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Characterization of Violetto di Toscana, a typical Italian variety of artichoke (*Cynara scolymus* L.)

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Abstract

This work regards the characterization of the phenolic compounds and the mineral composition of the variety Violetto di Toscana, which is typically cultivated in Tuscany for the flavour of its edible part. All the results achieved are compared with those of var. Terom, which is widely used in Italy. A systematic study was performed on the different plant parts: the edible head and leaves, outer bracts and stems. The data for the whole plant showed that the polyphenol content of var. Violetto exceeds that of var. Terom by about 25%, so this variety could be regarded as a functional food and also as an interesting source of antioxidant phenolic compounds. A decoction of bracts and leaves was also carried out in order to evaluate the amount of polyphenols which may be extracted by a simple hot water extraction.

Keywords: *Cynara scolymus* L.; Polyphenol antioxidants; Mineral composition; Violetto di Toscana; Terom

1. Introduction

Artichoke is an herbaceous perennial plant (*Cynara scolymus* L.) belonging to the Compositae family (Asteraceae) cultivated in the Mediterranean area. The heads of the artichoke are edible and used worldwide; the leaves are an herbal medicine recognized for a long time for their beneficial effects against liver complaints and for their antioxidant action. In particular, the leaf extract exhibits different effects: it lowers blood cholesterol, exerting a potent anticholestatic activity (Coon & Ernst, 2003; Gebhardt, 2001), and it shows antioxidant, bile expelling, and hepatoprotective effects (Englisch, Beckers, Unkauf, Ruepp, & Zinserling, 2000; Jimenez-Escrig, Gragsted, Daneshvar, Pulido, & Saura-Calixto, 2003; Saenz Rodriguez, Garcia Gimenez, & De La Puerta Vazquez, 2002). Moreover, many studies have focused on its anti-oxidant properties (Adzet, Camarasa, & Laguna, 1987; Gebhart, 1997; Perez-Garcia, Adzet, & Canigueral, 2000; Wang et al., 2003) and most of these actions are related to the presence of the polyphenolic fraction.

On the other hand, from an agronomical point of view, each region preserves its peculiar cultivars which may differ in chemical composition and may, therefore, exhibit different properties (Alamanni & Cossu, 2003; Hammouda, El-Nasr, Ismail, & Shahat, 1993). Recently, Lanteri et al. (2004) performed the analysis of the artichoke genome by means of molecular markers on different artichoke accessions, as measured by amplified fragment length polymorphism, to evaluate *Cynara* genetic diversity and classify the accessions to phylogenetic groups.

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In Tuscany, artichoke was first introduced in the 15th century and "Violetto di Toscana" is one of the more widespread and peculiar varieties. This paper deals with the characterization of the Violetto di Toscana variety from the point of view of the phenolic composition of the different plant parts: the edible head and leaves, outer bracts and stems, which may be regarded as byproducts providing anti-oxidant material (Llorach, Espin, Tomas-Barneran, & Ferreres, 2002). The mineral composition of the different parts has also been studied so that a window on the nutritional value of this variety could be opened. All the results achieved are compared with those of var. Terom, which is widely used in Italy.

2. Experimental

2.1. Sample preparation

Leaf, outer bract, head and stem of the two accessions (Violetto di Toscana and Terom) were collected in Pian di Rocca area (Grosseto, Italy). The raw material was lyophilized and then homogenized. A quantity of 10–30 g was used for the extraction with 3×100 ml of 70% v/v ethanol (pH 2); the extract was then concentrated under vacuum (Rotavapor 144 R, Büchi, Switzerland) and rinsed with ultra pure acid water (MilliQ system, Waters Co., Milford, MA, USA) adjusted to pH 2 with formic acid. It was then completely defatted with *n*-hexane (4×20 ml) and concentrated under vacuum to a final volume of 5 ml. The lyophilized extracts were stored at −20 °C until use and remained stable for at least 12 months. Chlorogenic acid and cynarin were from Roth (Germany); luteolin 7-*O*-glucoside (cynaroside) and luteolin 7-*O*-rutinoside (scolymoside) were purchased from Extrasynthese S.A. (Lyon, Nord-Genay, France). All solvents were HPLC grade and were obtained from Merck (Darmstadt, Germany).

2.2. HPLC/DAD and HPLC/MS analysis

The analysis was conducted using a HP-1100 liquid chromatograph equipped with a DAD detector and a HP 1100 MSD API-electrospray (Agilent Technologies, Palo Alto, USA) operating in positive and negative ionization mode under the following conditions: gas temperature 350 °C, nitrogen flow rate 10.0 l min^{-1}, nebulizer pressure 40 psi, quadrupole temperature 40 °C, and capillary voltage 3500 V. Fragmentors operated in the range 80–180 eV.

Polyphenol compounds were separated using a 250×4.6 mm (5 μm) LiChrosorb RP18 (Merck) maintained at 26 °C. Eluent comprised: (A) H_2O (pH 3.2 by HCOOH), (B) CH_3 CN. A four-step linear solvent gradient system was used, starting from 0% up to 100% of the solvent B during a 40-min period, at a flow rate of 0.8 ml min^{-1}. The percentage of B reached the 11% from 0 to 5 min, then 20% from 10 to 15 min, and finally 100% from 25 to 33 min.

UV–Vis spectra were recorded in the range 190–600 nm, and chromatograms were acquired at 254, 280, 330, and 350 nm. Identification of individual polyphenols was carried out using their retention times, and both spectroscopic and spectrometric data. Quantitation of the single polyphenol was directly performed by HPLC-DAD using a four-point regression curve built with the available standards. Curves with a correlation factor $r^2 > 0.9998$ were considered. Calibration was performed at the wavelength of maximum UV–Vis absorbance applying the correction for the molecular weight. In particular, caffeoylquinic mono and di-esters amounts were calculated at 330 nm using chlorogenic acid and cynarin as reference, respectively. Luteolin malonylglucoside was calibrated at 350 nm using cynaroside as reference. Finally, apigenin 7-*O*-glucuronide was calibrated at 350 nm using apigenin 7-*O*-glucoside as reference. As an example, in this latter case, the actual apigenin 7-*O*-glucuronide concentration was obtained applying a multiplication factor of 446/432, where 446 is the molecular weight of apigenin 7-*O*-glucuronide and 432 is the molecular weight of apigenin 7-*O*-glucoside.

2.3. Atomic absorption analysis

Sample dissolution: 10 g of plant material was treated with 10 ml of a 1:1 ethanol/glycerol mixture and held at 480 °C for 24 h. The ashes were recovered with 10 ml of 6 M HCl, were filtered and the solution was brought to 100 ml with bidistilled water. A Perkin–Elmer 1100B atomic absorption spectroscope was used for the quantitative determinations of metals.

3. Results and discussion

Table 1 lists some inorganic components of artichoke parts (i.e. water content, total mineral content, main inorganic mineral elements content). These data agree with those reported in the literature (Alamanni, Cossu, & Mura, 2001; Lopez et al., 1997) and with the dietetic characteristics (data not shown), and no significant differences were found between the two varieties and among the different plant parts, apart from a higher sodium content in the leaves of Terom.

The HPLC chromatogram of the extracts of the different parts of Violetto di Toscana is shown in Fig. 1. From a qualitative point of view, it is interesting to underline that leaves contain the highest amount of flavonoids, while they are completely lacking in stems. In both leaves and stems the most representative compound is 1,5-dicaffeoylquinic acid. The qualitative

Table 1
Mineral composition, water and organic matter content of artichoke parts of two varieties: Violetto di Toscana (V) and Terom (T)

Artichoke part	Humidity (%)	Minerals (%)	Organic matter (%)	Sodium (mg 100 g^{-1} fw)	Potassium (mg 100 g^{-1} fw)	Magnesium (mg 100 g^{-1} fw)	Calcium (mg 100 g^{-1} fw)	Copper (mg 100 g^{-1} fw)
Leaves V	79.0	2.85	86.4	163	388	44	53	0.52
Leaves T	80.6	2.71	86.0	201	362	43	50	0.54
Bracts V	83.9	1.17	95.3	51.7	326	19	47	0.33
Bracts T	75.6	0.75	95.2	31.7	378	24	46	0.37
Heads V	78.0	1.05	95.2	32.9	375	24	49	0.53
Heads T	85.7	0.70	95.1	38.6	349	23	51	0.38

Data are expressed as mg 100 g^{-1} of fresh weight (fw). Values are mean of three samples each one performed in triplicate. Standard error was omitted and it was under 1%.

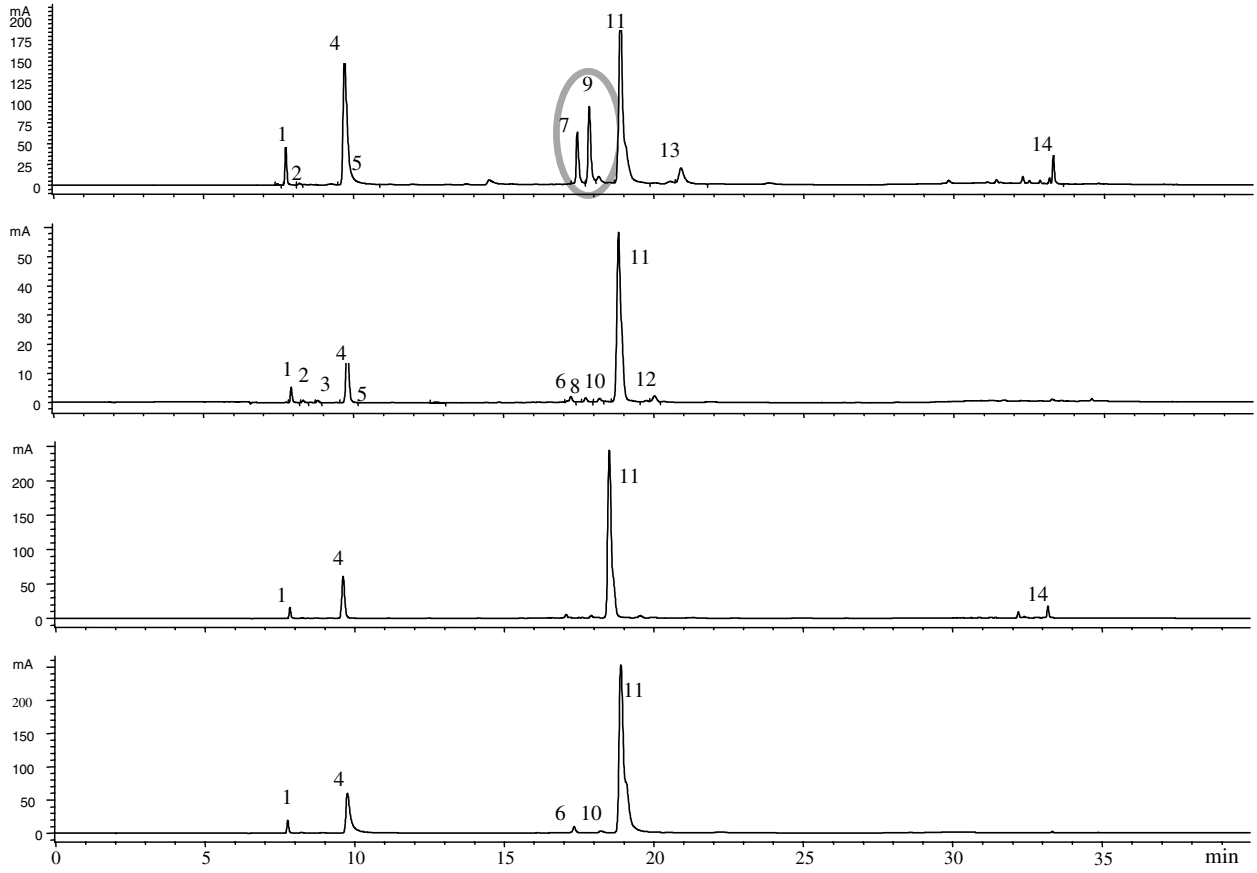

Fig. 1. Chromatographic profile, registered at 330 nm, of hydroalcoholic extracts obtained from different artichoke plant parts (var. Violetto di Toscana). Peaks: 1. 1-*O*-caffeoylquinic acid; 2. 3-*O*-caffeoylquinic acid; 3. 4-*O*-caffeoylquinic acid; 4. Chlorogenic acid; 5. Caffeic acid; 6. Dicaffeoylquinic acid; 7. Luteolin 7-*O*-rutinoside; 8. Luteolin 7-*O*-glucuronide; 9. Luteolin 7-*O*-glucoside; 10. Dicaffeoylquinic acid; 11. 1,5-*O*-dicaffeoylquinic acid; 12. Apigenin 7-O-glucuronide; 13. Luteolin malonylglucoside; 14. Luteolin. (a) leaves; (b) outer bracts; (c) heads; (d) stems.

composition does not differ from previously published findings (Hausler, Ganzera, Abel, Popp, & Stuppner, 2002; Schutz, Kammerer, Carle, & Schieber, 2004; Wang et al., 2003).

The quantitative data are reported in Table 2, where the values are the mean of three analyses. It can be pointed out that, as regards the edible part, the heads of var. Violetto contain more polyphenols than var. Terom. If the data for the whole plant are considered (leaves, stems, heads and bracts), the polyphenols content of var. Violetto exceeds that of var. Terom by about 25%.

The obtained data were compared with those reported in the literature (Table 3) and from the complex of results it emerges that var. Violetto, which is typically cultivated in Tuscany for the flavor of its edible part, may be regarded as a functional food and also as a source of antioxidant phenolic compounds.

Table 2
Polyphenols composition of different parts of artichokes from two varieties: Violetto di Toscana (V) and Terom (T)

	Leaves		Bracts		Heads		Stems	
	V	T	V	T	V	T	V	T
Chlorogenic acid	520 ± 4	1052 ± 7	872 ± 6	253 ± 2	3051 ± 20	1425 ± 10	3029 ± 18	1647 ± 12
Monocaffeoylquinic acids	606 ± 6	843 ± 6	70 ± 1	39 ± 0.4	406 ± 3	56 ± 0.5	324 ± 3	146 ± 2
Dicaffeoylquinic acids	1295 ± 10	721 ± 6	6357 ± 48	2754 ± 21	25335 ± 244	9502 ± 91	22774 ± 218	28325 ± 265
Total phenolic acids	*2421*	*2616*	*7299*	*3046*	*28792*	*10983*	*26127*	*30118*
Luteolin-7-*O*-glucoside	357 ± 4	934 ± 7	0	0	0	0	0	0
Luteolin-7-*O*-rutinoside	584 ± 6	372 ± 3	0	0	0	0	0	0
Luteolin 7-*O*-malonylglucoside	150 ± 2	176 ± 2	0	0	0	0	0	0
Luteolin-7-*O*-glucuronide	0	0	57 ± 1	97 ± 1	0	166 ± 2	0	0
Apigenin-7-*O*-glucuronide	0	0	109 ± 1	66 ± 0.8	116 ± 1	32 ± 0.4	0	0
Luteolin	126 ± 1	0	0	0	842 ± 10	0	0	0
Total flavonoids	*1217*	*1482*	*166*	*163*	*958*	*198*	*0*	*0*
Total polyphenols	*3638*	*4098*	*7465*	*3209*	*29750*	*11181*	*26127*	*30118*

Data are mean values of three determinations and are expressed as μg^{-1} of fresh weight.

Table 3
Amount of polyphenols in different parts of artichoke from different authors

Part of artichoke	Variety	Fresh or dry weight (mg g^{-1})	Reference
Bracts, receptacles and stems		30 (fresh)	Lorach et al., 2002
Leaves	Green globe	62 (dry)	Wang et al., 2003
Young heads	Green globe	14 (dry)	Wang et al., 2003
Mature heads	Green globe	8 (dry)	Wang et al., 2003
Heads	Spinoso sardo	4.8 (fresh)	Alemanni et al., 2001
Leaves	Violetto	3.7 (fresh)	This study
Heads	Violetto	29.8 (fresh)	This study

Table 4
Amounts of lyophilized extract (expressed as g of extract with respect to 100 g of fresh tissue) and of polyphenols (expressed as g of polyphenols with respect to 100 g of lyophilized extract)

	Violetto		Terom	
	Leaves	Bracts	Leaves	Bracts
Lyophilized extract	22.5	14.8	19.1	22.6
Polyphenols	2.4	2.7	1.4	0.6

In order to better evaluate the amount of polyphenols which may be recovered from artichoke byproducts, a decoction of bracts and leaves (Farmacopea Ufficiale Italiana, 1998) was performed and the data are reported in Table 4. The contents of these aqueous extracts may support the possibility of obtaining a lyophilized sample, rich in polyphenols, easily by simple hot water extraction.

Work is in progress to verify if there is a suitable correlation between the polyphenolic composition and the molecular markers, in different artichoke accessions, including clones belonging to the same varietal type, and different accessions of wild and cultivated cardoon.

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References

Adzet, T., Camarasa, J., & Laguna, C. J. (1987). Hepatoprotective activity of polyphenolic compounds from *Cynara scolymus* against CCl_4 toxicity in isolated rat hepatocytes. *Journal of Natural Products, 50*, 612–617.

Alamanni, M. C., & Cossu, M. (2003). Antioxidant activity of the extracts of the edible part of artichoke (*Cynara scolymus* L.) var. spinoso sardo. *Italian Journal of Food Science, 15*, 187–195.

Alamanni, M. C., Cossu, M., & Mura, M. (2001). Evaluation of the chemical composition and nutritional value of artichokes (*Cynara scolymus*) from Sardinia. *La Rivista di Scienza dell' Alimentazione, 30*, 345–351.

Coon, J. S. T., & Ernst, E. (2003). Herbs for serum cholesterol reduction: a systematic view. *Journal of Family Practice, 52*, 468–478.

Englisch, W., Beckers, C., Unkauf, M., Ruepp, M., & Zinserling, V. (2000). Efficacy of artichoke dry extract in patients with hyperlipoprotemia. *Arzneimittelforschung, 50*, 260–265.

Farmacopea Ufficiale Italiana. (10th ed). Istituto Poligrafico e Zecca dello Stato, Roma, p. 966 and 1332 (1998).

Gebhardt, R. (2001). Anticholestatic activity of flavonoids from artichoke and their metabolites. *Medical Science Monitor, 7*, 316–320.

Gebhart, R. (1997). Antioxidative and protective properties of extracts from leaves of the artichoke against hydroperoxide-induced oxidative stress in cultured rat hepatocytes. *Toxicology and Applied Pharmacology, 144*, 279–286.

Hammouda, F. M., El-Nasr, N. M. S., Ismail, S. I., & Shahat, A. A. (1993). Quantitative determination of the active constituents in Egyptian cultivated *Cynara scolymus*. *International Journal of Pharmacognosy, 31*, 299–304.

Hausler, M., Ganzera, M., Abel, G., Popp, M., & Stuppner, H. (2002). Determination of caffeoylquinic acids and flavonoids in *Cynara scolymus* L. by high performance liquid chromatography. *Chromatographia, 56*, 407–411.

Jimenez-Escrig, A., Gragsted, L. O., Daneshvar, B., Pulido, R., & Saura-Calixto, F. (2003). In vitro antioxidant activities of edible artichoke and effect on biomarkers of antioxidants in rats. *Journal of Agricultural and Food Chemistry, 51*, 5540–5545.

Lanteri, S., Saba, E., Cadinu, M., Mallica, G. M., Baghino, L., & Portis, E. (2004). Amplified fragment length polymorphism for genetic diversity assessment in globe artichoke. *Theoretical and Applied Genetics, 108*, 1534–1544.

Llorach, R., Espin, J. C., Tomas-Barneran, F. A., & Ferreres, F. (2002). Artichoke (*Cynara scolymus* L.) byproducts as a potential source of health-promoting antioxidant phenolics. *Journal of Agricultural and Food Chemistry, 50*, 3458–3464.

Lopez, G., Ros, G., Rincon, F., Periago, M. J., Martinez, M. C., & Ortuno, J. (1997). Mineral composition of isolated fibre fractions from artichoke and the effect of phosphate buffer on its structure and mineral content. *Food Chemistry, 60*, 541–547.

Perez-Garcia, F., Adzet, T., & Canigueral, S. (2000). Activity of artichoke leaf extract as reactive oxygen species in human leukocytes. *Free Radical Research, 33*, 661–665.

Saenz Rodriguez, T., Garcia Gimenez, D., & De La Puerta Vazquez, R. (2002). Choleretic activity and biliary elimination of lipids and bile acids induced by an artichoke leaf extract in rats. *Phytomedicine, 9*, 687–693.

Schutz, K., Kammerer, D., Carle, R., & Schieber, A. (2004). Identification and quantification of caffeoylquinic acids and flavonoids from artichoke (*Cynara scolymus* L.) heads, juice, and pomace by HPLC-DAD-ESI/MS. *Journal of Agricultural and Food Chemistry, 52*, 4090–4096.

Wang, M., Simon, J. E., Aviles, I. F., He, K., Zheng, Q. Y., & Tadmor, Y. (2003). Analysis of antioxidative phenolics compounds in artichoke. *Journal of Agricultural and Food Chemistry, 51*, 601–608.