



Influences of chitosan coatings on functional compounds of sweet cherries

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Abstract In this study, sweet cherries were coated with four chitosans (1%) [two of them produced from shrimp waste from Marmara Sea in Turkey (Chitosan-1, Chitosan-2) and two of them commercially produced (Commercial-1 and Commercial-2)] which have different deacetylation degree, and molecular weight (Chitosan-1, deacetylation degree: 78.20%, molecular weight: 182 kDa; Chitosan-2, deacetylation degree: 84.95%, molecular weight: 127 kDa; Commercial-1, deacetylation degree: 81.22%, molecular weight: 273 kDa; Commercial-2, deacetylation degree: 75.12%, molecular weight: 407 kDa) and stored at 4 °C for 25 days, and 20 °C for 15 days. Changes in the total phenolic content, antioxidant capacity, total anthocyanin content, ascorbic acid, total pectin content, firmness, and colour values were evaluated. The results revealed that Chitosan-1 had the highest firmness value; Chitosan-2 showed the highest total anthocyanin and total phenolic content and Commercial-1 exhibited the highest antioxidant capacity and ascorbic acid content at 4 °C. Furthermore, it was found that Chitosan-1 demonstrated the highest total phenolic content; Chitosan-2 displayed the highest total anthocyanin; Commercial-1 had the highest firmness value and C-2 exhibited the highest ascorbic acid

content at 20 °C. In conclusion, each tested chitosan coatings have different effects on different quality attributes at different storage temperatures.

Keywords Sweet cherry · Chitosan · Deacetylation degree · Molecular weight · Antioxidant activity

Introduction

Nowadays, there is an increasing interest in polyphenol-rich fruits because of their antioxidant properties. Sweet cherries are also a rich source of many nutrients and phytochemicals. Its attractive colour, sweetness, firmness, having antioxidants are the main characteristics for sweet cherry quality. The antioxidant properties of sweet cherry are associated with the ascorbic acid and polyphenolic content. Sweet cherries have various phenolic contents including hydroxycinnamic acids, hydroxybenzoic acids, flavonols, and anthocyanins (Ferretti et al. 2010; Kelebek and Selli 2011; Prvulović et al. 2011; Wani et al. 2014; Chiabrando and Giacalone 2015). The major anthocyanins identified in sweet cherry were the cyanidin-3-O-glucoside and cyanidin-3-O-rutinoside (Mulabagal et al. 2009; Kelebek and Selli 2011).

Sweet cherry (*Prunus avium* L.) is an important fruit in Turkey with a high commercial value. Studies for preserving sweet cherries quality until reach the consumer are gaining great importance. Proper packaging and storage are necessary for maintaining the quality of sweet cherries. One of these applications is coating sweet cherries with an edible coating. Edible coatings can act as barrier and inhibit gas exchanges, control mass transfers, improve mechanical properties of food, use as carriers for food additives or ingredients, improve sensorial characteristics

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such as colour and shininess (Debeaufort et al. 1998; Baldwin et al. 2009; Dutta et al. 2009).

Chitosan, consisting of (1,4)-linked 2-amino-deoxy- β -D-glucan, is a natural carbohydrate polymer having antimicrobial and antioxidant properties besides superior film forming properties. There are several mechanisms that explain the antioxidant activity of chitosan: (1) having high metal binding capacity due to its free amino groups. It is related to the fact that the amino and hydroxyl group in chitosan can react with unstable free radicals to form more stable and relatively non-toxic macromolecules (2) The chitosan chelate the iron ions in the system to retard lipid oxidation, thereby eliminating their prooxidant activity or their conversion to ferric ion (Kamil et al. 2002; Kim and Thomas 2007; Vinšová and Vavříková 2011). Besides all these properties, chitosan has been often used as an edible coating because of nontoxic, biodegradable, biofunctional and biocompatible polymer (Dutta et al. 2009).

The use of chitosan as edible coating has been examined in many studies and successfully used for extending shelf life of sweet cherry and many fruit and vegetables. Zam (2019) studied the influence of chitosan 1% and alginate 3% enriched with olive leaves extract on the quality of sweet cherries. In this study, it was verified to retard the ripening process of sweet cherries with a maximum retention of phenolic compounds compared with uncoated fruit samples. Nair et al. (2018) who coated guavas (*Psidium guajava* L.) with chitosan (1% w/v) and alginate (2% w/v) in combination with pomegranate peel extract (1% w/v) indicated that coatings enhance the quality of the guava fruit during 20 days of low temperature storage. Reyes-Avalos et al. (2019) researched on the effect of the treatment of an alginate–chitosan (A–Ch) coating on the bioactive compounds and the antioxidant capacity of fresh figs (*Ficus carica*). They found that the application of A–Ch coating can be an effective alternative to preserve the bioactive compounds and the organoleptic characteristics assuring the shelf life of the fresh figs during storage at low temperature.

The deacetylation degree and molecular weight of chitosan influences its physicochemical and biological properties (Cho et al. 1998; Matsugo et al. 1998; Tolaimate et al. 2000; Raafat and Sahl 2009; Rajalakshmi et al. 2013; Wan et al. 2013; Hossain and Iqbal 2014; Trung and Bao 2015). Some researchers have suggested that antioxidant activity of chitosan increased with increasing the degree of deacetylation (Park et al. 2004; Yen et al. 2007; Samar et al. 2013). Park et al. (2004) investigated free radical scavenging activity of chitosan which have different deacetylation degree (90%, 75% and 50%) on DPPH radical, alkyl radical, hydroxyl radical, and superoxide radical using electron spin resonance spectrometer. Chitosan with 90% degree of deacetylation showed the highest radical

scavenging effects on the hydroxyl radical and superoxide radical. It was concluded that the free amino groups in chitosan can react with free radicals to form stable macromolecule radicals. Furthermore, Kim and Thomas (2007) stated that chitosan with lower molecular weight (30 kDa) showed the highest scavenging activity compared to higher molecular weight chitosan (90–120 kDa).

The aim of this study were to investigate and compare the effects of different type chitosan edible coatings on the phytochemical and some quality attributes of sweet cherries during storage periods at 4 °C and 20 °C.

Materials and methods

Materials

Sweet cherries (*Prunus avium* L., 0900-Ziraat) used in this study were harvested from Tokat province of Turkey in July of 2015 and were stored at 4 °C until coating process. Sweet cherries were washed with tap water, dried at ambient temperature and physical damaged ones were eliminated before treatments.

Chitosan-1 (CH-1) and Chitosan-2 (CH-2) were produced from shrimp wastes sourced from Marmara Sea. Chitin and chitosan production were carried out by chemical extraction. Therefore, chitin production was obtained using 0.73 mol/L hydrochloric acid for 132.61 min at room temperature for demineralization, and 0.95 mol/L sodium hydroxide for 75.65 min at 60.49 °C for deproteinization. Then, CH-1 was achieved by deacetylation with 40% sodium hydroxide at 120 °C for 300 min, while CH-2 was carried out by deacetylation with 50% sodium hydroxide at 100 °C for 720 min (Tokatlı and Demirdöven 2018). CH-1 has a degree of deacetylation of 78.2%, and a molecular weight of 182 kDa. CH-2 has a degree of deacetylation of 84.95%, and a molecular weight of 127 kDa. Commercial-1 (C-1) (deacetylation degree: 81.22%, molecular weight: 273 kDa) and Commercial-2 (C2) (deacetylation degree: 75.12%, molecular weight: 407 kDa) were supplied from Sigma-Aldrich Chemical Company, USA.

Details on the determination of chitosan's degree of deacetylation, and molecular weight are given in the Tokatlı and Demirdöven (2018).

Preparing coating solution

The chitosan coating solutions were prepared by mixing 1% chitosan in 1% acetic acid solution at 40 °C for 2 h, filtering through coarse filter paper, adding 1.5 mL glycerol for each 1 g chitosan as plasticizer agent and stirring that solution for 15 min (Tokatlı and Demirdöven 2020).

Coating of sweet cherries

The cherries were divided into five groups: Control (uncoated), CH-1 (coated with Chitosan-1), CH-2 (coated with Chitosan-2), C-1 (coated with Commercial-1) and C-2 (coated with Commercial-2). Sweet cherries washed in distilled water and dried at ambient temperature were dipped in the coating solutions (CH-1, CH-2, C-1 and C-2) in two different steps. The first dipping was followed by drying step for 15 min and dried for 1 h under ambient temperature, and the second one for 15 min to create a uniform film on the fruit surface. Then cherries were dried for 3 h at ambient temperature, weighed, and placed in polyethylene boxes. The storage conditions in the study were determined based on the storage conditions after the consumer obtained the sweet cherries. So that, after coating, all of the samples (the coated and control samples) were stored at 4 °C for 25 days and 20 °C for 15 days. All analyses were performed in a five-day period.

Total phenolic content

The total phenolic content (mg/kg) was measured by Folin–Ciocalteu procedure, using gallic acid as the standard (Franke et al. 2004).

Antioxidant capacity

The antioxidant capacity of samples was determined by the ABTS (2,2-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) assay according to Re et al. (1999) with some modifications. ABTS radical cation was produced by mixing 2.5 mL of 7 mmol L⁻¹ ABTS stock solution and 44 µL of 140 mmol L⁻¹ K₂S₂O₈, both diluted with ultrapure water. This mixture was stored 12–16 h in darkness and then diluted with ethanol to an absorbance of 0.70 ± 0.02 at 734 nm. After addition of 300 µL of Trolox or the diluted sample to 3 mL of diluted ABTS solution, absorbance readings were taken after 6 min of the initial mixing. Trolox (± -6-Hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid) was used for the standard and the results were expressed in µM Trolox equivalents (TE)/kg.

Total anthocyanin content

Total anthocyanin content of samples was determined by pH differential method using two buffer systems: pH 1.0 (0.025 M potassium chloride) buffer and pH 4.5 buffer (0.4 M sodium acetate) according to Fuleki and Francis (1968). Absorbance was measured at 535 nm. Total anthocyanin content of samples was calculated using the following formula and was expressed as mg cyanidin-3-rutinoside/kg sweet cherry.

$$\text{Anthocyanin, (mg/kg)} = \frac{A}{\epsilon \cdot L} (\text{MW}) (\text{DF}) 1000$$

A: Absorbance difference at pH 1.0 and pH 4.5

ε: Molar absorptivity, 28,800

L: Layer thickness of the absorbance measurement cuvette

MW: Molecular weight, 595.2

DF: Dilution factor

Ascorbic acid

Ascorbic acid was determined by 2,6-dichloroindophenol titrimetric method according to Hışıl (2004). L(+) ascorbic acid was used as the standard and the results were expressed in mg/100 g sweet cherry.

Total pectin content

Total pectin content was determined according to Anonymous (1968). Total pectin content was calculated with the calibration curve which was performed by using galacturonic acid anhydrate as standard.

Firmness

Firmness of samples was carried out using a testing machine (Digital Force Gauge, SH-50). The maximum force required for 9 mm drilling of the sample with the needle pointed head was determined as newton (N).

Colour

The colour was evaluated in four different points of the randomly selected sweet cherries external surface. The colour of the samples was measured using a Minolta CR-300 (Japan) colorimeter to determine the L* (darkness, lightness), a* (redness, greenness) and b* (blueness, yellowness) values. To estimate the ΔE (total colour differences) of the samples, the following equation was used

$$\Delta E = \left[(L^*)^2 + (a^*)^2 + (b^*)^2 \right]^{1/2}$$

Statistical analyses

All experiments were carried out with two replicates and two parallels. The significant difference between the means was established by ANOVA variance analysis and Duncan Tests. The results were performed with the SPSS statistical package program (SPSS 17.0 for Windows Evaluation Version (17.0.3); SPSS Inc., Chicago, USA).

Results and discussion

Total phenolic content

The total phenolic content of sweet cherries varied in all storage conditions. At the end of storage at 4 °C, the lowest total phenolic content was determined in the control group (451.00 mg/kg) while the highest total phenolic content was observed at CH-2 coated sweet cherries (554.88 mg/kg). The difference between the total phenolic content of the sweet cherries coated with CH-2 and control group was significant ($P < 0.05$). However, contrary to the storage at 4 °C, the lowest total phenolic content was detected in CH-2 coated sweet cherries (467.11 mg / kg) at the end of storage at 20 °C. Decrease in total phenolic content can be attributed to the disintegration of the cell structure due to the ripening of the sweet cherries and the reason for the increase in total phenolic content is that chitosan causes the production of phenolic substances by increasing the phenylalanine ammonia lyase enzyme activity (Ghasemnezhad et al. 2013). Han et al. (2014) who coated sponge gourd with 0.5% and 1% chitosan with 90% deacetylation and stored for 12 days at 4 °C, indicated that total phenolic content varied during storage and the chitosan coated samples had high total phenolic content than control sample at the end of storage. Petriccione et al. (2015) coated cherries with chitosan (0.5%) which has degree of deacetylation of 90% and a molecular weight of 360 kDa and stated that coated fruits exhibited a lower decrease in total phenolics content values than did uncoated during cold storage.

Antioxidant capacity

Some researchers have suggested a correlation between the total phenolic content and antioxidant capacity, but no such correlation was found in this study. This result can be explained by the measured antioxidant capacity was not solely from the phenolic content or the exact amount of phenolic substances may not be determined by the Folin-Ciocalteu method (Çetin 2012). At the end of storage at 4 °C, the lowest antioxidant capacity was determined in the control group as 1.4993 $\mu\text{mol Tr/kg}$. In addition, the difference between the antioxidant capacity of the control and chitosan coated sweet cherries was significant ($P < 0.05$). This is due to the fact that the chitosan has high metal binding capacity due to its free amino groups and therefore has antioxidant activity (Vinšová and Vavříková 2011). The highest antioxidant capacity was determined in the C-1 and CH-2 coated sweet cherries at the end of storage at 4 °C as shown in Fig. 1, respectively. The difference between C-1 (81.22% deacetylated chitosan) and CH-2

(84.95% deacetylated chitosan) coated sweet cherries was statistically insignificant ($P > 0.05$). This result confirms that antioxidant activity of chitosan increases with increasing the degree of deacetylation. The antioxidant capacity of all samples increased throughout storage and the changes in antioxidant capacity of each sample groups throughout storage at 20 °C was found to be statistically significant ($P < 0.05$). At the end of storage at 20 °C, the highest antioxidant capacity was determined in the control group as 2.0725 $\mu\text{mol Tr/kg}$, but the difference between control, CH-1, CH-2 and C-1 coated sweet cherries was statistically insignificant ($P > 0.05$). On the contrary, some researchers observed a decrease in the antioxidant activity throughout storage with higher antioxidant capacity values in chitosan coated ones compared with uncoated fruits (Ghasemnezhad et al. 2013; Shiri et al. 2013). Petriccione et al. (2015) also reported that chitosan with 90% deacetylation and a molecular weight of 360 kDa delayed the changes in antioxidant capacity of sweet cherries.

Total anthocyanin content

The total anthocyanin content was expressed in cyanidin 3-rutinoside which predominant anthocyanin in the sweet cherries (Mozetič and Trebše 2004; Pappas et al. 2011). Differences between total anthocyanin content of the samples were determined at the beginning of storage. This is due to the difference in the maturity levels of the sweet cherries. At the end of storage at 4 °C, the total anthocyanin content showed an increase of 216.29% in the control group while 318.86% in CH-1 coated sweet cherries, 257.15% in CH-2 coated sweet cherries, 240.76% in C-1 coated sweet cherries and 199.61% in C-2 coated sweet cherries. The highest increase was observed in the control group (516.12%) after storage for 15 days at 20 °C and followed by CH-2 coated sweet cherries (369.38%), CH-1 coated sweet cherries (351.81%), C-1 coated sweet cherries (346.46%) and C-2 coated sweet cherries (332.44%). Although literature studies have shown that the stability of anthocyanin is reduced with increasing storage temperatures (Kalt et al. 1999; Ferretti et al. 2010), the total anthocyanin content of sweet cherries stored at 20 °C were found to be higher in each storage period than sweet cherries stored at 4 °C as seen in Table 1. It is observed that these increases in the total anthocyanin contents that occur during the storage period are due to the increase in maturity. The reason for this is that, the breakage of the cell wall depending on the maturation and then contributing to the release of anthocyanins in the cell by breaking down pectic substances (Mikkelsen and Poll 2002; Gonçalves et al. 2004; Díaz-Mula et al. 2012; Nabifarkhani et al. 2015). Similar results were reported by Nabifarkhani et al. (2015) working with sweet cherry treating with nano-

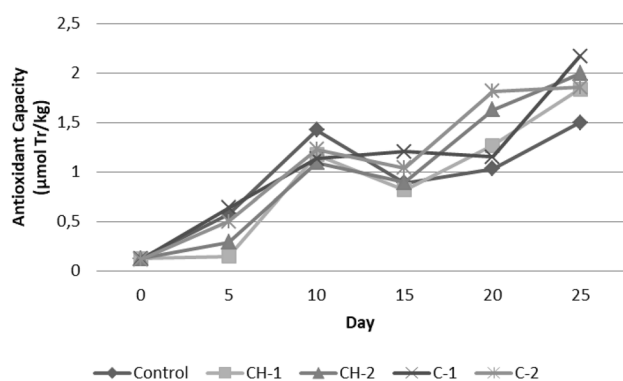


Fig. 1 Effect of chitosan coatings on antioxidant capacity of sweet cherries stored at 4 °C

composite (1% chitosan + 0.1% nano- cellulose) and thyme oil that anthocyanins increased during storage period. On the other hand, Çetin (2012), Ghasemnezhad et al. (2013) and Petriccione et al. (2015) reported a decrease in total anthocyanin content during the storage.

Ascorbic acid

Ascorbic acid degradation was accelerated with the increase of storage temperature and the ascorbic acid content of sweet cherries at 20 °C on the 5th day were lower than those stored at 4 °C. The ascorbic acid content of control group was fallen to zero on day 5 while the ascorbic acid content of chitosan coated sweet cherries was maintained until the 10th day at 20 °C. For all the samples and conditions, the ascorbic acid content of chitosan coated sweet cherries was higher than control group as seen in Table 1. Chitosan coatings may inhibit the activity of some enzymes that play a role in ascorbic acid oxidation due to their low O₂ permeability and as a consequence they can inhibit ascorbic acid oxidation (Dang et al. 2010; Kerch et al. 2011). The ascorbic acid is selected as the indicator vitamin in fruits which have a low amount of ascorbic acid and gives important information about shelf life. Similarly, Aday and Caner (2010), Dang et al. (2010) and Petriccione et al. (2015) also reported that chitosan coatings delayed the decrease in ascorbic acid content of sweet cherries. Furthermore, Chien et al. (2007), treated sliced mango with aqueous solutions of 0%, 0.5%, 1% or 2% chitosan with degree of deacetylation of 95–98, reported an increased in ascorbic acid content in chitosan coating ones at the end of a seven-day holding period.

Total pectin content

There is an increase in the amount of pectin during ripening and storage of fruits and vegetables. This may be caused by: 1—the pectin is degraded by natural pectinases and the

degree of esterification is reduced, 2—the protopectin is broken down by protopectinase and transformed into soluble pectin 3—microorganisms break down pectin by pectin lyase enzyme. However in the later stages of enzymatic degradation, methoxyl groups in the pectin chain are converted to galacturonic acid and the amount of pectin may decrease (Cemeroğlu et al. 2001; Aday, 2008). The total pectin content of sweet cherries varied in all storage conditions. At the end of the storage at 4 °C, the lowest total pectin content was determined in the C-1 coated sweet cherries (2044.64 mg/kg) while the highest total pectin content was observed at CH-1 coated sweet cherries (3084.15 mg/kg). There was no statistically significant difference in the total pectin content of sweet cherries coated with CH-1 at 4 °C until the 20th day ($P > 0.05$) but the total pectin content showed an increase because of the dry matter increased with the loss of water of sweet cherries on the 25th day. At the end of the storage at 20 °C, the lowest total pectin content was determined in the C-2 coated sweet cherries as 2046.20 mg/kg and the highest total pectin content was reported in the CH-2 coated sweet cherries as 2927.71.20 mg/kg. Additionally, the differences in the storage periods of CH-1 coated sweet cherries stored at 20 °C was not significant ($P > 0.05$).

Firmness

The coating of sweet cherries with chitosan caused an increase in firmness at the beginning of storage at both 4 °C and 20 °C. However, the difference between the samples at day 0 was not statistically significant ($P > 0.05$) and the firmness of sweet cherries varied considerably during storage in all storage conditions. The highest firmness value was observed in the CH-1 coated sweet cherries (0.450 N) after storage for 25 days at 4 °C. This value was 0.435 N in C-1 coated sweet cherries, 0.417 N in C-2 coated sweet cherries, 0.370 N in control group and 0.347 N in CH-2 coated sweet cherries. Minimal changes in firmness was observed in the CH-2 coated sweet cherries during storage at 4 °C and the difference in storage periods of sweet cherries coated with CH-2 was also found to be insignificant ($P > 0.05$). As a result, CH-2 was found to be the most effective coating material for preserving sweet cherry firmness when compared to all sample groups at 4 °C. The difference between storage periods of the control sample and between the samples on days 0, 10 and 15 of storage at 20 °C were not significant ($P > 0.05$). This indicates that different coating applications have no effect on firmness of sweet cherries at 20 °C. Similar results were reported by Yaman and Bayındurlu (2002) working with sweet cherry dipping in Semperfresh™ (composed of sucrose esters of fatty acids, sodium carboxymethyl cellulose and mono-diglycerides of fatty acids) and stored at

Table 1 Effect of chitosan coatings on some quality attributes (total anthocyanin content, total phenolic content, ascorbic acid, total pectin content and firmness) of sweet cherries stored at 4 °C and 20 °C

Treatment	Storage time (day)	Total anthocyanin content mg cyanidin-3-rutinoside/kg	Total phenolic content mg/kg	Ascorbic acid mg/100 g	Total pectin content mg/kg	Firmness N	
4 °C	Control	0	132.85 ± 1.05 ^{De}	440.00 ± 1.01 ^{Ba}	0.022 ± 0.045 ^{Ab}	1849.78 ± 265.06 ^{Cb}	0.402 ± 0.04 ^{ABa}
		5	223.27 ± 17.68 ^{Cc}	408.92 ± 20.18 ^{Da}	0.302 ± 0.604 ^{Ab}	2380.48 ± 124.8 ^{Bbc}	0.340 ± 0.04 ^{Ba}
		10	219.93 ± 26.85 ^{Cc}	572.14 ± 5.0 ^{Aa}	0 ± 0 ^{Aa}	2720.96 ± 53.9 ^{Aa}	0.422 ± 0.04 ^{ABa}
		15	354.81 ± 7.54 ^{Aa}	360.72 ± 3.99 ^{Ea}	0 ± 0 ^{Aa}	2836.02 ± 238 ^{Aa}	0.437 ± 0.08 ^{Aa}
		20	377.74 ± 1.82 ^{Ab}	423.90 ± 2.94 ^{Cc}	0 ± 0 ^{Aa}	2037.60 ± 200.6 ^{Cbc}	0.410 ± 0.06 ^{ABab}
		25	287.34 ± 18.69 ^{Bc}	451.00 ± 5.94 ^{Bd}	0 ± 0 ^{Aa}	2638.22 ± 283.3 ^{ABb}	0.370 ± 0.04 ^{ABab}
CH-1	0	182.78 ± 0.50 ^{De}	365.00 ± 2.02 ^{Db}	0.722 ± 0.522 ^{Aa}	2303.85 ± 526.42 ^{Bab}	0.428 ± 0.08 ^{Aa}	
	5	332.77 ± 27.61 ^{Ca}	423.57 ± 19.20 ^{Ca}	0.703 ± 0.611 ^{Aab}	1973.83 ± 98.81 ^{Bc}	0.385 ± 0.05 ^{ABa}	
	10	345.30 ± 3.56 ^{Cab}	571.78 ± 5.13 ^{Aa}	0 ± 0 ^{Ba}	2232.63 ± 410 ^{Bb}	0.395 ± 0.04 ^{ABa}	
	15	495.44 ± 205.16 ^{ABa}	359.61 ± 6.37 ^{Da}	0 ± 0 ^{Ba}	2546.90 ± 40.48 ^{Bb}	0.432 ± 0.02 ^{Aa}	
	20	387.55 ± 19.51 ^{BCb}	434.65 ± 6.99 ^{Cb}	0 ± 0 ^{Ba}	2279.77 ± 529.1 ^{Bab}	0.327 ± 0.02 ^{Bc}	
	25	582.82 ± 44.60 ^{Aa}	513.22 ± 5.13 ^{Bb}	0 ± 0 ^{Ba}	3085.13 ± 117.6 ^{Aa}	0.450 ± 0.05 ^{Aa}	
CH-2	0	231.62 ± 0.50 ^{Ca}	358.57 ± 1.01 ^{Cc}	0.454 ± 0.409 ^{Bab}	1888.83 ± 348.14 ^{Cub}	0.411 ± 0.03 ^{Aa}	
	5	298.01 ± 33.99 ^{Cb}	323.57 ± 32.47 ^{Db}	0.858 ± 0.173 ^{Aab}	2991.73 ± 68.71 ^{Aa}	0.387 ± 0.07 ^{Aa}	
	10	417.04 ± 120.10 ^{Ba}	516.78 ± 41.90 ^{Bb}	0 ± 0 ^{Ca}	2104.39 ± 31.2 ^{Cb}	0.397 ± 0.03 ^{Aa}	
	15	410.43 ± 71.64 ^{Ba}	353.77 ± 1.9 ^{CDa}	0 ± 0 ^{Ca}	2232.16 ± 57.4 ^{Cc}	0.385 ± 0.05 ^{Aa}	
	20	576.60 ± 67.80 ^{Aa}	489.65 ± 1.25 ^{Ba}	0 ± 0 ^{Ca}	2641.20 ± 174.3 ^{Ba}	0.392 ± 0.03 ^{Abc}	
	25	595.62 ± 83.36 ^{Aa}	554.88 ± 6.44 ^{Aa}	0 ± 0 ^{Ca}	2591.68 ± 407.5 ^{Bb}	0.347 ± 0.04 ^{Ab}	
C-1	0	174.78 ± 0.98 ^{Dd}	327.85 ± 2.02 ^{Dd}	0.892 ± 0.379 ^{Aa}	2404.46 ± 222.21 ^{ABa}	0.476 ± 0.06 ^{Aa}	
	5	254.13 ± 16.50 ^{Cc}	413.57 ± 49.04 ^{Ba}	1.259 ± 0.839 ^{Aa}	2719.00 ± 282.19 ^{Aab}	0.407 ± 0.04 ^{ABa}	
	10	287.25 ± 18.41 ^{Cbc}	425.00 ± 6.70 ^{Bd}	0 ± 0 ^{Ba}	2179.96 ± 277 ^{BCb}	0.397 ± 0.02 ^{ABa}	
	15	387.18 ± 4.45 ^{ABa}	361.00 ± 6.66 ^{Ca}	0 ± 0 ^{Ba}	2574.31 ± 218 ^{Ab}	0.477 ± 0.05 ^{Aa}	
	20	325.32 ± 55.59 ^{Bcb}	427.90 ± 3.55 ^{Bbc}	0 ± 0 ^{Ba}	1694.48 ± 79.77 ^{Dc}	0.342 ± 0.08 ^{Bbc}	
	25	420.80 ± 103.80 ^{Ab}	520.16 ± 4.29 ^{Ab}	0 ± 0 ^{Ba}	2044.64 ± 66.11 ^{Cc}	0.435 ± 0.07 ^{ABa}	
C-2	0	193.63 ± 0.49 ^{Cb}	272.85 ± 1.01 ^{Ee}	0.993 ± 0.468 ^{Aa}	2131.71 ± 57.25 ^{ABab}	0.441 ± 0.007 ^{ABa}	
	5	235.05 ± 5.48 ^{BCc}	387.50 ± 6.42 ^{Ca}	0.740 ± 0.237 ^{Aab}	2414.51 ± 733.4 ^{Abc}	0.342 ± 0.02 ^{Ca}	
	10	272.13 ± 26.18 ^{Bbc}	479.28 ± 10.75 ^{Bc}	0 ± 0 ^{Ba}	2177.91 ± 280 ^{ABb}	0.430 ± 0.03 ^{ABa}	
	15	388.18 ± 53.48 ^{Aa}	343.22 ± 0.90 ^{Db}	0 ± 0 ^{Ba}	2239.65 ± 129 ^{ABc}	0.392 ± 0.09 ^{BCa}	
	20	350.57 ± 36.11 ^{Ab}	389.15 ± 10.43 ^{Cd}	0 ± 0 ^{Ba}	1797.26 ± 369.1 ^{Bbc}	0.472 ± 0.02 ^{Aa}	
	25	386.50 ± 42.97 ^{Abc}	501.55 ± 2.12 ^{Ac}	0 ± 0 ^{Ba}	2147.23 ± 81.55 ^{ABc}	0.410 ± 0.04 ^{ABcab}	

Table 1 continued

Treatment	Storage time (day)	Total anthocyanin content mg cyanidin-3-rutinoside/kg	Total phenolic content mg/kg	Ascorbic acid mg/100 g	Total pectin content mg/kg	Firmness N
20 °C						
Control	0	132.85 ± 1.05 ^{De}	440.00 ± 1.01 ^{Ca}	0.022 ± 0.045 ^{Ab}	1849.78 ± 265.06 ^{Bb}	0.402 ± 0.04 ^{Aa}
	5	279.04 ± 71.40 ^{Cb}	422.14 ± 7.99 ^{Da}	0 ± 0 ^{Aa}	2658.18 ± 245.58 ^{Aa}	0.400 ± 0.07 ^{Aa}
	10	444.53 ± 66.17 ^{Bc}	588.92 ± 9.64 ^{Ac}	0 ± 0 ^{Aa}	2669.75 ± 213.52 ^{Aa}	0.377 ± 0.01 ^{Aa}
	15	685.67 ± 5.12 ^{Ab}	496.83 ± 13.34 ^{Bb}	0 ± 0 ^{Aa}	2492.72 ± 491.02 ^{Aa}	0.332 ± 0.01 ^{Aa}
	0	182.78 ± 0.50 ^{De}	365.00 ± 2.02 ^{Db}	0.722 ± 0.522 ^{Aa}	2303.85 ± 526.42 ^{Aab}	0.428 ± 0.08 ^{Aa}
CH-1	5	278.72 ± 71.23 ^{Cb}	388.21 ± 3.75 ^{Ca}	0.159 ± 0.319 ^{Ba}	2724.38 ± 467.28 ^{Aa}	0.307 ± 0.02 ^{Bb}
	10	500.48 ± 32.96 ^{Bbc}	558.92 ± 4.57 ^{Ad}	0 ± 0 ^{Ba}	2756.25 ± 194.99 ^{Aa}	0.415 ± 0.06 ^{Aa}
	15	643.03 ± 36.98 ^{Abc}	518.77 ± 2.02 ^{Ba}	0 ± 0 ^{Ba}	2514.52 ± 297.73 ^{Aa}	0.362 ± 0.01 ^{ABa}
	0	231.62 ± 0.50 ^{Ca}	358.57 ± 1.01 ^{Cc}	0.454 ± 0.409 ^{Ab}	1888.83 ± 348.14 ^{Bab}	0.411 ± 0.03 ^{Aa}
	5	276.27 ± 59.38 ^{Cb}	263.92 ± 39.18 ^{Db}	0.158 ± 0.316 ^{ABa}	1939.92 ± 148.73 ^{Bb}	0.367 ± 0.02 ^{ABab}
CH-2	10	545.01 ± 105.19 ^{Bb}	543.21 ± 7.03 ^{Ac}	0 ± 0 ^{Ba}	2785.30 ± 239.82 ^{Aa}	0.362 ± 0.04 ^{ABa}
	15	855.57 ± 42.82 ^{Aa}	467.11 ± 1.43 ^{Bc}	0 ± 0 ^{Ba}	2927.71 ± 66.46 ^{Aa}	0.337 ± 0.02 ^{Ba}
	0	174.78 ± 0.98 ^{Dd}	327.85 ± 2.02 ^{Cd}	0.892 ± 0.379 ^{Aa}	2404.46 ± 222.21 ^{BCa}	0.476 ± 0.06 ^{Aa}
	5	444.43 ± 31.39 ^{Ca}	259.28 ± 29.98 ^{Db}	0.312 ± 0.399 ^{Ba}	2882.62 ± 148.73 ^{Aa}	0.377 ± 0.01 ^{Ba}
	10	649.58 ± 31.96 ^{Ba}	647.85 ± 10.87 ^{Ab}	0 ± 0 ^{Ba}	2185.19 ± 85.90 ^{Cb}	0.387 ± 0.01 ^{Ba}
C-2	15	605.54 ± 1.79 ^{Ac}	492.94 ± 6.75 ^{Bb}	0 ± 0 ^{Ba}	2587.20 ± 86.91 ^{Ba}	0.365 ± 0.06 ^{Ba}
	0	193.63 ± 0.49 ^{Db}	272.85 ± 1.01 ^{De}	0.993 ± 0.468 ^{Aa}	2131.71 ± 57.25 ^{Bab}	0.441 ± 0.007 ^{Aa}
	5	337.76 ± 0.99 ^{Cb}	386.42 ± 8.08 ^{Ca}	0.456 ± 0.912 ^{ABa}	2808.51 ± 201.41 ^{Aa}	0.372 ± 0.04 ^{BCa}
	10	491.50 ± 14.22 ^{Bbc}	665.71 ± 1.84 ^{Aa}	0 ± 0 ^{Ba}	2673.98 ± 95.30 ^{Aa}	0.385 ± 0.03 ^{Ba}
	15	643.70 ± 53.29 ^{Abc}	497.94 ± 1.66 ^{Bb}	0 ± 0 ^{Ba}	2046.20 ± 156.48 ^{Bb}	0.325 ± 0.03 ^{Ca}

Values are expressed as means ± standard deviations, $n = 4$.

CH-1: Chitosan-1, CH-2: Chitosan-2, C-1: Commercial-1, C-2: Commercial-2

^{A, B} different capital letters indicate statistical differences of the same samples at the same column ($p < 0.05$); each storage temperature was assessed among themselves

^{a, b} different lowercase letters indicate statistical differences of the same storage times at the same column ($p < 0.05$); each storage temperature was assessed among themselves

Table 2 The colour values (L*, a*, b*, ΔE) of sweet cherries stored at 4 °C and 20 °C

Treatment	Storage time (day)	L*	a*	ΔE
4 °C				
Control	0	23.13 ± 0.82 ^{Ba}	34.59 ± 1.12 ^{Bb}	59.19 ± 0.82 ^{Bb}
	5	21.17 ± 2.44 ^{Ba}	47.82 ± 3.62 ^{Aa}	65.06 ± 1.83 ^{Aa}
	10	29.01 ± 2.21 ^{Aa}	27.92 ± 2.53 ^{Ca}	42.21 ± 2.71 ^{Da}
	15	13.19 ± 3.73 ^{Cb}	49.17 ± 5.64 ^{Aab}	58.35 ± 3.14 ^{Bab}
	20	26.76 ± 1.46 ^{Ab}	23.65 ± 0.52 ^{Ca}	36.52 ± 1.53 ^{Eab}
CH-1	25	21.26 ± 0.76 ^{Ba}	27.66 ± 3.05 ^{Ca}	50.52 ± 2.93 ^{Ca}
	0	23.02 ± 1.33 ^{BCa}	34.09 ± 1.82 ^{Bb}	55.60 ± 2.11 ^{Bc}
	5	14.01 ± 4.30 ^{Eb}	48.10 ± 4.10 ^{Aa}	56.20 ± 3.27 ^{Bbc}
	10	27.24 ± 1.92 ^{Aab}	22.72 ± 1.72 ^{CDb}	36.55 ± 1.88 ^{Cb}
	15	17.69 ± 3.05 ^{DEab}	51.17 ± 6.45 ^{Aa}	62.58 ± 2.35 ^{Aa}
CH-2	20	25.01 ± 2.16 ^{ABb}	20.87 ± 5.51 ^{Da}	33.03 ± 4.34 ^{Cb}
	25	19.44 ± 1.72 ^{CDab}	27.60 ± 1.61 ^{Ca}	48.79 ± 2.57 ^{Cab}
	0	21.06 ± 1.06 ^{CDab}	36.73 ± 2.72 ^{Bb}	55.71 ± 2.13 ^{Ac}
	5	22.13 ± 0.76 ^{BCa}	40.27 ± 1.36 ^{ABb}	59.78 ± 2.04 ^{Ab}
	10	28.89 ± 1.08 ^{ABb}	24.92 ± 4.36 ^{Cab}	39.02 ± 2.99 ^{Cab}
C-1	15	14.62 ± 2.71 ^{Eab}	43.30 ± 2.36 ^{Abc}	51.40 ± 3.61 ^{Bc}
	20	24.66 ± 2.24 ^{Bb}	22.21 ± 2.63 ^{Ca}	33.45 ± 3.32 ^{Db}
	25	18.31 ± 2.64 ^{Dab}	25.33 ± 1.69 ^{Ca}	42.38 ± 2.52 ^{Cc}
	0	17.21 ± 5.20 ^{Bb}	34.86 ± 1.64 ^{Bb}	51.56 ± 3.19 ^{Bd}
	5	21.18 ± 1.32 ^{Ba}	39.55 ± 3.85 ^{Ab}	58.16 ± 2.58 ^{Abc}
C-2	10	27.13 ± 1.99 ^{Aab}	25.41 ± 1.71 ^{Cab}	38.00 ± 2.26 ^{Db}
	15	18.23 ± 3.25 ^{Ba}	37.22 ± 1.12 ^{ABc}	51.94 ± 3.27 ^{Bc}
	20	30.24 ± 0.65 ^{Aa}	23.65 ± 3.15 ^{Ca}	40.04 ± 2.15 ^{Da}
	25	17.31 ± 2.20 ^{Bb}	26.15 ± 3.24 ^{Ca}	45.17 ± 3.65 ^{Cbc}
	0	22.64 ± 2.09 ^{Ba}	44.08 ± 2.98 ^{Aa}	62.68 ± 0.70 ^{Aa}
Control	5	23.64 ± 2.36 ^{ABa}	33.31 ± 1.07 ^{Bc}	54.55 ± 1.75 ^{Bc}
	10	26.06 ± 1.26 ^{Ab}	25.57 ± 2.22 ^{Cab}	37.89 ± 1.61 ^{Cb}
	15	14.88 ± 1.86 ^{Cab}	43.10 ± 5.78 ^{Abc}	53.56 ± 5.64 ^{Bbc}
	20	25.58 ± 1.53 ^{Ab}	20.10 ± 1.53 ^{Da}	33.29 ± 1.63 ^{Db}
	25	21.07 ± 1.53 ^{Ba}	28.37 ± 2.34 ^{Ca}	50.69 ± 1.16 ^{Ba}
20 °C				
Control	0	23.13 ± 0.82 ^{Aa}	34.59 ± 1.12 ^{Cb}	59.19 ± 0.82 ^{Ab}
	5	11.29 ± 2.36 ^{Bc}	53.92 ± 1.22 ^{Aa}	59.03 ± 1.80 ^{Aab}
	10	24.81 ± 1.65 ^{Ac}	20.14 ± 1.65 ^{Da}	32.22 ± 1.64 ^{Cb}
	15	13.56 ± 1.98 ^{Ba}	41.45 ± 2.59 ^{Bbc}	50.27 ± 1.81 ^{Babc}
	0	23.02 ± 1.33 ^{Aa}	34.09 ± 1.82 ^{Bb}	55.60 ± 2.11 ^{Ac}
CH-1	5	16.37 ± 3.41 ^{Bab}	39.95 ± 5.37 ^{Bc}	50.78 ± 5.62 ^{Ac}
	10	27.07 ± 1.82 ^{Abc}	21.50 ± 2.41 ^{Ca}	34.97 ± 2.26 ^{Bab}
	15	9.78 ± 4.30 ^{Cab}	53.73 ± 5.76 ^{Aa}	55.16 ± 5.78 ^{Aa}
CH-2	0	21.06 ± 1.06 ^{Bab}	36.73 ± 2.72 ^{Bb}	55.34 ± 0.44 ^{Ac}
	5	13.31 ± 4.10 ^{Cbc}	48.75 ± 4.16 ^{Aab}	59.88 ± 2.86 ^{Ba}
	10	29.93 ± 1.49 ^{Aa}	20.67 ± 2.90 ^{Ca}	36.87 ± 2.57 ^{Da}
	15	8.59 ± 2.56 ^{Db}	40.43 ± 3.84 ^{Bbc}	45.51 ± 2.79 ^{Cbc}
C-1	0	17.21 ± 5.20 ^{Bb}	34.86 ± 1.64 ^{Bb}	52.30 ± 3.31 ^{Ad}
	5	17.97 ± 1.86 ^{Ba}	41.27 ± 4.64 ^{ABc}	56.46 ± 4.70 ^{Aabc}
	10	29.05 ± 2.20 ^{Aab}	21.26 ± 0.43 ^{Ca}	36.42 ± 1.96 ^{Bab}
	15	11.13 ± 2.21 ^{Cab}	46.82 ± 8.34 ^{Aab}	52.07 ± 7.44 ^{Aab}

Table 2 continued

Treatment	Storage time (day)	L*	a*	ΔE
C-2	0	22.64 ± 2.09 ^{Ba}	44.08 ± 2.98 ^{Aa}	62.68 ± 0.70 ^{Aa}
	5	19.34 ± 1.49 ^{Ca}	45.33 ± 4.14 ^{Abc}	52.92 ± 3.41 ^{Bcd}
	10	28.03 ± 1.36 ^{Ab}	19.37 ± 5.72 ^{Ca}	34.77 ± 3.97 ^{Dab}
	15	11.91 ± 0.52 ^{Dab}	37.67 ± 2.68 ^{Bc}	44.53 ± 2.05 ^{Cc}

Values are expressed as means ± standard deviations, $n = 4$.

CH-1: Chitosan-1, CH-2: Chitosan-2, C-1: Commercial-1, C-2: Commercial-2

^{A, B} different capital letters indicate statistical differences of the same samples at the same column ($p < 0.05$); each storage temperature was assessed among themselves

^{a, b} different lowercase letters indicate statistical differences of the same storage times at the same column ($p < 0.05$); each storage temperature was assessed among themselves

cold temperature had a strong effect on the retention of firmness values than ambient temperature.

Colour

Skin colour is the most important indicator of quality and maturity of sweet cherry. The colour values (L^* , a^* , and ΔE) of sweet cherries stored at 4 °C and 20 °C were shown in Table 2. At the beginning and at the end of the storage periods in all samples, the highest L^* value was determined in the control group. The L^* values of the control group at the end of storage at 4 °C and 20 °C are 21.26 and 13.56, respectively.

Similarly, Han et al. (2014) who treated Sponge gourd with 0.5% and 1% chitosan and stored at 25 °C also determined the highest L^* value in the control group at the end of the storage which was 12.7% higher than 0.5% chitosan treated fruits and 17.7% higher than 1.0% chitosan treated fruits.

The a^* values, which have reached the highest values on the 15th day, have started to decrease since the 20th day with the beginning of microbial activities and deterioration reactions at 4 °C. On day 5 at 20 °C, an increase on a^* value of the control sample was observed depending on the increase in maturity. It has been found that the coating sweet cherries with chitosan causes a slower increase in a^* values by retarding maturity due to slowing the respiration rate. Chien et al. (2007), treated sliced mango with aqueous solutions of 0%, 0.5%, 1% or 2% chitosan (deacetylation degree is 95–98%), also stated that increase in the a^* values of chitosan treatment samples was lower than the control group. The fluctuations in the b^* values of the all samples have occurred under all storage conditions.

At the end of the storage, the minimum change in total colour differences (ΔE) occurred in the CH-2 coated sweet

cherries (42.38) stored at 4 °C and in the C-2 coated sweet cherries at 20 °C with 44.53. Çetin (2012) also reported that total colour changes were higher in the control group than coated groups.

Conclusion

This study revealed the positive influence of all chitosan coatings on functional compounds of sweet cherries. CH-1 showed significant effects in the preservation of firmness; CH-2 exhibited the highest total anthocyanin (an increase of 257.15%) and total phenolic content (554.88 mg/kg); C-1 delayed changes in ascorbic acid content and displayed the highest antioxidant capacity (2.1690 μM Trolox equivalents (TE)/kg) at 4 °C. Furthermore, it was found that CH-1 demonstrated the highest total phenolic content (518.77 mg/kg); CH-2 displayed the highest total anthocyanin content (an increase of 369.38%); C-1 had the highest firmness value (0.365 N) and C-2 exhibited the highest ascorbic acid content at 20 °C. As a result, each chitosan coating tested has different effects on different quality properties at different storage temperatures. So, we suggest that application of the coating could be considered for commercial application during storage and marketing. For all that, in this study no external packaging material was used except chitosan during storage of sweet cherries. However, it is considered that chitosan coating application may be more effective in increasing the commercial value and shelf life of sweet cherries and preserving the quality characteristics by using combined modified atmosphere packaging or controlled atmosphere storage.

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