



Changes in the Quality of Black Mulberry and Blueberry Sherbets During Storage

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ABSTRACT

This study was evaluated the quality properties of traditional drink sherbets that are prepared from black mulberry and blueberry fruits. After production sherbets were investigated to determine their pH, acidity, °brix and colour values, total sugar, phenolic, anthocyanin and antioxidant contents. Moreover the sherbets stored at 4°C during 2 months and the changes in these quality properties were examined per month. As a result statistically significant changes were observed in the quality properties of these sherbets of black mulberry and blueberry fruits which are known with their rich content of phytochemical compounds. The results show that in blueberry sherbet the degradation of phenolics was faster than black mulberry sherbet. Anthocyanins that are higher in black mulberry sherbets after production were preserved better in blueberry sherbets at the end of 2nd month. L* and a* values decreased for blackberry and blueberry sherbets during storage. b* value decreased from 5.59 to 4.92 for blackberry sherbet while it increased from 0.62 to 0.79 for blueberry sherbet at the end of the storage time.

Introduction

The black mulberry (*Morus nigra*), which originates from Iran, is cultivated for its fruits in South Europe, Southwest Asia and is the most important species in the Mediterranean countries (Tutin, 1996). The black mulberry (Turkish name 'Kara Dut') is widely grown in Turkey (Yaltirik, 1982). Along with Mediterranean conditions, Northeast part of Turkey, in particular Coruh valley has notable populations of black mulberry, which have been cultivated in gardens for their delicious edible fruits (Ercisli and Orhan, 2007).

Blueberry fruits belong to the genus *Vaccinium*, fam (Grace and et al., 2009). The blueberry (*Vaccinium* spp.) is a fruit native to North America and Europe, where it is widely cultivated and commercialized (Prior et al., 1998).

Berries are rich in phenolic compounds, such as phenolic acids, tannins, stilbenes, flavonoids and anthocyanins, but berries, in particular, have been the focus of considerable research regarding their anthocyanin-rich properties and according to Seeram (2008), there are many studies claim that the dietary intake of berry fruits has a positive and profound impact on human health, performance, and disease (Rios de Souza and et al., 2014). Blueberry and mulberry juice

prevent obesity with the rich content of anthocyanins and phenolics (Wu et al., 2013). In addition in previous studies, antimicrobial activity of these fruits and juices on some microorganisms were investigated (Yang et al., 2014).

The red-coloured fruits which taste very pleasant when eaten fresh also have other uses as marmalades, juices, liquors, natural dyes and in the cosmetics industry (Ercisli and Orhan, 2007).

Sherbet is sweet and served chilled. Popular sharbats are made of one or more of the following: rose water, sandalwood, bael, gurhal (*Hibiscus*), lemon, orange, mango, pineapple, and falsa (*Grewia asiatica*). Most of the sharbats are very common in Indian, Turkish, Iranian, Arab, Afghan, Pakistani, and Bangladeshi homes. Black mulberry and Blueberry Sherbets are the traditional foods of İzmir (Turkey).

There are several studies about these fruits but the traditional drink sherbet has not been investigated before. The aim of this study was to evaluate the changes in the quality of black mulberry and blueberry sherbets during storage.

Materials and Methods

Preparation of Black mulberry and Blueberry Sherbets

Black mulberry and blueberry sherbets were produced at Yudumla Sherbet Food Marketing and Industry. The fruits were processed after the pretreatment such as washing and sorting. After addition of sugar mashing was made, citric acid was added then pasteurization was made at 85°C, for 10 min. Then the sherbets were hot filled and cooled before storage. The flow charts were shown in figure 1.

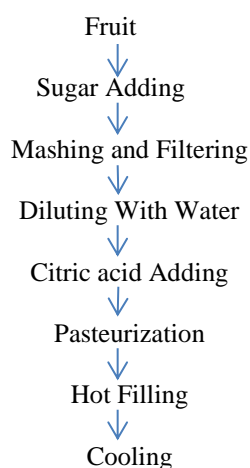


Figure 1 Process flow chart of sherbets

Methods of Analyses

Quality characteristics were investigated by physical and chemical analyses. Samples were analysed to determine the following: pH values were determined with pH meter (WTW InoLab, Weilheim, Germany).

Total acidity was determined by means of a potentiometric titration of the acidity of the juice by placing 10 mL of juice into 90 mL deionized water after filtration 10 mL filtrate was taken to titration, with a solution of 0.1 N NaOH up to pH 8.1. The results were expressed as g / 100 mL with reference to citric acid (Anon, 1995).

The total soluble matter (°Brix) of juices was measured with a refractometre at 20°C (RFM 330 Bellingham + Stanley Limited, Tunbridge Wells, Kent, UK) (Anon, 1995).

Total sugar was determined according to the Luff-Schoorl method (Lees, 1975). No reducing sugar (sucrose) content was calculated according to the relationship $\text{sucrose} = [\text{total sugar} - \text{reducing sugar}] \times 0.95$.

Total phenolic content was measured using the Folin-Ciocalteu method (Singleton and Rossi, 1965). Results were expressed as milligrams of gallic acid equivalents per gram of fresh weight.

Total anthocyanin content was determined with the pH differential method (Wrolstad, 1976) and expressed as milligrams of cyanidin-3-glucoside equivalents per gram of fresh weight, on the basis of the molar absorptivity (29.600) of cyn-3-glu for blackberry. Calculations were based on malvidin-3-glucoside with molar absorptivity 28.000 for blueberry. All spectrophotometric analyses

were performed by using Varian Cary 50 Scan (Australia) model spectrophotometer.

Total antioxidant activity of juices was determined by the ABTS (2,2-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt) assay (Re et al., 1999). The method gives a measure of the antioxidant activity of the juice, determined by the decolorization of the ABTS (Sigma-Aldrich Corp.) performed by a reaction with potassium persulfate, and monitoring the reduction of the radical cation as the percentage inhibition of absorbance at 734 nm during 6 min. The Trolox equivalent antioxidant capacity (TEAC) is the concentration of antioxidant giving the same percentage inhibition of absorbance of the radical cation at 734 nm as 1-mM Trolox (Fluka Chemical, Switzerland) solution. The TEAC was obtained by the ratio between the gradient of the plot of the absorbance inhibition percentage compared with the concentration for the analyzed antioxidant and the corresponding gradient of the plot for Trolox.

The colour (L*, a*, b*) values were measured at HunterLab Colourflex colourimetre (Hunter Associates Laboratory Inc., Reston, VA, USA).

Statistical analyses were performed by analysis of variance (ANOVA) using the software SPSS 16 (SPSS Inc., Chicago, IL, USA) with Duncan test to evaluate the differences between months at level of significance ($P \leq 0.05$). Each experiment was repeated at least three times; means and standard deviations of results were calculated.

Results and Discussion

Physical and chemical properties of black mulberry sherbet were investigated during 2 months of storage at refrigerator conditions. Results of physical and chemical analyses were shown in table 1 for the samples of black mulberry sherbet.

pH value and acidity did not change significantly during storage ($P > 0.05$). There are limited studies about black mulberry beverage (sherbet or drink) but previous studies were focused on the properties of black mulberry fruit. For example Erciřli and Orhan (2008) who investigated the physico-chemical characteristics of black mulberry cultivars found that the pH values of black mulberries vary between 3.43-3.66 which are comparable with our study. Gündođdu et al. (2011) determined the acidity of mulberry species as 1.084±0.003% as citric acid for black mulberry fruit. The pH value was found as 3.30 and the acidity as 1.42% in the research of Kara and Ercelebi (2013) who studied with the Urmu mulberry juice concentrate.

°Brix value didn't change during storage. The change in the total sugar content was statistically not significant ($P > 0.05$). Aramwit et al. (2010) found the total sugar content as 7.60±1.80 g/100 g for purple, 5.1 ± 1.4 for purple-red and 1.8 ± 0.8 for red coloured mulberries. Gundogdu et al. (2011) reported that the total sugar content was changed between 10.4±0.1-13.2±0.1 (g/100 g) and they stated that glucose (52%) and fructose (48%) composed main part of the sugar composition whereas sucrose is only the 1% part of the total sugar in *Morus nigra* black mulberry fruit.

Table 1 Changes in the quality of black mulberry sherbet during storage

Analyses	0.day	30 th day	60 th day
pH	3.75±0.01 ^a	3.75±0.02 ^b	3.75±0.01 ^c
Acidity (%)	0.47±0.10 ^a	0.48±0.01 ^b	0.48±0.01 ^c
Total sugar (%)	18.17±0.04 ^a	18.15±0.07 ^b	18.13±0.04 ^c
°Brix	18.85 ±0.07 ^a	18.80 ±0.14 ^b	18.70 ±0.14 ^c
Phenolic content (ppm)	383.98±0.25 ^a	299.98±0.40 ^b	199.90±0.20 ^c
Anthocyanin content (mg cy-3-glu/g)	300 ±0.10 ^a	250 ±0.12 ^b	150±0.15 ^c
Antioxidant capacity (TEAC mM trolox/ml)	12.5 ±0.20 ^a	11.4 ±0.10 ^b	9.2 ±0.30 ^c
L*	6.96±0.07 ^a	6.75±0.01 ^b	6.24±0.01 ^c
a*	19.49±0.19 ^a	18.07±0.54 ^b	17.86±0.60 ^b
b*	5.59±0.29 ^a	5.36±0.35 ^b	4.92±0.30 ^c

^{a to c} Different lower case letter in the same line for each month indicates significant differences amongst samples during storage (P<0.05).

Phenolics which are important with the antioxidant, antimutagenic and anticarcinogenic properties, are found in high amounts in deep-colored fruits. The total phenolic content for different cultivars of black mulberries was ranged between 1943 and 2237 mg gallic acid equivalents/100g fresh mass (Ercişli and Orhan, 2007). Kara and Ercelebi (2013) studied with the Urmu mulberry juice concentrate and found the total phenolics as 2650 (mg GAE/L). We found this content as 383.98±0.25 ppm in the sherbet. Phenolic compounds showed a degradable character during storage. The difference between the values were statistically significant (P<0.05). In a different study the blackberry-blueberry fermented beverage was studied. They determined the total phenolics of blackberry wine as 4029.0±68.4 (mg EAE²)/L. Altuner and Tokusoglu (2013) demonstrated that phenolic compound composition in fruits is affected by some intrinsic factors, such as using different genus, species or cultivars, and extrinsic factors, such as the time of the collection of fruits, location, environmental factors and storage. Besides these intrinsic and extrinsic factors, some food-processing technologies can also affect the plant phenolics' composition. They treated the juice with high hydrostatic pressure and found that this method preserves the compounds well after processing.

Anthocyanins (mg cy-3-glu/g fw) having a role in the colour of berry fruits, and this phytochemicals are also known with positive health effects by their antioxidant properties (Johnson et al., 2013). Anthocyanins were decreased by half at the end of 2nd month compared to the initial value of black mulberry sherbet. Similar to phenolics, anthocyanins are highly perishable compounds. Fazaeli et al. (2013) studied with black mulberry juice concentrate at 42° Brix which is treated with conventional and microwave heating, results showed that anthocyanin degradation and consequently decrease in antioxidant activity were more pronounced in rotary evaporation compared to microwave heating method. Nikkha et al. (2007) emphasized that cyanidin-3-glucoside which is common anthocyanin in blackberry has also been reported to have the highest antioxidant capacity of 14 different anthocyanin tested (Wang et al., 1997; Mazza and Miniati, 1993). They also indicated that lowering water activity with sugar and especially sucrose concentration of 20% has a protective effect on anthocyanins, but higher concentrations has negative effect. If the sugar content of

the sherbet was taken into consideration the degradation of total anthocyanins could be much more slower rather than to be a half. But on the other hand, Hellstrom et al. (2013) who studied the stability of anthocyanins in blackcurrent, chokeberry, bilberry and crowberry juices stored at different temperatures, decided that the anthocyanin stability was effected by a number of factors including the type of anthocyanin, the origin of the juice, and especially the storage temperature. They also indicated that in the berry smoothie the total anthocyanin content was 40.3±1.2 mg/100 ml quantified as cyanidin 3-glucoside equivalents. They explained that the commercial berry juice drinks had much lower anthocyanin contents than the other berry juice samples. The reason for this situation is their rather low juice content but probably also because of the intense processes applied commonly in the food industry. Anthocyanin degradation was faster in the commercial juice drinks stored at room temperature than in the smoothie stored at 4°C. Indeed, they explain the anthocyanin decrease with the changes in pH. In the commercial berry juice drinks the initial pH value (approximately 2.9) was lower and therefore, more favourable for anthocyanins than in the laboratory-made juices (pH 3.3–3.5). Similar to their study, we can say that the black mulberry sherbet has a pH value of 3.75 so the degradation was faster.

Antioxidant capacity due to phenolic and anthocyanin content decreased from 12.5 ±0.2 to 9.2 ±0.30 TEAC mM trolox/ml sherbet. Gündoğdu et al., (2011) found the total antioxidant capacity of black mulberry as 13.999 ± 0.008 TEAC µmol TEg⁻¹ fw. In another reaserch the antioxidant contents were changed between 6.8-14.4 TEAC (mmol TE/g fw) for black mulberry fruits (Ozgen et al., 2009).

Color of berry type fruits were provided by the anthocyanin compounds. These compounds can be intensive on the skin or leaves of the fruits (Riihinen et al., 2008). It was stated that fruit color values were changes in the ranges for L*, a* and b* value as 14.79-17.47, 9.18-17.30 and 2.43-4.86 respectively (Ercişli and Orhan, 2008). Aramwit et al. (2010) measured the red colored mulberries and reported the lightness, redness and blueness to be as 27.75 ± 5.32, 20.62 ±3.56, 9.28 ±2.87 respectively. The L* value was lower in the sherbet. Redness was found as 19.49±0.19 and decreased during storage. But this decrease is not statistically significant

($P>0.05$). The b^* value was found in the range of 5.59-4.92. The decreases can be explained by the degradation of colour pigments. Anthocyanins especially Cyanidin ($R_1 = OH, R_2 = H$) which is the dominant one in the black berries (Hellstrom et al., 2013; Bae and Suh, 2007) was instable. Due to losses in anthocyanins the colour was affected negatively.

Chemical and physical characteristics of blueberry sherbets were investigated and shown in the table 2. In blueberry sherbet samples pH value was lower compared to black mulberry sherbet. Giovanelli & Buratti (2009) who studied with blueberry indicated that pH value was in the range of 2.92-3.22. Giovanelli et al. (2013) found the pH value of blueberry as 3.48 ± 0.02 . Acidity and pH value didn't change during the storage time.

Table 2 Changes in the quality of blueberry sherbet during storage

Analyses	0. day	30 th day	60 th day
pH	2.29±0.06 ^a	2.30±0.06 ^b	2.31±0.06 ^c
Acidity (%)	0.41±0.01 ^a	0.41±0.01 ^b	0.41±0.01 ^c
°Brix	18.95±0.07 ^a	18.90±0.07 ^b	18.88±0.10 ^c
Total sugar (%)	18.74±0.06 ^a	18.65±0.07 ^b	18.68±0.11 ^c
Phenolic content (ppm)	536.79±0.15 ^a	435.34±0.10 ^b	100±0.12 ^c
Anthocyanin content (mg cy-3-glu/g)	200±0.20 ^a	165±0.15 ^b	160±0.17 ^c
Antioxidant capacity (TEAC mM trolox/ml)	17.93±0.02 ^a	10±0.04 ^b	7.86±0.01 ^c
L*	4.20±0.09 ^a	4.68±0.01 ^b	3.95±0.28 ^c
a*	11.66±0.01 ^a	11.46±0.02 ^b	10.49±0.03 ^c
b*	0.62±0.10 ^a	1.12±0.01 ^b	0.79±0.20 ^c

^{a to c} Different lower case letter in the same line for each month indicates significant differences amongst samples during storage ($P<0.05$).

°Brix value was decreased to 18.68% at the end of two months. It was indicated that blueberry has a °Brix value in the range of 11.6-12.9. Giovanelli et al. (2013) found the °Brix value as 11.4 ± 0.4 . Soluble solid content was determined as 14.9 ± 0.6 by Gunathilake et al., (2014) in the blueberry juice. Sugar content was significantly decreased during 2 months of storage similar to black mulberry sherbet samples.

The losses in the phenolics increased after 1st month, approximately 77% losses were determined after 1st month. These compounds were susceptible to degradation during processing and storage (Syamaladevi et al., 2012). Giovanelli and Buratti (2009) found the total phenolics 251-298 (gallic acid mg/100 g) in different cultivars of blueberry. The phenolic content of blueberry was indicated as 317 ± 18 mg/100g (2013). Total polyphenols of partially concentrated to 15 °Brix of blueberry juice has phenolic content of 2627 ± 25 mg GAE/L mg gallic acid equivalent/L. Syamaladevi et al. (2012) found a 117% increase in the phenolic content of in canned blueberry syrup during 13 month of storage. They explain this increase to the improved extraction efficiency due to cell destruction during storage.

Anthocyanin content was lower compared to black mulberry sherbet. It was explained that there is relationship between the sugar content and anthocyanins due to the role of sugars in the anabolism of anthocyanins by Aramwit et al., (2010). It is possible to verify this opinion with the data that black mulberry sherbet has higher total sugar than blue berry sherbet samples initially. But during storage lower reduction of these compounds were obtained in the samples of blueberry sherbet. Giovanelli et al. (2013) indicated that total anthocyanin content for the blueberry was 97 ± 10 mg/100 g. Syamaladevi et al. (2012) found 73–79% decreases in

anthocyanins in blueberry juices after 5 months of storage in comparison with 0 month of storage. Mohideen et al. (2015) investigated the effect of continuous ultrasonication on microbial counts and physico-chemical properties of blueberry (*Vaccinium corymbosum*) juice and showed that ultrasonic treatment did not change the total anthocyanin content of blueberry juice. It was previously highlighted that deterioration of anthocyanins can be effected by the enzymes such as polyphenoloxidase, peroxidase and β -glucosidase (Cano et al. 1997; Altuner and Tokuşoğlu, 2013).

The antioxidant activity decreased during storage and the difference between the antioxidant activities of samples were statistically significant ($P<0.05$). In a similar study; Syamaladevi et al. (2012) found in the canned blueberry syrup, total anthocyanins and total antioxidant activity decreased by up to 68 and 15% respectively. They observed that the antioxidant activity in canned blueberry syrup showed greater consistency than total anthocyanins and total phenolics during storage. They also researched the effect of blanching on the blueberries during the storage of juice, total antioxidant activity changed from 1st month to 3rd month as 9.49. The berries processed to juice without blanching have lower antioxidant values. The antioxidant activity was reduced from 11.14 ± 0.18 by 39.85% in blueberry juice stored under refrigerator for 10 days. In a different study researches investigated the blueberry by products and decided that acidification significantly reduced the total anthocyanin content, total polyphenols and antioxidant activities. In addition they demonstrated that by-products especially juice pomace and wine pomace still retained important phenolic concentrations and antioxidant activities (Su and Silva, 2006).

Color values were shown in table 2 for the blueberry sherbet samples. L* and b* values were increased in the 1st month and then decreased in the 2nd month. a* value was more stable than the L* and b* values. Mohideen et al. (2015) found the colour values of blueberry juice comparable to our results as L*: 3.03 ± 0.03 and a* value: 15.49 ± 0.39. It is suggested that a decreases in a* values of berry juices are related to anthocyanin degradation and formation of Maillard reaction products (Aguilo-Aguayo et al., 2009; Mohideen et al., 2015).

Conclusion

In summary this study demonstrated that sherbets produced from black mulberry and blueberry fruits are rich in phenolic and anthocyanin compounds which have high antioxidant properties comparable with the raw fruits. These fruits and fruit products have health benefits associated with these nutritious phytochemicals. However these valuable compounds are favourable to deterioration reactions. At the end of storage at refrigerator conditions (+4°C) the quality parameters were decreased significantly. It can be said that although blueberry drink has higher phenolic and antioxidant activity, the losses in these compounds are faster and higher than the black mulberry sherbet. Novel technologies such as ohmic heating, high hydrostatic pressure can be used for blocking phenolic and anthocyanin compounds loses that is caused from temperature.

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