JOURNAL OF FOOD COMPOSITION AND ANALYSIS

ELSEVIER

Journal of Food Composition and Analysis 21 (2008) 199–210

www.elsevier.com/locate/jfca

Original Article

Food content of ubiquinol-10 and ubiquinone-10 in the Japanese diet

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Received 18 April 2007; received in revised form 17 October 2007; accepted 28 October 2007

Abstract

Seventy food items (8 types of meat, 16 types of fish and shellfish, 21 vegetables, 7 fruits, 6 pulses, 3 potatoes, 3 dairy products and 6 others) were analyzed using a simple and reliable method that can detect the reduced form of coenzyme Q_{10} (ubiquinol-10) and the oxidized form of coenzyme Q_{10} (ubiquinone-10) simultaneously. This method employed direct 2-propanol extraction and high performance liquid chromatography (HPLC) equipped with a reduction column and an electrochemical detector (ECD). Ubiquinol-10 was found in 63 out of 70 food items, while ubiquinone-10 was found in 66 of the 70 food items. In the food items in which ubiquinol-10 was found, its content ranged from 2.63 to 84.8 μg/g in meat, 0.38 to 23.8 μg/g in fish and shellfish, 0.17 to 5.91 μg/g in vegetables, 0.22 to 3.14 μg/g in fruits, 0.68 to 1.82 μg/g in potatoes, 0.72 to 4.3 μg/g in pulses and 0.18 to 33.3 μg/g in other food items including seeds, eggs, dairy products, soybean oil and miso (fermented soybean paste). Pork (shoulder), bovine liver, chicken heart, horse mackerel, young yellowtail and soybean oil showed a high ubiquinol-10 content of more than 20 μg/g. On the other hand, total coenzyme Q_{10} content ranged from 13.8 to 192 μg/g in meat, 1.25 to 130 μg/g in fish and shellfish, 0.08 to 7.47 μg/g in vegetables, 0.51 to 9.48 μg/g in fruits, 1.05 to 3.01 μg/g in potatoes, 2.31 to 6.82 μg/g in pulses and 0.26 to 53.8 μg/g in other food items. The estimated average daily intakes of ubiquinol-10 and total coenzyme Q_{10} calculated from our results and data on Japanese daily food consumption were 2.07 and 4.48 mg, respectively. Thus, intake of ubiquinol-10 accounted for 46% of the total coenzyme Q_{10} intake.

Keywords: Coenzyme Q_{10}; Ubiquinol-10; Ubiquinone-10; Food; Dietary intake; HPLC; Japan; Japanese daily food consumption

1. Introduction

Coenzyme Q_{10} (CoQ_{10}) is a fat-soluble, vitamin-like substance present in nearly all human tissues. Its primary biochemical function is as a mobile electron carrier in the mitochondrial electron-transfer process of cellular respiration and energy production (Ernster and Dallner, 1995; Turunen et al., 2004).

Although CoQ_{10} is a redox molecule, which exists in both a biochemically reduced form (ubiquinol-10) and an oxidized form (ubiquinone-10) in biological tissues (Crane, 2001), ubiquinol-10 is the more common form of CoQ_{10} in vivo and accounts for more than 80% of the total CoQ_{10} (ubiquinol-10 + ubiquinone-10) pool in human plasma, intestine and liver (Okamoto et al., 1989; Frei et al., 1990; Åberg et al., 1992). It seems reasonable that a major portion of CoQ_{10} exists as ubiquinol-10 in vivo because ubiquinol-10 plays an important role as an antioxidant in both mitochondria and lipid membranes either by scavenging free radicals directly or in conjunction with α-tocopherol (Quinn et al., 1999; Lass and Sohal, 2000; Forsmark-Andree et al., 1997; Noack et al., 1994).

Because of its important biological roles, ubiquinone-10 has been widely used as a dietary supplement by health-conscious individuals and those who have ailments including various cardiac disorders (Overvad et al., 1999; Greenberg and Frishman, 1990; Hendler and Rorvik, 2001; Tran et al., 2001; Jones et al., 2002).

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doi:10.1016/j.jfca.2007.10.003

Some studies have assessed the ubiquinone content of various food items. It has been reported that most foods contained ubiquinone (Weber et al., 1997; Kamei et al., 1986; Mattila and Kumpulainen, 2001) and the daily intake of CoQ_{10} was estimated to be 3–5 mg (Weber et al., 1997) or 3.8–5.4 mg (Mattila and Kumpulainen, 2001). However, there have been few studies of the ubiquinol-10 content of foods. In these studies, ubiquinol-10 was reported at a level of 11–189 μg/mL in extra virgin olive and in seed oils (peanut, soybean, corn and sunflower; Cabrini et al., 2001), at 0.18–8.37 μg/g in the flesh of 21 species of fish and shellfish caught in the Mediterranean Sea (Passi et al., 2002), and at 0.2–18.9 μg/g in some meats (beef, pork and chicken), some fish (salmon, eel and squid) and some vegetables (Chinese cabbage, eggplant and parsley) (Kettawan et al., 2007).

While ubiquinol-10 is major form of CoQ_{10} in the human and animal body, there is little information on the ubiquinol-10 content of foods and on the dietary intake of ubiquinol-10. One reason may be that ubiquinol-10 has been difficult to measure quantitatively in previous studies because it is rapidly oxidized to ubiquinone-10 during extraction. However, because of its biological importance the content of ubiquinol-10 in foods is of nutritional significance. In this study, we determined the ubiquinol-10 and ubiquinone-10 content of various foods using a simple and reliable high performance liquid chromatography (HPLC) method with an electrochemical detector (ECD) and using data on the consumption of foods we then estimated the average dietary intake of ubiquinol-10 and of total CoQ_{10} in the Japanese population.

2. Materials and methods

2.1. Food samples, standards and reagents

Food samples used for analysis were selected considering the ordinary intake of the Japanese population. They were obtained from nearby supermarkets, fish shops, greengrocers and butchers, and prepared for analysis on the day of purchase or on the following day. They were preserved under the same conditions as in the stores (refrigerated or at room temperature) until preparation. Samples were purchased as about 300 g of meat, as half a fish, a whole fish, a number of shellfish or as 150–300 g fillets for the fish and shellfish samples, as half a vegetable, one whole vegetable or as a bunch for vegetable samples, as one whole piece of fruit or as a number of servings of fruit, as one whole potato and as one small package for beans, dairy products and processed food samples (details are noted in Tables 3–6). Ubiquinol-10 and ubiquinone-10 were prepared in our Finechemical Research Laboratories with purities greater than 99% by HPLC analysis. Methanol, ethanol, 2-propanol and hexane were of HPLC grade from Wako Pure Chemical (Osaka, Japan) or Nacalai Tesque (Kyoto, Japan).

2.2. HPLC analysis

The ubiquinol-10 and ubiquinone-10 content of foods were determined by a minor modification of the HPLC method with electrochemical detection published by Yamashita and Yamamoto (1997). The HPLC system comprised an auto injector (SIL-10AXL, Shimadzu, Kyoto, Japan) with a sample cooler set at 4 °C, a pump (LC-10AD, Shimadzu), a degasser (DGU-20A, Shimadzu), a column oven (CTO-6A, Shimadzu) set at 40 °C, an analytical column (YMC-Pack ODS-A303, 5 μm, 250 × 4.6 mm i.d., YMC, Kyoto, Japan), a reduction column (RC-10, 15 × 4.0 mm i.d., Shiseido, Tokyo, Japan), and an amperometric electrochemical detector (SI-2, Shiseido). The mobile phase consisted of 0.05 M sodium perchlorate in methanol/hexane (88:12, v/v) at a flow rate of 1.0 mL/min. The oxidation potential of the ECD was 600 mV (vs. Ag/AgCl). Ubiquinol-10 and ubiquinone-10 were quantified by an external standard method based on peak area. Ubiquinol-10 and ubiquinone-10 content of food samples were measured in triplicate.

2.3. Preparation of standard solutions

Stock standard solutions (50 μg/mL) of ubiquinol-10 and ubiquinone-10 were prepared in ethanol and stored at −80 °C. Each working day, they were diluted stepwise with 2-propanol to prepare working standard solutions.

2.4. Preparation of food samples

To obtain representative data for each food sample, three subsamples (approximate 1 g aliquots or 1 basic unit) were taken from three different parts of the original food sample or three different units (single servings), and these were minced individually (details are noted in Tables 3–6). To each minced 50 mg food sample, 1.95 mL of 2-propanol was added and the mixture was homogenized with a Polytron homogenizer. The homogenate was diluted 2.5- or 5-fold with 2-propanol and was centrifuged at 16,000 × g for 3 min at 4 °C. The supernatant was separated and 50 μL of the extract was injected into the HPLC apparatus.

2.5. Validation of the method

To assess linearity, calibration curves were constructed in triplicate for each analyte. The calibration curves were obtained by weighted (1/concentration) least-squares linear regression analysis.

Recovery was assessed by spiking two known concentrations (high and low) of each analyte or solvent (as blank) to homogenates from three representative matrices of beef (thigh), five-ray yellowtail and white potato in triplicate. To calculate recovery, the areas found in the blank matrices were subtracted from those of the spiked matrices, and then the resulting areas were compared with those found by direct injection of standard solutions at the same concentration. Within-day and between-day accuracy and precision were evaluated by analyzing the spiked samples that were prepared separately using the same procedure adopted for recovery assessment in triplicate, based on the daily calibration curves. Within-day accuracy and precision were assessed on 1 day while between-day accuracy and precision were assessed on three separate days.

2.6. Calculation of average intake per day

Estimates of the average intake of ubiquinol-10, ubiquinone-10 and total CoQ_{10} per day were based on the average contents for individual food groups (e.g., beef, chicken, citrus, etc.) calculated from the data obtained in this study and using data on the consumption of individual food groups taken from the 2003 National Nutrition Survey conducted by the Japan Ministry of Health, Labour and Welfare (2005).

3. Results and discussion

In order to establish a simple and sensitive analytical method for the simultaneous detection of ubiquinol-10 and ubiquinone-10 in foods, we employed direct 2-propanol extraction and quantification using HPLC equipped with a reduction column and an ECD as key features of the measurement technique.

Sample preparation for the determination of ubiquinone-10 or ubiquinone-9 in foods in previous studies involved homogenization in an aqueous solvent, extraction with a non-aqueous solvent, washing and evaporation of the non-aqueous solvent and dissolution in an aqueous solvent for HPLC injection (Kamei et al., 1986; Weber et al., 1997; Mattila and Kumpulainen, 2001; Mattila et al., 2000). Since ubiquinols are readily oxidized into ubiquinones by oxygen in the air, this procedure is unsatisfactory for the quantitation of ubiquinols. To protect ubiquinol-10 from oxidation during sample preparation we used a simple and fast isolation procedure which consisting of homogenization in 2-propanol and centrifugation to separate the supernatant for HPLC injection with no concentration or solvent substitution steps (Galinier et al., 2004).

To analyze ubiquinols and ubiquinones simultaneously in biological materials with HPLC, various procedures which take advantage of ECD's high sensitivity have been used including the combination of ultra-violet detection and ECD (Takada et al., 1984; Lang and Packer, 1987), the combination of a reduction column and amperometric ECD (Wakabayashi et al., 1994; Yamashita and Yamamoto, 1997), and the use of coulometric electrochemical cells (Edlund, 1988; Tang et al., 2001). We employed a combination of a reduction column and amperometric ECD due to its easy maintenance. This technique counteracts the drawback of the direct 2-propanol extraction method we used for sample preparation that cannot be used to concentrate analytes of ubiquinol-10 and ubiquinone-10, which exist at low concentrations in foods.

Representative chromatograms of standards for ubiquinol-10 and ubiquinone-10, blank and spiked food samples (beef and potato) are shown in Fig. 1. Peaks of ubiquinol-10 and ubiquinone-10 were clearly separated and there was no interference at the retention time of the analyte. The retention times for ubiquinol-10 and ubiquinone-10 were 17.8 and 27.7 min with confidence intervals of 17.7–17.9 min, and 27.6–27.9 min, respectively, which were obtained from 27 analyses of the standard conducted once a day on separate days.

Calibration curves were constructed with five calibration solutions for both analytes. The ECD response was linear over the range 0.040–50 ng/injection corresponding to 0.08–100 μg/g or 0.16–200 μg/g of the analytes in foods. The calibration curves were obtained by weighted (1/concentration) least-squares linear regression analysis. The mean equations and correlation coefficients of triplicate determinations were $Y = 71054x - 852$ and 0.999 for ubiquinol-10, and $Y = 70703x - 592$ and 0.999 for ubiquinone-10, respectively, where Y is peak area and x is amount of injected analyte (ng). Coefficients of variation (CVs) of linearity (correlation coefficients) and slopes of the calibration curves were 0.003% and 1.54% for ubiquinol-10, and 0.001% and 0.96% for ubiquinone-10, respectively. Confidence intervals (95%) for the intercept of the calibration curves were −7726 to +4170 for ubiquinol-10 and −6282 to +4626 for ubiquinone-10, each containing the zero point. The detection limits of ubiquinol-10 and ubiquinone-10 were 38 pg/injection each, corresponding to 0.07 or 0.15 μg/g of the analytes in foods (signal-to-noise ratio = 3).

Analytical recoveries were evaluated for the analytes by spiking known concentrations to three representative matrices of white potato, five-ray yellowtail and beef (thigh). As shown in Table 1, recovery and the CVs of ubiquinol-10 ranged from 87% to 112% and 2.2% to 21%, respectively, and those of ubiquinone-10 ranged from 97% to 106% and 0.9% to 4.9%, respectively. The high recovery values for the analytes (more than 87%) and the finding that there was no difference in the values between the analytes indicated that there was little chance that ubiquinol-10 was oxidized and ubiquinone-10 reduced during the extraction process. Souchet and Laplante (2007) developed a simple and efficient extraction procedure for HPLC determination of CoQ_{10} content in pelagic fish tissues including mackerel by using homogenization with 0.1 M sodium dodecyl sulfate and 0.15 M NaCl, subsequent extraction with ethanol-hexane, hexane layer evaporation, and reconstitution in isopropanol. In contrast to our results, however, they reported that no ubiquinol-10 peak was detected at 290 nm, which is the maximum absorbance of the molecule, indicating that ubiquinol-10 was completely converted into ubiquinone-10 during the sample preparation procedures.

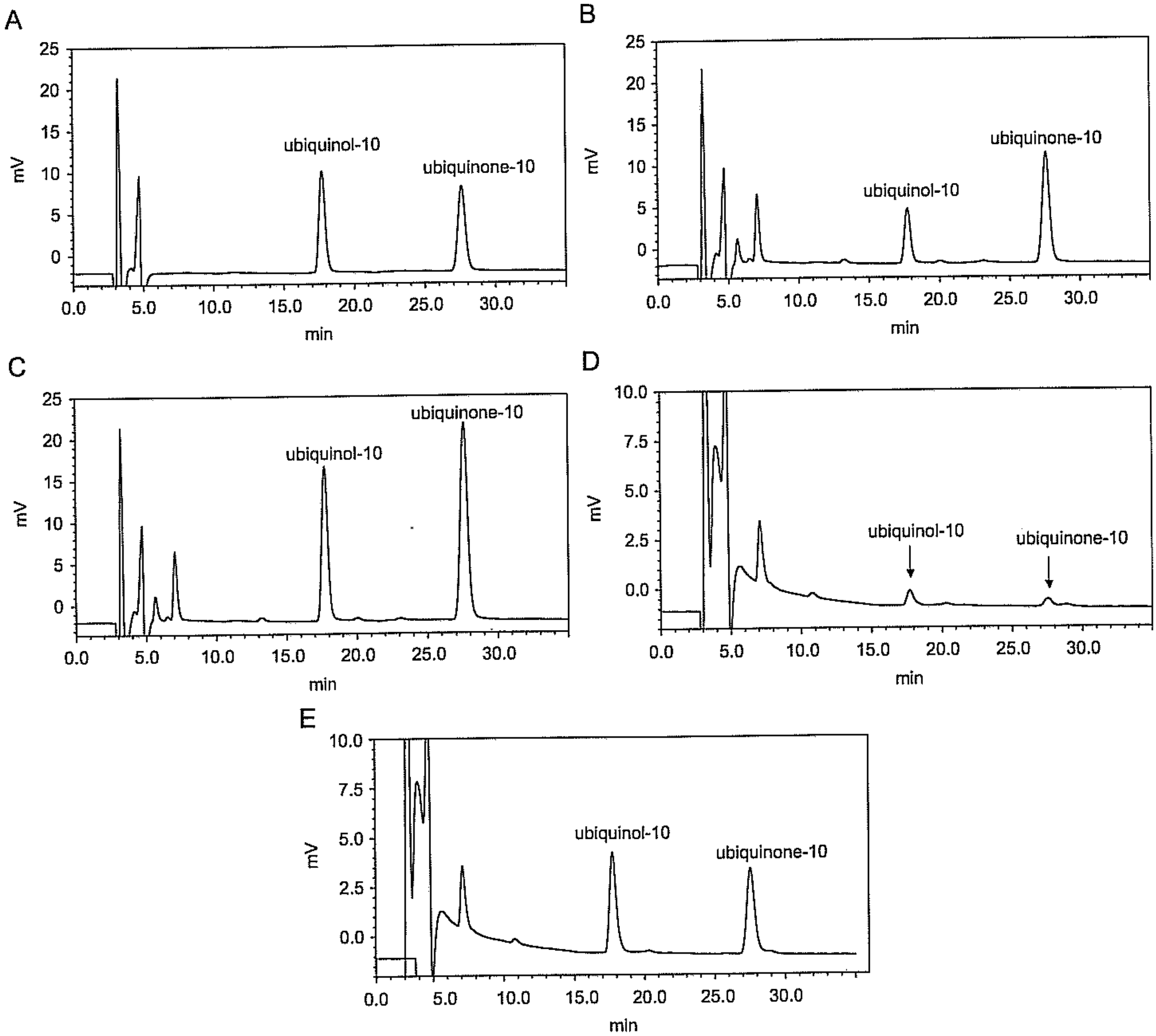

Fig. 1. HPLC chromatograms of (A) a standard mixture of ubiquinol-10 and ubiquinone-10 at 0.10 µg/mL each, (B) beef shoulder extract with concentrations of ubiquinol-10 and ubiquinone-10 at 12.2 and 29.7 µg/g, respectively, (C) beef shoulder extract spiked with ubiquinol-10 and ubiquinone-10 at 20 µg/g each, (D) white potato extract with concentrations of ubiquinol-10 and ubiquinone-10 at 0.73 and 0.43 µg/g, respectively, and (E) white potato extract spiked with ubiquinol-10 and ubiquinone-10 at 4.0 µg/g each.

Within-day and between-day accuracy and precision (CV%) were evaluated by analyzing three representative matrices spiked at two known concentrations. The results are shown in Table 2. Within-day accuracy and precision were assessed on 1 day while between-day accuracy and precision were assessed on three separate days. Within-day accuracy and precision were 101–109% and 0.3–9.9% for ubiquinol-10, and 106–115% and 0.6–10.7% for ubuquinone-10, respectively. Between-day accuracy and precision were 101–110% and 2.4–15.4% for ubiquinol-10, and 106–114% and 3.0–13.8% for ubiquinone-10, respectively.

The results obtained for analytical recovery, within-day and between-day accuracy and precision for both ubiquinol-10 and ubiquinone-10 confirmed the reliability of the method.

In addition, we confirmed the presence of ubiquinol-10 in each food sample by treating the homogenate with *p*-benzoquinone, an oxidizing agent, before 2-propanol extraction during the actual analysis of food samples. In all food samples, the ubiquinol-10 peak disappeared and all the CoQ_{10} was recovered with a retention time corresponding to that of ubiquinone-10. Representative chromatograms of food samples with or without treatment with *p*-benzoquinone are shown in Fig. 2.

Table 1
Recovery of ubiquinol-10 and ubiquinone-10 from representative foods

Food	Ubiquinol-10			Ubiquinone-10		
	Spiked concentration (μg/g)	Recovery (%)		Spiked concentration (μg/g)	Recovery (%)	
		Mean±S.D.[a]	CV (%)		Mean±S.D.[a]	CV (%)
White potato	0.44	112±3.2	2.9	0.22	101±1.1	1.1
	4.0	104±2.3	2.2	1.96	98.2±1.8	1.9
Five-ray yellowtail	2.0	105±3.1	3.0	2.96	106±5.2	4.9
	18.0	98.8±2.8	2.9	26.8	97.5±0.9	0.9
Beef (thigh)	2.0	94.7±19.6	20.7	8.8	97.4±4.1	4.2
	18.0	87.8±3.4	3.8	80.0	101±1.0	1.0

[a]Result obtained from triplicate analyses.

Table 2
Within-day and between-day variations in analysis of ubiquinol-10 and ubiquinone-10 in representative foods

Food	Spiked substance	Spiked concentration (μg/g)	Within-day		Between-day	
			Accuracy (%)[a]	CV (%)	Accuracy (%)[a]	CV (%)
White potato	Ubiquinol-10	0.44	107±3.3	3.1	110±8.7	7.9
		4.0	109±0.7	0.6	107±2.6	2.4
	Ubiquinone-10	0.22	111±10.4	9.4	114±15.7	13.8
		1.96	115±2.9	2.5	111±3.3	3.0
Five-ray yellowtail	Ubiquinol-10	2.0	101±10.0	9.9	105±16.2	15.4
		18.0	105±4.5	4.3	102±3.5	3.4
	Ubiquinone-10	2.96	106±11.3	10.7	110±12.0	10.8
		26.8	107±3.6	3.4	106±3.5	3.3
Beef (thigh)	Ubiquinol-10	2.0	107±9.3	8.7	101±5.4	5.4
		18.0	107±0.3	0.3	101±5.2	5.2
	Ubiquinone-10	8.8	110±7.6	7.0	110±3.9	3.5
		80.0	109±0.7	0.6	106±4.4	4.2

[a]Mean±S.D. of triplicate analyses.

The content of ubiquinol-10, ubiquinone-10 and total CoQ_{10} (ubiquinol-10+ubiquinone-10) in various food items are shown in Tables 3–6. Ubiquinol-10 was found in 63 out of 70 food items, while ubiquinone-10 was found in 66 out of 70 food items. In the food items in which ubiquinol-10 was found, its content ranged from 2.63 to 84.8 μg/g in meat, 0.38 to 23.8 μg/g in fish and shellfish, 0.17 to 5.91 μg/g in vegetables, 0.22 to 3.14 μg/g in fruits, 0.68 to 1.82 μg/g in potatoes, 0.72 to 4.3 μg/g in pulses and 0.18 to 33.3 μg/g in other food items including seeds, eggs, dairy products, soybean oil and miso (fermented soybean paste). Moderately high ubiquinol-10 content was found in pork (shoulder), bovine liver, chicken heart, horse mackerel, young yellowtail and soybean oil, at more than 20 μg/g. Furthermore, in 51 out of 63 food items, ubiquinol-10 accounted for 31.0–98.1% of the total CoQ_{10}. A high ubiquinol-10 ratio ranging from 70.2% to 98.1% was observed in 14 food items, i.e., chicken (breast), bovine liver, canned tuna, garlic, lotus root, cabbage, Japanese radish, parsley, orange, grapefruit, persimmon, roasted sesame seed, cow milk and miso. Canned tuna showed a high ubiquinol-10 ratio at 98.1% and a relatively high ubiquinol-10 content of 14.6 μg/g, whereas the values for raw tuna were 10.5% and 0.51 μg/g, respectively. To confirm these results, two other different brands of canned tuna were also analyzed. The data showed similar ubiquinol-10 content (17.2 and 21.8 μg/g) and ubiquinol-10 ratios (94.5% and 95.5%, respectively); therefore, it is likely that canned tuna has a relatively high ubiquinol-10 content and a high ubiquinol-10 ratio. Vegetable oil used for canning tuna may at least partly explain these difference, as vegetable oils made from seeds such as soybean, corn and sunflower have been shown to have an ubiquinol-10 ratio of 73.5–90.3% and an ubiquinol-10 content of 13–189 μg/mL (Cabrini et al., 2001).

On the other hand, ubiquinone-10 content ranged from 3.24 to 107 μg/g in meat, 0.29 to 106 μg/g in fish and shellfish, 0.08 to 3.17 μg/g in vegetables, 0.14 to 6.34 μg/g in fruits, 0.38 to 1.19 μg/g in potatoes, 1.19 to 2.52 μg/g in pulses and 0.07 to 20.6 μg/g in other food items. Therefore,

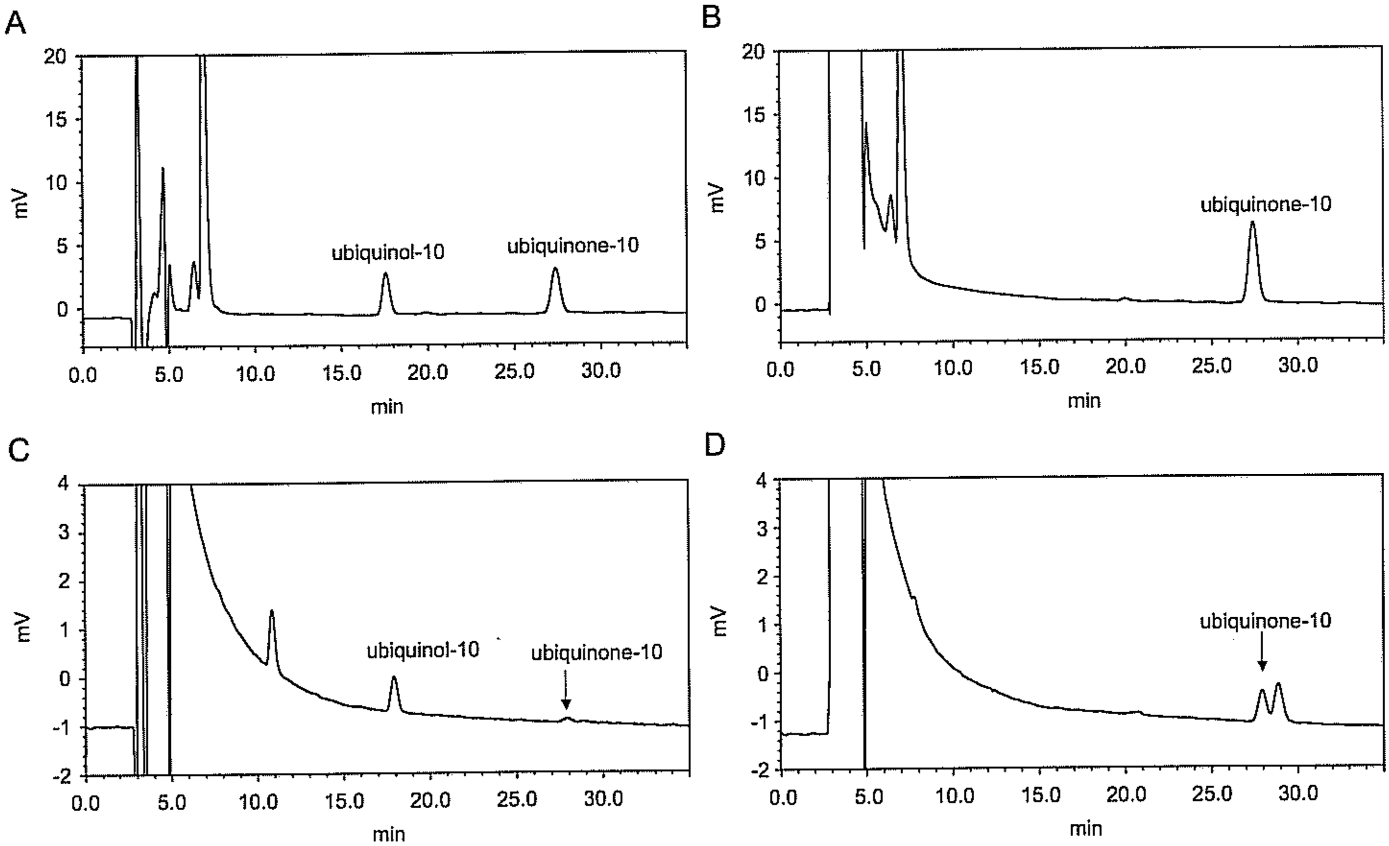

Fig. 2. HPLC chromatograms of (A) five-ray yellowtail extract, (B) five-ray yellowtail extract treated with *p*-benzoquinone, (C) Japanese radish extract and (D) Japanese radish extract treated with *p*-benzoquinone.

Table 3
The content of ubiquinol-10, ubiquinone-10 and total CoQ_{10} in meat

Food	Scientific name	Content (μg/g wet weight)			% Ubiquinol-10[c]	Sampling	Preparation
		Ubiquinol-10[a]	Ubiquinone-10[a]	Total CoQ_{10}[a,b]			
Beef (thigh)	*Bos taurus*	5.36±0.59	25.0±3.4	30.3±3.9	17.6	300 g serving[d,e]	–
Beef (shoulder)		11.1±0.9	29.0±0.7	40.1±1.5	27.7	300 g serving[d,e]	–
Pork (thigh)	*Sus scrofa domesticus*	2.63±0.48	11.2±2.0	13.8±2.4	19.0	300 g serving[d,e]	–
Pork (shoulder)		25.4±1.5	19.6±1.6	45.0±3.0	56.4	300 g serving[d,e]	–
Chicken (breast)	*Gallus gallus domesticus*	13.8±0.5	3.24±0.41	17.1±0.1	81.0	300 g serving[d,e]	–
Chicken (thigh)		12.6±3.6	12.4±3.5	25.0±6.7	50.4	400 g serving[d,e]	Skinned
Bovine liver		40.1±4.5	10.4±2.1	50.5±3.5	79.3	300 g serving[d,e]	–
Chicken heart		84.8±5.0	107±8.2	192±6.6	44.2	One whole heart[d,e]	–

[a]Mean±S.D. of triplicate analyses.
[b]Total CoQ_{10} = ubiquinol-10 + ubiquinone-10.
[c]% Ubiquinol-10 = (ubiquinol-10/total CoQ_{10}) × 100.
[d]Sold refrigerated.
[e]Domestic product.

the total CoQ_{10} content ranged from 13.8 to 192 μg/g in meat, 1.25 to 130 μg/g in fish and shellfish, 0.08 to 7.47 μg/g in vegetables, 0.51 to 9.48 μg/g in fruits, 1.05 to 3.01 μg/g in potatoes, 2.31 to 6.82 μg/g in pulses and 0.26 to 53.8 μg/g in other food items. Our data were generally in agreement with those reported by Kamei et al. (1986), Weber et al. (1997) and Mattila and Kumpulainen (2001) regardless of the analytical method used such as the extraction and detection techniques (Table 7).

The estimated average daily intakes of ubiquinol-10 and total CoQ_{10} in Japanese people were 2.07 and 4.48 mg, respectively, and intake of ubiquinol-10 accounted for 46% of the total CoQ_{10} intake (Table 8). Meat, fish and shellfish, vegetables and other foods, especially soybean oil, are

Table 4
The content of ubiquinol-10, ubiquinone-10 and total CoQ_{10} in fish and shellfish (muscle tissue)

Food	Scientific name	Content (μg/g wet weight)			% Ubiquinol-10[c]	Sampling	Preparation
		Ubiquinol-10[a]	Ubiquinone-10[a]	Total CoQ_{10}[a,b]			
Mackerel		0.52±0.16	10.1±1.3	10.6±1.3	4.9	One whole fish[d,e,f]	Skinned
Sardine	*Sardinops melanostictus*	0.70±0.04	11.2±0.9	11.9±0.9	5.8	One whole fish[d,e,g]	–
Horse mackerel		23.8±4.1	106±4.3	130±0.7	18.3	One whole fish[d,e,g]	–
Five-ray yellowtail	*Seriola quinqueradiata*	6.25±0.50	6.59±0.52	12.8±0.6	48.7	A 150 g serving[d,e]	Skinned
Young yellowtail		20.9±7.3	12.5±4.2	33.4±11.5	62.6	A 300 g serving[d,e]	–
Cod		1.71±0.26	1.99±0.42	3.70±0.65	46.2	A 150 g serving[d,e]	Skinned
Flatfish		0.61±0.13	1.19±0.09	1.80±0.16	34.0	One whole fish[d,e]	–
Salmon	*Oncorhynchus keta*	0.38±0.16	5.35±0.44	5.73±0.57	6.6	Half a fish[d,e]	Skinned
Tuna		0.51±0.04	4.36±0.26	4.87±0.22	10.5	A 300 g serving[d]	–
Scallop	*Patinopecten yessoensis*	1.33±0.23	3.62±0.34	4.95±0.22	27.0	One scallop[d,e]	Adductor muscle minced
Oyster	*Crassostrea gigas*	1.94±0.09	1.47±0.44	3.42±0.54	56.9	About 10 oysters[d,e]	–
Cuttlefish		1.84±0.67	2.83±0.32	4.67±0.97	39.4	One cuttlefish[d,e]	Skinned
Octopus		2.15±0.36	1.27±0.17	3.42±0.53	62.9	Half an octopus[d,e]	Legs minced
Shrimp		0.75±0.11	0.91±0.10	1.66±0.21	45.4	About 10 shrimps[d,e]	Skinned (head excluded)
Canned tuna		14.6±3.0	0.29±0.07	14.9±3.1	98.1	One can[d,e]	–
Fish sausage		0.45±0.03	0.80±0.07	1.25±0.09	35.9	One serving[d,e]	–

[a]Mean±S.D. of triplicate analyses.
[b]Total CoQ_{10} = ubiquinol-10 + ubiquinone-10.
[c]% Ubiquinol-10 = (ubiquinol-10/total CoQ_{10}) × 100.
[d]Sold refrigerated.
[e]Domestic product.
[f]Filleted.
[g]Excludes the internal organs.

Table 5
The content of ubiquinol-10, ubiquinone-10 and total CoQ_{10} in vegetables

Food	Scientific name	Content (μg/g wet weight)			% Ubiquinol-10[c]	Sampling	Preparation
		Ubiquinol-10[a]	Ubiquinone-10[a]	Total CoQ_{10}[a,b]			
Spinach	*Spinacia oleracea*	<0.07	0.44±0.16	0.44±0.16	n.d.[d]	A bunch[e,g]	Excludes core
Broccoli	*Brassica oleracea* var. *italica*	3.83±0.06	3.17±0.40	7.01±0.42	54.7	A bunch[e,g]	Minced with core
Perilla	*Perilla frutescens*	0.17±0.03	1.91±0.04	2.07±0.04	8.1	A bunch[f,g]	–
Pumpkin	*Cucurbita moschata*	<0.07	<0.07	n.d.	n.d.	Half a pumpkin[e,g]	Peeled, excludes seeds
Mustard spinach	*Brassica rapa* var. *peruviridis*	1.30±0.33	0.73±0.04	2.03±0.32	64.0	A bunch[e,g]	–
Cauliflower	*Brassica oleracea* var. *botrytis*	4.16±0.83	2.46±0.09	6.63±0.89	62.8	A bunch[e,g]	–
Eggplant	*Solanum melongena*	<0.07	0.99±0.12	1.01±0.10	n.d.	One whole eggplant[e,g]	Peeled
Garlic	*Allium sativum*	2.53±0.23	0.92±0.22	3.45±0.45	73.4	One clove of garlic[e,g]	Excludes core
Lotus root	*Nelumbo nucifera*	0.74±0.08	0.22±0.05	0.96±0.08	77.2	One whole lotus root[e,g]	Peeled
Chinese cabbage	*Brassica rapa* var. *glabra*	2.95±0.07	1.54±0.05	4.48±0.04	65.7	Half a Chinese cabbage[e,g]	Excludes core

Table 5 (*continued*)

Food	Scientific name	Content (μg/g wet weight)			% Ubiquinol-10[c]	Sampling	Preparation
		Ubiquinol-10[a]	Ubiquinone-10[a]	Total CoQ_{10}[a,b]			
Tomato	*Solanum lycopersicum*	<0.07	<0.07	n.d.	n.d.	One whole tomato[e,g]	Peeled, excludes seeds
Cabbage	*Brassica oleracea* var. *capitata*	2.22±0.27	0.85±0.12	3.07±0.33	72.2	Half a cabbage[e,g]	–
Japanese radish	*Raphanus sativus*	0.62±0.12	0.10±0.03	0.71±0.15	86.3	Half a Japanese radish[e,g]	Peeled
Onion	*Allium cepa*	0.38±0.04	0.29±0.00	0.67±0.04	57.1	One whole onion[e,g]	Peeled, excludes core
Cucumber	*Cucumis sativus*	<0.07	0.08±0.01	0.08±0.01	n.d.	One whole cucumber[e,g]	Peeled
Rape		4.26±0.30	2.43±0.07	6.70±0.34	63.6	A bunch[e,g]	–
Asparagus	*Asparagus* L.	1.11±0.28	1.05±0.35	2.16±0.55	51.4	A bunch[e,g]	–
Okra	*Abelmoschus esculentus*	<0.07	<0.07	n.d.	n.d.	One whole okra[e,h]	Excludes calyx
Parsley	*Petroselium crispum*	5.91±0.30	1.57±0.08	7.47±0.37	79.0	One whole parsley[e]	Excludes core
Soybean sprout		0.35±0.09	0.72±0.06	1.06±0.14	32.7	A bunch[e,g]	–
Welsh onion	*Allium fistulosum*	0.59±0.10	0.48±0.27	1.07±0.31	55.1	One whole welsh onion[e,g]	Excludes core

[a]Mean±S.D. of triplicate analyses.
[b]Total CoQ_{10} = ubiquinol-10 + ubiquinone-10.
[c]% Ubiquinol-10 = (ubiquinol-10/total CoQ_{10}) × 100.
[d]n.d.: not determined.
[e]Sold at room temperature.
[f]Sold refrigerated.
[g]Domestic product.
[h]Imported from the Philippines.

Table 6
The content of ubiquinol-10, ubiquinone-10 and total CoQ_{10} in fruits, potatoes, pulses, dairy products and other foodstuffs

Food	Scientific name	Content (μg/g wet weight)			% Ubiquinol-10[c]	Sampling	Preparation
		Ubiquinol-10[a]	Ubiquinone-10[a]	Total CoQ_{10}[a,b]			
Fruits							
Orange		0.88±0.22	0.14±0.07	1.02±0.28	86.5	One whole orange[e,g]	Peeled
Grapefruit	*Citrus X paradisi*	1.05±0.22	0.26±0.06	1.30±0.16	80.3	One whole grapefruit[e,h]	Peeled
Strawberry		0.33±0.09	0.18±0.02	0.51±0.11	65.3	20 strawberries[e,g]	Excludes calyx
Banana	*Musa spp.*	0.22±0.02	0.60±0.03	0.82±0.05	26.7	A bunch[e,i]	Peeled
Apple	*Malus pumila* var. *domestica*	0.62±0.09	0.59±0.09	1.21±0.02	51.0	One whole apple[e,g]	Peeled
Persimmon	*Diospyros kaki* L.	0.60±0.08	0.17±0.03	0.77±0.09	77.3	One whole persimmon[e,g]	Peeled, excludes seeds
Avocado	*Persea americana*	3.14±0.40	6.34±0.48	9.48±0.84	33.1	One whole avocado[e,j]	Peeled, excludes seeds
Potatoes							
Sweet potato	*Ipomoea batatas* L.	1.82±0.10	1.19±0.09	3.01±0.15	60.5	One whole sweet potato[e,g]	Peeled
White potato	*Solanum tuberosum* L.	0.68±0.08	0.38±0.05	1.05±0.11	64.1	One whole white potato[e,g]	Peeled
Japanese taro	*Colocasia esculenta*	1.18±0.11	0.62±0.06	1.80±0.17	65.6	One whole Japanese taro[e,g]	Peeled
Pulses							
Natto		3.60±0.61	1.97±0.63	5.57±1.24	64.6	A 45 g pack[f,g]	–
Tofu		1.10±0.23	1.76±0.07	2.86±0.20	38.6	A 350 g pack[f,g]	–
Adzuki bean	*Vigna angularis*	0.72±0.04	1.60±0.23	2.31±0.22	31.0	A 250 g pack[e,g]	–

Table 6 (*continued*)

Food	Scientific name	Content (μg/g wet weight)			% Ubiquinol-10[c]	Sampling	Preparation
		Ubiquinol-10[a]	Ubiquinone-10[a]	Total CoQ_{10}[a,b]			
Soybean	*Glycine max*	4.30±0.48	2.52±0.12	6.82±0.60	63.1	A 250 g pack[e,g]	–
Soy milk		0.94±0.05	1.56±0.09	2.50±0.14	37.6	200 mL[f,g]	–
Peas	*Pisum sativum* L.	1.15±0.11	1.19±0.04	2.34±0.08	49.2	A 50 g pack[e,g]	–
Dairy products							
Milk		0.24±0.01	0.07±0.01	0.31±0.01	78.2	200 mL[f,g]	–
Yogurt		0.18±0.01	0.09±0.00	0.26±0.01	66.9	500 g [f,g]	–
Cheese		0.75±0.05	0.66±0.04	1.41±0.02	53.1	100 g[f,g]	–
Others							
Roasted sesame seed	*Sesamum indicum*	12.4±2.3	5.24±0.38	17.6±2.1	70.2	A 75 g pack[e,g]	–
Almond	*Prunus dulcis*	1.10±0.46	3.89±0.49	4.99±0.10	22.1	A 1 kg pack[e,h]	–
Hen's egg		0.32±0.03	0.41±0.04	0.73±0.05	44.0	One egg[e,g]	Excludes shell, beaten
Mushroom	*Agaricus bisporus*	<0.07	<0.07	n.d.[d]	n.d.	6 mushrooms[e]	–
Soybean oil		33.3±5.2	20.6±2.5	53.8±7.7	61.8	500 mL[e]	–
Miso		2.36±0.50	0.10±0.00	2.45±0.50	96.1	500 g[e]	–

[a]Mean±S.D. of triplicate analyses.
[b]Total CoQ_{10} = ubiquinol-10 + ubiquinone-10.
[c]% Ubiquinol-10 = (ubiquinol-10/total CoQ_{10}) × 100.
[d]n.d.: not determined.
[e]Sold at room temperature.
[f]Sold refrigerated.
[g]Domestic product.
[h]Imported from the USA.
[i]Imported from Mexico.
[j]Imported from Chile.

Table 7
Comparison among studies of total CoQ_{10} reported in foods

Food	Total CoQ_{10} content (μg/g wet weight)			
	This study	Mattila and Kumpulainen (2001)	Weber et al. (1997)	Kamei et al. (1986)
Beef	30.3–40.1	36.5	31	31.0
Chicken	17.1–25.0	14.0	17	21.0
Fish	1.80–130	8.5–15.9	4.3–27	5.5–64.3
Tomato	n.d[a]	0.9	0.19	n.d.
Broccoli	7.01	n.d.	6.6	8.6
Cauliflower	6.63	2.7	4.9	1.4
Orange	1.02	1.4	2.2	n.d.
Apple	1.21	1.3	1.1	n.d.
Potato	1.05	0.5	0.52	1.0
Milk	0.31	0.1	n.d.	0.4
Cheese	1.41	1.2–1.3	<0.16	2.1
Yogurt	0.26	2.4	1.2	n.d.
Hen's egg	0.73	1.2	1.5	3.7

[a]n.d.: not determined.

major contributors to ubiquinol-10 intake and to total CoQ_{10} intake. The estimated average daily intake of total CoQ_{10} in the Japanese population was similar to the estimates of 3–5 mg/day and 3.8–5.4 mg/day, respectively, reported for the Danish (Weber et al., 1997) and Finnish (Mattila and Kumpulainen, 2001) populations.

Although ubiquinol-10 itself is rapidly oxidized to ubiquinone-10 in air, significant amounts of ubiquinol-10

Table 8
Mean daily intake of ubiquinol-10, ubiquinone-10 and total CoQ_{10}

Individual food group	Consumption (g/day)	Mean daily intake (mg/day)[a]		
		Ubiquinol-10	Ubiquinone-10	Total CoQ_{10}[b]
Meat				
Beef	15.8	0.130	0.426	0.556
Pork	26.9	0.377	0.414	0.791
Chicken	20.4	0.270	0.159	0.429
Offal meat	1.6	0.100	0.094	0.194
Subtotal		0.88	1.09	1.97
Fish and shellfish				
Mackerel and sardine	11.7	0.098	0.498	0.595
Salmon	3.9	0.001	0.021	0.022
Sea bream and right eye flounder	7.0	0.008	0.011	0.019
Tuna	6.5	0.003	0.028	0.032
Other fish	9.7	0.132	0.093	0.224
Shellfish	4.5	0.007	0.011	0.019
Cuttlefish	6.2	0.012	0.013	0.025
Prawns, shrimps and crabs	5.4	0.004	0.005	0.009
Canned seafood	2.3	0.034	0.001	0.034
Fish paste products	11.3	0.005	0.009	0.014
Subtotal		0.31	0.69	0.99
Vegetables				
Tomato	14.9	0.000	0.000	0.000
Spinach	20.5	0.000	0.009	0.009
Other deep yellow vegetables	36.1	0.069	0.046	0.115
Cabbage	21.3	0.047	0.018	0.065
Cucumber	10.3	0.000	0.001	0.001
Japanese radish	38.6	0.024	0.004	0.028
Onion	27.2	0.010	0.008	0.018
Chinese cabbage	19.1	0.056	0.029	0.086
Other yellow vegetables	44.4	0.070	0.047	0.116
Subtotal		0.28	0.16	0.44
Fruits				
Cherries	0.10	0.000	0.000	0.000
Citrus	29.2	0.028	0.006	0.034
Banana	11.7	0.003	0.007	0.010
Apple	24.2	0.015	0.014	0.029
Other fruits	35.8	0.067	0.117	0.184
Subtotal		0.11	0.14	0.26
Potatoes				
White potatoes	7.1	0.013	0.008	0.021
Sweet potatoes	28.5	0.019	0.011	0.030
Other potatoes	22.4	0.027	0.014	0.040
Subtotal		0.06	0.03	0.09

Table 8 (*continued*)

Individual food group	Consumption (g/day)	Mean daily intake (mg/day)[a]		
		Ubiquinol-10	Ubiquinone-10	Total CoQ_{10}[b]
Pulses				
Soybeans	2.1	0.009	0.005	0.014
Tofu	36.1	0.040	0.063	0.103
Natto	6.6	0.024	0.013	0.037
Soy milks	3.4	0.003	0.005	0.008
Other beans	1.7	0.002	0.002	0.004
Subtotal		0.08	0.09	0.17
Dairy products				
Milk	96.1	0.023	0.007	0.030
Cheese	1.9	0.001	0.001	0.003
Fermented milk	20.9	0.004	0.002	0.005
Subtotal		0.03	0.01	0.04
Others				
Seeds	2.1	0.014	0.010	0.024
Mushrooms	15.0	0.000	0.000	0.000
Eggs	36.6	0.012	0.015	0.027
Vegetable oils	8.3	0.276	0.171	0.447
Miso	12.4	0.029	0.001	0.030
Subtotal		0.33	0.20	0.53
Total intake		2.07	2.42	4.48

[a]Estimates of the average intake of ubiquinol-10, ubiquinone-10 and total CoQ_{10} per day were calculated based on their average contents for individual food groups (e.g., beef, chicken, citrus, etc.) determined in the present study and data on the consumption of individual food groups reported in the 2003 National Nutrition Survey conducted by the Japan Ministry of Health, Labor and Welfare.

[b]Total CoQ_{10} = ubiquinol-10 + ubiquinone-10.

were found in some processed foods such as tofu (made from ground soybeans that are subsequently boiled), natto (made from soybeans by boiling and subsequent fermentation), roasted sesame seeds, cheese, canned tuna and fish sausage. The reasons for the existence of ubiquinol-10 in these processed foods may be that they are processed in closed containers which would limit exposure to oxygen and minimize oxidation of ubiquinol-10, and that endogenous natural antioxidants such as ascorbic acid and tocopherol protect ubiquinol-10 in these foods from oxidation. The same reasons may apply to some cooked foods, because cooking processes such as boiling, baking, grilling and frying would not always cause exposure of ubiquinol-10 located inside the foods to oxygen; only ubiquinol-10 on the surface of the foods would be exposed. Also, most foods would contain natural antioxidants. It has been reported that frying meat (pork heart, pork chop) causes a loss of CoQ_{10} in the range of 14–32%, whereas CoQ_{10} in vegetables and eggs is unchanged upon boiling; this indicates a relatively low degree of CoQ_{10}-destruction during heat treatment of food items and suggests that the data for raw foods may be considered representative of a cooked meal (Weber, 2001). In addition to these findings, given that seafoods and vegetables are often eaten raw as sashimi or salad, respectively, and that oxidation of ubiquinol-10 during food processing and cooking is uncertain, as mentioned above, our estimation of dietary intake of ubiquinol-10 may be plausible, at least in part.

Regarding the potential importance of ubiquinol-10 content in foods, it has been suggested that ubiquinol-10 acts as a natural lipophilic antioxidant that protects sensitive substances such as polyunsaturated fatty acids in foods from oxidation while ubiquinone-10 does not act in this way (Passi et al., 2002). In addition, from the point of view of CoQ_{10} supply sources and benefits for humans, ubiquinol-10 in foods seems to be more important than ubiquinone-10 because ubiquinol-10 has a higher bioavailability than ubiquinone-10 (Mae et al., 2001), and ubiquinol-10 is also more effective than ubiquinone-10 in decreasing the degree of senescence in SAMP1 mouse, a model for accelerated senescence and severe senile amyloidosis (Yan et al., 2006).

Although 70 food items with a wide range of CoQ_{10} content were analyzed in this study, our data was not powerful enough to make assessments accounting for the diversity of sources and breed varieties or seasonal and inter-individual variability in the food items and further studies are necessary to make a complete assessment of ubiquinol-10 and ubiquinone-10 content in foodstuffs that make up the Japanese diet.

In summary, we describe a simple and reliable method using 2-propanol extraction and HPLC-ECD for the determination of ubiquinol-10 and ubiquinone-10 contents in various foods and the results of its application to analyze their contents. Most of the 70 food items contained ubiquinol-10 as well as ubiquinone-10. The estimated average intake of ubiquinol-10 was 2.07 mg/day and that of total CoQ_{10} 4.48 mg/day in the Japanese population.

Acknowledgment

The authors thank Dr. Hidaka for reviewing the manuscript.

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