Serum Antioxidants and Skin Cancer Risk: An 8-Year Community-Based Follow-up Study

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Abstract

Background: **Antioxidant nutrients can help prevent skin damage caused by ultraviolet radiation from sunlight, but it is not clear whether serum concentrations of such nutrients influence skin cancer risk.**
Methods: **We carried out a prospective study of the associations between serum concentrations of antioxidant nutrients and incidence (person-based and tumor-based) of basal cell carcinoma (BCC) and squamous cell carcinoma (SCC) of the skin among a random subsample of 485 adults from an Australian community. Participants were divided into thirds, ranked according to their serum concentrations of carotenoids, α-tocopherol, and selenium measured in 1996 and were monitored for incident, histologically confirmed BCC and SCC tumors until 2004.**
Results: **Although there were no associations between baseline serum carotenoids or α-tocopherol concentrations and incidence of BCC or SCC, baseline serum selenium concentrations showed strong inverse associations with both BCC and SCC tumor incidence. Compared with participants with lowest selenium concentrations at baseline (0.4-1.0 μmol/L), those with the highest serum selenium concentrations (1.3-2.8 μmol/L) had a decreased incidence of BCC tumors (multivariate relative risk, 0.43; 95% confidence interval, 0.21-0.86; P_{trend} = 0.02) and SCC tumors (multivariate relative risk, 0.36; 95% confidence interval, 0.15-0.82; P_{trend} = 0.02).**
Conclusion: **Relatively high serum selenium concentrations are associated with an ~60% decrease in subsequent tumor incidence of both BCC and SCC, whereas serum concentrations of carotenoids or α-tocopherol are not associated with later skin cancer incidence. A possible U-shaped association between serum selenium concentrations and SCC of the skin needs confirmation.** (Cancer Epidemiol Biomarkers Prev 2009;18(4):1167–73)

Introduction

Basal cell carcinoma (BCC) and squamous cell carcinoma (SCC) are the most commonly occurring skin cancers in Caucasian populations. The costs of screening and treatment of these cancers continue to burden health systems of many countries around the world; thus, prevention could cause substantial public health and economic benefits (1-3).

The epidermis contains an array of antioxidants that help protect this outer layer of the skin against damage caused by exposure to ultraviolet radiation from sunlight, the main environmental cause of skin cancer. Ultraviolet radiation causes direct damage to DNA and the immune system (4, 5) and indirect damage through formation of free radicals such as reactive oxygen species (6). Carotenoids, α-tocopherol (vitamin E), and selenium (an important component of the antioxidant enzyme glutathione peroxidase) are found in the epidermis where they have been shown to protect against oxidative damage by neutralizing reactive oxygen species and other free radicals (7-11).

There is little consistent evidence about the relation between serum biomarkers of antioxidant nutrients and skin cancer risk in the general population. In a prospective study of antioxidant status and BCC and SCC risks, Dorgan et al. reported a positive association between serum lutein, zeaxanthin, and β-carotene and SCC risk in a 5-year follow-up study of skin cancer patients who participated in a drug trial in the United States (12). No such associations were found in another prospective study of persons who also all had a previous history of skin cancer (13). Results from a placebo-controlled trial, however, showed that selenium supplementation in persons with a history of skin cancer was associated with an increased incidence of BCC and SCC (14).

We have investigated associations between serum concentrations of antioxidant nutrients in a longitudinal study of a population sample of Australian adults whose sun exposure and skin cancer histories have been fully characterized.

Materials and Methods

Study Population. As part of the ongoing Nambour Skin Cancer Study, we conducted an 8-year prospective cohort study among adults who had been selected at

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random from the residents of the township of Nambour, a subtropical community in Queensland, Australia, for a baseline study of skin cancer (15). Between 1992 and 1996, 1,621 of these adults participated in a field trial to evaluate the role of a daily 30 mg β-carotene supplement and daily sunscreen use in skin cancer prevention (16, 17). In a two-by-two factorial design, participants were randomly allocated to taking daily either the β-carotene supplement or a placebo tablet (blinded) and daily sunscreen use or continuation of their usual sunscreen use habits (discretionary sunscreen use). At the end of the trial in 1996 (baseline of the current investigation), participants provided a blood sample, and during the course of the trial, they completed questionnaires on smoking habits, education, occupation, presence of selected medical conditions, and skin cancer risk factors such as skin color, tanning ability of the skin, and occupational and leisure-time sun exposure (17). During a physical examination in 1996, elastosis of the neck was recorded as a measure of long-term sun exposure history. Detailed descriptions of the community sample, the field trial, and its outcomes have been reported previously (16, 17).

Study participants were followed-up from 1996 until the end of 2004 to ascertain all occurrences of BCC and SCC. In the analyses, we considered all participants who provided a blood sample in 1996 and those who were randomized to the placebo tablets during the trial.

Ascertainment of skin cancers took place through an intensive surveillance system that had been set up during the Nambour trial and was continued during the complete post-trial follow-up period. Questionnaires were mailed twice yearly to participants and any reported skin cancers were confirmed through histologic reports. Independent pathology laboratories throughout Queensland provided pathology reports for all skin cancers diagnosed among study participants. These methods ensured virtually 100% ascertainment of histologically confirmed skin cancers in the study population (18).

There were no differences between the participants in the present study and the original 1,621 trial participants in terms of randomized sunscreen allocation during the trial, age, sex, education, occupation, smoking, use of dietary supplements, and skin cancer risk factors such as skin color, lifetime number of sunburns and other measures of sun exposure, and skin cancer history before 1996. This study was approved by the Ethics Committee of the Queensland Institute of Medical Research and all participants provided written informed consent.

Outcomes. We considered two outcomes in the analysis: (*a*) incidence of persons affected by a new basal or squamous cell cancer calculated as the number of persons affected by these cancers after the 1996 skin examination survey through December 31, 2004, divided by the person-years of follow-up accumulated between these dates and expressed per 100,000 person-years, and (*b*) incidence of basal or squamous cell tumors during the same person-years follow-up time as calculated for the person-based analysis. Tumors diagnosed during the 1996 skin survey were not included in the analyses to exclude disease that already existed at baseline. Tumors and person-years of follow-up were counted until date of withdrawal from the study, date of death, or December 31, 2004, whichever came first.

Serum Biomarkers. In 1996, nonfasting venous blood samples of 30 mL were collected using standard venipuncture techniques done by experienced phlebotomists. Blood was not taken from subjects who had consumed more than a light breakfast (e.g., toast or cereal, no cooked breakfast). Blood samples were processed at the time of collection and serum samples were stored in ~1 mL aliquots at -70°C until analysis. Measurements of carotenoids, α-tocopherol, selenium, and cholesterol were conducted by Queensland Health Pathology Services at the Royal Brisbane Hospital. Measurements of carotenoids and α-tocopherol were conducted simultaneously by high-performance liquid chromatography using the method of Sowell et al. (19). Total concentrations of lutein and zeaxanthin have been presented as a combined estimate because these carotenoids were not separated in the high-performance liquid chromatography analysis. Total cholesterol was measured using an enzymatic colorimetric test (20). Serum selenium was analyzed by atomic absorption spectrometry using a graphite furnace and Zeeman background correction (21). Coefficients of variations of pooled serum measured in duplicate in every run (n = 21) were α-carotene 10.7%, β-carotene 9.7%, β-cryptoxanthin 12.1%, lutein and zeaxanthin 17.3%, α-tocopherol 4.7%, selenium 9.8%, and cholesterol 2.6%. Due to an error in the automated reading of chromatograms, results for lycopene serum concentrations could not be included.

Stability of these analytes over time was investigated in 45 participants of this study population in 1992 to 1993. This was done in participants who received the placebo and thus not the β-carotene supplement. Spearman correlation coefficients (r) of serum concentrations measured in two blood samples that were taken 16 months apart were α-carotene 0.72, β-carotene 0.70, β-cryptoxanthin 0.60, lutein and zeaxanthin 0.85, and α-tocopherol 0.63.

Statistical Models. Serum carotenoids and α-tocopherol were adjusted for serum cholesterol using the residual method to account for variations in serum carotenoids and α-tocopherol that are due to variations in serum lipids (22). Distributions of the biomarkers were identified as skewed and variables were log transformed to improve normality before calculation of the residuals. Tertiles were calculated for each biomarker and used as cut points for grouping. For person-based analysis, relative risks (RR) with 95% confidence intervals (95% CI) for increasing concentrations of the biomarker compared with the lowest group were derived from generalized linear models specifying Poisson distribution with a robust error variance (23) and person-years of follow-up as offset. For tumor-based analyses, RR (95% CI) were derived using generalized linear models with negative binomial distribution. The negative binomial distribution has been recommended for analyzing nonnegative integer data with variance greater than the mean (24) and provided the best fit to our tumor-count data.

We first applied models controlling for age and sex. The expanded multivariate models also controlled for usual time spent outdoors on weekdays, history of skin cancer before 1996, pack-years of smoking until 1996, and alcohol intake (continuous variable). These confounders were selected based on their association with the

exposure and outcome variables and on previous studies of diet and skin cancer. Covariates were retained in the model if they changed the risk estimate by >10%, whereas age and sex were retained in all multivariate models. There was no additional confounding by sunscreen treatment allocation during the trial, skin color, tanning ability of skin, elastosis of the neck, number of painful sunburns during life, other indicators of past sunlight exposure, or use of dietary supplements (yes/no).

To test for linear trends, we assigned the median to each tertile group and modeled these values as a continuous variable for each biomarker. All analyses were done with SAS statistical software (version 9.1; SAS Institute). All reported P values are two-sided.

Results

Serum antioxidants were measured in 485 participants. In the 8-year follow-up of these participants, a total of 173 histologically confirmed new BCC tumors were diagnosed in 77 participants during 3608 person-years of follow-up (tumor-based incidence: 4,795/100,000; person-based incidence: 2,134/100,000). For SCC, a total of 124 histologically confirmed new tumors were diagnosed in 59 participants during the same person-years of follow-up (tumor-based incidence: 3,437/100,000; person-based incidence: 1,635/100,000). In general, participants who had either an incident SCC or BCC in the follow-up period were more likely ($P < 0.05$) to be male, to be older, to have fair skin, and to have a history of skin cancer than those who did not develop a skin cancer (Table 1). Those who developed SCC were also more likely ($P < 0.05$) to have a tendency to sun-burn and to have smoked, but those who developed BCC did not differ by these characteristics compared with participants who did not develop BCC. The number of painful sunburns during life was not associated with developing BCC or SCC in this study population (although borderline significant for SCC; $P = 0.06$).

There were no associations between serum carotenoids or α-tocopherol concentrations and person-based incidence of BCC or SCC (Tables 2 and 3). However, participants with the highest compared with the lowest selenium concentrations appeared less likely to be affected by BCC (highest versus lowest group multivariate-adjusted RR, 0.58; 95% CI, 0.32-1.07; $P_{trend} = 0.08$) and SCC (highest versus lowest group multivariate-adjusted RR, 0.49; 95% CI, 0.24-0.99; $P_{trend} = 0.05$; Table 4). The tumor-based analyses confirmed and strengthened these inverse associations, such that participants with the highest compared with the lowest selenium concentrations had a substantially decreased incidence of BCC (highest versus lowest group multivariate-adjusted RR, 0.43; 95% CI, 0.21-0.86; $P_{trend} = 0.02$) and SCC (highest versus lowest group multivariate-adjusted RR, 0.36; 95% CI, 0.15-0.82; $P_{trend} = 0.02$; Table 4). None of the other serum concentrations showed associations with skin cancer incidence in the tumor-based analyses (results not shown).

Discussion

In this prospective community-based study of Australian adults, those with relatively high serum selenium concentrations at baseline had decreased incidence rates of both BCC and SCC tumors in the 8-year follow-up period. Previously, few, if any, nutrients have been shown to influence BCC occurrence (in contrast to SCC).

Table 1. Characteristics by skin cancer status of 485 participants of the Nambour Skin Cancer Study, 1996-2004

	SCC			BCC		
	Yes (n = 59), n (%)	No (n = 426), n (%)	P*	Yes (n = 77), n (%)	No (n = 408), n (%)	P*
Sex						
Male	38 (64)	185 (43)	0.002	46 (60)	177 (43)	0.008
Female	21 (36)	241 (57)		31 (40)	231 (57)	
Age						
Age (mean) in 1996 (y)	63 y	54 y	<0.0001	61 y	54 y	<0.0001
Skin color						
Fair	41 (69)	226 (53)	0.04	53 (69)	214 (53)	0.04
Medium	15 (25)	172 (41)		19 (25)	168 (41)	
Olive	3 (5)	27 (6)		5 (6)	25 (6)	
Propensity to burn/tan after acute sun exposure						
Always burn	20 (34)	77 (18)	0.004	21 (27)	76 (19)	0.12
Burn then tan	36 (61)	299 (70)		49 (64)	286 (70)	
Tan only	3 (5)	49 (12)		7 (9)	45 (11)	
Painful sunburns during life						
None	14 (25)	74 (18)	0.06	19 (25)	69 (17)	0.33
1 Burn	12 (21)	58 (14)		7 (9)	63 (16)	
≥2 Burns	31 (54)	285 (68)		49 (65)	267 (67)	
Smoking status in 1996						
Never	22 (37)	236 (55)	0.005	36 (47)	222 (55)	0.12
Current	5 (8)	38 (9)		5 (6)	38 (9)	
Ex	32 (54)	152 (36)		36 (47)	148 (36)	
History of skin cancer before 1996						
No	17 (29)	309 (73)	<0.0001	19 (25)	307 (75)	<0.0001
Yes	42 (71)	117 (27)		58 (75)	101 (25)	

*P from Mantel-Haenzel χ^2 test (categorical data) or ANOVA (continuous data).

These results reflected the inverse associations observed between serum selenium concentrations and overall incidence of persons newly affected by BCC and SCC, although mostly these associations did not reach statistical significance. This apparently protective effect of selenium on skin cancer incidence is consistent with *in vivo* and *in vitro* studies that have shown that topical and oral selenium can provide protection against ultraviolet-induced sunburn, tanning, and skin cancer (7, 25-27). Previous human data are sparse and conflicting, however. A clinic-based study showed lower plasma selenium concentrations in patients with SCC or BCC compared with controls (28), whereas other studies found no associations (13, 29). Notably, our observations are in apparent contrast to the results of a selenium supplementation trial in which a daily 200 μg selenium supplement was associated with an increased risk of SCC after 13 years of follow-up (hazard ratio for selenium supplement versus placebo, 1.25; 95% CI, 1.03-1.51; $P = 0.03$; refs. 14, 30). The authors reported that the increased risk of SCC in their trial was greatest among participants with the highest baseline concentrations of plasma selenium (>105.6 ng/mL or 1.3 μmol/L). These concentrations were equivalent to those in the highest serum selenium group in our study population, in which we found a protective association. It is possible that selenium supplementation in people who already have adequate levels of this nutrient increases rather than decreases skin cancer risk. Such unintended adverse effects have also occurred in supplementation trials of β-carotene (31, 32). Furthermore, the investigators of the selenium supplementation trial have indicated that ~60% of the trial participants had punctate keratoses of the palms, characteristic of arsenic exposure (14). Selenium can enhance the toxic effects of arsenic by increasing its retention in tissues and by suppressing its methylation (14, 33), although the risk of premalignant skin lesions in arsenic-exposed populations appears to be higher among persons with lower than average whole-blood selenium concentrations (34). Thus, the generalizability of the findings of the selenium supplementation trial is not clear. No arsenic exposure-related lesions have been seen in our study population who had no unusual background exposures.

The majority of our study participants had selenium concentrations in the normal range (0.5-2.5 μmol/L) including those in the lowest group. The selenium content of serum and plasma is generally comparable and both respond to short-term changes in dietary selenium intake (35, 36). A general cancer-preventive effect of selenium supplementation has been described in persons who were initially in the lowest two tertiles of serum selenium concentrations within their study population (<121.6 ng/mL or 1.6 μmol/L; ref. 37). Furthermore, experimental study has shown that the selenium plasma concentration needed to achieve optimal platelet glutathione peroxidase activity lies between 1.3 and 1.5 μmol/L (38). In our study, such concentrations were achieved only by the participants in the highest group of serum concentrations, for whom the observed protection against SCC was strongest.

In previous analysis of nutrient intake in this study population, selenium intake per se was not associated with BCC or SCC incidence (39), and correlations

Table 2. RR (95% CI) of BCC by group of serum concentration of antioxidants at baseline, person-based analyses in 485 participants of the Nambour Skin Cancer Study, 1996-2004

Serum biomarker	Biomarker concentration			P_{trend}
	Group 1	Group 2	Group 3	
α-Carotene				
Median (μmol/L; min-max)	0.05 (0.01-0.07)	0.11 (0.08-0.15)	0.22 (0.16-1.04)	
No. tumors/participants	18/153	35/171	24/161	
Basic RR (95% CI)*	1.00	1.87 (1.05-3.30)	1.33 (0.72-2.46)	0.91
Multivariate RR (95% CI) †	1.00	1.70 (0.95-3.03)	1.28 (0.68-2.41)	0.69
β-Carotene				
Median (μmol/L; min-max)	0.30 (0.02-0.44)	0.59 (0.45-0.79)	1.10 (0.80-5.10)	
No. tumors/participants	23/163	30/159	24/163	
Basic RR (95% CI)*	1.00	1.33 (0.77-2.30)	1.06 (0.59-1.90)	0.99
Multivariate RR (95% CI) †	1.00	1.30 (0.74-2.30)	1.07 (0.59-1.96)	0.95
β-Cryptoxanthin				
Median (μmol/L; min-max)	0.10 (0.02-0.16)	0.29 (0.18-0.42)	0.73 (0.43- 3.20)	
No. tumors/participants	23/159	30/168	24/158	
Basic RR (95% CI)*	1.00	1.10 (0.64-1.90)	0.90 (0.50-1.63)	0.63
Multivariate RR (95% CI) †	1.00	1.07 (0.61-1.87)	0.91 (0.50-1.67)	0.68
Lutein and zeaxanthin				
Median (μmol/L; min-max)	0.20 (0.04-0.28)	0.37 (0.29-0.47)	0.64 (0.48-1.60)	
No. tumors/participants	26/162	21/158	30/165	
Basic RR (95% CI)*	1.00	0.85 (0.48-1.51)	1.05 (0.62-1.77)	0.78
Multivariate RR (95% CI) †	1.00	0.81 (0.45-1.46)	1.06 (0.62-1.83)	0.71
α-Tocopherol				
Median (μmol/L; min-max)	24.0 (9.0-27.0)	30.0 (28.0-33.0)	38.5 (34.0-130.0)	
No. tumors/participants	22/163	29/158	26/164	
Basic RR (95% CI)*	1.00	1.12 (0.63 1.98)	0.90 (0.50-1.65)	0.65
Multivariate RR (95% CI) †	1.00	0.88 (0.49-1.56)	0.95 (0.51-1.75)	0.92

*Adjusted for age, sex, and allocation of a β-carotene supplement during the Nambour trial. All RRs from multivariate Poisson regression.
† Adjusted for age, sex, pack-years of smoking, β-carotene supplement allocation during the trial, alcohol intake (continuous), time spent outdoors on weekdays, and history of skin cancer before 1996.

Table 3. RR (95% CI) of SCC by group of serum concentration of antioxidants at baseline, person-based analyses in 485 participants of the Nambour Skin Cancer Study, 1996-2004

Serum biomarker	Biomarker concentration			P_{trend}
	Group 1	Group 2	Group 3	
α-Carotene				
Median (μmol/L; min-max)	0.05 (0.01-0.07)	0.11 (0.08-0.15)	0.22 (0.16-1.04)	
No. tumors/participants	19/153	26/171	14/161	
Basic RR (95% CI)*	1.00	1.28 (0.71-2.32)	0.74 (0.37-1.49)	0.59
Multivariate RR (95% CI) †	1.00	1.30 (0.70-2.39)	0.71 (0.35-1.45)	0.49
β-Carotene				
Median (μmol/L; min-max)	0.30 (0.02-0.44)	0.59 (0.45-0.79)	1.10 (0.80-5.10)	
No. tumors/participants	20/163	22/159	17/163	
Basic RR (95% CI)*	1.00	1.09 (0.59-2.01)	0.88 (0.45-1.69)	0.64
Multivariate RR (95% CI) †	1.00	1.08 (0.57-2.03)	0.92 (0.47-1.81)	0.78
β-Cryptoxanthin				
Median (μmol/L; min-max)	0.10 (0.01-0.17)	0.29 (0.18-0.42)	0.73 (0.43- 3.20)	
No. tumors/participants	16/159	22/168	21/158	
Basic RR (95% CI)*	1.00	1.18 (0.62-2.25)	1.11 (0.56-2.16)	0.87
Multivariate RR (95% CI) †	1.00	1.16 (0.60-2.24	1.17 (0.59-2.33)	0.70
Lutein and zeaxanthin				
Median (μmol/L; min-max)	0.20 (0.04-0.28)	0.37 (0.29-0.47)	0.64 (0.48-1.60)	
No. tumors/participants	17/162	21/158	21/165	
Basic RR (95% CI)*	1.00	1.34 (0.71-2.55)	1.04 (0.55-1.98)	0.96
Multivariate RR (95% CI) †	1.00	1.26 (0.66-2.42)	1.01 (0.53-1.93)	0.91
α-Tocopherol				
Median (μmol/L; min-max)	24.0 (9.0-27.0)	30.0 (28.0-33.0)	38.5 (34.0-130.0)	
No. tumors/participants	19/163	20/158	20/164	
Basic RR (95% CI)*	1.00	0.84 (0.44-1.60)	0.76 (0.39-1.47)	0.44
Multivariate RR (95% CI) †	1.00	0.78 (0.41-1.50)	0.85 (0.43-1.65)	0.68

*Adjusted for age and sex. All RRs from multivariate Poisson regression.
† Adjusted for age, sex, pack-years of smoking, alcohol intake (continuous), time spent outdoors on weekdays, and history of skin cancer before 1996.

between intake and serum concentrations were very weak ($r = 0.09$; $P < 0.01$). None of our study participants have reported use of selenium-containing supplements. Due to large variations in food selenium content, serum selenium concentration is a better indicator of selenium status than estimates of dietary intake, although correlations with functional indicators such as glutathione peroxidase are progressively weaker for blood concentrations >1.0 μmol/L (40). Globally soil levels are highly variable and this is also the case within Australia. The selenium content of the foods is known to vary up to 5-fold between states but also can vary up to 5-fold within states (41). Furthermore, the food supply in Australia is very mobile, and produce grown in a region tends to get distributed to many different areas of the country. Thus, dietary intake is generally not strongly

Table 4. RR (95% CI) of BCC and SCC by group of serum selenium concentration at baseline in 485 participants of the Nambour Skin Cancer Study, 1996-2004

	Selenium concentration			P_{trend}
	Group 1	Group 2	Group 3	
Median (μmol/L*; min-max)	0.9 (0.4-1.0)	1.1 (1.1-1.2)	1.4 (1.3-2.8)	
BCC: person-based				
No. persons affected/total participants	33/194	28/163	16/128	
Basic RR (95% CI) †	1.00	1.02 (0.61-1.69)	0.67 (0.37-1.22)	0.19
Multivariate RR (95% CI) ‡	1.00	0.97 (0.58-1.61)	0.58 (0.32-1.07)	0.08
BCC: tumor-based				
No. tumors/total participants	20/163	22/159	17/163	
Basic RR (95% CI) †	1.00	1.09 (0.56-2.10)	0.57 (0.28-1.17)	0.14
Multivariate RR (95% CI) ‡	1.00	1.02 (0.56-1.87)	0.43 (0.21-0.86)	0.02
SCC: person-based				
No. persons affected/total participants	27/194	21/163	11/128	
Basic RR (95% CI) †	1.00	0.92 (0.52-1.63)	0.54 (0.27-1.08)	0.08
Multivariate RR (95% CI) ‡	1.00	0.89 (0.49-1.60)	0.49 (0.24-0.99)	0.05
SCC: tumor-based				
No. tumors/total participants	17/162	21/158	21/165	
Basic RR (95% CI) †	1.00	0.97 (0.48-1.95)	0.44 (0.19-1.00)	0.06
Multivariate RR (95% CI) ‡	1.00	0.86 (0.44-1.67)	0.36 (0.15-0.82)	0.02

*1 μmol/L ≈ 79 μg/L.
† Adjusted for age and sex. RRs from Poisson regression (person-based) or negative binomial regression (tumor-based).
‡ Adjusted for age, sex, pack-years of smoking, alcohol intake (continuous), time spent outdoors on weekdays, and history of skin cancer before 1996.

dependent on local selenium soil concentrations, although direct evidence for that is lacking.

Serum selenium was the only biomarker of selenium status that was measured in our study; toenail or erythrocyte selenium concentrations are better indicators of long-term selenium status (34). Although in a previous nested case-control study of BCC in this population we found no associations between serum concentrations of selenium (or carotenoids or α-tocopherol) and BCC (42), the previous study was based only on cases identified after a 4.5-year follow-up period, which would explain the different earlier findings.

A major strength of this study was its prospective nature and the extensive characterization of past sun exposure and of skin cancer occurrence in all participants. Our study was based on analysis of histologically confirmed BCC and SCC, which were ascertained through an extensive surveillance system. We consider any bias due to misclassification of participants due to misdiagnosis or missed diagnosis of skin cancer very unlikely. Any confounding effect of the β-carotene supplement taken during the Nambour field trial that preceded this study was ruled out by including only those who received placebo treatment in the present analyses. Although only one measurement of serum antioxidant concentrations was available, in a randomly selected subgroup of our study population, repeated measurements of serum antioxidant concentrations were very stable ($r \geq 0.60$). Furthermore, serum concentrations of antioxidants have shown high correlations with concentrations in the skin, with both a single-blood analysis and the average of repeated blood analyses (43). One third of the participants had already had skin cancer before the start of the study. The results from all multivariable models were adjusted for and are thus independent of the presence or absence of a skin cancer history before baseline.

A drawback in our study was the high coefficient of variation in our serum measurements of lutein and zeaxanthin, which may have caused dilution of associations with skin cancer incidence. We measured serum concentrations of carotenoids, α-tocopherol, and selenium because of their possible antioxidant properties. However, the antioxidant capacity of human tissues is influenced by a wide variety of nutrients and other factors (44). Thus, the effect of the antioxidants measured in this study on the overall antioxidant defense capabilities of the skin may be limited. Although serum selenium concentrations were not included in our study of stability over time, food is the main source of selenium, so we expect that the stability of selenium concentrations was as high as that of the other five measured analytes, indicating the generally stable overall dietary intake of our study population. We tested a large number of associations in this study; thus, caution needs to be exercised in its interpretation due to the possibility of chance findings. However, our observation that selenium concentrations specifically showed inverse associations with both BCC and SCC tumor incidence, and moreover these reflected the results based on people newly affected in the study population at large, suggest that chance is an unlikely explanation for this finding.

From this population-based prospective study, we conclude that although concentrations of α-carotene, β-carotene, β-cryptoxanthin, lutein and zeaxanthin, and α-tocopherol appear not to be associated with skin cancer incidence, relatively high serum selenium concentrations at baseline are associated with ~60% lower incidence of BCC and of SCC tumors in this population. The inverse association with BCC is especially salient given the dearth of existing evidence about the possible dietary links of BCC. Given the results of a previous intervention trial, our findings suggest that a U-shaped relationship between serum selenium concentrations and cutaneous SCC may exist. Evidence from other populations, particularly those that include persons with relatively low serum selenium concentrations, is needed to confirm this.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

Acknowledgments

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