Original Article

Increases in plasma lycopene concentration after consumption of tomatoes cooked with olive oil

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Lycopene is the main carotenoid in tomatoes and it has been hypothesised to be responsible for reducing the risk of some cancers and heart disease. The cooking of tomatoes with olive oil is a characteristic combination in the Southern Mediterranean diet. Previous studies have shown that the absorption of lycopene is greater from processed tomatoes than fresh tomatoes, since the processing breaks down the tomato cell matrix and makes the lycopene more available. The aim of the present study was to determine whether consumption of diced tomatoes cooked with olive oil resulted in higher plasma lycopene concentrations than consumption of diced tomatoes cooked without olive oil. Plasma lycopene concentrations were measured after 5 days on a low lycopene diet and again after a five-day dietary intervention, in healthy subjects, who consumed one meal per day of tomatoes (470 g) cooked with or without extra virgin olive oil (25 ml olive oil). There was an 82% increase in plasma *trans*-lycopene ($P < 0.001$) and a 40% in *cis*-lycopene ($P = 0.002$) concentrations in the 11 subjects who consumed tomatoes cooked in olive oil. There was no significant change in *trans*-lycopene ($P=0.684$) and a 15% increase in *cis*-lycopene ($P = 0.007$) concentrations in 12 subjects consuming tomatoes cooked without olive oil. We conclude that the addition of olive oil to diced tomatoes during cooking greatly increases the absorption of lycopene. The results highlight the importance of cuisine (i.e how a food is prepared and consumed) in determining the bioavailability of dietary carotenoids such as lycopene.

Key Words: antioxidant, lycopene, tomatoes, olive oil, cuisine, bioavailability, Mediterranean cuisine.

Introduction

Cardiovascular disease (CVD) and cancers are major causes of premature mortality in industrialized countries. Fruit and vegetable consumption and increasing concentrations of dietary carotenoids have been linked to a lower incidence of cardiovascular disease (CVD) and some cancers.[1] Epidemiological studies have shown inverse associations of chronic disease not only with dietary fruit, vegetable and carotenoid intake but also with circulating concentrations of carotenoids.[2-5] Giovannucci *et al.*,[6] demonstrated an inverse relationship between dietary intake of lycopene and prostate cancer incidence. Others have reported an inverse association between lycopene concentrations in adipose tissue and risk for myocardial infarction.[7]

The Mediterranean diet is an example of a dietary regime associated with a reported low incidence of cancers and CVD and the typical fruit and vegetables of this region (leafy green vegetables and tomatoes) are rich sources of a range of antioxidant vitamins, minerals and carotenoids.[8] Other characteristics of the Mediterranean diet include low consumption of saturated fat and high consumption of monounsaturated fat. An important component of the Mediterranean diet is the combination of olive oil with tomatoes in food preparation. Tomatoes and tomato products are typical components of the Mediterranean diet and these are a major dietary source of lycopene.[9,10] Several studies have reported that lycopene from fresh tomatoes or tomato juice is poorly absorbed in comparison with lycopene from processed tomatoes (e.g tomato paste).[11-13] Other studies have reported that boiling tomato juice in corn oil increased the uptake of lycopene compared with straight tomato juice.[14]

No studies have investigated the effects on lycopene bioavailability following the heating of diced tomatoes with olive oil (a traditional combination amongst Southern European populations). The aim of this study was to determine whether the plasma lycopene level is significantly increased following consumption of diced tomatoes cooked with olive oil compared with consumption of diced tomatoes cooked without olive oil.

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Subjects and methods

Subjects

Subjects (N = 23) were 18-60 yrs of age, of Anglo-Celtic origin and in good health. They had no history of bowel, liver or pancreatic disease and were not taking any medication that would interfere with the absorption of fat-soluble vitamins and carotenoids. Participants did not take vitamin or minerals supplements for four weeks prior to the beginning of the study. None of the subjects smoked. Subjects were randomly assigned to olive oil (N =11, 5 males, 6 females) or no olive oil (N =12, 6 males, 6 females) groups. Exclusion criteria were plasma cholesterol ≥ 7.0 mmol/L or triacylglycerol ≥ 3.0 mmol/L. The study was approved by the Deakin University Ethics Committee. All subjects were informed of the purpose and procedure of the study and gave written consent.

Study design

The study duration was two weeks. Participants completed a food frequency questionnaire to assess lycopene intake over the previous month. The volunteers were instructed on how to adhere to a low background lycopene diet (avoidance of tomatoes, watermelon, pink grapefruit, pink guava, apricots, rose hip, paw paw and their products) which commenced 5 days prior to the test tomato diet periods. Thereafter, for the next five days the subjects consumed 470g of cooked tomatoes daily at lunchtime. The subjects were instructed to consume their typical evening and morning meals except for exclusion of sources of lycopene. During the five day intervention periods, subjects completed a 5-day, semi-quantitative food record, which was analysed using Diet 3 (Version 3.22; Xyris Software Australia, Highgate Hill, Queensland, Australia). Blood samples were taken after an overnight fast at the end of the 5-day low lycopene period and on the morning following the last tomato meal.

Preparation of test meals

Sun-ripened tomatoes of medium to large size, and 3/4 to full color were used for the study. They were purchased from a local distributor as a single batch. Each tomato was chopped into sixteenths and 500g of raw tomatoes were cooked by stewing in a stainless steel pot over a gas burner at medium heat for 10 min. The tomatoes cooked with olive oil were prepared in exactly the same way except for the addition of 25 ml of cold-pressed olive oil (Caroli; Enoteca Sileno, North Carlton, VIC.) before the tomatoes were cooked. After cooking, the two different types of tomato meals were packaged, weighed (average 470g) in containers and stored frozen at -20^{o}C. The lycopene content of each meal was approximately 20mg.[9,10,15,16] The subjects were instructed to adhere to the following protocol for all the tomato meals: the container with tomatoes was thawed overnight in the refrigerator. Meals were then heated for 2 min in a microwave oven set on high heat, with stirring at 1 min intervals. The whole of the meal was consumed along with 2 slices of white bread to mop up remaining juice and/or oil. Water and weak black coffee were allowed to be consumed with the meal. Volunteers waited two hours before consuming any other foods or beverages. An instruction booklet was provided to subjects in order to standardise thawing, heating and consumption of meals.

Analytical procedure

Plasma cholesterol and triacyglycerol levels were measured using standard enzymatic techniques. Inter-assay CV's for cholesterol and triacylglycerols were 7% and 6%, respectively. Lipid soluble antioxidants were measured as described previously.[17] Briefly, analytes were extracted from 200µl of plasma, after the addition of 200µl ethanol containing internal standards (tocopherol acetate and retinol acetate), using 1ml of hexane. Extracts were dried under nitrogen and reconstituted first in 30µl chloroform and then 70µl methanol: acetonitrile added. All procedures were carried out under red light conditions. Fifty µl of sample was injected onto a 250 x 4.6 mm Spherisorb column (5 µm; Activon, Melbourne, Vic) using a Waters 2690 solvent delivery system (Waters Australia, Box Hill, Vic). The following conditions were applied[17]: solvent A consisted of methanol plus 0.05% ammonium acaetate, solvent B was acetonitrile plus 0.1% triethylamine and solvent C was chloroform. Three linear gradient steps were used: from 0 to 5 min, solvent A remained at 50%, solvent B decreased from 50% to 44% and solvent C increased from 0% to 6%; from 5 to 16 min, solvent A increased from 50 to 55%, solvent B decreased from 6% to 15%; and from 18 to 21 min the solvent combinations returned to 50% solvent A and 50% solvent B. The recovery of the column using chloroform as a modifier was greater than 97%.[17] Absorbance data was collected at three wavelengths using a model 996 Photoiode Array detector (Waters): retinol and retinol acetate at 325 nm; α-tocopherol and tocopherol acetate at 295nm; and lutein/zeaxanthin, cryptoxanthin, lycopene, α-carotene and β-carotene at 450 nm. Interassay CV was < 9 % for all analytes except *cis*-lycopene (CV = 11%).

Statistical analyses

An independent t-test (2-tailed) was used to compare the equality of means for the two groups with respect to age, total cholesterol, HDL, triacylglycerol, BMI and dietary intake of selected nutrients at baseline. Repeated measures analysis of variance was used to examine changes in plasma lycopene concentrations over time, within and between the two diets. Age, BMI, total cholesterol, HDL cholesterol, dietary fibre intake and other antioxidants were entered as covariates in the model. These were found to be non significant and so were omitted from the final analysis unless otherwise noted. SPSS (Version 8, SPSS Inc, Chicago IL, USA) was used for all statistical analyses. Since there was large interindividual variations for plasma carotenoid, tocopherol and triacylglycerol concentrations, the data for these were log-transformed prior to statistical analysis, and the results for these are presented as geometric mean (95% confidence interval [CI]).

Results

The two diet groups were similar with respect to plasma total cholesterol, HDL cholesterol and triacylglycerol concentrations at the end of the low lycopene period (Table 1). BMI was greater for the no olive oil group however this was not significant (P <0.1). Food records

of the foods consumed for the five days of the tomato intervention period, other than the daily tomato meals, were analysed for specific nutrients and this showed there was no difference in the mean daily intake between the control and test group with respect to intake of total fat, monounsaturated fat (except for that used in the tomato meals), polyunsaturated or saturated fat (Table 2). Dietary fibre intake was higher for the no olive oil group however this was not significant (Table 2).

There was a significant increase in plasma concentration of lycopene in the olive oil group compared with the no olive oil group, $P<0.008$ (Fig. 1). The magnitude of the change in the plasma lycopene concentration in the olive oil group ranged from 28 to 531 nmol/L.

There was also a significant increase in the plasma concentration of *trans*-lycopene over the five day dietary period for the olive oil diet group (Table 3). There was an 82% increase in *trans*-lycopene among the olive oil group ($P<0.001$) but no significant change in the no olive oil group ($P=0.684$). There was a significant change in plasma concentration of *cis*-lycopene over the dietary period but the interaction term between change in concentration and dietary group was significant only at $P<0.1$ (Table 3). There was a 40% increase in the plasma *cis*-lycopene in the olive oil group ($P = 0.002$) and a 15% increase in the no olive oil group ($P = 0.007$). At the end of the low lycopene period (baseline), the ratio of *trans: cis*-lycopene in the plasma was similar for each group (1.4:1) Following the five day tomato diet periods, the ratio of *trans:cis*-lycopene in plasma was significantly higher in the olive oil group (1.8:1) compared with the no olive oil group (1.4:1, $P = 0.004$).

There were increases in plasma concentrations of lutein plus zeaxanthin and β-carotene during the dietary period but there was no significant interaction between these changes and dietary group (Table 3), indicating similar increases in both the olive oil and no olive oil groups. The percentage increases for lutein and zeaxanthin and α-carotene in both diet groups were approximately 12%. There were no significant changes in α-tocopherol, retinol or α-carotene concentrations throughout the study in either group. There was a significant decrease in the plasma γ-tocopherol concentration in both diet groups (Table 3).

Discussion

The major finding of the present study was that the addition of a moderate amount of olive oil to diced tomatoes during cooking significantly increased the plasma concentration of lycopene, which could be interpreted to mean there was an increased bioavailability of lycopene. This was demonstrated by increases in plasma concentrations of both *cis*- and *trans*-lycopene after 5 days on the diet which included tomatoes cooked with olive oil. In the absence of olive oil, there were no significant increases in the plasma concentrations of total lycopene or *trans*-lycopene and only a small increase in *cis*-lycopene, despite an identical intake of tomatoes in both diet groups. Although plasma lycopene concentrations increased in all subjects consuming tomatoes with olive oil, there was wide variability in responsiveness, consistent with observations of others.[12,14]

Table 1. Characteristics of subjects in the two diet groups[1]

	Diet group		
	Tomato and no olive oil	Tomato with olive oil	*P* value
N	12	11	
Gender			
Female	6	6	
Male	6	5	
Age (yr)	34 ± 4	39 ± 4	0.459
BMI (kg/m^2)	25.7 ± 1.0	22.8 ± 1.2	0.070
Total cholesterol (mmol/L)	4.7 ± 0.2	5.1 ± 0.3	0.259
HDL cholesterol (mmol/L)	1.09 ± 0.19	0.93 ± 0.13	0.492
Triacylglycerols (mmol/L)	1.27 (1.03 – 1.58)	1.58 (1.22 – 2.04)	0.220

[1]Data shown as Mean ± SEM except for triglycerides which are geometric mean (95% CI). The plasma analytes were measured following the five day low lycopene diet.

Table 2. Dietary fat and fibre intake of the background diet during the five day diet periods where subjects consumed a tomato meal daily which had been cooked with and without olive oil[1, 2]

	Diet group		
Nutrient	Tomato & no olive oil (*N*=12)	Tomato with olive oil (*N*=11)	*P* value[3]
Total fat (g/MJ)	8.0 ± 1.3	8.5 ± 0.4	0.735
Monounsaturated fat (g/MJ)	2.9 ± 0.4	3.0 ± 0.1	0.671
Polyunsaturated fat (g/MJ)	1.0 ± 0.1	1.1 ± 0.1	0.413
Saturated fat (g/MJ)	3.3 ± 0.7	3.6 ± 0.3	0.701
Dietary fibre (g/MJ)	4.4 ± 0.9	2.6 ± 0.3	0.079

[1]Data shown as nutrient density, Mean ± SEM; [2] The five day food record was calculated excluding the tomato meals and the olive oil. [3]Unpaired t-test.

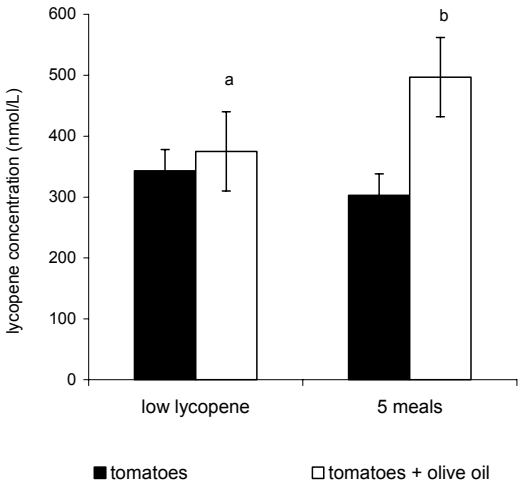

Figure 1. Total plasma lycopene concentrations before and after consuming tomato meals prepared with and without olive oil for five days. [ab] $P=0.008$, ANOVA (repeated measures)

Table 3. Plasma carotenoid and tocopherol concentrations, pre- and post- 5 day tomato diet periods[1]

Antioxidant	Time	Diet group		P_{time}[2]	$P_{oil\ x\ time}$[2]
		Tomato & no olive oil	Tomato with olive oil		
trans-lycopene	*Pre*	201 (146 - 278)	175 (125 - 245)		
	Post	209 (147 - 298)	319 (220 - 462)	<0.001	<0.001
cis-lycopene	*Pre*	142 (105 - 193)	128 (93 - 175)		
	Post	166 (119 - 231)	178 (126 - 252)	<0.001	0.072
Lutein/zeaxanthin	*Pre*	395 (275 - 569)	350 (239 - 512)		
	Post	448 (308 - 650)	388 (263 - 573)	0.013	0.804
β-carotene	*Pre*	367 (250 - 539)	424 (283 - 634)		
	Post	413 (272 - 627)	473 (306 - 731)	0.010	0.905
α-carotene	*Pre*	133 (96 - 184)	123 (88 - 173)		
	Post	132 (92 - 190)	118 (81 - 172)	0.608	0.639
cryptoxanthin	*Pre*	171 (107 - 275)	189 (115 - 309)		
	Post	182 (119 - 279)	177 (114 - 276)	0.999	0.155
α-tocopherol	*Pre*	24.1 (19.0 - 31.8)	24.7 (18.9 - 32.4)		
	Post	24.7 (18.7 - 32.6)	25.4 (18.9 - 34.0)	0.528	0.647
γ-tocopherol	*Pre*	2.1 (1.6 - 2.9)	2.1 (1.5 - 3.0)		
	Post	1.8 (1.3 - 2.4)	1.7 (1.2 - 2.5)	0.022	0.841
retinol	*Pre*	3.1 (2.6 – 3.5)	2.7 (2.3 – 3.2)		
	Post	3.1 (2.6 – 3.6)	2.7 (2.2 – 3.3)	0.822	0.915

[1]Data are geometric mean (95% CI) except for retinol which are arithmetic mean (95% CI). Concentrations are nmol/L except for α- and γ-tocopherol and retinol which are μmol/L; [2]Repeated measures ANOVA.

Factors other than the presence of olive oil, which may contribute to variations in plasma lycopene are age, gender, adiposity, plasma lipid concentrations and intake of macronutrients such as dietary fibre.[18-22] The groups were well matched for all these factors except that the olive oil group had a lower BMI than the no olive oil group. However, the inclusion of BMI in the ANOVA model suggested it was not a significant determinant of plasma total lycopene concentrations in these subjects. In any case, an association between adiposity and plasma lycopene has not been consistently observed.[7,20,22] The groups were also well matched for their background diets whilst consuming the tomato meals.

The enhanced absorption of lycopene from tomatoes cooked with olive oil is presumably due to its lipophilic nature and the extraction of lycopene into the lipophilic phase during cooking, as previously reported.[12] Cooking may allow the fat to act as a vehicle for absorption.[23] Fat is required for the incorporation of lycopene into the lipid micelles in the small intestine, and the lipid micelles will only form when sufficient fat is present to stimulate the release of bile acids from the gall bladder. The heating and chopping processes may also breakdown the cell matrix and make the lycopene more physically available for absorption. This has been supported by bioavailability studies on β-carotene from carrots.[24] In addition, co-ingestion of β-carotene, which is also found in tomatoes, may enhance the absorption of lycopene, at least acutely.[25] The present results are consistent with those of Stahl and Sies[14] who investigated the uptake of lycopene from processed and unprocessed tomato juice. They found that the uptake of lycopene was greater from heat processed tomato juice (boiled with 1% corn oil) than from unprocessed tomato juice. Other studies have also shown that processing carotenoid-rich foods with heat and the addition of oil increases carotenoid (lycopene and β-carotene) concentrations in chylomicra and serum.[12,26]

The ratio of *trans:cis*-lycopene in the plasma samples in all subjects was 1.4:1 after the five day low lycopene period, which increased significantly to 1.8:1 in the olive oil group at the end of the tomato period compared with 1.4:1 in the no olive oil group. By way of comparison, the ratio of *trans:cis* lycopene in raw tomatoes is typically 9:1.[11] If the lycopene isomers were absorbed in the ratio in which they exist in tomatoes, the *trans:cis* lycopene values in plasma after the tomato meal might be expected to have been higher than 1.8:1. As both *cis*- and *trans*-lycopene are stable in plasma for at least 24 h *ex vivo*,[17] stereoisomerisation in plasma after collection does not explain this disparity between food and plasma. This observation therefore suggests either preferential absorption of the *cis*-isomer into the human body, *in vivo* isomerisation or more rapid degradation/excretion of *trans*-lycopene. Data in the literature report that *cis*–lycopene is more soluble in bile acid micelles and is also preferentially incorporated into the chylomicrons compared with *trans*-lycopene.[26] The small increase in plasma *cis*-lycopene concentration even in the no-olive oil group, where there was no change in *trans*-lycopene, supports the possibility of preferential absorption. Total lycopene concentration in foods is not affected by cooking[12,23,27] but the heating of tomatoes via the process of stewing (1 hr) has been reported to alter the geometric isomer profile of lycopene, resulting in isomerisation from *all trans*- to *cis*-lycopene.[11,23] However, other studies have reported no such change.[28] The *cis*-isomer is apparently more soluble in lipid than the *all trans* form.[29] In the present study, the tomatoes were cooked in the presence of the olive oil for only 10 minutes which may not have been sufficient to alter the proportion of *cis*-lycopene in the sample.[11]

The functional implications of increased plasma levels of lycopene have been explored by Hadley *et al.,*[30] and Agarwal *et al.*[11] Both groups showed that consumption of tomatoes increased the plasma lycopene levels and significantly reduced LDL oxidation,[11,30,31] and improved the antioxidant activity of the plasma. In another study by Lee *et al.*,[32] tomato products (canned tomatoes and tomato

soup) were consumed with either extra virgin olive oil or sunflower oil. While the plasma lycopene levels increased in both treatment groups, only the group using the olive oil showed improvements in the plasma antioxidant activity.

In conclusion cooking tomatoes with olive oil increased bioavailability of dietary lycopene. This occurred when tomatoes and olive oil were combined in quantities similar to that used by Southern European migrants in Australia. The results highlight the importance of cuisine (i.e how a food is prepared and consumed) in determining the bioavailability of dietary carotenoids. This research may have important implications for food processing and for populations with limited access to fresh foods.

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References

1. Hercberg S, Galan P, Preziosi P, Roussel AM, Arnaud J, Richard MJ, Malvy D, Paul-Dauphin A, Briancon S, Favier A. Background and rationale behind the SU.VI.MAX Study, a prevention trial using nutritional doses of a combination of antioxidant vitamins and minerals to reduce cardiovascular diseases and cancers. SUpplementation en VItamines et Mineraux AntioXydants Study. Int J Vitam Nutr Res 1998; 68:3-20.
2. Burney PG, Comstock GW, Morris JS. Serologic precursors of cancer: serum micronutrients and the subsequent risk of pancreatic cancer. Am J Clin Nutr 1989; 49: 895-900.
3. Gey KF. Prospects for the prevention of free radical disease, regarding cancer and cardiovascular disease. Br Med Bull 1993; 49: 679-99.
4. Steinmetz KA, Potter JD. Vegetables, fruit, and cancer prevention: a review. J Am Diet Assoc 1996; 96: 1027-39.
5. Coudray C, Roussel AM, Mainard F, Arnaud J, Favier A. Lipid peroxidation level and antioxidant micronutrient status in a pre-aging population; correlation with chronic disease prevalence in a French epidemiological study (Nantes, France). J Am Coll Nutr 1997; 16:584-91.
6. Giovannucci E, Ascherio A, Rimm EB, Stampfer MJ, Colditz GA, Willett WC. Intake of carotenoids and retinol in relation to risk of prostate cancer. J Natl Cancer Inst 1995;87:1767-76.
7. Kohlmeier L, Kark JD, Gomez-Gracia E, Martin BC, Steck SE, Kardinaal AF, Ringstad J, Thamm M, Masaev V, Rimersma R, Martin-Moreno JM, Huttunen JK, Kok FJ. Lycopene and myocardial infarction risk in the EURAMIC Study. Am J Epidemiol 1997;146:618-26.
8. Ferro-Luzzi A, Sette S. The Mediterranean Diet: an attempt to define its present and past composition. Eur J Clin Nutr 1989; 43 Suppl 2:13-29.
9. Mangels AR, Holden JM, Beecher GR, Forman MR, Lanza E. Carotenoid content of fruits and vegetables: an evaluation of analytic data. J Am Diet Assoc 1993; 93: 284-96.
10. Khachik F, Beecher GR, Goli MB, Lusby WR. Separation and quantitation of carotenoids in foods. Methods Enzymol 1992; 213: 347-59.
11. Agarwal A, Shen H, Agarwal S, Rao AV. Lycopene content of tomato products: its stability, bioavailability and in vivo antioxidant properties. J Med Food 2001;4:9-15.
12. Gartner C, Stahl W, Sies H. Lycopene is more bioavailable from tomato paste than from fresh tomatoes. Am J Clin Nutr 1997; 66: 116-22.
13. Richelle M, Bortlik K, Liardet S, Hager C, Lambelet P, Baur M, Applegate LA, Offord EA. A food-based formulation provides lycopene with the same bioavailability to humans as that from tomato paste. J Nutr 2002; 132: 404-8.
14. Stahl W, Sies H. Uptake of lycopene and its geometrical isomers is greater from heat-processed than from unprocessed tomato juice in humans. J Nutr 1992; 122: 2161-6.
15. Hart DJ, Scott KJ. Development and Evaluation of an HPLC Method for the Analysis of Carotenoids in Foods, and the Measurement of the Carotenoid Content of Vegetables and Fruits Commonly Consumed in the UK. Food Chemistry 1995; 54:101-111.
16. Scott KJ, Thurnham DI, Hart DJ, Bingham SA, Day K. The correlation between the intake of lutein, lycopene and beta-carotene from vegetables and fruits, and blood plasma concentrations in a group of women aged 50-65 years in the UK. Br J Nutr 1996; 75: 409-18.
17. Su Q, Rowley KG, O'Dea K. Stability of individual carotenoids, retinol and tocopherols in human plasma during exposure to light and after extraction. J Chromatogr B Biomed Sci Appl 1999; 729: 191-8.
18. Albanes D, Virtamo J, Taylor PR, Rautalahti M, Pietinen P, Heinonen OP. Effects of supplemental beta-carotene, cigarette smoking, and alcohol consumption on serum carotenoids in the Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study. Am J Clin Nutr 1997; 66:366-72.
19. Vogel S, Contois JH, Tucker KL, Wilson PW, Schaefer EJ, Lammi-Keefe CJ. Plasma retinol and plasma and lipoprotein tocopherol and carotenoid concentrations in healthy elderly participants of the Framingham Heart Study. Am J Clin Nutr 1997; 66: 950-8.
20. Brady WE, Mares-Perlman JA, Bowen P, Stacewicz-Sapuntzakis M. Human serum carotenoid concentrations are related to physiologic and lifestyle factors. J Nutr 1996; 126: 129-37.
21. Olmedilla B, Granado F, Blanco I, Rojas-Hidalgo E. Seasonal and sex-related variations in six serum carotenoids, retinol, and alpha-tocopherol. Am J Clin Nutr 1994; 60: 106-10.
22. Virtanen SM, van't Veer P, Kok F, Kardinaal AF, Aro A. Predictors of adipose tissue carotenoid and retinol levels in nine countries. The EURAMIC Study. Am J Epidemiol 1996; 144: 968-79.
23. Stahl W, Sies H. Lycopene: a biologically important carotenoid for humans? Arch Biochem Biophys 1996; 336: 1-9.
24. Rock CL, Lovalvo JL, Emenhiser C, Ruffin MT, Flatt SW, Schwartz SJ. Bioavailability of beta-carotene is lower in raw than in processed carrots and spinach in women. J Nutr 1998; 128: 913-6.
25. Johnson EJ, Qin J, Krinsky NI, Russell RM. Ingestion by men of a combined dose of beta-carotene and lycopene does not affect the absorption of beta-carotene but improves that of lycopene. J Nutr 1997; 127:1833-7.
26. Boileau AC, Merchen NR, Wasson K, Atkinson CA, Erdman JW, Jr. Cis-lycopene is more bioavailable than trans-lycopene in vitro and in vivo in lymph-cannulated ferrets. J Nutr 1999;129: 1176-81.

27. Khachik F, Goli MB, Beecher GR, Holden J, Lusby WR, Tenorio MD.. Effect of food preparation on qualitative and quantitative distribution of major carotenoid constituents of tomatoes and several green vegetables. J Agric Food Chem 1992; 40: 390-398.
28. Nguyen ML, Schwartz SJ. Lycopene: Chemical and biological properties. Food Technology 1999; 53:38-45.
29. Gerster H. The potential role of lycopene for human health. J Am Coll Nutr 1997; 16:109-26.
30. Hadley CW, Clinton SK, Schwartz SJ. The consumption of processed tomato products enhances plasma lycopene concentrations in association with a reduced lipoprotein sensitivity to oxidative damage. J Nutr 2003; 133: 727-32.
31. Agarwal S, Rao AV. Tomato lycopene and low density lipoprotein oxidation: a human dietary intervention study. Lipids 1998; 33: 981-4.
32. Lee A, Thurnham DI, Chopra M. Consumption of tomato products with olive oil but not sunflower oil increases the antioxidant activity of plasma. Free Radic Biol Med 2000; 29: 1051-5.