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Proceeding Article

Aronia Berry Tea as Antioxidant Functional Drink: Bioactive Phenolics By HPLC-DAD and LC-ESIQTOFF-Mass Spectrometry

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ABSTRACT

In this proceeding, aronya tea was produced as healthy functional products of Turkish aronia fruit (Aronia melanocarpa sp.Viking). Their antioxidant activity, antioxidant phenolic profiles, HPLC (High Pressure Liquid Chromatography) profiles and Q-TOFF-MS (Quadrupole Time of Flight Mass Spectrometry) profiles of major active bioactives were determined.With analytical determinations, it has been determined phenolic acids, gallic acid, epigallocatechin, catechin, chlorogenic acid, hydroxycinnamic acid, neochlorogenic acid, vanillic acid, siringic acid, caftaric acid, p-coumaric acid, ferulic acid, t-sinapic acid as flavanols (catechins); rutin (quercetin-3-glycoside), kaempferol-3-rutinoside, quercitrin (quercetin-O-glycoside), quercetin, astragalin (kaempferol-O-glycoside), kaempferol as flavonols and their glycosides ($p< 0.05$).

Keywords: Aronia Berry, Black Chokeberry, Aronia melanocarpa (Michx.), Aronia Tea, Powder

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Introduction

Black chokeberry or aronia (Aronia melanocarpa) is a shrub of the Rosaceae family that is a berry plant native to North America and was transferred to Europe about a century ago (Chrubasik et.al.,2010). Edible parts of black chokeberry are mainly small cherry-like berry parts. The genus Aronia (Rosaceae family) includes two species of shrubs, native to eastern North America and Eastern Canada: *Aronia melanocarpa* (Michx.) Ell., known as black chokeberry and *Aronia arbutifolia* (L.) Pers. (red chokeberry). The aronia berries contain high levels of flavonoids, mostly proanthocyanidins and anthocyanins, and in vitro and in vivo studies indicate that the berries may have potential health benefits, e.g. hepatoprotective effects, cardioprotective effects, antidiabetes effect and anticancer effects on selected CA cells. The consumption of low levels of antioxidants in the form of fruit and vegetables has been shown to more than double the incidence of certain cancers. (Tokusoglu and Boz, 2019; Tokusoglu 2019; Sidor et.al. 2019; Gavaric et.al.2019; Tolić et.al 2015; Savikin et.al.2014).

Unprocessed fresh black chokeberry fruits are generally not consumed routinely owing to their so astringent taste; aronia berries widely consumed as juices, syrups, jams, fruit teas and dietary supplements (Tokusoglu,2019; Tokusoglu and Boz,2019). Chemical composition and biological activity of berries and their products have been widely reported but there are limited works dealing with berry fruit teas. Powder forms of berries and industrial ice-cream form of berries are also utilized as functional food products for nutrition. Tea is popular beverage and currently, herbal infusions based on dried fruit products have gained in popularity because of their fragrance, fruity flavor, lower amounts of caffeine, and low astringent and bitter taste.

In this proceeding content, aronia based product aronia berry teas was manufactured as functional drink and its and bioactive phenolics were determined by HPLC-DAD and LC-ESIQTOFF-Mass Spectrometry.

Material and Methods

Aronia berry [*Aronia melanocarpa* (Michx.)] (black chokeberry) was harvested at Yalova Research Institute, Yalova, Turkey. In our current research, aronia based new products including aronia berry teas (as decoction and infusion types), was developed by Dokuz Eylul University Technology Development Zone Depark Technopark Spil Innova LLC, Izmir Project.

For aronia (chokeberry) powder production, aronia berries were subjected to freze drying (FD) and spray drying process (B-290, Buchi Labour Technik,AG,Flawil, Switzerland) based on our determined conditions (Tokusoglu,2019) (Figure 1).

Figure 1. Aronia Berry, Aronia Powder and Aronia Herbal Tea

In manufacturing, decoction method was applied by boiling of aronia berry material in a non-aluminum pot during 8 min until up to two-thirds of the water was evaporated and was strained by home-made tea strain apparatus.

Phenolics were extracted according to Tokusoglu (2019). The obtained extract was used for determination of total phenolic content (TPC), for antioxidant capacity assay by DPPH method and chromatographic analyses including HLPC-DAD and Q-TOFF-MS.

In Q-TOFF-MS Analyses, 30 °C of column temperature, 2 µL of injection volume, flow rate 0.5 ml/min was performed. Gradient elution was as 0–0.5 min, 5% B; 0,5–2 min, 25% B; 2–4 min, 50% B; 4–6 min, 75% B; 6–10 min, 95% B; for column conditioning için 10-16 min, 5%B. For MS analyses, dryer gas flow rate 140 L/min; nebulizer gas pressure as 35 psi, dryer gas temperature as 290 °C; sheath ggas temperature as 400°C; sheath gaz flow as 12 L/min, Agilent Dual Jet Stream elektrosprey ionization (Dual AJS ESI) intermediate surface unit 6550 iFunnel was utilized.

Table 1. Retention Times (RT) of Phenolic Acids and Flavanols (Catechins) in Aronia Tea (mg/100g).

Phenolic Acids and Flavanols (Catechins)	Retention Time (R.T.) Aronia Tea
DAD-Signal 280 nm	
Gallic Acid	12.88
Epigallocatechin	14.10
Catechins	19.44
Chlorogenic Acid	22.76
Hydroxycinnamic Acid	26.98
Neochlorogenic Acid	30.97
Vanilic Acid	32.54
Syringic Acid	36.66
DAD-Signal 320 nm	
Caftaric Acid	—
Cafeic Acid	—
p-Cumaric Acid	—
Ferulic Acid	—
t-Sinapic Acid	—
Rosemarinic Acid	59.47

Table 2. Phenolic Acid Concentrations of Aronia Teas in Different Brewing Time (mg/100g).

Phenolic Acids and Flavanols (Catechins)	mg/100 g Tea
DAD-Signal 280 nm	
Gallic Acid	0.52 (3 min brewed)
	0.76 (5 min brewed)
	1.12 (10 min brewed)
	1.33 (15 min brewed)
Epigallocatechin	8.45 (3 min brewed)
	13.32 (5 min brewed)
	19.26 (10 min brewed)
	20.44 (15 min brewed)
Catechins	38.16 (3 min brewed)
	41.55 (5 min brewed)
	30.78 (10 min brewed)
	30.96 (15 min brewed)
Chlorogenic Acid	90.28 (3 min brewed)
	95.63 (5 min brewed)
	188.42 (10 min brewed)
	195.33 (15 min brewed)
Hydroxycinnamic Acid	3.08 (3 min brewed)
	3.44 (5 min brewed)
	4.69 (10 min brewed)
	4.90 (15 min brewed)
Neochlorogenic Acid	42.55 (3 min brewed)
	58.03 (5 min brewed)
	67.62 (10 min brewed)
	65.58 (15 min brewed)
Vanilic Acid	10.96 (3 min brewed)
	11.56 (5 min brewed)
	15.67 (10 min brewed)
	12.78 (15 min brewed)
Syringic Acid	—
DAD-Signal 320 nm	
Caftaric Acid	—
Cafeic Acid	—
p-Cumaric Acid	—
Ferulic Acid	—
t-Sinapic Acid	0.55 (3 min brewed)
	1.02 (5 min brewed)
	1.25 (10 min brewed)
	1.22 (15 min brewed)
Rosemarinic Acid	0.60 (3 min brewed)
	0.62 (5 min brewed)
	0.89 (10 min brewed)
	0.54 (15 min brewed)

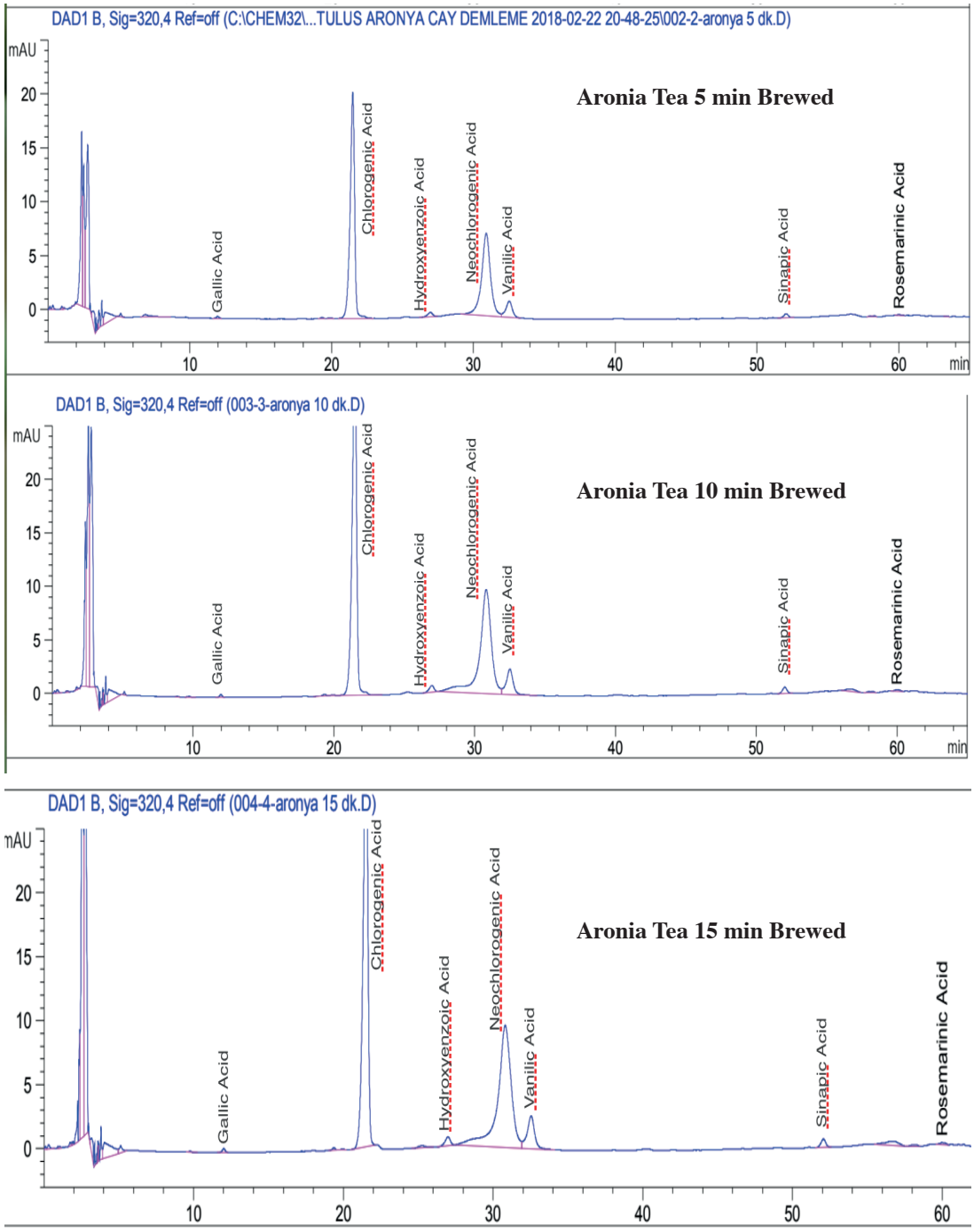
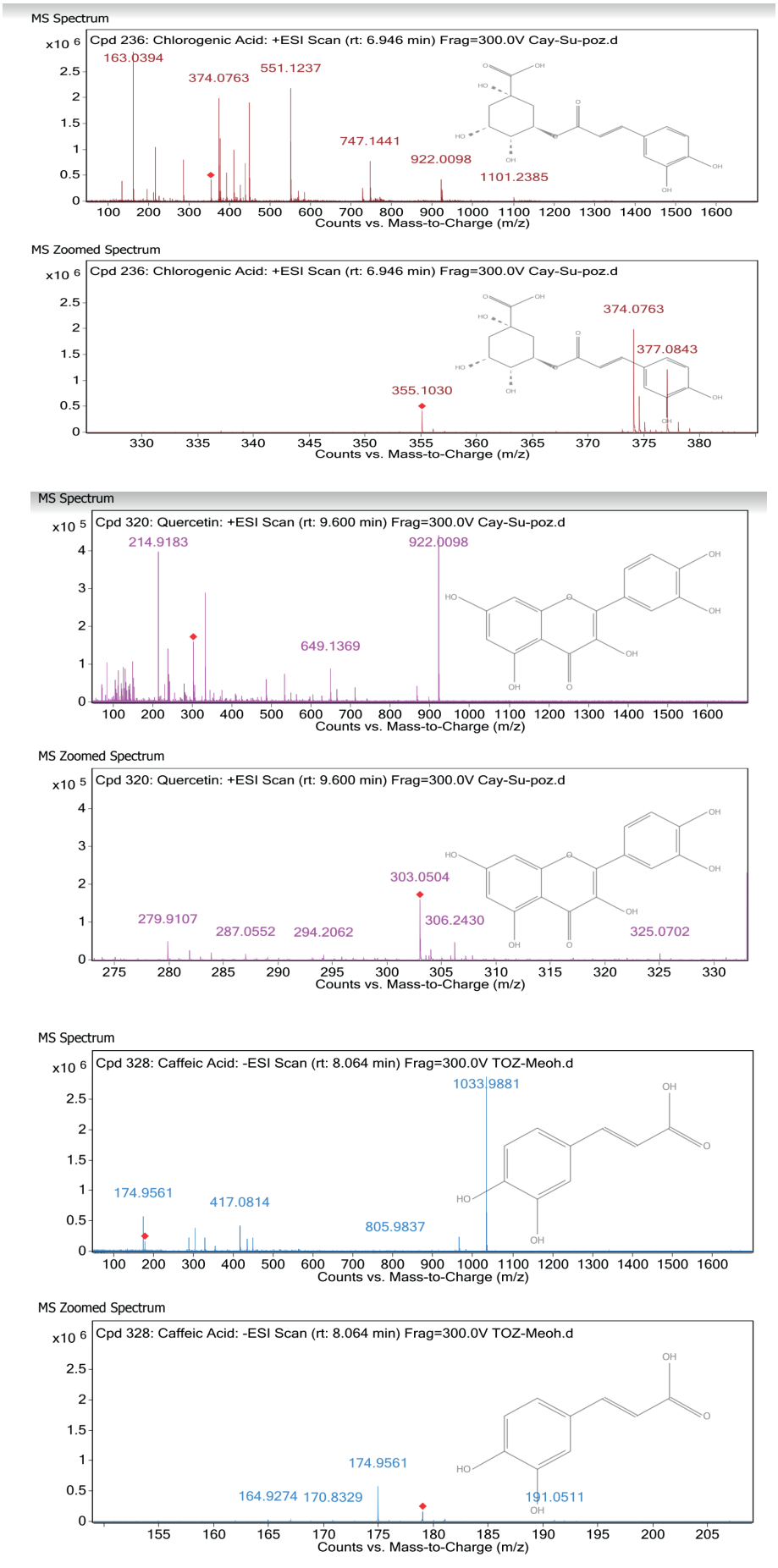


Figure 1. Simultaneously Chromatograms of Phenolic Acids in Aronia Berry Teas Brewed as 5,10,15 min

Figure 2. Chlorogenic Acid, Quercetin, Caffeic Acid Analyses by Q-TOFF-MS



m/z	z	Abund
163,0394	1	2983721,75
217,5422	2	1046018,62
287,0555	1	802338,39
374,0763	2	2137319,3
377,0843	1	1243901,82
411,1263	1	1059906,68
449,1079	1	1929603,27
551,1237	2	2181225,03
551,6252	2	1155425,98
747,1441	2	800334,93

m/z	z	Abund
84,96		106839,01
125,9865		93879,21
149,953		111970,06
214,9183	1	396562,88
238,8847		141247,95
303,0504	1	170687,52
333,5559	2	292666,13
333,5573	2	93553
649,1369	1	92471,28
922,0098	1	440147,43

m/z	z	Abund
174,956	1	662285,12
304,9137	1	454407,98
340,1026	1	644351,47
434,8705		132023,96
441,196	1	148100,68
481,0976	1	289538,08
966,0001	1	503079,33
1033,9881	1	3052135,77
1034,9907	1	701877,95
1035,9927	1	112167,13

Results and Discussion

After harvesting, the content of total polyphenols of fresh aronia berry was 1012.67 ±!34.62 mg GAE/100 ml (*n*=3) and the monomeric anthocyanin level was 425.65 ±3.65 mg/100 ml (*n*=3). The content of total polyphenols in aronia powder product was 444.72 ±!4.33 mg GAE/100 ml (*n*=3) whereas the anthocyanin level of powder was 151.30 ±!1.53 mg/100 ml (*n*=3). Total phenolics was found as 87.72 ±! 0.83 mg GAE/100 ml (*n*=3) whereas total anthocyanin content was measured according to European Pharmacopoeia 6.0 method with slight modifications.

In the study, aronia tea infusion was also carried out. Infusion means achieving a desired taste and aroma results of aronia berry by dissolving a certain proportion of the tea materials into water. This application was performed by using a certain combination of teaware, steeping process, water temperature, water to aronia berry tea ratio. The total phenolics and the anthocyanin level of infusion was determined as 101.02 ±!0.55 mg GAE/100 ml (*n*=3) and 9.05 ±!0.05 mg/100 ml (*n*=3), respectively.

In aronia tea, retention times of phenolic acids and flavanols in Table 1 and their analytical concentrations are in Table 2. Figure 1 shows the simultaneously chromatograms of gallic acid (5-caffeoylyquinic acid), hydroxycinnamic acid, neochlorogenic acid (3-caffeoylyquinic acid), vanillic acid, syringic acid, rosemarinic acid in aronia teas with 5,10,15 min brewed (Figure 1). Figure 2 indicate chlorogenic acid, quercetin, caffeic acid analyses by Q-TOFF-MS (Figure 2). It has been determined phenolic acids, gallic acid, epigallocatechin, catechin, chlorogenic acid, hydroxycinnamic acid, neochlorogenic acid, vanillic acid, siringic acid, caftaric acid, p-coumaric acid, ferulic acid, t-sinapic acid as flavanols (catechins); rutin (quercetin-3-glycoside), kaempferol-3-rutinoside, quercitrin (quercetin-O-glycoside), quercetin, astragalin (kaempferol-O-glycoside), kaempferol as flavonols and their glycosides. It has been identified that chlorogenic acid (ChA) was found major phenolic matter in 3/5/10/15 min brewed aronia teas that were determined as 90.28 mg/100g, 95.63 mg/100g, 188.42 mg/100 g, and 195.33 mg/100g, respectively and it has been provided strong antioxidative property ($p< 0.05$). Quercetin (QE) was found major flavonol in 3/5/10/15 min brewed aronia teas that were determined as 307.20 mg/100g; 328.96 mg/100g; 380.56 mg/100g; and 387.25 mg/100g, respectively ($p< 0.05$).

Aronia berry (black chokeberry) fruit teas was found as valuable source of flavonoids and anthocyanins compared to the most of commonly consumed berry teas. Aronia berry based new nutritive tea products could be utilized in functional food industry as valuable antioxidant sources and could be evaluated as innovative teas.

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