RESEARCH COMMUNICATION

Cancer Mortality and Serum Levels of Carotenoids, Retinol, and Tocopherol: a Population-based Follow-up Study of Inhabitants of a Rural Area of Japan

Yoshinori Ito[1], Mio Kurata[1], Risa Hioki[1], Koji Suzuki[1], Junichi Ochiai[2], Kunio Aoki[3]

Abstract

A total of 3,182 subjects (1,239 males and 1,943 females) aged from 39y to 79y, were recruited from the inhabitants of a rural area in Japan who participated in health check-up programs from 1988 to 1995. During the 10.5 year follow-up, 287 deaths (175 males and 112 females) from all causes, 134 (81 males and 53 females) from cancer of all sites, 31 from lung cancer, 21 from colorectal cancer, 20 from stomach cancer, and 62 from other cancers, were identified among the cohort subjects. Fasting serum samples were taken at the time of the health check-ups, and serum levels of carotenoids, retinol and tocopherols were separately determined by HPLC. Statistical analyses were performed using Cox's proportional hazard model after adjusting for sex, age, and other confounding factors. High serum levels of α- and β- carotenes and lycopene were found to marginally significantly or significantly reduce the risk for mortality rates of cancer of all sites and of colorectal cancers. High serum levels of β-cryptoxanthin also showed an inversely relation with the risk of mortality from lung and stomach cancers, but this was not statistically significant. High intake of green-yellow vegetables contributing to serum levels of α- and β- carotenes, as well as lycopene, may reduce the risk of cancer mortality, especially from colorectal cancer, in rural Japanese.

Key Words: Follow-up study - colorectal cancer - lung cancer - stomach cancer - β-carotene - lycopene

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Introduction

Many epidemiological studies have shown that dietary intake of vegetables and fruits rich in carotenoids such as β-carotene (BC) can reduce a risk of human cancer mortality such as lung cancer and stomach cancer (Hirayama, 1979; Block et al.,1992, Vainio et al., 1998). Recent studies have found that serum carotenoids such as β-cryptoxanthin (CR) and lutein have also clear inverse associations with lung cancer risk (Yuan et al., 2001). We previously found that high serum levels of α-carotene (AC) and BC among male inhabitants can reduce significantly the risk of cancer deaths such as lung cancer death, in a case-control study nested in a large-scale Japanese cohort (Ito et al., 2003). Serum levels of AC and BC are affected by daily lifestyle elements such as diet (Micozzi et al., 1992; Olsen, 1999), smoking and alcohol consumption (Aoki et al., 1987; Stryker et al., 1988). However, a recent intervention study of BC administration did not show the expected inverse association with lung cancer incidence among non-Asians (Vainio et al., 1998).

The present study was conducted to investigate the effects of serum levels of carotenoids, retinol (RE) and tocopherol on deaths from lung, colorectal, or stomach cancer in the follow-up study of inhabitants of a rural area of Japan.

Subjects and Methods

Subjects

The study subjects were recruited from inhabitants of a rural area of Hokkaido, Japan, who participated in health check-up programs from 1988 to 1995.The followed-up cohort subjects were 3,182 participants (1,239 males and 1,943 females), aged from 39y to 79y. During the 10.5y follow-up, 287 deaths (175 males and 112 females) from all causes, 134 (81 males and 53 females) from cancer of all sites, 31 from lung cancer, 21 from colorectal cancer, 20

[1]Department of Environmental Health, Fujita Health University Graduate School of Health Sciences [2]Department of Medical Electronics, Fujita Health University School of Health Sciences [3]Aichi Cancer Center
Correspondence: Dr. Yoshinori Ito, Department of Public Health, Fujita Health University School of Health Sciences, Dengakugakubo, Kutsukake-cho, Toyoake city, Aichi prefecture, 470-1192, Japan Tel&Fax: +81-562-93-9405 E-mail: yoshiito@fujita-hu.ac.jp

Table 1. Charasteristics of the Study Subjects

Item		Males	Females	Overall
Subjects	Total	1,239(100.0)	1,943(100.0)	3,182(100.0)
Age	39 - 49	441 (35.6)	717 (36.9)	1,158 (36.4)
	50 - 59	351 (28.3)	617 (31.8)	968 (30.4)
	60 - 69	349 (28.2)	497 (25.6)	846 (26.6)
	70 - 79	98 (7.9)	110 (5.7)	208 (6.5)
Smoking status	Current smoker	637 (51.5)	217 (11.2)	854 (26.8)
	Former smoker	318 (25.7)	82 (4.2)	400 (12.6)
	Never smoker	284 (22.9)	1,644 (84.6)	1,928 (60.6)
Alcohol drinking habit	Regular drinker	935 (75.5)	560 (28.8)	1,495 (47.0)
	Irreguar drinker	63 (5.1)	38 (2.0)	101 (3.2)
	Non drinker	241 (19.4)	1,344 (69.2)	1,585 (49.8)
Deaths	All causes	175 (100.0)	112 (100.0)	287 (100.0)
	Cancer of all sites	81 (46.3)	53 (47.3)	134 (46.7)
	Lung cancer	26 (14.9)	5 (4.5)	31 (10.8)
	Colorectal cancer	10 (5.7)	11 (9.8)	21 (7.3)
	Stomach cancer	17 (9.7)	3 (2.7)	20 (7.0)
	Liver cancer	6 (3.4)	5 (4.5)	11 (3.8)
	Other cancer	22 (12.6)	29 (25.9)	51 (17.8)
	Circulatory diseases	52 (29.7)	32 (13.4)	84 (29.3)
	Accidents	10 (5.7)	8 (3.3)	18 (6.3)
	Others	32 (18.3)	19 (7.9)	51 (17.8)
Moved		112 (9.0)	239 (12.3)	351 (11.0)

from stomach cancer, 62 from other cancers, 84 from circulatory diseases, 18 from accidents, and 51 from others, were identified among the cohort subjects (Table 1).

Methods

Fasting serum samples were taken at the time of the health check-ups, and then serum levels of carotenoids, RE, and α-tocopherol (AT) was separately determined by the HPLC (Ito et al., 1990). Serum levels of zeaxanthin & lutein (ZL) and total carotenes (TCR) were the sums of zeaxanthin and lutein levels or the sums of AC, BC, and lycopene (LY) levels, respectively. Data for serum levels of CR, ZL, RE, and AT from the subjects at 1988 were not included in the analyzed data because values for samples collected could not be determined. Informed consent to use the information obtained from questionnaires and sera collected at the time

Table 2-1. Comparison of Serum Levels of Carotenoids Between Alive and Dead from Cancers

Serum components (μmol/l)	Alive (AL)	Dead from Cancer: All sites (AC)	Lung cancer (LC)	Colorectal cancer (CC)	Stomach cancer (SC)	Differences (probability) (p-values): (AL)vs(AC)	(AL)vs(LC)	(AL)vs(CC)	(AL)vs(SC)
Total carotenes	1.244	0.952	0.906	0.911	0.817	<0.001	0.004	0.018	0.003
α-Carotene	0.141	0.115	0.104	0.111	0.105	0.005	0.010	0.098	0.044
β-Carotene	0.698	0.515	0.517	0.500	0.385	<0.001	0.016	0.029	0.001
Lycopene	0.328	0.244	0.229	0.227	0.242	<0.001	0.007	0.023	0.074
Number	2,891	132	31	21	19				

Data are geometric means calculated after adjusting for sex and age.

Table 2-2. Comparison of Serum Levels of Carotenoids, Retinol, and Tocopherol Between Alive and Dead from Cancer

Serum components (μmol/l)	Alive (AL)	Dead from Cancer: All sites (AC)	Lung cancer (LC)	Colorectal cancer (CC)	Stomach cancer (SC)	Differences (probability) (p-values): (AL)vs(AC)	(AL)vs(LC)	(AL)vs(CC)	(AL)vs(SC)
β-Cryptoxanthin	0.311	0.266	0.249	0.286	0.145	0.043	0.165	0.644	<0.001
Zeaxanthin&lutein	0.985	0.872	0.950	0.886	0.810	0.035	0.775	0.436	0.195
Retinol	2.234	2.353	2.142	2.651	2.521	0.200	0.875	0.070	0.261
α-Tocopherol	23.61	23.77	23.82	25.11	20.71	0.860	0.890	0.491	0.181
Number	2,092	82	18	15	12				

Data are geometric means calculated after adjusting for sex and age.

of health check-ups was obtained using the protocol for epidemiological studies of life-style-related disease prevention by individual signature, as previously reported (Ito et al., 2002).

Statistical analyses were performed using Student t-tests for mean value differences, after controlling for sex and age, and the Cox's proportional hazard model (JMP ver 4.0) for analysis of hazard risk, after adjusting for sex and age (model 1), and sex, age, smoking habit, and serum levels of total cholesterol and alanine aminotransferase (ALT) activity (model 2).

Results

1. Comparison of Serum Levels of Carotenoids, Retinol, and Tocopherol Between Alive and Dead Among the Cohort

Geometric mean values for serum carotenoids such as AC, BC and LY were marginally significantly or significantly lower for the cancer deaths in all sites, and for lung, colorectum and stomach, than for the survivors among the cohort (Table 2-1). Values for TCR were also significantly lower for cancer mortality in all sites and all three specific sites. Moreover, geometric mean values for serum ZX for all sites and for serum CR for stomach cancer were significantly lower (Table 2-2). However, there were no apparent differences in serum levels of RE and AT between those suffering mortality due to cancers and the surviving group.

2. Hazard Ratios of Serum Carotenoids, Retinol, and Tocopherol for Cancer Mortality Among the Cohort

The hazard ratios of high serum levels of carotenes such as BC and LY appeared to be lower with regard to cancer mortality in all sites (Table 3-1). Those for AC and LY, but not BC, also were lower for lung cancer mortality. The hazard ratios for high serum levels of carotenoids, such as AC, BC, and LY appeared to be lower for colorectal cancer death and those for high serum levels of carotenoids were statistically significant in model 2 (Table 3-2). Those for high serum levels of AC and BC also tended to be lower for stomach cancer death. High serum levels of CR, ZL, RE, and AT tended to be lower for cancer mortality of all sites and lung (Table 4-1). Moreover, hazard ratios of high serum levels of CR and ZL for mortality rates of colorectal and stomach cancer also tended to be lower in model 2, but not statistically significant (Table 4-2). However, values for high serum

Table 3-1. Hazard Ratios for Serum Levels of Carotenoids for Cancer Deaths of All Sites and Lung

Serum component (μmol/l)	Group	Person year	All sites Deaths	Model 1 HR(95%CI)	trend p	Model 2 HR(95%CI)	trend p	Lung Deaths	Model 1 HR(95%CI)	trend p	Model 2 HR(95%CI)	trend p
Total carotenes	High	10060.5	37	0.54 (0.36-0.83)	0.02	0.67 (0.43-1.06)	0.23	10	0.91 (0.34-2.43)	0.27	1.34 (0.47-3.77)	0.58
	Moderate	10170.5	43	0.75 (0.50-1.13)		0.82 (0.54-1.24)		13	1.70 (0.69-4.22)		1.63 (0.64-4.14)	
	Low	9908.0	52	1.00		1.00		8	1.00		1.00	
α-Carotene	High	10243.0	47	0.58 (0.69-0.87)	0.004	0.68 (0.48-1.05)	0.05	17	0.84 (0.38-1.86)	0.15	0.97 (0.41-2.30)	0.14
	Moderate	9935.0	31	0.52 (0.34-0.81)		0.59 (0.38-0.93)		4	0.33 (0.10-1.03)		0.34 (0.10-1.09)	
	Low	9960.0	54	1.00		1.00		10	1.00		1.00	
β-Carotene	High	9995.0	34	0.51 (0.33-0.81)	0.004	0.64 (0.39-1.03)	0.04	11	1.05 (0.38-2.91)	0.10	1.55 (0.53-4.56)	0.17
	Moderate	10027.5	52	1.01 (0.68-1.51)		1.16 (0.77-1.74)		14	2.23 (0.86-5.74)		2.43 (0.92-6.43)	
	Low	10116.5	46	1.00		1.00		6	1.00		1.00	
Lycopene	High	10390.5	33	0.54 (0.35-0.83)	0.01	0.61 (0.39-0.97)	0.01	11	0.72 (0.31-1.66)	0.57	0.93 (0.39-2.24)	0.76
	Moderate	9953.0	41	0.71 (0.48-1.06)		0.78 (0.52-1.17)		8	0.64 (0.27-1.56)		0.72 (0.29-1.78)	
	Low	9759.0	58	1.00		1.00		12	1.00		1.00	

Model: Cox proportional hazard model; adjusting for sex and age (model 1) and for sex, age, smoking habit, and serum levels of total cholesterol and ALT activity (model 2). HR (95%CI): hazard ratio (95% confidence interval) Total carotenes: the sum of α- and β-carotenes, and lycopene levels.

Table 3-2. Hazard Ratios of Serum Levels of Carotenoids for Cancer Deaths of Colorectal Cancer and Stomach

Serum component (μmol/l)	Group	Person year	Colorectal Deaths	Model 1 HR(95%CI)	trend p	Model 2 HR(95%CI)	trend p	Stomach Deaths	Model 1 HR(95%CI)	trend p	Model 2 HR(95%CI)	trend p
Total carotenes	High	10060.5	4	0.29 (0.09-0.91)	0.08	0.21 (0.06-0.68)	0.02	5	0.77 (0.22-2.71)	0.37	1.20 (0.32-4.53)	0.38
	Moderate	10170.5	6	0.48 (0.18-1.32)		0.40 (0.15-1.12)		9	1.65 (0.55-4.92)		2.07 (0.68-6.29)	
	Low	9908.0	11	1.00		1.00		5	1.00		1.00	
α-Carotene	High	10243.0	4	0.33 (0.10-1.10)	0.18	0.25 (0.07-0.87)	0.09	4	0.35 (0.10-1.18)	0.22	0.48 (0.13-1.72)	0.43
	Moderate	9935.0	8	0.85 (0.32-2.22)		0.72 (0.27-1.92)		7	0.84 (0.30-2.32)		1.04 (0.37-2.96)	
	Low	9960.0	9	1.00		1.00		8	1.00		1.00	
β-Carotene	High	9995.0	5	0.37 (0.12-1.09)	0.16	0.28 (0.09-0.85)	0.07	4	0.43 (0.12-1.49)	0.29	0.62 (0.17-2.31)	0.50
	Moderate	10027.5	5	0.53 (0.19-1.46)		0.46 (0.16-1.28)		8	1.08 (0.39-2.98)		1.29 (0.46-3.66)	
	Low	10116.5	10	1.00		1.00		7	1.00		1.00	
Lycopene	High	10390.5	3	0.29 (0.08-1.04)	0.16	0.23 (0.06-0.84)	0.08	4	0.77 (0.20-2.87)	0.17	1.07 (0.27-4.20)	0.19
	Moderate	9953.0	7	0.67 (0.26-1.72)		0.58 (0.22-1.52)		10	2.06 (0.70-6.04)		2.39 (0.80-7.10)	
	Low	9759.0	11	1.00		1.00		9	1.00		1.00	

Model: Cox proportional hazard model; adjusting for sex and age (model 1) and for sex, age, smoking habit, and serum levels of total cholesterol and ALT activity (model 2) HR (95%CI): hazard ratio (95% confidence interval) Total carotenes: the sum of α- and β-carotenes, and lycopene levels.

Table 4-1. Hazard Ratios of Serum Levels of Carotenoids, Retinol and Tocopherol for Cancer Deaths of All Sites and Lung

Serum component (μmol/l)	Group	Person year	All sites Deaths	Model 1 HR(95%CI)	trend p	Model 2 HR(95%CI)	trend p	Lung Deaths	Model 1 HR(95%CI)	trend p	Model 2 HR(95%CI)	trend p
β-Cryptoxanthin	High	7643.0	28	0.78 (0.46-1.34)	0.61	0.96 (0.54-1.69)	0.93	5	0.59 (0.18-1.95)	0.59	0.66 (0.18-2.36)	0.79
	Moderate	5909.0	28	0.98 (0.57-1.67)		1.06 (0.61-1.84)		7	1.03 (0.35-3.07)		0.93 (0.29-2.97)	
	Low	6057.0	26	1.00		1.00		6	1.00		1.00	
Zeaxanthin	High	6719.0	22	0.64(0.37-1.10)	0.26	0.68 (0.39-1.17)	0.36	7	1.24 (0.42-3.67)	0.86	1.27 (0.42-3.87)	0.71
&lutein	Moderate	5615.0	24	0.90(0.54-1.53)		0.93 (0.54-1.59)		4	0.90 (0.26-3.10)		0.71 (0.18-2.81)	
	Low	7274.0	36	1.00		1.00		7	1.00		1.00	
Retinol	High	8328.0	34	0.70 (0.41-1.18)	0.22	0.73 (0.43-1.27)	0.32	7	0.47 (0.16-1.43)	0.21	0.46 (0.14-1.50)	0.21
	Moderate	5787.5	19	0.62 (0.35-1.11)		0.65 (0.36-1.18)		3	0.34 (0.09-1.29)		0.33 (0.09-1.29)	
	Low	5505.0	29	1.00		1.00		8	1.00		1.00	
α-Tocopherol	High	7431.0	32	0.87 (0.50-1.50)	0.81	0.95 (0.52-1.72)	0.92	8	0.67 (0.23-1.96)	0.36	0.56 (0.16-2.03)	0.33
	Moderate	6240.0	26	1.01 (0.58-1.77)		1.06 (0.59-1.88)		3	0.37 (0.09-1.46)		0.34 (0.08-1.43)	
	Low	5925.0	24	1.00		1.00		7	1.00		1.00	

Model: Cox's proportional hazard model; adjusting for sex and age (model 1) and for sex, age, smoking habit, and serum levels of total cholesterol and ALT activity (model 2). HR (95%CI): hazard ratio (95% confidence interval) Total carotenes: the sum of α- and β-carotenes, and lycopene levels.

Table 4-2. Hazard Ratios of Serum Levels of Carotenoids, Retinol and Tocopherol for Cancer Deaths from Colorectal and Stomach

Serum component (μmol/l)	Group	Person year	Colorectal Deaths	Model 1 HR(95%CI)	trend p	Model 2 HR(95%CI)	trend p	Stomach Deaths	Model 1 HR(95%CI)	trend p	Model 2 HR(95%CI)	trend p
β-Cryptoxanthin	High	7643.0	5	0.90 (0.24-3.39)	0.79	0.76 (0.19-3.00)	0.7	3	0.41 (0.10-1.64)	0.35	0.69 (0.16-3.03)	0.75
	Moderate	5909.0	6	1.35 (0.38-4.79)		1.28 (0.35-4.61)		3	0.47 (0.12-1.88)		0.59 (0.14-2.45)	
	Low	6057.0	4	1.00		1.00		6	1.00		1.00	
Zeaxanthin&lutein	High	6719.0	5	1.06 (0.30-3.77)	0.86	0.95 (0.26-3.43)	0.85	2	0.44 (0.09-2.29)	0.29	0.55 (0.10-2.88)	0.29
	Moderate	5615.0	5	1.38 (0.39-4.84)		1.34 (0.38-4.73)		5	1.64 (0.47-5.71)		1.92 (0.55-6.77)	
	Low	7274.0	5	1.00		1.00		5	1.00		1.00	
Retinol	High	8328.0	9	1.56 (0.45-5.35)	0.35	1.31 (0.37-4.60)	0.39	4	0.78 (0.19-3.23)	0.93	1.16 (0.26-4.79)	0.97
	Moderate	5787.5	2	0.51 (0.09-2.81)		0.45 (0.08-2.46)		4	0.98 (0.24-3.95)		1.19 (0.28-4.97)	
	Low	5505.0	4	1.00		1.00		4	1.00		1.00	
α-Tocopherol	High	7431.0	5	1.73 (0.33-9.15)	0.22	1.26 (0.22-7.14)	0.12	4	0.54 (0.15-1.95)	0.33	1.12 (0.26-4.95)	0.56
	Moderate	6240.0	8	3.91 (0.82-18.56)		3.44 (0.71-16.68)		2	0.32 (0.06-1.59)		0.45 (0.08-2.41)	
	Low	5925.0	2	1.00		1.00		6	1.00		1.00	

Model: Cox's proportional hazard model; adjusting for sex and age (model 1) and for sex, age, smoking habit, and serum levels of total cholesterol and ALT activity (model 2) HR (95%CI): hazard ratio (95% confidence interval) Total carotenes: the sum of α- and β-carotenes, and lycopene levels.

levels of RE and AT were not shown to be lower.

Discussion

In the present study cohort, serum levels of carotenoids, RE and AT were approximately similar to those seen in other populations (Ito et al., 1994), although serum carotenoid levels among Japanese do different from those of Caucasians (Ito et al., 1999). Validity of serum carotenoid determination was less than 15% CV for day to day variation with the applied assay method (Ito et al., 1990) but serum carotenoid levels tended to be similar to those estimated among the same individuals after 3 years (Ito et al., 2000). In addition, serum levels of carotenoids such as AC, BC, CR and ZL were lower among current smokers and regular alcohol drinkers, a finding also reported in a previous study (Ito et al., 1991). Furthermore, serum levels of carotenoids and tocopherols are associated with serum levels of total cholesterol, and serum enzyme activities reflecting liver function closely correlate, because serum carotenoids are carried by lipoprotein in the blood and metabolized in the liver (Bendich et al., 1987; Olsen, 1999). We estimated the risk of cancer deaths associated with these serum levels, controling for sex, age, smoking habit, and serum levels of total cholesterol and ATL activity, using Cox's proportional hazard model.

In the present study, analyses indicated that high serum levels of carotenoids such as AC, BC, LY, and TCR were significantly or marginally significantly associated with lower mortality of all sites and colorectal cancer, as in a previous report (Ito et al., 2002). However, no such clear relationships with lung cancer death were observed for these serum levels. In addition, although the subjects recruited in 1988 were not included, associations between lung cancer mortality and serum levels of CR, RE, and AT tended to be inverse between high and low levels, but without statistical significance. The relations between stomach cancer mortality and serum levels of carotenoids, including CR and ZL, were also similar.

Previous studies have demonstrated serum BC levels to be low in lung cancer cases (Comstock et al., 1997; Ito et al., 2003). In an earlier study of Japanese, there was no trend for decreased risk of lung and colorectal cancer of women

with the highest serum BC levels (>1.21 μmol/l). Although the mechanisms of carcinogenesis are complex, BC (which has particularly high provitamin A activity) is considered to be a crucial factor. The finding that BC has antioxidant activity and enhances immunity related to carcinogenesis also suggests that it can protect against oxidative stress such as damage to cell membranes and DNA caused by activated oxygen species and free radicals (Bendich, 1990). According to some reports, most carotenoids possess antioxidant activity (Gerster, 1992; McCall et al., 1999) and examples such as AC and BC can enhance cell-mediated immune responses (Hughes, 1999). This is consistent with the inverse association between cancer death of all sites and high serum levels of AC and BC observed among subjects in the present study. There have been reports indicating that higher serum levels of carotenoids other than BC obtained through high intake of vegetables and fruits are also associated with lower risk of lung cancer (Eicholzer et al., 1996; Yuan et al., 2001). High intake of vegetables and fruits can increase serum levels of carotenoids by about a few tenths of a percent, and bio-factors such as other carotenoids and vitamins (including vitamin B and C) appear to play a linked role in lowering of cancer incidence (Comstock et al., 1997). In contrast, intervention trials have found that high-dose administration of synthetic BC is associated with an increased incidence of lung cancer in smokers (ATBC Study Group, 1994) and industrial workers (Omenn et al., 1996). A trial conducted by American physicians also found no inverse association between synthetic BC administration and lung cancer incidence (Hennekens et al., 1996). A high dose of synthetic BC elevates serum BC levels more than 10-fold, and then produces prooxidant activities in biological systems (Palozza, 1998). It has been reported that synthetic BC administration also increases levels of cell proliferation indicators such as c-jun and c-fos proteins in the lungs of ferrets (Wolf, 2002). Thus, the available data suggest that high-dose administration of synthetic BC alone may be associated with an elevated risk of lung cancer (Wang et al., 1999).

Serum BC levels have been shown to reflect high intake of colored vegetables, and BC consumed together with fat is incorporated effectively into the body (Prince et al., 1993). We previously found that, after synthetic BC administration to our staff, serum levels of thiobarbituric acid–reactive substances (TBARS; a class of lipid peroxides) were significantly elevated and were more than 3-fold greater than serum BC levels (Ito et al., 1996). In the present study, the hazard ratios for lung cancer mortality of the subjects with the high serum BC levels may be due in part to prooxidant effects of BC, because synthetic BC has been readily available for intake from 1990 in Japan.

In the present study, the hazard ratios for cancer mortality of lung, colorectal, and stomach were lower for the subjects with higher serum CR levels, but this was not statistically significant. Mandarin juice, which is rich in CR, has chemo-preventive effects against mouse lung tumorigenesis (Kohno et al., 2001). There have furthermore been some reports that high serum levels of CR are associated with reduced risk of lung or colorectal cancer (Eicholzer et al., 1996; Yuan et al., 2001). In light of the available reports, our finding that serum CR levels demonstrate an inverse association with mortality of cancers such as lung or stomach cancer should be followed up, because of the unclear potential for application of this protective substance in prevention of cancer mortality.

In addition, some authors have provided evidence that serum levels of ZL and CR are not inversely associated with the risk for stomach cancer (Tubono et al., 1999; Yuan et al., 2004). However, while high serum levels of ZL did not appear to be lower risk for lung and colorectal cancer in the present study, they were associated with lower risk for stomach cancer. ZL possesses antioxidant activities and immune-promoting function (McCall et al., 1999; Hughes, 1999). In addition, the fine bioavailability of lutein, a major carotenoid in green-leaf vegetables, was reported to be 5 times higher than that of BC (Hof et al., 1999). Further studies, with larger populations and measurement of serum levels of a wider range of carotenoids is now needed for clarification.

In this study, no apparent associations, except for lung cancer, have been found between the risk of colorectal and stomach cancer and high serum levels of AT and RE. In nine population studies, only a few cases of lung cancer death had lower serum levels of RE and AT (Comstock et al., 1992). In a previous follow-up study of Japanese subjects, we found that higher serum RE levels were not significantly associated with mortality from cancer of all sites (Ito et al., 2002), a finding similar to the present results. The available evidence suggests that higher serum RE levels reduce the risk for lung cancer by preventing certain processes of carcinogenesis, although AT is an antioxidant substance (Ricciarelli et al., 2001).

In conclusion, the present results indicate that serum carotenoids such as AC, LY, and CR are associated with reduced risk of death from lung, colorectal and stomach cancer, especially in the colorectal case. In addition, the risk for lung cancer appears to be lower for the subjects exhibiting high serum levels of RE and AT. Of particular interest is that serum levels of carotenoids such as AC and LY may be particularly promising as biomarkers to predict mortality from colorectal and stomach cancer in rural inhabitants in Japan.

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