

Original Article

Serum concentrations of antioxidant nutrients in healthy American, Chinese and Korean adults

Kyung-Jin Yeum¹ PhD, Yang Cha Lee-Kim² PhD, Shunshi Zhu³ MD, Shudong Xiao³ MD, Joel Mason¹ MD and Robert M Russell¹ MD

¹Jean Mayer United States Department of Agriculture, Human Nutrition Research Center on Ageing, Tufts University, Boston, MA, USA

²Department of Food and Nutrition, Yonsei University, Seoul, Korea

³Institute of Shanghai Second Medical University, Shanghai Ren-Ji Hospital, Shanghai Institute of Digestive Diseases, Shanghai, China

Population variabilities of serum carotenoid and tocopherol levels using reverse-phase HPLC in healthy American (Caucasian), Chinese and Korean adults were determined. Considerable variation in serum nutrient concentrations was found among these groups. The mean serum concentration of lutein/zeaxanthin was significantly higher in the Chinese than in the white Americans ($P < 0.001$), whereas serum α -carotene and lycopene concentrations were significantly higher in white Americans than in Chinese ($P < 0.001$). Serum β -carotene concentrations in Koreans were significantly higher than in either white Americans or in Chinese. The mean serum lycopene concentration in Koreans was lower than in Americans. The mean serum retinol concentration was significantly higher in white Americans than in Chinese or Koreans ($P < 0.005$). American serum α -tocopherol concentrations were significantly higher ($P < 0.005$) than those of the Chinese, whereas γ -tocopherol values were significantly lower ($P < 0.001$) than those of the Chinese. These differences probably reflect differences in dietary intakes among these population groups.

Key words: antioxidant nutrients, American adults, Chinese adults, Korean adults, carotenoids, tocopherols.

Introduction

There is continued interest in dietary antioxidant nutrients given that they may prevent oxidative damage in target tissues.¹ It is well known that carotenoids and tocopherols are important antioxidant nutrients.^{2–4} Epidemiological studies have indicated that these dietary fat-soluble nutrients are related to a lower risk of certain types of chronic diseases such as cancer,^{5–7} cardiovascular disease^{8,9} and eye disease.^{10–12}

Preformed vitamin A occurs in foods of animal origin, and the absorption of retinol is more efficient than that of carotenoids. However, the circulating retinol concentration is controlled by a homeostatic mechanism; therefore, the plasma retinol concentration does not reflect vitamin A nutritional status, unless there is vitamin A deficiency or excess.^{13,14} In contrast, it has been clearly demonstrated that serum carotenoid concentrations mainly reflect immediate dietary carotenoid intake.^{15–17} Similarly, it has been shown that the serum α -tocopherol concentration reflects dietary vitamin E,¹⁴ even though tocopherol-binding protein appears to regulate plasma/serum α -tocopherol levels within a narrow range of concentrations.⁴

Food habits as well as the cultural and social environments of Asian people are different from those of Western people.¹⁸ Therefore, we measured serum concentrations of carotenoids, retinol and tocopherols in healthy American, Chinese and Korean adults to determine the variability of antioxidant nutrients among these population groups.

Materials and methods

Chemicals

All-*trans*- β -carotene (type IV), α -carotene, all-*trans*-retinol, retinyl palmitate, γ -tocopherol and α -tocopherol were purchased from Sigma Chemical Co. (St. Louis, MO, USA). Lutein was purchased from Kemin Industries, Inc. (Des Moines, IA, USA). Zeaxanthin, cryptoxanthin, tocol, and γ -carotene were gifts from Hoffmann-La Roche Inc. (Nutley, NJ, USA). Solutions of carotenoids and retinoids were prepared under red light immediately before use. All high performance liquid chromatography (HPLC) solvents were obtained from JT Baker Chemical Co. (Philipsburg, NJ, USA) and were filtered through a 0.45 μ m membrane filter before use.

Subjects

Fasting blood samples from human volunteers (56 American Caucasian, 25 Chinese and 53 Korean) were analyzed (Table 1). Subjects were recruited from each community: Boston in America, Shanghai in China and Seoul in Korea. American and Korean blood samples were drawn in the autumn, and Chinese samples were collected in the spring. Written informed consent was obtained from each volunteer.

Correspondence address: Kyung-Jin Yeum, Jean Mayer USDA Human Nutrition Research Center on Ageing, Tufts University, Boston, MA 02111, USA.
Tel: +1 617 556 3128, Fax: +1 617 556 3344,
E-mail: Yeum_GI@HNRC.Tufts.edu

Table 1. Characteristics of the subjects

	Male	Female	Age
American	12	44	38.4 ± 1.6
Chinese	22	3	39.9 ± 2.8
Korean	20	33	52.8 ± 2.0

Chinese and Korean serum samples were frozen in dry ice before being transported to the Nutrition Research Center at Tufts University. Serum was stored at -80°C until analysis.

Carotenoids, retinoids and tocopherol analysis

Serum antioxidant nutrients were extracted using a modified method previously described.¹⁹ American and Chinese samples were analyzed for β-carotene, α-carotene, cryptoxanthin, lycopene, lutein/zeaxanthin, retinol, α-tocopherol and γ-tocopherol, whereas Korean samples were analyzed for β-carotene, lycopene, retinol and α-tocopherol. Briefly, 150 μL of serum was extracted twice with 2 mL of CHCl₃/CH₃OH (2:1, v/v) and 3 mL of hexane. γ-Carotene, retinyl acetate and tocol were added as internal standards. All sample processing was done under red light.

The HPLC system consisted of a Series 410 LC pump (Perkin-Elmer Inc. Norwalk, CT, USA), a Waters 717 plus autosampler (Millipore, Milford, MA, USA), a Pecosphere-3 C18 0.46 × 8.3 cm cartridge column (Perkin-Elmer Inc., Norwalk, CT, USA), a Waters 994 programmable photodiode array detector and a Waters 840 digital 350 data station. The HPLC mobile phase was CH₃CN:THF:H₂O (50:20:30, v/v/v, 1% ammonium acetate in H₂O, solvent A) and CH₃CN:THF:H₂O (50:44:6, v/v/v, 1% ammonium acetate in H₂O, solvent B). The gradient procedure at a flow rate of 1 mL/min was as follows: 60% solvent A and 40% solvent B were used for 2 min followed by a 9 min linear gradient to 83% solvent B, then a 2 min linear gradient to 60% solvent B, a 2 min hold at 100% solvent B, and finally a 2 min gradient back to 100% solvent A and 40% solvent B. The Waters 994 programmable photodiode array detector was set at 340 nm for retinoids and 450 nm for carotenoids. A fluorescence detector (Waters 470, Millipore, Ex:292 nm, Em:330 nm) was connected for tocopherol analysis. Using this method, lutein and zeaxanthin coelute, but other major carotenoids are adequately separated. Carotenoids, retinoids and tocopherols were quantified by determining peak areas in the

HPLC chromatograms calibrated against known amounts of standards. Levels were corrected for extraction and handling losses by monitoring the recovery of the internal standards. The lower limit of detection was 0.2 pmol for carotenoids, 2.0 pmol for retinoids and 8 pmol for tocopherols.

Statistical analysis

Results are expressed as mean ± SEM. The significance of difference was determined by Student's *t*-test or analysis of variance using the Statview II program (Abacus Concepts Inc., Berkeley, CA, USA).

Results

The mean ± SEM ages of each group were 38.4 ± 1.6, 39.9 ± 2.8 and 52.8 ± 2.0 years for American, Chinese and Korean subjects, respectively. The mean serum concentration of lutein/zeaxanthin was significantly higher in Chinese subjects (31.0 ± 2.7 μg/dL) as compared with American subjects (15.7 ± 1.0 μg/dL, *P* < 0.001), whereas serum concentrations of α-carotene (4.8 ± 0.6 vs 0.9 ± 0.1) and lycopene (32.2 ± 1.8 vs 4.8 ± 1.6) were significantly higher in American subjects as compared with Chinese subjects (*P* < 0.001). Serum β-carotene concentrations were similar in American and Chinese subjects. Concentrations of the total major carotenoids were 78.9 ± 3.4 and 59.1 μg/dL for American and Chinese, respectively. The mean American serum α-tocopherol concentration was significantly higher (*P* < 0.005) than the Chinese value, whereas the mean American serum γ-tocopherol value was significantly lower (*P* < 0.001) than the Chinese value (Table 2). Lycopene and β-carotene were major carotenoids in the American subjects, whereas lutein was the predominant carotenoid in the Chinese subjects. Lycopene had the lowest concentration of all carotenoids in the Chinese serum (Fig. 1). The mean serum retinol concentration was significantly higher in American subjects (52.6 ± 2.1 μg/dL) than in Chinese (36.8 ± 2.6 μg/dL *P* < 0.001) and Korean subjects (43.3 ± 2.41 μg/dL, *P* < 0.005). Serum β-carotene levels were significantly higher in Korean subjects (37.4 ± 2.7 μg/dL) than in American (17.1 ± 1.5 μg/dL) or Chinese (15.6 ± 2.6 μg/dL) subjects. The mean serum lycopene concentration in Koreans (2.5 ± 0.4 μg/dL) was the lowest among the three groups, and significantly lower than the value for Americans. Serum lycopene concentration showed no significant difference between Chinese subjects and

Table 2. The serum concentration of antioxidant nutrients in healthy adults (unit: μg/dL)

	American (<i>n</i> = 56)	Subjects Chinese (<i>n</i> = 25)	Korean (<i>n</i> = 53)
Provitamin A carotenoids			
β-carotene	17.1 ± 1.5	15.6 ± 2.6	37.4 ± 2.7
α-carotene	4.8 ± 0.6	0.9 ± 0.1***	NA
Cryptoxanthin	9.1 ± 0.6	6.8 ± 1.1*	NA
Non-provitamin A carotenoids			
Lycopene	32.2 ± 1.8	4.8 ± 1.6***	2.5 ± 0.4
Lutein and Zeaxanthin	15.7 ± 1.0	31.0 ± 2.7***	NA
Total carotenoid	78.9 ± 3.4	59.1 ± 5.4*	NA
Retinol	52.6 ± 2.1	36.8 ± 2.6***	43.3 ± 2.4
α-tocopherol	947.1 ± 37.5	690.4 ± 51.6**	911.0 ± 65.7
γ-tocopherol	145.9 ± 8.7	399.5 ± 63.6***	NA

Values are mean ± SEM. Significantly different from Americans, **P* < 0.05, ***P* < 0.005, ****P* < 0.001. NA, data are not available.

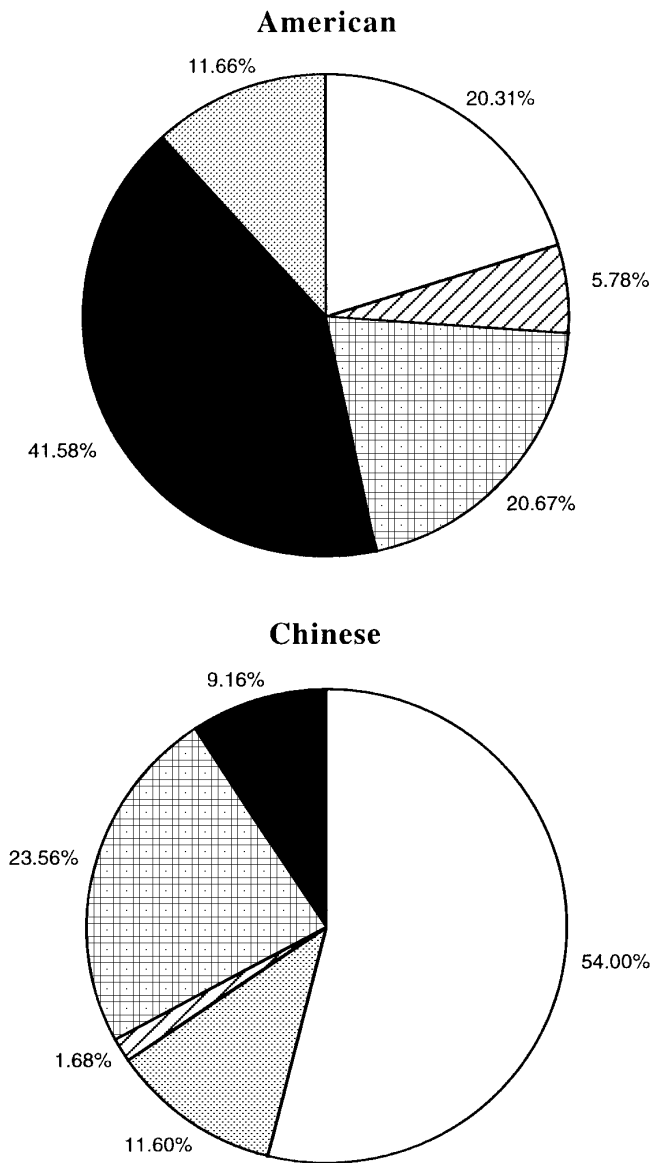


Figure 1. Serum carotenoid composition in American and Chinese healthy adults. (□), lutein/zeaxanthin; (▨), cryptoxanthin; (▧), α -carotene; (▩), *trans* β -carotene; (■), lycopene.

Korean subjects. There was no significant difference in serum α -tocopherol concentration between American and Korean subjects, but the mean Chinese serum α -tocopherol concentration was significantly lower than the value for Koreans ($P < 0.05$; Fig. 2).

Discussion

The data shows great variability of serum antioxidant nutrients among the three study populations. Serum concentrations of retinol and tocopherol in American subjects were comparable with those found among British adults.²⁰ However, the concentrations of these fat-soluble vitamins in Chinese subjects were significantly lower than in American subjects. Even though homeostatic mechanisms tightly control the transport of retinol to target tissue,^{13,14} and the serum

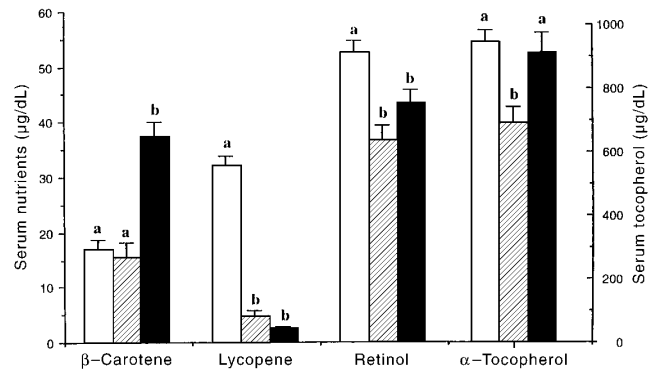


Figure 2. Serum carotenoid, retinol, and α -tocopherol concentrations in (□) American ($n = 56$), (▨) Chinese ($n = 25$) and (■) Korean ($n = 53$) healthy adults. Bars not sharing the same letter are statistically different, $P < 0.05$ (ANOVA).

α -tocopherol level is regulated by tocopherol-binding protein,⁴ their serum concentrations differed in the various groups. In contrast with serum α -tocopherol, γ -tocopherol (which has about one-half the antioxidant activity and one-tenth the biologic activity of α -tocopherol²¹) was significantly lower in American sera than in Chinese sera.

Lutein, cryptoxanthin, β -carotene, and lycopene were the major carotenoids in both American and Chinese sera, as reported earlier.^{14,22} However, compared with the American adults, serum lutein/zeaxanthin concentrations were approximately two-fold higher, and lycopene approximately six-fold lower, in the Chinese adults. Seasonal variations of serum/plasma carotenoids in several population groups have been reported.^{23–25} Because season affects the pattern of food intake in China (data not shown), a seasonal variability in Chinese subjects cannot be overlooked. Serum lycopene concentrations in Koreans were more than 10-fold lower than those found in white Americans. It is well known that individual fruit and vegetables provide specific carotenoids. Yellow or orange vegetables, such as carrot and squash, have high levels of α -carotene and β -carotene; green leafy vegetables, such as spinach and broccoli, contain lutein, zeaxanthin, and β -carotene;²⁶ while tomatoes contain high levels of lycopene.²⁷ Dietary data was not available in our study; however, given that serum carotenoid concentrations reflect recent dietary intake of fruits and vegetables,^{14–17,28} these differences probably reflect a higher consumption of green leafy vegetables and a lower intake of tomatoes and their products in Oriental people as compared with Western Caucasian people.

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*Asia Pacific Journal of Clinical Nutrition (1999) Volume 8, Number 1: 4-8***美国, 中国, 和韩国的健康人群血清中抗氧化 营养素的差异****摘要:**

为调查不同种族间血清中的类胡萝卜素和维生素E的差异, 我们运用反向高效液相色谱法测定了美国白人, 中国人, 和韩国人健康人群血清中的类胡萝卜素和维生素E的水平。研究结果显示, 三组不同人群血清中的营养素浓度明显不同。中国人血清中的叶黄素/玉米黄素的平均浓度显著高于美国人 ($p < 0.001$)。 美国人血清中的 α -胡萝卜素和蕃茄红素浓度显著高于中国人 ($p < 0.001$)。 韩国人血清中的 β -胡萝卜素浓度显著高于美国人和中国人, 而其蕃茄红素浓度显著低于美国人。研究结果还显示, 美国人血清中的视黄醇浓度显著高于中国人和韩国人($p < 0.005$), 其 α -维生素E浓度显著高于中国人($p < 0.005$), 但是, 其 γ 维生素 E 的浓度却显著低于中国人 ($p < 0.001$)。 这些结果的差异可能反映了这些人群饮食的不同。

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*Asia Pacific Journal of Clinical Nutrition (1999) Volume 8, Number 1: 4-8***요 약 문**

인구집단에 따른 혈청 영양소 농도의 다양성을 연구하였다. 미국인, 중국인, 그리고 한국인 정상 성인을 대상으로, 이들의 혈청 카로티노이드, 비타민 A, 그리고 비타민 E 농도를 HPLC로 분석하였다. 중국인의 평균 혈청 루테인/지아잔틴 농도는 미국인의 혈청 농도보다 유의적으로 높은 반면 ($p < 0.001$), α -카로틴과 라이코펜 농도는 중국인의 혈청 농도가 미국인의 혈청 농도보다 유의적으로 낮았다 ($p < 0.001$). 한국인의 혈청 β -카로틴 농도는 미국인, 그리고 중국인의 혈청 농도보다 유의적으로 높았다. 한국인의 혈청 라이코펜 농도 역시 미국인의 혈청 농도보다 낮았다. 미국인의 혈청 비타민 A 농도는 중국인 또는 한국인의 혈청 농도보다 유의적으로 높았다 ($p < 0.005$). 미국인의 혈청 α -토코페롤 농도는 중국인보다 유의적으로 높은 반면 ($p < 0.005$), γ -토코페롤 농도는 중국인보다 유의적으로 낮았다 ($p < 0.001$). 이와 같은 혈청 영양소 농도의 다양성은 이들 인구 집단의 식품섭취의 다양성과 연관되는 것으로 생각된다.

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