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Breast cancer and dietary and plasma concentrations of carotenoids and vitamin A

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KEY WORDS Carotenoids, vitamin A, breast cancer, diet, plasma, benign breast disease, epidemiology

Introduction

The roles of vitamin A and carotenes in carcinogenesis have been investigated in a large number of experimental systems (I). Supplemental retinyl acetate in animal diets may cause either a lower (2-8) or higher (9) incidence of mammary tumors whereas supplemental fj-carotene in the diet was shown to inhibit mammary tumors (10, 1 1) and transplantable tumors (12).

Epidemiologic studies have indicated an inverse association between plasma retinol and risk of cancer of all sites (13, 14), with only suggestive evidence for this association in breast cancer (15). Three studies with dietary data (16-18) and three studies with plasma data (19-21) showed no association between breast cancer and retinol whereas one study with plasma showed a positive association (16).

An inverse association was observed between breast cancer and estimates of dietary intake of carotenes (17) and vegetables (22). One dietary study (18) showed no association whereas other dietary studies (23, 24) reported an inverse association between breast cancer and total vitamin A intake, with contri-

butions from both preformed (retinol) and precursor (carotenes) vitamin A. Although results from some plasma studies (16, 20) indicated no association between breast cancer and fJ· carotene or total carotenoids. respectively, results from other blood studies (19, 25. 26) showed a nonsignificant inverse association between breast cancer and fj-carotene.

Overall, previous studies indicate either no association or a direct association between breast cancer and retinol and no association or an inverse association between carotenes and breast cancer. Some studies were not specifically designed to investigate breast cancer and suffered from inadequate power (19-21. 25) whereas others did not control for important breast cancer risk factors (20. 21, 23, 25. 26) or plasma lipids (19. 21, 25, 26). These limitations may have diminished the ability to observe the effects of nutrients on cancer risk.

This case-control study was designed to assess the independent and interactive effects of preformed vitamin A and carotenoids in the plasma and in the diet, controlling for other risk factors and other nutrients.

Methods

Between September 1985 and September 1986, study subjects were enrolled from the breast clinic at Roswell Park Memorial Institute and from the offices of two private surgeons in Buffalo. Eligible patients were women aged 30-80 y who were being evaluated for a breast mass but who had no previous history of cancer. All participants had lived in upstate New York or Pennsylvania for I y. After subjects were scheduled for a diagnostic biopsy, informed consent was obtained and they were given a questionnaire and directions for the blood-collec-

tion procedure. The study was approved by the Clinical Investigations Committee at Roswell Park Memorial Institute and by Institutional Review Boards at the State University of New York at Buffalo and the Buffalo General Hospital. Thirty-four patients who were referred to the specialists because of a suspicious mammogram or lump but were determined not to have masses requiring biopsy were included in the control group.

Patients were classified into one of three groups on the basis of their pathology reports from the breast biopsies: subjects with breast cancer *(n = 83)*, control subjects *(n = 1 13)*, or excluded *(n = 40)*. Women with breast cancer had cancers classified as 7% in situ, 32% stage I, 46% stage II, and 15% stages III or IV. The 1 13control subjects were those 79 women who were found to have benign lesions on biopsy , plus the 34 women who did not require a biopsy . The 40 patients with biopsy reports that described lesions and who were thought to be at increased risk of breast cancer (predominantly of an atypical hyperplastic nature) were excluded from the control group.

The self-administered questionnaire requested information about the subjects' dietary practices and medical history. A food frequency table with five frequency categories (never, rarely, 1-3 times/mo, 1-4 times/wk, and 5-7 times/wk) contained a list of 30 food items that previous analysis indicated would account for 90% of the variance associated with estimated vitamin A intake (27). Standard portion sizes were assumed to convert the frequency data into estimates of nutrient intake.

Fasting blood samples were drawn from subjects before their biopsies were taken or on a designated day for the 34 patients who did not have biopsies taken . The samples were centrifuged at 2000 X *g* for 12 min and plasma was preserved in sodium ascorbate (I 0 mg/mL plasma) and frozen at -80 °C until the end of the study. Blood was stored from *5* to 17 mo; storage duration was similar for women with breast cancer and control subjects. In addition, there were no trends observed concerning nutrient concentrations with length of storage. Blood specimens were thawed once for portioning of samples and once for the laboratory analysis. Retinol and carotenoids were analyzed by HPLC, retinol by a modification of a standard method (28), and carotenoids by a modification of a new method (29).

Statistical analysis

Statistical analyses were conducted with *SYSTAT* (version 3.0, Systat, Inc, Evanston, IL) and the supplementary logit program on an AT&T 6300 personal computer. Differences in means were tested with the Student's *t* test and differences in proportions were tested by logit analysis. In the latter analyses, case status was the independent variable, the breast-cancer risk factor was the dependent variable, and other breast-cancer risk factors were added to the model as needed. Nutrients were log-transformed and categorized into quartiles on the basis of their distribution in the control subjects. Multivariate analyses used logit analysis with case status as the dependent variable; there were three possible outcome categories but only the results for subjects with breast cancer vs control subjects are presented here. In the multivariate analyses the nonnutrient, breast-cancer risk factors used as covariates were age, age at menarche, age at menopause, age at first birth, parity, menopausal status, family history of breast cancer in a first-degree relative, Quetelet index (kg/m2), marital status, and income. Analyses

TABLE I

Descriptive statistics of potential confounding variables for subjects with breast cancer and control subjects•

	Breast-cancer patients *(n* = 83)	Control subjects *(n* = 1 13)
Age (y)	57.78 ± 1 1.0lt	50.47 ± I0.81
Weight (kg)	69.79 ± 13.49	67.85 ± 12.00
Height (cm)	164.75 ± 7.33	165.19 ± 7.93
Quetelet index (kg/m2)	25.82 ± 5.16	24.89 ± 4.52
Age at menarche (y)	12.83 ± 1.30	12.70 ± 1.57
Age at first birth (y)	23.43 ± 3.95	23.37 ± 4.29
Parity	3.03 ± 1.36	3.36 ± 1.63
Age at menopause (y)	47.66 ± 6.27	47.02 ± 5.92
Duration of breast-feeding (mo)t	5.21 ± 9.01	6.70 ± I0.47
Menopausal (%)§	66	57
Surgical menopause (%)11	40	33
Family history of breast cancer (%)	28	39
Married (%)	71	87
Never married (%)	13	9
Widowed (%)	16	4
Nulliparous (%)	16	17
History ofBBD (%)	29	40
Two highest income groups (%)"*	29	42

•*X* ± SD. BBD, benign breast disease.

t Significantly different from control subjects, *P* < 0.00 I .

* Total months in lifetime spent breast-feeding.

§ Percent of diagnostic group.

U Percent of those menopausal.

Self-reported history of a previous diagnosis of BBD.

•• Two highest of seven income categories.

of the plasma retinol and carotenoid variables also controlled for plasma lipids to evaluate the effect of the nutrient of interest after differences in lipid status were eliminated. In addition, both triglycerides and cholesterol were associated with case status and therefore were potential confounding variables. Tests for trend used the continuous, log-transformed nutrient variable. The models and variables were assessed by use of the log likelihood ratio test and all significant predictors of case status were included in the full models, as well as the nonnutrient, breast-cancer risk factors.

Results

The characteristics of the subjects with breast cancer and control subjects at baseline are presented in Table 1. Subjects with breast cancer were significantly older than the control subjects but were not statistically different from control subjects in any of the other potentially confounding variables. Subjects with breast cancer had significantly lower P-carotene and lycopene values than did control subjects *(P* < 0.01 and *P* < 0.05, respectively) (Table 2); however, they did not have different mean values for retinol, a-carotene, or dietary estimates of vitamin A intake.

Multivariate analysis showed that low P-carotene was associated with an increased risk of breast cancer, with and without adjustment for plasma lipids (Table 3). Further analyses showed that the inverse association between P-carotene and breast cancer was restricted to the postmenopausal cases (test for trend *P* < 0.00 I). Alpha-carotene was not associated with breast-cancer risk (Tables 2 and 3), and low concentrations of

TABLE 2
Comparison of plasma and dietary variables among subjects with breast cancer and control subjects*

	Breast-cancer patients (n = 81)	Control subjects (n = 101)
Plasma		
Retinol (μmol/L)	1.95 ± 0.62	1.93 ± 0.44
P-carotene (μmol/L)	0.21 ± 0.16	1.26 ± O.t 5t
a-carotene (nmol/L)	57 ± 41	64 ± 37
Lycopene (nmol/L)	417 ± 188	491 ± 238t
Diet		
Total vitamin A (IU/d)	1 1 900 ± 6 800	12 300 ± 6 800
Vegetable vitamin A (IU/d)	I770 ± 2 400	1 900 ± 2 900

*x ± SD.
tt Significantly different from cases: tP < 0.01, *tP* < 0.05.

lycopene appeared to be associated with increased risk of breast cancer before controlling for plasma lipids. Lycopene was not associated with case status after controlling for these lipids.

The main effect of plasma retinol values indicated no association with breast-cancer risk (Table 3); however, a statistically significant interaction with plasma P-carotene (Table 4) showed a trend of increasing risk for breast cancer with increasing retinol within the low P-carotene quartile. The wide confidence intervals indicated few individuals with higher retinol and low P-carotene; even so, the results were statistically sig

nificant. Although the other confidence intervals included unity, the trends indicated that within the other P-carotene quartiles, risk decreased as retinol increased.

Multivariate analyses of the dietary data indicated no association between breast cancer and either total or vegetable vitamin A intakes (Table 5). There was no statistically significant interaction between total vitamin A and vegetable vitamin A.

Total dietary vitamin A was not correlated with plasma retinol *(r* = 0.1 1) or P-carotene *(r* = 0.16), nor was dietary vitamin A from vegetable sources correlated with plasma P-carotene *(r* = -0.02). Plasma a-carotene, however, was marginally correlated with total dietary vitamin A *(r* = 0.26).

Discussion

Control group

Many of the known breast-cancer risk factors were not more prevalent among subjects with breast cancer than among control subjects. Therefore, these control subjects would appear to be at high risk for breast cancer, according to known risk-factor classification for breast cancer, if they were compared with women in the general population. Thus, this study compared women with breast cancer with a group who had similarly high risk but who did not have breast cancer.

There are two possible biases of concern with this control group. The results may be biased towards a false-positive finding if the breast-cancer risk factors prevalent in this control group were also associated with nutrient status (eg, high carot-

TABLE 3
Odds ratios (subjects with breast cancer vs control subjects) for plasma nutrient quartiles with adjustments for potential confounding variables with and without lipid adjustment*

	Quartilest				
Nutrient	QI	Q2	Q3	Q4	*pt*
Lipid unadjusted					
P-Carotene	4.42§	t.90	1.41	1.00	<0.01
	(1.33. 14.7)11	(0.55, 6.55)	(0.43, 4.64)		
a-Carotene	2.69	1.12	0.96	1.00	0.18
	(0.88. 8.23)	(0.36, 3.51)	(0.31.2.99)		
Lycopene	2.40	2.45	1.55	1.00	0.09
	(0.83, 6.95)	(0.87, 6.93)	(0.51, 4.72)		
Retinol	I.II	1.21	0.64	1.00	0.67
	(0.38, 3.26)	(0.41. 3.55)	(0.23. 1.83)		
Lipid adjusted					
P-Carotene	3.15	1.79	1.18	1.00	0.02
	(0.90, I 1.04)	(0.50, 6.39)	(0.34, 4.09)		
a-Carotene1!	0.63	0.45	0.61	1.00	0.19
	(0.13, 2.95)	(0.1 I, 1.80)	(0.18, 2.06)		
Lycopene	t.60	1.94	1.39	1.00	0.43
	(0.50, 5.18)	(0.64, 5.88)	(0.45. 4.32)		
Retino111	1.06	1.01	0.60	1.00	0.41
*					
	(0.31. 3.64)	.32, 3.25)	(0.18, I.97)		

*Full model included age, age at first birth, family history, age at menarche, Quetelet index, parity. age at menopause, income, and marital status. Lipids include plasma cholesterol and triglyceride.
t Qt is low and Q4 is high.
t Significance of trend.
§Odds ratio of quartile compared with the fourth quartile.
II 95% confidence limits.
11 Full model also contained P-carotene.

TABLE 4
Case-control adjusted odds ratios for the retinol, by ,8-carotene interaction•

Retinol level[t]	{:I-carotene quartiles			
	Q1	Q2	Q3	Q4
Low (25th percentile)	3.41 (0.75. 15.6)§ (9.7)ll	4.07 (0.90. 18.5) (5.5)	3.64 (0.66. 20.1) (5. 3)	3.85 (0.84. 17.6) (4, 4)
Median (50th percentile)	5.50 (1.30. 23.2) (14. 14)	2.90 (0.71. 1 1.9) (9. 14)	1.92 (0.45. 8.21) (9. 14)	1.92 (0.92, 4.00) (4. 10)
High (75th percentile)	8.58 (1.82. 40.5) (12. 4)	2.11 (0.48. 9.30) (3. 6)	1.06 (0.23. 4.91) (4.8)	1.00 (3.8)

• Odds ratios adjusted for cholesterol, triglyceride . tl<arotene. the retinol-by- tl-carotene interaction. age. age at first birth, family history. age at menarche. Quetelet index. parity. age at menopause, income, and marital status.
t Percentile based on ranking of all study subjects. Low, 0.81 µmol/L; median.
0.98 µmol/L: high. 1 .17 µmol/ L.
t QI is low and Q4 is high.
§ 95% confidence limit.
ll Number of cases. control subjects.
Reference group.

enoid consu mption). No evidence was found supporting an association between the benign breast disease prevalent in this control group and vitamin A or carotenoids (30), and it is unlikely that other components of the high-risk classification (eg, age at menarche, age at first birth) are related to these nutrients. In contrast. the control subjects were very similar to subjects with breast cancer, by risk-factor classification which would bias the results towards a false-negative finding of no association. This is only of concern with the findings for a-carotene and lycopene. which were nonsignificant , but is not of concern with findings for ,B-carotene and retinol, which were significant. Indeed , the similarity between the groups may be a strength of the study because the analysis focused on factors that allowed control subjects to be free of cancer even though they were very similar to subjects with breast cancer in terms of other risk factors. A difference in nutritional status between these two groups would be of considerable interest because it would suggest a nutritional advantage among the control subjects very late in the promotional stages of the disease process. These results not only indicated that such a difference existed between these groups but also that the difference was in the same direction (low ,B-carotene, higher risk) as that frequently observed in earlier etiologic stages.

Plasma ,B-carotene

The concentrations of carotenoids in this study were relatively low compared with those in similar groups of women in the United States (31, 32). Quality control plasma samples were exchanged between laboratories, which determined that our assay yielded values approximately one-half the values obtained by other standard methods (33, 34) because of lower extraction efficiency of the solvent. There was no evidence of differential loss by concentration of carotenoids, rather a loss of -50% at all concentrations. Therefore, the ordinal ranking of individuals would be similar to that provided by the higher absolute values. Carotene values are reported here in the original form

because relative and not absolute values were used to support these types of statistical analyses.

The association of lower ,B-carotene concentrations with breast cancer may be confounded by alcohol intake, which was not measured in this study. Alcohol was associated with breast cancer in 12 studies (35-46) but not in 5 other studies (47-51). In addition, alcohol may be associated with lower plasma ,B-carotene values (32, 52) although this effect has only been observed in men and it is unclear whether alcohol is directly related to lower ,B-carotene concentrations or is coincident with lower ,B-carotene intake. Further research is warranted to determine whether the association of plasma ,B-carotene values with breast cancer persist after controlling for alcohol consumption. Although smoking has been associated with lower plasma ,B-carotene (32, 52), it has not been associated with breast cancer (53) and therefore was not controlled in the analyses.

Lower plasma ,B-carotene values in the breast-cancer patients may also be a result of rather than a precursor of disease. A marginally significant trend (P = 0.08) of decreasing ,B-carotene values with increasing stage of disease was observed; this suggests that the disease may alter this blood variable. Alternatively, the patients with later-stage disease may have altered their intake to a greater degree than did the patients with earlier-stage disease. Although it is difficult to disentangle such results in a case-control study, there is some evidence from other studies against a metabolic alteration due to the disease. A prospective study in Britain (19) demonstrated that the subjects with breast cancer diagnosed within 2 y of blood collection did not have lower ,B-carotene concentrations than did subjects diagnosed later. In addition, there is no evidence from animal studies that tumors affect the concentrations of this nutrient whereas there is evidence that low ,B-carotene intake promotes mammary tumors (I 0). However, further studies are needed to elucidate the possible affect of the disease on ,B-carotene concentrations.

Plasma ,B-carotene was shown to reflect dietary intake of ,B-carotene (26, 54, 55) and total carotene intake (32, 52, 56); therefore, the lower ,B-carotene concentrations observed to be associated with case status may be of dietary origin. Although the dietary data from this study did not support this conclusion, results from larger dietary studies indicated that lower estimated intakes of total vitamin A (23) or ,8-carotene (17) were

TABLE 5
Odds ratios (subjects with breast cancer vs control subjects) for dietary nutrient quartiles with adjustments for potential confounding variables in the full model*

Nutrient	Quartiles			Q4t	P§
	QI	Q2	Q3		
Total vitamin A	1.35[ll] (0.50, 3.60)[ll]	1.43 (0.53, 3.87)	0.45 (0.14, 1.45)	1.00	0.56
Vegetable vitamin A	1.27 ((0.44, 3.67)	1.72 (0.66, 4.48)	1.48 (0.50, 4.36)	1.00	0.52

Adjusted for age, age at first birth, family history. menarche, Quetelet index, parity, age at menopause , income, and marital status.
t QI is low and Q4 is high.
t Reference group.
§ Significance of trend.
ll Odds ratio of quartile compared with the fourth quartile. 95% confidence limits.

associated with breast cancer. Vitamin supplements were not considered in this study because only six women reported taking /j-carotene supplements and the common multiple-vitamin supplement did not contain (j-carotene at the time of data collection.

There have been four other investigations of breast cancer that have measured plasma carotene values. In a prospective study in England (19) the authors found lower plasma P-carotene values (NS) in women later diagnosed with breast cancer than in women who remained disease free. Although these findings were later questioned because of storage considerations (57), the present study corroborates these findings. In a preliminary report (26) the mean plasma Jj-carotene concentration of 30 women with metastatic breast cancer was 21% lower than in control subjects. Willett et al (20) did not find an association between breast cancer and plasma carotenoids; however. this study was limited by sample size $(n = 14$ cases) and did not control for important confounding factors. In a case-control study in Italy (16), the authors did not find an association between breast cancer and plasma P-carotene values. The mean P-carotene values were relatively high in this population, which suggests that there were few individuals in the low range where the effect was observed in the present study (even if our values were doubled to allow for analytical extraction efficiency).

Alpha-carotene and lycopene

There was weak evidence of an association between a-carotene and the disease in the model that did not include Jj-carotene and lipids. However, the collinearity between P-carotene and a-carotene $(r = 0.71)$ made it difficult if not impossible to assess the independent a-carotene effect when both carotenoids were necessary in the full model. The larger range of Jj-carotene allowed it to predominate but this does not exclude the possibility of a contribution from a-carotene.

Plasma lycopene values were associated with case status only before plasma cholesterol and triglycerides were controlled in the analysis. Adjustment for the these lipids allowed observation of the independent effect of lycopene (or lack of an effect) after differences in lipid status and the association of the lipids with case status were eliminated. Stryker et al (32) demonstrated a better correlation between calorie-adjusted dietary intake of fat-soluble nutrients (carotenoids and tocopherols) and the lipid-adjusted concentrations of these nutrients in plasma compared with the unadjusted concentrations. Therefore, the inference of risk estimates from plasma data to dietary intake is stronger after the plasma variables are adjusted for these lipids.

Plasma retinal

The higher risk observed with higher retinol within the low P-carotene quartile is consistent with studies that associated breast cancer with low dietary intake of total vitamin A (23, 24) or P-carotene (17) and high plasma retinol (16). The significance of the high plasma retinol is u nclear, perhaps it is a marker of particular dietary habits or other metabolic factors. Retinol has been positively associated with plasma estrogens (58) and, because estrogens may be associated with promotion of breast cancer, higher retinol concentrations may merely be a marker for the estrogenic environment or, perhaps, part of the mechanism by which estrogens exert their effect. Nonetheless, the pattern of risk observed suggests that P-carotene is not

acting solely through its conversion to retinol. and that these two nutrients may have some interdependent functions in carcinogenesis. The lack of a main effect of plasma retinol and the presence of an interactive effect between plasma retinol and P-carotene demonstrates the importance of evaluating n utritional interactions in studies of cancer etiology.

Dietary data

The results from the dietary data were consistent with one study that did not find an association between breast cancer and dietary Jj-carotene (16). Other dietary studies (17, 23, 24) with larger sample sizes (451, 2024, and 120 cases. respectively) than the present study did show associations when dietary indicators of vitamin A or (j-carotene were used.

The questionnaire used in this study was an abbreviated form of a larger food frequency questionnaire. Although individuals should rank approximately the same by the use of either questionnaire, the abbreviated questionnaire may be more prone to misclassification than the more complete questionnaire. This misclassification would have more impact on small studies and therefore could account for the lack of findings.

Preliminary analysis of the reproducibility of the dietary data from this study suggests poor agreement in frequency categories between first and second questionnaires (59% exact agreement, 71% agreement plus or minus one frequency category). Thus, these dietary data included a large amount of variability that was unaccounted for, which biased the results toward the null hypothesis. The positive findings when the plasma indicators were used, the lack of a correlation between the plasma and dietary indicators of vitamin A, and the lack of reproducibility of the dietary data suggest that dietary indicators were not as sensitive as were plasma indicators. Therefore, a larger sample size would be required for the food frequency methodology employed here to be useful.

In conclusion, the findings concerning P-carotene are in accord with studies that found an association between low Jj-carotene concentrations and cancers of epithelial origin (59). It is important to note, however, that high plasma P-carotene may also be a marker for carotene-rich foods so that other carotenes, or other constituents of these foods, may be the more important risk factors. Low plasma P-carotene in combination with high retinol was found to be associated with breast cancer, which was consistent with diets low in carotene-containing vegetables and may be a marker of a dietary pattern. These findings are consistent with the National Research Council's (60) dietary recommendations for increased consumption of fruits and vegetables. As interest in the association between nutrition and cancer rises (61), additional studies of this type will be warranted. ○

References

I . Sporn MB, Robens AB. Role of retinoids in differentiation and carcinogenesis. Cancer Res 1983;43:3034-40.
2. Moon RC. Grubbs CJ, Sporn MB. Inhibition of 7,12-dimethyl-benz-(a)-anthracene-induced mammary carcinogenesis by retinyl acetate. Cancer Res 1976;36:2626-30.
3. Moon RC. Grubbs CJ. Sporn MB, Goodman 00. Retinyl acetate inhibits mammary carcinogenesis induced by N-methyl-N-nitro-sourea. Nature 1977;267:620-1.
4. Thompson HJ, Meeker LD, Tagliaferro AR. Becci PJ . Effect of

retinyl acetate on the occurence of ovarian hormone-responsive and -nonrespnsive mam mary cancers in rats. Cancer Res 1 982:42:903-5 .

5. McCormick DL. Burns FJ. Albert RE. Inhibition of benzo[a]pyrene-induced mammary carcinogenesis by retinyl acetate. JNCI 1981:66:559-64 .

6. McCormick DL. Sowell ZL. Thompson CA. Moon RC. Inhibition by retinoid and ovariectomy of additional primary malignancies in rats following surgical removal of the first mammary cancer. Cancer 1983:51:594-9.

7. Stone J P. Shellabarger CJ, Holtzman S. Life-span retinyl acetate inhibition of spontaneous and induced rat mammary carcinogenesis.Carcinogenesis 1987;28:141(abstr).

8. Holtzman S. Retinyl acetate inhibits estrogen-induced mammary carcinogensis i n female ACI rats. Carcinogenesis 1988:9:305-7.

9. Welsch CW. Goodrich-Smith M . Brown CK. Crowe N. Enhancement by reti nyl acetate of hormone-induced mammary tumorigenesis in female GR/A mice. J NCI 1981:67:935-8.

10. Rettura G. Duttaqupta C. Listowsky P. Levenson SM, Seifter E. Dimethylbenz(a)anthracene (DMBA) induced tumors: prevention by supplemental (:I-carotene (BC).Fed Proc 1983;42:786(abstr).

1 1. Seifter E. Rettura G. Levenson SM. Supplemental 13-carotene (BC): prophylactic action against 7.12-dimethylbenz(a)anthracene (DMBA) carcinogensis. Fed Proc 1984;43:662(abstr).

12. Rettura G. Stratford F, Levenson SM. Seifter E. Prophylactic and therapeutic actions of supplemental ti-carotene in mice inoculated with C3HBA adenocarcinoma cells: lack of therapeutic action of supplemental ascorbic acid. JNCI 1982:69:73-7.

13. Kark JD. Smith AH . Switzer BR. Hames CG. Serum vitamin A (retinol) and cancer incidence in Evans County. Georgia. JNCI 1981 :66:7-16.

14. Wald N. Idle M. Boreham J. Bailey A. Low serum vitamin A and subsequent risk of cancer. Lancet 1980:2:813-5.

15. Peleg l. Heyden S. Knowles M. Hames CG. Serum retinol and risk of subsequent cancer: extension of the Evans County. Georgia, study. J NCI 1984:73: 1455-8.

16. Marubini E. Decarli A. Costa A. et al. The relationship of dietary intake and serum levels of retinol and beta-carotene with breast cancer. Cancer 1988:61 :I 73-80.

17. Rohan TE. McMichael AJ. Baghurst PA. A population-based case-control study of diet and breast cancer in Australia . Am J Epidemiol 1988: 128:478-89.

18. Toniolo P. Riboli E. Protto F. Charrel M . Cappa APM . Calorie-providing nutrients and risk of breast cancer. J NCI 1989;8 l:278-86.

19. Wald NJ. Boreham J . Hayward JL, Bulbrook RD. Plasma retinol. beta-carotene and vitamin E levels in relation to the future risk of breast cancer. Br J Cancer 1984:49:321-4.

20. Willett WC. Polk BF. Underwood BA. et al. Relation of serum vitamins A and E and carotenoid to the risk of cancer. N Engl J Med 1984:310:430-4.

21 . Russell MJ. Thomas BS. Bulbrook RD. A prospective study of the relationship between serum vitamins A and E and risk of breast cancer. Br J Cancer 1988:57:213-5.

22. . Katsouyanni K. Trichopoulos D, Boyle P. et al. Diet and breast cancer: a case-control study in Greece. Int J Cancer 1986;38:815- 20.

23. Graham S. Marshall J. Mettlin C. Rzepka T. Nemoto T. Byers T. Diet in the epidemiology of breast cancer. Am J Epidemiol 1982:1 16:68-75.

24. Katsouyanni K. Willett W. Trichopoulos D. et al. Risk of breast cancer among Greek women in relation to nutrient intake. Cancer 1988;61:181-5.

25. Basu TK. Hill GB. NG D. Abdi E. Temple N . Serum vitamins A and E, .B-carotene. and selenium in patients with breast cancer. J Am Coll Nutr 1989;8:524-8.

26. Cantin L. Daignault-Gelinas M. Latrielle J. Bhat PY, Lacroix A. Retinol, carotenes and nutritional status in metastatic breast cancer. Union Med Can 1988;premier trimestre:29-36.

27. Byers T, Marshall J, Fiedler R, Zielezny M, Graham S. Assessing nutrient intake with an abbreviated dietary interview. Am J Epidemiol 1985; 122:41-50.

28. Driskell WJ, Neese JW, Bryant CC, Bashor MM . Measurement of vitamin A and vitamin E in human serum by high-performance liquid chromatography. J Chromatogr 1982;231 :439-44.

29. Wang G, Root MM, Ye X, Chen J, Campbell TC. Routine assay of plasma carotenes by HPLC with an internal standard.J Micronutr Anal 1989;5:3-14.

30. Hislop TG, Band PR, Deschamps M, et al. Diet and histologic types of benign breast disease defined by subsequent risk of breast cancer. Am J Epidemiol 1990; 131:263-70.

31. Comstock GW, Menkes MS, Schober SE, Vuilleumier JP, Helsing KJ. Serum levels of retinol, beta-carotene, and alpha-tocopherol in older adults. Am J Epidemiol 1987; 127:114-23.

32. Stryker WS, Kaplan LA, Stein EA, Stampfer MJ, Sober A, Willett WC. The relation of diet, cigarette smoking, and alcohol consumption to plasma beta-carotene and alpha-tocopherol levels. Am J Epidemiol 1988;127:283-96.

33. Bieri JG, Brown ED, Smith JC Jr. Determination of individual carotenoids in human plasma by high performance liquid chromatography. J Liq Chromatogr 1985;8:473-84 .

34. Stacewicz-Sapuntzakis M, Bowen PE, Kikendall JW, Burgess M. Simultaneous determination of serum retinol and various carotenoids: their distribution in middle-aged men and women . J Micronutr Anal 1987;3:27-45.

35. Williams RR, Horm JW. Association of cancer sites with tobacco and alcohol consumption and socioeconomic status of patients: interview study from the Third National Cancer Survey. J Natl Cancer Inst 1977;58:525-47.

36. Rosenberg L, Slone D, Shapiro S, et al. Breast cancer and alcoholic-beverage consumption . Lancet 1982; l:267-71.

37. Hiatt RA, Bawol RD. Alcoholic beverage consumption and breast cancer incidence. Am J Epidemiol 1984; 120:676-83.

38. Le MG, Hill C, Kramar A, Flamant R. Alcohol beverage consumption and breast cancer in a French case-control study. Am J Epidemiol 1984; 120:350-7.

39. Talamini R, La Vecchia C, Decarli A, et al. Social factors, diet and breast cancer in a northern Italian population. Br J Cancer l 984;49:723-9.

40. La Vecchia C, Decarli A, Franceschi S, Pampallona S, Tognoni G. Alcohol consumption and the risk of breast cancer in women. JNCI 1985;75:61-5.

41. Harvey EB, Schairer C, Brinton LA, Hoover RN, Fraumeni JF Jr. Alcohol consumption and breast cancer. JNCI 1987;78:657-61 .

42. O'Connell DL, Hulka BS, Chambless LE, Wilkinson WE, Deubner DC. Cigarette smoking, alcohol consumption , and breast cancer risk . JNCI 1987;78:229-34.

43. Schatzkin A, Jones Y, Hoover RN, et al. Alcohol consumption and breast cancer in the epidemiologic follow-up study of the first National Health and Nutrition Examination Survey. N Engl J Med 1987;316:1 169-73.

44. Willett WC, Stampfer MJ, Colditz GA. Rosner BA, Hennekens CH, Speizer FE. Moderate alcohol consumption and the risk of breast cancer. N Engl J Med 1987;316:1 174-80.

45. Hiatt RA, Klatsky Al, Armstrong MA . Alcohol consumption and the risk of breast cancer in a prepaid health plan. Cancer Res 1988;48:2284- 7.

46. Rohan TE, McMichael AJ . Alcohol consumption and risk of breast cancer. Int J Cancer 1988;41:695-9.

47. Begg CB, Walker AM , Wessen B, Zelen M. Alcohol consumption and breast cancer. Lancet 1983; I :293-4(letter).

48. Byers T, Funch DP. Alcohol and breast cancer. Lancet 1982;I: 799-800(letter).

49. Paganini-Hill A. Ross R K. Breast cancer and alcohol consumption. Lancet 1983;2:626-7(1etter).

50. Webster LA. Layde PM. Wingo PA. et al. Alcohol consumption and risk of breast cancer. Lancet I983;2:724-6.

51. Harris RE. Wynder EL. Breast cancer and alcohol consumption. A study in weak associations. JAMA 1988;259:2867-71.

52. Russell-Briefel R. Bates MW. Kuller LH. The relationship of plasma carotenoids to health and biochemical factors in middle-aged men. Am J Epidemiol 1985;122:741-9.

53. Baron JA. Smoking and estrogen-related disease. Am J Epidemiol 1984; 1 19:9-22.

54. Buzina R, Jusic M, Brodarec A, et al. The assessment of dietary vitamin intake of 24 lstrian farmers: II . Comparison between the dietary intake and biochemical status of ascorbic acid. vitamin A, thiamine. ribonavin. and niacin. Int J Vitam Nutr Res 1971;41: 289-300.

55. Peto R. The marked differences between carotenoids and retinoids: methodological implications for biochemical epidemiology. Cancer Surv 1983;2:327-40.

56. Willett WC. Stampfer MJ. Underwood BA. Speizer FE. Rosner B. Hennekens C. Validation of a dietary questionnaire with plasma carotenoid and alpha-tocopherol levels. Am J Clin Nutr 1983;38:631-9.

57. Wald NJ. Hudson GA. Plasma retinol. beta-carotene and vitamin E levels in relation to future risk of breast cancer. Br J Cancer 1988;57:235(1etter).

58. Wolf G. Vitamin A. In: Alfin-Slater RB, Kritchevsky D, eds. Nutrition and the adult: micronutrients. New York: Plenum Press. 1980:108-1 1 1, 190-1.

59. Peto R, Doll R, Buckley JD, Sporn MB. Can dietary beta-carotene materially innuence human cancer rates? Nature 1981;290:201-8.

60. Committee on Diet. Nutrition and Cancer, National Research Council. Diet, nutrition and cancer. Washington, DC: National Academy Press, 1982.

61. Rohan TE, Bain CJ. Diet in the etiology of breast cancer. Epidemiol Rev 1987;9:120-45.