



Refrigerated storage of blackberry cultivar ‘BRS Cainguá’ harvested at different ripeness stages

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ABSTRACT

Blackberries have a short post-harvest conservation period, making it necessary to optimize harvest and storage conditions. The objective of this study was to evaluate the impact of refrigerated storage and maturation stage on the physicochemical quality of ‘BRS Cainguá’ blackberries, produced in an organic system. Blackberries from an experimental orchard, were harvested at three ripeness stages (RS), with RS1 being fruits with 100% red skin; RS2 skin 50% red and 50% black; and EM3 100% black skin. The samples were stored in a cold chamber at $4\pm 0.5^{\circ}\text{C}$ and 90-95% RH for 12 days. The evaluations were carried out after 0, 3, 6, 9 and 12 days of storage, for: weight loss (WL), soluble solids (SS), pH, titratable acidity (TA), ratio (SS/TA) and color parameters. Blackberries harvested in RS3 had the highest SS content, lowest TA and the highest SS/TA ratio and pH. During storage, TA and WL showed decreasing and increasing linear responses, respectively, regardless of the RS of the fruits. Harvesting based on the skin color of ‘BRS Cainguá’ blackberries influences the physicochemical quality and postharvest conservation. Fruits harvested with 100% black skin, have the best quality and are the most suitable for subsequent refrigerated storage.

Keywords: *Rubus* spp.; physicochemical; small fruits; postharvest; flavor; harvest point.

INTRODUCTION

The blackberry (*Rubus* spp.), which belongs the group of small fruits, along with strawberries, blueberries and raspberries is considered a functional food, due to its high content of phytochemical compounds, such as anthocyanins and phenolic compounds. These are responsible for the high antioxidant potential of these fruits and help to fight and prevent diseases (Acosta-Montoya *et al.*, 2010; Vizzotto *et al.*, 2012).

The BRS Cainguá cultivar was launched in 2019, by Embrapa Clima Temperado Blackberry Genetic Improvement Program, its fruits are elongated, have a good balance between acidity and sugar, which gives them

good acceptance for the *in natura* market. The plant has a lower density of thorns and is also less susceptible to rust, when compared to ‘Tupy’, the main cultivar used in Brazil (Raseira *et al.*, 2020). Which makes it an excellent option for cultivation in alternative production techniques, such as organic and agroecological systems (Cavender *et al.*, 2019).

The physicochemical, morphological, and physiological characteristics of the blackberry make it very perishable, posing a challenge to overcome the barrier that it presents in relation to its shelf life, as it is very susceptible to mechanical damage and has a high respiratory rate (Soethe *et*

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al., 2019; Carvajal *et al.*, 2021). The use of techniques that increase the shelf life of fruits, without altering the physicochemical, sensory and nutritional characteristics, are of great importance to reduce post-harvest losses (Bersaneti *et al.*, 2021). In this sense, one of the main factors that affect the conservation and quality of fruits is the storage temperature, as it regulates the speed at which metabolic processes occur and the storage time (Antunes *et al.*, 2003). The use of low temperature during post-harvest storage delays the occurrence of fungi, reduces respiratory rate and water loss, prolonging fruit conservation (Oliveira *et al.*, 2013). By reducing respiration, the aroma, color, flavor, textures and other quality attributes of the stored fruits are preserved (Chitarra & Chitarra, 2005). On the other hand, if fruits are harvested before optimal maturity, their postharvest life is extended, but the physicochemical quality and acceptability are reduced (Brackmann *et al.*, 2017).

Seeking to supply more distant markets, some blackberry producers harvest at less advanced ripeness stages, as they are less susceptible to mechanical damage, prolonging their conservation. However, if fruits are harvested before optimal maturity, their postharvest life is extended, but the physicochemical quality and acceptability by the consumer marked are reduced (Brackmann *et al.*, 2017). According to Threlfall *et al.* (2016) blackberries whose objective is the in natura market should have a soluble solids content around 9-10 °Brix, titratable acidity of 0.9% to 1% citric acid and a soluble solids/titratable acidity (ratio) from 10 to 13. Therefore, it is necessary to find a balance between the ripeness stage, quality, and postharvest conservation of blackberries.

Being a recently launched cultivar, there is no information available in the extant literature about the physicochemical quality of 'BRS Caingua', harvested at different ripeness stages, as well as its postharvest conservation. Given the above, the objective of this work was to evaluate the impact of refrigerated storage and ripeness stage on the physicochemical quality of 'BRS Caingua' blackberries, produced in an organic system.

MATERIAL AND METHODS

Blackberries of the BRS Caingua cultivar were harvested from an experimental orchard of Embrapa Clima Temperado, Pelotas-RS (31°37'09" S and 52°31'33" W and altitude of 70 m) during the 2019/2020 crop. The climate classification of the region is classified, according to W. Köppen as "Cfa"- humid subtropical climate, that

is, humid temperate with hot summers. The local soil is moderately deep with medium texture on the A horizon and clayey on the B horizon, classified as Red Yellow Argisol (Santos *et al.*, 2006). The orchard was managed in an organic production system, with spacing between plants of 1 m x 5 m (2000 plants per hectare), conducted in a "T" espalier system.

Fruits were harvested manually, between 6 and 7 am, separated into three ripeness stages according to the visual assessment of the skin color, with the ripeness stages being defined as: ripeness stage one (RS1), where the fruits had a 100% red skin (Figure 1A); ripeness stage two (RS2), when the skin was 50% red and 50% black (Figure 1B); and ripeness stage three (RS3), with 100% black fruit skin (Figure 1C).

As soon as the fruits were harvested, they were taken to the Post-harvest Laboratory of the Fruit Growing sector of the Federal University of Pelotas, where the blackberry selection process was carried out, and those with mechanical injuries, fungi and/or insect attacks, or other defects were discarded, separating them into uniform batches according to the ripeness stages. Samples composed of ten fruits were placed in plastic containers measuring 19 x 12 x 4.9 cm and stored in a cold chamber at a temperature of 4±0.5°C and a relative humidity of 90-95%, for 12 days.

The experimental design used was completely randomized, in a 3 x 5 factorial scheme (three ripeness stages and five storage periods), with four replications, each consisting of ten fruits.

The storage periods were the day of harvest (day 0), 3, 6, 9 and 12 days of refrigerated storage. The physicochemical analyses of the blackberries harvested at different ripeness stages were carried out during each of the storage periods. The mass loss was determined by the difference between the fruit mass on the day of harvest (day zero) and at the time of removal from the cold chamber, and the results are expressed in percentage. The soluble solids content was obtained with a digital refractometer (Atago® brand model PAL-1), with the results expressed in °Brix. The pH was determined in a pHmeter (AZ brand model 86505) and the determination of total acidity was performed by titration of a sample, consisting of 5 mL of juice diluted in 90 mL of distilled water, using a standardized 0.1 N NaOH solution, adopting pH 8.1 as the end point of the titration, and the result expressed as a percentage of citric acid.

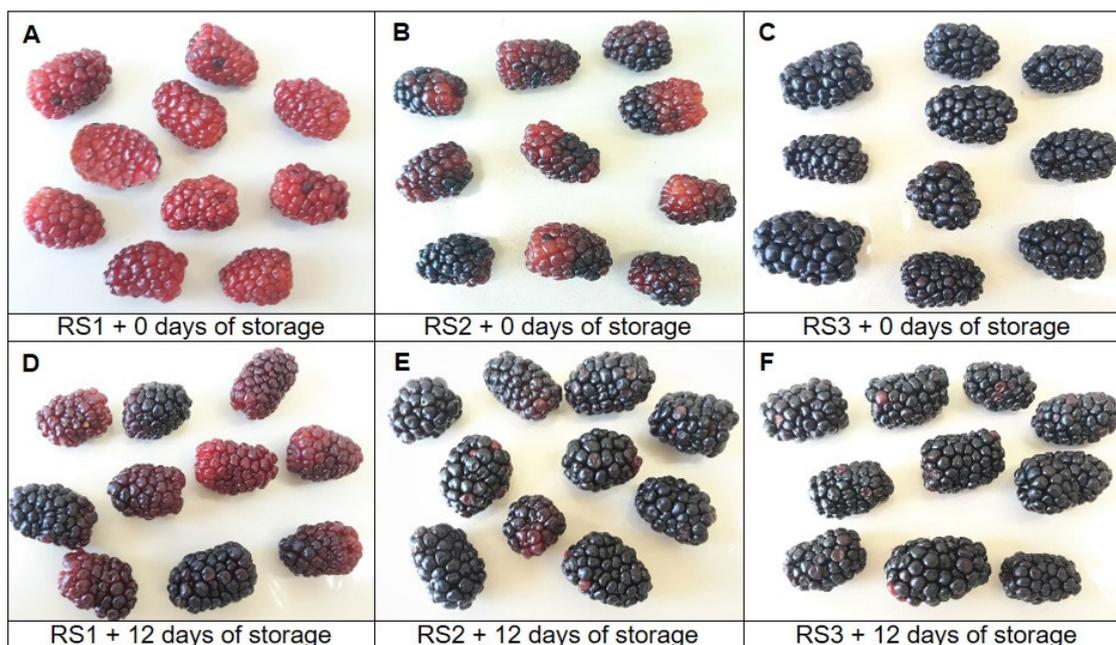


Figure 1: ‘BRS Caingua’ blackberries harvested at different stages of ripeness and submitted to refrigerated storage. A: fruits with a 100% red skin (RS1); B: fruits with an 50% red and 50% black skin (RS2); C: fruits with 100% black skin (RS3) on harvest day (day 0). D: fruits with a 100% red skin (RS1); E: fruits with an 50% red and 50% black skin (RS2); F: fruits with 100% black skin (RS3) after 12 days of refrigerated storage.

The SS/TA ratio was determined by obtaining the quotient of the soluble solids content and the titratable acidity. To assess the color of the skin, a colorimeter was used (Minolta Chroma Meter CR-400/410), with a D65 light source, expressed by the rectangular coordinate system L^* , a^* and b^* according to the CIE (Commission Internationale de E’clairage), where L^* expresses the luminosity (0 = black and 100 = white), a^* represents the red (+) or green (-) colors and b^* the yellow (+) or blue (-) colors. From these data, the color intensity (chroma) was calculated using the equation $C = [(a^*)^2 + (b^*)^2]^{\frac{1}{2}}$, and the color tone (Hue angle), through the equation $^{\circ}Hue = \tan^{-1}b^* / a^*$

The data obtained were subjected to analysis of variance and the variables whose results revealed significance ($p < 0.05$), a regression analysis was performed for the quantitative factor (storage periods) and for the qualitative factor (ripeness stages), the means were compared by the Tukey test at 5% significance.

RESULTS AND DISCUSSION

Significant variation was observed in all but soluble solids (SS) content due to the interaction between the

ripeness stages and the storage periods. There was significant variation for the ripeness stages only when the factors were analyzed separately (Table 1). Fruits harvested at the RS3 had the highest levels of soluble solids, while those from RS1 had the lowest. According to Schulz *et al.* (2019), this is related to a series of biochemical and physiological processes that occur during maturation, including starch hydrolysis, in which carbohydrates are transformed into simpler, water-soluble sugars. In addition to organic acids, including citric acids, which are also converted into sugars as maturation progresses (Moreno & Oyla, 2016) and together lead to an increase in the soluble solids content of fruits. It is worth noting that the fruits harvested in RS3 had an average SS content of 11.54 °Brix, which is higher than that found by Soethe *et al.* (2019), who at the end of 8 days of storage in modified atmosphere, found an SS content of 8.5 °Brix for the Tupy cultivar, which is currently the most cultivated in Brazil. This shows the great marketing potential of the BRS Caingua cultivar for *in natura* consumption, due to the high SS content of its fruits.

During the storage period, there were no changes in the soluble solids content (Table 1), with an average value of 9.66 °brix. According to Oliveira *et al.* (2014), the use of

low temperatures during storage makes it possible to reduce the metabolic activity of fruits, preventing the degradation of soluble solids during the respiratory process, and minimizing the loss of fruit mass, due to

loss of water, which can increase the concentration of SS. Showing that the conservation method used in this study was suitable for the SS conservation of 'BRS Caingua' blackberries.

Table 1: Soluble solids, titratable acidity of blackberries harvested at different ripeness stages and submitted to refrigerated storage for 12 days

| Soluble solids (°brix) | | | | | | | | |
|------------------------------------|----------------|--------|--------|--------|--------|----------------------|-----------|-----------|
| Ripeness stage | Storage (days) | | | | | Mean | Linear | Quadratic |
| | 0 | 3 | 6 | 9 | 12 | | | |
| RS1 | 8.35 | 8.08 | 8.35 | 7.95 | 8.02 | 8.15 c | - | - |
| RS2 | 9.72 | 9.38 | 8.75 | 9.48 | 9.18 | 9.30 b | - | - |
| RS3 | 11.42 | 11.58 | 11.58 | 11.32 | 11.88 | 11.54 a | - | - |
| Mean | 9.83 | 9.68 | 9.53 | 9.58 | 9.69 | | ns | ns |
| p (ripeness) | | | | | | 0.0001** | | |
| p (storage) | | | | | | 0.6052 ^{ns} | | |
| p (ripeness x storage) | | | | | | 0.1886 ^{ns} | | |
| CV (%) | | | | | | 4.99 | | |
| Titratable acidity (% Citric Acid) | | | | | | | | |
| Ripeness stage | Storage (days) | | | | | Linear | Quadratic | |
| | 0 | 3 | 6 | 9 | 12 | | | |
| RS1 | 2.13 a | 1.98 a | 1.83 a | 1.89 a | 1.74 a | *(1) | ns | |
| RS2 | 1.62 b | 1.50 b | 1.37 b | 1.09 b | 1.06 b | *(2) | ns | |
| RS3 | 0.97 c | 1.01 c | 0.93 c | 0.85 c | 0.78 c | *(3) | ns | |
| p (ripeness x storage) | | | | | | 0.0013** | | |
| CV (%) | | | | | | 7.09 | | |

Means followed by the same letter in the columns do not differ significantly from each other, by the Tukey test at 5% significance. ^{ns}non-significant for regression analysis; * significant to 5% probability; ** significant to 1% probability; ⁽¹⁾ $y = -0.0292x + 2.089$ ($R^2 = 0.86$); ⁽²⁾ $y = -0.0502x + 1.629$ ($R^2 = 0.95$); ⁽³⁾ $y = -0.0175x + 1.0141$ ($R^2 = 0.83$) CV: Coefficient of variation; RS1: fruits with 100% red skin; RS2: 50% red and 50% black skin; RS3: 100% black skin.

Titratable acidity is an indirect measure of the organic acids present in fruits, which together with sugars are responsible for the flavor of blackberries. According to Horvitz *et al.* (2017), the citric acid is the main acid found in blackberries, representing more than 90% of the total organic acids present. Therefore, by expressing the titratable acidity of blackberries as a percentage of citric acid, a good parameter is obtained in relation to organic acids and fruit flavor.

Fruits from RS1, harvested with a 100% red skin, had the highest titratable acidity in all periods of refrigerated storage, and fruits harvested at RS3 had the lowest acidity (Table 1). This is due to the ripeness stage at which the fruits were harvested, because with the advancement of maturation, there is a decrease in titratable acidity and an

increase in soluble solids, due to the conversion of acids into sugars (Cavender *et al.*, 2019). This same phenomenon was observed by Samaniego *et al.* (2020), for the Brazos cultivar harvested at different ripeness stages, with the blackberries harvested with 100% black skin had the lowest titratable acidity and the highest SS content.

During refrigerated storage, there was a reduction in the titratable acidity, with a decreasing linear response (Table 1), for all ripeness stages. According to Brackmann *et al.* (2017), the reduction in titratable acidity during storage is associated with the respiratory metabolism of fruits, where substrates such as organic acids are degraded, and is directly related to storage temperature, since the lower the temperature, the greater the reduction in the fruit respiratory rate and the degradation of acids will be lower.

The soluble solids content (SS) and the titratable acidity (TA) ratio is related to the balance between sugars and organic acids present in the fruit and is an important indicator of flavor (Arroyo *et al.* (2020). It is more appropriate to use the relationship between them, than just one of them independently. Fruits harvested with 100% black skin (RS3) had the highest SS/TA ratios, followed by RS2, regardless of the storage period (Table 2). According to Skrovankova *et al.* (2015), the best quality in relation to

the flavor of blackberries is obtained when the fruit reaches its full maturity, when the skin acquires a 100% black color. Corroborating the results observed in table 1, in which the fruits harvested RS3 (100% black) had the highest SS contents and the lowest titratable acidity contents, which consequently resulted in fruits with a high SS/TA ratio. According to Schulz *et al.*, (2019), blackberries with higher SS/TA ratios have a more pleasant taste, which increases consumer acceptance.

Table 2: Ratio of soluble solids content and titratable acidity, pH and weight loss of blackberries harvested at different ripeness stages and stored in a refrigerated environment for 12 days

| SS/TA Ratio | | | | | | | |
|------------------------|----------------|--------------------|--------------------|--------------------|---------|--------|-----------|
| Ripeness stage | Storage (days) | | | | | Linear | Quadratic |
| | 0 | 3 | 6 | 9 | 12 | | |
| RS1 | 3.92 c | 4.07 c | 4.56 c | 4.22 c | 4.65 c | ns | ns |
| RS2 | 6.03 b | 6.29 b | 6.42 b | 8.66 b | 8.64 b | *(1) | ns |
| RS3 | 11.84 a | 11.53 a | 12.40 a | 13.35 a | 15.13 a | *(2) | ns |
| p (ripeness x storage) | | | | 0.0001** | | | |
| CV (%) | | | | 8.29 | | | |
| pH | | | | | | | |
| Ripeness stage | Storage (days) | | | | | Linear | Quadratic |
| | 0 | 3 | 6 | 9 | 12 | | |
| RS1 | 2.19 c | 2.18 c | 2.29 c | 2.51c | 2.56 c | *(3) | ns |
| RS2 | 2.44 b | 2.43 b | 2.55 b | 2.91 b | 3.09 b | *(4) | ns |
| RS3 | 2.63 a | 2.71 a | 2.78 a | 3.11 a | 3.18 a | *(5) | ns |
| p (ripeness x storage) | | | | 0.0001** | | | |
| CV (%) | | | | 2.03 | | | |
| Weight loss (%) | | | | | | | |
| Ripeness stage | Storage (days) | | | | | Linear | Quadratic |
| | 0 | 3 | 6 | 9 | 12 | | |
| RS1 | - | 0.62 ^{ns} | 1.18 ^{ns} | 1.91 ^{ns} | 2.99 a | *(6) | ns |
| RS2 | - | 0.58 | 0.99 | 1.72 | 2.49 b | *(7) | ns |
| RS3 | - | 0.36 | 0.82 | 1.67 | 1.76 c | *(8) | ns |
| p (ripeness x storage) | | | | 0.0053** | | | |
| CV (%) | | | | 23.57 | | | |

Means followed by the same letter in the columns do not differ significantly from each other, by the Tukey test at 5% significance. ^{ns}not significant for regression analysis; *significant to 5% probability; ** significant to 1% probability; ⁽¹⁾ $y = 0.2523x + 5.6968$ ($R^2 = 0.82$); ⁽²⁾ $y = 0.2799x + 11.172$ ($R^2 = 0.84$); ⁽³⁾ $y = 0.0357x + 2.131$ ($R^2 = 0.88$); ⁽⁴⁾ $y = 0.0584x + 2.334$ ($R^2 = 0.88$); ⁽⁵⁾ $y = 0.0498x + 2.583$ ($R^2 = 0.92$); ⁽⁶⁾ $y = 0.2426x - 0.1165$ ($R^2 = 0.98$); ⁽⁷⁾ $y = 0.2037x - 0.0643$ ($R^2 = 0.98$); ⁽⁸⁾ $y = 0.1607x - 0.042$ ($R^2 = 0.96$); CV: Coefficient of variation; RS1: fruits with 100% red skin; RS2: 50% red and 50% black skin; RS3: 100% black skin.

During refrigerated storage, the SS/TA ratio of RS3 and RS2 fruits show an increasing linear adjustment at the end of 12 days of storage, reaching values of 15.13 and 8.64, respectively, (Table 2). This increase in SS/TA is due to the reduction in TA during fruit storage, as organic acids are used as substrates in respiratory metabolism (Soethe *et al.*,

2019) and the SS content did not change during storage as seen in table 1. Although fruits harvested in RS2 showed changes in the SS/TA ratio during storage, in no period was the SS/TA ratio equal to or greater than that presented by RS3 fruits on the day of harvest, or even after 12 days of storage. This shows that despite changes in fruit flavor,

these did not reach a quality equal to or superior to those harvested with 100% black skin, a stage considered ripe for blackberry (Skrovankova *et al.*, 2015).

However, RS1 fruits, harvested with 100% red skin, did not show significant changes in the SS/TA ratio during storage, with an average ratio of 4.28 (Table 2). However, when compared with the mean values obtained for the ratio of fruits harvested in RS3, which is 12.85, it is observed that it is three times greater than those in RS1. Therefore, RS1 is not suitable for harvesting, as the acidity is still very high and the soluble solids content low. What characterizes fruits of low organoleptic quality, because even after long periods of storage, there are no significant improvements (Brackmann *et al.*, 2017).

The pH varied according to the ripeness stage at which the fruits were harvested, where RS3 had the highest values (Table 2) in all storage periods evaluated, indicating less acidic fruits, corroborating the titratable acidity analysis, where RS3 had the lowest values. RS1 on the other hand had the lowest pH and the highest titratable acidity. This same trend, of increase in pH values and decrease in acidity as ripeness progresses, was observed by Rutz *et al.* (2012) and Souza *et al.* (2017), when evaluating 'Tupy' blackberries harvested at different ripeness stages.

During the refrigerated storage period, all ripeness stages showed an increasing linear response in relation to fruit pH (Table 2). Storing blackberries produced in an organic system for 12 days at 4°C and 85% RH, Cavender *et al.* (2019) also observed an increase in pH and a decrease in the titratable acidity of the fruits. In our study there was an increase of 16.89%, 26.63% and 20.91% in the pH values of fruits collected in RS1, RS2 and RS3, respectively, while TA a reduction of 18.30%, 34.56%, 19.58% for fruits harvested in RS1, RS2 and RS3, respectively, when comparing data from the day of harvest and at the end of the 12 days of refrigerated storage. It is observed that as the pH increases, the degradation of organic acids occurs and the TA values decrease, this reduction is associated with the natural ripeness and senescence process of the fruits (Oliveira *et al.*, 2014).

Fruit mass loss only varied between ripeness stages and 12 days of storage, in which RS1 and RS3 showed the highest and lowest losses, respectively (Table 2). According to Kim *et al.* (2015), weight loss is related to the size of the berry, smaller blackberries have a higher surface/volume ratio and consequently greater weight loss. At harvest, the blackberries from RS1, RS2 and RS3 had an average mass

of 5.7 g.fruit⁻¹, 6.6 g.fruit⁻¹ and 8.1 g.fruit⁻¹, respectively, and due to the smallest average fruit size, RS1 showed the greatest weight loss. However, regardless of the ripeness stage at which the blackberries were harvested and the period of refrigerated storage, all had a lower weight loss than what is considered acceptable for the sale of blackberries, which is a 5% maximum (Salgado & Clark, 2016).

During refrigerated storage, weight loss linearly increased for all ripeness stages (Table 2). The weight loss is related to the rate of transpiration and carbon losses caused by respiration of fruits during storage (Brackmann *et al.*, 2014) and significant reductions in weight loss can be obtained through the storage of fruits at low temperatures (Kim *et al.*, 2015; Horvitz *et al.*, 2017).

For luminosity, it was found that blackberries harvested from the 100% red skin (RS1) obtained the highest values, differing from the other ripeness stages in all storage periods evaluated (Table 3). While RS2 and RS3 only differed at 0 and 6 days of storage. This same trend, decreasing luminosity with advancing ripening, was verified by Horvitz *et al.* (2017), where luminosity was significantly higher in the less mature blackberries, when compared to those harvested at a more advanced ripeness stage. During ripening, the colors tend to become more intense or darker and the luminosity of the fruit's skin decreases, (Samaniego *et al.*, 2020).

During the refrigerated storage, the blackberries harvested at the RS1 and RS2 ripeness stages showed a decreasing linear response to luminosity (Table 3), showing that there was a darkening of the fruit skin, as can be seen in Figure 1. Which is possibly associated with the content of anthocyanins, as according to Wang *et al.* (2009), significant increases in the synthesis of this compound can occur during refrigerated storage, even in those fruits harvested at less advanced ripeness stages. Rigolon *et al.* (2020) observed that, as the anthocyanin content of fruits increases, the luminosity decreases, making the color darker. However, the luminosity of fruits harvested in RS3 did not change during storage, with an average of 20.43. This shows that the refrigerated storage of blackberries harvested at this ripeness stage (RS3) at a temperature of 4 °C and 95% relative humidity, made it possible to preserve the visual appearance of the fruits, especially in relation to the brightness of the skin, even after 12 days of storage.

As for the color of the epidermis, the fruits harvested in RS3 presented the highest values for °Hue, differing significantly from the others, in all analyzed refrigerated

storage periods (Table 3). According to Samaniego *et al.* (2020), as the ripening process of blackberries progresses, the color tends to turn purplish-red, due to the accumulation of anthocyanins in the fruits. However, RS1 and RS2 only differed from each other after 9 and 12 days of storage, where fruits harvested with 50% black and 50% red epidermis (RS2) had higher °Hue than those harvested with 100%

red epidermis (RS1). Samaniego *et al.* (2020) verified significant increases in the skin color (°Hue), as well as in the levels of total anthocyanins of blackberries from the Brazos cultivar, as the maturation progressed. In the cited work, the contents were 0.67 mg cy-3-glu.g⁻¹, 1.21 mg cy-3-glu.g⁻¹ and 8.63 mg cy-3-glu.g⁻¹ in fruits considered green, semi-ripe and ripe, respectively.

Table 3: Color parameters of blackberries harvested at different ripeness stages and stored in a refrigerated environment for 12 days

| Luminosity (L) | | | | | | | |
|------------------------|----------------|---------|---------|----------|---------|--------|-----------|
| Ripeness | Storage (days) | | | | | Linear | Quadratic |
| stage | 0 | 3 | 6 | 9 | 12 | | |
| RS1 | 32.72 a | 28.87 a | 27.14 a | 24.27 a | 23.68 a | *(1) | ns |
| RS2 | 24.15 b | 23.06 b | 22.37 b | 20.43 b | 21.31 b | *(2) | ns |
| RS3 | 20.87 c | 21.43 b | 20.49 c | 19.77 b | 19.63 b | ns | ns |
| p (ripeness x storage) | | | | 0.0001** | | | |
| CV (%) | | | | 4.53 | | | |
| Color (°Hue) | | | | | | | |
| Ripeness | Storage (days) | | | | | Linear | Quadratic |
| stage | 0 | 3 | 6 | 9 | 12 | | |
| RS1 | 32.23 b | 31.05 b | 28.88 b | 28.69 c | 25.07 c | *(3) | ns |
| RS2 | 28.06b | 29.93b | 30.37 b | 40.65 b | 35.05 b | *(4) | ns |
| RS3 | 41.94 a | 42.26 a | 43.42 a | 47.09 a | 46.84 a | *(5) | ns |
| p (ripeness x storage) | | | | 0.0001** | | | |
| CV (%) | | | | 7.33 | | | |
| Chroma | | | | | | | |
| Ripeness | Storage (days) | | | | | Linear | Quadratic |
| stage | 0 | 3 | 6 | 9 | 12 | | |
| RS1 | 35.66 a | 29.53 a | 25.21 a | 22.63 a | 20.19 a | *(6) | ns |
| RS2 | 14.36 b | 8.10 b | 8.71 b | 5.35 b | 5.37 b | *(7) | ns |
| RS3 | 4.42 c | 4.29 b | 4.82 b | 4.61 b | 4.39 b | ns | ns |
| p (ripeness x storage) | | | | 0.0001** | | | |
| CV (%) | | | | 19.13 | | | |

Means followed by the same letter in the columns do not differ significantly from each other, by the Tukey test at 5% significance. ns not significant for regression analysis; *significant to 5% probability; **significant to 1% probability; (1) $y = -0.7556x + 31.872$ ($R^2 = 0.95$); (2) $y = -0.2744x + 23.903$ ($R^2 = 0.81$); (3) $y = -0.5558x + 32.518$ ($R^2 = 0.93$); (4) $y = 0.8232x + 27.872$ ($R^2 = 0.59$); (5) $y = 0.488x + 41.383$ ($R^2 = 0.87$); (6) $y = -1.2606x + 34.21$ ($R^2 = 0.96$); (7) $y = -0.6914x + 12.528$ ($R^2 = 0.79$); CV: Coefficient of variation; RS1: fruits with 100% red skin; RS2: 50% red and 50% black skin; RS3: 100% black skin.

Analyzing the changes in the color parameter (°Hue) during refrigerated storage, it was found that the fruits harvested in RS3 and RS2 showed increasing and those in RS1 decreasing linear responses (Table 3). According to Oliveira *et al.* (2014), the reduction of °Hue together with the reduction of titratable acidity, suggest increases in anthocyanin contents. Horvitz *et al.* (2017) observed that the content of total anthocyanins increased significantly during

refrigerated storage when storing blackberries harvested at different maturity stages, regardless of the fruit ripeness stage. In fruits harvested in RS1, there was a reduction in the titratable acidity (Table 1), as well as in the °Hue (Table 3), making the skin of the fruits more purplish red (Figure 1), which is probably associated with the increase in the anthocyanin content of these fruits.

However, despite changes in the color of the blackberry

skin at different ripeness stages, those harvested in RS1 and RS2 did not show values greater than or equal to those harvested with 100% black skin (RS3) even after 12 days of refrigerated storage. Quantification of epidermal color is very important because consumer acceptance is generally based on the black color of blackberries, which is used to draw conclusions about their ripeness and freshness (Krüger *et al.*, 2011; Hirsch *et al.*, 2012).

Chromaticity indicates the color saturation, the higher the chroma, the greater the saturation of the colors perceptible to humans. Pure colors have high saturation, while neutral colors have low saturation, so they are less bright to human perception (Pathare *et al.*, 2013). Fruits harvested in RS1, 100% red skin, had the highest chroma values, differing significantly from the others, regardless of the storage period analyzed (Table 3). This indicates high saturation and are thus brighter than those collected in RS2 and RS3, according to human perception. Samaniego *et al.* (2020) observed this same tendency, where chroma values decrease with the advancement of maturation of blackberries of the Brazos cultivar.

Regarding refrigerated storage, fruits harvested in RS1 and RS2 show a reduction in chroma values over time, with decreasing linear adjustment (Table 3). Mikulic-Petkovsek *et al.* (2017) found that unripe blackberry fruits were slightly red and with advancing maturation, they became dark purple, with a decrease in saturation and, consequently, a reduction in chroma values. However, there was no variation in relation to the chroma of fruits harvested in RS3 during refrigerated storage, with an average of 4.51. Pérez-Gallardo *et al.* (2015) did not observe changes either, in relation to the chroma of blackberries harvested with 100% black skin during refrigerated storage, even after 16 days of refrigerated storage at 4 °C. Showing that, in general, blackberries harvested at a ripeness stage considered mature (RS3), practically do not present changes in relation to the skin color parameters, as they have already reached their complete maturation (Table 3). However, it is worth emphasizing that, through the refrigerated storage of the fruits at 4 ± 0.5 °C and RH of 90-95%, it was possible to preserve the quality of the fruits in relation to the colorimetric parameters.

CONCLUSIONS

The ripeness stage at which the 'BRS Caingua' blackberries are harvested influences the physicochemical quality and postharvest conservation.

Fruits harvested with 100% black skin (RS3) have the best quality and can be stored in a refrigerated environment for up to 12 days, without significant loss of quality or making them unsuitable for consumption.

The storage of fruits harvested with 100% red or 50% red and 50% black (RS1 and RS2) for 12 days was not enough for them to develop physicochemical characteristics close to or equal to those presented by fruits harvested with the epidermis 100% black, mainly in relation to parameters related to flavor. Therefore, harvesting of 'BRS Caingua' blackberries should not be carried out when the skin is 100% red or 50% red and 50% black (RS1 and RS2).

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