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Role of Biotin and its effects on health

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Introduction

Biotin is contained in various food products, but little is known about it. Recently, biotin has been found to be greatly involved in our health. In this report, we will discuss the role of biotin in a living body, particularly, the involvement of biotin in dermatopathies, such as atopic dermatitis, which is attracting great attention in recent years. Furthermore, since biotin will soon be approved as a food additive, we will introduce the importance of biotin as a dietary supplement.

1. What's Biotin?

Biotin is a water-soluble vitamin found in a wide range of food such as liver, yolk and cereals. It is also synthesized by enteric bacteria. Therefore, except for people with extremely unbalanced diet or intestinal disorders, generally, biotin deficiency is rarely found among people with ordinary diet. Biotin deficiency is possibly caused by reduction of biotin intake, inhibition of biotin absorption, inborn errors of metabolism, etc.

In our bodies, biotin acts as a coenzyme, supporting the function of carboxylase. Since carboxylase is an enzyme involved in glyconeogenesis, fatty acid synthesis, amino acid metabolism, etc., biotin deficiency disturbs the energy metabolism as well as various physiological functions. Deficiency of biotin also results in immune depression and reduced collagen synthesis. As a result, biotin deficiency is known to induce dermatological symptoms such as seborrheic eczema and psilosis, opportunistic infection and neuritis in human, although its direct involvement is still unclear. A recent report has suggested that biotin levels decrease along with the gestational stage in pregnant women. In an animal experiment, biotin deficiency in the mother induced fetal anomaly.

Biotinidase is an enzyme involved in biotin metabolism, which liberates biotins bound to proteins within the digestive tract. As a carrier protein, it is also involved in biotin absorption and transport. Therefore, symptoms of biotin deficiency are also observed in cases of carboxylase- or biotinidase-related impairment. "Egg white injury", found in those consuming a large amount of raw egg over a long period of time, is also caused by biotin deficiency.

Consequently, biotin has been otherwise known as "vitamin H" in which "H" stands for "Haut", the German word for "skin". Since the old days, biotin has been known as a vitamin closely related to the skin. Therefore, biotin has been used as a remedy for dermatoses, but rarely after the recent development of new drugs. Chinese medicine formulations used for dermatoses also contain a large amount of free biotin.

2. Diabetes and Biotin

Palmoplantar pustulosis and palmoplantar pustular osteoarthropathy are diseases in which many rashes and pustules appear on the palms and soles. To date, there is no definitive treatment for such diseases. The blood biotin levels in these patients are less than half of the normal level, and about 60% of the patients have diabetes as complications. Oral administrations of a large dose (9 mg/day) of biotin to the patients not only eliminated the rashes and bone aches but also reduced their blood glucose levels, indicating the efficacy of biotin dosing against diabetes.

When biotin was continuously administered in combination with an antidiarrheal to insulin-independent diabetes patients, the blood glucose levels were actually decreased to a normal range in all of the patients tested. Even in insulin-dependent diabetes patients, biotin dosing resulted in normal blood glucose levels. The mechanism of biotin's action is still unclear, but since biotin is involved in sugar metabolism, it is suggested that biotin administration has promoted sugar metabolism, which lead to reduction in the blood glucose levels.

3. Infant and Biotin

Infants having milk allergy or inborn error of metabolism are fed with special therapeutic milk preparations. In our country, biotin cannot be added to food products, because it has not been approved as a food additive.

Therefore, biotin is not added to the milk powders or special therapeutic milk preparations that are currently in use.

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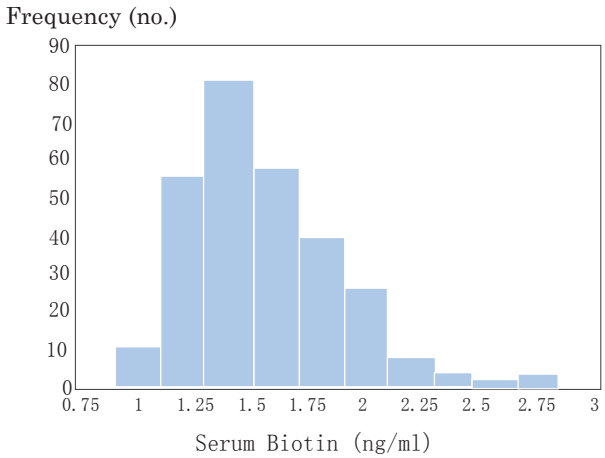


Fig. Serum Biotin of a diabetes close examination appropriate person
(Standard 1.6-3.7 ng/ml)

Especially, therapeutic special milk preparations are prepared using purified raw materials, so some products barely contain biotin. The average biotin content in the commercially available powdered milk and special therapeutic milk preparations are 1.04 and 0.45 μg/100 kcal, respectively, which are extremely low compared to the level (1.5 μg/100 kcal) recommended by FAO/WHO (United Nations Food and Agriculture Organization). Therefore, insufficient biotin intake is greatly concerned for artificially fed infants who are fed with such powdered milk in our country.

Recently, it has been reported that serum biotin levels in atopic dermatitis patients are less than half of the normal level. Moreover, when infants diagnosed as milk allergy were fed with special therapeutic milk preparations, diaper rash-like rashes and erythema around the eyes and mouths appeared. The erythema disappeared when biotin was administered to such infants. These reports indicate that biotin is involved in the development of atopic dermatitis. The biotin content in baby food is within the range of 0−58.5 μg/100g. In general, biotin content is higher in cereal- or meet-based food but lower in juice beverages and refreshments based on dairy products. Some products contain no biotin at all.

In general, infant meals have poor variety, and powdered milk has low biotin content. In addition, functions of digestive tracts are immature in infants, allowing little production and absorption of biotin. Therefore, the risk of biotin deficiency is higher in infants. Biotin deficiency hinders skin formation, resulting in a state susceptible to external stimuli. Therefore, a great attention must be paid to the biotin intake in infants.

4. Bitoin dysbolism

In some patients who have been diagnosed as atopic dermatitis, remission cannot be observed for more than 5 years. Among such patients, 3−4% are found to have low biotinidase activities. In addition to the reduced enzyme activity, the biotin levels in such patients are less than 10% of those of healthy subjects. Biotin administration (5 mg/day) resulted in the elimination of the eczema formed on their faces, indicating that some of the intractable cases of atopic dermatitis are caused by deficiencies of biotin-related enzymes such as biotinidase and carboxylase.

5. Biotin as supplement

In the United States and European countries, biotin is used in daily food and vitamin tablets, and its importance is widely accepted. Meanwhile, in our country, biotin is not used as a dietary supplement, since it is not approved as a food additive. The reason for its disapproval is not because biotin has any toxicity, but because its nutritional importance has not been sufficiently acknowledged. This is obvious from the fact that biotin is approved as a remedy for dermatoses.

The recommended dietary allowance for biotin has been first established in the sixth revision of nutritional requirements for the Japanese. The recommended dietary allowance for biotin is 30 μg/day for adults. The maximum acceptable intake has not been established, since an excessive dosing (10−100 mg) of biotin showed no side effects, although it has not been tested in healthy subjects. Nevertheless, biotin was not included in the fifth revision of Standard tables of food consumption in Japan, although folic acid and vitamin B12 were newly included.

In our country, health-promoting food has been formulated in April 2001, which has clearly determined the specified health food and the food with nutrient function claims. For the food with nutrient function claims, labeling standards and specifications have been established for 12 vitamins including biotin and 2 minerals. According to the ingredient specification for biotin, the maximum and minimum allowable dosages are 500 μg and 10 μg, respectively. Moreover, the nutrient function claim approved for biotin is "biotin is a nutrient which supports the health maintenance of the skin and the mucous membrane". As mentioned above, biotin is greatly involved in the health of our skin, so biotin supplement should have a great value.

6. Conclusion

It is generally considered that biotin deficiency does not exist, but the possibility of latent or non-manifest biotin deficiency still remains. Moreover, it has been suggested that biotin deficiency can worsen the intractable cases of atopic dermatitis or diabetes. Nevertheless, the physiological function of biotin has not been sufficiently understood, requiring further research in this area. At this occasion of the approval of biotin as a food additive, it is expected that the importance of biotin will be reviewed, and that biotin supplement developed based on the latest research results will be available.

Comparison of the amount of Biotin intake in each country

References	μg/day	
'78 Hoppne et al	62	Canada; Diet survey
	60	Analysis from diet
'82 Bul & Buss	35.5	UK; Diet survey
'86 Murphy & Calloway	39.9±26.9	USA (women, 18-24 aged)
'88 Lewis & Buss	35-70	Calculation from foods
Author et al.	37.2	Healthy adults
		(Tohoku-area)
Adequate Intake (AI)	30	Adults (Japan, USA)

The alpha-amino group of L-arginine mediates its antioxidant effect

(S. Wallner et al, Eur. J Clin. Invest. 2001, 31, 98-102)

L-arginine has been shown to reduce atherogenesis, both in animal and human experiments. It affects the platelet aggregation or the attachment of monocytes (leukocytes) to blood vessels, which are essential steps in atherogenesis. Conventionally, the effect was attributed to arginine's role as a NO (nitrogen monoxide) precursor, which results in an increased NO production and therefore leading to functional improvement of vascular endothelium. However, our recent study has shown that administration of external arginine delays the internal peroxide eliminative action and the cell-mediated NO degradation, and also decreases lipoprotein oxidation which is induced in the presence of copper.

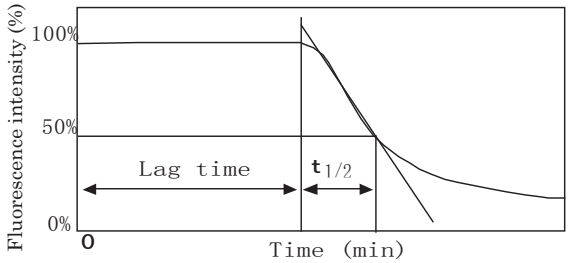
Furthermore, vitamins C and E also improve the vascular endothelium function and inhibit atherogenesis. Therefore, it is considered that not only the NO production but also the antioxidative effect of arginine is involved in such inhibitory effect.

The antioxidative effect of arginine, characterized in the lipoprotein oxidation model, may play a role in atherosclerosis resistance. Furthermore, copper ion is involved in the initiation of LDL oxidation by binding to ApoB-100 (the main protein in LDL (low-density lipoprotein)) at a specific saturated state. It is believed that free radicals penetrate and diffuse into lipid layers from this oxidation initiation point by transferring from surface to surface of lipid particles.

While monitoring the serum lipoprotein level using fluorescent probes, the antioxidative effects of L-arginine, N-alpha-acetylarginine and vitamin E in combination with arginine, were determined after the free radical generation induced by copper (CuSO4) or AAPH (2,2'-acobis(2-amidinonpropane) hydrochloride). The propagation rate of copper-induced lipoprotein oxidation increased in a dose-dependent manner by L-arginine treatment but not by N-alpha-acetylarginine treatment.

The propagation of copper-induced lipoprotein oxidation was represented by a reduction in the fluorescence level. Concerning the time required for this, the time required to reach half of the initial level (t 1/2) increased by L-arginine treatment in a dose-dependent manner, but not by N-alpha-acetylarginine treatment.

Fig. Original tracing of serum lipoprotein oxidation initiated by 50 μ M CuSO4 monitored by the decline of fluorescence intensity



Vitamin E and L-arginine show different effects on copper-induced oxidation, the former increasing only lag-time, the latter increasing only propagation rate, and do not have reciprocal effects.

Effect of Vitamin E (15 μ M) and L-arginine (0.8mM)	
Vitamin E	
$t_{1/2}$ (min)	not significantly changed
Lag time	38±6 (control) ⇒ 72±4 (min) (p<0.01)
Vitamin E + L-arginine	
$t_{1/2}$ (min)	28±5(control) ⇒ 121±9 (min) (p< 0.001)
Lag time	38±6(control) ⇒ 68±8 (min) (p< 0.01)

The effect on copper-induced oxidation

L-arginine
t 1/2 (min): 38±5 (0mM) ⇒335±45 (3.2mM)
*N-alpha-acetyl-L-arginine did not result in effect
Lag time: not significantly changed

In contrast to copper-induced oxidation, L-arginine increased the lag-time of AAPH-induced lipoprotein oxidation, with no effect on the propagation rate at physiological concentrations. Again, N-alph-acetyl-arginine did not show any antioxidation effects.

The effect on AAPH-induced oxidation

L-arginine (0 ⇒ 3.2mM)
t 1/2 (min) : 72±11 (0mM) ⇒ 119±19 (3.2mM)
*N-alpha-acetyl-L-arginine did not result in effect
Lag time: 85±12⇒ 178±19 (p<0.01)

This suggest that the effect of the combination of L-arginine and vitamin E on the lag time of AAPH-induced lipid peroxidation was additive rather than synergistic.

Effect of Vitamin E(15 μ M) and L-arginine(0.8mM) on lag time	
Vitamin E alone	
Lag time	85±12 (control⇒ 128±19 (min) (p< 0.05)
Vitamin E + L-arginine	
Lag time	129±18(L-arginine alone) ⇒ 168±15 (min) (p<0.05)

Our experiments provide further evidence, that mechanisms other than serving as a substrate for the NO-synthase could be involved in L-arginine's antiatherosclerotic effect. In addition, our experiments clearly show, that the antioxidant effect of L-arginine is due to a chemical moiety differenr from that necessary for NO biosynthesis.

Basic knowledge on amino acids

Proteins are important components of our body accounting for about 20% of our body weights, forming the cells, hormones and enzymes in our organs and muscles. Approximately 500 kinds of amino acids have been found in the nature. Among these, merely 20 kinds of amino acids are used in various combinations to form 100,000 kinds of proteins.

Among the 20 amino acids, eleven can be synthesized in our body to make up for their deficiencies, while the other nine must be taken up from our meal. The former eleven are called nonessential amino acids, while the latter are called essential amino acids.

Within our bodies, some amino acids are re-synthesized into proteins, while the others are stored within cells or blood, which are called free amino acids. Most free amino acids, including the nonessential amino acids, are extremely important for maintaining our lives.

Essensial amino acids	Non essensial amino acids
Methionine	Glycine
Tryptophan	Glutamic acid
Isoleucine	Thyrosine
Threonine	Alanine
Phenylalanine	Asparagine
Lysine	Proline
Valine	Arginine
Leucine	Glutamine
Histidine	Serine
	Asprtic acid
	Cysteine

Relation of short-term pyridoxine-HCl supplementation to plasma vitamin B6 vitamers and amino acid concentrations in young women

(Soon et al, Am J Clin Nutr 1992; 55: 865-872)

Pyridoxal 5'-phosphate (PLP) is a coenzyme in many metabolic transformations of amino acids and may play a role in their absorption and transport. This investigation analyzed the effects of large oral doses (27mg/day) of pyridxine (PN)-HCl over 2 wk on plasma PLP and amino acid concentration in 10 young women.

Methods and results

Table 1 Influence of short-term supplementation with vitamin B6 on plasma vitamin B6 vitamersand 4-pyridoxic acid concentrations in the young women

B6 vitamers	before supplementation	after 7 days supplementation		after 14 days supplementation	
PLP (tyrosine decarboxylase method)	45±2	377±12	§	429±16	§
PLP (HPLC)	57±7	424±40	§	455±47	§
4-PA	21±4	108±13	*	113±13	**
PMP	3±1	2±1		2±0	
PL	16±4	77±10	***	117±18	§, ∫
PN	13±3	11±1	§§	21±3	§§, ∫∫
PM	4±3	7±3	§	8±2	§
Total (PLP (HPLC)+PMP+PL+PN+PM)	93±11	521±42		604±53	

Significantly diffrent from before supplementation: § : p<0.0001、* : p<0.002、** : p<0.004、*** : p<0.001、§§ : p<0.01
Significantly diffrent from after 7 days supplementation : ∫ : p<0.05、∫∫ : p<0.005

Plasma PLP was 45±2nmol/L initially and reached 377±12nmol/L after 7 d of supplementation. A steady-state PLP concentration remained as long as daily PN-HCl supplementation was continued.

Table 2 Plasma amino acid concentrations and other N-containing compounds that were by short-term supplementation with high doses of vitamin B6

	before supplementation	after 7 days supplementation		after 14 days supplementation	
Phosphoserine	12±3	16±3	§	17±3	§
Urea	4421±508	5108±1145	*	5198±444	*
Aspartic acid	4± 1	5±1	*	4±1	*
Glutamic acid	51±17	35±14	§	27±12	§
Glutamine	971±182	1127±148	*	1023±97	*
Alanine	369±97	451±107	*	440±128	*
α-amino-N-butyric acid	20±5	32±6	**	32±3	**
Cysteine	20±3	29±5	**	30±4	**
Methionine	26±4	30±4	*	26±3	*
Isoleucine	65±9	74±8	∫	62±6	
Tyrosine	62±9	73±12	∫∫	62±8	
Arginine	71±20	110±23	§§	103±24	§§

Significantly diffrent from before supplementation: § : p<0.005、* : p<0.05、** : p<0.0001、∫ : p<0.01
∫∫ : p<0.02、§§ : p<0.00 1
Amino acids and N-containing compounds not altered by a high dose of vitamin B6 : Taurine, Hydroxy-proline, Threonine, Serine, Asparagine, Proline, Glycine, Citrulline, Valine, Leucine, Phenylalanine, Tryptophan, Lysine, Histidine

Plasma glutamic acid concentration was significantly lower after 7 and 14 d of supplementation whereas alpha-amino-N-butyric acid, alanine, cysteine, arginine, phosphoserine, and urea concentrations were significantly higher.

Table 3 Responses of plasma amino acid to short-term oral supplementation with high doses of vitamin B6

	before supplementation	after 7 days supplementation		after 14 days supplementation	
Total essential amino acids (μM/L)	966±33	1078±38	§	969±33	**
Total amino acids (μM/L)	3072±141	3525±127	§	3217±125	
Total essential amino acids / total amino acids	0.30±0.0 1	0.32±0.01		0.31±0.01	
Branched-chain amino acids (μM/L)	394±14	455±21	§	415±11	§
Aromatic amino acids (μM/L)	175±6	193±8	§	170±5	**
Sulfur-containing amino acids (μM/L)	46±2	59±3	*	56±2	*

Significantly diffrent from before supplementation § : p<0.05、* : p<0.01、** : p<0.05

Discussion

In our study, cases of vitamin B6 deficiency in young females were accompanied by increased blood levels of glycine, serine, glutamic acid and alpha-amino-N-butyrate and decreased blood levels of alanine, cysteine and arginine. In other reporting on the effect of B6 deficiency on amino acid levels in male subjects, B6 deficiency resulted in temporary changes in blood levels of some amino acids, which was considered to derive from the secretion or the repartitioning of such amino acids among tissues. Vitamin B6 deficiency may inhibit the tissue absorption of some amino acids, which would explain the increased fasting blood levels of glycine and serine, which were observed under vitamin B6 deficiency. In fact, vitamin B6 was found to stimulate the absorption of some amino acids by the small intestine, so its deficiency may lead to decreased absorption of such amino acids. Vitamin B6 is a coenzyme that acts in the enzymatic reaction involved in various non-oxidative degradation and dissimilation of free amino acids. An administration of 30 mg of pyridoxin hydrochloride under B6 deficiency resulted in a temporary increase and a subsequent decrease of blood levels of free amino acids. The blood amino acid level changed temporally, along with the time from B6 intake. An increased urea level was also observed, which indicated the acceleration of protein or amino acid metabolism.

Published Safety Observations for Vitamin E Supplementation

(The Established Safety of Supplements of Vitamin E and C: The Scientific Evidence Nov. 15, 2002 CRN)

Reference	Subjects	Dosage	Duration	Safety Observation
1	Symptomatic Coronary disease n=36	3,200 IU/day	9 weeks	No significant subjective or objective adverse effects
2	Heart disease n=75	200 mg/day	4-6 weeks	No significant adverse effects
3	Volunteers n=30	800 IU/day	16 weeks	No reported adverse effects
4	Diabetes n=25	2,000 IU/day	6 weeks	No subjective adverse effects
5	Male smokers aged 50-69, n=29,133	50 mg/day	5-8 years	vitamin E associated with somewhat higher incidence of hemorrhagic stroke and lower incidence of ischemic stroke as compared to placebo (degree of statistical significance not reported)
6	Symptomatic coronary disease n=2,002	400 or 800IU/day	510 days median	Only 0.55% of patients discontinued treatment due to adverse effects (no difference between active and control groups)
7	Volunteers n=88	60-800 IU/day	4 months	No subjective adverse effects
8	Patients with Parkinson's n=800	2,000 IU/day	8.2 years	No adverse effects reported
9	Recent heart attack n=11,324	300 mg/day	3.5 years mean	No reported adverse effects
10	Multiple cardiovascular risk factors n=9,541	400 IU/day	4.5 years mean	No significant adverse effects
11	Cardiovascular patients on hemodialysis n=196	800 mg/day	519 days mean	No reported adverse effects
12	Smokers, non-smokers, men and post-menopausal n= 520名	91 mg/day	3 years	No significant adverse effects
13	AMD and vision loss n=3,640	400 IU/day	6.3 years mean	Antioxidant cocktail significantly associated with rate increases in skin yellowing
14	Vascular disease or diabetes n=732	400 IU/day	4.5 years mean	No significant adverse effects
15	Cardiovascular risk factors n=4,495	300 mg/day	3.6 years mean	No reported adverse effects
16	Healthy elderly subjects n=1,193	500 IU/day	4 years	No significant adverse effects

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Dietary Supplements Research

(Annual Bibliography of Significant Advances in Dietary Supplement Research 2001)

The Office of Dietary Supplements at the National Institutes of Health published the Annual Bibliography of Significant Advances in Dietary Supplement Research 2001. Two examples are introduced as follows.

B Vitamins and Homocysteine

(Low dose vitamin B6 effectively lowers fasting plasma homocysteine in healthy elderly persons who are folate and riboflabin replete; Am J Clin Nutr, 2001, 73:759-764)

Aging is associated with an increase in homocysteine blood levels and a decline in vitamin B6 status, which may increase the risk for cardiovascular disease in the elderly. The aim of this study was to see if vitamin B6 would independently homocysteine levels in older individuals who were not deficient in folate, riboflavin, or vitamin B12. To ensure that they were not deficient in these B-vitamins, 22 healthy older adults between 62 and 80 years of age were given riboflavin (1.6 mg) daily for 12 weeks followed by a combination of folic acid (400 μg) and riboflavin (1.6 mg) daily for an additional six weeks. Vitamin B12 supplements were not provided, as none of the individuals was deficient in vitamin B12. They were then given a low-dose level of vitamin B6 (1.6 mg) or a placebo daily for an additional 12 weeks while continuing to take the riboflavin and folic acid supplements. Folic acid supplementation lowered fasting blood levels of homocysteine by 19.6 percent. Vitamin B6 supplementation lowered these levels by an additional 7.5 percent. These data suggest that adding low-dose vitamin B6 to a B-vitamin supplementation regimen will result in additional reductions in blood levels of homocysteine and thus prove helpful in protecting older individuals from cardiovascular disease.

Table Response of plasma total homocysteine and B Vitamins status to 12wk of supplementation with 1.6mg B6

	Before	After (12wk)
Plasma tHcy(μmol/L)	9.90±3.03	9.16±2.34
Plasma PLP (nmol/L)	26.2±17.6	47.4±29.7
Serum folate (nmol/L)	36.6±10.9	45.9±20.2
Serum vitamin B12 (pmol/L)	269.5±44.0	267.2±51.5

Serum carotenoids and breast cancer

(Serum Carotenoids and Breast Cancer; Am J Epidemiol 2001;153:1142-1147)

Carotenoids may contribute to the prevention of cancer by counteracting oxidative processes that could damage and interfere with the normal functioning of cells. While it is documented that adequate intake of fruits and vegetables protects against some forms of cancer, the evidence in the case of breast cancer is not compelling. This study examined the association between the etiology of breast cancer and blood biochemical markers that indicate intakes of fruits, vegetables, and carotenoids in supplements. Blood concentrations of the carotenoids lutein, zeaxanthin, β-cryptoxanthin, lycopene, α-carotene, and β-carotene were compared to the incidence of breast cancer among 270 women with and 270 women without a history of breast cancer. Analyses of the data showed an increased risk of breast cancer with decreasing levels of lutein, β-cryptoxanthin, α-carotene, and β-carotene. The risk of breast cancer for women with blood levels of β-carotene in the lowest quartile was double that of those in the highest quartile. The results of this observational study indicate that low intakes of carotenoids, from either foods or supplements, are associated with an increased risk of breast cancer and may have public health relevance for women.

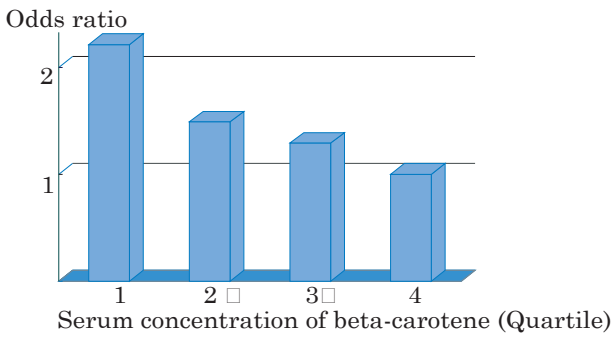

Fig. Odds ratios of breast cancer for quartiles of serum beta-carotene

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