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Chemical composition, nutritional value and antioxidant properties of *Allium caepa* L. Var. *tropeana* (red onion) seeds

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Abstract

Chemical analysis of *Allium caepa* L. var *Tropeana* (red onion) seeds showed high amounts of oil (20.4%), fibre (22.4%), crude protein (24.8%), calcium (175.0 mg/100 g), potassium (1010 mg/100 g), low amounts of sodium (11.2 mg/100 g) and six cysteine derivatives, of which the *S*-propylmercapto-cysteine has never been reported in onion before. The antioxidant capacity of seed extracts containing cysteine derivatives (SECCD), before and after boiling the seeds, and of cooking water extracts containing cysteine derivatives (CWECCD), was also evaluated, by the ferric reducing antioxidant power (FRAP) and 1,1-diphenyl-2-picrilhydrazyl (DPPH) assays. The extracts showed discrete antioxidant capacity which increased after boiling, although cooking methods caused significant losses of the cysteine derivatives in water.

Keywords: Alliaceae; *Allium caepa*; Antioxidant activity; Chemical composition; Red onion seeds

1. Introduction

Onion is one of the most important vegetable crops, with a world production of about 55 million tonnes (FAO, 2004). Its consumption is attributed to several factors, mainly heavy promotion that links flavour and health and the popularity of onion-rich ethnic foods. Onion bulbs are the main edible part, with a distinctive strong flavour and pungent odour. Onion seeds are also eaten, especially in some indian dishes, they do not affect the breath as strongly as bulbs do, nevertheless, their commercial availability is currently limited. Perhaps, if consumers were much more acquainted with onion seed nutritional and functional properties, there would be a boost in the trade market for this product. At the moment, no data about onion seed chemical value are available in literature. Onion represents a source of cysteine derivatives, which makes it a good antioxidant additive for food (Ostrowska et al., 2004), increasing its potential usability as a functional food and in ethnomedicine (Tram Ngoc et al., 2005). Oxidation is one of the major causes of chemical spoilage, resulting in rancidity and/or deterioration of the nutritional quality, colour, flavour, texture and safety of foods (Antolovich, Prenzler, Patsalides, McDonald, & Robards, 2002). Dietary antioxidants are important components because they protect against free radicals, such as reactive oxygen species in the human body. Free radicals are known to be the major contributors to degenerative diseases of aging and are recognised as major factors causing cancer, cardiovascular disorders and diabetes. At present, there is an increasing interest both in industry and scientific research in spices and aromatic herbs because of their strong antioxidant and antimicrobial properties. These properties are due to many substances, including some vitamins, flavonoids, terpenoids, carotenoids, phytoestrogens, minerals, etc. and render spices and some herbs or their antioxidant components as preservative agents in food (Calucci,

Abbreviations: SECCD, seed extracts containing cysteine derivatives; CWECCD, cooking water extracts containing cysteine derivatives.

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Pinzono, Zandomeneghi, & Capocchi, 2003). The spices, in view of the many promising health beneficial physiological effects, have assumed the status of "Nutraceuticals" and are considered as a natural and necessary components of our daily nutrition. Moreover, since modern consumers are increasingly asking for natural products, free of synthetic additives, the application of natural antioxidants will probably continue in the future and it will be necessary to study their changes and interactions in more details. The aim of this work is the study of the nutritional value, including cysteine derivative quality, of the seeds of a low pungency or so-called "sweet" onion variety, the Tropea red onion, to make available appropriate nutritional labelling so that consumers can be better informed towards this product. Since these seeds are generally consumed cooked, the antioxidant activity of seed extracts containing cysteine derivatives (SECCD) before and after boiling the seeds, and of cooking water extracts containing cysteine derivatives (CWECCD), was also evaluated, by using DPPH and FRAP tests, in order to establish how a traditional cooking method could affect it. Several in vitro analytical methods are available to characterise the antioxidant propensity of bioactive compounds in plant foods and supplements. The use of more than one method is recommended to give a comprehensive prediction of antioxidant efficacy. A factor that provides a distinct challenge in the assay of antioxidant capacity is that within biological systems, there are at least four general sources of antioxidants: enzymes (superoxide dismutase, glutathione peroxidase and catalase); large molecules (albumin, ceruloplasmin, ferritin and other proteins); small molecules [ascorbic acid, glutathione, uric acid, tocopherol, carotenoids, (poly)phenols] and some hormones (estrogen, angiotensin, melatonin, etc.). Furthermore, there are multiple free radical and oxidant sources [e.g. $O_2^{\bullet}$, $HO^{\bullet}$, $NO^{\bullet}$, $ONOO^{-}$, $HOCl^{\bullet}$, $RO(O)^{\bullet}$, $LO(O)$] and both oxidants and antioxidants have different chemical and physical characteristics. Therefore, there is no simple universal method by which antioxidant capacity can be measured accurately and quantitatively. The mechanism of antioxidant action in vitro may involve direct inhibition of the generation of reactive oxygen species, or the scavenging of free radicals. We used FRAP assay because it is the only one that directly estimates the capacity of antioxidants or reductants in a sample and is based on the ability of the analyte to reduce Fe^{3+}/Fe^{2+} couple. The reaction detects compounds with redox potentials of <0.7 V (the redox potential of Fe^{3+}-TPTZ), so the FRAP test is a reasonable screen for the ability to maintain redox status in cells or tissues. Reducing power appears to be related to the degree of hydroxylation and extent of conjugation in polyphenols (Ou, Huang, Hampsch-Woodill, Flanagan, & Deemer, 2002). However, the FRAP test cannot detect compounds which act by radical quenching (H transfer), particularly thiols and proteins (Cao, Sofic, & Prior, 1997). In addition, since reduced metals are active propagators of radical chains via hydroperoxide reduction to RO**.**, it would be interesting to evaluate whether high FRAP values correlate with the tendency of polyphenols to become pro-oxidants under some conditions. This has been shown for some flavones and flavanones (Delgado-Andrade, Rufián-Henares, & Morales, 2005), which also have high FRAP values. The DPPH test determines radical scavenging activities of compounds by measuring the inactivation potential of radicals in an aqueous mean. It is simple and rapid but it has generally a relatively small linear reaction range of only 2-3-fold and small molecules that have better access to the radical site have higher apparent antioxidant activity with this test. Furthermore, the DPPH radical is decolourised by reducing agents as well as H transfer, which also contributes to inaccurate interpretations of antioxidant capacity (Beretta, Granata, Ferrero, Orioli, & Maffei Facino, 2005; Noruma, Kikuchi, & Kawakami, 1997). Interpretation of data is further complicated when the tested compounds, carotenoids in particular, have UV spectra which overlap with those of the DPPH complex at 515 nm (Benzie & Strain, 1996).

2. Materials and methods

2.1. Plant material and chemicals

Red onion seeds, available in the market, from the Capo Vaticano (VV-Italy), were identified (and reference specimens deposited) at the Dipartimento di Chimica delle Sostanze Naturali, University of Federico II, Napoli (Italy). DPPH (1,1-diphenyl-2-picrilhydrazyl), ferric chloride dry, 2,4,6-tris-2,4,6-tripyridyl-2-triazine (TPTZ), 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxilic acid (Trolox), *S*-methyl-L-cysteine (99%) and *S*-ethyl-L-cysteine (99.5%) were purchased from Sigma Chemical (St. Louis, MO). *S*-allyl-L-cysteine (99%), *S*-propyl-L-cysteine (99%) and *S*-propylmercapto-L-cysteine were supplied by Wakunaga Pharmaceutical (Osaka, Japan). *S*-2-hydroxyethyl-cysteine was synthesised by the reaction of L-cysteine with 2-bromoethanol, as described by Shiraiwa et al. (1998).

2.2. Methods

2.2.1. Cooking regimens

Red onion seeds were heat-treated by boiling. For boiling, 10 g seeds were added to 100 ml of distilled boiling water, after 20 min, the seeds were drained and processed for the analyses.

2.2.2. Determination of crude protein, moisture, ash, oil and dietary fibre

The recommended methods of the Association of Official Analytical Chemists (AOAC, 1990) were adopted to determine the levels of crude protein, moisture, ash and oil. Nitrogen content was determined by using the Kjeldahl method (Kjeldahl, 1883) and multiplied by a factor of 6.25 to determine the crude protein content. The moisture content was determined by drying the samples, at 105 °C, to a constant weight. Crude fat was determined by the Soxhlet

method. Crude fat was obtained by exhaustively extracting 5.0 g of each sample in a Soxhlet apparatus using petroleum ether (boiling point range 40–60 °C) as the extractant. Dietary fibre content of the defatted samples was determined by decomposing the starches with acids and the proteins with base and then filtering (Nielsen, 1998). Total carbohydrate content was obtained by difference.

2.2.3. Determination of sugars

Sugar contents (raffinose, sucrose) were determined after extraction of the seed powder with warm H_2O, for 2 h 30 min and centrifugation, at 18.000*g*. The clarified extract was filtered through a 0.45 μm filter (Millipore Millex-HV). A Waters-Millipore Sugar-pak cartridge thermostatted at 90 °C was used. EDTA-Ca (50 mg/L), at a flow-rate of 0.5 ml/min, served as the elution solvent. Individual sugars were identified by comparison with an internal standard (sucrose 0.1–0.2%; glucose 0.1–0.2%; fructose 0.05–0.1%; raffinose 0.02–0.04%). A Waters-Millipore 600 E liquid chromatograph was employed and a differentia1 refractometer (Model 410) was utilised as a detector for sugar analysis (Voi, Impembo, Fasanaro, & Castaldo, 1995).

2.2.4. Determination of minerals

Mineral elements (Na, K, Ca, Mg, Cu, Zn, Fe) were measured by atomic absorption after seed flour mineralisation, performed with a sulfo–nitric mixture according to the National Italian Standards [Ministero dell'Agricoltura e delle Foreste (1989). Official Italian Methods. "Metodi Ufficiali di Analisi per le Conserve Vegetali- parte generale" Gazzetta Ufficiale della Repubblica Italiana no. 168 del 20-07-1989 (Rome, Italy: Istituto Poligrafico dello Stato)] using standard reference materials (BDH Chemical Ltd., Poole England).

2.2.5. Determination of anions

Anion contents (Cl^-, F^-, NO_3^-, PO_4^{3-}, SO_4^{2-}, $C_2O_4^{2-}$) were determined using the same tools used to measure sugars, except for the detector which was a Dionex Pulser electrochemical detector provided with Suppressed Conductivity ASRS-II™ Autosuppression™ Recycle Mode. The analyses were carried out on an Ion Pac AS4A-SC Analytical 4 × 250 mm column and samples eluted with buffer solution ($Na_2CO_3 = 1.7$; $NaHCO_3 = 1.8$ mM) (Louidice et al., 1995).

2.2.6. Total amino acid analysis

Total amino acid contents were determined after hydrolysis with 6 N HCl at 100 °C in vacuum hydrolysis tubes, following the Waters AccQ-Tag Method (1993) [manual number WATO-52874 TP, Rev. April, 1993 WATERS]. The analysis was performed by reverse-phase HPLC using a Waters system with Alliance 2690 Separation Module pump, an auto sampler provided with injector programme, a fluorescence detector Mod. 474 ($\lambda_{ecc} = 250$ nm; $\lambda_{em} = 395$ nm) and AccQ-Tag 3.9 × 150 mm column thermostatted at 35 °C. The derivatization procedure was done with "AccQ-Fluor Reagent Kit Waters" (borate buffer, AQC, CH_3CN; Cat. No. 052880). The eluents were a gradient of phosphate buffer pH 5.80 (A), CH_3CN (B) and H_2O (C) (flow-rate 1.0 ml/min) (Strydom & Cohen, 1994).

2.2.7. Free amino acid analysis

Free amino acid samples were prepared for HPLC analysis by flour (1 g) extraction with 0.1 M HCl (about 10 ml), then filtered by 0.45 μm filter (Millipore Millex-HV). One millilitre was applied to a cation exchange (100 × 6 mm) column [AG 50W-X8(H^+), Bio-Rad]. After the column was washed with 50 ml of milli-Q Water, the amino acids were eluted with 3.0 M NH_3 (about 10 ml). The sample was evaporated and recovered with 0.01 M HCl (9.2 ml) and internal standard (BABA 2.5 mM in 100 ml 0.1 M HCl). Free amino acids were determined using the same tools used to measure total amino acids.

2.2.8. Extraction and characterisation of cysteine derivatives

Red onion seeds were extracted with MeOH:H_2O-8:2. The extract was partitioned between $CHCl_3$ and H_2O. The aqueous portion was partitioned between BuOH and H_2O. The aqueous layer, containing the crude mixture of cysteine derivatives, was analysed by MS and MS–MS spectra. Results were compared to analytical standards. Positive mode electrospray ionisation ESI–MS spectra were recorded in CH_3OH on an AB Applied Biosystems mass spectrometer API 2000 MS/MS system. Operational parameters were as follows: vaporiser, 350 °C; heated capillary, 150–200 °C; carrier gas, nitrogen was at a sheath gas pressure of 70 psi and auxiliary gas was on a 30 psi to assist in nebulisation; fragmentor voltage was set at 200 V; ions were decomposed in the collision cell, at 0.8 m Torr, by using an optimised collision energy of 55.0 eV.

2.2.9. Determination of antioxidant capacity of SECCD and CWECCD

SECCD before and after boiling the seeds, and CWECCD, were obtained with the same procedure as described in the paragraph of the extraction and characterisation of cysteine derivatives. For antioxidant activity determination, SECCD and CWECCD were adjusted to 1 mg/ml (on dry basis), which was chosen as an appropriate concentration for assessing antioxidant activity after preliminary studies of the different concentrations. For each antioxidant assay, a trolox aliquot was used to develop a 50–500 μmol/L standard curve. All data were then expressed as trolox equivalents (TEs, μM). Assay results were obtained using a Jasco V-530 UV–Vis spectrophotometer (Tokyo, Japan) set at wavelengths appropriate to each assay. All assays were performed in triplicate.

2.2.9.1. Determination of Ferric reducing/antioxidant power. The total antioxidant potential of samples was determined by measuring the ferric reducing antioxidant power (FRAP assay) according to Benzie and Strain (1996). A solution of

10 mM TPTZ and 12 mM ferric chloride in 40 mM HCl was diluted in 300 mM sodium acetate buffer (pH 3.6) at a ratio of 1:1:10. SECCD and CWECCD solutions (50 μl) were added to 3 ml of the FRAP solution and the absorbance at 593 nm was determined after assay samples were allowed to react for 90 min.

2.2.9.2. Determination of free radical scavenging ability by the use of a stable DPPH radical (1,1-diphenyl-2-picrilhydrazyl). The ability of SECCD and CWECCD solutions to scavenge the DPPH radical was measured using the method of Brand-Williams 1995. An aliquot of 50 μl of each solution was added to 3 ml of DPPH solution (6×10^{-5} mol/L) and the absorbance was read at 515 nm after 90 min.

2.2.9.3. Statistics. Triplicate analyses for each measurement were conducted for each sample. To establish the reproducibility of the analytical method, sample preparation was repeated three times. Differences between the means were evaluated with ANOVA, using the Graf Pad Instat 3 (Microsoft Software) statistics program. The significance of the model was evaluated by ANOVA. The significance of the regression coefficients was evaluated by the Student's *t* test. The significance level was fixed at 0.05 for all the statistical analyses.

3. Results and discussion

3.1. Proximate and fibre analyses

The proximate composition is shown in Table 1. These values were compared to corresponding data for several typical spices of italian cuisine, *Allium caepa* bulb raw as reported in USDA National Nutrient Database for Standard Reference (2004) and other *A. caepa* seeds. The protein level in Tropea red onion seeds was 24.8% and the total lipid content was found to be 20.4%, the total sugar content was found to be 21.9% and the amount of fibre was 22.4%. All nutrient and fibre contents were higher than the values reported for typical spices used in italian cuisine and bulbs, considering the higher water content, except carbohydrates (Table 1). The high fat content, similar to sinapis alba may also suggest, for Tropea red onion, the possibility of using its seeds for obtaining an aromatic oil useful to make dishes more flavoury.

3.2. Amino acid analysis

Quantitative determination of amino acid concentrations was conducted by HPLC. Sixteen amino acids were detected and the separation of the amino acids in the samples was reasonably good. All of the essential amino acids, namely leucine, lysine, cystine, tyrosine, isoleucine, threonine, histidine and valine, were found to be present in Tropea red onion seeds. Table 2 shows the essential amino acid pattern of Tropea red onion seeds compared to the FAO/WHO (1990) for evaluating proteins (FAO/WHO, 1990). The amino acid profile of Tropea red onion seeds showed that all the essential amino acids were at lower concentrations, when compared with the reference pattern (FAO/WHO, 1990), which implied that the proteins present had a low biological value and are therefore called incomplete proteins. Nevertheless, the essential amino acid profile was similar to that characteristic of other typical spices of italian cuisine and lower than other *A. caepa* seeds.

3.3. Free aminoacid analysis

Table 3 shows the free amino acids present in Tropea red onion seeds. In particular, these seeds revealed large amounts of glutamic acid (97.3 mg/100 g), arginine (88.9 mg/100 g) along with lesser amounts of tyrosine (69.3 mg/100 g) and asparagine (52.3 mg/100 g). These four amino acids constitute 68.8 % of the total free amino acids in Tropea red onion seeds. Although individual free amino acids have been reported to impart distinct taste and flavour to foods, the free amino acids found in higher amounts in Tropea red onion seeds may be responsible for their colour and/or bitter, sour or sweet taste (Basha & Young, 1985; Botta, Gianotti, Richardson, Suwanagul, & Sanz, 1994; Macleod, 1998; Nishimura & Kato, 1988).

Table 1
Chemical composition of commercial *Allium caepa* L. Var. *tropeana* seeds and some typical spices of italian cuisine (%)

	Water	Crude protein	Fat	Carbohydrate	Fibre
*Allium caepa*var. *tropeana* (Tropea red onion seeds)[a]	10.5 ± 0.1	24.8 ± 0.1	20.4 ± 0.7	21.9 ± 0.9	22.4 ± 0.4
Allium caepa bulb raw[*]	89.11	1.1	0.1	9.34	1.7
Allium caepa seeds[b]	6–8	27.9	23.6	20.4	16.1
Anethum graveolens (dill seeds)[*]	7.7	16.0	14.5	55.2	21.1
Carum carvi[*] (caraway seeds)	9.9	19.8	14.6	49.9	38.0
Faeniculum vulgare[*] (fennel seeds)	8.8	15.8	14.9	52.3	39.8
Sinapis alba[*] (mustard)	6.9	24.9	28.8	34.9	14.7

[a] Values are the mean ± SD of three different determinations ($p < 0.0001$).
[b] Udayasekhara, P.R. (1994). Nutrient composition of some less-familiar oil seeds. Food Chemistry. 50, 379–380.
[*] Source: USDA National Nutrient Database for Standard Reference, Release 16.1 (2004).

Table 2
Essential amino acid pattern of *Allium caepa* L. Var. *tropeana* seeds compared to some typical spices of italian cuisine and the FAO/WHO reference pattern (1990) for evaluating proteins (mg/100 grams)

Amino acid	*Allium caepa* var. *tropeana* (Tropea red onion seeds)[a]	FAO**	Essential aminoacid score	*Allium caepa* bulbs raw*	*Allium caepa* seeds[b]	*Anethum graveolens* (dill seeds)*	*Carum carvi** (caraway seeds)	*Faeniculum vulgare** (fennel seeds)	*Sinapis alba** (mustard)
Isoleucine	0.7± 0.4	2.8	0.25		4.4	0.8	0.8	0.7	1.1
Leucine	1.2 ± 0.1	6.6	0.18		6.1	0.9	1.2	1.0	1.8
Lysine	1.3± 0.2	5.8	0.22		5.0	1.0	1.0	0.8	1.5
Phenylalanine	0.8 ± 0.6				3.9	0.7	0.9	0.6	1.1
Tyrosine	0.5 ± 0.5	6.3	0.07	0.014	2.3	Missing	0.6	0.4	0.7
Cystine	0.9 ± 0.5	2.5	0.36			Missing	0.3	0.2	0.6
Threonine	0.8 ± 0.7	3.4	0.23	0.021	2.5	0.6	0.7	0.6	1.1
Valine	1.0 ± 0.6	3.5	0.28	0.021	4.4	1.1	1.0	0.9	1.3
Arginine	2.3 ± 0.4			0.104	9.4	1.3	1.2	0.7	1.7
Alanine	0.9 ± 0.4			0.021	4.1	Missing	0.9	0.8	1.2
Serine	1.5 ± 0.3			0.021	5.4	Missing	0.9	0.9	1.1
Proline	0.7 ± 0.5			0.012	1.9	Missing	0.9	0.9	1.9
Aspartic acid	1.7 ± 0.3			0.091	6.5	Missing	2.1	1.8	2.0
Glycine	1.0 ± 0.4			0.025	3.8	Missing	1.3	1.1	1.3
Histidine	0.5 ± 0.4	1.9	0.26	0.014	1.9	0.3	0.5	0.3	0.8
Glutamic acid	4.7 ± 0.6			0.258	21.3	Missing	3.2	2.9	5.0

[a] Values are the mean ± SD of three different determinations ($p < 0.0001$).
[b] Udayasekhara (1994).
* Source: USDA National Nutrient Database for Standard Reference, Release 16.1 (2004).
** FAO/WHO, 1990 Protein quality evaluation in: Report of Joint FAO/WHO expert consultation; Food and Agricultural Organization of the United Nations: Rome 1990, 23.

3.4. Cysteine derivative analysis

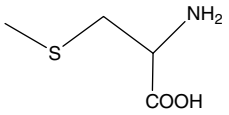
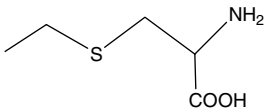
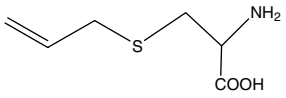
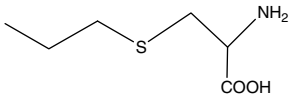
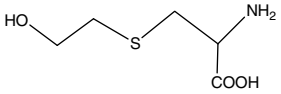
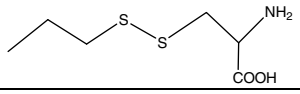
Table 4 shows cysteine derivatives in red onion seeds. Particularly interesting is the presence of *S*-propylmercapto-cysteine, whose occurrence has never been reported in onion. Dietary intake of cysteine derivatives is important because extensive research in the last few years has revealed that regular consumption of *S*-allyl-cysteine can reduce the risk of acquiring specific cancers. This chemopreventive agent has also very recently been found to reverse chemoresistance and radioresistance in patients undergoing cancer treatment. Thus, it has potential to be used as an adjunct to current cancer therapies (Thambi & Aggarwal, 2004). Moreover, *S*-substituted L-cysteine and derivatives have shown cardiovascular-protective properties because some of these protect partially oxidised and glycated LDL or plasma against further oxidative and glycative deterioration, which might benefit patients with diabetic-related vascular diseases (Chien-Ning, Joeu-Shyan, & Mei-Chin, 2004), and inhibit fatty acid, triglyceride (Lijuan and Yu-Yan, 2001) and cholesterol synthesis (Yu-Yan & Lijuan, 2001). Finally, *S*-substituted L-cysteine and derivatives are useful to improve conditions and to alleviate the symptoms of dermatologic disorders related to the impairment of lipid metabolism, suitable for the treatment of edematous-fibrosclerotic panniculopathy, ichthyosis, hyperkeratosis, Darier disease, lichen simplex chronicus, keloid, scar, acne, rosacea and couparose (Ghisalberti, 2003). *S*-allyl-cysteine, *S*-ethylcysteine, *S*-methylcysteine and *S*-propylcysteine have been reported to improve glycemic control, delay oxidation damage, down-regulate inflammatory cytokines and enhance anticoagulant activity in diabetic mice via their antioxidant activities (Ostrowska et al., 2004); in particular, propylmercapto-cysteine caused decreased food intake (Hatono & Wargovich, 1997) and reduced cancer risk (Pinto, Krasnikov, & Cooper, 2006). Finally, *S*-methylcysteine, *S*-ethylcysteine and *S*-allyl-cysteine should be use for treatment of obesity because of induced adipose tissue cell death (Li, Dellafera, & Baile, 2006).

Table 3
Free amino acid pattern of *Allium caepa* L. Var. *tropeana* seeds

Free amino acid	(mg/100 g of red onion seeds)
Threonine	22.8 ± 0.3
Aspartic acid	25.1 ± 0.6
Valine	8.9 ± 0.7
Asparagine	52.3 ± 0.3
Glutamic acid	97.3 ± 0.2
Serine	6.7 ± 0.4
Glycine	11.0 ± 0.4
Glutamine	19.6± 0.6
Hystidine	3.0 ± 0.4
Arginine	88.9 ± 0.6
Alanine	30.5 ± 0.6
Proline	5.9 ± 0.4
Tyrosine	69.3 ± 0.3
Gaba	5.8 ± 0.5

Values are the mean ± SD of three different determinations ($p < 0.0001$).

Table 4
Cysteine derivatives in *Allium caepa* L. Var. *tropeana* seeds

Compound	Structure	MW	MS–MS spectral data of $[M+H]^+$ (ion relative abundance)
S-methyl-cysteine		135	136 (100); 74 (62); 38 (38)
S-ethyl-cysteine		149	150 (100); 74 (14); 61 (7)
S-allyl-cysteine		161	145 (100); 74 (55); 116 (27); 87 (16); 162 (14); 41 (1)
S-propylcysteine		163	164 (100); 74 (50); 136 (33); 89 (20)
S-2-hydroxyethyl-cysteine		165	166 (100); 74 (5)
S-propylmercapto-cysteine		195	196 (100); 74 (83); 75 (75); 107 (22); 88 (25); 43 (8)

3.5. Dietary fibre analysis

Noteworthy is dietary fibre content (22.4%) (Table 1). Dietary fibre (indigestible carbohydrate) is not a nutrient but it still plays a very important role in maintaining good health (Anderson & Bridges, 1998; Johnson & Southgate, 1994; Kritchevsky, 1998; Wisker, Daniel, & Felddheim, 1996). Diets rich in dietary fibre have been associated with beneficial effects on human health and are sometimes considered to be useful for the prevention of obesity (Wisker, Daniel, Felddheim, 1996).

3.6. Anion analysis

Six anions were positively identified in Tropea red onion seeds (Table 5). These included Cl^-, F^-, NO_3^-, PO_4^{3-}, SO_4^{2-}, and $C_2O_4^{2-}$. Oxalates ($C_2O_4^{2-}$) and phosphates (PO_4^{3-}) form water-soluble salts with Na^+, K^+ and NH_4^+ ions and also bind Ca^{2+}, Fe^{2+} and Mg^{2+}, rendering these minerals biologically unavailable (Noonan & Savage, 1999).

3.7. Mineral analysis

Tropea red onion seeds showed high levels of copper and zinc and a lower level of sodium (Table 6) (Konishi, Hirano, Tsuboi, & Wada, 2004). Calcium, iron and zinc bioavailability is low due to the high fibre content and low mineral/P ratio (Belitz & Grosch, 1999). A high potassium/sodium ratio (90.2) makes Tropea red onion seeds interesting for diets with a defined electrolytic balance (Stamler, 1994). The high content of potassium can be utilised beneficially in the diets of people who take diuretics, to control hypertension and suffer from excessive excretion of potassium. Minerals are also important as constituents of bones, teeth, soft tissues, haemoglobin, muscle, blood and nerve cells and are vital to overall mental and physical well being (MAFF, 1995; Sardesai, 1998).

3.8. Free sugar analysis

Two sugars were positively identified in Tropea red onion seeds. These included sucrose (1.60 g%), and its

Table 5
Anions in *Allium caepa* L. Var. *tropeana* seeds

Anions	mg/100 g
Cl^-	879.70 ± 0.7
F^-	203.33 ± 0.5
NO_3^-	35.66 ± 0.5
PO_4^{3-}	1712.39 ± 1.1
SO_4^{2-}	605.51 ± 0.5
$C_2O_4^{2-}$	209.40 ± 0.8

Values are the mean ± SD of three different determinations ($p < 0.0001$).

galactoside, namely raffinose (0.20 g%). Sugars are responsible for the sweetness of foods. Individual sugars possess different relative sweetness scores; fructose has been reported to be the sweetest sugar (1.1–1.8), followed by sucrose (1.0) and glucose (0.5–0.8) (Alexander, 1998).

3.9. The antioxidant capacity of SECCD and CWECCD

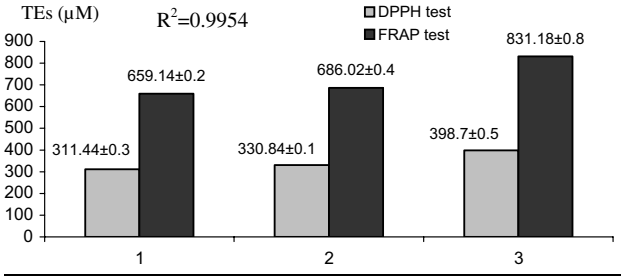
The antioxidant capacity of SECCD before and after boiling the seeds, and CWECCD, was evaluated in order to establish how a traditional cooking method could affect it, by using two different methods: FRAP and DPPH tests (Table 7). Seeds were cooked by boiling for 20 min, considered as the traditional cooking time. Interestingly, SECCD $TEAC_{FRAP}$ values were consistently higher than $TEAC_{DPPH}$ ones (Table 7). The probable reason for the lower $TEAC_{DPPH}$ values could be due to the presence of compounds not reactive towards DPPH (Przybylski, Lee, & Eskin, 1998). This could be explained from the basic concept that antioxidants are reducing agents for their ability of donating a single electron or hydrogen atom for reduction, but not all reducing agents are antioxidants. In this study, the strong correlation ($R = 0.9954$) between the mean values of $TEAC_{DPPH}$ and $TEAC_{FRAP}$ indicated that compounds present in the extracts capable of reducing DPPH radicals were also able to reduce ferric ions. Finally, our results showed that cysteine derivatives have discrete antioxidant capacity, which increased after boiling for 20 min. It was in agreement with the results reported by Gazzani, Papetti, Massolini, and Daglia (1998), that the antioxidant activity of vegetables increased by boiling because pro-oxidant activity was due to peroxidases which were inactivated at high temperatures (Gazzani et al., 1998). In contrast to what has been reported by Hunter and Fletcher, 2002, our results showed that cooking by boiling caused significant losses of antioxidant compounds in water.

Table 6
Contents of microelements in *Allium caepa* L. Var. *tropeana* seeds compared to some typical spices of italian cuisine

	Calcium	Magnesium	Potassium	Sodium	Iron	Copper	Zinc
Allium caepa L. Var. *tropeana* seeds[a]	175 ± 1.2	325 ± 1.1	1010 ± 1.1	11.2 ± 0.3	6.60 ± 0.4	0.92 ± 0.04	7.25 ± 0.02
Allium caepa L bulb raw[*]	23	10	146	4	0.21	0.039	0.17
Allium caepa seeds[b]	279	414			43	0.5	5.6
Anethum graveolens (dill seeds)[*]	1516	256	1186	20	16.3	0.78	5.20
Carum carvi[*] (caraway seeds)	689	258	1351	17	16.2	0.91	5.50
Faeniculum vulgare[*] (fennel seeds)	1196	385	1694	88	18.5	1.07	3.70
Sinapis alba[*] (mustard)	521	298	682	5	9.98	0.41	5.70

Value (mg) per 100 g of edible portion.
[a] Values are the mean ± SD of three different determinations ($p < 0.0001$).
[b] Udayasekhara (1994).
[*] Source: USDA National Nutrient Database for Standard Reference, Release 16.1 (2004).

Table 7
Antioxidant tests of SECCD before and after boiling *Allium caepa* L. Var. *tropeana* seeds and of CWECCD



Values are the mean ± SD of three different determinations ($p <+ 0.0001$).

4. Conclusion

In conclusion, our results showed that red onion seeds contained high amounts of oil (20.4%), fibre (22.4%) and crude protein (24.8%). The mineral content was high in calcium (175.0 mg/100 g) and potassium (1010 mg/100 g) and low in sodium (11.2 mg/100 g). The high fat content may suggest the possibility of using these seeds for obtaining an aromatic oil useful to make dishes more flavour. Noteworthy is the presence of six cysteine derivatives for their biological properties, with special regard to *S*-propylmercapto-cysteine whose occurrence has never been reported in onion. Their presence should be important for obesity treatment because they improve glycemic control, cause decreased food intake and induce adipose tissue cell death (Li et al., 2006). Finally, SECCD revealed a discrete antioxidant effect that increased after boiling the seeds, although cooking methods caused significant losses of antioxidant compounds in water.

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