Food and Nutrition Sciences, 2017, 8, 457-464
http://www.scirp.org/journal/fns
ISSN Online: 2157-9458
ISSN Print: 2157-944X

Nutrients and Bioactive Molecules of the *Early* and *Late* Cultivars of the Treviso Red Chicory (*Cichorium intybus* L.)

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How to cite this paper: D'Evoli, L., Lucarini, M., del Pulgar, J.S., Aguzzi, A., Gabrielli, P., Azzini, E. and Lombardi-Boccia, G. (2017) Nutrients and Bioactive Molecules of the *Early* and *Late* Cultivars of the Treviso Red Chicory (*Cichorium intybus* L.). *Food and Nutrition Sciences*, **8**, 457-464.
https://doi.org/10.4236/fns.2017.85031

Received: January 11, 2017
Accepted: May 7, 2017
Published: May 10, 2017

Abstract

The study provides original data on the compositional profile (macronutrients, dietary fiber, mineral and trace elements, bioactive molecules) of an Italian typical plant foods, *Treviso Red Cichory,* studying the two cultivars (*Early, Late*) grown following two different traditional cultivation systems. For two consecutive years plants from three growing areas, were studied. Major, significant differences, between *Early* and *Late* cultivars, were observed in minerals (Ca, Mg, P), trace elements (Fe, Zn) and bioactive molecules content. The *Late* cv. was found the richest in minerals and trace elements content than the *Early* cv., but for Ca. *Treviso Red Cichory* was found a valuable source of bioactive molecules such as ascorbic acid, anthocyanins and total polyphenols. The *Early* cv. showed a significantly higher ($p < 0.05$) ascorbic acid and total anthocyanins content (8.63 and 92.15 mg/100g, respectively) than the *Late* cv. (6.15 and 24.38 mg/100g, respectively), by contrast total polyphenols content was significantly higher ($p < 0.05$) in the *Late* cv. A marked variability in bioactive molecules content among the growing areas was found only for the *Late* cv. The observed differences between the *Early* and *Late* cultivars could be strictly related to the traditional growing systems applied during plant's growth.

Keywords

Treviso Red Cichory, Ascorbic Acid, Anthocyanins, Polyphenols, Minerals, Dietary Fiber

1. Introduction

Italy has a wide range of local plant foods that evoke cultural traditions strongly representative of cultural realities of specific territories. The enhancement of local foods production has increasingly becoming a key component for the improvement of an agro-food system strongly linked to the history and culture of the specific geographical area to which these foods belong. The cultivation of local ecotypes has also a strategic importance for both germoplasm and biodiversity preservation. An example is the ancient typical Italian red leafy plants belonging to the chicory family (*Cichorium intybus* L.) locally named "Radicchio", the *Treviso Red Cichory.* This local plant food, typical of the North-East Italy, received the attribution of Protected Geographical Indication (PGI) according to EU rules Council Regulation (2081/92) (http://eur-lex.europa.eu) for their peculiar cultivation system and the nutritional and organoleptic characteristics. *Treviso Red Cichory,* named after the Italian town where it comes from, is one of the most distinctive Italian vegetables, it is characterized by elongated red leaves with white ribs and by a typical slightly bitter taste. Two cultivars of *Treviso Red Cichory*, named *Early* and *Later,* are still cultivated according to different traditional growing systems that involve leaf tying during growth; this induce a bleaching process and give rise to the typical red leaves. The *Early* cultivar is harvested in autumn; the *Late* cultivar, after the harvest, is placed in vats filled with resurgent running water where new shoots appear, after about twenty days the plants are harvested. *Treviso Red Cichory* is particularly resistant to low temperature, and it is grown and consumed during the fall and winter seasons.

Data on the qualitative and quantitative composition of nutrients and bioactive compounds generally refer almost exclusively to a common cultivar (*Radicchio of Chioggia*) used in an extensive crops while, to date, there are no systematic studies representing the compositional figure of the two typical cultivars (*Early, Late*) of the *Treviso Red Cichory*. The red color of the leaves of the two *Treviso Red Cichory* ecotypes originated by the synthesis of anthocyanin pigments during growth [1] [2]. D'Evoli *et al.* [2] found cyanidin-3-*O*-(6"malonyl)-glucoside the predominant anthocyanin in *Treviso Red Cichory*. Studies carried out on the same ecotypes demonstrated that anthocyanins exerted a high peroxyl radical scavenging activity [3], furthermore D'Evoli *et al.* [4] highlighted a direct scavenging effect against ROS formation in terms of antioxidant, cytoprotective activities and antiproliferative activity in Caco-2 cell exerted by the red part of the leaf compared with the whole leaf. Azzini *et al.* [5] found beneficial effects of polyphenol-rich extracts from *Treviso Red Cichory* in counteracting the oxidative stress and cellular damage in *in vitro* Caco-2 cells model. Furthermore, *Treviso Red Cichory* also received attention because of its lower nitrate content compared to other leafy vegetables. Lucarini *et al.* [6] reported a lower nitrate content in *Treviso Red Cichory* compared to other leafy vegetables suggesting a great influence of the cultivation methods developed for the cultivation of this plant on nitrogen metabolism.

This study was addressed to characterize the compositional profile of *Treviso Red Cichory,* studying the two cultivars (*Early* and *Later*) commonly consumed in Italy. Plants were collected, for two consecutive years, in three different areas of the largest PGI production area and analysed for macronutrients (moisture, ash, protein, lipid, carbohydrates), total dietary fiber, minerals (Ca, Mg, K, P, Na) and trace elements (Fe, Zn, Cu, Mn) content. The study also aimed to evaluate the influence of the two different cultivation methods applied for the growth of the *Early* and *Late* cultivars on the concentration of some bioactive molecules, such as ascorbic acid, total anthocyanins and total polyphenols.

2. Materials and Methods

2.1. Materials

Treviso Red Cichory (*Cichorium intybus* L.) has intense red coloured leaves and white ribs, during the plant growth leaves are tied together. Two cultivars of *Treviso Red Cichory* were studied: *Early* cv. which was harvested in field on autumn; *Late* cv. which was removed from field in late autumn and then transferred in nylon covered "bleaching tanks" with roots bathed in circulating springwater, no fertilizer treatments were applied until plants were harvested after at least 20 days when new roots and new leaves sprouted up.

2.2. Methods

The plants of both the cultivar (*Early* and *Late* cv.) of the *Treviso Red Cichory* were collected for two consecutive years from three respective growing areas: Scorzè, Quinto, Zero Branco. For two consecutive years, 5Kg of plants from each selected area were collected at the harvesting time and delivered to the laboratory. Only the edible portion of the plants was utilised for analysis. Plants from each growing area (500 g) were weighed, washed in distilled water and dried with filter paper. The collected samples were freeze-dried before analysis, or immediately analysed. The analyses were carried out in triplicate. Each experimental data refers to two growing years, a total of six analyses for each area were performed.

Proximate composition*:* Moisture, protein, lipid and ash were determined according to AOAC methods [7].

Carbohydrates: carbohydrates were quantified by High-Performance Anion-Exchange Chromatography [8].

Total dietary fiber*:* Total dietary fiber was determined following the method of Prosky *et al.* [9].

Minerals and Trace Elements*:* Samples were analyzed for mineral (Ca, Mg, Na, K, P) and trace element (Fe, Zn, Cu, Mn) contents by ICP-OES (Optima 3200XL-Perkin-Elmer) after liquid ashing (4 mL HNO_3 + 1 mL H_2O_2) of the samples in a microwave digestion system (Milestone, 1200 Mega). Standard Reference Materials: Cabbage (IAEA-359, International Atomic Energy Agency Reference Materials Group) and Haricots vert (BCR 383, Community Bureau of Reference, Brussels) were analyzed as a check on the accuracy of the analysis.

Ascorbic acid: Ascorbic acid was determined by RP-HPLC (Waters 996, PAD detector) as described by Valls *et al.* [10].

Total Anthocyanins*:* About 100 g of Treviso red chicory were homogenized with cold methanol (HCl 0.1%) using an Ultra Turrax homogenizer for 5 min.

The extraction was repeated until the residue was uncoloured. Subsequently, homogenates were filtered through a Whatman paper under vacuum, and the MeOH in the filtrate was evaporated at 35˚C. Total anthocyanin content was quantified by RP-HPLC (Waters 996, PAD detector) [11].

Total Polyphenols*:* Total polyphenols content was determined following the method by Singleton *et al.* [12]. Gallic acid was used as standard phenol and results expressed as milligrams of gallic acid equivalents (GAE) per 100 g.

2.3. Statistics

All experimental data are presented as the Mean ± Standard Deviation. Statistical analysis was performed utilizing the Student's *t*-test; the effect was considered significative at $p < 0.05$. Data were statistically processed by XL-STAT software.

3. Results

The macronutrients profile and total dietary fiber content of the *Early* and *Late* cultivars of *Treviso Red Chicory* is reported in Table 1*.* Both the cultivars of the *Treviso Red Chicory,* as all horticultural products, showed a high water content with an average value of about 92.5%. The ash content was found significantly higher ($p < 0.05$) in the *Early* cv. compared with the *Late* cv. (Table 1)*.*

The protein content of the *Early* cv. showed not significant differences among the three cultivation areas. By contrast the protein content in the *Late* cv. was significant different ($p < 0.05$) among the three cultivation areas, with the highest value in Scorzè area followed by Zero Branco and Quinto areas (Table 1). The differences in protein content was not significant between the two cultivars studied (Table 1). Lipid content was negligible, while charbohydrates, resulted about 2.9 g/100 g in both the cultivars (Table 1). Carbohydrates content resulted significantly higher ($p < 0.05$) in the *Late* cv. compared to the *Early* one; no substantial differences in their content was observed among the growing areas for each cultivar (Table 1). The quantitative analysis of total dietary fiber showed similar amounts in both the *Early* and the *Late* cultivars

Table 1. Moisture, ash, protein, lipid charbohydrates and total dietary fiber in the *Early* and *Late* cultivars of *Treviso Red Chicory* (*Cichorium intybus* L.) (mg/100 g) (f.w.).

	Early cv.			*Early cv.*	*Late cv.*			*Late cv.*
	Quinto	Scorzè	Zero Branco	*Mean value*	Quinto	Scorzè	Zero Branco	*Mean value*
Moisture	92.4 ± 0.3	91.5 ± 0.06	92.4 ± 0.2	92.1	93.6 ± 0.5	92.1 ± 1.1	93.2 ± 051	92.9
Ash	0.71 ± 0.04a	0.61 ± 0.13b	0.66 ± 0.11	0.66A	0.55 ± 0.05	0.56 ± 0.12	0.56 ± 0.05	0.56B
Protein	1.54 ± 0.06	1.77 ± 0.22	1.63 ± 0.07	1.65	1.20 ± 0.1c	1.78 ± 0.25a	1.46 ± 0.05b	1.48
Lipid	0.1 ± 0.01	0.1 ± 0.01	0.1 ± 0.01	0.10	0.1 ± 0.01	0.1 ± 0.01	0.1 ± 0.01	0.10
CHO*	1.13 ± 0.04	0.93 ± 0.05	0.821 ± 0.02	0.96B	1.45 ± 0.31	1.55 ± 0.2	1.32 ± 0.2	1.44A
Total Fiber	2.50 ± 0.2	2.40 ± 0.1	2.50 ± 0.5	2.40	1.76 ± 0.1	2.58 ± 0.1	1.97 ± 0.2	2.10

Values are the M ± SD of two growing years. Values in the same row (among growing areas) followed by different small letters are statistically significant ($p < 0.05$). Values (between *Mean values*) in the same row followed by different capital letters are statistically significant ($p < 0.05$). *From "Banca dati BIOVITA"-ISBN 978-88-96597-02-6.

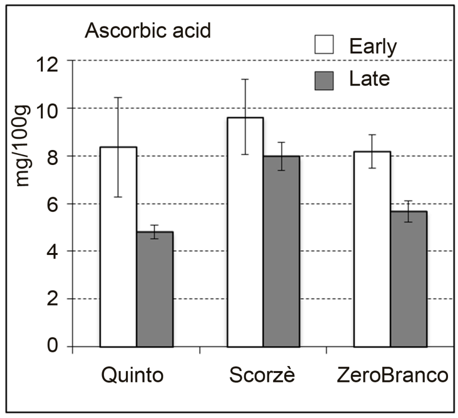
(2.4 and 2.1 mg/100g, respectively). No differences minerals and trace elements content within the growing areas of each cultivar were found (Table 2); on the other hand major differences were observed between the *Early* and *Late* cultivars (Table 2). Among minerals, Ca content was significantly higher ($p < 0.05$) in the *Early* cv. compared to the *Late* cv. (23.2 and 15.0 mg/100g, respectively) (Table 2). By contrast Mg and especially P content was significangtly higher in the *Late* cv., P was twice that found in the *Early* cv. (53 and 25.7 mg/100 g, respectively) (Table 2). The differences observed in both K and Na content between the cultivars were not significant. Among trace elements, significant differences ($p < 0.05$) in both Fe and Zn content between the two cultivars were found, the *Late* cv. having the highest value compared with the *Early* cv. (Table 2).

The study of bioactive molecules of nutritional interest in *Treviso Red Chicory* included the analysys of ascorbic acid, total anthocyanins and total polyphenols content. Ascorbic acid content in both the cultivars of *Treviso Red Chicory* from the three growing areas is reported in Figure 1(a). The mean value of the ascorbic acid content in *Treviso Red Chicory* was 8.6 mg/100g for the *Early* cv. grown in open field, a value significantly higher ($p < 0.05$) than that found for the *Late* cv. (6.15 mg/100g). The effect of the growing areas on ascorbic acid content was not significant for the *Early* cv. (Figure 1(a)). On the other hand, the *Late* cv. showed significant differences ($p < 0.05$) in ascorbic acid content among the growing areas: Scorzè area showed the highest values (7.15 mg/100g) with respect to Zero Branco (5.67 mg/100g) and Quinto (4.81 mg/100g) areas (Figure 1(a)). Total anthocyanins content in both the cultivars of *Treviso Red Chicory* from the three respective growing areas is reported in Figure 1(b). As previously found for ascorbic acid, also for total anthocyanins content the mean value for the three growing areas was found significantly higher

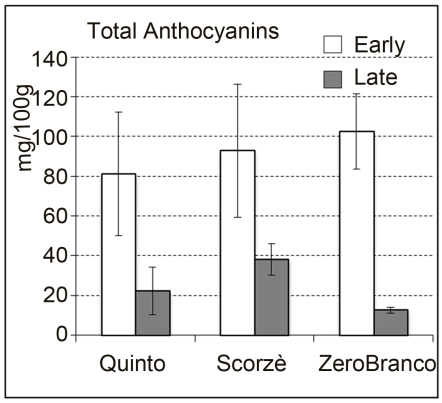
Table 2. Minerals and trace elements content (two cultivation years) in the *Early* and *Late* cultivars of *Treviso Red Chicory* (*Cichorium intybus* L.) (mg/100g) (f.w.).

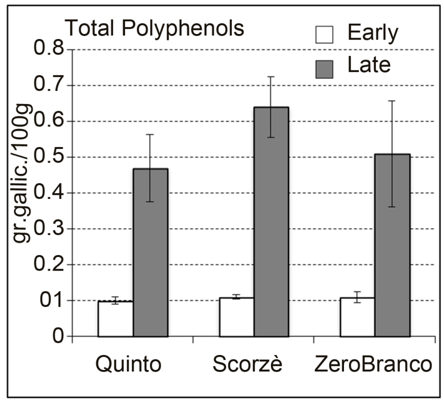
	Early cv.			*Early cv.*	*Late cv.*			*Late cv.*
	Quinto	Scorzè	Zero Branco	*Mean value*	Quinto	Scorzè	Zero Branco	*Mean value*
Ca	23.9 ± 5.5	22.9 ± 6.2	22.7 ± 2.9	23.2a	13.8 ± 5.5	14.5 ± 5.8	16.8 ± 2.1	15.0b
Mg	11.6 ± 3.9	11.7 ± 4.3	10.7 ± 2.5	11.3b	17 ± 8.4	16 ± 6.2	18 ± 7.7	17.0a
K	253 ± 36	246 ± 12	261 ± 20	260	380 ± 108	383 ± 118	354 ± 93	372
P	25.7 ± 2.6	26.0 ± 7.6	25.5 ± 0.4	25.7b	52.8 ± 19	53.4 ± 13	53.2 ± 15	53.0a
Na	9.2 ± 8.0	11.9 ± 6	6.6 ± 3.1	9.30	4.94 ± 1.8	7.4 ± 2.7	8.3 ± 5.5	6.90
Fe	0.42 ± 0.09	0.39 ± 0.03	0.48 ± 0.01	0.43b	0.50 ± 0.14	0.52 ± 0.2	0.58 ± 0.05	0.53a
Zn	0.15 ± 0.02	0.17 ± 0.04	0.15 ± 0.05	0.10ba	0.21 ± 0.16	0.30 ± 0.1	0.27 ± 0.08	0.18a
Cu	0.14 ± 0.06	0.12 ± 0.01	0.11 ± 0.03	0.12	0.14 ± 0.06	0.18 ± 0.01	0.18 ± 0.01	0.17
Mn	0.09 ± 0.03	0.08 ± 0.01	0.13 ± 0.04	0.10	0.16 ± 0.06	0.10 ± 0.01	0.12 ± 0.01	0.13

Values are the M ± SD of two growing years. Values (between Mean values) in the same row followed by different letters are statistically significant ($p < 0.05$).



(a)





(b) (c)

Figure 1. Ascorbic acid, total Anthocyanins and Total polyphenols content in the *Early* and *Late* cultivars of *Treviso Red Chicory* (*Cichorium intibus* L.) (f.w.). Values are the M ± SD of two growing years.

($p < 0.05$) in the *Early* cv., than in the *Late* cv. (92.1 and 24.4 mg/100g, respectively). Also in this case the *Early* cv. did not show significant differences in total anthocyanins content among the three growing areas (**Figure 1(b)**). By contrast significant differences ($p < 0.05$) in total antocyanins content among the three growing areas of the *Late* cv. were observed: plants from Scorzè area showed the highest total anthocyanins content (38.15 mg/100g) compared to both Quinto and Zero Branco areas (22.4 and 12.6 mg/100g, respectively) (**Figure 1(b)**).

A previous HPLC and NMR study (D'Evoli *et al.*, 2012) carried out on the same *Treviso Red Chicory* cultivars described cyanidin-3-*O*-(6"malonyl)-glucoside as the main anthocyanin in the cultivars, contributing with 70% of total anthocyanins. Total polyphenols content in *Treviso Red Chicory* is reported in Figure 1c. A marked difference in total polyphenols content between the two cultivars of the *Treviso Red Chicory* was found. The *Early* cv. had a very low content in total polyphenols (mean value 0.11 g gallic ac./100 g) compared with *Late* cv. where a mean value of 0.54 g gallic ac./100 g was found ($p < 0.05$) (**Figure 1(c)**). Furthermore, whilst the total polyphenols content was very similar in the three areas of the *Early* cv., the *Late* cv. showed a high variability in total polyphenols content among the three growing areas, it ranged from 0.47 to 0.64 g gallic ac. /100 g, a significant difference ($p < 0.05$) was observed only between Scorzè and Quinto areas (**Figure 1(c)**). Azzini *et al.* (2015) found, among flavonols, a significantly higher Kaempferol content in the *Late* cv. compared with the *Early* one.

4. Conclusion

Our findings provide a fingerprint of the two cultivars, *Early* and *Late*, of the *Treviso Red Chicory* (*Chicorium intibus* L.), that is a basic element for both recognition of cultivars and market competitiveness. The daily consumption of these traditional red-leafy cultivars significantly contributes to the dietary intake of relevant minerals, trace elements, dietary fiber and of some bioactive molecules, such as ascorbic acid, anthocyanins and total polyphenols. The *Early* and *Late* cultivars of the *Treviso Red Chicory* showed substantial differences in the content of these molecules, the *Early* cv. was the richest in anthocyanins and ascorbic acid, whereas the *Late* cv. was richest in total poplyphenols. The *Late* cv. was the only showing a high variability in the concentration in these molecules among the three growing areas. These differences between the two cultivars (*Early* and *Late*) could be strictly related to the traditional growing systems applied during plant's growth, the different approach may greatly influence, other than the minerals and trace elements content, the synthesis of the bioactive molecules analysed. This effect was noticeable in the *Late* cv., grown in the last months in spring-water without any further fertilization. Further studies are needed to elucidate the contribution of these peculiar cultivation systems to the high variability in bioactive molecules content observed.

Acknowledgements

This study was funded by Italian Ministry of Agriculture, Food, and Forestry Policies (MiPAAF), Research Project "BIOVITA".

References

[1] Lante, A., Nardi, T., Zocca, F., Giacomini, A. and Corich, V. (2011) Evaluation of Red Chicory Extract as a Natural Antioxidant by Pure Lipid Oxidation and Yeast Oxidative Stress Response as Model System. *Journal of Agriculture and Food Chemistry*, **59**, 5318-5324. https://doi.org/10.1021/jf2003317

[2] D'Evoli, L., Lucarini, M., Valentini, M., Ritota, M., Sequi, P. and Lombardi-Boccia, G. (2012) Anthocyanins Profile of Two Italian *Cichorium intybus* L. Cultivar. *Acta Horticulture*, **939**, 337-343. https://doi.org/10.17660/ActaHortic.2012.939.44

[3] Rossetto, M., Lante, A., Vanzani, P., Spettoli, P., Scarpa, M. and Rigo, A. (2005) Red Chicories as Potent Scavengers of Highly Reactive Radicals: A Study on Their Phenolic Composition and Peroxyl Radical Trapping Capacity and Efficiency. *Journal of Agriculture and Food Chemistry*, **53**, 8169-8175. https://doi.org/10.1021/jf051116n

[4] D'Evoli, L., Lombardi-Boccia, G., Lucarini, M., Morroni, F., Hrelia, P., Cantelli-Forti, G. and Tarozzi, A. (2013) Red Chicory (*Cichorium intybus* L. Cultivar) as a Potential Source of Antioxidant Anthocyanins for Intestinal Health. *Oxidative*

Medicine and Cell Longevity, **2013**, Article ID: 704310.

[5] Azzini, E., Maiani, G., Garaguso, I., Polito, A., Foddai, M.S., Venneria, E., Durazzo, A., Intorre, F., Palomba, L., Rauseo, M.L., Lombardi-Boccia, G. and Nobili, F. (2016) The Potential Health Benefits of Polyphenol-Rich Extracts from *Cichorium intybus* L. Studied on Caco-2 Cells Model. *Oxidative Medicine and Cell Longevity*, **2016**, Article ID: 1594616.

[6] Lucarini, M., D'Evoli, L., Tufi, S., Gabrielli, P., Paoletti, S., Di Ferdinando, S. and Lombardi-Boccia, G. (2012) Influence of Growing System on Nitrate Accumulation in Two Varieties of Lettuce and Red Radicchio of Treviso. *Journal of the Science of Food and Agriculture*, **92**, 2796-2799. https://doi.org/10.1002/jsfa.5526

[7] AOAC (2012) Official Methods of Analysis. 19th Edition, Association of Official Analytical Chemists, Arlington, VA.

[8] Lee, Y.C. (1996) Carbohydrate Analyses with High-Performance Anion-Exchange Chromatography. *Journal of Chromatography A*, **720**, 137-149.

[9] Prosky, L., Asp, N.G., Schweizer, T.F., De Vries, J.W. and Furda, I. (1988) Determination of Insoluble, Soluble and Total Dietary Fiber in Foods and Food Products: Interlaboratory Study. *Association of Analytical Chemistry*, **71**, 1017-1023.

[10] Valls, F., Rancho, M.T., Fernandez-Muino, M., Alonso-Torre, S. and Chec, M.A. (2002) High Pressure Liquid Chromatography Determination of Ascorbic Acid in Cooked Sausages. *Journal of Food Protein*, **65**, 1771-1774. https://doi.org/10.4315/0362-028X-65.11.1771

[11] Wang, S.Y. and Wang, W.Z. (2001) Effect of Plant Growth Temperature on Antioxidant Capacity in Strawberry. *Journal of Agriculture and Food Chemistry*, **49**, 4977-4982. https://doi.org/10.1021/jf0106244

[12] Singleton, V.L., Orthofer, R. and Lamuela-Raventós, R.M. (1999) Analysis of Total Phenols and Other Oxidation Substrates and Antioxidants by Means of Folin-Ciocalteu Reagent. *Methods in Enzymolology*, **299**, 152-178.