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Nutritional Value of Cherry Tomatoes (*Lycopersicon esculentum* Cv. Naomi F1) Harvested at Different Ripening Stages

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The average content of some classes of antioxidants is generally higher in cherry tomatoes than in normal-sized berries. The aim of this work was to assess the nutritional value of cherry tomato (cv. Naomi F1) by investigating the compositional pattern of berries harvested at different ripening stages and evaluating, in particular, all of the main antioxidants (carotenoids, ascorbic acid, phenolic compounds, and α-tocopherol) and the antioxidant activity of the water-soluble and water-insoluble fractions. Results confirmed the relatively high level of carotenoids in cherry tomato but showed that not all biologically active compounds necessarily increase in tomatoes picked at later stages of ripeness. Cherry tomatoes harvested at full ripeness exhibited the highest level of carotenoids and antioxidant activity in the water-insoluble fraction. On the other hand, no significant differences in ascorbic acid content were observed at different ripening stages, whereas the main phenolics content and the antioxidant activity of water-soluble fraction showed slight, but significant, decreases at later stages of ripeness.

KEYWORDS: Cherry tomato (*Lycopersicon esculentum* var. *cerasiforme*); ripening; carotenoids; ascorbic acid; phenolic compounds; α-tocopherol; antioxidant activity

INTRODUCTION

Several epidemiological studies indicated a beneficial effect of tomato consumption in the prevention of some major chronic diseases, such as some types of cancer and cardiovascular diseases (*1*, *2*). It has been postulated that the protective role is due to tomato antioxidants that could contribute to the inhibition of abnormal oxidative processes (*3*). Tomato contains different classes of antioxidants such as carotenoids, ascorbic acid, phenolic compounds, and α-tocopherol, and, due to its high consumption rates, it can provide significantly to the total intake of these components (*4*−*7*).

The antioxidant content of fresh tomatoes can be affected by many pre- and postharvest factors. The influence of cultivar (*8*), cultural practices (*9*), ripening stage at harvest (*10*), and storage conditions (*11*) on antioxidants accumulation has been studied during the past decade.

Moreover, it is well-known that the positive effect on health associated with the consumption of fresh fruits and vegetables is exerted by the pool of antioxidants, with noticeable synergistic effects. Therefore, to assess the nutritional quality of fresh tomatoes, it is important to study all of the main compounds having antioxidant activity.

Cherry tomato is one of the most important types for fresh consumption. In Italy cherry tomato is mainly cultivated in the Sicily region and grown in cold greenhouses. The peculiar conditions of climate (relatively high radiation level and temperature) and irrigation water (particularly high salinity) yield tomatoes that are tasty and now constitute >25% of the market of tomatoes for fresh consumption, so their commercial importance is continuously increasing (*12*, *13*). Cherry tomato varieties are generally characterized by higher dry matter and soluble solids levels than normal-sized fresh market cultivars; these differences are due to the higher content of sugars (fructose and glucose) and organic acids (citric and malic), which, in turn, are major factors in determining the greater sweetness, sourness, and overall flavor intensity of most cherry varieties (*14*−*18*). Many studies have dealt with organoleptic quality and related

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compositional parameters, but a only few works have investigated the nutritional properties of these tomato varieties. Leonardi et al. found that carotenoid content as well as lipophilic antioxidant activity of tomato was more affected by ripening stage than by cultivar, which nevertheless determined significant effects; cherry tomatoes showed a relatively high level of carotenoids and higher lipophilic and hydrophilic antioxidative abilities than other typologies of tomatoes commonly used for fresh consumption in Italy (*19*). On the other hand, in a study on tomato cultivars grown in the United Kingdom, cherry tomatoes showed intermediate lycopene and β-carotene levels when compared with other fresh marketed varieties (*4*). Crozier et al. found higher flavonol concentrations in Spanish and British cherry tomatoes compared to normal-sized fruits (*6*); in a recent paper this observation was only partially confirmed by Stewart et al., who, however, suggested that the greater skin/volume ratio of cherry tomatoes could enhance their flavonol content, because these compounds occur within the skin of the fruit (*20*). A detailed assessment of cherry tomato compositional profile, with regard to antioxidant compounds, is still lacking, and it is not clear if possible differences between cherry and normal-sized varieties are due to a dilution effect or a metabolism effect. This prompted us to perform an in-depth study, focusing attention on compounds playing a significant role in determining the nutritional value of cherry tomato. The aim of this study was to investigate changes in the nutritional value of cherry tomatoes (cv. Naomi F1) harvested at different ripening stages after turning by evaluating all major compounds having antioxidant properties, as well as the antioxidant activity of the water-soluble and water-insoluble fractions.

MATERIALS AND METHODS

Fruit Sampling. Tomato seeds (*Lycopersicon esculentum* cv. Naomi F1, Hazera Seeds) were sown (February 1999) four times (each separated by one week), and fruits were harvested at the same time, from the same truss, at various ripening stages. This procedure controls the effects of both fruit position on the plant and climatic conditions prior to harvest (*21*, *22*). About 45 days after sowing, when the seedlings had reached the stage of four true leaves, tomato plants were transplanted to a cold greenhouse. Growth to maturity and fruit development took place in an unheated greenhouse located in the coastal area of Sicily; plant nutrition and chemical pest and disease control followed commercial practices. In the month preceding harvesting the mean temperature was 26.5 °C and the mean solar radiation at crop level was 19.6 MJ m^{-2} d^{-1}.

All fruits were harvested when the first fruits of the second truss reached the full ripening stage (beginning of July). At harvest the four proximal fruits of each second truss were pooled; fruits were then sorted to five groups corresponding to the following ripening stages: green−yellow (G−Y, ∼30% yellow skin); green−orange (G−O, ∼50% orange skin); orange−red (O−R, >90% orange or red skin); light red (L−R, fully orange or red skin); red (R, fully red skin). These categories encompass the full range of tomato ripening stages. Tomatoes from each maturity stage had about the same size and weight. After harvesting, tomatoes were kept for 2 days at ambient temperature to simulate the normal time span from harvest to consumption. Then carpometric characteristics and the contents of sugars, organic acids, and antioxidant compounds were determined separately on three groups of fruits at each ripening stage, consisting of 30 fruits chosen at random from each sample.

Carpometric Characteristics. The following measurements were made on each sample: dry matter (%), obtained by drying the fruits in a thermoventilated oven at 70 °C until constant weight was reached; soluble solids by a refractometer (Atago), results reported as Brix degrees at 20 °C; chromatic coordinates (L^*, a^*, and b^*) on the equatorial part of the fruit as described by McGuire (*23*), using a tristimulus Minolta Chroma meter (model CR-200, Minolta Corp.). Color was described by the ratio a^*/b^*, lightness (L^*), and chroma ($C^* = \sqrt{a^{*2}+b^{*2}}$).

Biochemical Analyses. Whole tomatoes were homogenized in a Waring blender for 1 min. The homogenate was then frozen at −20 °C and stored until analyzed.

Organic Acids. Organic acids were extracted with 20 mM KH_2PO_4 (pH 2.8) and quantified by HPLC, with UV detection, according to the method of Lee (*24*).

Sugars. One gram of homogenate was combined with 9 mL of CH_3CN/H_2O (4:1) and rehomogenized in an Ultra Turrax T25 blender at 8000*g* for 3 min. The samples were centrifuged at 3000 rpm for 10 min, and the supernatants were filtered through 0.20-μm syringe cellulose filters before HPLC analysis. Samples were injected onto a 150 × 4.6 mm i.d. Hypersil 5 APS 2 HPLC column (Chrompack), using CH_3CN/H_2O (4:1) as mobile phase, at a flow of 0.5 mL/min. Sugars were quantified by refractometric index detection (Knauer).

Antioxidants Content. α-Tocopherol from raw tomatoes was measured according to the method of Baldini et al. (*25*). Briefly, the extraction consists of base hydrolysis of the sample followed by $C_2H_4Cl_2$ addition. The organic phase was dried, and the residue was resuspended in 1 mL of CH_3OH (1 mL) and run on the HPLC column (Phenomenex Prodigy C18, 5 μm; 4.6 × 250 mm). The mobile phase was isocratic CH_3OH/H_2O (98:2) with a flow rate of 1 mL/min. α-Tocopherol was identified by comparison with a pure standard (Sigma) and quantified by measuring absorbance at 290 nm.

Phenolics were both hydrolyzed and not, to obtain free and conjugated forms, and extracted as described by Hertog et al. (*26*). Quantitative analysis was performed using an ESA HPLC system with an eight-channel coulometric electrode array detector (ESA). HPLC separation was carried out at a flow rate of 1 mL/min, at a temperature of 30 °C, using a Supelcosil LC-18 column (250 × 4.6 mm) with a Perisorb Supelguard LC-18 (Supelco); the mobile phase and the elution program were as described in ref *20*. The calibration curve for quantification was obtained using authentic standards (Sigma).

Ascorbic acid, reduced and total (ascorbic plus dehydroascorbic acid), was extracted and quantified by HPLC according to the method of Margolis et al. (*27*). HPLC separation was carried out using the ESA system previously mentioned, on a Capcell Pak NH_2 column (250 × 4.4 mm) (Shiseido) at a flow rate of 1 mL/min at 40 °C; the mobile phase was as described in ref *27*. Pure standards (Sigma) were used for identification of peaks and for quantification.

Carotenoid content was determined by HPLC with diode array detection as described by Leonardi et al. (*19*).

Antioxidant Activity. Ten milliliters of deionized water was added to 5 g of tomato homogenate, and the suspension was centrifuged at 15000 rpm for 15 min (4 °C). The supernatant (water-soluble fraction) was recovered, filtered, and, after suitable dilution (1:5) with phosphate buffer (10 mM, pH 7.0), used for the crocin bleaching inhibition test (*28*). Antioxidant activity was expressed as equivalent millimolar Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) for 100 g of tissue fresh weight. The pulp resulting from centrifugation of the homogenate was extracted with 10 mL of CH_2Cl_2, centrifuged at 15000 rpm for 5 min (4 °C), and filtered, and the supernatant was recovered; this extraction step was repeated three times, and supernatant fractions were pooled. The extract (water-insoluble fraction) was used for the 2,2′-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) test (*29*), and antioxidant activity was expressed as equivalent millimolar Trolox for 100 g of fresh tissue.

Statistical Evaluation of Data. A completely randomized experimental design with three replications was adopted. We performed analysis of variance (ANOVA) in order to test the significance of the observed differences. When the effects of ripening stage were significant ($P \leq 0.05$), we performed the Duncan test; in the tables significant differences are indicated. Regression analysis was performed to evaluate the relationship between all of the considered parameters and ripening stage, which is expressed by the ratio a^*/b^*.

Table 1. Chromaticity Values, Dry Matter, Soluble Solids, Sugars, and Organic Acids at Different Ripening Stages

parameter	ripening stage[a]					relationship vs *a**/*b**	
	G–Y	G–O	O–R	L–R	R	type	*P*[b]
chromaticity values							
*a**/*b**	0.04 a	0.60 b	0.96 c	1.41 d	1.76 e		
*L**	49.2 e	46.3 d	44.8 c	43.2 b	41.6 a	linear	≤0.001
*C**	13.4 a	13.1 a	13.7 a	14.1 a	15.6 b	quadratic	≤0.05
dry matter (%)	6.74 a	6.94 ab	7.02 b	7.29 c	7.38 c	linear	≤0.01
soluble solids (°Brix)	5.03 a	5.27 a	5.80 b	6.07 c	6.07 c	linear	≤0.01
sugars (g/100 g)							
fructose	1.53 a	1.63 b	1.65 b	1.77 c	1.86 d	quadratic	≤0.05
glucose	1.34 a	1.44 ab	1.50 bc	1.62 c	1.79 d	cubic	≤0.001
organic acids (g/100 g)							
malic acid	0.12 b	0.10 ab	0.09 ab	0.09 a	0.12 b	–[c]	–
citric acid	0.79 c	0.80 c	0.75 b	0.69 a	0.67 a	cubic	≤0.05
oxalic acid	0.03 a	0.03 a	0.04 a	0.04 a	0.05 b	linear	≤0.05

[a] In this and the following tables different letters, within each parameter, indicate significant differences according to the Duncan test. No letter was reported when ANOVA did not evidence significant differences ($P \leq 0.05$) between ripening stages. [b] In this and the following tables, the significance of the relationship was reported as *P* values. [c] If not indicated, there was no relationship significant at least at $P \leq 0.05$.

RESULTS AND DISCUSSION

The aim of this work was to characterize the nutritional value of fresh cherry tomatoes harvested at different ripening stages by examining their compositional pattern, with particular attention to the different classes of tomato antioxidants. In our study we have considered the *a**/*b** ratio determined on the skin as a reference parameter for ripening stage, so that we could compare our results with literature data. Tomato berries picked at five different ripening stages showed *a**/*b** values significantly increasing from 0.04 to 1.76, whereas both color lightness (*L**) and chroma (*C**) values varied only slightly (**Table 1**). Lightness declined during ripening, showing an inverse linear relationship with respect to *a**/*b** values. Firmness, another physical parameter related to ripening stage, declined significantly but was poorly correlated with *a**/*b** values (data not reported).

Dry matter and soluble solids increased during ripening from 6.74 to 7.38% and from 5.03 to 6.07 °Brix, respectively, showing a linear relationship with ripening stage ($R^2 = 0.95$ and 0.92) (**Table 1**). Proteins (1.0−1.3 g/100 g), fats (0.1−0.2 g/100 g), fiber (1.4−1.7 g/100 g), and ash (0.6−0.7 g/100 g) did not vary significantly during ripening.

Monosaccharide content showed a 27% increase from 2.87 to 3.65 g/100 g (**Table 1**); sugar accumulation represented the main contribution to the dry matter increase during ripening, as confirmed by the increase of sugar content expressed on a dry weight (dw) basis (from 42.6 to 49.5 g/100 g, dw). Glucose and fructose both increased noticeably, whereas sucrose was not present in detectable amounts. Dry matter, soluble solids, and, in particular, glucose and fructose (1.79 and 1.86 g/100 g) in the tomato cultivar examined at full ripeness were high when compared with ranges reported for tomatoes in international food composition tables (0.90−1.62 and 1.25−1.70 g/100 g for glucose and fructose, respectively) (*30*); this confirmed the high value of these parameters in cherry tomatoes compared to other tomato types, already observed in different periods, places of production, and cultivars (*14*−*19*).

The pattern of sugar accumulation differed from that observed in other cultivars. In fact, only slight monosaccharides accumulation and no increase of glucose were previously observed during the color change from G−Y to R (*31*).

With regard to organic acids, citric acid content declined slightly from the G−Y stage (0.79 g/100 g) to the R stage (0.67 g/100 g), whereas malic and oxalic acid did not change markedly during ripening. It was well established that their content is considerably affected by cultivar (*31*), and in this case also our data confirmed the high acids content in cherry tomatoes compared with normal-sized varieties, previously evidenced by other authors (*14*−*16*, *18*). The trend of citric acid content during ripening was similar to that reported in the literature, whereas malic acid did not exhibit the declining pattern previously observed (*31*).

All of these factors play a key role in determining the peculiar sensory profile of cherry tomatoes (*18*). On the other hand, sugars and organic acids, which form a substantial fraction of tomato dry matter, are relevant more to taste attributes than to the nutritional value of tomato, with tomato antioxidants playing a major role in the latter aspect.

Lipophilic Antioxidants. At the breaker stage the content of carotenoids was very low (1084 μg/100 g), increasing >10-fold during ripening and reaching 12705 μg/100 g at full ripeness (**Table 2**). Lycopene, which represented >80% of total carotenoids at the R stage, increased during ripening ∼20-fold. Its content at the R stage expressed on a fresh weight basis, 10440 μg/100 g, was relatively high when compared with published data on different varieties for fresh consumption. In a general review on the carotenoid content of vegetables, Mangels et al. reported the range from 880 to 4200 μg/100 g (*32*). Hart and Scott determined the lycopene content in 11 varieties cultivated for the fresh market in the United Kingdom (tomatoes purchased from various retail outlets in England between April and October) and found concentrations from 1710 to 5650 μg/100 g and, in particular, 3780 μg/100 g in a cherry variety (*4*). According to Abushita et al., the lycopene content in 12 salad tomato varieties cultivated in Hungary (grown outside) ranged from 5180 to 8470 μg/100 g (*8*). In eight medium- and large-sized varieties grown in Florida and harvested in June, Thompson et al. found lycopene concentrations from 2620 to 5790 μg/100 g (*33*). In a more recent paper on nine commercial varieties grown in Spain the lycopene content (simply obtained by measuring the absorbance of a tomato extract) ranged from 3150 to 6500 μg/100 g (*34*). By comparing tomato varieties (Naomi F1, a cherry type; Felicia, a cluster type; Italdor, with elongated fruit; ES200, a salad type) grown in Sicily in a greenhouse and harvested during May and June, Leonardi et al. found that cherry tomatoes had relatively high levels of carotenoids and lycopene but that on a dry weight basis the cluster-type variety showed the highest contents (147 mg/100

Table 2. Carotenoid Content at Different Ripening Stages (Micrograms per 100 g of Fresh Weight)

	ripening stage					relationship vs *a*/b**	
compound	G–Y	G–O	O–R	L–R	R	type	*P*
lycopene	453 a	2232 b	4510 c	6920 d	10440 e	quadratic	≤0.01
β-carotene	339 a	713 b	898 d	841 c	1073 e	linear	≤0.05
phytoene	49 a	187 b	404 c	462 c	575 d	linear	≤0.01
phytofluene	19 a	116 b	261 c	268 cd	297 d	linear	≤0.05
ζ-carotene	188 c	114 b	92 b	0 a	0 a	linear	≤0.01
γ-carotene	0 a	0 a	22 b	34 c	46 d	linear	≤0.05
5,6-dihydroxy-5,6-dihydrolycopene	0 a	11 b	17 c	23 d	39 e	linear	≤0.01
lycopene 1,2-epoxide	0 a	20 b	73 c	85 d	175 e	linear	≤0.05
lycopene/β-carotene	1.3	3.1	5.0	8.2	9.7	quadratic	≤0.01
total carotenoids	1084 a	3393 b	6278 c	8655 d	12705 e	quadratic	≤0.01

Table 3. Phenolic Compounds at Different Ripening Stages (Milligrams per 100 g of Fresh Weight)

	ripening stage					relationship vs *a*/b**	
compound	G–Y	G–O	O–R	L–R	R	type	*P*
chlorogenic acid	4.19 c	3.26 bc	2.73 bc	2.25 ba	0.91 a	quadratic	≤0.05
caffeic acid	0.74 c	0.51 b	0.35 ab	0.28 ab	0.13 a	linear	≤0.01
p-coumaric acid	0.37	0.31	0.26	0.36	0.30	–	–
ferulic acid	0.20	0.16	0.14	0.17	0.12	–	–
rutin	0.85 b	1.53 c	0.58 ab	0.82 b	0.40 a	–	–
quercetin	0.46 c	0.45 c	0.37 bc	0.21 ab	0.12 a	quadratic	≤0.05
naringenin	3.66	4.55	4.27	3.35	3.19	–	–

g, dw, lycopene in the cluster-type tomatoes vs 96−114 mg/100 g, dw, in the cherry tomatoes) (*19*). Generally, published data on carotenoid content have been reported on a fresh weight basis, and the dry matter content has not been given; consequently, it is not possible to establish comparison on a dry weight basis in order to know if the higher concentrations in cherry tomatoes are due to dilution or metabolism effects. The only study in which data on dry matter were reported was that on Spanish tomatoes (*34*): lycopene content expressed on a dry weight basis ranged from 35 to 123 mg/100 g, dw, and was significantly lower than in cherry tomatoes examined in our study (141 mg/100 g, dw). The rate of lycopene accumulation relative to the change in *a*/b** was similar to that observed in two different genotypes of cv. Moneymaker (Normal Red and Crimson) (*11*). β-Carotene showed a less marked increase (∼3-fold) compared to lycopene and was linearly correlated with the ripening index; it accounted for 31% of total carotenoids at the breaker stage and only 8.4% at full ripeness (the ratio lycopene/β-carotene increased from 1.3 to 9.7). Thus, in cv. Naomi tomatoes the great increase of carotenoids was substantially due to the accumulation of lycopene and β-carotene, which at full ripeness accounted for ∼90% of the total carotenoid fraction. β-Carotene content at full ripeness (1073 μg/100 g) also fell within the high range when compared with published data of Mangels et al. (*32*), Hart et al. (*4*), and Abushita et al. (*8*) (115−660, 350−1700, and 285−615 μg/100 g, respectively). Although the regression analysis showed a linear relationship between β-carotene content and *a*/b** ratio, the accumulation rate of this compound was higher at the first stages than later. Conflicting data concerning the accumulation rate of β-carotene during ripening have been reported: in some cases its concentration increased throughout ripening (*5*, *11*), whereas in other studies, it was reported to reach its maximum level prior to full ripeness (*31*, *36*). These differences have been attributed to different growing conditions and cultivars (*19*, *35*, *37*). Even phytoene and phytofluene accumulated considerably during ripening, and their contents were linearly correlated with *a*/b** values; their sum formed 6.8% of the total carotenoid fraction at the R stage. Among the minor carotenoids, γ-carotene was detectable only at later ripening stages, whereas ζ-carotene, which represented 17% of total carotenoids at the breaker stage, markedly decreased during ripening. It is worth noticing that at all stages β-carotene content was higher than that of the two colorless carotenoids phytoene and phytofluene, at variance with what has been reported for other cultivars (*31*, *36*, *38*). Furthermore β-carotene content was markedly higher than γ-carotene as already observed in other varieties (*31*, *36*). From the nutritional point of view, this means that the vitamin A activity of Naomi tomatoes, as for fresh tomatoes from other cultivars, derives essentially from β-carotene, and the contribution of γ-carotene is quite negligible, contrary to what has been reported for processed tomato products (*7*).

Table 4. Reduced and Total Ascorbic Acid and α-Tocopherol at Different Ripening Stages (Milligrams per 100 g of Fresh Weight)

	ripening stage					relationship vs *a*/b**	
compound	G–Y	G–O	O–R	L–R	R	type	*P*
ascorbic acid							
reduced	2.9	3.7	5.8	4.3	5.5	–	–
total	13.5	11.5	11.1	11.8	11.0	–	–
α-tocopherol	0.57 b	0.57 b	0.48 a	0.59 c	0.75 d	–	–

Finally, two compounds derived from lycopene, 5,6-dihydroxy-5,6-dihydrolycopene and lycopene 1,2-epoxide, accumulated similarly to their precursor but at much lower levels.

With regard to the other important tomato lipophilic antioxidant, α-tocopherol, we observed no clear relationship to the ripening index (**Table 4**) but simply higher values at full ripeness than at the first stages (∼30% increase), similar to that observed in other varieties (*5*).

The antioxidant activity of the water-insoluble fraction increased 2.5-fold during ripening (**Table 5**); the relationship between antioxidant activity and the ripening index was similar to that observed for lycopene. Carotenoids and α-tocopherol are the main lipophilic antioxidants of tomato, but the former

Table 5. Antioxidant Activity at Different Ripening Stages (Equivalent Millimolar Trolox per 100 g of Fresh Weight)

	ripening stage					relationship vs *a*/b**	
fraction	G–Y	G–O	O–R	L–R	R	type	*P*
water-soluble	0.270 b	0.220 a	0.196 a	0.174 a	0.186 a	quadratic	≤0.05
water-insoluble	0.015 a	0.023 b	0.024 b	0.033 c	0.036 c	quadratic	≤0.05

Table 6. Correlation Coefficients and Related Significance between Antioxidants Content and Antioxidant Activity

compound	corr coeff	*P*
Water-Soluble Fraction		
chlorogenic acid	0.831	ns[a]
caffeic acid	0.933	≤0.05
p-coumaric acid	0.358	ns
rutin	0.339	ns
quercetin	0.769	ns
naringenin	0.259	ns
ascorbic acid		
reduced	–0.655	ns
total	0.750	ns
Water-Insoluble Fraction		
total carotenoids	0.961	≤0.01
lycopene	0.962	≤0.01
β-carotene	0.885	≤0.05
phytoene	0.939	≤0.05
phytofluene	0.888	≤0.05
5,6-dihydroxy-5,6-dihydrolycopene	0.956	≤0.05
lycopene 1,2-epoxide	0.901	≤0.05
α-tocopherol	0.662	ns

[a] ns, no significant correlation.

are clearly predominant (12.7 vs 0.75 mg/100 g at full ripeness); so in our samples the antioxidant activity of the water-insoluble fraction essentially resulted from carotenoids, particularly lycopene, as confirmed by the relatively high value of the correlation coefficient between their content and the antioxidant activity of the fraction (**Table 6**).

Hydrophilic Antioxidants. The changes of phenolic compound content of tomato, as for other fruits and vegetables, may be influenced by external factors, such as light and average temperature, although genetic control is the primary factor (*39*). Among simple phenols, the main compound was chlorogenic acid (5′-caffeoylquinic acid), a hydroxycinnamic acid conjugate; its concentration during ripening gradually declined (**Table 3**) and at full ripeness (0.91 mg/100 g) was lower than in Spanish tomato varieties (1.43−3.28 mg/100 g) (*34*). Chlorogenic acid has a potential protective effect on human health, being characterized by a medium antioxidant activity (*40*), and due to its relatively high concentration, it could contribute significantly to the antioxidant activity of the tomato water-soluble fraction. Free caffeic acid, an immediate precursor of chlorogenic acid, was present at lower concentrations but also noticeably declined during ripening.

Naringenin is the main flavonoid in Naomi tomatoes. We quantified naringenin as the aglycon, both before and after hydrolysis; its content before hydrolysis (in the free state) was negligible, indicating that it was present as chalconaringenin or in a conjugated form, so we report its content after hydrolysis (**Table 3**). Naringenin concentration has previously been reported to increase rapidly in the early stages of maturation and to accumulate within the cuticular membrane of the fruit (*39*, *41*). In our samples naringenin increased from the G−Y to the G−O stage, whereas a slight decrease was observed in the following stages; at full ripeness the naringenin level was higher than in Spanish varieties (3.19 vs 0.45−1.25 mg/100 g) (*34*). With regard to flavonols, during the later stages, we observed a declining trend of both rutin (quercetin 3-*O*-rhamnosylglucoside), the main flavonol derivative in tomato, and free quercetin, which was found at low levels. Total quercetin content (expressed as aglycon, 0.37−1.41 mg/100 g), deriving from rutin and free quercetin, was medium to low when compared with that of other cherry tomato cultivars (*6*, *20*), whereas its trend during ripening resembled that previously reported (*10*, *39*). With regard to nutritional implications, the absorption rate of quercetin glycosides is strongly influenced by the nature of the sugar moiety; for example, pure rutin has been reported to be 70% less bioavailable than the quercetin glucosides contained in onions (*42*). Moreover, because of their long half-lives of elimination, repeated consumption of quercetin-containing foods causes accumulation of quercetin in the blood. Thus, it can contribute significantly to blood antioxidant defenses. The in vitro antioxidant activity of rutin has been reported to be relatively high but significantly lower compared to that of free quercetin, whereas naringenin, as aglycon, evidenced a medium value (*40*). The declining content of quercetin and naringenin after the first ripening stages could be associated with their involvement in fruit defense mechanisms against reactive oxygen species, which are produced in high amounts during the climacteric peak as a consequence of increasing rate of respiration (*43*).

In Naomi tomatoes the pattern of variation of phenolic compounds was similar to that observed in other cultivars (*10*, *31*, *39*). Published data on phenolics content were also generally expressed on a fresh weight basis only, so that it is not possible to establish comparison on a dry matter basis. In particular, tomatoes have been considered as food plants with a medium content of flavonoids, with an average value of 5 mg/100 g (*44*); in Naomi tomatoes the highest flavonoid content was observed at the green−orange stage, 5.96 mg/100 g, expressed as aglycon, whereas at full ripeness it decreased to 3.56 mg/100 g.

Ascorbic acid content, both the reduced form and total ascorbic acid (ascorbic plus dehydroascorbic acid, both of which exhibit vitamin activity and contribute to vitamin C content), did not change significantly during ripening (**Table 4**). Moreover, the reduced form accounted for between 22 and 54% of the total vitamin C, so the simple ascorbic acid quantification would lead to a substantial underestimation of vitamin C content. Together with phenolic compounds, ascorbic acid represents the main water-soluble antioxidants of tomatoes and contributes to the antioxidant activity of the water-soluble fraction. In our samples the antioxidant activity of the water-soluble fraction exhibited a slight decline during ripening, and at the G−Y stage it was significantly higher than at later stages (**Table 5**). This trend could be only partly ascribed to changes in phenolics and ascorbic acid content, and it was not unequivocally related to any single component (**Table 6**). It is plausible that the total antioxidant activity depended also upon synergistic effects among all water-soluble antioxidants and their interactions with other constituents of the fraction. Finally, in our study we did not consider glutathione, another water-soluble antioxidant; its level in tomato tissue has been recently determined by Jimenez et al. (*45*). According to these authors, glutathione content increased during ripening and significantly contributed to the antioxidative system of tomato fruits.

In conclusion, fruit harvested at full ripeness exhibited the highest level of lipophilic antioxidants, primarily lycopene, but also other carotenoids and α-tocopherol. This was correlated

with the highest antioxidant activity of the water-insoluble fraction. In particular, in Naomi tomatoes, in contrast to other varieties, β-carotene accumulation extended to later stages of ripening, so that one serving of tomatoes (100 g) provided ~8% of the recommended daily intake of vitamin A (*46*) at the green−yellow stage and ~25% at full ripeness. The comparison with other tomato varieties confirmed the relatively high level of lycopene content (on a fresh weight basis) in cherry varieties when compared to other types of tomatoes; nevertheless, on the basis of available data it is not possible to establish if this is due to a dilution effect. α-Tocopherol content showed a less marked increase, providing ~5% of the vitamin E recommended daily intake (*46*) in the first stages of ripening and 7.5% at full ripeness. On the other hand, fully ripened fruit were not richer in water-soluble antioxidants than mature green fruit, but rather the antioxidant activity of the water-soluble fraction decreased during ripening. Vitamin C content did not change significantly throughout ripening, one serving of tomatoes contributing 24−28% of the recommended daily intake (*46*), whereas the main phenolic compounds, nonvitamin antioxidants, decreased from the green−yellow to the red stage. Nevertheless, even if their concentration at full ripeness was lower than that in other fruits and vegetables, the relatively high average daily consumption of tomatoes can result in a significant phenolics intake. Finally, the level of flavonols we found in Naomi tomatoes was not particularly high when compared to that in other cherry and normal-sized varieties.

There is a common belief that during ripening an improvement of quality attributes related to consumer acceptance and nutritional value occurs in many fruits and vegetables. Data presented here demonstrate that not all biologically active compounds necessarily increase during the ripening process, the accumulation of distinct compounds depending on different biosynthetic pathways and mechanisms of metabolic control. Thus, from a nutritional point of view, it is important to investigate in detail the pattern of variation of all bioactive compounds.

ABBREVIATIONS USED

Ripening stages of tomatoes: G−Y, green−yellow; G−O, green−orange; O−R, orange−red; L−R, light red; R, red.

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