DE GRUYTER OPEN

ACTA FACULTATIS MEDICAE NAISSENSIS

UDC:634.74:543:615.27

DOI: 10.2478/afmnai-2014-0030

Scientific Journal of the Faculty of Medicine in Niš 2014;31(4):245-252

Original article ■

Bagged *Aronia Melanocarpa* tea: Phenolic Profileand Antioxidant Activity

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SUMMARY

While there is a large number of scientific papers reporting chemical composition and biological activities of *Aronia melanocarpa*, there is a lack information regarding the commercially available bagged tea. In order to supply new information on the antioxidant activity of the *Aronia melanocarpa* tea infusions, the aim of this study was to evaluate individual phenolic compounds which could be responsible for antioxidant activities of these beverages.

Selected anthocyanins (cyanidin-3-*O*-galactoside, cyanidin-3-*O*-glucoside, cyanidin-3-*O*-arabinoside, and cyanidin-3-*O*-xyloside), gallic acid, caffeic acid, rutin, morin, and protocatechuic acid were simultaneously detected from commercially available tea infusions using a High Performance Liquid Chromatographic (HPLC) method. The antioxidant activity was measured using five *in vitro* spectrophotometric methods: 1,1-diphenyl-2-picrylhydrazyl radical scavenging activity (DPPH), 2,2'-azino-bis (3-ethylbenzthiazoline-6-sulphonic acid) radical cation scavenging activity (ABTS), ferric reducing-antioxidant power (FRAP) and reduction power (RP)Fe(III) to Fe(II).

Obtained results showed that anthocyanins, predominantly of cyanidin-3-O-galactoside, are the major class of polyphenolic compounds in tea infusions. Among phenolic acids the most abundant is caffeic acid. A significant correlation between DPPH and ABTS and FRAP and RP suggested that antioxidant components in these beverages were capable scavenging free radicals and reducing oxidants.

Generally, these beverages had relatively high antioxidant capacities and could be important dietary sources of antioxidant phenolics for the prevention of diseases caused by oxidative stress.

***Key words:* *Aronia melanocarpa*, tea infusions, polyphenolic compounds, antioxidant activity, HPLC**

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INTRODUCTION

The genus *Aronia* belong to the *Rosaceae* family, native to eastern North America, has become popular in Eastern Europe and Russia. It includes two species: *Aronia melanocarpa* (Mich.) Ell., known as black chokeberry, and *Aronia arbutifolia* (L.) Pers., known as red chokeberry. Most information concerning *Aronia* cultivation refers to black chokeberry. Their berries are used for production of juice, jam, wine, and tea as well as anthocyanin colorant (1). In recent years *A. melnocarpa* has been highlighted with respect to its particularly rich source of antioxidants. Those compounds are mainly represented by vitamin C and polyphenols such as anthocyanins, phenolic acids, flavanols, flavonols and tannins (2-4). They are known as natural antioxidants and due to their high concentration and qualitative diversity, chokeberry fruit is increasingly referred to as natural functional foods.

These findings have been confirmed in the review research of Szajdek and Borowska (5) and Kokotkiewicz et al. (6). Chokeberry fruit and its products supplement the treatment of hypertension, atherosclerosis and gastrointestinal tract disorders. The bioactive compounds found in chokeberry strengthen blood vessel walls and improve their elasticity. Chokeberry juice improves peripheral circulation of the blood and boosts the body's resistance to infections (7). Many researchers are examining the chemical nature and activity of natural antioxidants in *A. melanocarpa* (2-7). Little is known about the phenolic profiles and antioxidant activity of tea infusions of *A. melanocarpa* (8) and there is a lack information regarding the phenolic content and antioxidant activity of infusions obtained from commercially available bagged tea. Tea as well as herbal and fruit tea infusions contribute to the major source of phenolic compounds in our diet (9).Chokeberry tea infusions are found to have beautiful and aromatic taste.

The aim of this paper was to estimate the phenolic content and evaluate the antioxidant activity of commercially available bagged tea infusions of *A. melanocarpa*.

MATERIALS AND METHODS

Chemicals

The 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) was purchased from Acros Organics (New Jersey, USA). 2,2-azinobis (3-ethylbenzothiazoline-6-sulfonic acid) diamonium salt (ABTS), 2,2-diphenyl-1-picrylhidrazyl (DPPH), 2,4,6-Tri(2-pyridyl)-S-tirazine (TPTZ), gallic acid, protocatechuic acid, caffeic acid, rutin, morin,quercetin, were purchased from Sigma Aldrich (Steineheim, Germany). Cyanidin-3-*O*-galactoside chloride, cyanidin-3-*O*-glucoside chlorideand cyanidin-3-*O*-arabinosidewere purchased from Extrasynthese (Cedex, France). All standards were of >95% purity. Potassium peroxodisulfate, ammonium iron(II) sulfate hexahydrate, iron (III) chloride, potassium hexacyanoferrate(III), sodium acetate, sodium dihydrogen phosphate, sodium hydrogen phosphate, ascorbic acid, trichloroacetic acid, hydrochloric acid, acetic acid, formic acid and acetonitrile (HPLC grade) were purchased from Merck® (KGaA, Darmstadt, Germany). Ethanol (96% by vol.) and methanol (HPLC grade) were from J.T. Baker.

Preparation of tea infusions

Five commercially available tea infusions (TI) were purchased at local markets. Tea infusions were prepared in the following manner: (2.0000±0.0001)g of each tea sample (1 tea bag) was weighed and infusing with 200 ml deionised water heated to 95°C for 10 min. The solutions were filtered through cotton wool, and then the residue was washed with deionised water, cooled to room temperature and finally diluted to 250 ml with deionised water.

Instruments

An Agilent 8453 UV/V spectrophotometer was used for absorbance measurements and spectra recording, using optical cuvettes of 1 cm optical path. The pH measurements were made with Hanna Instruments pH-meter equipped with glass electrode. A model 1200 Agilent Technologies was used for HPLC analysis. The analytical column was C_{18} (Zorbax Eclipse XDB-C18, 5 μm, 4.6×150 mm).

General characteristics

The pH was measured with a pH meter previously calibrated with buffer solutions (pH 4.00 and 7.00). The density measurement was carried out using a pycnometer, comparing the density of the infusion with the density of pure water at 20ºC (10). The moisture determination was carried out in the dry sample by the method of weight loss by evaporation in an oven at (100± 2)ºC until constant weight (11).

Antioxidative assays

For DPPH method (12), which is slightly modified, a solution of DPPH (1×10^{-4} mol·l^{-1}) was prepared in methanol. 5.0 ml of this solution and 100 μl of tea infusion were mixed in 10 ml volumetric flask and filled with methanol to the mark. The mixture was shaken and left at room temperature for 30 min. The absorbance was measured spectrophotometrically at 520 nm. The Trolox calibration curve was plotted as a function of the decrease in absorbance of DPPH radical scavenging activity.

$$(\Delta A = A_{blank} - A) \quad (1)$$

The final results were expressed as milimoles of Trolox equivalents (TE) pergram of tea sample.

The ABTS radical scavenging activity was measured using the method of Re et al.(13) and Arts et al. (14). ABTS stock solution (7mM) and 2.45 mM potassium persulfate were left at room temperature for 16 h to produce ABTS radical cation ($ABTS^{\cdot+}$). Then, $ABTS^{\cdot+}$ solution was diluted with distilled water to an absorbance of 0.700±0.02 at 734 nm. An aliquot of tea infusion (100 μl) was mixed with 3.9 ml of diluted $ABTS^{\cdot+}$ solution. The absorbance was monitored at 734 nm after 6 min. The Trolox calibration curve was plotted as a function of the decrease in absorbance of ABTS radical cation scavenging activity. The final results were expressed as milimoles of (TE) per gram of tea samples.

Ferric reducing-antioxidant power (FRAP) assay was performed as previously described by Benzie and Strain (15). In the FRAP assay, antioxidants in the sample reduce Fe^{3+}-TPTZ complex to the ferrous form at low pH (pH=3.6) with an increase in absorbance at 595 nm. Briefly, 3.0 ml of freshly prepared FRAP reagent was mixed with 20 μl of infuse tea sample along with 380 μl (total volume was 3.4 ml). The absorbance at 595 nm was recorded after 5 min incubation at 37°C. FRAP values were expressed as milimoles of Fe^{2+} equivalents (FE) per gram of tea sample.

Reducing power (RP) assay Fe(III) to Fe(II) was determined as described by Oyaizu (16). Reducing power was expressed in relation to the reducing power of ascorbic acid as a positive control (Ascorbate Equivalent Antioxidant Capacity, AEAC). 100 µl of tea infusion were mixed with 1.5 ml of phosphate buffer (NaH_2PO_4-Na_2HPO_4,0.2 mol·l^{-1}, pH 6.6) and 2.5 ml of 1% potassium ferricyanide ($K_3Fe(CN)_6$). The mixture was incubated at 50 °C for 20 min. 1.5 ml of 10% trichloroacetic acid was added to the mixture, which was then centrifuged for 10 min at 3000 rpm. The upper layer of solution (1.5 ml) was mixed with 1.5 ml of distilled water and 0.3 ml of 0.1% of $FeCl_3$. Absorbance was measured at 700 nm. Increased absorbance of the reaction mixture indicated increased reducing power. RP values were expressed as milimoles of ascorbic acid equivalents (AAE) per gram of tea sample.

Separation of individual phenolic compounds in tea infusions

An Agilent chromatograph equipped with autosampler and photodiode-array and fluorescence detector (1200 Series) was used for the HPLC analysis. The separation was performed with a Zorbax Eclipse C_{18} column kept at 25 °C, at a flow rate of 0.8 ml/min, and an injection volume of 20 μl. Detection was performed by scanning from 200 to 600 nm. For the gradient elution, the following programme, slightly modified, was used: solvent A (5% HCOOH-H_2O) and solvent B (80% AcN and 5% HCOOH-H_2O) as follows: in 0-10 min, solvent B increased by 0% , in 10-28 min, it gradually increased by 0-25% , in 28-30 min solvent B increased by 25% B, in 30-35 min, it gradually increased by 25-50% B, in 35 to 40 min, solvent B gradually increased by 50-80% B, and finally in the last 5 min it gradually decreased by 80-0% B (17). The individual phenolic compounds were separated within 40 min. Identification was carried out by comparing the retention times and spectral data with those of standards. Quantitative determination of individual phenolic compounds in tea infusions was calculated using calibration lines. For compounds lacking standards, quantification was carried out using similar compounds as standards. Thus, cyanidin-3-*O*-xyloside was quantified in equivalents of cyanidin-3-*O*-glucoside. All experiments were repeated three times.

Statistical analysis

Data are presented as the mean ± standard deviation (SD) for triplicate determinations. Statistical analysis was performed by paired Student *t* test, using a statistical package running on a computer (Statistica 8.0, StatSoft, Tulsa, Oklahoma, USA). A probability of $p<0.05$ was considered to be statistically significant (18).

RESULTS

General characteristics of the tea infusion are given in the Table 1. The results of antioxidative activity and quantitative phenolic composition in tea infusions are presented in Table 2 and 3, respectively.

The typical HPLC profile of the polyphenols of selected tea infusion is given in Figure 1.

Table 1. *General characteristics of tea infusions*

Tea infusion	pH	Density g/ml	Moisture %
TI1	4.48	0.9964	7.3
TI2	3.74	0.9981	7.3
TI3	3.58	1.0001	7.1
TI4	4.68	0.9977	7.4
TI5	4.56	0.9990	7.8

Table 2. *The antioxidant capacities* of tea infusions*

Tea infusion	DPPH	ABTS	FRAP	Fe(III)/Fe(II)
	mmol TE/g	mmol TE/g	mmol FE/g	mmol AAE/g
TI1	0.067 ± 0.001[b]	2.715 ± 0.007[b]	0.147 ± 0.001[b]	1.36 ± 0.03[c]
TI2	0.074 ± 0.002[b]	2.744 ± 0.006[b]	0.136 ± 0.001[a]	0.53 ± 0.02[a]
TI3	0.068 ± 0.001[b]	2.731 ± 0.006[b]	0.147 ± 0.001[b]	0.88 ± 0.02[b]
TI4	0.055 ± 0.001[a]	0.076 ± 0.001[a]	0.153 ± 0.001[b]	3.48 ± 0.04[e]
TI5	0.058 ± 0.001[a]	0.089 ± 0.002[a]	0.144 ± 0.001[b]	2.14 ± 0.03[d]

*Values are the mean ± SD (n = 3). Values with different letters within columns are statistically different at $p<0.05$ by paired Student *t* test

Table 3. *Individual polyphenols content (mg/g)* in tea infusions*

	TI1	TI2	TI3	TI4	TI5
Gallic acid	0.476±0.001	ND**	0.596±0.003	ND	0.372± 0.001
Caffeic acid	0.095±0.002	0.067±0.001	1.26±0.03	0.072±0.002	0.727±0.002
Protocatechuic acid	0.176±0.004	ND	0.023±0.001	ND	ND
Rutin	0.738±0.006	0.329±0.006	0.032±0.001	0.279±0.007	0.252±0.002
Morin	ND	ND	0.501±0.003	ND	ND
Quercetin	ND	ND	0.243±0.002	ND	ND
Cyanidin-3-*O*-galactoside	37.6±0.4	29.5±0.3	22.2±0.2	11.6±0.1	3.8±0.1
Cyanidin-3-*O*-glucoside	1.69±0.07	1.38±0.07	5.6±0.2	0.52±0.01	3.1±0.1
Cyanidin-3-*O*-arabinoside	17.7±0.3	13.9±0.3	9.3±0.2	5.5±0.1	1.9±0.1
Cyanidin-3-*O*-xyloside	2.1±0.1	1.62±0.03	4.4±0.1	0.77±0.03	1.71±0.04

*Mean ± SD (n = 3), **ND - not detected

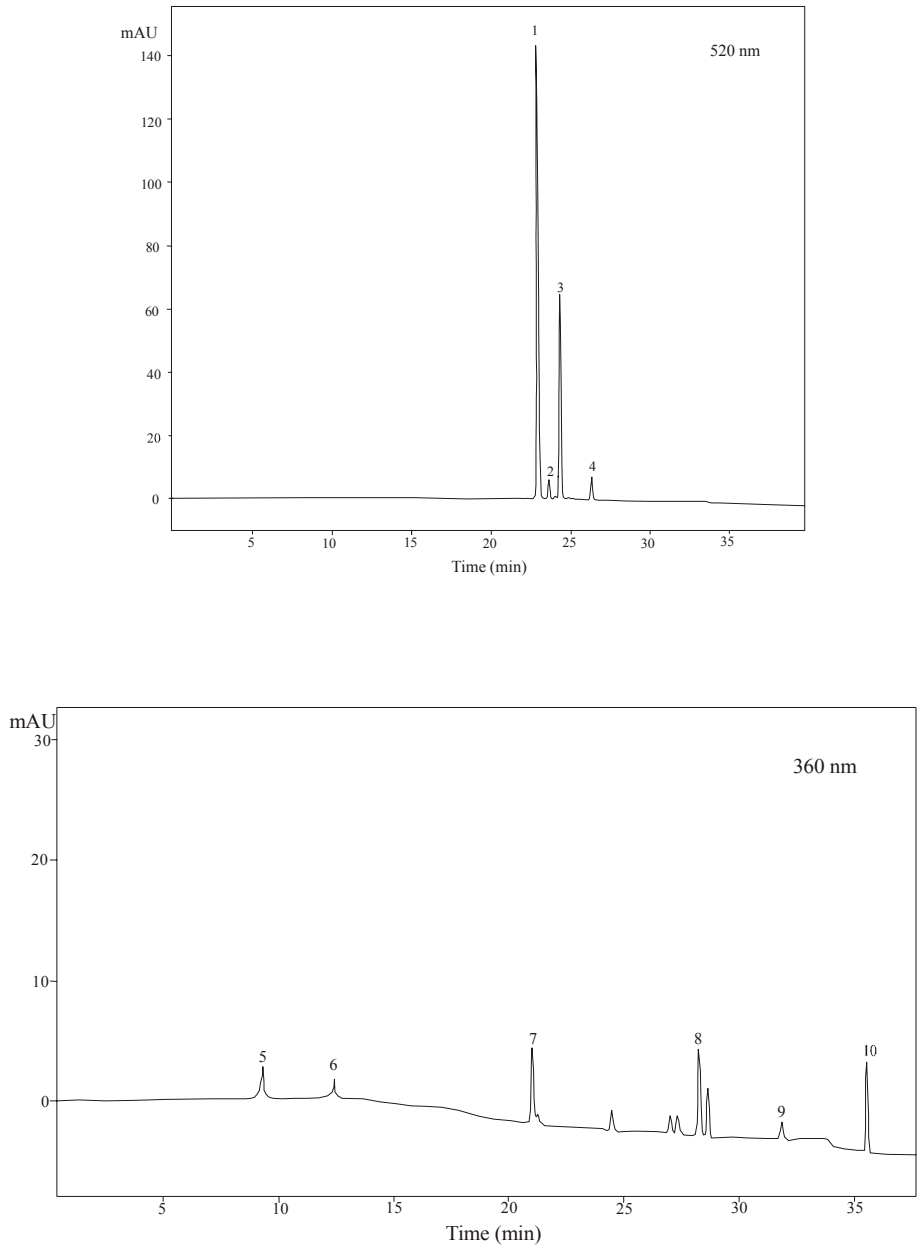

Figure 1. *Chromatogram of tea infusion (TI3) recorded at 520 nm: 1. cyanidin-3-O-galactoside, 2. cyanidin-3-O-glucoside, 3. cyanidin-3-O-arabinoside, 4. cyanidin-3-O-xyloside; recorded at 360 nm: 5. gallic acid, 6. protocatechiuc acid, 7. caffeic acid, 8. rutin, 9. morin, 10. quercetin*

DISCUSSION

The pH values in the tea infusions ranged from 3.58-4.68. Tea infusions TI2 and TI3 have relative acid pH values of 3.74 and 3.58, respectively. The density was around 1 g/ml in all samples. According to the Serbian legislation (19), a maximum of moisture for this kind of products to ensure a good preservation is 12%. None of the samples exceeded this level. The moisture content was between 7.1-7.8%.

Taking into account that the most natural antioxidants are multifunctional, the antioxidant capacities of samples cannot be completely described by one single method. In addition, currently, there is no single antioxidant assay for food labeling because of the lack of standard quantification methods. Each antioxidant assay has a different mechanism, redox potential, pH, reaction media, etc. Antioxidant assays may be broadly classified as the electron transfer (ET)- and hydrogen atom transfer (HAT)-based assays (20). According to Hengst et al. (21), a combination of HAT and ET assays or of reduction and free radical scavenging based assays is strongly recommended to use. Also, according to Apak et al. in IUPAC Technical Report (20), using values of antioxidant

tests to make dietary decisions is unwarranted and more difficult. In the present study, spectrophotometric techniques such as DPPH radical scavenging activity, 2,2'-azino-bis (3-ethylbenzthiazoline-6-sulphonic acid) radical cation scavenging activity (ABTS), ferric reducing-antioxidant power (FRAP) and reduction power (RP) were used to determine antioxidant activity of tea infusions. DPPH and ABTS assays are based on the ability of antioxidants to scavenge free radicals, while the FRAP and RP assays are based on the capacity of antioxidants to reduce ferric(III) ions to ferrous(II) ions. DPPH radical scavenging activity ranged from 0.038 mmolTE/g to 0.074 mmolTE/g. ABTS activity varied from 0.076 mmolTE/g to 2.744mmolTE/g. The lowest and the highest ABTS radical cation scavenging activities were found in TI4 and TI2, respectively, which was consistent with the results from DPPH radical scavenging activity assay. In addition, the values obtained by the ABTS assay were consistently higher than those obtained by the DPPH assay. Different reaction kinetics between phenol and the ABTS radical cation and DPPH radical over a similar range of contents might lead to the different results from two methods (22). The antioxidant activity of tea infusions measured by the FRAP and RP assays is ranged from 0.136 mmol FE/gto 0.153 mmolFE/gand from 0.53 mmolAAE/gto 3.48 mmolAAE/g, respectively. The lowest and the highest FRAP and RP activities were found in TI2 and TI4, respectively. Furthermore, among the applied antioxidant methods, DPPH and ABTS methods correlated strongly (R^2=0.93131, p<0.0500), followed by the FRAP and RP methods (R^2=0.85863, p<0.0500).This might be explained by the mechanism by which phenols scavenge the stable DPPH and ABTS radical in these assays as well as capacity of antioxidants to reduce ferric(III) ions to ferrous(II) ions. When the analyzed antioxidant activity of tea infusions were compared with those of *A. melanocarpa* berries and their products, such as juice and pomace(4) and tea (8, 23), the obtained results indicated higher antioxidant activity and higher content of anthocyanins and flavonoids (rutin). Also, the obtained results are in accordance with literature data obtained for antioxidant activity of green, black, herbal and fruit tea infusions (24).

Identification of the individual phenolic compounds has given more information about chemical characteristics of tea infusions and their antioxidant activity. The obtained results showed that tea infusion samples contain anthocyanins, rutin, morin, and a number of well-known non-flavonoid polyphenols, such as gallic acid and protochatechuic acid, as well as a number of chlorogenic acid such as caffeic acid. Relatively high levels of anthocyanins were found while the values were lower for rutin, morin, gallic acid, protochatechuic acid and caffeic acid. Anthocyanins in tea infusions are a mixture of four different cyaniding glycosides: cyanidin-3-*O*-galactoside, cyanidin-3-*O*-glucoside, cyanidin-3-*O*-arabinoside and cyanidin-3-*O*-xyloside of which cyanidin-3-*O*-galactoside is the main one. Individual content of determined phenols are given in Table 3. The obtained results show that tea infusions are rich in o-diphenolics as cyanidinderivates and caffeic acid. According to Rice-Evans et al. (25), o-dihydroxy structure in the B ring leads to higher stability of the radical form and participates in electron delocalization. Because of that, these compounds are the most active as antioxidants. According to the literature, the anthocyanin amount in chokeberries varies from 3166 to 14800 mg/kg (26-31). These results are similar with those obtained for chokeberries tea infusions (Table 3).

CONCLUSION

The obtained results showed that *A. melanocarpa* leaves tea infusions contained a high level of polyphenols with high antioxidant activity. When individual phenolic contents of these infusions were compared with those of *A. melanocarpa* products such as pomace and juice, these infusions could contribute the same health benefit as other chokeberry products in terms of polyphenols. Regarding the important place that tea infusions have as a popular beverages, this could be commercially exploited. Because of their high antioxidant activities, it could be speculated that these infusions will be beneficial for the diseases caused by oxidative stress.

Acknowledgement

This research was supported by grant numbers 172047 from the Ministry of Education, Science and Technological Development of the Republic of Serbia. The authors are grateful for the financial support provided by this Ministry.

References

1. Jeppsson N. The effectsof fertilizer rate on vegetative growth, yield and fruit quality, with special respect to pigments, in black chokeberry (*Aronia melanocarpa*) cv. “Viking”. SciHort Amsterdam 2000; 83: 127-37. http://dx.doi.org/10.1016/S0304-4238(99)00070-9
2. Oszmiański J, Sapis JC. Anthocyanins in fruits of Aronia melanocarpa (chokeberry). J Food Sci 1988; 53: 1241 -2. http://dx.doi.org/10.1111/j.1365-2621.1988.tb13577.x
3. Slimestad R, Torskangerpoll K, Nateland HS, Johannessen T, Giske NH. Flavonoids from black chokeberries, Aronia melanocarpa. J Food Compost Anal 2005; 18:61-8. http://dx.doi.org/10.1016/j.jfca.2003.12.003
4. Oszmianski J, Wojdylo A. Aronia Melanocarpaphenolics and their antioxidant activity. Eur Food Res Technol 2005; 221: 809-13. http://dx.doi.org/10.1007/s00217-005-0002-5

5. Szajdek A, Borowska EJ. Bioactive Compounds and Health-Promoting Properties of Berry Fruits: A Review. Plant Foods Hum Nutr 2008; 63:147-56. http://dx.doi.org/10.1007/s11130-008-0097-5
6. Kokotkiewicz A, Jaremicz Z, Luczkiewicz M. *Aronia* plants: A Review of traditional use, biological activities and perspectives for modern medicine. J Med Food 2010; 13: 255-69. http://dx.doi.org/10.1089/jmf.2009.0062
7. Ara V. The black chokeberry: a healthy fruit that will soonbe "on all tongues"? Fruit Process 2002; 12: 500-6.
8. Šavikin K, Zdunić G, Janković T, Gođevac D, Stanojković T, Pljevljakušić D. Berry fruit tea: Phenolic composition and cytotoxic activity. Food Res Int 2014; 62: 677-83. http://dx.doi.org/10.1016/j.foodres.2014.04.017
9. Shahidi F. Antioxidants in food and food antioxidants. Nahrung. 2000; 44: 158-63. http://dx.doi.org/10.1002/1521-3803(20000501) 44:3<158::AID-FOOD158>3.0.CO;2-L
10. SRPS EN 1131: 2005, Sokovi od voća i povrća, Određivanje relativne gustine
11. AOAC method 930.15, Loss on drying (moisture) for feeds (at 135ºC for 2 hours) dry metter on oven drying for feeds (at 135ºC for 2 hours) In: Official Methods of Analysis, Association of Official Analytical Chemists, Washington, D.C., 1990: 69-70.
12. Brand-Williams W, Cuvelier ME, Berset C. Use of a free radical method to evaluate antioxidant activity. LWT-Food SciTechnol 1995; 28: 25-30. http://dx.doi.org/10.1016/S0023-6438(95)80008-5
13. Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. Antioxidant activity applying an improved ABTS radical cation decolonization assay. Free Radical BioMed1999; 26: 1231-7. http://dx.doi.org/10.1016/S0891-5849(98)00315-3
14. Arts MJTJ, Haenen GRMM, VossHP, Bast A. Antioxidant capacity of reaction products limit the applicability of the trolox equivalent antioxidant capacity (TEAC) assay. FoodChem Toxicol2004; 42: 45-9. http://dx.doi.org/10.1016/j.fct.2003.08.004
15. Benzie IFF, Strain JJ. Ferric reducing/antioxidant power assay: Direct measure of total antioxidant activity of biological and modified version for simultaneous measurement of total antioxidant power and ascorbic acid concentration. Methods Enzymol 1999; 299: 15-27. http://dx.doi.org/10.1016/S0076-6879(99)99005-5
16. Oyaizu M. Studies on products of browning reaction: antioxidative activity of products of browning reaction prepared from glucosamine. Jpn JNutr 1986; 44: 307-15. http://dx.doi.org/10.5264/eiyogakuzashi.44.307
17. Naczk M, Shahidi F. Extraction and analysis of phenolics in food. JChromatogr A,2004; 1054: 95-111. http://dx.doi.org/10.1016/j.chroma.2004.08.059
18. Miller JN, Miller JC. Statistics and Chemometrics for Analytical Chemistry. Pearson Education, London, 2005: 288.
19. Pravilnik o kvalitetu čaja, biljnog čaja i njihovih proizvoda, Službeni glasnik Republike Srbije br. 4/2012.
20. Apak R, Gorinstein S, Bohm V, Schaich KM, Ozyurek M, Guclu K. Methods of measurement and evaluation of natural antioxidant capacity/activity (IUPAC Technical Report). Pure ApplChem 2013; 85: 957-98. http://dx.doi.org/10.1351/PAC-REP-12-07-15
21. Hengst C, Werner S, Muller L, Frohlich K, Bohm V. Determination of the antioxidant capacity: influence of the sample concentration on the measured values. Eur Food Res Technol 2009; 230: 249-54. http://dx.doi.org/10.1007/s00217-009-1166-1
22. Campos AM, Lissi EA. Kinetics of the reaction between 2,2-azinobis (3-ethylbenzothiazoline)-6-sulfonic acid (ABTS) derived radical cation and phenols. Int J Chem Kinet 1996; 29: 219-24. http://dx.doi.org/10.1002/(SICI)1097-4601(1997)29:3<219::AID-KIN9>3.0.CO;2-X
23. Piljac-Žegarac J, Belščak A, Piljac A. (2009). Antioxidant capacity and polyphenolic content of blueberry (Vaccinium corymbosum L.) leafinfusions. J Med Food 2009; 12:608-14. http://dx.doi.org/10.1089/jmf.2008.0081
24. Veljković JN, Pavlović AN, Mitić SS, Tošić SB, Stojanović GS, Kaličanin BM, Stanković DM, Stojković MB, Mitić MN, Brcanović JM. Evaluation of individual phenolic compounds and antioxidant properties of black, green, herbal and fruit tea infusions consumed in Serbia: spectrophotometrical and electrochemical approaches. J Food Nutr Res 2013; 52: 12-24.
25. Rice-Evans CA, Miller NJ, Bolwell PG, Bramley PM, Pridham JB. The relative antioxidant activities of plant-derived polyphenolic flavonoids. Free Radical Res 1995; 22: 375-83. http://dx.doi.org/10.3109/10715769509145649
26. Zheng W, Wang SY. Oxygen radical absorbing capacity of phenolics in blueberries, cranberries, chokeberries and lingonberries. J Agric Food Chem 2003; 51: 502-9. http://dx.doi.org/10.1021/jf020728u
27. Maatta-Riihinen KR, Kamal-Eldin A, Mattila PH, Gonzalez-Paramas AM, Torronen AR. Distribution and content of phenolic compounds in eighteen Scandinavian berry species. J Agric Food Chem 2004; 52: 4477-86. http://dx.doi.org/10.1021/jf049595y
28. Wu X, Gu L, Prior RL, McKay S. Characterization of anthocyanins and proanthocyanidins in some cultivars of *Ribes*, *Aronia* and *Sambucus* and their antioxidant capacity. J Agric Food Chem 2004; 52: 7846-56. http://dx.doi.org/10.1021/jf0486850
29. Slimestad R, Torskangeroll K, Nateland HS, Johannessen T, Giske NH. Flavonoids from black chokeberries, *Aronia melanocarpa*. J Food Comp Anal 2005; 18: 61-8. http://dx.doi.org/10.1016/j.jfca.2003.12.003
30. Kulling SE, Rawel HM. Chokeberry (*Aronia melanocarpa*) - A review on the characteristic components and potential health effects. Planta Med 2008; 74: 1625-34. http://dx.doi.org/10.1055/s-0028-1088306
31. Jakobek L, Šeruga M, Krivak P. The influence of interaction among phenolic compounds on the antiradical activity of chokeberries (*Aronia melanocarpa*). 2011; 62: 345-52.

ARONIA MELANOCARPA FILTER ČAJ: FENOLNI PROFIL I ANTIOKSIDATIVNA AKTIVNOST

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Sažetak

U cilju dobijanja informacija o antioksidativnoj aktivnosti infuz čajeva ploda aronije (*Aronia melanocarpa*) određena je koncentracija pojedinačnih fenolnih jedinjenja za koje se smatra da su odgovorna za antioksidativnu aktivnost ispitivanih čajeva.

Sadržaj antocijana (cijanidin-3-*O*-galaktozid, cijanidin-3-*O*-glukozid, cijanidin-3-*O*-arabinozid, cijanidin-3-*O*-ksilosid), galne kiseline, kafene kiseline, protokatehinske kiseline, rutina imorina određen je korišćenjem tečne hromatografije pod visokim pritiskom (HPLC) sa UV/Vis ifluorescentnim detektorom. Antioksidativna aktivnost je ispitana korišćenjem četiri *in vitro* spektrofotometrijske metode: metoda uklanjanja 2,2-difenil-1-pikrilhidrazil (DPPH) radikala, metoda vraćanja 2,2'-azino-bis (3-etilbenzotiazolin-6-sulfo katjon radikala) u svoj neutralni oblik (ABTS), redukcija gvožđe(III)-2,4,6-tripiridil-S-triazin kompleksa do gvožđe(II)-2,4,6-tripiridil-S-triazin kompleksa (FRAP) metoda redukcije Fe^{3+} do Fe^{2+}jona (RP).

Na osnovu dobijenih rezultata može se zaključiti da su u ispitivanim infuz čajevima najviše zastupljeni antocijani, a zatim slede fenolne kiseline. Od antocijana najviše je zastupljen cijanidin-3-*O*-galaktozid, a od fenolnih kiselina kafena kiselina. Visok stepen korelacije između DPPH i ABTS metode i FRAP I RP metode ukazuje na to da su prisutna fenolna jedinjenja, kao redukujući agensi u ispitivanim uzorcima, dobri hvatači slobodnih radikala.

Generalno, dobijeni rezultati ukazuju da infuz čajevi od lista aronije pokazuju visok stepen antioksidativne aktivnosti i mogu biti dopuna svakodnevnoj ishrani u prevenciji bolesti izazvanih oksidativnim stresom.

***Ključne reči: Aronia melanocarpa*, infuz čaj, polifenolna jedinjenja, antioksidativna aktivnost, HPLC**