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Antioxidants and Mineral Contents of Chicory as Coffee Additive

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ABSTRACT

In this study, roots of Turkish origin wild chicory *(Cichorium intybus* L.) genotypes were investigated for total flavonoid and phenolic contents, radical cation scavenging activity (ABTS), Free radical scavenging activity (DPPH), and radical, mineral content. These characteristics were also compared with other coffee varieties. The total flavonoid and phenolic contents ranged between 0.290-4.350 mg QE/g dry weight (DW) and 0.943-13.860 mg GAE/g DW. The DPPH was listed here from high to low value: raw coffee beans = roasted coffee beans > roasted fruits of turpentine tree > instant coffee = roots of chicory. The content of P, Ca, Mg, Zn, B, Cr, Co and Mo ranged between 0.71-2.78%, 0.25-0.46%, 7.29-20.66, 4.44-11.07, 0.40-1.67, 0.49-5.48 and 5.69-14.46 ppm, respectively. As a result, chicory roots exhibited low antioxidant activity, but higher mineral content compared to the other tested coffee varieties which indicates that chicory could be used a coffee additive.

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Introduction

Cichorium sp. genus is widely distributed in Africa, Asia, Europe, Australia, Northern America, and Southern America. *Cichorium intybus* L. and *Cichorium endivia* L. are mainly cultivated. *C. intybus* differs from *C. endivia* in terms of short capsules and persistency. *C. intybus* is common species compared to *C. endivia*. *C. intybus* is extremely tolerant to high temperature and drought due to its capability of elongation, and protecting its greenery during the summer and capability of growing without being watered in marginal areas (Kiers et al., 1999).

C. intybus is cultivated as a medicinal plant. Judzentienne and Udien (2008) reported that chicory roots contain 40% inulin. Its compounds such as, bitter sesquiterpene lactones, coumarins, flavonoids and vitamins are also used as anti-hepatotoxic, anti-inflammatory, liver tonic, cholagogue, depurative, diuretic, emmenagogue, alexeteric, and used as tonic, anticancer and other medicinal purposes (Franck, 2002; Nandagopal and Ranjitha 2007; Denev et al., 2012).

Previous studies showed that chicory roots have constituents such as caffe-oylquinic acid, quercedina, anti-diabetic, antihepatotoxic, anti-inflammatory and antioxidant activities (Ahmad et al., 2002; Schumacher et al., 2011; Ghamarian et al., 2012; Jurgonski et al., 2012; Kaskos, 2012). On the other hand, chicory roots have been considered as coffee additives since ancient times. Chicory coffee which is caffeine-free is the source of plant phenolics, and phenolic content is correlated with antioxidant activity (Lavelli, 2008). Chicory coffee is a source of caffeic acid and a healthy person is recommended to consume 300 ml chicory coffee per week (Schumacher et al., 2011). Besides, the plant has anti-inflammatory, antiviral and anti-cancer characteristics.

The coffee contains significant nutrients vital for energy and health, but the biggest problem in coffee is additives. These substances are not organic and create health risk (Yiteyal and Tilahun, 2017). Therefore, there is a need for additives of plant origin. Chicory is used as

coffee additives, preservatives, and digestive stimulant action. The aim of this study is to determine the antioxidant composition and mineral contents of wild chicory roots collected from 27 different locations in Turkey, and to compare with commercial or traditional coffee varieties (roasted fruits of turpentine tree, instant coffee, roasted coffee beans, and raw coffee beans).

Material and Methods

In this study, roots of *C. intybus* "Chicory" genotypes, roasted fruits of turpentine tree (*Pistacia terebinthus* L.), instant coffee, roasted coffee beans and raw coffee beans were used. (The coffee varieties used as control were provided from markets). The roots of chicory genotypes were obtained from the field experiments in Yozgat/Turkey. Initially, chicory seeds were collected from 27 different locations of Turkey (Table 1) in 2014.

Seeds were sown in peat media and were transferred to the field with 50x50 m of dimension distance in May 2015 in Yozgat/Turkey, and roots were harvested at the end of October, the same year. The experimental area soil gathered from 0-30 cm deep included pH of 8.20, 7.93% CaCO3, 86.2 kg ha^{-1} phosphorus, 484.7 kg ha^{-1} potassium, and 1.91% organic matter. Mean, long term annual precipitation of Yozgat is about 574.4 mm and the mean temperature is 9.0 °C. Mean rainfall of Yozgat throughout 2015 (717.1 mm) was higher than the precipitation means for a long time. Mean temperature in growing season in 2015 was 10.0 °C (Anonoymous, 2019).

After the harvesting, the roots were cleaned and roasted at 140 °C temperature for 2 hours, then crushed using a conventional method. Roasted fruits of turpentine tree (*P. terebinthus*), instant coffee, roasted coffee beans and raw coffee beans were provided from commercial coffee suppliers and analyzed for same traits.

Harvested, dried and finely ground samples (weighing about 5g per plant) of chicory roots, roasted fruits of turpentine tree, instant coffee, raw and roasted coffee beans were extracted in methanol at 40 °C for 24 h. The mixtures were filtered through Whatman paper and, methanol was separated with a rotary evaporator (Heidolph, laborota 4000) to obtain extract yields. Then, extracts were dissolved in methanol. Besides, different ratio of the roasted coffee bean and chicory root extracts (1:4, 2:3, 3:2, 4:1) were prepared as total 1000 µg mL^{-1} in methanol for synergistic activity.

The total phenolic contents of samples were determined with slight modification according to the Folin-Ciocalteu reagent (FCR) method of Singleton et al. (1999). Samples (200 µL) were mixed with diluted FCR (200 µL) and shaken vigorously for 3 min. Then, 600 µL Sodium Carbonate (Na_2CO_3) solutions (20%) were added, and absorbance of each sample was measured at 760 nm after incubating in dark at room temperature for 2 h. The total phenolic contents were expressed as mg equivalents of Gallic acid (GAE) g^{-1} dry weight (DW) according to the equation obtained from the standard Gallic acid graph and calculated from the calibration curve (R^2= 0.9994).

Total flavonoid content of each sample was determined with a method which was partially modified and adopted by Arvouet-Grand et al. (1994). Briefly, each sample (500 µL) were mixed with 100 µL of aluminum nitrate (10%) and 100 µL of potassium acetate (1 M). Total volume of the solution was adjusted to 5mL with ethanol. Similarly, a blank was prepared by adding methanol in place of sample. Absorbance measurements were read at 417 nm after 40 min incubation at room temperature in dark conditions. Total flavonoid content was expressed as mg equivalents of quercetin (QE) g^{-1} DW according to the equation obtained from the standard quercetin graph and calculated from the calibration curve (R^2= 0.9994).

The effect of each sample on 2.2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH) radical was identified according to Gezer et al. (2006). Two hundred microliter from each sample in methanol was added to 3.2 mL of 0.004% methanol solution of DPPH. Absorbance of each sample was read at 517 nm after 30 min incubation at room temperature in dark.

ABTS radical cation scavenging activity was estimated according to Miller et al. (1993) and Re et al. (1999) with a partial modification. $ABTS^+$ radical cation was obtained directly by reaction 30 mg ABTS with 6.6 mg potassium persulfate in 7.8 mL double-distilled water, and allowing the mixture to remain for 12-16 h in dark at the room temperature. Then, ABTS solution was diluted with bi-distilled water to an absorbance of 0.700±0.020 at 734 nm. Each sample in methanol (100 µL from 1000 µg mL^{-1}) was added to ABTS solution (2.8 mL) and mixed. A blank was prepared by adding methanol instead of sample solution. Absorbance of each sample was read at 734 nm after at 30 min incubation at room temperature.

One-gram powdered samples were burned at 550 ^{0}C then 4 ml 3 N HCL added and the researchers waited 30 minutes for the ash to subside. Ash solution was filtered and pure water was added until the solution became 50 ml. Phosphorus (P), calcium (Ca), magnesium (Mg), zinc (Zn), iron (Fe), manganese (Mn), sulphur (S), boron (B), chromium (Cr), cobalt (Co) and molybdenum (Mo) concentrations in chicory root. Coffees (roasted fruits of turpentine tree, instant coffee, roasted coffee beans and raw coffee beans) were determined by inductively coupled plasma mass spectrometry (ICP-MS) using a Thermo Scientific- iCAPQc (Bremen, Germany).

The data were analyzed using the statistical package SPSS 16.0 V. Probabilities less than 0.05 were considered significant. Duncan's multiple range tests was used to separate the treatment means. All significant main effects were considered.

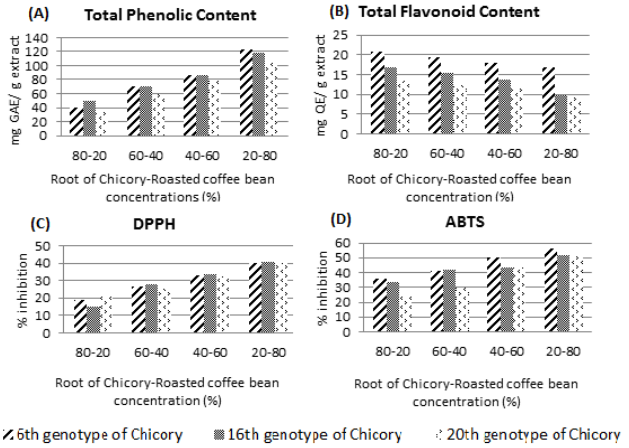

Figure 1. Total phenolic content (A), total flavonoid content (B), DPPH (C), ABTS (D) of mixtures (1 mg/mL) belong to variation ratio of roasted coffee bean and root of *C. intybus* (6^{th}, 16^{th} and 20^{th} genotype of Chicory)

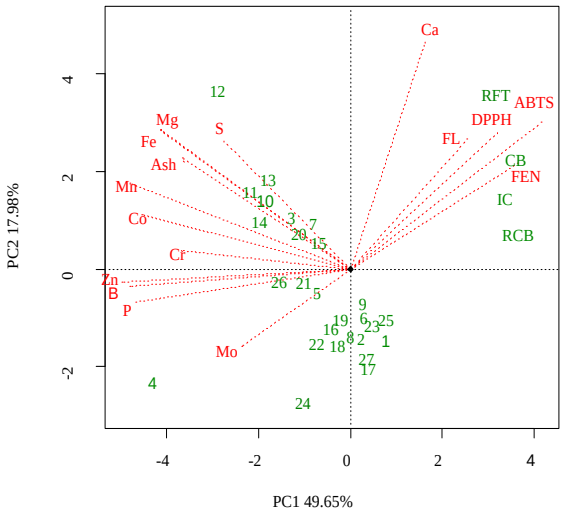

Figure 2. Principal component analysis of antioxidant properties and mineral contents of chicory roots and coffee varieties. 1-27, roots of chicory plants (*Cichorium intybus*) collected from different localities; RFT, roasted fruits of turpentine tree (*Pistacia terebinthus*); IC, instant coffee; RCB, roasted coffee beans; CB, raw coffee beans.

Result and Discussion

Total bioactive components in chicory roots (RCP), roasted fruits of turpentine tree (*P. terebinthus*) (RFT), instant coffee (IC), raw coffee beans (CB) and roasted coffee beans (RCB) showed the presence of phenolics and flavonoids, and results were compared with each other.

Total flavonoid content of root extracts of chicory varied from 0.300 mg to 1.146 mg QE/g DW, while total phenolic contents varied from 0.943 mg to 3.363 mg GAE g^{-1} DW (Table 1). It was also observed that there are significant differences among localities. The statistically highest total phenolic and flavonoid content in root extracts of chicory was emerged from extracts of the 15th and the 13rd genotypes (3.363 mg GAE g^{-1} DW- 1.120 mg QE g^{-1} DW and 3.533 mg GAE g^{-1} DW – 1.063 QE g^{-1} DW). The extract of the 18th genotype (0.943 mg QE g^{-1} DW - 0.296 mg QE g^{-1} DW) was lowest.

Chicory has rich polyphenols content (Heimler et al., 2009), therefore the total phenolic content in the roots of chicory was higher than the total flavonoids. Besides, coffee varieties compared and the highest total phenolic content was found out to be instant coffee (13.860 mg GAE g-1 DW), while the lowest was roasted coffee beans (4.433 GAE g-1 DW). It was reported that instant coffee contains 4 mg/g of chlorogenic acid as nitrosatable compound, and also other phenolic compounds such as catechol, caffeic acid (Duarte et al., 2000).

Table 1. Origin of the chicory genotypes under investigation.

Genotypes	Location	Latitude	Longitude
1	Nevşehir-Avanos	38° 42′ 33.81″N,	34° 50′ 50.54″E
2	Yozgat-Yerköy	39° 39′ 10.7028"N	34° 29′ 15.13"E
3	Samsun Central	41° 50′ 27.39″N	36° 06′ 43.98″E
4	Konya-Meram	37° 40' 24.89"N	32° 28' 08.23"E
5	Yozgat-Boğazlıyan	39° 24′ 20″N	35° 0′ 24″E
6	Yozgatlı-Central	39° 50′ 24.89″N	34° 51′ 58.77″E
7	Yozgat-Sarıkaya	39° 32′ 22.12″N	35° 15′ 15.70″E
8	Yozgat-Çandır	39° 14′ 36.15″N	35° 31′ 01.91″E
9	Konya-Kulu	39° 05′ 20.98″N,	33° 02′ 40.04″E
10	Yozgat-Yerköy	39° 38′ 53.82″N	34° 33′ 14.20″E
11	Yozgat-Yerköy	39° 38′ 20.45″N	34° 27′ 48.98″E
12	Yozgat-Central	39° 49′ 54.33″N	34° 48′ 32.43″E
13	Yozgat-Sorgun	39° 54′ 08.78″N	35° 02′ 12.17″E
14	Yozgat-Sarıkaya	39° 34′ 54.63″N	35° 25′ 43.97″E
15	Yozgat-Kadışehri	39° 57′ 22.28″N	35° 39′ 52.68″E
16	Yozgat-Yerköy	39° 40′ 57.65″N	34° 35′ 25.75″E
17	Yozgat-Boğazlıyan	39° 8′ 19.30.20"N	35° 22′ 41.22"E
18	Kırşehir-Kaman	39° 21′ 24.93″N	33° 41′ 51.41″E
19	Yozgat-Sarıkaya	39° 33′ 34.57″N	35° 23′ 11.62″E
20	Yozgat-Central	39° 48′ 16″N	34° 48′ 40″E
21	Yozgat-Boğazlıyan	39° 19′ 55″N	35° 08′ 35″E
22	Yozgat-Central	39° 49′ 26.25″N	34° 51′ 58.77″E
23	Antalya-Akseki	37° 02' 11.43"N	31° 46' 50.60"E
24	Yozgat-Central	39° 50′ 28.22″N	34° 48′ 22.05″E
25	Amasya-Merzifon	40° 52′ 06.65″N	35° 28′ 46.51″E
26	Yozgat-Sorgun	39° 50' 44.86"N	35° 65′ 44.68"E
27	Yozgat-Çandır	39° 14′ 47.68″N	35° 32′ 07″E

Table 2. Total bioactive compounds and radical scavenging activities of methanol extracts of roots of chicory, roasted fruits of turpentine tree, instant coffee, raw roasted coffee beans.

Genotypes	Total Flavonoid Contents (mg QE/g DW)	Total Fenolic Contents (mg GAE /g DW)	ABTS Radical Cation (% inhibition)	DPPH Radical (% inhibition)
1	0.396 klma	2.077 j-m	33.65 efg	8.437 cd
2	0.570 hij	1.873 k-p	28.64 ij	7.787 cd
3	0.636 ghi	2.423 hij	26.79 jk	7.607 cd
4	0.413 kl	2.870 fg	24.14 lm	7.223 cde
5	0.673 fg	1.703 l-p	20.83 n	2.810 gh
6	0.653 gh	3.203 ef	25.79 klm	8.507 cd
7	0.360 k-n	1.567 nop	24.99 klm	7.697 cd
8	0.336 k-n	2.527 ghi	13.36 p	8.327 cd
9	0.513 i	1.860 k-p	29.39 i	8.910 c
10	0.880 e	2.170 ijk	25.04 klm	5.290 ef
11	0.310 m	1.457 p	26.29 kl	7.200 cde
12	0.300 n	1.550 op	38.61 d	8.170 cd
13	1.063 d	3.533 e	33.30 fg	8.573 cd
14	0.426 k	1.983 j-o	20.43 n	8.077 cd
15	1.120 d	3.363 e	31.95 gh	8.417 cd
16	0.383 k-n	2.157 ijk	34.50 ef	nd
17	0.326 lmn	1.623 m-p	29.34 i	7.967 cd
18	0.296 n	0.943 q	16.63 o	3.823 fg
19	0.616 ghi	2.737 gh	35.77 e	1.620 h
20	0.390 k-n	2.137 i-k	23.59 m	8.370 cd
21	0.516 j	1.803 k-p	24.79 klm	8.057 cd
22	0.863 e	1.793 k-p	24.94 klm	nd
23	1.146 d	2.677 gh	26.69 jk	7.920 cd
24	0.570 hij	1.433 p	23.84 m	nd
25	0.526 ij	2.000 j-n	30.04 hi	8.010 cd
26	0.740 f	2.067 j-n	33.95 efg	6.680 de
27	0.353 k-n	1.423 p	26.64 jk	4.647 fg
RFT	4.350 a	5.550 c	88.63 a	13.727 b
IC	1.163 b	13.860 a	69.80 c	8.413 cd
RCB	0.567 hij	4.433 d	77.97 b	21.917 a
CB	1.443 c	7.090 b	76.26 b	20.913 a

In each column different letters indicate significant difference (P<0.01); nd: not determined; QE: Quercetin equivalents; GAE: Gallic acid equivalents; 1-27: Roots of chicory plants (*Cichorium intybus*) collected from different localities; RFT: Roasted fruits of turpentine tree (*Pistacia terebinthus*); IC: instant coffee; RCB: Roasted coffee beans; CB: Raw coffee beans.

According to previous studies, raw coffees beans have high polyphenol content, which is particularly rich in chlorogenic acid and related compound (Clifford 1999, Suzuki et al., 2002), reaching up to 14 % (dry matter basis). However, it has been determined that processing, especially roasting modifies the phenolic composition of coffee, producing aroma, flavor and color compounds characteristics of coffee beverage significantly (Farah and Donangelo, 2006), and this shows similarity with the present study. Besides, total flavonoid content of roasted coffee beans was less than raw coffee beans' similar to the total phenolic content. On the other hand, it was assumed that roasted fruits extract of turpentine tree had the highest total flavonoid content (4.350 mg QE g^{-1} DW).

All the assessed extracts for DPPH activity were able to reduce the initial stable blue/purple DPPH radical to a yellow DPPH-H (Cavar et al., 2012, Khadhri et al., 2017). In the present study, the DPPH radical scavenging activity was changed based on the genotypes, coffee varieties, raw-roasted coffee, and was listed here from high to low value: raw coffee beans = roasted coffee beans > roasted fruits of turpentine tree > instant coffee = roots of chicory. Unlike total phenolic and flavonoid contents, higher DPPH radical scavenging activities were obtained from raw and roasted coffee beans. The capability of different phenolic substances to scavenge various types of oxidation-initiating radicals has been reported, demonstrating different effects (Rive-Evans et al., 1996, Yen and Duh, 1994). For example; polyphenolic compounds have antioxidant activity in many studies (Okuda et al., 1994).

The ABTS free radical scavenging effects of all the tested samples of the plant were denoted in Table 2. The highest ABTS radical scavenging activity was found in extract of roasted fruits of turpentine tree (88.63%), indicating more accumulation of flavonoids in this extract. However, the lowest was found in extracts of roots of chicory. Raw and roasted coffee beans (76.26%, 77.97%, respectively) were statistically placed in the same group and exhibited more activity than instant coffee (69.80 %).

The total phenolic and flavonoid content, DPPH and ABTS free radical scavenging activities of mix extracts of roasted coffee bean and root of chicory (20-80, 40-60, 80-20 as % in 1 mg/ml extract) were given in Figure 1. It was determined that the total phenolic contents, DPPH and ABTS activities in the mix extract increased with increasing ratio of roasted coffee bean, but, the total flavonoid contents decreased. This result was observed in all investigated genotypes of chicory (6^{th}, 16^{th} and 20^{th} genotypes). Specific synergistic effect was not observed in mix extracts.

Table 3. Crude ash ratio and P, Ca, Mg (%), Zn, Fe (ppm) content of chicory, roasted fruits of turpentine tree, instant coffee, raw roasted coffee beans

Genotypes	Crude ash ratio	P content	Ca content	Mg content	Zn content	Fe content
1	4.75 $^{k-n}$	2.66 $^{f-j}$	0.18 $^{j-n}$	0.62 ^{klm}	13.40 ^{j}	19.93 ^{lmn}
2	4.66 $^{l-o}$	2.44 $^{h-k}$	0.20 $^{h-n}$	0.65 $^{i-m}$	18.70 $^{e-h}$	27.92 $^{h-k}$
3	5.54 ^{gh}	3.02 $^{c-g}$	0.27 $^{d-j}$	1.05 ^{bc}	19.51 $^{e-h}$	43.63 ^{e}
4	4.53 $^{n-q}$	2.96 $^{d-g}$	0.24 $^{e-m}$	0.76 $^{e-j}$	16.14 ^{hij}	26.84 ^{ijk}
5	4.93 $^{j-m}$	3.08 $^{c-f}$	0.27 $^{d-k}$	0.90 ^{de}	20.13 ^{efg}	37.49 ^{f}
6	4.49 $^{n-q}$	2.87 $^{d-h}$	0.20 $^{h-n}$	0.67 $^{h-m}$	14.95 ^{ij}	27.33 $^{h-k}$
7	9.16 ^{b}	2.61 $^{f-j}$	0.38 ^{bc}	0.76 $^{e-l}$	20.25 ^{efg}	52.95 ^{d}
8	5.60 ^{gh}	1.96 ^{kl}	0.17 $^{k-n}$	0.62 ^{klm}	16.88 $^{g-j}$	29.20 $^{g-j}$
9	5.10 ^{ijk}	2.53 $^{g-j}$	0.18 $^{j-n}$	0.81 $^{e-i}$	14.91 ^{ij}	34.70 ^{fg}
10	7.33 ^{d}	3.92 ^{ab}	0.32 $^{c-g}$	1.20 ^{ab}	25.15 ^{bc}	63.08 ^{c}
11	8.03 ^{c}	3.02 $^{c-g}$	0.34 ^{cd}	1.13 ^{abc}	24.36 ^{bcd}	71.06 ^{b}
12	9.95 ^{a}	2.73 $^{f-i}$	0.43 ^{b}	1.21 ^{a}	25.05 ^{bc}	92.52 ^{a}
13	6.83 ^{e}	3.29 ^{cde}	0.32 $^{c-f}$	1.17 ^{ab}	28.73 ^{a}	45.07 ^{e}
14	6.15 ^{f}	3.34 ^{cd}	0.33 ^{cde}	1.14 ^{ab}	27.79 ^{ab}	55.93 ^{d}
15	4.58 $^{m-p}$	3.34 ^{cd}	0.27 $^{d-k}$	0.99 ^{cd}	22.11 ^{cde}	32.29 $^{f-i}$
16	5.88 ^{fg}	2.23 ^{ijk}	0.23 $^{f-n}$	0.70 $^{g-m}$	21.96 ^{cde}	29.56 $^{g-j}$
17	4.67 $^{l-o}$	2.22 ^{ijk}	0.14 ^{n}	0.60 $^{k-n}$	17.45 $^{f-i}$	13.39 ^{opq}
18	5.52 ^{h}	2.24 ^{ijk}	0.20 $^{h-n}$	0.63 $^{j-m}$	19.51 $^{e-h}$	33.42 ^{fgh}
19	4.95 ^{ijk}	2.79 ^{fgh}	0.18 $^{i-n}$	0.79 $^{e-j}$	17.06 ^{ghi}	25.22 ^{jkl}
20	7.41 ^{d}	2.65 $^{f-j}$	0.28 $^{d-i}$	0.86 ^{def}	18.72 $^{e-h}$	54.27 ^{d}
21	4.33 $^{o-r}$	3.06 $^{c-f}$	0.26 $^{d-l}$	0.89 ^{de}	25.01 ^{bc}	28.08 $^{h-k}$
22	5.72 ^{gh}	2.82 $^{e-h}$	0.19 $^{h-n}$	0.61 $^{k-n}$	25.12 ^{bc}	23.30 $^{j-m}$
23	4.19 ^{qr}	2.41 $^{h-k}$	0.19 $^{h-n}$	0.70 $^{f-m}$	17.88 $^{f-i}$	25.58 ^{jkl}
24	5.13 ^{ij}	2.44 $^{h-k}$	0.16 ^{lmn}	0.60 ^{lmn}	20.08 ^{efg}	22.07 ^{klm}
25	5.42 ^{hi}	1.64 ^{lm}	0.16 ^{lmn}	0.58 ^{mn}	16.05 ^{hij}	24.91 ^{jkl}
26	4.78 $^{j-n}$	4.30 ^{a}	0.24 $^{e-m}$	0.84 $^{d-g}$	28.89 ^{a}	24.74 ^{jkl}
27	3.85 ^{t}	2.28 ^{ijk}	0.17 $^{k-n}$	0.73 $^{f-m}$	21.86 ^{cde}	15.48 ^{nop}
RFT	4.85 $^{j-n}$	0.69 ^{n}	0.54 ^{a}	0.83 $^{e-i}$	6.95 ^{l}	18.13 ^{mno}
IC	4.34 $^{o-r}$	0.55 ^{n}	0.40 ^{bc}	0.64 $^{j-ml}$	6.16 ^{l}	8.63 ^{qr}
RCB	4.23 ^{pqr}	0.37 ^{n}	0.28 $^{d-h}$	0.47 ^{no}	6.00 ^{l}	7.45 ^{s}
CB	2.61 ^{u}	1.22 ^{m}	0.61 ^{a}	0.35 ^{o}	10.02 ^{j}	23.23 $^{j-m}$

In each column different letters indicate significant difference (P<0.01); nd: not determined; QE: Quercetin equivalents; GAE: Gallic acid equivalents; 1-27: Roots of chicory plants (*Cichorium intybus*) collected from different localities; RFT: Roasted fruits of turpentine tree (*Pistacia terebinthus*); IC: instant coffee; RCB: Roasted coffee beans; CB: Raw coffee beans.

Crude ash ratio and mineral element contents (P, Ca, Mg, Zn, Fe, Mn, S, B, Cr, Co and Mn) in the chicory genotypes and coffees are presented in Table 3 and 4. The highest crude ash ratios (9.95%) was found in 12th genotype, while the lowest (2.61) was found in raw coffee beans. Kim et al. (1978) reported the crude ash ratio of chicory root ranged between 3.0-4.2%.

The content of P, Ca and Mg ranged between 1.22-4.30%, 0.14-0.61% and 0.35-1.21% respectively. Durrani et al. (2010) reported that potassium is an essential element for normal body growth, while Calcium is an important constituent of bones and teeth and it is actively involved in the regulation of nerve and muscle functions. It was reported that Ca and Mg content in instant coffee were 0.78% and 0.21% respectively (Demir et al., 2015). Yiteyal and Tilahun (2017) reported that the average amount of calcium and phosphorus required for infants is 230-187.5 mg/day, for children is 850-480 mg/day and for other age groups is 1150-975 mg/day. The present study shows that the Ca and P contents of chicory roots are sufficient. Previous researchers indicated that coffees made from other plants have less Mg than chicory (Suseela et al., 2001; Chaves et al., 2012; Samsonowicz et al., 2019).

The highest Fe and Mn contents were obtained from the 12th genotype (92.52 and 61.86 ppm, respectively), while the lowest were found in roasted coffee beans (7.45 and 4.38 ppm, respectively). Kim et al. (1978) reported the Fe content of chicory root ranged between 90-120 ppm. Yiteyal and Tilahun (2017) reported that Mn is necessary for the development and growth of organisms, and 0.6-2.6 mg of Mn should be consumed daily.

The content of B, Cr, Co and Mo ranged between 2.23-14.81 ppm, 0.15-3.99 ppm, 0.22-12.37 ppm and 4.28-20.47 ppm, respectively. Comparing the results obtained in this study with the data in the literature (Table 4), chicory root can be said to have higher cobalt content than others plant. (Suseela et al., 2001; Chaves et al., 2012; Samsonowicz et al., 2019).

The biplot graphic analyses of the 27 chicory genotypes and coffee varieties are shown in Figure 2. According to the antioxidant properties and mineral contents that sum of values in PCA 1 (49.65%) and PCA 2 (17.98%) were 67.63%. In the present study, the coffee varieties had higher values compared to the chicory genotypes in terms of the antioxidant properties, while chicory genotypes had higher mineral content compared to the coffee varieties. Besides, genotypes of 3th, 5th, 7th, 10th, 11th, 12th, 13th, 14th, 15th, 20th, 21th and 26th had higher mineral contents compared to the other chicory genotypes.

Table 4. Mn, S, B, Cr, Co and Mo content of chicory, roasted fruits of turpentine tree, instant coffee, raw roasted coffee beans (ppm)

Genotypes	Mn content	S content	B content	Cr content	Co content	Mo content
1	21.19 hij	745.7 i-l	10.501 f-k	0.158 o	2.281 hi	11.558
2	30.08 f	692.8 i-l	9.881 g-l	1.127 mn	1.286 ij	15.465
3	36.73 de	1596.4 a	13.400 abc	1.650 g-j	5.069 ef	20.473
4	18.88 ijk	700.5 i-l	12.502 b-e	1.638 g-k	3.168 gh	11.730
5	29.34 fg	919.5 f-i	12.962 bcd	1.569 h-l	4.320 fg	12.154
6	18.94 ijk	875.7 g-j	10.230 f-k	2.071 de	2.463 hi	11.209
7	36.56 de	633.8 kl	9.317 i-l	1.360 im	7.476 cd	14.256
8	21.32 hij	564.2 l	8.960 jkl	2.324 cd	7.680 cd	11.412
9	21.45 hij	640.4 kl	9.438 h-l	1.547 h-l	4.199 fg	8.630
10	46.80 c	1199.4 cde	11.599 c-g	1.716 fgh	8.835 bc	14.901
11	52.38 b	1003.7 e-h	11.954 b-f	2.560 c	9.787 b	12.876
12	61.86 a	1258.0 cd	11.979 b-f	3.579 b	12.379 a	12.534
13	49.67 bc	1347.5 bc	14.818 a	1.334 j-m	7.332 cd	16.745
14	51.52 bc	1082.8 d-g	13.187 a-d	1.951 efg	8.007 cd	16.863
15	29.38 fg	1088.0 d-g	13.097 a-d	2.407 c	4.748 fg	13.509
16	27.56 fg	717.6 i-l	10.713 e-j	1.689 f-i	6.939 d	12.575
17	21.63 hij	758.9 i-l	9.386 i-l	1.960 efg	3.171 gh	10.031
18	31.71 ef	794.1 h-l	9.677 h-l	0.848 o	6.536 de	10.379
19	30.43 f	736.5 i-l	9.297 i-l	1.593 h-k	6.951 d	13.669
20	38.64 d	821.5 h-k	11.360 d-h	1.253 lm	7.101 d	11.355
21	24.26 ghi	1095.2 def	13.510 ab	3.995 a	3.462 fgh	15.160
22	36.68 de	736.6 i-l	11.071 e-i	1.585 h-k	4.564 fg	13.643
23	24.33 ghi	573.0 l	10.112 f-k	1.051 mn	4.412 fg	11.037
24	26.49 fgh	748.9 i-l	9.484 h-l	1.520 h-l	6.312 de	49.388
25	16.76 jkl	749.5 i-l	8.556 kl	1.935 efg	3.297 gh	9.332
26	53.57 b	1466.9 ab	12.385 b-e	1.496 h-l	3.680 fgh	17.549
27	16.18 jkl	887.2 f-j	9.543 h-l	1.128 mn	2.383 hi	12.083
RFT	13.72 klm	863.9 h-k	6.697 m	0.871 n	0.669 j	6.374
IC	12.27 lm	766.0 i-l	4.370 n	0.204 o	0.653 j	6.653
RCB	4.38 n	857.8 h-k	2.232 o	0.242 o	0.425 j	5.465
CB	8.65 mn	719.4 i-l	4.444 n	0.274 o	0.221 j	4.284

In each column different letters indicate significant difference (P<0.01); nd: not determined; QE: Quercetin equivalents; GAE: Gallic acid equivalents; 1-27: Roots of chicory plants (*Cichorium intybus*) collected from different localities; RFT: Roasted fruits of turpentine tree (*Pistacia terebinthus*); IC: instant coffee; RCB: Roasted coffee beans; CB: Raw coffee beans.

Conclusion

In the present study, the 6th, 13th. 15th genotypes for total phenolic content and 13th, 15th, 23th genotypes for total flavonoid content had the highest total bioactive content with regards to the genotypes of *C. intybus.* Likewise, the 1th, 6th, 13th, 15th genotypes for DPPH, and the 1st, 12th, 16th, 19th genotypes for ABTS had the highest radical scavenging activity. Moreover, the 3th, 5th, 7th, 10th, 11th, 12th, 13th, 14th, 15th, 20th, 21th and 26th genotypes had the highest value in terms of mineral contents. Chicory roots showed low antioxidant activity, but, higher mineral content compared to the other tested coffee varieties. As a result, the 13th and the 15th genotypes can be recommended as a coffee additive in terms of antioxidant properties and mineral contents.

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