory III, J.F., and Hiner, M.E.: Thermal Stability of Vitamin B₆ Compounds in Liquid Model Food Systems, *J. Food Sci.*, **48**, 1323-1327 (1983)
 - 31) Ang, C.Y.W., Cenciarelli, M., and Eitenmiller, R.R.: A Simple Liquid Chromatographic Method for Determination of B₆ Vitamers in Raw and Cooked Chicken, *J. Food Sci.*, **53**, 371-375 (1988)
 - 32) Andon, M.B., Reynolds, R.D., Moser-Veillon, P.B., and Howard, M.P.: Dietary Intake of Total and Glycosylated Vitamin B₆ and the Vitamin B₆ Nutritional Status of Unsupplemented Lactating Women and Their Infants, *Am. J. Clin. Nutr.*, **50**, 1050-1058 (1989)
 - 33) Nakano, H., McMahon, L.G., and Gregory III, J.F.: Pyridoxine-5'- β -D-Glucoside Exhibits Incomplete Bioavailability as a Source of Vitamin B₆ and Partially Inhibits the Utilization of Co-Ingested Pyridoxine in Humans, *J. Nutr.*, **127**, 1508-1513 (1997)

調理法による食品中のビタミン B₆ 保持と 日常の食事における調理損失

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実際のビタミン B₆ (B₆) 摂取量を考慮して, 日常の食事の中の B₆ におけるおおよその調理損失の見積もりに適用するデータが得られた. 我々の 19 名の健康な女性の栄養調査によるデータより, HPLC により測定した調理後の食事の中の B₆ 含量は食品成分表を用いて計算した調理前の B₆ 含量のおよそ 13% であった. 更に実際の B₆ 損失を確認するため, 食事記録による食事を再現調理し, 調理前後の食物中の B₆ の測定より得られた結果もまたおよそ 13% であることを示した. 日常の食事において, 動物性食品のほとんどが B₆ 損失の少ない乾式加熱により調理されており, 一方, 植物性食品の多くが湿式加熱であった. 調理損失は「煮る」で最大となり, 続いて「揚げる」, 「焼く」の順であった. しかし, 食材と共にゆで汁に移行した B₆ を摂取すれば, 「煮る」は比較的 B₆ 損失が少ないことが認められた.

キーワード: ビタミン B₆, 保持率, 食品摂取状態, 調理損失, 乾式加熱, 湿式加熱.