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Effect of Altitude and Location on Compositions and Antioxidant Activity of Laurel Cherry (*Prunus Laurocerasus* L.)

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Abstract: The compositional and antioxidant properties of cherry laurel *(Prunus laurocerasus* L.) fruit, which grow in two different locations (Trabzon and Rize) and altitudes, were investigated. The results indicated that antioxidant activity, total phenolic content, citric acid, sugars and phenolic compounds were affected by location and altitude. While fruits of Trabzon province have higher composition content than those of Rize province, fruits collected at low altitudes in both regions were found to have superior properties. Total phenolic content in fruits from Trabzon and Rize provinces increased from 21.90 to 23.32 and from 16.84 to 18.91 mg gallic acid equivalent / 100 g dry weight (DW), β-carotene increased from 5.19 to 6.75 and from 4.16 to 5.61 mg / kg DW, and total sugar increased from 81.68 to 131.99 and from 86.44 to 99.58 mg / g DW when altitude decrease from 351 to 49 m and 316 to 14 m, respectively. Chlorogenic acid (1404.46-7358.63 mg / kg DW) and rutin hydrate (1491.05-2712.91 mg / kg DW) were major phenolic compounds in all samples.

Keywords: Altitude, Antioxidant activity, Cherry laurel, Phenolic composition, Location

Karayemiş Meyvesinin (*Prunus Laurocerasus* L.) Bileşimi ve Antioksidan Aktivitesi Üzerine Yükseklik ve Konum Etkisi

Öz: Bu çalışma kapsamında Trabzon ve Rize bölgelerinin farklı yüksekliklerinde yetiştirilen karayemiş (*Prunus laurocerasus* L.) meyvesinin bileşim ve antioksidan özellikleri incelenmiştir. Elde edilen sonuçlara göre meyvenin toplam fenolik madde miktarı, bireysel fenolik bileşenleri, şeker, sitrik asit içeriği ve antioksidan aktivitesinin yetiştirildiği yer ve rakımdan oldukça etkilendiği belirlenmiştir. Trabzon bölgesinde yetiştirilen meyve Rize bölgesinde yetiştirilene kıyasla daha yüksek bileşen miktarına sahip iken, en yüksek bileşen kompozisyonu her iki bölgenin düşük rakımında yetiştirilen meyvelerinde belirlenmiştir Trabzon ve Rize bölgelerinde rakımın sırası ile 351 den 49 m'ye ve 316'dan 14 m'ye düşmesi ile toplam fenolik içeriğinin 21.90 dan 23.32'ye ve 16.84'den 18.91 mg gallik asit miktarı eşdeğeri / 100 g kuru madde (KM)'ye yükseldiği, β-karoten miktarının 5.19'dan 6.75'e ve 4.16'dan 5.61 mg / kg KM'ye yükseldiği, toplam şeker miktarının ise 81.68'den 131.99'a ve 86.44'den 99.58 mg / g KM'ye yükseldiği belirlenmiştir Klorojenik asit (1404.46-7358.63 mg /kg KM) ve rutin (1491.05-2712.91 mg / kg KM) tüm örneklerde baskın fenolik bileşik olarak belirlenmiştir.

Anahtar Kelimeler: Antioksidan aktivite, Bölge, Fenolik bileşim, Karayemiş, Yükseklik

1. Introduction

Cherry laurel (*Prunus laurocerasus* L.), belongs to *Rosaceae* family and Prunus genus, generally grows wildly in Europe, Asia, the Balkans, Iran and the East Black Sea Region of Turkey. The fruit is used in various products such as jam, pekmez (a boiled fruit juice), marmalade and fruit juice as well as fresh or dried consumption (Alasalvar et al., 2005; Ozturk et al., 2017). In addition, cherry laurel has been also used for medicinal purposes in Turkey for many years due to its positive effects on health (Alasalvar et al., 2005). Cherry laurel is used in many countries as a traditional medicine to treat disorders abdominal pain, cough, bronchitis, stomach ulcers, hemorrhoids and eczema nausea as well as strengthening of bones, reduction of kidney stones, the establishment of acid-base balance in bloodstream (Ozturk et al., 2017). Diets with a high amount of fruits and vegetables are recommended to reduce the risk of various chronic diseases like cancer, coronary heart disease, and cardiovascular disease (Mphahlele et al., 2014). Recently, many researchers have been focused on elucidating mechanisms of bioactive compounds in fruits. These studies contribute to understanding the effect of fruit genotype and growth conditions on compositional properties (Crespo et al., 2010). Although the biosynthesis and accumulation of phytochemicals in fruits (mainly phenolic compounds) can be endogenously (such as genotype) controlled during developmental differentiation, the differences in the amount and variety of them emerge with the effect of many exogenous factors. The main exogenous factors are as follows: growing conditions (light, temperature,

irrigation, altitude, etc.), pre-harvest environmental conditions, level of maturity and storage conditions (Alasalvar et al., 2005; Crespo et al., 2010; Topalovic and Mikulic-Petkovsek, 2010; Mphahlele et al., 2014).
In literature, there are a lot of studies on different properties of cherry laurel such as chemical composition, antioxidant activity (Alasalvar et al., 2005; Celik et al., 2011;Yıldız et al., 2014), bioactive contents (Celik et al., 2011; Yıldız et al., 2014) and change in fruit quality at different storage temperatures (Ozturk et al., 2017). However, no study has been conducted on the changes in composition and antioxidant capacity of fruit which grow in different locations and altitudes. This study aims to investigate the fruit composition (phenolic content, β-carotene, organic acids and sugars) and antioxidant activity of cherry laurel grown in different locations and altitudes.

2. Material and methods

2.1. Samples preparation

The samples were collected from two different districts of Trabzon and Rize provinces of the East Black Sea Region located in the northeastern part of Turkey. At least 5 kg of fruit from 6 tree per locations collected on August 2018 from Trabzon Low (TL; 1° 0' 9.70" N latitude, 39° 43' 0.34"E longitude and altitude is 49 meters,), Trabzon High (TH; 40° 48' 51.19" N latitude 39° 36' 38.37" E longitude and altitude is 351 meters), Rize Low (RL; 41° 1' 31.84"N latitude 40° 31' 3.59"E longitude and altitude is 14 meters) and Rize High (RH; 1° 2' 53.53"N latitude 40° 53' 56.61"E longitude and altitude is 316 meters). Samples were packed in ice in an insulated container and transferred to the laboratory in the cold chain. Upon arrival to the laboratory, samples were homogenized with a Warring blender (Model HGB2WTS3, Connecticut, USA) after pre-treatments such as manual sorting, washing, removing of seeds by hand and used for analysis.

2.2. Total phenolic content and antioxidant capacity

The extraction was performed according to the method described by Veberic et al. (2009). Total phenolic contents were determined by the Folin–Ciocalteau procedure described by Krayjalyte et al. (2013) with minor modifications. In brief, 1 mL of 0.2 N Folin–Ciocalteau regent and 400 µL of distilled water were added to 100 µL extract. After 3 min incubation, 1 mL of Na_2CO_3 solution (7%) was added and incubated for 90 min at room temperature in dark. At the end of incubation time, the absorbance of the sample was measured at 725 nm using an UV-1700 spectrophotometer (Shimadzu, Kyoto, Japan). Total phenolic content was calculated by a five-point calibration curve prepared by gallic acid and results were expressed as mg gallic acid equivalents (GAE) per 100 g of dry weight (DW) samples.

The radical scavenging capacity of samples against DPPH (1,1-diphenyl-2-picrylhydrazyl) radical was determined according to the method of Brand-William et al. (2005) with minor modifications. In brief, extract (100 µL) was mixed with 1.9 mL of DPPH methanolic solution (1mg/mL) and incubated for 60 min in the dark at room temperature. After the incubation period, absorbances were measured at 520 nm. A calibration curve was constructed by measuring the absorbance of five concentrations of trolox. Results were expressed in µmol trolox / 100g DW.

The ABTS (2.2'-azinobis [3-ethylbenzthiazoline-6-sulphonic acid]) antioxidant activity was determined according to Re et al. (1999). In brief, The ABTS•+ was prepared by mixing an ABTS stock solution (7 mM in water) with 2.45 mM potassium persulfate in dark for 16h. The stock solution was diluted with ethanol to an absorbance of 0.7000 ± 0.020 at 734 nm. A total of 200 µL extract was added to the ABTS radical solution (3.8 mL). The absorbance at 734 nm was measured after incubation for 60 min at room temperature. A calibration curve was constructed by measuring the absorbance of five concentrations of trolox. Results were expressed in µmol trolox / 100g DW.

2.3. HPLC analyses

Shimadzu HPLC system integrated with an autosampler (DGU2A 5R) including a column oven (CTO-10AS VP), a degasser system (DGU2A 5R), a gradient pump (LC-20AR), 175 a diode-array detector (DAD, SPD-M20A), a refractive index detector (RID-10A), and a 176 software package for system control and data acquisition (LC solution) was used for the following analyses. The analysis was performed in triplicate for each sample and results were expressed as mean ± SD

2.3.1. β-carotene analysis

The β-carotene extraction of samples was carried out according to the method of Sadler et al. (1990) with slight modifications. Obtained extract (20 µL) was injected into HPLC system, separations were carried out according to method of Barba et al. (2006). Isocratic elution was used on inertsil ODS-2 column (250x4.6mm, 5µm, GL Sciences INC., Tokyo, Japan) for separation at the flow rate of 1 mL/min, column

temperature at 32°C, with mobile phase consisting of methanol: acetone (70:30 v / v). Quantification was performed by using a calibration curve prepared with the five concentrations of β-carotene standard solutions and results were expressed as mg β-carotene / g DW.

2.3.2. Organic acid and Sugar

For extraction, 5 g of fruit puree was diluted fivefold with ultra-pure water and the mixture was homogenized with Ultra Turrax T18 (Ika Staufen, Germany) at 13000 rpm for 1 min. After centrifugation (25155 g) for 20 min at 4°C, the supernatant was filtered through 0.45 µm nylon (Lubitech, Songjiang, China) syringe filter. The filtrate was used in sugar and organic acid analyses.

Organic acids were determined with HPLC system using Rezex ROA column (300 x 7.8; Phenomennex, Torrance, CA), UV dedector (210 nm for malic and citric acids, 245 nm for ascorbic acid), according to the method described by Sturm et al. (2003). The flow rate was 0.5 mL/min, the mobile phase was 0.0025 M sulphuric acid, column temperature was 55°C and isocratic flow was used for elution. For quantification, calibration curve that was prepared with malic acid, citric acid and ascorbic acid standards at five different concentrations was used. The results were expressed as mg organic acid / g DW.

Sugar and sorbitol analyses were performed according to HPLC method of Sturm et al. (2003) with slight modifications. A refractive index (RI) detector was used for determinations of sugar and sorbitol contents. Isocratic elution was used on Rezex RCM-monosaccharide column (300 x 7.8; Phenomennex, Torrance, CA) separation at the flow rate of 0.6 mL/min, column temperature at 80°C, with mobile phase consisting of ultra-pure water (100%). Quantification was performed by using a calibration curve prepared with the five concentrations of a mixture consisting of glucose, fructose, sucrose and sorbitol and results were expressed as mg / g DW.

2.3.3. Phenolic compounds

The extraction of phenolic compounds was carried out according to the method of Kim and Lee (2002).Separation of phenolic compounds was performed according to the method of Campbell and Padilla-Zakour (2013) with minor modifications. Aliquots (20 µL) of each extract were injected into HPLC system consists of Inertsil ODS-3 column (250 nm x 4.6mm, 5µm: GL, Sciences INC., Tokyo, Japan). A linear solvent gradient was composed of a binary mobile-phase system with solvent A, 0.1% phosphoric acid in ultrapure water, and solvent B, 0.1% phosphoric acid in HPLC-grade acetonitrile at a flow rate of 1 mL/min and column temperature of 25°C. Elution program was applied for 55 min as follows: 92% A at 0 min, 89% A at 4 min, 65% A at 25 min, 40% A-60% B at 30 min, 40% A-60% B at 40 min, 65% A-35% B at 45 min, 89% A at 50 min, 92% A at 55 min. Phenolic compounds were detected by using the DAD and data collection was monitored at 280, 320 and 360 nm. The identification of phenolic compounds was done by comparing the retention time of authentic standards. The calibration curve prepared by using five different concentrations of phenolic standards mixture was used for the quantification. Results were expressed as mg phenolic compound / kg DW.

2.4.Statistical analysis

The effect of different locations and altitudes on the fruit quality parameter was analyzed by analysis of variance (ANOVA). Duncan's multiple-comparison test was used as a tool for comparisons of means at a level of $p < 0.05$ using the SPSS package program version 16.0 (SPSS Inc., Chicago, IL, USA).

3. Results and Discussion

3.1. Total phenolic content and antioxidant capacity

The total phenolic contents of four cherry laurel samples are presented in Table 1. Total phenolic contents were varied from 16.84 to 23.32 mg GAE / 100 g DW. An increase in the total phenolic content of both locations was noted with lower altitude. The phenolic contents of the fruits grown at low and high altitudes were in the range of 18.91 (RL)-23.32 (TL) mg GAE / 100 g DW and 16.84 (RH)-21.90 (TH) mg GAE / 100 g DW respectively. Contradictory results in total phenolic contents of cherry laurel are available in the literature about the total phenolic contents of cherry laurel. Karabegović et al. (2014) found higher total phenolic content (36.2 to 46.3 mg GAE / g DW) than those reported (1.094 mg GAE / 100 g DW) by Karahalil and Şahin (2011). This may be due to geographical factors and analytical differences. The amount of phenolic substances in the fruit is governed by a few factors such as temperature, growing location, light intensity, altitude and age of tree (Mphahlele et al., 2014; Karabegović et al., 2014; Coklar, 2017). Total phenolic contents of cherry laurel samples collected from both altitudes of Trabzon province higher than those from Rize province ($p < 0.05$). This may be related to the difference in the annual rainfall amounts of two

provinces, therefore, there will be a difference in exposing time to sunlight. According to the Turkish State Meteorological Service report for 2017 year, Rize province takes approximately 1.5 times higher rainfall than that of Trabzon province. (Turkish State Meteorological Service, 2018). This indicates that Trabzon province receives more sunshine than that of Rize province. In this context, Pereira at al. (2006) reported that the sun exposed (high light intensity) berries have more phenolic content than those growing in the shade (low light intensity). Additionally, as shown in Figure 1, the color of the fruits from Trabzon province at both altitudes is darker than the fruits from Rize province. It is well known that the phenolic constituents contribute to the color in fruits (Topalovic and Mikulic-Petkovsek, 2010).

Figure 1. Cherry laurel samples from different location and altitudes; Trabzon High (TH), Trabzon Low (TL), Rize High (RH), Rize Low (RL)
Şekil 1. *Farklı bölge ve yüksekliklerde yetiştirilen karayemiş örnekler; Trabzon Yüksek (TY), Trabzon Düşük (TD), Rize Yüksek (RY), Rize Düşük (RD)*

Antioxidant activities determined by DPPH and ABTS of the samples are shown in Table 1. DPPH scavenging activities of the samples from Trabzon and Rize provinces were varied from 23.09 (TH) to 24.60 (TL) µmol trolox / 100 g DW and 19.71 (RH) to 22.12 (RL) µmol trolox / 100 g DW, respectively. There was a statistically significant difference in DPPH radical scavenging activity of the samples from the standpoint of harvesting province and altitude ($p < 0.05$). The fruits collected from Trabzon province at high and low altitudes have higher ABTS values (14.22 and 14.42 µmol trolox / 100 g DW, respectively) than those collected from Rize province (11.54-11.81 µmol trolox / 100 g DW, respectively). ABTS radical scavenging activities of Trabzon samples were higher than those of Rize province, while there was no significant change ($p > 0.05$) with the changes in altitudes. Mditshwa et al. (2013) reported that there were significant differences in DPPH antioxidant activity and total phenolic content of pomegranate fruit grown increased as altitude decreased. In accordance with our findings, Coklar (2017) found that the DPPH antioxidant activity of Eksikara grape decreased from 53.86 to 23.34 mmol TE / kg DW with altitude increased from 1000 to 1500 m. After approximate conversion of fresh weight (FW) to DW, antioxidant activity results of cherry laurel fruit are in accordance with previous studies who reported that DPPH scavenging activity of the fruit was in the range of 14.0 and 43.54 µmol trolox / 100 g FW (Celik et al., 2011; Yıldız et al., 2014; Ozturk et al., 2015). However, in a previous study, considerably higher values, which changed from 17.56 to 23.21 µmol trolox /g FW, were reported by ABTS assay (Ozturk et al., 2017).

There was a strong correlation between the total phenolic content and antioxidant capacity by DPPH ($r= 0.941$) and ABTS ($r= 0.944$), respectively. The phytochemicals like phenolic compounds, ascorbic acid, β-carotene, etc are known for their high antioxidant activity potential) (Mphahlele et al., 2014; Ozturk et al., 2017; Coklar , 2017).

3.2. HPLC analysis

3.2.1. β-carotene content

β-carotene contents of four cherry laurel samples are presented in Table 1. The β-carotene contents of the samples collected at low and high altitudes were in the range of 5.61 (RL) – 6.75 (TL) mg / kg DW and 4.16 (RH) -5.19 (TH) mg / kg DW, respectively. In accordance with the observations for the total phenolic content, cherry laurel fruit growing at low altitude conditions contain higher amounts of β-carotene than those growing at higher altitude conditions ($p < 0.05$). Similarly, cherry laurel fruit collected from Trabzon province has more β-carotene compared to those collected from Rize province. It is thought that the time of exposing sunlight and the amount of rainfall have an

impact on beta carotene content as discussed in the phenolic substance change. This result agrees with that of reported by Macar and Macar (2018) who concluded that the carotenoid content of Polygonum cognatum plant was 1.5 times higher from low altitude, compared with the higher one.

Previous studies have indicated that cherry laurel fruit is a good source of carotenoid with the total carotenoid concentration ranged from 206 to 274 mg 100 g FW (Alasalvar et al., 2005; Celik et al., 2011; Yıldız et al., 2014). In this study, only β-carotene content was determined and the total amount of carotenoid was expected to be higher. However, the differences between the findings may be explained by the environmental factors, plant varieties, age of tree, maturity level of fruit and post-harvest conditions or storage (Karabegović et al. 2014) as well as the differences in analytical techniques. It is well known that the β-carotene acts as an antioxidant by quenching free radicals and singlet oxygen. In this regard, as shown in Table 1, the higher the amount of beta carotene, the higher the DPPH antioxidant activity was found. There was a positive correlation between the DPPH scavenging activity and β-carotene (r=0. 882). A similar result was reported by Celik et al. (2011) who reported a positive correlation between the DPPH antioxidant activity and total carotenoids (r= 0.843).

Table 1. Antioxidant (DPPH and ABTS) capacity, total phenolic and β-carotene contents of cherry laurel samples grown in different locations and altitudes

Çizelge 1. *Farklı bölge ve yüksekliklerde yetiştirilen karayemiş örneklerinin, antioksidan (DPPH ve ABTS) kapasiteleri, toplam fenolik ve β-karoten içeriği*

Samples	Antioxidant Properties			
	Total Phenolic content (mg GAE/ 100 g DW)	DPPH (µmol trolox / 100 gDW)	ABTS (µmol trolox / 100 g DW)	β-carotene (mg /kg DW)
TH	21.90± 0.04c	23.09±0.08bc	14.22± 0.12b	5.19± 0.44b
TL	23.32± 0.04d	24.60±0.02c	14.42± 0.02b	6.75± 0.02c
RH	16.84± 0.40a	19.71±1.23a	11.54± 0.07a	4.16± 0.17a
RL	18.91± 0.21b	22.12±0.05b	11.81± 1.02a	5.61± 0.08b

Means with different letters in the column for each position and cherry laurel samples are significantly different ($p < 0.05$), Trabzon High (TH), Trabzon Low (TL), Rize High (RH), Rize Low (RL), Dry Weight (DW)

Table 2. Organic acids (mg g DW-1), sugar and sorbitol (mg g DW-1) contents of cherry laurel samples grown in different locations and altitudes

Çizelge 2. *Farklı bölge ve yüksekliklerde yetiştirilen karayemiş örneklerinin organic asit (mg g KM-1), şeker ve sorbitol (mg g KM-1) içeriği*

Samples	Organic Acids (mg /g DW)			Sugars and Sorbitol (mg / g DW)			
	Malic acid	Citric acid	Ascorbic acid	Glucose	Fructose	Sorbitol	Total Sugar
TH	124.72 ± 0.13c	26.93 ± 0.98c	0.46 ± 0.00c	36.87 ± 3.48a	30.54 ± 1.73a	14.26 ± 0.30b	81.68 ± 1.44a
TL	117.01 ±2.71 b	27.16 ± 1.03c	0.43 ± 0.00a	51.75 ± 0.51b	47.32 ± 0.01c	32.93 ± 0.08d	131.99 ± 0.60d
RH	119.86 ± 1.02bc	12.97 ± 0.57a	0.45 ± 0.00b	38.68 ± 0.10a	35.82 ± 0.18b	11.94 ± 0.12a	86.44 ± 0.10b
RL	89.93 ± 0.32a	20.56 ± 1.81b	0.43 ± 0.00a	38.34 ± 0.14a	38.19 ± 0.12b	23.05 ± 0.07c	99.58 ± 0.02c

Means with different letters in the column for each position and cherry laurel samples are significantly different ($p < 0.05$), Trabzon High (TH), Trabzon Low (TL), Rize High (RH), Rize Low (RL), Dry Weight (DW)

3.2.2. Organic acids and sugar

The contents of malic acid, citric acid and ascorbic acids in cherry laurel fruits grown at two different locations and altitudes are presented in Table 2. The major organic acid was malic acid (89.93-124.72 mg / g DW), followed by citric acid (12.97-27.16 mg / g DW) and ascorbic acid in small concentrations (0.43-0.46 mg / g DW) in cherry laurel fruit collected from Trabzon and Rize provinces. Malic acid content of fruit collected from Trabzon and Rize provinces decreased from 124.72 to 117.01 mg / g DW and from 119.86 to 89.93 mg / g DW, respectively, with the decrease in altitude ($p < 0.05$). The cherry laurel fruit collected from Trabzon at high altitude (TH) had the highest malic acid content (124.72 mg / g DW), whilst the lowest content was detected in that of Rize at low altitude (RL) (89.93 mg / g DW). Conversely, the amount of citric acid increased in Rize samples from 12.97 to 20.56 mg / g DW ($p < 0.05$) with a decrease in altitude. The highest citric acid content was detected in the TL sample (27.16 mg g DW-1), while the lowest citric acid content was detected in RH (12.97 mg / g DW). Amount of ascorbic acid in the sample from both location was decreased from 0.46 to 0.43 mg / g DW and 0.45 to 0.43 mg / g DW for Trabzon and Rize samples, respectively, with the decrease in altitude. In general, the organic acid concentrations are in agreement with the previous values of the cherry laurel fruit collected from Turkey (Celik et al., 2011;

Yıldız et al., 2014; Ozturk et al., 2017). In general, the amounts organic acids were higher in cherry laurel fruit collected from Trabzon province than those of collected from Rize province. The results are in agreement with the previous studies which report the malic acid and ascorbic acid contents decreased with decrease in altitude (Mphahlele et al., 2014).

Sugars have a powerful impact on fruit taste and consumer acceptance. In accordance with the previous studies (Ayaz et al., 1997; Var and Ayaz, 2004), it was found that the cherry laurel fruit contains high amounts of sugar compounds, mainly glucose, fructose and sorbitol, and the results are given in Table 2. The effect of altitude on each sugar and total sugar concentrations was found to be statistically significant ($p < 0.05$). Total sugar concentrations of the cherry laurel fruit collected from low altitudes was higher than those collected at high altitude. In accordance with this finding, the amount of sugar was substantially high at low altitude in strawberry (Crespo et al., 2010), and Polygonum cognatum (Macar and Macar, 2018). Concentrations of glucose, fructose, sucrose and sorbitol in cherry laurel fruits were reported to be in the range of 0.8 - 27.62 g 100 g FW-1, 1.3 - 27.3 g 100 g FW-1, 0-0.6 g 100 g FW-1 and 0.5-14.5 g 100 g FW-1, respectively (Ayaz et al. 1997; Ayaz et al. 1998; Var and Ayaz 2004). After approximate conversion of FW to DW, the amount of sugars for both locations and altitudes are higher than these reported concentrations, while sucrose was not detected in the current study. There is contradictory information about the sucrose content of cherry laurel fruit. The absence of sucrose may be explained by factors such as its decomposition to glucose and fructose due to invertase activity, maturation level genotype (Var and Ayaz, 2004; Crespo et al., 2010). There were statistically significant differences in the total sugar content of the samples, regardless of location and altitude ($p < 0.05$). The fruit collected from Trabzon at low altitudes (TL) has the highest total sugar content (131.99 mg / g DW). In agreement with this finding, higher total sugar content was also reported in golden berry fruit grown at low altitudes (Fisher et al., 2007).

3.2.3. Phenolic compounds

Phenolic compounds determined at various wavelengths in cherry laurel fruit are shown in Table 3. Among the fourteen phenolic compounds, Procyanidin B1, Epigallocatechin, Procyanidin B2, Epicathechin, Neochlorogenic acid, Quercetin 3-β-D-glucoside and Kaempferol-3-glucoside were identified and quantitated in cherry laurel fruit for the first time. Rutin hydrate (300.12-579.80 mg / kg DW) and chlorogenic acid (282.69-944.16 mg / kg DW) were the most abundant phenolic compounds in all cherry laurel samples regardless of their location and altitude. Also, quercetin 3-β-D-glucoside was only detected in fruit collected from Trabzon province. Chlorogenic acid was found as a major phenolic compound in cherry laurel fruit by many researchers (Alasalvar et al., 2005; Karahalil and Şahin, 2011; Ozturk at al., 2017). There is no agreement on the number and amount of phenolic compounds in the previous reports. For example, in a previous study by Ayaz et al. (2004) who reported the amount of ρ-coumaric acid and ferulic acid was reported by in the range of 0.01-7.14 mg/100 mg DW and 0.14-1.0 mg 100 mg /DW, respectively, while Karahalil and Şahin (2011) found that the average amount of these compounds were 2.55 and 0.58 mg / 100 FW. In general, the amount of phenolic compounds in this study were considerably higher than those data reported for the cherry laurel fruit.

The concentrations of phenolic compounds in fruit collected at both altitudes of Trabzon province were significantly higher than those of Rize province ($p < 0.05$). As mentioned before, compared to Rize province, Trabzon province has much exposure time to sunlight. In previous reports, good correlations were observed between the concentrations of phenolic compounds and exposing time to sunlight (Mphahlele et al., 2014). In cherry laurel fruits collected from Rize province, quercetin glycoside, which is a member of flavonol that forms on the fruit peel by the effect of sunlight (Topalovic and Mikulic-Petkovsek, 2010), was not detected in Rize samples due to low exposing time to sunlight. In accordance with the observations for the total phenolic content and β-carotene content, cherry laurel fruit growing at low altitude conditions contains higher amounts of phenolic compounds than those growing at higher altitude conditions ($p < 0.05$). In agreement with current findings, amounts of phenolic compounds increased in strawberry (Crespo et al., 2010), pomegranate fruit (Mphahlele et al., 2014) and elderberry (Senica et al., 2016), when growing altitudes decreased.

Regarding the effect of phenolic compounds on antioxidant activity determined by DPPH and ABTS assays were evaluated by taking into consideration the results from both of the location and altitude. All the phenolic compounds were well correlated with DPPH and ABTS values. Positive correlations were observed between the DPPH scavenging activity and phenolic compounds which ranged from $r= 0.636$ (neochlorogenic acid) to $r= 0.976$ (rutin hydrate). There

were also good correlations between the ABTS results and phenolic compounds which ranged from r= 0.506 (gallic acid) to r= 0.942 (epigallocatechine).

Table 3. Individual phenolic compounds (mg / kg DW) at 280 nm, 320 nm and 360 nm in cherry laurel samples grown in different locations and altitudes
Çizelge 3*. Farklı bölge ve yüksekliklerde yetiştirilen karayemiş örneklerinin 280, 320 ve 360 nm de bireysel fenolik bileşenleri (mg / kg KM)*

Wavelength	Phenolic Compounds (mg / kg DW)	TH	TL	RH	RL
280 nm	Gallic acid	83.51± 0.24b	111.43±0.40d	63.67± 1.63a	99.29± 1.70c
	Procyanidin B1	1.80± 0.33b	2.40± 0.63c	0.07± 0.00a	1.38± 0.25b
	Epigallocatechin	153.29±6.05b	217.49± 2.53c	52.04± 2.75a	51.49± 0.02a
	Catechin	19.70±0.93b	35.21± 1.38c	11.24± 1.27a	17.06± 1.11b
	Procyanidin B2	55.42± 1.02c	56.99± 0.27c	10.43± 1.62a	34.10± 1.28b
	Epicathechin	61.48± 0.22b	123.60± 0.44d	41.23± 0.72a	86.40± 0.14c
320 nm	Neochlorogenic acid	0.21± 0.04a	2.09± 0.09c	0.14± 0.02a	1.64± 0.02b
	Chlorogenic acid	447.67± 1.30b	944.16± 4.01d	282.69± 0.02a	509.70± 3.45c
	Caffeic acid	8.13± 0.21c	12.54± 0.00d	3.99± 0.01a	4.47± 0.23b
	ρ-Coumaric acid	2.76± 0.00b	5.88± 0.07d	0.46± 0.19a	4.94± 0.16c
	Ferulic acid	0.58± 0.01a	16.69± 0.46c	0.10± 0.02a	12.68± 1.05b
360 nm	Rutin hydrate	517.94± 3.52d	579.80± 0.07c	300.12± 2.86a	434.39± 0.82b
	Quercetin 3-β-D-glucoside	2.91± 0.14a	5.13±0.09b	0	0
	Keampherol 3-gliKaempferol-3-glucoside	5.06± 0.00c	5.31± 0.00d	4.86± 0.03a	4.96± 0.07b

Means with different letters in the line for each position and cherry laurel samples are significantly different ($p < 0.05$), Trabzon High (TH), Trabzon Low (TL), Rize High (RH), Rize Low (RL), Dry Weight (DW)

4. Conclusion

The present study exhibited that the location and altitude had a great effect on the compositional and antioxidant properties of cherry laurel fruit. The some bioactive components of cherry laurel fruit (malic acid from 117.01 to 124.72 mg/g DW, ascorbic acid from 0.43 to 0.46 mg/g DW) increased with increasing altitude while some bioactive contents of fruit (Total phenolic content from 23.32 to 21.90 mg GAE/100g DW, antioxidant activity from 24.60 to 23.09 µmol trolox / 100 g DW, β-carotene from 6.75 to 5.19 mg/kg DW) decreased with increasing altitude . The fruit from Trabzon province had superior features in terms of some quality properties (total phenolic content, organic acid, sugar content and all the individual phenolic compounds examined) and antioxidant characteristics. This may be due to differences in exposing time of sunlight and the amount of rainfall. These results are considered to be of practical importance in the nutritive value and bioactive properties of cherry laurel fruit.

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