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Summary

Zusammenfassung

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Characteristics of Myrtle (*Myrtus communis* L.) Fruit and Oils

Eigenschaften von Myrte (Myrtus communis L.) Beeren und Öle

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In this study, chemical analysis, amino acids, sugar pofile, fatty acid composition and mineral contents of white and black myrtle (Myrtus communis L.) fruits were determined. The crude protein and oil contents of white and black myrtle fruits were determined between 5.99 and 5.45 % to 8.31 and 6.17 % respectively. In addition, total phenol, flavonoid and anthocyanin contents of white and black fruits were determined between 4253 and 4060 mg gallic acid equivalent (GAE)/g, 94.4 and 121.1 mg catechol equivalent (CE)/g, and 0.096 and 1.667 mmol/g, respectively. Aspartic acid contents of white and black myrtle fruits were determined as 0.50 and 0.45 % respectively. In addition, the highest glutamic acid and arginin were found in white myrtle fruits. Fructose and glucose contents of white and black myrtle fruits were reported as 11.26 and 11.53 % to 13.02 and 13.74 % respectively. Linoleic, palmitic, and oleic acid contents of white and black myrtle fruit oils were determined as 72.48 and 68.34 %, 9.99 and 10.43 % to 8.52 and 9.02 % respectively. P, K, Ca, and Mg contents of black and white myrtle fruits were measured as 1335.10 and 1165.15 mg/kg to 2714.16 and 2631.04 mg/kg, 191.13 and 245.28 mg/kg to 837.86 and 720.38 mg/kg respectively. In conclusion, myrtle fruits were found to be important sources of nutrients and essential elements.

Keywords: phenol, anthocyanin, amino acids, sugars, fatty acids, minerals

In dieser Studie wurden chemische Analysen durchgeführt sowie die Aminosäuren-, Zucker- und Fettsäurezusammensetzung und den Mineralgehalte von weißen und schwarzen Myrtenbeeren (Myrtus communis L.) bestimmt. Der Rohproteingehalt der weißen und schwarzen Myrtenbeeren lag bei 5,99 und 5,45 % während der Ölgehalt mit 8,31 und 6,17 % bestimmt wurde. Zusätzlich wurden Phenol-, Flavonoid- und Anthocyangehalte der weißen und schwarzen Beeren zwischen 4253 und 4060 mg Gallus-Säureäquivalent/g, 94,4 und 121,1 mg Catechol-Äquivalent/g und 0,096 bzw. 1,667 mmol/g bestimmt. Der Asparaginsäuregehalt der weißen und schwarzen Beeren wurde mit 0,50 bzw. 0,45 % bestimmt. Darüber hinaus wurden die höchsten Glutaminsäure und Arginin Werte in weißen Myrtenfrüchten analysiert. Der Fructoseund Glukosegehalt von weißen und schwarzen Myrtenfrüchten lag bei 11,26 bzw. 11,53 % bis 13,02 bzw. 13,74 %. Der Linolein-, Palmitin- und Oleinsäuregehalt von weißen und schwarzen Myrtenfruchtölen wurde mit 72,48 und 68,34 %, 9,99 und 10,43 % bis 8,52 bzw. 9,02 % bestimmt. Der Phosphor-, Kalium-, Calcium- und Magnesium-Gehalt der schwarzen und weißen Beeren wurden mit 1335,10 und 1165,15 mg/kg bis 2714,16 und 2631,04 mg/kg, 191,13 und 245,28 mg/kg bis 837,86 bzw. 720,38 mg/kg bestimmt. Zusammenfassend kann festgehalten werden, dass Myrtenbeeren eine wichtige Quelle von Nährstoffen und essentiellen Spurenelementen ist

Schlüsselwörter: Phenol, Anthocyane, Aminosäuren, Zucker, Fettsäuren, Mineralien

Introduction

Myrtle (Myrtus communis L.) is an evergreen shrub (belonging to the Myrtaceae family), and it is found in Europe, Asia, Africa and America (Davis,1982). The myrtle fruits are used as a condiment (Canhoto et al., 1998). Minor elements have very important functions due to a key component of proteins which play a role in biochemical functions. The leaves, flowers and barks of the myrtle plants are important in the food and cosmetic industries (Chalchat et al.1998; Senatore et al. 2006). In addition, the essential oils of fresh and/or dried leaves are used in cosmetics, sauces, confectionary and beverage industries (Buhner,1998; Özcan and Chalchat, 2004). The chemical composition of myrtle leaves and berries essential oils were determined by several researchers (Bradesi et al. 1997; Asllani, 1998; Wannes et al., 2009; Ghannadi and Dezfuly, 2011). Myrtle had been used since ancient times for medicinal, food and spices purposes (Asllani, 1998; Ghannadi and Dezfuly, 2011; Sümbül et al. 2011; Ghnaya et al. 2013). However, there are limited studies on oil contents, fatty acid composition, protein and amino acid, composition, sugar composition, mineral contents of white and black myrtle fruits growing in Turkey. The objective of this study was to determine chemical analysis, amino acids, sugar pofile, fatty acid composition and mineral contents of white and black myrtle (Myrtus communis L.) fruits.

Material and methods

The black and white fruits of myrtle plants were provided from the Antalya (Serik) province in Turkey. The fruits were transported to a laboratory in cool polypropylene bags (+4 °C), and were dried to constant weight in room temperature for analyses. About 1 kg homogenized and dried fruits were kept at +4 °C till analysing. A specimen was deposited in the department of Food Engineering, University of the Selçuk in Konya in Turkey.

Moisture and protein contents of myrtle fruits were determined according to AACC approved methods 44–15.02 and 46–30.01 respectively. Moisture was measured at 135 °C in a gravity oven for 1 h (AACC International, 1999a). The protein determination was made in a Leco combustion analyzer and 6.25 was used as the conversion factor (AACC International,1999b). For oil concentration analysis, about 10 g of the dried fuits were ground in a ball milled, and extracted with petroleum ether in a Twisselmann apparatus for 6 h. The solvent was removed by a rotary evaporator at 40 °C and 25 Torr. The oil were dried by a stream of nitrogen and stored at –20 °C until used (AOCS, 1998).

The sugar contents were determined by chromatographic methods (Churms et al., 1982; Kakehi and Honda, 1989). The amino acid contents of myrtle fruits was determined according to AOAC Official Method 982.30 E(a,b,c) (International AOAC et al., 2006).

Fatty acid composition for myrtle fruit oils was determined using a modified fatty acid methyl ester method. The oil sample (50–100 mg) was converted to its fatty acid methyl esters (FAME). The methyl esters of the fatty acids (1 μ l) were analysed in a gas chromatography (HP 6890) equipped with a flame ionising detector (FID), a fused silica capillary column (60 m x 0.25 mm i. d.; film thickness 0.20 micrometer). It was operated under the following conditions: oven temperature program. 175 °C for 7 min. Raised to 250 °C at a rate 5 °C/min and than kept at 250 °C for 15 min); injector and detector temperatures, 250 and 250 °C; respectively, carrier gas. nitrogen at flow rate of 1.51 ml/min; split ratio. 1/50 μ l/min (International AOAC et al., 2006).

For mineral concentration analysis about 0.5 g sample dried at 70 °C in a drying cabinet with air-circulation ground samples were digested by using 5ml of 65% HNO₃ and 2 ml of 35 % H_2O_2 in a closed microwave system (Cem-MARS Xpress). The volumes of the digested samples were completed to 20 ml with ultra-deionized water, and mineral contents were determined by inductively coupled plasma atomic emission spectroscopy (ICP AES) (Varian-Vista, Australia). Measurements of mineral concentrations were checked using the certified values of related minerals in the reference samples received from the National Institute of Standards and Technology (NIST; Gaithersburg, MD, USA) (Skujins, 1998).

Working conditions of ICP-AES

8	
Instrument:	ICP-AES (Varian-Vista)
RF Power:	0.7-1.5 kw (1.2-1.3 kw for Axial)
Plasma gas flow rate (Ar):	10.5–15 L/min. (radial) 15 " (Axial)
Auxilary gas flow rate (Ar):	1.5 "
Viewing height:	5–12 mm
Copy and reading time:	1- 5 s (max. 60 s)
Copy time:	3 s (max. 100 s)

Anthocyanin contents of myrtle fruits were analyzed according to the method of Ticconi et al. (2001). 0.5 g fresh weight (FW) was homogenized in a solution containing propanol, chlorhydric acid and water (18:1:81). The resulting homogenates were boiled in a water bath for 3 min, and then left in darkness for 24 h at room temperature. 3 mL of the supernatants were centrifuged at 6500 rpm for 40 min as the accelaeration depends on the rotor of the centrifuge. Finally, the absorbancies of the samples were measured at 535 and 650 nm. The absorbance value was calculated and corrected by the following formula:

 $A = A_{535} - A_{650}$

The phenols of the myrtle fruits were extracted with MeOH. Total phenolic content was assayed quantitatively by absorbance at 765 nm with Folin-Ciocalteau reagent according to the method of Madaan et al. (2011). Firstly, a standard curve of known concentrations of gallic acid was prepared to calculate the total phenolic content to be expressed as gallic acid equivalent (GAE). Ten mg of gallic acid were dissolved in 100 mL of 50 % methanol (100 $\mu\text{g/mL})$ and then diluted to 12.5, 25, 50 or 100 $\mu\text{g/mL}.$ 0.076 mL aliquot of each dilution was taken in a test tube and diluted to 0.76 mL of distilled water. Then 0.12 mL FolinCiocalteu's reagent (1 N) was added and allowed to incubate at room temperature for 5 min. 0.32 mL of 20 % (w/w) Na₂CO₂ was added in each test tube, adjusted with distilled water up to the mark of 2 mL, vortexed and left to stand for 30 min at room temperature. Absorbance of the standard was measured at 765 nm using UV/VIS spectrophotometer (Schimadzu, Japan) against blank. For measurement of plant samples, appropriately diluted methanolic extracts of 0.76 mL were taken into test tubes and then a similar procedure was followed with the standards.

Total flavonoid contents of myrtle fruits were determined according to Dewanto et al. (2002). Methanol

extracts of fruits were diluted with distilled water. 5 % $NaNO_2$ solution was added to each test tube; after five minutes, 10 % AlCl₃ solution was added and then after six minutes 1.0 M NaOH was added. Finally, total volume was filled up to 5 mL with water and the test tubes were mixed well. Absorbance of the resulting pink-colored solution was measured at 510 nm versus blank. Calibration curve was prepared using Catechol as standard. The flavonoid content was expressed as mg Catechol equivalents (CE) per g of dry weight (mg CE/g DW).

A complete randomized split plot block design was used analysis of variance (ANOVA) was performed by using JMP version 9.0 (SAS Inst. Inc., Cary, N.C.U.S.A). The results are mean±standard deviation (MSTAT C) of independent myrtle fruit samples (Püskülcü and İkiz, 1989).

Results and Discussion

The chemical properties of white and black myrtle fruits are given in Table 1. The moisture, protein and oil contents of white and black myrtl fruits were determined as 8.20 and 8.59 %, 5.99 and 5.45 % to 8.31 and 6.17 % respectively (p<0.05). Total phenol, flavonoid and anthocyanin contents of white and black myrtle fruits were reported as 4253 and 4060 mg GA/g, 94.4 and 121.1 mg catechol/g to 0.096 and 1-667 µmol/g respectively. It was observed opposite relation between total phenol and flavonoid contents of myrtle fruit extracts, and statistically differences were observed (p<0.05 While phenolic acid values of myrtle fruit changed between 67.9 mg/100 g and 322.8 mg/100 g, total polyphenol values of myrtle fruits ranged from 2186.7 mg/100 g to 6743.3 mg/100 g (Barboni et al. 2010). The highest total flavonoid and anthocyanin contents were found in black myrtle berries. In previous study, polyphenol compounds were extracted from Myrtus communis L. berries (Myrtaceae) by maceration in 70 % ethanol and analysed by HPLC-DAD and electrospray mass spectrometry (Barboni et al. 2010). The polyphenol composition of Corsican Myrtus berries was characterized by two phenolic acids, four flavanols, three flavonols and five flavonol glycosides. The major compounds of Myrtus communis L. berries were myricetin-3-O-arabinoside and myricetin-3-O-galactoside. Piras et al. (2009) reported that the differences in the concentration of many compounds can be probably due to geoclimatic factors, genetic and/or environmental factors, the quantitative composition of myrtle berry extract and irrigation.

The amino acid composition of white and black myrtle fruits are shown in Table 2. Twenty-three amino acids in both samples were established. While amino acid contents of white myrtle fruits change between 0.01 % (ornithine) and 0.87 % (glutamic acid), amino acid contents of black myrtle ranged from 0.01 % (ornithine) to 0.74 % (glutamic acid) (p<0.05). Aspartic acid contents of white and black myrtle fruits were determined as 0.50 and 0.45 %, respec-

TABLE 2: Amino acid profile of white and black myrtle fruits (g/100; dw).

Amino acids	White myrtle	Black myrtle
Taurine	0.05±0.01*a	0.04±0.01b
Hydroxyproline	0.07±0.02a**	0.07±0.01a
Aspartic Acid	0.50±0.07a	0.45±0.03b
Threonine	0.13±0.02a	0.13±0.03a
Serine	0.18±0.03a	0.16±0.01b
Glutamic Acid	0.87±0.09a	0.74±0.07b
Proline	0.19±0.03a	0.18±0.05b
Lanthionine	_***	-
Glycine	0.38±0.03a	0.32±0.07b
Alanine	0.20±0.01a	0.19±0.03b
Cysteine	0.15±0.02a	0.12±0.01b
Valine	0.22±0.05a	0.20±0.03b
Methionine	0.06±0.02a	0.06±0.01a
Isoleucine	0.19±0.03a	0.17±0.05b
Leucine	0.38±0.07a	0.33±0.02b
Tyrosine	0.17±0.02a	0.14±0.03b
Phenylalanine	0.21±0.05a	0.19±0.03b
Hydroxylysine	0.03±0.01a	0.03±0.01a
Ornithine	0.01±0.00a	0.01±0.00a
Lysine	0.19±0.03a	0.19±0.01a
Histidine	0.13±0.02a	0.12±0.01b
Arginine	0.56±0.09a	0.47±0.05b
Tryptophan	0.04±0.01a	0.04±0.01a
Total	4.87	4.31

*: mean±standard deviation (n:3), **: Values within each column followed by different letters are significantly different (p<0.05), ***: non identified

tively. In addition, the highest glutamic acid and arginin were found in white myrtle fruits. Also, aspartic acid, glycine, leucine and arginine in both myrtle samples were found partly high. As a total amino acid, white and black myrtle berries contained 4.87 and 4.01 %, respectively.

The sugar compositions of myrtle fruits are presented in Table 3. While the fructose and glucose contents of white myrtle change between 11.26 and 11.53 %, fructose and

TABLE 3: Sugar composition of white and black myrtle fruits (%, w/w).

Sugars	White myrtle	Black myrtle	
Fructose	11.26±1.12*b	13.02±1.27a	
Glucose	11.53±1.32b**	13.74±1.18a	
Sucrose	0.02±0.01b	0.03±0.01a	
Raffinose	0.02±0.01a	0.02±0.01a	
Stachyose	0.03±0.01b	0.08±0.01a	

*: mean±standard deviation (n:3), **: Values within each column followed by different letters are significantly different (p<0.05)

TABLE 1:	Proximate ana	lysis of white and	l black myrtle fruits	(dwb: dry weight basis).

Samples	Moisture (%)	Protein (%)	Oil (%)	Ash (%)	Anthocyanin (µmol/g)	Total phenol content (mg Gallic acid/g)	Total flavonoid content (mg catechol g ⁻¹)
Black myrtle	8.59 ± 0.95*a	5.45 ± 0.73b	4.58 ± 0.98b	3.15 ± 0.01a	1.67 ± 0.53a	4060 ± 166b	121.1 ± 10.5a
White myrtle	8.20 ± 0.78b**	5.99 ± 0.47a	5.55 ± 0.79a	2.99 ± 0.02b	0.096 ± 0.00b	4253 ± 96a	94.4 ± 3.3b

*: mean±standard deviation (n:3), **: Values within each column followed by different letters are significantly different (p<0.05)

glucose contents of black myrtle ranged from 13.02 to 13.74 %, respectively (p<0.05). Fructose and glucose contents of white and black myrtle fruits were reported as 11.26 and 11.53 % to 13.02 and 13.74 %, respectively. Other sugars were found at low levels (Table 3). Generally, sugar content of black myrtle was found higher compared with results of white myrtle berry.

The fatty acid composition of myrtle fruit oils were summarized in Table 4. The highest fatty acid had linoleic acid for both myrtle fruit oils, followed by palmitic, oleic and stearic acids. Linoleic, palmitic, oleic and stearic acid contents of white and black myrtle fruit oils were determined as 72.48 and 68.34 %, 9.99 and 10.43 %, 8.52 and 9.02 % and 3.61 and 3.54 % respectively (p<0.05). Linoleic acid content of white myrtl oil was found partly high (72.48 %) compared to other myrtle oil (p<0.05). In previous study, the predominant fatty acids of myrtle beries were linoleic (12.21–71.34 %), palmitic (13.58–37.07 %) and oleic (6.49-21.89 %) acids (Wannes et al., 2009). In a previous studies, literature values illustrated that polyunsaturated fatty acid (PUFA) helped prevent to the cardiovascular inflammatory, heart diseases, atherose lerosis, autoimmune disorder, diabetes and other diseases (Finley and Shahidi, 2001). The fatty acid composition and the high contents of PUFA contents of the myrtl fruit lipids are very important for a variety of healthy applications. Generally, fatty acid compositions of myrtle oil were found partly similar. Results were found similar with literature values. Minor differences can be probably due to locations, climatic factor and harvest time.

The mineral contents of myrtle fruits are given in Table 5. P, K, Ca, Mg and S contents of black and white myrtle fruits were determined as 1335.10 and 1165.15 mg/kg, 2714.16 and 2631.04 mg/kg, 191.13 and 245.28 mg/kg, 837.86 and 720.38 mg/kg to 757.90 and 807.81 mg/kg respectively (p<0.05). Na contents of both samples had been found as 32.11 and 33.66 mg/kg respectively. Cd was not found in myrtle samples. Cr, Ni, Pb, Mo and Se were found at too low levels. Pb contents of black and white fruits were found as 0.66 and 0.72 mg/kg respectively. Fe, Zn and Mn contents of black and white fruits changed between 25.51 and 26.23 mg/kg, 6.54 and 25.75 mg/kg to 19.01 and 18.79 mg/kg respectively (p<0.05). Cu contents of both samples were found as 3.06 and 5.11 mg/kg, respectively. Özcan and Akbulut (2007) reported that myrtle berry contained 65.25 ppm A1, 12.40 ppm B, 5639.70 ppm Ca, 0.77 ppm Cd, 2.66 ppm Cr, 44.83 ppm Fe, 5849.05 ppm K, 1937 ppm Mg, 15.81 ppm Mn, 983.66 ppm Na, 3.72 ppm Ni, 443.60 ppm P and 14.01 ppm Zn (p<0.05). Results were found partly similar. Lead and Cadmium cause both acute and chronic poisoning, adverse effects on the kidney, liver, heart, vascular and immune system (Heyes, 1997). Decreasing these toxic element contents is an advantage for human consumption.

Conclusion

The highest total flavonoid and anthocyanin contents were found in black myrtle berry. Other sugars were found at low levels (Table 3). Generally, sugar content of black myrtle was found higher compared with results of white myrtle berry. Twenty-three amino acids in both samples were established. The highest glutamic acid and arginin were found in white myrtle fruits. In addition, aspartic acid,

TABLE 4:	Oil contents and fatty acid composition of white
	and black myrtle fruits (%, w/w).

Fatty acid and oil	White myrtle	Black myrtle
Palmitic (C16:0)	9.99±0.87*b	10.43±1.07a
Stearic (C18:0)	3.61±0.45a**	3.54±0.89b
Oleic (9c-18:1)	8.52±1.13b	9.02±1.27a
Linoleic (18:2n6)	72.48±2.38a	68.34±2.56b
Linolenic (18:3n3)	0.73±0.11b	1.28±0.13a
Arachidic (C20:0)	0.88±0.13b	0.95±0.07a
Behenoic (C22:0)	0.47±0.19b	0.70±0.11a
Lignoceric (C24:0)	0.37±0.09b	0.52±0.05a
Oil contents	8.31±0.98a	6.17±1.13b

*: mean±standard deviation (n:3), **: Values within each column followed by different letters are significantly different (p<0.05)

TABLE 5: *Mineral contents of myrtle fruits (mg/Kg; dwb*).*

Minerals	White myrtle	Black myrtle
Р	1165.15±11.43b	1335.10±55.03**a
К	2631.04±8.16b	2714.16±9.00a***
Ca	245.28±7.66a	191.13±2.15b
Mg	720.38±10.46b	837.86±6.94a
S	807.81±4.50a	757.90±4.99b
Fe	26.23±2.04a	24.51±1.80b
Zn	25.75±1.61a	6.54±2.11b
Mn	18.79±1.08b	19.01±1.42a
В	2.35±1.96b	5.51±1.21a
Cu	5.11±0.66a	3.06±0.40b
Мо	0.35±0.06b	0.59±0.02a
Na	33.66±0.32a	32.11±1.79b
Cd	-	_****
Cr	0.14±0.01b	0.15±0.01a
Ni	0.35±0.04b	0.36±0.02a
Pb	0.72±0.01a	0.66±0.01b
Se	0.58±0.00b	2.12±0.01a

*dwb: Dry weight basis; **: mean±standard deviation (n:3), ***: Values within each column followed by different letters are significantly different (p<0.05), ****: non identified

glycine, leucine and arginine in both myrtle samples were found partly high. The highest fatty acid had linoleic acid for both myrtle fruit oils, followed by palmitic, oleic and stearic acids. Linoleic acid content of white myrtl oil was found partly high compared to other myrtle oil. P, K, Ca, Mg and S were the major elements of black and white myrtle fruits. Cd was not found in myrtle samples. Cr, Ni, Pb, Mo and Se were found at too low levels. The results also show that myrtl berries contain several bioactive components and minerals of vital importance in human metabolism and that are needed for growth and developments prevention and healing of diseases. This study is to contribute to knowledge of the nutritional properties of some aromatic plants growing wild in Turkey. In conclusion, myrtle fruits were found to be important sources of nutrients and essential elements.

Conflict of interest

No conflict of interest among authors.

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