

Report on Effects of Beta-carotene Supplementation in Combination with Tocopherol and Ascorbate in Clinical and Chemopreventive Trials (Adopted by the SCF on 19/3/98)

Terms of Reference

The committee was asked by the Commission to review on recent reports of the studies on health effects of high doses of beta-carotene, retinol and alpha-tocopherol as well as ascorbate in controlled studies.

Introduction

Epidemiological studies of the last ten years have indicated that beta-carotene is a potential agent for the chemical prevention against carcinogenesis. In contrast, the recent prospective study performed by Heinonen et al. (1) has strikingly suggested that supplementation with beta-carotene significantly increased the incidence of lung, prostate and stomach cancer.

The rising question is the possible dose dependency of a preventive or harmful action of beta-carotene and possible synergistic effects with other antioxidants.

To answer this question clinical trials for therapy and intervention studies in healthy populations dealing with beta-carotene supplementation in different dosages below the GRAS daily intake of 200 mg have to be regarded.

Clinical trials

Therapeutic dosages of beta-carotene applied varied in a relative wide range of 20 mg/d to 180 mg/d.

The highest daily dosage of 180 mg beta-carotene in combination with 100.000 IU vitamin A (30 mg retinol equivalents) was applied 1988 by Stich et al. (2) in tobacco chewers who had already sustained premalignant leukoplakia.

After 6 months the single treatment with beta-carotene induced a significant remission of leukoplakias compared to the placebo group, although the combination of beta-carotene and retinol was much more effective.

In a similar study, 20 patients with oral leukoplakia were administered 30 mg beta-carotene, 1000 mg ascorbic acid plus 800 IU vitamin E (354 mg alfa-tocopherol equivalents) daily (3). After 3 months supplementation, 60% of the patients had experienced partial or complete regression of their oral clinical symptoms.

For the follow-up study, the same supplementation dosages were given 79 patients with oral lesions over a total period of 9 months (4). The antioxidant supplementation significantly increased serum and oral mucosa levels of the supplemented vitamins. However, these changes did not correlate with the clinical improvement in 56 % of the patients as observed for the reduction of the use of alcohol and tobacco.

The majority of patients with the genetic disease erythropoietic protoporphyria (EPP) benefit from high-dose supplementation of beta-carotene and/or canthaxanthin. Recommended beta-carotene doses for adults with EPP are approximately 180 mg/d, substantially higher than the doses investigated for cancer prevention purposes (15 Å– 50 mg/d). No serious side effects from high dose beta-carotene supplements have been reported in EPP patients, and no long-term toxicity has been observed (5).

The high dosage of 120 mg beta-carotene was applied in 10 HIV-positive patients in combination with a whole body hyperthermia (42 °C, 1 h) daily.

Beside one patient, who died after 4 months, all the others underwent an HIV burden diminution and clinical improvement after the combined physical and beta-carotene treatment. No beneficial effect was observed for the treatment with the beta-carotene alone (6).

In patients with alcohol induced liver disease the interactions of supplemented beta-carotene and alcohol were investigated by Ahmed et al. (1994). Before starting the daily supplementation of 30 mg and 60 mg beta-carotene for 3 days in alcoholics, the patients showed lower plasma concentrations of beta-carotene than healthy control subjects. However, heavy drinkers (> 200 g ethanol/d) had about twice the beta-carotene plasma level of those drinking less, with a significant correlation between the plasma beta-carotene and the alcohol intake. After beta-carotene administration patients with cirrhosis had a lower plasma beta-carotene response than those without. An improvement of clinical symptoms was neither observed for the dosage of 30 mg/d nor for that of 60 mg/d of beta-carotene supplementation (7).

A further placebo-controlled clinical trial of a 4 year beta-carotene and retinol supplementation (50 mg/d and 25 000 IU/d (7.5 mg retinol equivalents), resp.) in 755 former asbestos workers with sputum atypia was done by McLarty et al. 1995. The supplementation resulted in significant increases of the serum vitamin concentrations with no clinical toxicity. Baseline analysis revealed that smoking and drinking were associated with lower concentrations of serum beta-carotene, even after dietary intake of beta-carotene was adjusted for. After treatment no significant reduction of sputum atypia was observed (8).

In cancer patients undergoing a bone marrow transplantation an intervention study with high-dose supplementation of beta-carotene (45 mg daily) in combination with alpha-tocopherol (825 mg daily) and ascorbic acid (400 mg daily) for 3 weeks prior to chemo- and radiotherapy was done by Clemens 1994 (9).

After chemo-/radiotherapy in patients without vitamin supplementation the serum levels of the supplemented vitamins were significantly decreased and lower than those of the supplemented patients. The chemo-/radiotherapy induced increase of lipid peroxides in serum of unsupplemented patients could not be observed in the supplemented patients. However, the high-dose supplementation of beta-carotene, alpha-tocopherol and ascorbic acid lead to a lower hepatotoxicity of the chemo-/radiotherapy.

A 6 day supplementation of 30 mg beta-carotene and 300 mg ascorbic acid in 17 healthy volunteers showed a significant influence on the x-ray induced damage of isolated lymphocytes shown by Umegaki et al. 1994 (10). For 12 d, three groups of healthy volunteers were given a beta-carotene deficient diet containing 100 mg ascorbic acid. In the first 6 days there was no supplementation, and in the last 6 days the respective groups were given the vitamin supplementation (group A: beta-carotene, group B: ascorbic acid), or placebo. Overnight fasted blood samples were taken on day 13 and were exposed either to x-ray radiation (0.3 Gy/min for 2 min) for micronucleus induction or left unexposed and were cultured. In the blood samples of the beta-carotene supplemented group the lymphocytes containing x-ray induced micronuclei became less frequent compared to the placebo and ascorbic acid supplemented groups. This indicates that beta-carotene is able to prevent x-ray induced DNA damage.

The results of the clinical trials did not show a close correlation between the beta-carotene dosage ingested and the remission of clinical symptoms or the mortality. However, for healthy populations the more important question is the beta-carotene potential to reduce the risk of developing cancer, cardiovascular diseases or even mortality.

Intervention studies

In the last decade several chemoprevention trials were conducted to verify a possible preventive effect of beta-carotene. At the present, three large-scale randomised trials of beta-carotene were completed.

Beta-carotene and alfa-tocopherol in the prevention of cancer disease

The alpha-tocopherol beta-carotene (ATBC) project was conducted by the National Cancer Institute (NCI) in collaboration with the National Public Health Institute of Finland (1). The purpose of this randomised, double-blind, placebo-controlled study was to evaluate, if certain vitamin supplements would prevent lung and other cancers in a group of 29.133 male smokers in Finland. The 50- to 69 year old participants took a pill containing either 50 mg alpha-tocopherol, 20 mg beta-carotene, both or a placebo for 5 to 8 years daily. In participants taking beta-carotene, 18 % more lung cancers were diagnosed and 8 % more overall death. With respect to the incidence of lung cancer no evidence of an interaction of beta-carotene and alfa-tocopherol was found. The total mortality was 8 % higher among the participants who received beta-carotene than among those, who did not, primarily because of more death from lung cancer and from ischemic heart disease. The incidence of angina pectoris was monitored in a subgroup of 22.269 participants who were diagnosed free of coronary heart disease (11).

The supplementation with alpha-tocopherol was associated with only a minor decrease in the incidence of angina pectoris, while the beta-carotene supplementation was associated with a slight increase. Therefore, no preventive effect of the daily supplementation with 20 mg beta-carotene over a period of 5 to 8 years could be seen.

In the United States the large-scaled beta-carotene and retinol efficacy chemoprevention trial (CARET) is conducted in six areas and founded by the NCI.

The purpose of this study was to see, if the combination of beta-carotene (50 mg/d) and retinol (25.000 IU/d) supplements would prevent lung cancer and other cancers in 14.254 men and women aged 50 to 69 who are smokers or former smokers and in 775 men aged 45 to 69 who have been exposed to asbestos (8, 12, 13).

After an average of 4 years of receiving supplements, 28 % more lung cancers were diagnosed and 17 % more deaths occurred in participants taking beta-carotene and retinol than in those taking placebos. Because these interim results were similar to those of the ATBC study, the intervention was stopped 21 months early on January 18, 1996. Although there was evidence for a harmful effect of beta-carotene, a dose dependency reflected by high serum levels of beta-carotene and high incidence of lung cancer was not observed. The investigators will follow up the study group for five more years to determine the long-term effects of this intervention trial.

One of the critical points of the ATBC and the CARET study was that both studies involved people who were specifically invited to participate because of their high risk for developing lung cancer and that other influence factors like the genetic disposition, environmental factors or life style were not regarded.

Second, the dosage of supplementation might be too high for a population whose nutritional status is adequate or better. The cancer preventive effect of beta-carotene which was seen in animal or clinical human studies could be dose dependent (14).

In the nutritionally deficient population of Linxian / China as one of the worlds highest area at risk for oesophageal and stomach cancer a randomised, placebo-controlled chemoprevention trial was conducted. A sample of 29.584 adults were supplemented with four nutrient combinations of 1) beta-carotene (15 mg/d), alfa-tocopherol (60 mg/d) plus selenium or 2) retinol plus zinc or 3) riboflavin and niacin or 4) ascorbic acid plus molybdenum over a total period of 5.25 years (15).

Statistically significant reductions of oesophageal and stomach cancers and in cancer mortality were observed only in participants receiving the combination of beta-carotene plus alfa-tocopherol plus selenium. For the other nutrient combinations, no preventive effect was seen. Unfortunately, the effect of the individual agents beta-carotene or alfa-tocopherol could not be assessed. Since the trial was carried out among a nutritionally deficient population, its results may not have direct relevance to well-nourished individuals.

As a part of the Health Professional Study, which started 1986 in the USA, two chemopreventive trials, the "Physicians Health Study" and the "Women's Health Study", which both are based at Brigham and Women's Hospital / Boston, were conducted. In the Physicians Health Study the impact of a 12 year beta-carotene (50 mg/d) and aspirin (100 mg/d) supplementation was examined among 22.071 male physicians on cancer at all sites. The supplementation of the aspirin component was stopped in early 1988 due to a 44 % reduction in risk of first heart attack among those taking aspirin.

The beta-carotene supplementation ended December 31, 1995.

In this trial among healthy men, 12 years of supplementation with beta-carotene produced neither benefit nor harm in terms of the incidence of malignant neoplasms, cardiovascular disease, or death from all causes (16).

Data in women will be available from the Women's Health Study, which started in 1992. This chemopreventive trial will investigate the effect of beta-carotene (50 mg/d), alpha-tocopherol (600 mg/d) and aspirin (100 mg/d) on the incidence of lung and breast cancer in about 40,000 US female health professionals age 45 years and older, mostly nurses. Considering the evidence of harmful effects of a beta-carotene supplementation, which were observed in the ATBC and the CARET study, the beta-carotene supplementation is being dropped (17).

To compare tissue and plasma carotenoids status of healthy subjects and subjects with pre-cancer and cancer lesions Pappalardo et al. 1997 (18) examined eighteen subjects which were divided into three groups (control group A: healthy subjects; group B with pre-cancer lesions; group C with colonic cancer). All groups received a daily dose of 30 mg beta-carotene for 15 to 43 days. The subjects with cancer show tissue levels for each carotenoid lower than those of healthy subjects. Patients with colonic cancer seemed to undergo a significant reduction in their antioxidant reserves with respect to the normal subjects. In all groups oral beta-carotene supplementation induced an increase in plasma alpha-carotene.

One of the lowest dosages used in a chemoprevention trial was 20 mg beta-carotene daily administered in the placebo-controlled Australian Polyp Prevention Project (19). The 411 patients, whose eligibility was contingent upon confirmation of at least one colorectal adenoma and removal for all polyps, reduced their dietary fat to 25 % of total calorie intake and supplemented the diet with 25 g wheat bran and 20 mg beta-carotene daily. After 24 months, there was a statistically significant preventive effect of the low fat diet in combination with the supplementation of wheat bran. No preventive effect was observed for beta-carotene.

The failure to see a protective effect of supplemental beta-carotene could be due to many factors: 1) dietary, blood, or tissue carotenoids might primarily serve as a marker for other protective factors, 2) beta-carotene may interact synergistically with other carotenoids and phytochemicals found in a natural food matrix, 3) beta-carotene may have been administered too late in the carcinogenic process, and 4) the duration of supplementation may have been inadequate.

beta-carotene in the prevention of atherogenicity

Four studies have examined the ability of beta-carotene to inhibit the oxidation of LDL, which is suggested to increase atherogenicity. Data of these studies are conflicting.

Two in vitro studies have reported that supplementation of LDL with beta-carotene inhibited oxidation induced by copper and human monocyte macrophages (20,21). In contrast, two researcher groups found that beta-carotene supplementation did not confer increased protection to ex vivo copper-mediated oxidation of LDL despite 17- to 20-fold increase in LDL beta-carotene levels (22,23). It remains possible that beta-carotene contained within the LDL particle may inhibit LDL oxidation in vivo, but the ex vivo assay of LDL oxidation does not readily mimic the in vivo processes. On the other hand, beta-carotene may inhibit oxidation of LDL not from within the particle but by reducing oxidative stress in the atherosclerotic plaque where carotenoids clearly accumulate.

Recent intervention trials with beta-carotene have not confirmed the hypothesis that when given in large doses, the single antioxidant nutrient will protect against coronary artery disease.

The Physicians Health Study (PHS) did not show any differences in cardiovascular disease death, myocardial infarction and stroke between treatment group and placebo group. The Beta-carotene and Retinol Efficacy Trial (CARET) noticed an excess of cardiovascular death among those assigned beta-carotene and vitamin A (24). Rapola et al. (1997) studied the frequency of major coronary events in men enrolled in the ATBC-Study who had a previous myocardial infarction. In fact the risk of fatal coronary heart disease increased in the groups that received either beta-carotene or the combination of alpha-tocopherol and beta-carotene. Therefore they do not recommend the use of alpha-tocopherol or

beta-carotene supplements in this group of patients (25).

The results of the chemopreventive trials show, that there is no need to recommend the supplementation of beta-carotene for healthy individuals consuming a mixed, well balanced diet.

Effects of beta-carotene supplementation in combination with Retinol, Tocopherol and Ascorbate in Clinical and Chemopreventive Trials

Study Author	Design	Purpose	Collective / Population	Dosage / Application form	Findings
Stich et al. (2) (Int J Cancer 1988; 42:195-9)	Therapy randomised double-blind, placebo-controlled	Therapeutic effect of beta-C and retinol on oral leukoplakia	Tobacco chewers with premalignant oral leukoplakia	180 mg beta-C / d + 100.000 IU Vit. A / d for 6 months; orally	beta -C induced remission of leukoplakias compared to placebo; most effective: combination of beta-C and retinol
Brandt et al. (3) (Pennington Symposium "Vitamins and Cancer Prevention" 1991)	Therapy randomised double-blind, placebo-controlled	Therapeutic effect of a combination of beta-C, Vitamin C and E on oral leukoplakia	Patients with premalignant oral leukoplakia (n = 20)	30 mg beta-C / d + 1000 mg Vit. C / d + 800 IU Vit. E / d for 3 months; orally	The vitamin combination lead to partial or complete regression of oral clinical symptoms in 60 % of the patients
Kaugars et al. (4) (Oral Surg Oral Med Oral Pathol 1994; 78:462-8)	Therapy randomised double-blind, placebo-controlled	Therapeutic effect of a combination of beta-C, Vitamin C and E on oral leukoplakia	Patients with premalignant oral leukoplakia (n = 79)	30 mg beta-C / d + 1000 mg Vit. C / d + 800 IU Vit. E / d for 9 months; orally	No effect of the vitamin supplementation on clinical improvement, which was associated with reduction of alcohol and tobacco
Mathews-Roth (5) (Ann N Y Acad Sci 1993; 691:127-38)	Therapy randomised double-blind, compared with unsupplemented control group	Therapeutic effect of beta-C on clinical symptoms of patients with EPP	EPP patients (n = 38)	180 mg beta-C orally / d for 10 weeks	Administration of beta-C benefit photosensitivity disorders
Pontiggia et al. (6) (Biomed-Pharmacother 1995; 49(5): 263-5)	Therapy randomised double-blind, compared with unsupplemented control group	Therapeutic effect of beta-C on clinical symptoms of HIV- positive patients	HIV positive patients (n = 10)	120 mg beta-C orally / d + whole body hyperthermia (42 °C, 1h / d) for 6 months	Combination of beta-C-supplementation and hyperthermia induced clinical improvement of all patients
Ahmed et al. (7) (Am J Clin Nutr 1994; 60:430-6).	Therapy randomised double-blind, compared with	Therapeutic effect of beta-C on alcohol induced liver disease	Patients with alcohol induced liver disease (n = 34)	30 mg beta-C orally / d or 60 mg -C orally /	No effect of the beta-C supplementation on clinical improve-

	unsupplemented control group			d for 3 days	ment; patients with cirrhosis had a lower plasma beta-C response
McLarty et al. (8) (Am J Clin Nutr 1995; 62(S6): S1431-8)	Therapy randomised double-blind, placebo-controlled	Therapeutic effect of beta-C on sputum atypia	Former asbestos workers with sputum atypia (n = 755)	50 mg beta-C / d + 25.000 IU Vit. A / d for 4 years; orally	Increased serum levels of beta-C and retinol without clinical signs of toxicity and without beneficial effects on sputum atypia
Umegaki et al. (10) (Am J Clin Nutr 1994; 59:409-12).	In vitro model, double-blind	Effects of beta-C on x-ray induced damage of isolated lymphocytes from healthy volunteers	Isolated lymphocytes of healthy volunteers (n = 17) after x-ray radiation (0.3 Gy/min for 2 min)	30 mg beta-C / d + 300 mg Vit. C / d for 6 days; orally	Supplementation of beta-C prevents x-ray induced DNA damage in lymphocytes (less number of x-ray induced micronuclei)
The alpha-tocopherol beta-carotene (ATBC) project. (1) Conducted by the National Cancer Institute (NCI) in collaboration with the National Public Health Institute of Finland (Heinonen et al., New England J Med 1994; 330(15): 1029-1035).	Intervention, randomised, double-blind, placebo-controlled	Preventive effects of vitamin supplements on lung and other cancers.	Finnish healthy men aged between 50 and 69 and smoking (n = 29.133)	20 mg beta-C / d or 50 mg Vit. E / d or the combination of beta-C and Vit. E for 5 to 8 years; orally	In the beta-C supplemented group, 18 % more lung cancers were diagnosed and 8 % more overall death. No preventive effect of beta-C on the incidence of lung cancer was observed. For the interaction of beta-C and Vit. E no evidence was found.
The beta-carotene and retinol efficacy chemoprevention trial (CARET). (12, 13) Conducted in six areas of the USA and founded by the NCI (Goodman et al.; Cancer Epidemiology, Biomarkers & Prevention 1993; 2:389-396 /	Intervention, randomised, double-blind, placebo-controlled	Preventive effects of beta-C and Vit. A on lung and other cancers	Healthy smoking volunteers aged between 50 and 69 years (n = 14.254 men and women)	50 mg beta-C / d + 25.000 IU Vit. A / d for 4 years; orally	Among the supplemented group, 28 % more lung cancers were diagnosed and 17 % more deaths occurred. Because these results were similar to those of the ATBC study, the intervention was stopped 21 months early.

Omenn et al.; J Natl Cancer Inst 1996; 88:1550-9).					
The Linxian Study (15) (Blot et al.; Am J Clin Nutr 1995; 62(S6): S1424-6).	Intervention, randomised, double-blind, placebo-controlled	Preventive effects of beta-C, Vit. E and selenium on oesophageal and stomach cancer	Adult nutritionally deficient volunteers (n = 29.584 men and women)	15 mg beta-C / d + 60 mg Vit. E / d + selenium for 5.25 years; orally	In the supplemented group, significant reductions of oesophageal and stomach cancers and in cancer mortality were observed.
Physicians Health Study (16) (Hennekens et al.: N Engl J Med 1996; 334:1145-9)	Intervention, randomised, double-blind, placebo-controlled	Preventive effect of beta-C and aspirin on cancer at all sites	Adult healthy physicians (n = 22.071 male)	50 mg beta-C / d + 100 mg aspirin / d; for 12 years; orally	The supplementation of the aspirin component was stopped in early 1988 due to a 44 % reduction in risk of first heart attack. The beta-carotene supplementation ended December 31, 1995.
Women's Health Study, (17) (Women's Health Study Research Group: J Myocardil Ischemia 1992; 4:27-9)	Intervention, randomised, double-blind, placebo-controlled	Effects of beta-C, Vit. E and aspirin on the incidence of lung and breast cancer	Female health professionals aged 45 years and older (n = 40.000)	50 mg beta-C / d + 600 mg Vit. E / d + 100 mg aspirin / d; ongoing; orally	Considering the harmful effects of beta-C supplementation observed in ATBS and CARET, the beta-C supplementation is being dropped.
Australian Polyp Prevention Project (19) (Mac Lennan; J Natl Cancer Inst 1995; 87:1760-6).	Intervention, randomised, double-blind, placebo-controlled	Preventive effect of beta-C, wheat bran and restriction of dietary fat on colorectal adenoma and polyps	A group of 411 patients, whose eligibility was contingent upon confirmation of at least one colorectal adenoma and removal for all polyps	20 mg beta-C / d + 25 g wheat bran/d + dietary fat 25 % of total calories; for 24 month	A significant preventive effect of the low fat diet in combination with the supplementation of wheat bran was found. No preventive effect was observed for the beta-C supplementation.
Clemens (9) (Ther Umschau 1994; 51(7):483-488)	Intervention, compared with unsupplemented control group	Effects of beta-C, Vit. E and Vit. C supplementation on clinical symptoms induced by chemo/radiotherapy in cancer patients	Cancer patients undergoing a bone marrow transplantation (n = 10)	45 mg beta-C / d + 825 mg Vit. E / d + 450 mg Vit. C / d for 3 weeks; orally prior to chemo- and radiotherapy	Increased the serum levels of the supplemented vitamins after therapy were associated with a lowered hepatotoxicity of chemo-

					/radiotherapy
Stratta et al. (26) (Gynecol Obstet Invest 1994; 37(4):246-249)	Intervention, non-randomised, compared with unsupplemented control group	Effects vit. E on fetal and maternal outcome of preeclamptic pregnants	Preeclamptic patients undergoing a conventional therapy (n = 36)	100 - 300 mg Vit. E / d; orally	No beneficial effects were observed for the vit. E supplementa-tion on the foetal outcome and on maternal hyperten- sion and proteinuria.
Jakeman and Maxwell (27) (Eur J Appl Physiol & Occ Physiol 1993; 67(5): 426-430).	Intervention, randomised, double-blind, placebo- controlled	Effect of antioxidant vitamin supplementation on muscle function after eccentric exercise	A group of 24 healthy physically active adults	400 mg Vit. E / d or 400 mg Vit. C / d 21 d prior to and for 7 d after performing a 60 min box-stepping exercise; orally	No changes were observed for the maximal voluntary contraction after the vitamin supplemen- tation. The initial phase of recovery was less in subjects supplemented with vit. C than in those supplemented with vit. E or in the placebo group.
Nierenberg et al. (28) (J Natl Cancer Inst 1994; 86(2):117-120).	Intervention, randomised, double-blind placebo- controlled	Interactive effect of beta-C and vit. E	A group of 500 patients, who had and a complete removal for colonic polyps	400 mg Vit. E / d or 25 mg beta-C / d or 1 g Vit. C / d or combination of all three agents for 9 month; orally	Altered serum concentrations were neither observed after beta-C- supplementation for vit. C nor after supplementation with vit. E plus vit. C for beta-C.
Singh et al. (29) (Am J Cardiol 1996; 77(4):232- 236)	Intervention, randomised, double-blind placebo- controlled	Effects of antioxidant vitamin supplementation on incidence of suspected myocardial infarction	Patients with suspected acute myocardial infarction (n = 63)	400 mg Vit. E / d + 1 mg Vit. C / d + 125 mg beta-C/d + 150 00 IU Vit. A / d for 28 d; orally	Angina pectoris and total arrhythmias occurred less often in patients with the combined treatment of the antioxidants beta-C, Vit. A, E and C compared to the control group.
Delamaza et al. (30) (J Am Coll Nutr 1995; 14(2): 192- 196).	Intervention, randomised, double-blind, placebo- controlled	Effect of long term vitamin E supplementation in alcoholic cirrhosis	Patients with decompensated ambulatory alcoholic liver cirrhosis (n = 33)	500 mg Vit. E / d for 1 y; orally	The vit. E supplementation did not influence hepatic laboratory parame-ters, mortality or hos- pitalisation rates, although the vit. E serum levels increa- sed in the supple- mented patients.
Greenberg et al.	Intervention,	Preventive effect	A group of 864	400 mg Vit. E / d	Neither the combi-

(31) (New England J Med 1994; 331(3): 141-147).	randomised, double-blind controlled	of beta-C and vit. E and vit. C on colorectal adenoma	patients, who had adenomatous polyps	or 1 mg Vit. C / d or 25 mg beta-C / d or combination of all three agents for 4 y; orally	ned supplementation nor the single treatment appeared to be effective in the prevention of any subtype of polyp defined by size or location.
Kessopoulou et al. (32) (Fertility & Sterility 1995; 64(4): 825-831)	Therapy, randomised, double-blind placebo-controlled cross over	Effect of vitamin E on reactive oxygen species-associated male infertility	Male patients with high levels of reactive oxygen spe-cies genera-tion in semen and a normal female partner (n = 30)	600 mg Vit. E / d for 3 months; cross over after a 1 month wash-out period; orally	Increased serum levels of vit. E were found after the sup-plementation and were accompanied by improvement of the zone binding assay as the one of the sperm function test.
Meydani et al. (33) (Am J Clin Nutr 1994; 60(5): 704-709).	Intervention, randomised, double-blind, placebo-controlled	Assessment of the safety of high-dose, short-term supplementation with vitamin E in healthy older adults	Healthy elderly subjects (> 60 y) (n = 32)	800 mg Vit. E / d for 30 d; orally	The subjects reported no side effects due to the supple-ment. No effect on body weight, serum parameters (albumin, to-tal cholesterol, triglycerides), renal and hepatic function and on creatinine clearance was observed after treatment.
Chung et al. (34) (Clinical Hemorheology 1995; 15(5): 775-782).	Therapy, randomised, controlled	Effects of vit. E on retinal capillary blood flow in female NIDDM with retinopathy	Female NIDDM with retinopathy (n = 7)	900 mg Vit. E / d for 3 months; orally	The vit. E supple-mentation resulted in an improvement of blood viscosity, red cell deformability and retinal capillary blood flow velocity.
The Cambridge Heart Antioxidant Study (CHAOS) (35) (Stephens et al; Lancet 1996; 347: 781-786).	Intervention, randomised, double-blind, placebo-controlled	Effects of vitamin E in patients with coronary disease	Patients with angiographically proven coronary athero-sclerosis (n = 2002); end-point of treat-ment: cardio-vascular death and/or myocar-dial infarction	800 IU Vit. E / d or 400 IU Vit. E / d for a median of 510 d (range 3 - 981 d); orally	The vit. E supple-mentation resulted in a risk reduction of myocardial infarction and in a statistically non-significant excess of cardiovascular death.
Paolisso et al. (36)	Therapy, randomised,	Effect of pharmacological	Elderly patients (> 70 y) with	900 mg Vit. E / d for 4 months;	The vit. E adminis-tration was associa-

(Am J Clin Nutr 1995; 61(4): 848-852)	double-blind placebo-controlled cross over	doses of vitamin E on coronary heart disease in elderly	stable effort angina (n = 30)	orally	ted with higher non oxidative glucose metabolism. Plasma concentrations of insulin, triglycerides and the ratio of LDL to HDL cholesterol were lowered.
Hartmann et al. (37) (Mut Res Lett 1995; 346(4): 195-202)	Therapy, compared to control group	Effect of vitamin E on exercise induced white blood cell DNA damage	Healthy volunteers who did a treadmill run until exhaustion (n = 5)	1200 mg Vit. E/d for 14 d prior to the run; orally	The vit. E treatment prevented the exercise induced DNA damage completely in 4 out of 5 subjects.

Summary and conclusion

The Committee has reviewed in detail the published studies on the use of beta-carotene supplementation in combination with tocopherol, retinol and ascorbate in clinical and chemopreventive trials. In clinical trials doses up to 180 mg/day and in chemopreventive trials up to 50 mg/day were given.

Most of the chemopreventive trials with beta-carotene alone and in combination with tocopherol, retinol or ascorbate in well-nourished population groups showed no protective effects against malignant neoplasms, cardiovascular disease or death from all causes. On the contrary, an increase of lung cancer incidence (18-28 %) and more overall death (8-17 %) were seen in smokers ingesting over long periods of time (4-8 years) supplements of 20 mg beta-carotene per day.

The Committee cannot identify any specific explanation for these unexpected findings and therefore reconfirms its earlier concern expressed at the 107th meeting regarding the use of beta-carotene supplements.

The Committee recommends that research is initiated to resolve the issue as a matter of urgency and thereby allowing the establishment of an upper safe limit for beta-carotene intake both alone and in combination with other antioxidants to be used for the general public and for special population groups at risk.

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