

Effects of Different Cooking Methods on Nutritional and Physicochemical Characteristics of Selected Vegetables

CRISTIANA MIGLIO,[†] EMMA CHIAVARO,[‡] ATTILIO VISCONTI,[§]
VINCENZO FOGLIANO,[§] AND NICOLETTA PELLEGRINI^{*,†}

Department of Public Health, University of Parma, via Volturno 39, 43100 Parma, Italy, Department of Industrial Engineering, University of Parma, viale GP Usberti 181/A, 43100 Parma, Italy, and

Department of Food Science, University of Napoli “Federico II”,
Parco Gussone, 80055 Portici, Napoli, Italy

The objective of the present study was to evaluate the effect of three common cooking practices (i.e., boiling, steaming, and frying) on phytochemical contents (i.e., polyphenols, carotenoids, glucosinolates, and ascorbic acid), total antioxidant capacities (TAC), as measured by three different analytical assays [Trolox equivalent antioxidant capacity (TEAC), total radical-trapping antioxidant parameter (TRAP), ferric reducing antioxidant power (FRAP)] and physicochemical parameters of three vegetables (carrots, courgettes, and broccoli). Water-cooking treatments better preserved the antioxidant compounds, particularly carotenoids, in all vegetables analyzed and ascorbic acid in carrots and courgettes. Steamed vegetables maintained a better texture quality than boiled ones, whereas boiled vegetables showed limited discoloration. Fried vegetables showed the lowest degree of softening, even though antioxidant compounds were less retained. An overall increase of TEAC, FRAP, and TRAP values was observed in all cooked vegetables, probably because of matrix softening and increased extractability of compounds, which could be partially converted into more antioxidant chemical species. Our findings defy the notion that processed vegetables offer lower nutritional quality and also suggest that for each vegetable a cooking method would be preferred to preserve the nutritional and physicochemical qualities.

KEYWORDS: Phytochemicals; antioxidant capacity; texture; color; carrots; courgettes; broccoli; cooking methods

INTRODUCTION

There is substantial evidence for the role of diet in the prevention of cancer (1), cardiovascular, and other degenerative diseases (2). Fruits and vegetables are considered particularly protective thanks to their content of phytochemicals. These naturally occurring compounds have attracted great attention from the scientific community for their antioxidant properties and their implication in a variety of biological mechanisms at the base of degenerative processes (2). Such compounds are secondary plant metabolites responsible for plant food color, smell, flavor, and bitterness and consist of a wide variety of different molecules, such as carotenoids, polyphenols, vitamins, and glucosinolates.

Most vegetables are commonly cooked before being consumed. It is known that cooking induces significant changes in

chemical composition, influencing the concentration and bioavailability of bioactive compounds in vegetables. However, both positive and negative effects have been reported depending upon differences in process conditions and morphological and nutritional characteristics of vegetable species (3–7).

Physical properties of vegetables are also greatly affected by heat treatments (8, 9). Texture and color are considered very important parameters in the cooking quality of vegetables, and they may strongly influence consumer purchases of these food items. Changes in texture are often dramatic because of the membrane disruption and the associated loss of turgor (8). In addition, cooked vegetables exhibit poor color quality in comparison with fresh ones (9).

Although consumption of fresh unprocessed plant food is widely advocated, evidence is emerging that *in vivo* bioavailability of many protective compounds is enhanced when vegetables are cooked (1). However, data on the effect of cooking on nutritional properties of vegetables are still incomplete (1). In fact, literature data on nutritional properties of cooked vegetables often deal with a single vegetable (10), a family of vegetables (7), or a single phytochemical group (4).

* To whom correspondence should be addressed. Telephone ++39-0521-903907. Fax: ++39-0521-903832. E-mail: nicoletta.pellegrini@unipr.it.

[†] Department of Public Health, University of Parma.

[‡] Department of Industrial Engineering, University of Parma.

[§] Department of Food Science, University of Napoli “Federico II”.

A more integrated analysis of nutritional and physical properties of vegetables is needed to obtain insight into the effect of cooking.

In the present study, the effect of cooking practices (i.e., boiling, steaming, and frying) on phytochemical contents (i.e., ascorbic acid, carotenoids, polyphenols, and glucosinolates) and total antioxidant capacities of three vegetables (carrots, courgettes, and broccoli), chosen on the basis of their different morphological feature, nutritional profiles, and antioxidant capacities (11, 12), was evaluated. Nutritional data are also compared to changes of texture and color induced by cooking.

MATERIALS AND METHODS

Chemicals. The 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), 2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), 2,4,6-tripyridyl-s-triazine (TPTZ), β -carotene, lutein, quercetin, rutin, chlorogenic acid, caffeic acid, *p*-coumaric acid, ferulic acid, sinapic acid, kaempferol, morin, 2,6-di-*tert*-butyl-*p*-cresol (BHT), and sinigrin (allyl glucosinolate) were purchased from Sigma-Aldrich (Sigma Chemical Co., St. Louis, MO). *R*-phycoerythrin (*R*-PE) was purchased from Prozyme (San Leandro, CA), and 2,2'-azobis(2-amidinopropane) dihydrochloride (ABAP) was purchased from Waco Chemicals (Richmond, VA).

All chemicals and solvents used were HPLC-grade and purchased from Carlo Erba (Milan, Italy). High-purity water was produced in the laboratory using an Alpha-Q system (Millipore, Marlborough, MA).

Preparation of Vegetables. Freshly harvested carrots (*Daucus carota* L.), courgettes (*Cucurbita pepo* L.), and broccoli (*Brassica oleracea*, var. *botrytis caput* L.) of a single batch were purchased from a local market. Carrots, peeled before processing, and courgettes were prepared by cutting off the top and bottom ends with a knife and extracting cylindrical specimens (diameter, 25 mm; height, 25 mm) from each sample. Broccoli were cleaned by removing the inedible parts and then chopped into homogeneous pieces, leaving a stem of 25 mm.

To obtain more homogeneous samples, each vegetable was prepared in batches of 400 g. Each batch was then divided into four equal portions. One portion was retained raw, and the others were cooked in three different methods in triplicate, as given below.

Cooking Treatments. Three of the most common cooking methods used by the Italian population, i.e., boiling, steaming, and frying, were used. Cooking conditions were optimized by preliminary experiments carried out for each vegetable. For all cooking treatments, the minimum cooking time to reach a similar tenderness for an adequate palatability and taste, according to the Italian eating habits, was used.

Boiling. Vegetable material was added to boiling tap water in a covered stainless-steel pot (1:5 food/water) and cooked on a moderate flame. For each cooking trial, 10 samples were boiled. Then, samples were drained off for 30 s.

Steaming. Steaming treatments were carried out in a Combi-Steal SL oven (V-Zug, Zurich, Switzerland). Nine specimens were placed in the oven equilibrated to room temperature before each cooking trial. Eight samples were arranged in a circle, and one was placed at the center to ensure uniform heating conditions in all samples for each cooking trial. The samples were cooked under atmospheric pressure.

Frying. Vegetable was added to 2.2 L of peanut oil in a domestic deep fryer (DeLonghi, Italy) at 170 °C. A total of 10 samples were fried for each cooking trial. At the end of each trial, samples were drained off and dabbed with blotting paper to allow for the absorption of exceeding oil.

After all cooking experiments, samples were cooled rapidly on ice for antioxidant analyses. The texture analyses were performed on cooked samples at 50 °C, referred as the temperature of consumption, while color analyses were performed at room temperature (25 °C). Both temperatures were controlled inserting a thermocouple (K-type; Ni/Al-Ni/Cr) connected to a multimeter acquisition system (Keithley Instruments, Inc., Cleveland, OH) to the thermal center of one sample for each cooking trial.

Dry Matter Determination. For the determination of the moisture, 3–4 g of raw or cooked homogenized sample (as triplicate) was dried in a convection oven at 105 °C for at least 16 h until reaching a constant weight.

Texture Analysis. Texture of the raw and cooked samples was analyzed by a shear force test using a TA.XT2 Texture Analyzer equipped with a 25 kg load cell (Stable Micro Systems, Goldalming, U.K.), and the parameters were quantified using the application software provided (Texture Expert for Windows, version 1.22).

Shear force analysis was performed using a Warner-Bratzler blade (3 mm thick), which cut the cylindrical specimen of carrot and courgette cylinders parallel to their major axis, whereas broccoli was cut between the flower heads and the end of the stems at a constant speed of 60 mm/min and pushed through the slot (4 mm wide). The maximum force (*N*) required to shear the sample was measured. Softening (%) was calculated as

$$\text{percent softening} = \left(1 - \frac{\text{shear force of cooked sample}}{\text{shear force of raw sample}}\right) \times 100$$

A total of 15 determinations was performed for each cooking treatment.

Color Analysis. Color determination was carried out using a Minolta Colorimeter (CM 2600d, Minolta Co., Osaka, Japan) equipped with a standard illuminant D₆₅. Both raw and cooked samples were analyzed. The assessments were carried out both on the external and internal surfaces of the cylindrical specimens of carrots and courgettes cut parallel to their major axis and on the florets and stems of broccoli. *L** (lightness; black = 0, white = 100), *a** (redness > 0, greenness < 0), *b** (yellowness > 0, blue < 0), *C* (chroma, 0 at the center of the color sphere), and *hue*° (hue angle, red = 0°, yellow = 90°, green = 180°, blue = 270°) were quantified on each sample using a 10° position of the standard observer (13).

A total of 15 determinations was performed for each cooking treatment.

Determination of Antioxidant Compounds and Total Antioxidant Capacity (TAC). For the analyses of antioxidant compounds, with the exception of ascorbic acid, the samples were freeze-dried using a Brizzio-Basi instrument (Milan, Italy). Dried sample material was finely ground, kept in sealed bags, and stored at -20 °C. Analyses of ascorbic acid and TAC were performed on fresh samples within 24 h of cooking.

For all of the parameters considered (e.g., TAC and carotenoid and polyphenol content), the cooking effect was reported as the percent variation with respect to uncooked vegetable and calculated according to the following equation: [(value of cooked sample - value of raw sample)/value of raw sample] × 100.

Carotenoids. The determination of carotenoids was carried out by high-performance liquid chromatography (HPLC) analysis as previously described by Leonardi et al. (14). Briefly, 0.1 g of lyophilized sample was extracted with tetrahydrofuran containing 0.01% BHT as the antioxidant agent, dried under nitrogen flow in dark tubes, resuspended in dichloromethane, and analyzed using a HPLC (Shimadzu LC10, Japan) controlled by Class VP software (Shimadzu, Japan) with a diode array detector (SPD-M10A Shimadzu, Japan) and a Prodigy column (5 μ m ODS3 100A, 250 × 4.6 mm; Phenomenex, Torrance, CA). The injection volume was 20 μ L, and the carotenoids were eluted with a flow of 0.8 mL/min, following this linear gradient: starting condition, 82% A and 18% B; at 20 min, 76% A and 24% B; at 30 min, 58% A and 42% B; at 40 min, 40% A and 60% B; and at 45 min, 82% A and 18% B. Phase A was a mixture of acetonitrile, *n*-hexane, methanol, and dichloromethane (2:1:1:1, v/v/v/v), while phase B was acetonitrile. Identification of the peaks in the HPLC chromatogram of the carotenoid extract was carried out by a comparison of UV-vis spectra or with retention times of eluted compounds with pure standards at 450 nm for α - and β -carotene, β -cryptoxanthin, and lutein, at 350 nm for phytofluene and at 290 nm for phytoene. To quantify phytofluene, phytoene, β -cryptoxanthin, and α -carotene, their respective peak areas were compared to the ones of standard β -carotene at known concentrations, established by the molar extinction coefficient in acetone reported in the literature and corrected by the molar extinction coefficient relative at each compound. The identification of *cis*-carotene isomers was based on spectral characteristics as described by Chen et al. (15). Because

no standards for *cis* isomers are available, the quantification was carried out using the calibration curve of all *trans* isomers.

Polyphenols. Polyphenols before and after deglycosylation, were determined following the procedure described by Crozier et al. (16), with few modifications. Briefly, 1 g of lyophilized sample was extracted with 10 mL of 60% aqueous methanol solution containing 0.25 mg of morin as an internal standard. About 1.5 mL of this solution was kept down; the remaining part was hydrolyzed by adding with 20 mM sodium diethyldithiocarbamate and 5 mL of 6 M HCl, and then it was refluxed at 90 °C for 2 h. A total of 20 μ L of the extract, taken both before and after hydrolysis, was analyzed by HPLC (Shimadzu LC 10, Shimadzu, Japan) with a diode array detector and a Prodigy column (5 μ m ODS3 100A, 250 \times 4.60 mm; Phenomenex, Torrance, CA) at a flow rate of 1 mL/min. The mobile phase was a mixture of water/formic acid (95:5, v/v) (A) and methanol (B). Flavonoid elution was achieved using the following linear gradient: starting condition, 70% A and 30% B; at 3 min, 50% A and 50% B; at 18 min, 40% A and 60% B; at 23 min, 20% A and 80% B; at 28 min, 10% A and 90% B; and at 33 min, 70% A and 30% B. Chromatograms were recorded at 256 nm for flavonols and at 325 nm for phenolic acids.

Glucosinolates. The determination of glucosinolates was carried out after the desulphation reaction according to the procedure described by Kiddle et al. (17) with some modifications. Briefly, 0.2 g of freeze-dried sample was extracted with 3.5 mL aqueous methanol (70:30, v/v) and heated at 70 °C in a heating bath for 10 min. The extracts were centrifuged at 2000g for 10 min at 4 °C; the supernatant was refrigerated, while the pellet was extracted a second time with 3 mL of aqueous methanol (70:30, v/v), heated at 70 °C, and centrifuged using the previous conditions. The two supernatants were combined and refrigerated. The desulphation reaction was performed with mini-columns prepared with 1 mL of Sephadex A25 and 2 M acetic acid (1:1, w/v) to have a 0.5 mL bed volume. Columns were washed with 6 M imidazole formate and with ultrapure water, and then 1 mL of the glucosinolate extract was added. The unbound material was removed washing with 0.1 M sodium acetate (pH 4); then 100 μ L sulfatase (EC 3.1.6.1) was loaded in the column, and desulphation was performed overnight (16 h) at room temperature. The desulphoglucosinolates were eluted with 1.5 mL of ultrapure water and stored at -20 °C before analysis. A total of 20 μ L of the extract was analyzed by HPLC (Shimadzu LC 10, Shimadzu, Japan) at a flow rate of 1 mL/min, using a Prodigy column (5 μ m ODS3 100A, 250 \times 4.60 mm; Phenomenex, Torrance, CA). Desulphoglucosinolates elution was achieved using the following linear gradient: starting condition, 2% B; at 5 min, 4% B; at 20 min, 20% B; at 30 min, 35% B; at 35 min, 40% B; at 45 min, 30% B; at 50 min, 10% B; and at 52 min, 2% B. The mobile phases were water (A) and methanol (B). Chromatograms were recorded at 227 nm. Sinigrin was used as an internal standard. The confirmation of the compound identity was achieved by HPLC MS-MS, as recently described (18).

Ascorbic Acid. Extraction (19) and analysis (20) of ascorbic acid were performed according to previously described methods.

TAC Determination. The TAC values were determined as previously described in Pellegrini et al. (12). Briefly, raw and cooked samples were homogenized under nitrogen flow in a high-speed blender (Brawn Multimix MX32). A weighed amount (~1 g) was extracted with 4 mL of water under agitation for 15 min at room temperature and centrifuged at 1000g for 10 min, and the supernatant was collected. The extraction was repeated with 2 mL of water, and the two supernatants were combined. The pulp residue was re-extracted by the addition of 4 mL of acetone under agitation for 15 min at room temperature and centrifuged at 1000g for 10 min, and the supernatant was collected. The extraction was repeated with 2 mL of acetone, and the two supernatants were combined. All food extracts were adequately diluted in the appropriate solvent (depending upon their activity) and immediately analyzed in triplicate for their antioxidant capacity by three different TAC assays: Trolox equivalent antioxidant capacity (TEAC) assay (21), total radical-trapping antioxidant parameter (TRAP) assay (22), and ferric reducing antioxidant power (FRAP) assay (23). The TEAC and TRAP values were expressed as millimoles of Trolox per 100 g of sample. FRAP values were expressed as millimoles of Fe^{2+} equivalents per 100 g of sample.

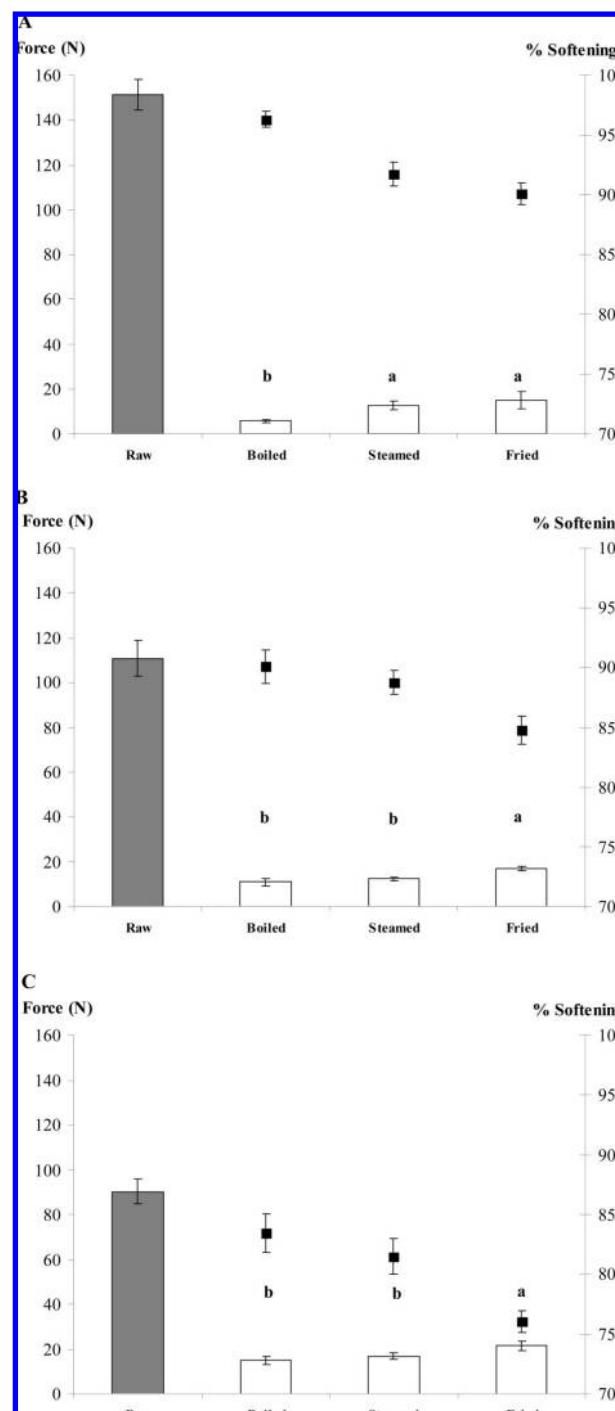


Figure 1. Shear force (histogram) and percent softening (■) of carrots (A), courgettes (B), and broccoli (C). Error bars represent ± 1 SD ($n = 15$). Bars of histograms (shear force) with the same letters are not significantly different ($p \leq 0.05$). Statistical significance of the raw sample was not considered.

Statistical Analysis. Means and standard deviations (SD) of data were calculated with SPSS (version 12.0, SPSS, Inc., Chicago, IL) statistical software. SPSS was used to perform one-way analysis of variance (ANOVA) and a least significant difference (LSD) test at a 95% confidence level ($p \leq 0.05$) to identify differences among groups.

RESULTS AND DISCUSSION

Carrots. Effect of Cooking on Physicochemical Characteristics. Figure 1A shows shear force values obtained for raw and cooked carrots. The degree of softening induced by the different cooking treatments as referred to the raw samples was also reported

Table 1. Color Determination Values (L^* , a^* , b^* , C , and Hue $^\circ$) of Raw and Cooked Vegetables Analyzed^a

| | raw | boiled | steamed | fried | raw | boiled | steamed | fried |
|--------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|
| carrots | | internal surface | | | | external surface | | |
| L^* | 60.1 \pm 1.2 a | 49.6 \pm 1.1 b | 50.5 \pm 1.5 b | 49.6 \pm 0.7 b | 56.8 \pm 1.5 a | 52.9 \pm 1.2 b | 50.5 \pm 0.9 c | 40.7 \pm 2.3 d |
| a^* | 31.8 \pm 0.9 a | 13.4 \pm 0.6 c | 15.7 \pm 1.4 b | 17.0 \pm 1.1 b | 30.7 \pm 2.0 a | 17.3 \pm 1.3 c | 20.2 \pm 0.9 b | 21.5 \pm 1.1 b |
| b^* | 41.9 \pm 2.5 a | 37.5 \pm 1.3 b | 37.7 \pm 2.4 b | 38.2 \pm 2.0 b | 37.2 \pm 2.6 a | 36.2 \pm 1.8 ab | 33.3 \pm 1.9 bc | 31.3 \pm 3.4 c |
| C | 35.6 \pm 1.0 b | 39.8 \pm 1.2 a | 40.8 \pm 2.3 a | 41.8 \pm 1.8 a | 48.3 \pm 3.3 a | 40.1 \pm 2.1 b | 39.0 \pm 1.9 b | 38.0 \pm 3.3 b |
| hue $^\circ$ | 67.6 \pm 1.7 ab | 70.3 \pm 1.1 a | 67.3 \pm 2.3 ab | 66.0 \pm 1.9 b | 50.4 \pm 0.6 c | 64.5 \pm 0.9 a | 58.8 \pm 1.3 b | 55.4 \pm 2.0 b |
| courgettes | | internal surface | | | | external surface | | |
| L^* | 85.5 \pm 1.1 a | 53.1 \pm 0.5 b | 54.0 \pm 0.8 b | 54.4 \pm 2.5 b | 36.8 \pm 1.8 a | 33.2 \pm 0.9 b | 33.8 \pm 1.4 b | 28.5 \pm 1.4 c |
| a^* | -5.8 \pm 0.7 a | -5.3 \pm 0.4 a | -5.8 \pm 0.5 a | -6.0 \pm 0.7 a | -5.8 \pm 0.5 c | -4.1 \pm 0.4 b | -2.2 \pm 0.4 a | -3.8 \pm 0.6 b |
| b^* | 31.3 \pm 1.6 a | 15.8 \pm 1.6 c | 18.1 \pm 1.5 bc | 18.8 \pm 2.2 b | 11.4 \pm 1.3 a | 7.3 \pm 0.8 c | 10.5 \pm 2.0 ab | 9.4 \pm 1.3 b |
| C | 31.9 \pm 1.7 a | 16.6 \pm 1.6 b | 19.0 \pm 1.6 b | 19.7 \pm 2.4 b | 12.8 \pm 1.4 a | 8.4 \pm 0.8 b | 10.8 \pm 2.1 ab | 10.2 \pm 1.3 ab |
| hue $^\circ$ | 100.4 \pm 0.9 b | 108.6 \pm 0.7 a | 107.7 \pm 0.8 a | 107.9 \pm 0.9 a | 117.1 \pm 1.4 a | 119.6 \pm 2.6 a | 102.1 \pm 1.2 c | 112.2 \pm 2.3 b |
| broccoli | | florets | | | | stems | | |
| L^* | 50.7 \pm 2.5 a | 36.6 \pm 0.9 c | 40.5 \pm 3.1 b | 33.5 \pm 1.0 d | 69.6 \pm 2.2 a | 54.4 \pm 2.5 b | 52.3 \pm 1.8 b | 53.3 \pm 2.9 b |
| a^* | -4.6 \pm 0.5 c | -7.4 \pm 1.5 d | -3.0 \pm 1.3 b | -0.4 \pm 0.3 a | -8.8 \pm 0.7 b | -9.0 \pm 1.2 b | -7.0 \pm 0.4 a | -6.6 \pm 2.2 a |
| b^* | 7.3 \pm 1.2 b | 11.9 \pm 2.6 a | 12.6 \pm 4.6 a | 2.8 \pm 1.5 c | 29.6 \pm 2.3 a | 23.7 \pm 3.6 c | 24.1 \pm 1.8 bc | 27.8 \pm 2.5 ab |
| C | 8.6 \pm 1.3 b | 14.0 \pm 2.9 a | 13.0 \pm 2.8 a | 2.9 \pm 1.5 c | 30.9 \pm 2.4 a | 26.8 \pm 2.1 ab | 25.1 \pm 1.7 b | 28.7 \pm 2.8 a |
| hue $^\circ$ | 122.3 \pm 3.2 a | 122.0 \pm 1.9 a | 103.3 \pm 2.2 b | 99.4 \pm 7.6 b | 106.6 \pm 0.8 b | 110.7 \pm 1.1 a | 106.3 \pm 1.2 b | 103.2 \pm 3.7 b |

^a Values are expressed in colorimetric units and presented as mean \pm SD ($n = 15$). Means in rows followed by different letters are significant ($p \leq 0.05$).

Table 2. Antioxidant Compounds of Raw and Cooked Carrots^a

| | raw | boiled | steamed | fried |
|-------------------------------------|-------------------|-------------------|-------------------|-------------------|
| carotenoids | | | | |
| lutein | 11.0 \pm 0.2 b | 12.2 \pm 0.1 a | 7.2 \pm 0.1 d | 6.3 \pm 0.1 d |
| α -carotene | 39.3 \pm 0.3 a | 33.0 \pm 0.2 b | 27.2 \pm 0.1 c | 26.1 \pm 0.0 d |
| β -carotene | 53.8 \pm 0.2 a | 54.6 \pm 0.2 a | 48.5 \pm 0.4 b | 41.0 \pm 1.0 c |
| cis-carotene | ND ^b | 17.7 \pm 0.1 | 12.2 \pm 0.1 | 11.6 \pm 0.2 |
| phytoene | 7.4 \pm 0.6 b | 8.3 \pm 0.1 ab | 8.0 \pm 0.4 b | 9.2 \pm 0.1 a |
| phytofluene | 6.4 \pm 0.0 d | 8.2 \pm 0.0 b | 7.5 \pm 0.0 c | 8.7 \pm 0.1 a |
| total carotenoids ^c | 117.9 \pm 0.4 b | 134.1 \pm 0.2 a | 110.6 \pm 0.4 c | 102.9 \pm 1.7 d |
| phenol compounds | | | | |
| chlorogenic acid | 44.0 \pm 1.5 a | ND ^b | 2.2 \pm 0.3 b | 3.1 \pm 0.0 b |
| caffeoic acid | 18.0 \pm 0.1 b | ND ^b | 29.9 \pm 3.2 a | 29.3 \pm 2.4 a |
| p -coumaric acid | 6.8 \pm 0.8 c | ND ^b | 7.6 \pm 0.3 b | 8.1 \pm 0.8 a |
| total phenol compounds ^c | 69.6 \pm 0.8 a | ND ^b | 39.6 \pm 3.2 c | 48.0 \pm 2.0 b |
| ascorbic acid | 30.8 \pm 1.5 a | 28.1 \pm 0.5 b | 19.2 \pm 0.3 c | ND ^b |

^a Values are presented as mean value \pm SD ($n = 3$) and expressed as mg/100 g of dry weight. Means in rows followed by different letters differed significantly ($p \leq 0.05$). ^b ND = not detected. ^c The total was obtained by summing each single replicate of each compound.

in the same graph. Raw carrots showed a shear force value of 151.4 \pm 6.8 N. Cooking of carrots caused a decrease in the force needed to shred the vegetable (**Figure 1A**), indicating a decrease of firmness and consequently softening of the vegetable for all of the three cooking methods. In particular, boiled carrots showed a significantly lower shear force value (higher degree of softening, $>96\%$) in comparison to both steamed and fried samples.

Color values are reported in **Table 1** for all of the vegetables considered. For carrots, both external and internal surfaces were considered. The color of the internal surface of raw carrots had a lightness of 60.1 \pm 1.2 (L^*), redness of 31.8 \pm 0.9 (a^*), and yellowness of 41.9 \pm 2.5 (b^*). L^* , a^* , and b^* values significantly decreased after all cooking treatments. The external color of the cooked samples was less bright (L^*), red (a^*), and yellow (b^*) than the raw sample color. A significant loss of vivid color (C decrease) with respect to raw carrots was observed for the external surface of all cooked samples, whereas a significant C increase was noticed internally. The hue angle significantly increased both for the external and internal surfaces of boiled samples in comparison to the raw product, resulting in a shift from red to orange. In the case of steamed and fried carrots, the hue angle also significantly increased but only on the external surfaces.

The color of carrots has been reported to be largely due to the presence of carotenes, which is in turn deeply affected by variety, maturity, and growing conditions (24). Thus, a comparison of color parameters obtained in this study for raw and

cooked carrots with data reported in the literature is difficult, because of the high variability of these vegetables.

The α - and β -carotene content was reported to influence the color of these vegetables (25). Although such carotenes are known to be relatively heat-stable (26), they isomerize into various *cis* isomers during cooking (15). The larger L^* , a^* , b^* , and C decrease observed in all cooked carrots may be related to the α - and β -carotene decrease and their isomerization (see data below), as already observed for heat-processed carrot juice (15). In addition, the hue angle increase could be related to the decrease of the carotene amount, as observed by Sulaiman and co-workers (27), who reported a high negative correlation between this color parameter and the carotene content of deep-fried carrots. The outer part of carrots was also reported to contain twice as much β -carotene as the inner part (24), and this may explain the remarkable loss of chroma (C) and the shift of the hue angle to yellow observed externally.

Effect of Cooking on the Phytochemical Profile and Antioxidant Capacities. The effects of cooking on carotenoids, polyphenols, and ascorbic acid are reported on a dry weight basis in **Table 2**, in comparison to the antioxidant concentrations measured in raw samples.

Raw carrots showed high concentrations of two vitamin A precursors, the carotenoids α - and β -carotene (4.6 and 6.4 mg/100 g of fresh weight corresponding to 39.3 and 53.8 mg/100 g on a dry weight basis).

Cooking had a small but significant effect on total carotenoids ($p \leq 0.05$): boiling determined a slight increase of 14% of their initial concentration, while the other two methods caused a slight but significant decrease, more evident in the case of frying (−13%). Among single carotenoids, lutein was slightly increased by boiling (+11%), whereas 34 and 43% of its initial concentration was lost during steaming and frying, respectively. α -Carotene decreased significantly after all cooking methods, even though its retention was lower after steaming and frying. β -Carotene was not significantly influenced by boiling ($p \geq 0.05$), but its concentration decreased slightly but significantly during steaming (−10%) and frying (−24%).

Better preservation of α - and β -carotene during boiling compared to steaming was also observed by Pinheiro-Sant'Ana et al. (28), who concluded that water boiling was the method that determines the greatest stability of these two compounds in carrots compared to water pressure cooking and steaming at 115–120 °C. The authors explained their findings by stating that temperature, instead of the presence of water, was the major factor influencing the carotenoids stability (28). In the present study, the temperatures during steaming and boiling were the same (100 °C), but steaming of carrots required a longer time in comparison to boiling to reach the appropriate tenderness. Thus, the prolonged exposure to oxygen and light may explain the lower carotenoid recovery observed after steaming than a temperature effect. In the case of frying, major losses may be explained by the lipophilic nature of carotenoids and their instability in the high temperatures reached in this process (i.e., 170 °C).

During all treatments, similar amounts of carotene *cis* isomer were formed. The *trans*–*cis*- isomerization of β -carotene has been well-documented during carrot processing involving thermal treatments, whereas that of α -carotene has been less investigated (15).

Phytoene and phytofluene concentrations increased after all three cooking treatments, especially in the case of frying. These two molecules are carotenoid precursors located inside the plant plastids, and their concentration increase may be the result of the release from plastids because of matrix softening during heat processing (29).

The predominant phenolic acids of raw carrots were chlorogenic acid, followed by caffeic and *p*-coumaric acids (Table 2).

Boiling had the most detrimental effect on carrot polyphenols, resulting in a complete loss of each compound likely because of their diffusion in the boiling water. Steaming and frying had a less negative effect on total phenolics (−43 and −31%, respectively), exclusively because of the loss of chlorogenic acid (−95 and −93% for the two processes, respectively). Phenolic acids are dissolved in vacuoles and apoplast (29). Cooking of vegetables determines softening and breaking of cellular components with the consequent release of these molecules into the boiling water. The higher softening observed for boiled carrots (Figure 1A) well explains the complete loss of polyphenols in comparison to steamed and fried samples.

During steaming and frying, moreover, the hydrolysis of chlorogenic acid into caffeic and quinic acids may also be occurred, justifying the significant increases of caffeic acid observed for both cooking methods (59 and 56%, respectively). Moreover, polyphenol losses could also be due to the covalent binding between oxidized phenols and proteins or amino acids as well as the polymerization of oxidized phenols (30). These losses could have also in turn affected the color of carrots, especially on the external surface, contributing to the lower a^*

Table 3. TEAC, TRAP, and FRAP Values of Raw and Cooked Vegetables Analyzed^a

| | TEAC (mmol of Trolox/100 g) | FRAP (mmol of Fe ²⁺ /100 g) | TRAP (mmol of Trolox/100 g) |
|------------|--------------------------------|---|--------------------------------|
| carrots | | | |
| raw | 0.40 ± 0.01 d | 0.68 ± 0.03 d | 0.03 ± 0.00 c |
| boiled | 0.83 ± 0.01 b (108%) | 1.45 ± 0.02 b (114%) | 0.20 ± 0.00 b (658%) |
| steamed | 0.70 ± 0.02 c (77%) | 1.23 ± 0.03 c (81%) | 0.04 ± 0.00 c (61%) |
| fried | 1.05 ± 0.02 a (165%) | 3.25 ± 0.05 a (379%) | 0.56 ± 0.02 a (1956%) |
| courgettes | | | |
| raw | 0.80 ± 0.00 c | 2.79 ± 0.11 c | 0.20 ± 0.02 d |
| boiled | 1.53 ± 0.11 ab (92%) | 6.32 ± 0.86 b (127%) | 0.29 ± 0.03 c (45%) |
| steamed | 1.40 ± 0.04 b (75%) | 5.92 ± 0.08 b (112%) | 0.36 ± 0.01 b (84%) |
| fried | 1.64 ± 0.04 a (106%) | 7.97 ± 0.12 a (186%) | 0.75 ± 0.01 a (277%) |
| broccoli | | | |
| raw | 1.10 ± 0.05 d | 5.23 ± 0.56 c | 1.61 ± 0.04 c |
| boiled | 2.17 ± 0.17 c (98%) | 8.91 ± 0.57 b (70%) | 1.95 ± 0.09 c (21%) |
| steamed | 3.51 ± 0.15 a (221%) | 11.98 ± 0.74 a (129%) | 3.59 ± 0.21 a (123%) |
| fried | 2.88 ± 0.32 b (163%) | 9.02 ± 1.68 b (73%) | 2.41 ± 0.26 b (49%) |

^a The percent variation because of cooking is given in parentheses. Values are presented as mean value ± SD ($n = 3$) and referred to the dry weight. Means in columns followed by different letters differed significantly ($p \leq 0.05$).

and higher hue angle values observed for boiled carrots in comparison to steamed and fried products.

Raw carrots had low value of ascorbic acid (2.3 and 30.8 mg/100 g for fresh and dry matter, respectively) in comparison to other vegetables (11). The ascorbic acid concentration was slightly but significantly ($p \leq 0.05$) affected by boiling (−9%) and steaming (−38%), whereas it was not detectable in fried carrots. The loss of ascorbic acid can probably be ascribed to water leaching and its thermal degradation, as already reported (4).

The TAC values of raw carrots (Table 3) are in agreement with the literature data (12). All cooking methods significantly increased carrot TAC, except in the case of steamed carrots measured by the TRAP assay. Frying determined the highest TAC increases, followed by boiling and steaming (Table 3). These results are in agreement with Mayer-Miebach et al. (31), who reported a significant increase of carrot TAC during a thermal treatment at 130 °C for 20 min.

Looking at single antioxidant compounds, boiling determined the highest ascorbic acid and carotenoid retention, a complete loss of polyphenols, and the highest formation of carotene *cis* isomers. Among these isomers, those of α -carotene have been reported to have higher antioxidant capacity compared to *trans* counterparts (32). The enhancement of carotene availability and their transformation into more active compounds could be both responsible for the TAC increase. On the other hand, steaming induced major losses of ascorbic acid and carotenoids compared to boiling but induced a higher retention of phenolic acids. Frying determined similar carotenoid and polyphenol retentions to steaming but caused a complete loss of ascorbic acid. In this case, the TAC increment is probably also due to the formation of new molecules with high antioxidant capacities, such as Maillard reaction products, because oil absorption and its contribution to TAC were both negligible (data not shown).

Courgettes. *Effect of Cooking on Physicochemical Characteristics.* Shear force values were also obtained on raw and cooked courgettes (Figure 1B). The shear force of the raw samples was 110.9 ± 7.8 N. Cooking induced significantly higher softening (~90%) in boiled and steamed products (~88%) in comparison to frying (~84%). However, softening of courgettes was lower than that of carrots. Color determinations were carried out on both external and internal surfaces, as was done for carrots. The internal surface of cooked courgettes showed significantly lower L^* and b^* values, while

Table 4. Antioxidant Compounds of Raw and Cooked Courgettes^a

| | raw | boiled | steamed | fried |
|-------------------------------------|-----------------|-----------------|-----------------|-----------------|
| carotenoids | | | | |
| lutein | 45.4 ± 0.3 a | 40.6 ± 1.8 b | 30.5 ± 0.5 c | 25.8 ± 0.0 d |
| β-carotene | 4.8 ± 0.1 b | 5.7 ± 0.1 a | 5.8 ± 0.0 a | 4.3 ± 0.0 c |
| phytoene | ND ^b | 1.1 ± 0.0 | 1.4 ± 0.1 | 1.1 ± 0.0 |
| phytofluene | ND ^b | 1.2 ± 0.1 | 1.5 ± 0.0 | 1.4 ± 0.5 |
| total carotenoids ^c | 50.1 ± 0.2 a | 48.5 ± 1.8 a | 39.2 ± 0.5 b | 32.6 ± 0.4 c |
| phenol compounds | | | | |
| chlorogenic acid | 9.6 ± 0.3 | ND ^b | ND ^b | ND ^b |
| caffeic acid | 39.9 ± 1.3 a | 12.7 ± 0.3 d | 27.3 ± 2.3 b | 17.7 ± 2.1 c |
| p-coumaric acid | 9.6 ± 0.1 a | 3.2 ± 0.4 c | 8.0 ± 0.8 b | 4.2 ± 0.6 c |
| total phenol compounds ^c | 59.0 ± 1.1 a | 17.9 ± 1.0 d | 35.3 ± 1.5 b | 21.9 ± 1.5 c |
| ascorbic acid | 193.8 ± 9.5 a | 186.5 ± 8.6 a | 165.6 ± 6.3 b | 167.0 ± 6.2 b |

^a Values are presented as mean value ± SD ($n = 3$) and expressed as mg/100 g of dry weight. Means in rows followed by different letters differed significantly ($p \leq 0.05$). ^b ND = not detected. ^c The total was obtained by summing each single replicate of each compound.

$-a^*$ (greenness) did not significantly vary in comparison to raw samples. The external color of the cooked samples also showed significant changes. Frying induced the highest L^* decrement in comparison to raw samples, while a^* and b^* were more influenced by the other two cooking methods. In particular, steamed courgettes became less green (higher $-a^*$) and boiled samples became less yellow (lower b^*) than the other two treatments, respectively. Chroma values significantly decreased both externally and internally. The hue angle significantly decreased on the external surface only for steamed and fried products, resulting in a shift from green to yellow. On the contrary, this color parameter was found to increase internally.

Changes in the visual color observed on the skin of courgettes [loss of chroma (C) and greenness ($-a^*$)] could be mainly related to the conversion of chlorophyll into pheophytin because of heat treatment, as commonly referred for green vegetables (9). However, the shift of the hue angle to yellow was more pronounced for both steamed and fried courgettes than for boiled ones. A more consistent color retention of boiled green vegetables has also been attributed by several authors not only to a different pattern of chlorophyll conversion but also to a change in surface reflectance and depth of light penetration into tissues of boiled vegetables, caused by the loss of air and other dissolved gases by cells and their replacement by cooking water and cell juices (33).

Effect of Cooking on the Phytochemical Profile and Antioxidant Capacities. The content of carotenoids, polyphenols, and ascorbic acid of raw and cooked courgettes are reported on a dry weight basis in **Table 4**.

To our knowledge, no comprehensive report exists on phytochemical contents of courgettes. In this study, low concentrations of both carotenoids, mostly lutein (2.7 mg/100 g of fresh weight corresponding to 45.4 mg/100 g of dry matter), and polyphenols (3.5 mg/100 g of fresh weight corresponding to 59 mg/100 g of dry weight) and a high concentration of ascorbic acid (11.6 mg/100 g of fresh weight and 193.8 mg/100 g of dry weight) were measured in raw courgettes. Small quantities of β-carotene (0.3 mg/100 g of fresh weight and 4.8 mg/100 g in terms of dry weight) were observed. Among phenol compounds, caffèic acid had the highest concentration (2.4 mg/100 g of fresh weight and 39.9 mg/100 g on a dry weight basis), whereas similar quantities of chlorogenic and *p*-coumaric acids (0.6 mg/100 g of fresh weight corresponding to 9.6 mg/100 g on a dry weight basis) were detected.

Boiling did not affect the total carotenoid concentration ($p \geq 0.05$), while steaming and frying resulted in significant losses (22 and 35%, respectively). As already stated for carrots, losses

of carotenoids could be explained by a longer exposure of steamed samples to light and oxygen and by a higher temperature reached during frying, in comparison to boiling.

Boiling and steaming determined an increase of β-carotene but resulted in significant losses ($p \leq 0.05$) of lutein (11% for boiling and 33% for steaming). In addition, both β-carotene and lutein were negatively affected by frying, but minor β-carotene losses (only 10%) were observed, suggesting a different thermal stability of these compounds. This finding is in agreement with Aman et al. (34), who studied the thermal stability of chloroplast-bound carotenoids, demonstrating in this system a higher stability of β-carotene than lutein.

The loss of lutein could have also influenced color change (e.g., decrease of L^* and b^* and increase of the hue angle) observed not only on the external green skin but also in the yellowish-white internal portion of cooked courgettes.

In general, greater losses of courgette polyphenols were recorded after boiling and frying than after steaming (**Table 4**). As already observed for carrots, chlorogenic acid was completely lost during all cooking processes, but in the case of courgettes, no caffèic acid increments were recorded. This seems to indicate that losses of phenolic acids occur more quickly in cooked courgettes than in cooked carrots.

Ascorbic acid was not dramatically affected by cooking processes. Steaming and frying had a similar effect on the ascorbic acid concentration, determining losses of about 15%, whereas no significant losses were recorded after boiling ($p \geq 0.05$). As already noticed for carrots, boiling had a surprisingly lower detrimental effect on the ascorbic acid concentration than other cooking methods. It is well-known that heat induces a significant loss of ascorbic acid (29), but this loss was also found to be time-dependent in vegetables (35). Thus, the lower time of boiling in comparison to steaming could explain the lower losses of ascorbic acid content observed in carrots and courgettes.

TAC values for courgettes are showed in **Table 3**. Frying determined the highest TAC increases, as already observed for carrots, whereas boiling and steaming determined minor increases. During frying, the increases of TAC values were probably due to the formation of Maillard reaction products having antioxidant activities (3). This formation should also explain the color changes (e.g., decrease of L^* and C) of both external and internal portions of the fried courgettes (**Table 1**). Moreover, it has been suggested that processing can promote the oxidation of polyphenols to an intermediate oxidation state, which can exhibit a higher radical scavenging efficiency than the nonoxidized ones (3).

Table 5. Antioxidant Compounds of Raw and Cooked Broccoli^a

| | raw | boiled | steamed | fried |
|---------------------------------------|---------------|----------------|---------------|-----------------|
| carotenoids | | | | |
| lutein | 16.9 ± 0.2 c | 22.2 ± 0.3 a | 18.1 ± 0.5 b | 4.8 ± 0.1 d |
| β-carotene | 5.7 ± 0.2 b | 6.6 ± 0.2 a | 6.7 ± 0.0 a | 2.0 ± 0.1 c |
| phytoene | 1.3 ± 0.0 b | 2.9 ± 0.1 a | 3.1 ± 0.0 a | ND ^b |
| phytofluene | 1.1 ± 0.1 c | 2.6 ± 0.0 b | 3.6 ± 0.2 a | 2.6 ± 0.0 b |
| β-cryptoxanthin | 3.2 ± 0.0 a | 2.6 ± 0.1 b | 2.3 ± 0.2 b | ND ^b |
| total carotenoids ^c | 28.2 ± 0.3 c | 37.1 ± 0.2 a | 33.6 ± 0.4 b | 9.3 ± 0.1 d |
| phenolic compounds | | | | |
| chlorogenic acid | 35.7 ± 1.5 a | 12.6 ± 2.0 b | 14.8 ± 2.4 b | 9.8 ± 0.7 b |
| caffeic acid | 2.8 ± 0.6 a | 2.9 ± 0.9 a | 2.8 ± 0.8 a | 3.1 ± 0.4 a |
| sinapic acid | 9.4 ± 0.8 a | 4.3 ± 0.2 c | 5.9 ± 0.4 b | 6.1 ± 0.6 b |
| ferulic acid | 8.9 ± 1.0 a | 1.4 ± 0.0 c | 2.7 ± 0.5 b | 2.8 ± 1.5 b |
| quercetin | 18.9 ± 1.8 a | 2.2 ± 0.2 d | 11.7 ± 1.3 b | 5.3 ± 0.4 c |
| kaempferol | 24.1 ± 1.6 a | 3.7 ± 0.1 c | 23.9 ± 2.8 a | 13.3 ± 0.2 b |
| total phenolic compounds ^c | 99.8 ± 2.4 a | 27.0 ± 3.4 d | 61.8 ± 8.2 b | 40.3 ± 0.7 c |
| ascorbic acid | 847.0 ± 4.7 a | 437.6 ± 14.5 b | 575.5 ± 7.3 b | 113.3 ± 1.3 c |

^a Values are presented as mean value ± SD ($n = 3$) and expressed as mg/100 g of dry weight. Means in rows followed by different letters differed significantly ($p \leq 0.05$). ^b ND = not detected. ^c The total was obtained by summing each single replicate of each compound.

Broccoli. Effect of Cooking on Physicochemical Characteristics. The shear force value of raw broccoli was 90.2 ± 5.6 N (**Figure 1C**). All cooking treatments induced softening, even though lower than that was observed for other vegetables, more significantly in boiling (~84%) and steaming (~82%) than in frying (~76%).

Color measurements were obtained both for florets and stems, as presented in **Table 1**. L^* values significantly decreased for florets in all cooking treatments. Steamed and fried florets became less green ($-a^*$ increase). On the contrary, boiled florets showed a significant increase in greenness ($-a^*$ decrease). b^* and C values significantly increased for both boiled and steamed florets, while a significant decrease was observed for frying in comparison to the uncooked product. The hue angle significantly shifted toward yellow for steamed and fried florets. The stem color was characterized by a decrease of L^* and b^* values. Greenness ($-a^*$) increased for boiled stems, although not significantly, in comparison to raw broccoli. On the contrary, stems of steamed and fried products were significantly less green than raw ones. C significantly decreased only for steamed stems, whereas the hue angle did not significantly change for boiled stem in comparison to the raw product as well as for florets. The color of florets and stems of raw and cooked broccoli was comparable to those obtained in a previous study (36), although broccoli was treated differently prior to cooking.

It is noteworthy that boiled broccoli retained a green color both for florets and stems. The green color intensity of raw and cooked vegetables was reported to be related not only to the pigment concentration but also to light scattering and reflectance of green surfaces (37), as reported above for courgettes. An increase of the green color intensity because of a change in surface-reflecting properties, as air between cells was removed and expelled, was reported for broccoli after the first stage of blanching (33) or short time treatment (i.e., microwave cooking) in comparison to boiling and steaming (9). After prolong heating, chlorophyll degradation was reported to cause a decrease in greenness (33). Under the same temperature of treatment (100 °C), the shorter time of boiling in comparison to steaming (data not shown) could have partially prevented chlorophyll degradation, inducing only changes in surface-reflecting properties.

Effect of Cooking on the Phytochemical Profile and Antioxidant Capacities. The effects of cooking on carotenoids, polyphenols, and ascorbic acids of broccoli are reported on a dry weight basis in **Table 5**, in comparison to raw samples. Raw broccoli showed intermediate concentrations of carotenoids, mostly lutein

(2.1 mg/100 g of fresh weight and 16.9 mg/100 g on a dry weight basis), and both higher concentrations of polyphenols, mostly chlorogenic acid (4.5 mg/100 g of fresh weight and 35.7 mg/100 g on a dry weight basis), and ascorbic acid (106 mg/100 g of fresh weight corresponding to 847 mg/100 g in terms of dry matter) in comparison to other vegetables analyzed. The flavonols quercetin and kaempferol (2.4 and 3.0 mg/100 g of fresh weight, respectively, corresponding to 18.9 and 24.1 mg/100 g of dry weight) were found. These data are in agreement with those recently published in a review on *Brassica* vegetable antioxidants (7).

Cooking of green fresh vegetables has been reported to promote the release of carotenoids from the matrix because of the disruption of carotenoid–protein complexes, leading to better extractability and higher concentrations in cooked samples (6). Accordingly, in the present study, the contents of all carotenoid compounds significantly increased in boiled and steamed broccoli (+32 and 19%, respectively) in comparison to raw ones. The concentration of phytoene and phytofluene almost tripled (**Table 5**). The release of carotenoids, mainly lutein, from the cells could partially have contributed to the significant increase of the b^* value observed for boiled and steamed florets (**Table 1**) in addition to other phenomena described above. Conversely, frying determined a 67% loss of the initial carotenoid concentration, probably because of leaching into oil and to a higher processing temperature. In this case, phytoene was completely lost, whereas phytofluene exhibited a similar behavior as in boiling and steaming (**Table 5**).

As observed for carrots and courgettes, boiling and frying determined a higher loss of total phenolics than steaming. Among phenolic acids, caffeoic acid was not affected by the cooking process ($p \geq 0.05$), probably because of the hydrolysis of chlorogenic acid that determines a new formation of this compound. The decrease of other phenolic acids could be ascribed to their autoxidation that, in turn, determined the polymerization and/or browning reaction (30). These reactions have probably affected the color of fried broccoli, mainly florets (L^* decrease and a^* increase) (**Table 1**).

As far as flavonols, quercetin and kaempferol were in general better preserved by steaming than by boiling and frying, in agreement with Vallejo et al. (38).

The ascorbic acid concentration significantly decreased after all cooking treatments, especially after frying, where a loss of 87% was detected. In steamed broccoli, a reduction of 32% of ascorbic acid with respect to the uncooked sample was observed,

Table 6. Glucosinolate Compounds of Raw and Cooked Broccoli^a

| | raw | boiled | steamed | fried |
|-----------------------------------|--------------|--------------|--------------|--------------|
| glucoraphanin | 1.9 ± 0.4 a | 2.4 ± 0.1 a | 1.7 ± 0.4 a | 0.6 ± 0.0 b |
| glucobrassicin | 10.0 ± 0.6 b | 8.8 ± 0.8 b | 18.0 ± 0.6 a | 3.0 ± 0.0 c |
| 4-methoxy-glucobrassicin | 1.8 ± 0.0 b | 1.6 ± 0.1 b | 2.7 ± 0.2 a | 0.4 ± 0.0 c |
| 1-methoxy-glucobrassicin | 57.7 ± 0.1 b | 16.5 ± 2.8 c | 71.0 ± 8.7 a | 7.4 ± 0.4 c |
| total glucosinolates ^b | 71.4 ± 0.0 b | 29.3 ± 3.6 c | 93.4 ± 9.5 a | 11.4 ± 0.4 d |

^a Values are presented as mean value ± SD ($n = 3$) and expressed as $\mu\text{mol/g}$ of dry weight. Means in rows followed by different letters differed significantly ($p \leq 0.05$). ^b The total was obtained by summing each single replicate of each compound.

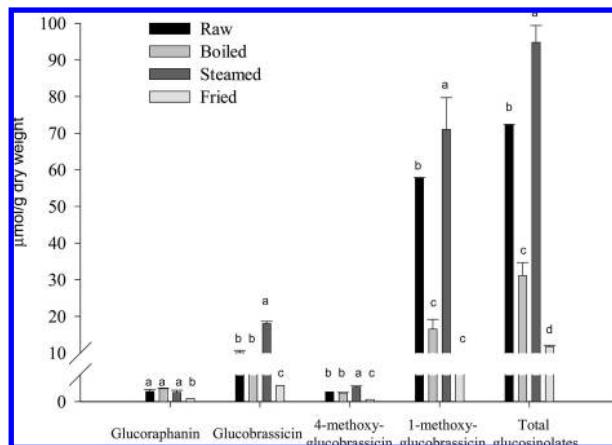


Figure 2. Concentration of the main glucosinolate compounds in broccoli cooked by steaming, boiling, and frying. Values are expressed as $\mu\text{mol/g}$ of dry weight.

in contrast to Glyszczyńska-Świglo et al. (10), who observed no change of the vitamin C content (both ascorbic and dehydroascorbic acids) during steaming.

The TAC values of raw broccoli (Table 3) were lower than those of a previous study (12). This is likely related to the high variability of this parameter observed in the *Brassica* family (5). Steaming determined the higher TAC increase probably because of the increase of carotenoids and the major retention of polyphenols and ascorbic acid with respect to the other two methods. These findings are in agreement with Glyszczyńska-Świglo et al. (10), who observed an increase of TEAC values of vitamin C, polyphenol, and carotenoid extracts in steamed broccoli.

Beside antioxidant compounds, vegetables belonging to the *Brassica* family, including broccoli, are characterized by the presence of glucosinolates, compounds that together with their breakdown products are considered biologically active and have cancer-protective effects (39). Thus, the glucosinolate content of raw and cooked broccoli was also determined and is presented in Table 6 and Figure 2. In agreement with data by Vallejo et al. (38), the predominant glucosinolates of raw broccoli were neoglucobrassicin (1-methoxy-glucobrassicin), which was present in a very high concentration, and glucobrassicin, followed by glucoraphanin, the only aliphatic compounds identified.

Data on the cooking effect showed that total glucosinolate concentrations were significantly modified by the cooking treatment (Figure 2). Steaming was the only cooking method that completely preserved glucosinolates and even significantly increased by 30% of their initial concentration ($p \leq 0.05$). On the contrary, boiling and frying caused a substantial degradation of these molecules, especially evident in the case of frying that determined a total loss of 84%.

Glucosinolates are water-soluble compounds and are usually lost during conventional cooking because of leaching into surrounding water (10). Moreover, degradation events at higher temperature, such as during frying, may also occur, leading to the formation of volatile compounds that were not determined in this study. On the contrary, steaming was reported to well-preserve (40) or increase (10) broccoli glucosinolates. The glucosinolate increase could be partially explained not only by the inactivation of myrosinase, as suggested by Vallejo et al. (40), but also by a disintegration of plant tissue upon heat because part of these molecules are bound to the cell walls and released only after a disintegration of cell structures (10).

Among single glucosinolates, glucoraphanin was the compound for which a lower decrease during frying was observed, confirming the higher thermostability of aliphatic glucosinolates than indolyl ones (40). The enzymatic hydrolysis of glucoraphanin produces the isothiocyanate sulforaphane that seems to be involved in the reduced incidence of a number of tumors in both *in vitro* and *in vivo* studies (10, 39). Thus, the retention of glucoraphanin during processing may be of great biological interest, because its breakdown into bioactive compounds has been reported to occur also within the intestinal tract by microflora (39).

In conclusion, the present study clearly indicates that physicochemical and nutritional qualities of vegetables are deeply modified by domestic cooking and that modifications of the evaluated parameters are also strongly dependent upon the vegetable species. However, the cooking conditions used here, chosen to reproduce the common Italian cooking practices, were less severe than thermal conditions usually applied in previous studies. These conditions would have promoted the release of antioxidant compounds from the vegetable matrix and determined the formation of new antioxidant compounds. Moreover, it is also likely that matrix softening and increased extractability upon cooking were accompanied by the conversion of polyphenol into very active chemical species, which were not yet identified and concurred synergistically to determine the high antioxidant capacity.

The overall increase of TAC values observed in the present study is in partial disagreement with the concept that processed vegetables have lower nutritional quality than the raw ones. Moreover, our results suggest that for each vegetable a preferential cooking method could be selected to preserve or improve its nutritional and physicochemical qualities. This selection may help consumers on the choice of cooking practices to improve the nutritional quality of foods, as well as their acceptability.

ACKNOWLEDGMENT

We thank Ms. Sara Manferdelli and Ms. Doralice Grossi for the excellent technical assistance.

LITERATURE CITED

- Link, L. B.; Potter, J. D. Raw versus cooked vegetables and cancer risk. *Cancer Epidemiol. Biomarkers Prev.* **2004**, *13*, 1422–1435.
- Kaur, C.; Kapoor, H. C. Antioxidants in fruits and vegetables—The millennium's health. *Int. J. Food Sci. Technol.* **2001**, *36*, 703–725.
- Nicoli, M. C.; Anese, M.; Parpinel, M. Influence of processing on the antioxidant properties of fruit and vegetables. *Trends Food Sci. Technol.* **1999**, *10*, 94–100.
- Lee, S. K.; Kader, A. A. Preharvest and postharvest factors influencing vitamin C content of horticultural crops. *Postharvest Biol. Technol.* **2000**, *20*, 207–220.

(5) Ou, B.; Huang, D.; Hampsch-Woodill, M.; Flanagan, J. A.; Deemer, E. K. Analysis of antioxidant activities of common vegetables employing oxygen radical absorbance capacity ORAC and ferric reducing antioxidant power FRAP assays. A comparative study. *J. Agric. Food Chem.* **2002**, *50*, 3122–3128.

(6) Bernhardt, S.; Schlich, E. Impact of different cooking methods on food quality: Retention of lipophilic vitamins in fresh and frozen vegetables. *J. Food Eng.* **2006**, *77*, 327–333.

(7) Podsedek, A. Natural antioxidants and antioxidant capacity of *Brassica* vegetables: A review. *Lebensm.-Wiss. Technol.* **2007**, *40*, 1–11.

(8) Waldron, K. W.; Smith, A. C.; Parr, A. J.; Ng, A.; Parker, M. L. New approaches to understanding and controlling cell separation in relation to fruit and vegetable texture. *Trends Food Sci. Technol.* **1997**, *8*, 213–221.

(9) Turkmen, N.; Poyrazoglu, E. S.; Sari, F.; Velioglu, Y. S. Effects of cooking methods on chlorophylls, pheophytins and colour of selected green vegetables. *Int. J. Food Sci. Technol.* **2006**, *41*, 281–288.

(10) Glyszcynska-Świglo, A.; Ciska, E.; Pawlak-Lemanska, K.; Chmielewski, J.; Borkowski, T.; Tyrakowska, B. Changes in the content of health promoting compounds and antioxidant activity of broccoli after domestic processing. *Food Addit. Contam.* **2006**, *23*, 1088–1098.

(11) Salvini, S. A food composition database for epidemiological studies in Italy. *Cancer Lett.* **1997**, *114*, 299–300.

(12) Pellegrini, N.; Serafini, M.; Colombi, B.; Del Rio, D.; Salvatore, S.; Bianchi, M.; Brighenti, F. Total antioxidant capacity of plant foods, beverages, and oils consumed in Italy assessed by three different *in vitro* assays. *J. Nutr.* **2003**, *133*, 2812–2819.

(13) Recommendations on uniform colourspaces-colour equations, psychometric colour terms. In CIE, Commission Internationale de l'éclairage15, Suppl. number 2; CIE Publisher, E-1.3.L 1971/9TC-1-3, CIE, Paris, France, 1978.

(14) Leonardi, C.; Ambrosino, P.; Esposito, F.; Fogliano, V. Antioxidant activity and carotenoid and tomatine contents in different typologies of fresh consumption tomatoes. *J. Agric. Food Chem.* **2000**, *48*, 4723–4727.

(15) Chen, B. H.; Peng, H. Y.; Chen, H. E. Changes of carotenoids, color and vitamin A contents during processing of carrot juice. *J. Agric. Food Chem.* **1995**, *43*, 1912–1918.

(16) Crozier, A.; Lean, M. E. J.; McDonald, M. S.; Black, C. Quantitative analysis of the flavonoid content of commercial tomatoes, onions, lettuce and celery. *J. Agric. Food Chem.* **1997**, *45*, 590–595.

(17) Kiddle, G.; Bennett, R. N.; Botting, N. P.; Davidson, N. E.; Robertson, A. A. B.; Wallsgrove, R. M. High-performance liquid chromatographic separation of natural and synthetic desulphoglucosinolates and their chemical validation by UV, NMR and chemical ionisation-MS methods. *Phytochem. Anal.* **2001**, *12*, 226–242.

(18) Barbieri, G.; Pernice, R.; Maggio, A.; De Pascale, S.; Fogliano, V. Glucosinolates profile of *Brassica rapa* L. subsp *Sylvestris* L. Janch. var. *esculenta* Hort. *Food Chem.* 2007, in press, doi: 10.1016/j.foodchem.2007.09.054.

(19) Dürust, N.; Sumengen, D.; Durust, Y. Ascorbic acid and element contents of foods of Trabzon Turkey. *J. Agric. Food Chem.* **1997**, *45*, 2085–2087.

(20) Gokmen, V.; Kahraman, N.; Demir, N.; Acar, J. Enzymatically validated liquid chromatographic method for the determination of ascorbic and dehydroascorbic acids in fruit and vegetables. *J. Chromatogr. A* **2000**, *881*, 309–316.

(21) Pellegrini, N.; Del Rio, D.; Colombi, B.; Bianchi, M.; Brighenti, F. Application of the 2,2'-azobis 3-ethylenbenzothiazoline-6-sulfonic acid radical cation assay to a flow injection system for the evaluation of antioxidant activity of some pure compounds and beverages. *J. Agric. Food Chem.* **2003**, *51*, 260–264.

(22) Ghiselli, A.; Serafini, M.; Maiani, G.; Azzini, E.; Ferro-Luzzi, A. A fluorescence-based method for measuring total plasma antioxidant capability. *Free Radical Biol. Med.* **1995**, *18*, 29–36.

(23) Benzie, I. F. F.; Strain, J. J. Ferric reducing antioxidant power assay, direct measure of total antioxidant activity of biological fluids and modified version for simultaneous measurement of total antioxidant power and ascorbic acid concentration. *Methods Enzymol.* **1999**, *299*, 15–27.

(24) Hart, D. J.; Scott, K. J. Development and evaluation of an HPLC method for the analysis of carotenoids in foods, and the measurement of the carotenoid content of vegetables and fruits commonly consumed in the UK. *Food Chem.* **1995**, *54*, 101–111.

(25) Bao, B.; Chang, K. C. Carrot juice color, carotenoids, and nonstarchy polysaccharides as affected by processing conditions. *J. Food Sci.* **1994**, *59*, 1155–1158.

(26) Khachik, F.; Beecher, G. R.; Goli, M. B. Separation, identification, and quantification of carotenoids in fruits, vegetables and human plasma by high performance liquid chromatography. *Pure Appl. Chem.* **1991**, *6*, 71–80.

(27) Sulaeman, A.; Giraud, D. W.; Keeler, L.; Taylor, S. L.; Driskell, J. A. Effect of moisture content of carrot slices on the fat content, carotenoid content and sensory characteristics of deep-fried carrot chips. *J. Food Sci.* **2004**, *69*, 450–455.

(28) Pinheiro San'Ana, H. M.; Stringheta, P. C.; Cardoso Brandão, S. C.; Cordeiro de Azeredo, R. M. Carotenoid retention and vitamin A value in carrot *Daucus carota* L. prepared by food service. *Food Chem.* **1998**, *61*, 145–151.

(29) Kalt, W. Effects of production and processing factors on major fruit and vegetable antioxidants. *J. Food Sci.* **2005**, *70*, R11–R19.

(30) Friedman, M. Food browning and its prevention: An overview. *J. Agric. Food Chem.* **1996**, *44*, 631–653.

(31) Mayer-Miebach, E.; Behsnilian, D.; Regier, M.; Schuchmann, H. P. Thermal processing of carrots: Lycopene stability and isomerisation with regard to antioxidant potential. *Food Res. Int.* **2005**, *38*, 1103–1108.

(32) Böhm, V.; Puspitasari-Nienaber, N. L.; Ferruzzi, M. G.; Schwartz, S. J. Trolox equivalent antioxidant capacity of different geometrical isomers of α -carotene, β -carotene, lycopene, and zeaxanthin. *J. Agric. Food Chem.* **2002**, *50*, 221–226.

(33) Tijskens, L. M. M.; Schijvens, E. P. H. M.; Biekman, E. S. A. Modelling the change in colour of broccoli and green beans during blanching. *Innov. Food Sci. Emerg. Technol.* **2001**, *7*, 303–313.

(34) Aman, R.; Schieber, A.; Carle, R. Effects of heating and illumination on trans–cis isomerization and degradation of β -carotene and lutein in isolated spinach chloroplast. *J. Agric. Food Chem.* **2005**, *53*, 9512–9518.

(35) Jiratana, T.; Liu, R. H. Antioxidant activity of processed table beets (*Beta vulgaris* var. *conditiva*) and green beans (*Phaseolus vulgaris* L.). *J. Agric. Food Chem.* **2004**, *52*, 2659–2670.

(36) Brewer, M. S.; Begum, S.; Bozeman, A. Microwave and conventional blanching effects on chemical, sensory, and colour characteristics of frozen broccoli. *J. Food Qual.* **1995**, *18*, 479–493.

(37) Ramesh, M. N. The performance evaluation of a continuous vegetable cooker. *Int. J. Food Sci. Technol.* **2000**, *35*, 337–384.

(38) Vallejo, F.; Tomas-Barberan, F. A.; Garcia-Viguera, C. Potential bioactive compounds in health promotion from broccoli cultivars grown in Spain. *J. Sci. Food Agric.* **2003**, *82*, 1293–1297.

(39) Holst, B.; Williamson, G. A critical review of the bioavailability of glucosinolates and related compounds. *Nat. Prod. Rep.* **2004**, *21*, 425–447.

(40) Vallejo, F.; Tomas-Barberan, F. A.; Garcia-Viguera, C. Glucosinolates and vitamin C content in edible parts of broccoli florets after domestic cooking. *Eur. Food Res. Technol.* **2002**, *215*, 310–316.

Received for review July 31, 2007. Revised manuscript received October 18, 2007. Accepted October 22, 2007. This work is supported by the EC project “PIPS—Personalised Information Platform for Life and Health Services” (IST-2004-507019) and by Laboratorio Regionale per la Sicurezza e la Qualità degli Alimenti (SIQUAL) [project number 9, Programma Regionale per la Ricerca Industriale, l’Innovazione e il Trasferimento Tecnologico (PRRIITT)].