

Change in Carotenoids and Antioxidant Vitamins in Tomato as a Function of Varietal and Technological Factors

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The change in the carotenoid and bioantioxidant content of tomato as a function of varietal and technological factors was investigated in the present work. No great differences were found between cultivars for fresh consumption (salad tomatoes) and those for processing in ascorbic acid content. The concentration of ascorbic acid ranged between 14.6 and 21.7 mg/100 g fresh weight of ripe tomato fruit. Processing cultivars contained higher amounts of tocopherols, particularly α -tocopherol than tomatoes for fresh consumption. Significant differences could be obtained between the examined varieties with regard to carotenoid concentration. The different tomatoes varied not only in the total carotenoid content but also in the qualitative distribution of some pigments such as lycopene, β -carotene and lutein. During heat-based processing, ascorbic acid, tocopherols, and carotenoids showed different role and response. Ascorbic acid, α -tocopherol quinone, and β -carotene were the most susceptible components toward thermal degradation.

Keywords: Carotenoids; vitamin; tomatoes; technology

INTRODUCTION

Tomato is an important agricultural commodity worldwide. Because of their year-round availability, tomato and tomato products merit attention, even in terms of value of micronutrients existing at low concentration. It contains in addition to the vital carotenoids considerable amounts of vitamin C and vitamin E (Abushita et al., 1997).

Recent epidemiological studies indicated that carotenoids, vitamin E, and vitamin C are among the constituents of diet postulated to play a preventive role in cancer (Hennekens, 1994; Garewal, 1995) and heart diseases (Gaziano, 1994; Pandey et al., 1995). Therefore, recommendations have been made to increase daily intake of fruits and vegetables rich in these nutrients to lower risk of cancer and cardiovascular diseases (American Cancer Society, 1984; Steinmetz and Potter, 1991; Block et al., 1992).

On the basis of these facts, many epidemiological studies have been conducted to investigate the role of tomato and tomato products in lowering risk of several cancers. Giovannucci (1999) reviewed in detail the results of these studies, which implied that intake of tomato products and plasma level of lycopene is consistently associated with a lower risk of a variety of cancers. In studies by Stahl and Sies (1992) and Gartner et al. (1997), lycopene has been found more bioavailable from heat-processed tomato products than from fresh tomatoes. The reasons for this are, so far, unknown.

The benefits of tomato and tomato products have been attributed mostly to their carotenoid content. Among carotenoids found in human serum, tomato products contribute to nine. In human diet, tomatoes and tomato products are the predominant sources of lycopene, which has been found to be available for antioxidant properties (Stahl and Sies, 1996). Due to its stereochemical proper-

ties (Britton, 1995) and ability to be efficient quencher of singlet oxygen and free radical (Di Mascio et al., 1989; Woodall et al., 1997; Mortensen and Skibsted, 1997), lycopene is regarded as bioantioxidant with high biological activity in the different tissues of human body.

The second predominant carotenoid in tomatoes is lycopene epoxide, an oxidation product of lycopene. Its biological and technological role is not clarified yet, but Khachik et al. (1992) reported that epoxides of carotenoids are not present in the extracts from human plasma.

β -Carotene and lutein are present in all of the tomato products. β -Carotene is of special interest due to its being the main provitamin A, and unusual antioxidative activity of β -carotene and lutein has been associated with reduced risk of lung cancer (Sies, 1991). A study by Oshima et al. (1996) showed that supplementation with such carotenoids inhibits singlet oxygen-mediated oxidation of human plasma low-density lipoprotein, thereby reducing risk of cardiovascular diseases.

The objective of this work was to extensively examine the differences between different genotypes of tomato in their carotenoid and antioxidant vitamin content and to study the effect of tomato processing on such vital micronutrients.

MATERIALS AND METHODS

Chemicals Used. All organic solvents used for the separation of carotenoids, tocopherols, and ascorbic acid were of HPLC grade and purchased from Merck (Germany). Other organic solvents and chemicals used in the extraction procedures were of analytical grade and from Reanal (Hungary). Standard ascorbic acid, tocopherols, and β -carotene as well as tetrabutylammonium hydroxide (TBA-OH) and butylated hydroxytoluene (BHT) were purchased from Sigma (St. Louis). Doubly distilled water was used in preparation of mobile phase of the paired-ion chromatography of ascorbic acid.

Different Varieties of Tomato. Ripe fruits of 12 tomatoes for fresh consumption (salad tomatoes) and 15 processing cultivars were obtained from the experimental station of the

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Table 1. Separation Conditions and Validation Parameters for the HPLC Determination of Carotenoids, Ascorbic Acid, and Tocopherols

parameters	conditions		
	carotenoids	ascorbic acid	tocopherols
separation column ^a	Lichrosorb C-18 6 μm , 46 \times 250 mm	Spherisorb ODS-2 10 μm , 46 \times 250 mm	Lichrosorb 10 μm , 46 \times 250 mm
eluent ^b	acetonitrile–2-propanol–methanol–water, 39:52:5:4	0.1 M KH_2PO_4 –methanol–TBAOH, ^c 97:3:0.05, pH 2.75	hexane–ethanol, 99.5:0.5
flow rate	0.9 mL/min	1 mL/min	1.2 mL/min
detection	visible 450 nm	UV 245 nm	fluorescence (ex = 295 nm; em = 320 nm)
validation			
linearity range ^d	0.0–60 $\mu\text{g}/\text{mL}$	0.0–66 $\mu\text{g}/\text{mL}$	0.0–50 $\mu\text{g}/\text{mL}$
precision (S_r) ^e	2.5–3.1	4.5–5.2	3.0–3.8
recovery (%) ^f	99–101	95–98	93–95
detection limit value ^g			
LOD	0.05	0.03	0.02
LOQ	0.17	0.11	0.09
reference	Biacs and Daood, 1994	Daood et al., 1994	Speek et al., 1985

^a The columns were obtained from Grom (Germany). ^b Different eluents were prepared on the basis of volume ratios (v/v). ^c TBAOH: tetrabutylammonium hydroxide. ^d Calibration range of ascorbic acid, β -carotene, and α -tocopherol with $r = 0.999$, 0.980, and 0.995, respectively. ^e Precision test was based on relative standard deviation (S_r) for five replications of the same sample. ^f Recovery test was based on spiking a well homogenized sample of ripe tomato fruit at 10 $\mu\text{g}/\text{g}$ with each standard material and estimating the difference between spiked and nonspiked (control) samples. ^g Limit of detection (LOD) and limit of quantification (LOQ) are the concentrations ($\mu\text{g}/\text{mL}$) of the solutions, which provide a signal/noise (S/N) ratio of 3/1 and 10/1, respectively.

Department of Horticulture, University of Gödöllő (Gödöllő, Hungary). Plants of different varieties were grown outside under the same field and agricultural conditions [calcareous sandy soil containing 5–10% clay and 1% humus, irrigation with a total of 130 mm and traditional fertilization] in 1998. Two kilogram ripe fruit samples were taken (in triplicate) from each variety and stored at refrigeration temperature during transfer to the laboratory.

Tomato Processing. Tomato processing was carried out under the conditions of Gold Pheasant (Aranyfácán) canning factory (Hatvan, Hungary). Tomato fruits of Draco variety were brought from one of the farms belonging to the factory (near research fields of University of Gödöllő) and directly processed. The processing included washing, chopping, hot-break extraction, sieving, vacuum evaporation, filling, sterilization, and storage. Hot-break extraction was performed at 90 °C for 5–10 min depending on the rate of feeding. The extract is continuously transferred to the roller sieves to remove seeds and peels. The sieved extract is kept at temperature between 70 and 80 °C in a stainless steel container before dehydration. The water was then evaporated under vacuum at 60–70 °C for approximately 4 h to reach total soluble solid content of 28% (Brix value). The final paste was packaged in canes of 1 kg net weight and sterilized at 100 °C for 30 min in a heated tunnel. Six samples of 2 kg weight were taken at 10 min intervals from washed raw material, hot-break extract, and sterilized paste and analyzed for their carotenoid, tocopherol, and ascorbic acid content.

Extractions. Brown-colored conical flasks, round-bottom flasks, and separatory funnels were used in the different analyses to avoid light-catalyzed degradation of photosensitive vitamins.

To extract carotenoids and tocopherols, tomato fruits were cut into quarters, and half of the batch was homogenized in a Waring Blender with maximum speed. Ten milliliters of the homogenate was taken, in duplicate, and disintegrated in a crucible mortar in the presence of 1 g of quartz sand. The extraction was carried out by a previously described (Abushita et al., 1997) method in which methanol was added first to catch water and make easier the transfer of lipophilic carotenoids and tocopherol to the less polar solvent in the subsequent step. A mixture of 60:20 v/v carbon tetrachloride-methanol containing (0.5%) butylated hydroxytoluene (BHT) was added, and the mixture was shaken for 15 min. The lower colored layer was separated in a separatory funnel and dried on anhydrous

Na_2SO_4 . The solvent was then evaporated under vacuum by rotary evaporator at maximum 40 °C. The residues were either redissolved in an aliquot of the HPLC eluent for carotenoid analysis or applied for saponification procedure for analyses of tocopherols.

Saponification of Tocopherols. To the extracted pigment and tocopherol fraction, 5 mL of saturated methanolic KOH, 0.5 g ascorbic acid, and 20 mL of methanol were added. The mixture was then saponified by refluxing for 30 min at the boiling point of methanol. After cooling the flask, 15 mL of salted water were added and the analogues of tocopherol were extracted twice with 40 mL of analytical-grade *n*-hexane in a separatory funnel. The hexane fractions were collected, washed twice with distilled water, and dried over anhydrous Na_2SO_4 . The solvent was evaporated under vacuum at 30 °C, and the residues were redissolved in 5 mL of HPLC-grade *n*-hexane for chromatographic analysis.

As for organic acid extraction, half of the tomato fruit batch was homogenized with equal weight of 4% metaphosphoric acid solution in a Waring Blender to avoid rapid oxidation of ascorbic acid. Ten milliliters of the homogenate was diluted two times with 4% metaphosphoric acid solution and shaken for 15 min. The mixture was eventually filtered through a Rudfilter MN 640 d filter paper. The first few milliliters (turbid solution) were discarded and the clear filtrate was kept at –20 °C when not directly analysed by HPLC.

HPLC Determinations. *Apparatus and Conditions.* A Beckman series liquid chromatography consisting of model 114 solvent delivery pump, a model 421 controller provided with 20 μL loop, and a model 165 variable wavelength UV–vis detector was used. To monitor tocopherols, a model RF-535 Shimadzu fluorescence detector was connected to the HPLC system. A Shimadzu model C-R2A or Waters model 740 integrator recorded the detector signals. For photodiode-array detection, a Waters model 990 chromatograph was used. The absorption spectra of carotenoid were displayed between 190 and 700 nm (for carotenoids) and 190–350 nm (for organic acids) at the rate of two spectra per second. Separation conditions for carotenoids, organic acids, and tocopherols are shown in Table 1.

Identification of Peaks. (1) Carotenoids. The peaks of the main carotenoids on a chromatogram were identified by comparing their spectral characteristics with those reported in the literature (Bauernfeind, 1981) after thin-layer chromatographic (TLC) separation on Silica gel (Daood et al., 1987).

Table 2. Ascorbic Acid and Tocopherols Contents of Different Salad Tomato Varieties Cultivated in Gödöllo (1998)

varieties	ascorbic acid (mg/100 g fresh weight)	tocopherol ($\mu\text{g}/100\text{ g fresh weight}$)			
		α -tocopherol	α -tocopherol quinone	β -tocopherol	γ -tocopherol
Monika	17	612	391	117	306
Delfine	21	326	191	110	205
Marlyn	15	322	171	93	112
Fanny	15	222	107	23	135
Tiffany	16	280	237	15	154
Alambra	15	238	137	12	109
Regulus	19	305	190	19	164
Petula	17	173	149	12	164
Diamina	15	164	115	trace	142
Brillante	18	207	96	trace	182
Furone	18	326	215	17	229
Linda	18	123	91	15	143
mean (\bar{x})	17	275	174	43	170
LSD _{5%} ^a	2.2	44	29	5.4	24
<i>P</i> ^b	≤ 0.05	≤ 0.001	≤ 0.001	≤ 0.001	≤ 0.001

^a LSD_{5%} = least significant difference at 5% probability level. ^b *P* = probability level, *n* = 3.

Identification of *cis* isomers of carotenoids was based on the appearance of extra maxima between 320 and 360 nm in the absorption spectrum of the individual peaks (Chandler and Schwartz, 1987). Epoxide and allylic hydroxyls, as in lutein, were identified by carrying out epoxide and hydroxyl tests (Bauernfeind, 1981) after thin-layer chromatographic separation. Yellow-colored carotenoids (lutein, β -carotene, and *cis*- β -carotene) were quantified as β -carotene equivalents, whereas the red-colored pigments (lycopene and lycopene epoxide) were quantified as lycopene equivalents. For this purpose, β -carotene standard and authentic lycopene prepared by TLC were used.

(2) *Tocopherols and Ascorbic Acid*. For identification of peaks the retention times and maximum absorption spectra of tocopherols and ascorbic acid were compared with those of standard materials, which were also used for quantification.

Statistical Analyses. Statistical program of Microsoft Excel computer package was used to analyze the obtained data. One-factor analysis of variance (ANOVA) was followed to determine the least significant differences (LSD) and degree of significance between varieties and processing steps. The standard deviation was calculated using Casio fx-115D scientific calculator.

RESULTS AND DISCUSSION

Data in Table 1 show the values of four parameters used in the validation of the analytical procedures that was applied for the quantitative determination of carotenoids, ascorbic acid, and tocopherols. The linearity of the six point calibration curves of standard β -carotene, ascorbic acid, and α -tocopherol was proven in a wide range of concentration. Regression analysis of the analytical data showed a linear response between peak area and concentration of each material tested in the examined ranges. The LOD values recorded for β -carotene, ascorbic acid, and α -tocopherol equaled to 0.25, 0.15, and 0.11% of the nominal concentrations, respectively. These values are enough low to allow for the sensitive and accurate quantitative analyses. Regarding the extraction recovery, a homogenized sample was fortified at 10 $\mu\text{g}/\text{g}$ for each component and extracted by the same procedure. The high recovery ranges obtained revealed the high extraction efficiency and acceptable limits of experimental loss of the tested materials. Precision of the assays as expressed in relative standard deviation (*S_r*) for five replicate measurements (extraction and HPLC separation) of the same sample supported the aforementioned conclusion on the accuracy of the assay methods used in this work.

Evaluation of Salad Tomatoes. Table 2 shows the results obtained from the analysis of ascorbic acid and tocopherol analogues in different salad tomatoes cultivated in 1998. The lowest values with regard to vitamin C in the salad cultivars were estimated with Alambra and Diamina, while the other tomatoes showed higher values ranging between 15 and 21 mg/100 g fresh weight). The values estimated for ascorbic acid in Monika and Delfine were well below those recorded for the same cultivars cultivated in 1995 in Kecskemét (Abushita et al., 1997). The reason for this variation can be ascribed to the differences in the techniques used for cultivation. In 1995, tomatoes were grown using hydro-technique with nutrient solution under plastic house conditions, while in 1998, tomatoes were grown outside in the soil with a traditional fertilization. However, the estimated values are in the range reported in the literature for some tomatoes for fresh consumption (Bajaja et al., 1990, De Serrano, 1993).

With regard to vitamin E content (α -tocopherol), among 12 varieties examined, Monika contained the highest level (612 $\mu\text{g}/100\text{ g}$). In the other varieties, α -tocopherol concentration ranged between 122 and 326 $\mu\text{g}/\text{g}$. On the other hand, concentration of α -tocopherol in Monika and Delfine cultivated in the field in 1998 was much higher than that found in the same cultivars cultivated in 1995 under plastic house and hydro-technique conditions (Abushita et al., 1997). The same held true for other analogues of tocopherol, particularly α -tocopherol quinone (the oxidation product of α -tocopherol), which merits attention because its concentration ranks number two among tocopherol derivatives in tomato fruit and has been found in human body as important metabolite (Vatassery and Smith, 1987).

The major carotenoids in all of the examined varieties were lutein, lycopene epoxide lycopene, neolycopene, and β -carotene in the order of chromatographic elution on a C18 RP-column. The epoxide test confirmed presence of lycopene epoxide and other epoxide derivatives existing at very small concentration. Because other identification possibilities are not available in our laboratories, it was difficult to achieve structural elucidation of the dominant epoxide of lycopene. According to literature, lycopene 5,6-epoxide is the major derivative of lycopene in raw and processed tomatoes (Ben-Aziz et al., 1973; Britton and Goodwin, 1975; Khachik et al., 1992; Tonucci et al., 1995). In a previous work,

Table 3. Carotenoid Content ($\mu\text{g}/100\text{ g}$ fresh weight) of Different Salad Tomato Varieties Cultivated in Gödöllo (1998)

varieties	carotenoids					total carotenoids
	lutein	lycopene epoxide	lycopene	cis-lycopene	β -carotene	
Monika	112	87	7222	112	617	8660
Delfine	77	105	6514	94	431	7659
Marlyn	83	83	5529	78	327	6534
Fanny	237	121	5260	101	285	6578
Tiffany	280	143	6229	120	337	7791
Alambra	79	96	5396	84	334	6274
Regulus	90	107	6592	79	548	7712
Petula	338	288	6681	114	471	8834
Diamina	98	131	6475	112	413	7656
Brillante	126	177	8474	136	384	9832
Furone	80	80	5182	92	572	6492
Linda	87	118	5693	87	374	6881
mean (\bar{x})	140	128	6032	101	424	7575
LSD _{5%} ^a	28	21	845	16	56	1061
<i>P</i> ^b	≤ 0.001	≤ 0.05	≤ 0.05	≤ 0.05	≤ 0.01	≤ 0.01

^a LSD_{5%} = least significant difference at 5% probability level. ^b *P* = probability level, *n* = 3.

Table 4. Ascorbic Acid and Tocopherol Contents of Different Industrial Tomato Varieties Cultivated in Gödöllo (1998)

varieties	ascorbic acid (mg/100 g fresh weight)	tocopherol ($\mu\text{g}/100\text{ g}$ fresh weight)			
		α -tocopherol	α -tocopherol quinone	β -tocopherol	γ -tocopherol
Amico	20	667	51	66	255
Casper	17	538	115	58	235
Góbé	17	737	567	61	198
Ispana	18	411	241	59	352
Pollux	17	599	317	51	184
Soprano	16	587	203	71	180
Tenger	17	418	54	60	92
Uno	19	494	171	45	171
Zaphyre	17	619	262	65	113
Draco	21	1164	707	42	263
Jovanna	21	1066	583	54	451
K-541	22	815	447	33	230
Nivo	20	965	601	29	225
Simeone	20	779	612	26	167
Sixtina	19	1002	463	35	428
mean (\bar{x})	19	731	360	50	236
LSD _{5%} ^a	3	84	48	11	42
<i>P</i> ^b	ns ^c	≤ 0.001	≤ 0.001	≤ 0.05	≤ 0.001

^a LSD_{5%} = least significant difference at 5% probability level. ^b *P* = probability level, *n* = 3. ^c ns = not significant.

such compound has been identified as lycoxanthin (Abushita et al., 1997). This variation may be due to some varietal or agricultural factors, since in the earlier work another variety cultivated under completely different conditions was used.

Data in Table 3 represent content of the major carotenoids in 12 salad tomato cultivars. Although total carotenoid content was maximal in Monika, Petula, and Brillante (86.6–98.3 $\mu\text{g}/\text{g}$), the highest values for β -carotene (the major provitamin A in tomato) were found in Monika, Furone, and Regulus. The latter two cultivars contained medium or low level of total carotenoids. Since no inverse correlation could be observed between lycopene and β -carotene, it is difficult to ascribe the reason for high β -carotene in some cultivars to only increased rate of lycopene cyclization to β -carotene. The lowest values of total carotenoids and lycopene were estimated in Marlyn, Fanny, and Diamina, while other varieties contained medium level of carotenoids. The β -carotene and total carotenoid contents of tomato fruits from Monika and Delfine were well above those obtained for the same varieties cultivated under different conditions (Abushita et al., 1997). This may point out to the fact that plant nutrition mode and environmental factors can affect, to a considerable extent, the overall

biosynthesis of carotenoids. It was remarkable that, in Petula cultivar, the fruits contained the highest level of lutein and lycopene epoxide, revealing that reactions of lycopene epoxidation and β -carotene hydroxylation are activated to a high extent, in this variety.

Evaluation of Processing Varieties. In this part of the work, 15 processing varieties were evaluated for their ascorbic acid, tocopherol, and carotenoid content (Tables 4 and 5).

Regarding the ascorbic acid content of tomato, there was no substantial difference between salad and processing varieties. The examined processing cultivars could be statistically divided into two groups, which showed significant difference at *p* < 0.05. In the first group, concentration of ascorbic acid ranged between 15.8 and 17.4 mg/g. Pollux, Soprano, Zaphyr, Casper, Ispana, Góbé, and Tenger are included in the first group. The second group, in which ascorbic acid content was between 18.6 and 21.7 mg/g, includes the other varieties examined in this work with Uno being the cultivar with the highest level of vitamin C.

Generally, fruits of processing varieties contained higher level of α -tocopherol than the salad tomatoes. The different cultivars exhibited marked variation with regard to α -tocopherol concentration. The highest con-

Table 5. Carotenoid Content ($\mu\text{g}/100\text{ g}$ fresh weight) of Different Industrial Tomato Varieties Cultivated in Gödöllo (1998)

varieties	carotenoids					total carotenoids
	lutein	lycopene epoxide	lycopene	cis-lycopene	β -carotene	
Amico	145	215	7726	133	447	9036
Casper	115	154	6614	96	245	7786
Góbé	143	116	5918	127	402	7150
Ispana	123	182	6222	100	317	7525
Pollux	348	148	5140	97	210	6799
Soprano	429	361	8646	107	321	11 026
Tenger	103	152	7656	114	228	8824
Uno	131	171	7086	91	323	8321
Zaphyre	303	173	6950	113	375	8907
Draco	76	208	6868	90	291	8191
Jovanna	138	282	11 606	82	339	13 205
K-541	118	256	9954	92	283	11 248
Nivo	116	231	8456	133	260	9722
Simeone	361	200	9879	103	296	11 882
Sixtina	332	249	10 510	111	318	12 521
mean (\bar{x})	199	307	7949	106	310	9476
LSD _{5%} ^a	29	35	1022	21	47	1442
P^b	≤ 0.001	≤ 0.05	≤ 0.05	ns ^c	≤ 0.05	≤ 0.05

^a LSD_{5%} = least significant difference at 5% probability level. ^b P = probability level $n = 3$. ^c ns = not significant.

centration ranged between 10 $\mu\text{g}/\text{g}$ in Sixtina and 11.6 $\mu\text{g}/\text{g}$ in Draco. The lowest values of α -tocopherol (4.1–4.2 $\mu\text{g}/\text{g}$) were recorded for Ispana and Tenger. The other cultivars are considered as tomatoes with medium level of vitamin E. The difference between the three groups in α -tocopherol content was significant ($p < 0.01$). The greatest variation between different varieties was noticed in the α -tocopherol quinone concentration. It ranged between 0.51 and 0.54 $\mu\text{g}/\text{g}$ in Amico and Tenger and 7.1 $\mu\text{g}/\text{g}$ in Draco. Concentration of γ -tocopherol, the most chemically reactive form of tocopherol fraction of tomato, was maximal (4.3–4.5 $\mu\text{g}/\text{g}$) in Jovanna and Sixtina, while a content of 0.9–3.5 $\mu\text{g}/\text{g}$ was determined in the other varieties.

The quantitative distribution of carotenoids in processing tomatoes is shown in Table 5. The highest concentration of carotenoids (total) was found in Jovanna and Sixtina followed by Simeone, K-541, and Soprano. These varieties, however, did not contain the highest level of β -carotene. Amico, Góbé, and Zaphyr were evaluated as varieties with high β -carotene level (3.7–4.5 $\mu\text{g}/\text{g}$). The lowest values with regard to the β -carotene and total carotenoid content were recorded for Pollux. Another difference between industrial tomatoes was in the formation of lutein via hydroxylation of β -carotene. Values of 4.3, 3.5, 3.6, and 3.0 $\mu\text{g}/\text{g}$ were estimated in Soprano, Pollux, Simeone, and Zaphyr, respectively. The other varieties have much lower quantities of lutein (0.8–1.5 $\mu\text{g}/\text{g}$).

From these data, it can be concluded that Jovanna merits high interest as an industrial variety containing high level of natural colorants and considerable amounts of vital antioxidants, so that the products from such a variety are of high technological and nutritional values.

Effect of Processing. In this work only paste processing was included. Under the conditions of the Gold Pheasant canning factory, the samples were taken at three stages of processing; raw tomato, crushed-sieved puree, and pasteurized paste. Table 6 shows the data obtained for ascorbic acid in tomato at different stages of processing. To avoid the effect of water evaporation and concentration of solids taking place during thermal processing on the quantification, the estimated values were dry weight-based (per g of dry

Table 6. Change in the Ascorbic Acid Content of Tomato as a Function of Processing

processing steps	ascorbic acid (mg/g dry matter)			
	\bar{x}^a	SD ^b	CV% ^c	n^d
raw material	3.17	0.06	1.9	5
hot-break extract	1.96	0.12	6.3	6
tomato paste ^e	1.45	0.21	14.3	6
Loss %		54.6		

^e 28–30% total soluble solids (Brix value). ^a \bar{x} = mean. ^b SD = standard deviation. ^c CV% = Percent coefficient of variation. ^d n = no. of samples examined.

matter). During hot-break extraction, tomato lost about 38% of the original ascorbic acid, while further processing to produce tomato paste by vacuum evaporation caused the product to lose more than 16% of its ascorbic acid content. This indicates that ascorbic acid can be greatly lost as a function of thermal processing. However, 45% of the initial content of ascorbic acid was retained by the final product (commercially sterilized paste). The retained quantity of ascorbic acid can play an important role in prevention of tomato paste against oxidative degradation during storage and/or subsequent preparation of meals (cooking).

The higher variation of coefficient found between replicate samples of hot-break extract and paste is probably due to fluctuation in the feeding speed of tomato extract between hot-break extraction and water evaporation. In case of high load, the extract feed is stopped for a short time that causes the temperature of the extract, particularly inside the extractor, to increase to higher degrees making such heat treatment more detrimental to ascorbic acid.

Data in Table 7 shows that α -tocopherol lost 20.3% of its content during thermal processing of tomato paste, while α -tocopherol quinone and γ -tocopherol lost 46.5 and 32.7% of their original content, respectively. On the basis of the quantitative changes (μg lost) contribution of the different form in the antioxidation processes is in the order of α -tocopherol > α -tocopherol quinone > γ -tocopherol. These results emphasize that the quinone derivative of tocopherol can play an important role in oxidation prevention in food system.

Table 7. Change in the Tocopherol Content of Tomato as a Function of Processing

processing steps	tocopherols ($\mu\text{g/g}$ dry matter)								
	α -tocopherol			α -tocopherol quinone			γ -tocopherol		
	\bar{x}^a	SD ^b	CV% ^c	\bar{x}	SD	CV%	\bar{x}	SD	CV%
raw material	202	10.0	4.9	113	10.9	9.6	42	4.7	11.0
hot-break extract	228	18.8	8.3	113	9.5	8.4	43	4.7	10.9
tomato paste ^d	161	9.0	5.6	61	8.2	13.5	28	3.1	10.9
loss %		20.3			46.5			32.7	

^a \bar{x} = mean. ^b SD = standard deviation. ^c CV% = percent coefficient of variation. ^d 28–30% total soluble solids (Brix value).

Table 8. Change in the Carotenoid Content ($\mu\text{g/g}$ dry matter) of Tomato as a Function of Processing^a

carotenoids	products								
	raw material			hot-break extract			tomato paste ^b		
	\bar{x}^c	SD ^d	CV% ^e	\bar{x}	SD	CV%	\bar{x}	SD	CV%
lutein	19.8	1.9	9.8	18.5	2.9	15.5	19.2	2.0	10.2
lycopene epoxide	41.4	3.6	8.6	37.0	4.9	13.2	47.3	3.0	6.4
<i>all-trans</i> -lycopene	1189.4	97.2	8.2	1219.5	90.6	7.4	1628.2	81.0	5.0
<i>cis</i> -lycopene	20.6	3.0	14.5	25.5	5.3	20.9	25.2	2.9	11.5
<i>all-trans-β</i> -carotene	37.2	4.0	10.8	38.5	4.5	11.6	26.3	3.5	13.5
<i>cis-β</i> -carotene	<1			3.9	0.6	16.5	9.7	1.5	15.4
total carotenoids	1430	96.2	6.7	1318.5	91.1	6.9	1848.8	90.2	4.9

^a Number of samples examined (*n*) was 5, 6, and 6 for raw material, hot-break extract, and tomato paste, respectively. ^b 28–30% total soluble solids (Brix value). ^c \bar{x} = mean. ^d SD = standard deviation. ^e CV% = percent coefficient of variation.

Table 8 contains carotenoids concentrations for whole tomato, hot-break extract and tomato paste. Compared to other carotenoids lycopene was detected in the highest concentration in all of the batches examined. High concentrations of lycopene have been found in different fresh and processed tomato products (Klauri and Bauernfeind, 1981; Daood et al., 1987; Tan, 1988; Tavares and Rodriguez-Amaya, 1994; Tonucci et al., 1995). High activity of lycopene, as singlet oxygen quencher (Di Mascio et al., 1989), makes its presence in the diet of considerable interest. The raw material contained 7.14 mg/100 g fresh weight *all-trans*-lycopene. The other major carotenoids were lycopene epoxide and *all-trans-β*-carotene. The estimated concentrations of lycopene in raw materials was lower than that reported by Tonucci et al. (1995), but well above the 3.11 mg/100 g found by Tavares and Rodriguez-Amaya (1994) in their work on processing tomatoes. Regarding β -carotene, it was at level of 0.22 mg/100 g in raw material. This level is close to the 0.23 mg/100 g estimated by Tonucci et al. (1995), but both are substantially lower than the 0.51 reported by Tavares and Rodriguez-Amaya (1994).

Although tomato processing was typically carried out at high temperature over an extended period with slight vacuum, the integrity of the carotenoids, except β -carotene; was unchanged, and the qualitative distribution of carotenoids in tomato paste remained identical to that of raw tomatoes. In general, these results agree with those reported by Khachik et al. (1992) and Tonucci et al. (1995), but disagree with the results of Tavares and Rodriguez-Amaya (1994) who investigated the changes in carotenoids of Brazilian tomatoes. This variation may be due to some varietal, agricultural, technological, and environmental factors.

It was remarkable that *all-trans*-lycopene and the total carotenoids content increased in the dry matter of tomato paste, most likely due to removal of seeds and peels and loss of soluble volatile compounds during water evaporation steps. This effect was not observable in case of ascorbic acid and tocopherols because the

magnitude of their degradation is much higher than the increase resulted from removal of seeds and peels.

As for *trans-β*-carotene, its content in the paste decreased substantially, meanwhile, concentration of the *cis* form increased, indicating that *trans* to *cis* isomerization of β -carotene is taking place during thermal processing of tomato particularly during dehydration step to produce the paste.

It is worthy to mention that under the given conditions lycopene underwent slight isomerization to form *cis* isomer. Similar result was found by Tonucci et al. (1995). This leads to the suspicion that isomerization of lycopene reflects longer and more drastic processing, particularly in the concentration step. The results also indicate that β -carotene is more sensitive than lycopene, and therefore, its *in vitro* antioxidative activity is higher in aqueous media.

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