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Influence of Organic versus Conventional Agricultural Practice on the Antioxidant Microconstituent Content of Tomatoes and Derived Purees; Consequences on Antioxidant Plasma Status in Humans

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The present study aims first to compare the antioxidant microconstituent contents between organically and conventionally grown tomatoes and, second, to evaluate whether the consumption of purees made of these tomatoes can differently affect the plasma levels of antioxidant microconstituents in humans. When results were expressed as fresh matter, organic tomatoes had higher vitamin C, carotenoids, and polyphenol contents (except for chlorogenic acid) than conventional tomatoes. When results were expressed as dry matter, no significant difference was found for lycopene and naringenin. In tomato purees, no difference in carotenoid content was found between the two modes of culture, whereas the concentrations of vitamin C and polyphenols remained higher in purees made out of organic tomatoes. For the nutritional intervention, no significant difference (after 3 weeks of consumption of 96 g/day of tomato puree) was found between the two purees with regard to their ability to affect the plasma levels of the two major antioxidants, vitamin C and lycopene.

KEYWORDS: Organic cultural practices; tomatoes (*Lycopersicon esculentum*); lycopene; *β*-carotene; vitamin C; polyphenols

INTRODUCTION

Because consumers are aware of their health and more and more conscious of environmental conditions, there is an increasing demand for food obtained from alternative cultural practices limiting the use of mineral soluble fertilizers and synthetic pesticides. However, organic farming remains the subject of controversies for claiming healthier and safer foods due to a lack of sound technical data. In fact, concerning the nutritional qualities of plant-food products, data are scarce and lead to difficulties in interpretation, because organic or conventional products were not obtained under comparable conditions (similar areas, climate, etc). Some studies have been conducted on the effect of organic food on animals' health, and four observation studies concerning human health examined semen quality (*1*). None of these studies provided strong evidence of any beneficial effect of organic food consumption. To date, there has been no study on the comparison of organic and conventional food products in nutritional intervention studies in humans.

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Tomato is widely consumed in Europe (it is the second most commonly consumed vegetable for all countries except Italy, where it remains the most commonly consumed vegetable in fresh and processed forms). Tomatoes contain a large variety of microconstituents (*2*, *3*), which certainly contribute to the protective effect of this vegetable against degenerative diseases. Many of these microconstituents display antioxidative properties. The main lipophilic antioxidants are carotenoids: β-carotene, and, essentially, lycopene, the red pigment. β-Carotene is a provitamin A, and both carotenoids are potentially implied in the protection against degenerative diseases (*4*, *5*). Epidemiological or intervention studies have shown a correlation between the consumption of lycopene and the diminution of the risk of prostate cancer (*6*−*8*). Tomatoes and tomato-derived products are the main source of lycopene in food in Western countries. Among hydrosoluble microconstituents, vitamin C is the major antioxidant for tomatoes consumed in fresh form. Vitamin C is known to be affected along with numerous other vitamins by heating and other technological treatments (*9*, *10*). At a lower level, phenolic compounds could contribute to the beneficial effect of tomato-based products mainly because of their antioxidant properties, demonstrated in numerous studies (*11*). However, it is important to consider phenols in tomatoes, because tomatoes have been shown to be the major source of phenols from vegetables in the diet in the United States, giving 41.7 mg of phenols/day (*12*). Chlorogenic acid (5′-caffeoylquinic acid), a hydroxycinnamic acid conjugate, is the main phenol in tomatoes (*13*, *14*). Rutin (quercetin-3-rhamnosylglucoside) and naringenin are representative flavonoids of tomatoes, respectively conjugated and nonconjugated (*14*, *15*). It was recently demonstrated that naringenin from cooked tomato paste is bioavailable in men (*16*).

The aim of the present work was first to compare the content in antioxidant microconstituents of tomatoes grown under organic and conventional conditions and then to assess whether the consumption of purees processed from organic or conventional tomatoes differently affects the antioxidant status in humans.

MATERIALS AND METHODS

Tomato Cultivation. Three tomato varieties (Félicia, Izabella, and Paola) were chosen for their capacities to be grown under different conditions (field and tunnel). All plants were grafted on Brigeor to improve their resistance to pathogens; they were planted on April 10, 1998, at the CTIFL Balandran Applied Research Centre for conventional plots and at an organic farm, within 1 km of the CTIFL with corresponding similar environmental conditions. Conventional and organic tomatoes were grown in a plastic tunnel (respectively, 324 and 560 m^2). A similar experimental design was used for both cultural practices. Plants (52/variety) were planted in two rows. Cultivation was conducted in soil improved by supplying commercial fertilizers for conventional conditions: NO_3 NH_4 = 116 kg/ha, P_2O_5 = 110 kg/ha, K_2O = 150 kg/ha, CaO = 43 kg/ha, and MgO = 141 kg/ha. Compost was used for organic conditions, corresponding to N = 340 kg/Ha, P_2O_5 = 406 kg/ha, and K_2O = 392 kg/ha. Soil analysis was carried out. Characteristics are reported in **Table 1**. Irrigation and mechanical crop protection were similar for both cultures. Integrated pest management was used for conventional cultivation. Vertimec was applied in nursery to prevent virus transmission by Western Flower Thrips. Applaud was applied at week 16 to reduce greenhouse whitefly populations before releases of *Encarsia formosa* at week 17 and *Macrolophus caliginosus* at week 21. Introduction of *Aphelinus abdominalis* and *Diglyphys isaea* were also performed at week 19 to control respectively aphids and leaf-miners. For organic cultivation, no chemical and no beneficial insect was used nor introduced.

Preparation of Plant Materials. Fruits from the three varieties, Félicia, Izabella, and Paola, were harvested twice a week from mid June to the end of July at red maturity stage, leading to 12 samples for each variety. All fruits were stored in a room regulated at 4 °C. Every day harvested fruits were checked. Firmness was evaluated with a Durofel apparatus. Firmness was reduced by 16% for conventionally grown tomatoes and by 7% for organic ones at the end of storage. Color was checked using a Minolta apparatus giving tristimulus coordinates L^*, a^*, and b^*. Fruits (90 pieces/variety/mode of culture) were randomly taken from the larger harvested sample and divided into three samples of 30 pieces. Color was checked at the beginning and at the end of the storage period. Within the subsample, each fruit was cut into four quarters and two opposite quarters were immediately frozen in liquid nitrogen and finely powdered using a Dangoumill crusher; powder was stored at −20 °C until analyzed. Two tomato purees were prepared at INRA Gruissan with a huge sample of tomatoes (one-third Félicia, one-third Izabella, and one-third Paola) grown either under conventional mode or the organic one at the end of storage (1 day after the last day of harvest due to the transport of fruits). The "Flash-Détente" process was applied to diminish the losses in microconstituents: heating for 8 min at 95 °C followed by a decrease of the pressure to 70 mbar. Purees obtained were kept in sealed aseptic bags. When bags were opened, containers (200 mL) were filled with tomato purees and flash-pasteurized to further store at room temperature and to better control the precise quantity taken by volunteers. For all of the analyses, tomato purees were submitted to liquid nitrogen and then finely powdered using a Dangoumill crusher (leading to similar conditions as described for fresh fruits); powder was stored at −20 °C until analyzed.

Table 1. Characteristics of Soils Used for Conventional and Organic Culture of the Three Varieties of Tomatoes

	conventional	organic
humidity (%)	9.2	15
organic matter (%)	1.8	4.1
pH	7.6	7.7
electric conductivity (mS/cm)	0.42	0.18
N (NH_4) (mg/kg)	2.5	1.5
N (NO_3) (mg/kg)	2.1	4.5
P (H_2PO_4) (mg/kg)	3	20.1
K (mg/kg)	66.6	142.8
Ca (mg/kg)	221	49
Mg (mg/kg)	38.1	9.3
Cl (mg/kg)	9.5	38.9
Na (mg/kg)	25.4	15.9

Dry matter content was determined after the tomatoes or tomato purees had been dried to constant weight, for ∼7 h, under vacuum at 70 °C.

Extraction and Analysis of Carotenoids in Tomatoes and Tomato Purees. The extraction and analysis of carotenoids from tomatoes were performed according to the procedure described by Cabibel and Ferry (*17*). Briefly, tomato powder or puree (5 g) was homogenized in deionized water (5 mL) using a Polytron blender. Extractions with acetone (275 mL in total) were performed until complete discoloration. Carotenoids were extracted using petroleum ether (50 mL). The petroleum ether phase was evaporated, and the residue was dissolved in 2 mL of petroleum ether before HPLC analysis. Separation and determination of carotenoids were performed by HPLC (Hewlett-Packard 1050 connected to a diode array detector) using a Vydac 201TP54 C_{18} 5 μm (250 mm × 4.6 mm, Interchim) column equipped with a precolumn (10 mm × 4.6 mm, 5 μm), both kept at 30 °C. The solvent system used was an isocratic mixture of acetonitrile/methanol/methylene chloride (60:38:2; v/v/v). The solvent flow rate was 1 mL/min. External calibration was performed using lycopene and β-carotene purchased from Sigma (Steinheim, Germany). Linearity was from 2 to 500 ng of carotenoid. Quantification was carried out in triplicate.

Extraction and Analysis of Phenolic Compounds in Tomatoes and Tomato Purees. Powder of tomato or tomato purees (10 g) was homogenized in 150 mL of cold aqueous ethanol (75%) containing sodium metabisulfite (0.5%) for 1 min using an Ultra-Turrax blender. Three successive extractions with ethanol (75%) were carried out at 4 °C for 15 min. After removal of the alcohol under vacuum at 35 °C,

ammonium sulfate (20%) and metaphosphoric acid (2%) were added to the aqueous phase. Pigments were eliminated by three successive extractions with petroleum ether (2:1, v/v). After methanol (20%) had been added to the aqueous phase, phenolic compounds were extracted three times successively with ethyl acetate (1:1, v/v). The three organic phases were combined, filtered on Whatman paper (silicone-treated phase separator) to eliminate aqueous phase, and evaporated to dryness under vacuum at 38 °C. The residue was dissolved in methanol (1 mL). Methanol extracts were filtered through an Acrodisc filter (0.45 μm) before analysis by HPLC. Extraction was carried out in triplicate on the same material. Separation and determination of phenolics were performed by HPLC (Varian 5500 connected to a diode array detector, Waters 990) using a Chrompack C_{18} 7 μm (200 mm × 3 mm i.d., Alltech) column. The mobile phase was a gradient of A (water; 2% acetic acid) and B (methanol). The best separation was obtained with the following gradient: at 0 min, 0% B; at 4.5 min, 7% B; at 13.5 min, 7% B; at 20 min, 10% B for 3 min; at 25 min, 14% B for 5 min; at 32 min, 20% B for 3 min; and at 60 min, 45% B for 15 min. After 75 min, the column was washed with 100% acetonitrile for 10 min and re-equilibrated to the initial conditions. The solvent flow rate was 0.8 mL/min, and the separation was performed at 35 °C. A diode array detector (Waters 990) was used for the characterization of each peak and for its quantification. The main phenolic compounds were assayed by external standard (ES) calculation at 320 nm for chlorogenic acid and rutin and at 280 nm for naringenin-7-*O*-glucoside (expressed as naringenin equivalent). Linearity was from 2.5 ng to 10 μg of phenolic standards purchased from Extrasynthèse (Genay, France). Quantification was carried out in triplicate.

Analysis of Vitamin C in Tomato and Tomato Purees. Powder of tomato or tomato puree (5 g) was suspended with Norit (Sigma) in 50 mL of a mixture of metaphosphoric acid 4%/methanol (3:1, v/v). The suspension was oxygenated with air during 3 min just before the analysis to transform ascorbic acid into dehydroascorbic acid. Concentration of dehydroascorbic acid was determined at 37 °C with a Technicon autoanalyzer using fluorometric detection according to the procedure described in the literature (*18*, *19*). Linearity was from 2 to 50 μg of L(+)-ascorbic acid (Prolabo, 99.7% purity).

Macronutrient and Energy Composition of Tomato Purees. Energy and macronutrient composition were estimated by using the French food composition table (*20*). One hundred grams of fresh matter corresponded to 215 kJ, protein = 2.3 g, fat = 0.5 g, and carbohydrates = 9.5 g.

Study Population. Twenty young nonsmoking females, aged 21−39 years, were recruited. None was taking oral medication, apart from oral contraceptives, or supplements of any kind during the month preceding the beginning of the study or during the study period. The study was approved by the regional committee on human experimentation of the University Hospital in Clermont-Ferrand (France): research project AU 237, approved May 15, 1998. Written consent was obtained from each volunteer. The subjects were healthy, according to clinical examination and disease history. Their lean and fat masses were measured by bioelectric impedance measurement using a BIA 101A instrument (RJL Systems, Mt. Clemens, MI). Fasting plasma triacylglycerol, cholesterol, and glycemia were measured to assess the normality of their lipid and carbohydrate metabolism. Their usual diet was estimated using a 5-day food diary (i.e., subjects wrote the food they ate, kind and serving, during 5 consecutive days including a weekend). This diary was analyzed for nutrient composition using diet analyzer software (GENI, Micro 6, Nancy, France). The software database was extended for carotenoids with a carotenoid food-composition database (*21*). The subjects' characteristics are given in **Table 2**.

Supplementation Experiment. Subjects were randomly divided into two groups. There was no significant difference between the two groups with regard to all of the variables measured (**Table 2**). The subjects were asked to ingest, during their meal (lunch or dinner), 100 g/day of tomato puree, during 3 weeks. The subjects did not know whether they ate the organically produced tomato puree or the conventional one. Note that the real intake of tomato puree was estimated at ∼96 g/day because of losses in the container. Group I received the conventionally produced tomato puree, and group II subjects received the organically produced tomato puree. Compliance was checked by regularly telephoning the subjects and by reviewing a notebook in which volunteers recorded when they ate the puree. After the supplementation period, the subjects were asked to avoid foods rich in tomato products as much as possible (a list of such foods was given to the volunteers) for a subsequent period of 3 weeks (the depletion period).

Table 2. Subject Characteristics[a] at the Beginning of the Study

	group I	group II
age (years)	25.80 ± 1.89	24.40 ± 0.67
BMI (kg/m^2)	20.54 ± 0.52	20.74 ± 0.49
glycemia (mmol/L)	4.18 ± 0.14	4.33 ± 0.14
triacylglycerol (mmol/L)	0.80 ± 0.07	0.80 ± 0.08
cholesterol (mmol/L)	4.74 ± 0.27	4.96 ± 0.11
daily macronutrient intake[b]		
energy (MJ)	7.05 ± 0.46	7.78 ± 0.36
protein (% of energy)	17.96 ± 0.95	15.75 ± 0.47
fat (% of energy)	36.78 ± 1.24	36.17 ± 0.67
carbohydrates (% of energy)	43.40 ± 1.49	46.63 ± 1.07
alcohol (% of energy)	1.86 ± 0.42	1.45 ± 0.53
daily vitamin C and carotenoid consumption (mg)		
vitamin C	92.76 ± 15.66	136.52 ± 21.96
lutein/zeaxanthin	0.89 ± 0.19	1.04 ± 0.33
β-cryptoxanthin	0.09 ± 0.02	0.17 ± 0.05
lycopene	1.93 ± 0.76	2.36 ± 0.79
α-carotene	0.34 ± 0.12	0.46 ± 0.17
β-carotene	5.28 ± 1.97	5.65 ± 1.33

[a] Mean ± SEM, $n = 10$ for each group. There was no significant difference between the two groups with regard to all of the measured parameters (Student's t test for unpaired values, $P < 0.05$). [b] Dietary habits were estimated using a 5 day food recall at the beginning of the experiment.

Plasma Preparation. Blood samples were collected, after at least a 12-h overnight fast, the day before the supplementation period was begun, the day after supplementation had finished, and 3 weeks later, that is, at the end of the depletion period. They were protected from light to prevent losses of carotenoids. Blood samples for carotenoid analysis were collected in disodium ethylenediaminetetraacetate (EDTA) containing tubes to prepare plasma. Tubes were cooled in an ice bath and centrifuged (1500*g* for 10 min at 10 °C) within 30 min after blood collection. Aliquots were kept at −80 °C until analysis of carotenoids. Aliquots for vitamin C analysis were separated within 30 min and deproteinized by the addition of 2 volumes of fresh 5% w/v metaphosphoric acid during 30 min. The supernatants were kept frozen at −70 °C until final assessment of vitamin C within 2 weeks.

Evaluation of Plasma Vitamin C. Vitamin C status was evaluated by measuring total plasma ascorbate concentration (*22*). This concentration was measured by HPLC using fluorometric detection at 360 nm (excitation) and 440 nm (emission) as previously described (*23*). Chromatography was performed using a model 422 pump (Kontron), a Rheodyne 7125 injection valve, and a model 2475 fluorometer (Waters). A reverse phase (5 μm, 250 × 0.46 mm) column supplied by Merck was eluted with a mobile phase consisting of 0.05 M sodium phosphate/methanol (50:50, v/v), pH 7.4. Ascorbic acid standard samples were prepared and diluted to 17.6 mg/100 mL in 5% HPO_3. The final solutions containing 0.25, 0.50, 1.0, and 1.5 mg/100 mL were prepared by diluting 0.14, 0.28, 0.57, and 1.14 mL of stock solution to 10 mL with 5% HPO_3. Standard curves were prepared by plotting ascorbic acid concentration against peak area. The linearity of the method with acceptable criteria of precision has been tested between 0.1 and 100 mg/100 mL. The detection limit was <0.05 mg/100 mL. The intraday assay coefficient of variation was <2% and the interday assay variability was <4% for the lower (0.25 mg/100 mL) and higher (1.5 mg/100 mL) concentrations tested.

Evaluation of Plasma Carotenoids. Carotenoid status was evaluated by measuring plasma carotenoid concentrations (*24*). Carotenoids were extracted twice with ethanol and hexane. Echinenone (Roche Vitamines France, Neuilly-sur-Seine, France) was used as internal standard. β-Carotene and lycopene were quantified by reverse phase HPLC on a Kontron apparatus (Zurich, Switzerland) with detection at 450 nm. They were separated using two columns placed in series (*25*): a 175

Table 3. Concentration of Microconstituents in Different Varieties of Tomatoes Grown under Conventional (conv) and Organic (org) Conditions, Expressed in Milligrams per 100 g of Fresh Matter

micro-constituent	Félicia		Izabella		Paola		variety effect[a]	culture effect[a]	variety × culture interactions
	conv	org	conv	org	conv	org			
lycopene	3.8 ± 0.10	4.2 ± 0.10	3.2 ± 0.10	3.6 ± 0.40	3.4 ± 0.10	4.1 ± 0.10	$P < 0.0001$	$P < 0.0001$	NS
β-carotene	0.92 ± 0.04	1.31 ± 0.12	0.86 ± 0.04	1.03 ± 0.04	0.83 ± 0.05	1.35 ± 0.12	NS	$P < 0.0001$	NS
vitamin C	13.2 ± 0.50	17.5 ± 0.60	9.6 ± 0.30	16.2 ± 0.70	13.7 ± 0.90	12.5 ± 0.90	$P < 0.05$	$P < 0.0001$	$P < 0.0001$
chlorogenic acid	0.64 ± 0.02	0.55 ± 0.02	0.17 ± 0.005	0.18 ± 0.01	0.65 ± 0.02	0.48 ± 0.01	$P < 0.0001$	$P < 0.0001$	$P = 0.0001$
rutin	0.15 ± 0.004	0.18 ± 0.01	0.04 ± 0.001	0.22 ± 0.01	0.10 ± 0.003	0.14 ± 0.004	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$
naringenin	0.08 ± 0.004	0.10 ± 0.003	0.08 ± 0.003	0.09 ± 0.003	0.09 ± 0.003	0.09 ± 0.003	NS	$P < 0.001$	$P < 0.05$

[a] Two-way ANOVA was performed to check whether there was a variety effect and/or a culture effect on microconstituent concentrations. NS, nonsignificant.

Table 4. Concentration of Microconstituents in Different Varieties of Tomatoes Grown under Conventional (conv) and Organic (org) Conditions, Expressed in Milligrams per 100 g of Dry Matter[a]

micro-constituent	Félicia		Izabella		Paola		variety effect	culture effect
	conv	org	conv	org	conv	org		
lycopene	62.29 ± 1.63	61.76 ± 1.47	54.24 ± 1.69	51.43 ± 5.71	57.63 ± 1.69	62.12 ± 1.51	$P < 0.001$	NS
β-carotene	15.08 ± 0.66	19.26 ± 1.76	14.58 ± 0.68	14.71 ± 0.57	14.07 ± 0.85	20.45 ± 1.82	NS	$P < 0.001$
vitamin C	216.39 ± 8.20	257.35 ± 8.80	162.71 ± 5.10	231.42 ± 10.00	232.20 ± 15.20	189.39 ± 13.60	$P < 0.05$	$P < 0.05$
chlorogenic acid	10.49 ± 0.33	8.09 ± 0.29	2.88 ± 0.08	2.57 ± 0.14	11.02 ± 0.34	7.27 ± 0.15	$P < 0.0001$	$P < 0.0001$
rutin	2.46 ± 0.06	2.65 ± 0.15	0.68 ± 0.02	3.14 ± 0.14	1.69 ± 0.05	2.12 ± 0.06	$P < 0.0001$	$P < 0.0001$
naringenin	1.31 ± 0.06	1.47 ± 0.04	1.36 ± 0.05	1.29 ± 0.04	1.52 ± 0.01	1.36 ± 0.04	NS	NS
dry matter (g/100 g)	6.10 ± 0.04	6.8 ± 0.04	5.9 ± 0.16	7.0 ± 0.09	5.9 ± 0.13	6.6 ± 0.07	$P < 0.05$	$P < 0.0001$

[a] For statistics see **Table 3** footnote. NS, nonsignificant.

× 4.6 mm RP C_{18}, 3 μm Nucleosil (Interchim, Montluçon, France), coupled with a Vydac 250 × 4.6 mm RP C_{18}, 5 μm Vydac TP54 (Hesperia, CA), and a Hypersil guard column. The mobile phase was an isocratic mixture of acetonitrile/methanol/dichloromethane/water (70:15:10:5, v/v/v/v). Lycopene and β-carotene were detected at 450 nm and identified by comparison of their retention time and spectral analysis (from 300 to 550 nm) with those of pure (>95%) standards (Roche Vitamines France). The range of the assay was 2−500 ng of carotenoids/sample. The linearity of the assay was $r > 0.99$. Internal standard allowed to calculate an overall recovery yield of 75−100%.

Statistical Analysis. Results are expressed as means ± SEM. The effect of variety and culture on microconstituent composition of tomatoes was assessed by using two-way analysis of variance (ANOVA). Concerning the human experiment, two-way ANOVA, with time and kind of tomato puree as factors, was used to determine whether there were significant differences between the measured parameters. When a significant ($P < 0.05$) difference was detected, means were compared either using the post-hoc Newman−Keuls test for time comparisons, that is, before supplementation, after supplementation, and after the depletion period, or using the Student t test for group comparisons at a given time. These statistical comparisons were performed using StatView software version 5.0 (SAS Institute, Cary, NC).

RESULTS AND DISCUSSION

Effect of Cultural Practices on Different Varieties of Tomatoes. Concentrations of microconstituents per 100 g of fresh matter for three varieties of tomatoes (Paola, which is a variety commonly cultivated in organic mode, and Félicia and Izabella, usually cultivated under conventional mode), are shown in **Table 3**. Comparing the composition of tomatoes on a fresh matter basis is essential when the nutritional value of the fruit is considered. The dry matter content of microconstituents is more pertinent when comparisons of the effect of the mode of culture on the composition of the plant are made; data on dry matter are given in **Table 4**.

On a fresh matter basis, the concentrations of all the microconstituents assayed, except that of chlorogenic acid, were significantly higher in organic tomatoes. The effect of organic farming appeared to be more pronounced when contents are expressed on fresh matter than on dry matter (**Tables 3** and **4**): +42% for β-carotene (+25% in dry matter), +14% for lycopene [nonsignificant (NS) in dry matter], +31% for vitamin C (+14% in dry matter), +170% for rutin (+132% in dry matter), +12.5% for naringenin (NS in dry matter), and −23% for chlorogenic acid (−11% in dry matter). The higher level in carotenoid for organic tomatoes was in good agreement with the higher red value a^* (20 for organic tomatoes vs 17 for conventional ones). The differences between the values expressed on fresh matter and those expressed on dry matter are directly related to the higher dry matter content of organic versus conventional tomatoes (**Table 4**). As previously reported in different crops (*1*), especially for leafy vegetables (*26*), dry matter is often higher in organically grown plants than in conventionally grown ones. In fact, in organic fertilization, nitrogen is absorbed by the plant after mineralization, and the development of the plant is not "forced"; as a consequence, higher dry matter content was found (*27*).

Controversial results have been reported in the literature on carotenoids. Lucarini et al. (*28*) found a higher concentration of β-carotene expressed as fresh matter in conventionally grown tomatoes, but no difference was observed for the concentration of lycopene, as also reported by Kopp et al. (*29*). Considering lycopene and β-carotene together in fresh tomato, Clarke and Merrow (*30*) showed no significant difference for one year of cultivation and higher contents in conventional cultures for the second year. Carotene content has been estimated in 27 studies on carrots cultivated according to either conventional or organic practices, as reviewed (*1*, *26*). Globally, no significant differences were found for the concentration of carotenes between the conventionally and organically grown carrots. It is also well-known that carotenoid content in fruits and vegetables, and particularly in tomatoes, is strongly dependent on other factors such as light exposure and temperature (*31*) and maturity stage (*17*). Our results expressed as fresh matter showing a higher carotene content in organic tomatoes have to be confirmed by

repeating the experiments over two to three years. Moreover, our results confirmed that variety significantly affects the content of lycopene, whereas no variety effect was found for β-carotene.

Fresh and dry matter contents of vitamin C were higher in organic tomatoes (**Tables 3** and **4**). Data reported in the literature for vitamin C were also controversial. For tomatoes, Lucarini et al. (*28*) found significantly higher vitamin C content expressed as dry matter in organic fruits, but Auclair et al. (*32*) found the contrary. Clarke and Merrow (*30*) reported opposite results between two cultivation years. In a study over six years, Fjelkner-Modig et al. (*33*) did not find significant differences in vitamin C content, also expressed as dry matter, of a range of vegetables grown in organic or conventional mode. As a general statement in published results on fruits and vegetables, no clear difference can be established between the contents of vitamin C in organic and conventional fruits and vegetables (*26*); although a tendency toward higher vitamin C content was found for leafy vegetables grown by organic mode compared to the conventional way (*1*, *34*, *35*). However, because these results were based on fresh matter content, the authors agreed that it could be due to higher water content in conventionally grown plant food products. Moreover, our results showed that the variety effect can have a significant influence on vitamin C content (**Tables 3** and **4**). Despite the significant positive effect of the organic mode of culture on the vitamin C content, both in fresh and in dry matter, a lower vitamin C content was observed for the Paola variety cultivated in the organic mode (**Tables 3** and **4**).

For phenolic compounds, the concentrations of chlorogenic acid expressed on both fresh and dry weight bases were lower in organically grown tomatoes. It is known that the content of hydroxycinnamic acids of fruits and vegetables rapidly decreases with maturity (*36*). It is possible that organic tomatoes were harvested at a slightly later stage of maturity than conventional ones as previously shown in regard to a^* values, which could be consistent with their lower content of chlorogenic acid. However, the effect of maturity stage was certainly minimized due to the high number of samples analyzed for each variety. Rutin and naringenin contents in fresh matter were significantly higher in organically grown tomatoes (**Table 2**). Data on phenolic composition of fruits and vegetables grown by either organic or conventional ways remain scarce in the literature, because these compounds have only recently been considered as interesting antioxidant microconstituents but not yet essential for human health. Among 11 recent papers mentioning total phenolic contents, 8 of these studies mentioned a higher content of phenolic concentration in organically grown fruits or vegetables (*37*). It is well-known that the biosynthesis of phenolic compounds in plants is strongly influenced by cultivar (*38*), maturity stage, environmental conditions, especially light, and mode of fertilization (*39*). As previously reported, the level of nitrogen influences the level of phenol (*40*, *41*). Even if the level of N as well as K and P was higher in organic conditions, the bioavailability of nitrogen from organic ferlilization remained lower than the synthetic one; the level of secondary metabolites with carbone (such as phenolic compounds and terpenes) is usually higher in organic plants (*40*, *41*). Such an increase is related to the well-known defensive role of phenolic compounds in plants under stressed conditions (*42*).

Composition in Microconstituents of Tomato Purees Made from Organic and Conventional Tomatoes. Purees of tomatoes were prepared for human studies according to a technologic process known to preserve the content in microconstituents, especially those with a high oxidizability such as vitamin C and phenolic compounds. Purees were prepared by mixing equal proportions of the three varieties of tomatoes obtained either in a conventional way or by organic culture. One kilogram of tomatoes was necessary to prepare 143 g of tomato puree. Concentrations of microconstituents for the two purees are given in **Table 5**.

Table 5. Concentration of Microconstituents in Tomato Purees, Expressed in Milligrams per 100 g of Fresh Matter[a]

microconstituent	conv	org	org vs conv
lycopene	15.57 ± 2.19	13.54 ± 0.60	NS
β-carotene	3.56 ± 1.02	1.71 ± 0.32	NS
vitamin C	22.53 ± 1.07	39.95 ± 0.44	$P < 0.0001$
chlorogenic acid	7.2 ± 0.2	10.6 ± 0.3	$P < 0.001$
rutin	2.30 ± 0.04	9.65 ± 0.28	$P < 0.0005$
naringenin	4.83 ± 0.14	6.18 ± 0.18	$P < 0.005$

[a] For statistics see **Table 3** footnote. NS, nonsignificant.

Our results showed a significantly higher content of vitamin C and phenols in organic tomato purees, whereas no difference was seen for carotenoid content. The higher content of vitamin C of puree made with organic tomatoes was consistent with the results on fresh tomatoes. Concerning phenolic compounds, their concentration was significantly higher in the puree made of organically grown tomatoes, even for chlorogenic acid, the concentration of which is very sensitive to the stage of maturity as mentioned above. However, the process did not allow keeping the differences in carotenoid content observed in fresh tomatoes and appeared to affect the content in lipid-soluble microconstituents. It is known that thermal processing generally causes some losses of lycopene in tomato-based foods (*43*, *44*). On the other hand, processing can have a beneficial effect on lycopene bioavailability by breaking down cell walls, thus making lycopene more accessible for absorption (*45*−*47*).

Effect of Long-Term Supplementation with Conventionally or Organically Produced Tomato Puree on Plasma Carotenoids and Vitamin C. The change in plasma concentrations of β-carotene and lycopene after supplementation with the tomato purees for 3 weeks is shown in **Figure 1A**,**B**. The consumption of organically or conventionally produced tomato puree for 3 weeks increased plasma β-carotene and lycopene. However, there was no significant difference in the concentrations measured in the two groups. This result is logical because the two purees had similar lycopene and β-carotene contents. In addition, it suggests that the higher content of water-soluble antioxidants in organic puree (i.e., vitamin C and polyphenols), which are theoretically able to protect carotenoids from oxidative degradation, had no influence on the bioavailability of carotenoids. Changes in plasma concentrations of vitamin C during the experiment are shown in **Figure 2**. After 3 weeks of tomato consumption, one can see no significant difference between the two groups, so the higher content of vitamin C in puree made from organically grown tomatoes was not sufficient to affect plasma vitamin C. We noted that the plasma level of vitamin C was significantly lower after the 3 week wash-out period than at the beginning of the study. This was not expected because subjects were asked to go back to their usual diet during the wash-out period. They probably ate fewer fruits and vegetables during this period, for no explainable reason.

Phenolics were not considered because of their low concentrations in tomatoes and their low bioavailability in human. The lack of difference between organic and conventional tomato puree can be explained by several hypotheses. First, one reason could be the small differences between microconstituent contents of the two purees. Second, it is possible that the duration of

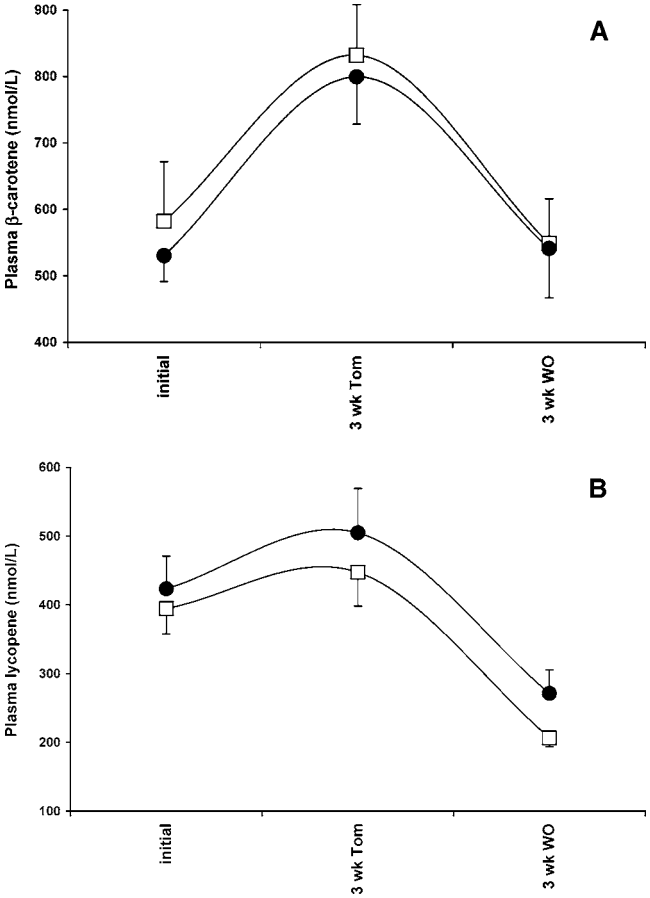

Figure 1. Variation of plasma carotenoids during supplementation: (**A**) β-carotene; (**B**) lycopene; (●) group supplemented with the conventional tomato puree; (□) group supplemented with the organic tomato puree; 3 wk Tom, after the consumption of tomato puree for 3 weeks; 3 wk WO, after a period of 3 weeks during which the subjects returned to their usual diet (wash-out). Note that there was no significant difference between the two groups with regard to the microconstituent concentration measured at each time (as assessed by two-way repeated measure ANOVA with group and time as factors).

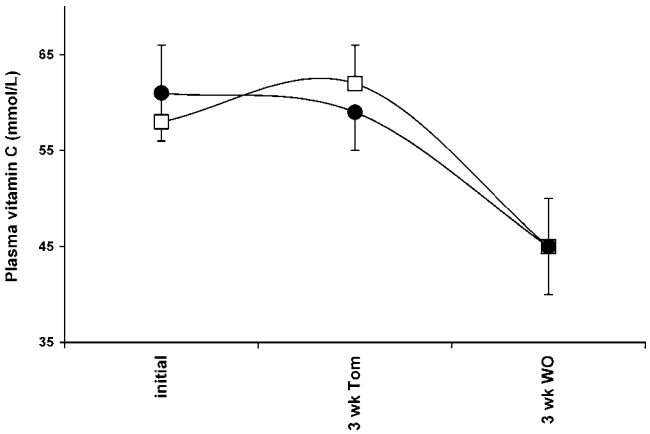

Figure 2. Variation of plasma vitamin C during supplementation. For details see **Figure 1** caption.

supplementation was too short to observe an effect. Finally, it is likely that the effect of organic puree was masked by the rest of the diet, which consisted of conventional foods in the two groups.

In conclusion, this study showed that organic cultivation can provide tomatoes and tomato-derived products with significantly higher contents of antioxidant microconstituents. However, it appeared to be difficult to get a beneficial effect with the consumption of only one organic food product, in regard to the antioxidant microconstituent content. Further human intervention studies need to be conducted with a diet richer in organic food products.

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