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ORIGINAL ARTICLE

Use of refrigeration in the conservation of black raspberries *Rubus niveus*

Uso da refrigeração na conservação de framboesas negras Rubus niveus

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ABSTRACT: Raspberries have limited durability after harvest due to their high respiratory rate. The use of low temperatures is one of the techniques used to extend this short period. While cooling is a low-cost method, knowledge of appropriate storage conditions is essential to ensure the quality of the raspberries. To add to this knowledge base, was evaluated black raspberries, *Rubus niveus,* packed in polyethylene terephthalate trays and stored for 12 days in BOD incubators (biochemical oxygen demand) at –1, 1; 3, 5 and 7 °C, with variations from 0.2 to 0.5 °C, and a relative humidity of 91.4±7.3%. Although the evaluations were extended until the 12th day of storage, the fruits showed different retention periods depending on the temperature. The best temperature for the conservation of fruits was –1 °C, at which the berries reached 12 days of storage without much variation in quality for consumption. On the other hand, when stored at temperatures of 1 and 3 °C, preservation of the berries lasted only three days. After this period, the organoleptic quality of the fruits was reduced due to high mass loss and the development of an unusual taste. These features were anticipated at 5 and 7 °C, and the presence of strange odours and microorganisms were found at 7 °C, all of which resulted in a short period of storage.

RESUMO: *Framboesas apresentam limitada durabilidade após a colheita devido à sua alta taxa respiratória. O uso de baixas temperaturas constitui uma das técnicas utilizadas para estender este curto período. Embora a refrigeração seja um método de baixo custo, o conhecimento das condições adequadas é essencial para manter a qualidade das framboesas. Para ampliar o conhecimento acerca deste assunto, framboesas negras, Rubus niveus, embaladas em bandejas de tereftalato de polietileno, foram armazenadas por 12 dias em câmaras do tipo BOD (demanda bioquímica de oxigênio) a –1, 1, 3, 5 e 7 °C, com variações de 0,2 a 0,5 °C, e umidade relativa de 91,4±7,3%. Embora as avaliações tenham sido realizadas somente até o 12.º dia de armazenamento, os frutos apresentaram períodos de conservação diferentes, dependendo da temperatura. A melhor temperatura para a conservação dos frutos foi –1 °C, em que as bagas permaneceram por 12 dias com pouca variação na qualidade para o consumo. Por outro lado, quando as framboesas negras foram armazenadas a temperaturas de 1 e 3 °C, a vida útil das bagas foi de apenas três dias. Após este período, a qualidade organoléptica dos frutos foi reduzida devido à elevada perda de massa e ao desenvolvimento de um sabor estranho. Estas características foram antecipadas a 5 e 7 °C, quando foi verificada a presença de odores estranhos e micro-organismos, os quais resultaram num curto período de armazenamento.*

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1 Introduction

The red raspberries, black raspberries and blackberries (*Rubus idaeus*) has been faming on south Minas Gerais States, Brazil. This area has a subtropical climate and is producing this fruit in crates quantities (CAMPAGNOLO; PIO, 2012a; MOURA et al., 2012; SILVA et al., 2012).

Red raspberries are greatly enjoyed for their flavour and aroma. In addition to these attractive characteristics, they have a high anti-oxidative capacity, thus providing significant benefits to health (WANG; LIN, 2000). These fruits also constitute a source of numerous photochemicals, such as ellagic acid, anthocyanins and phenolic compounds (BEEKWILDER et al., 2005).

Raspberries present limited useful life after harvest as they are highly perishable fruits (HAFFNER et al., 2002). This limitation is due to their high respiratory rate, firmness loss and deterioration by fungus such as *Botrytis cinerea*, *Rhizopus stolonifer* and *Cladosporium Herbarum* (ANTONIOLLI et al., 2001). To overcome these disadvantages, the storage of fruits at low temperatures is important for ensuring product quality until the fruits reach the final consumer.

Studying the effect of different temperatures on the storage of red raspberries, Kalt et al. (1999) found that temperatures above 10 °C significantly affect the raspberries' metabolism, thereby altering the content of the phenolic compounds and the integrity of the fruit. The reduction in the quality of raspberries when not storing them at low temperatures was also confirmed by Nunes, Emond and Brecht (2003), who found a marked darkening of colour, a decrease in the firmness and an off-flavour in raspberries stored at temperatures of 10, 15 and 20 °C. Further, Antunes, Duarte Filho and Souza (2003) and Campagnolo and Pio (2012b, c) suggest that blackberries, which are perishable fruits with a high respiratory rate and also belong to the genus *Rubus*, as raspberries, stored up to nine days at 2 °C. Another raspberry species, the black raspberry (*Rubus niveus*), also known as the Mysore black raspberry, possesses attractive fruits and can be grown commercially. Nevertheless, its post-harvest conservation is unknown, which can be a drawback in the marketing of fresh fruits.

The purpose of the present work is to study the use of refrigeration in the post-harvest conservation of black raspberries.

2 Materials and Methods

Black raspberries of the species *Rubus niveus* were collected when ripe, that is, when they presented a wholly black surface. The harvest occurred in May of 2011, soon after daybreak, when the environmental temperatures were relatively low. The plants used for this study were localised in the experimental orchard in the Agriculture Department of the Federal University of Lavras (Universidade Federal de Lavras), at Lavras – MG, Brazil. The fruits were of average mass of 1.01±0.47 g and were free of mechanical or physiological injuries.

In the harvest operation, the fruits were carefully placed directly on transparent terephthalate polyethylene trays with a 125 g capacity and dimensions of 11 cm in length × 10.5 cm in width × 4.5 cm in height. The lids had small circular holes to make gas circulation easier, and a moisture–absorbing paper was placed inside the trays. The trays were then stored for 12 days in BOD incubators (biochemical oxygen demand) at temperatures of –1, 1; 3, 5 and 7 °C, with variations of 0.2 to 0.5 °C and a relative humidity of 91.4±7.3%. At 3-day intervals, fruits were removed from the incubators and analysed.

The experiment was conducted in split plots with a plot for each of the five temperatures for storage and a subplot for each of the five sampling periods (three, six, nine and twelve days). The control consisted of the fruits stored immediately after harvest. Thus, the experimental unit was made up of a tray with approximately 125 g of fruits. The experimental design was completely randomised with four replicates. The data were analysed by means of the variance and regression analyses. The models adjusted by means of regression were chosen on the basis of the significance of the regression coefficient at the level of 1% probability using Student's "t" test for the determination coefficient to explain the biological phenomenon. The data were analysed with the aid of the SISVAR 5.3 *software* (FERREIRA, 2011).

The following analyses were performed in each sampling period. In the fresh mass loss *(FML)* analysis, the fruits were weighed on an electronic scale, accurate within 0.01 g, at the beginning of the storage and during each sampling period. The results were expressed as percentages based on the difference between the initial mass of the fruit and the mass obtained in each sampling period. The colorimetric analysis assessed the external colour of the fruits as determined using a colorimeter Minolta CR-400 operating in the CIE mode. The mean of the measures performed on ten fruits was utilised for each replicate. The measurements were performed on the on the median part and on opposite sides of the same fruit during the storage period. The total titratable acidity (*TTA*) was determined by titrating 5 g of pulp with 0.01 N NaOH until pH 8.1 was reached, and the TTA was expressed as mg of citric acid per 100 g of pulp. The total soluble solids content (*TSSC*) used a sample of fruits ground in a politron per experimental unit, and two readings per sample were performed. The TSSC content was determined with the aid of a digital handheld refractometer, the Atago N1 model, with a reading in the range of 0 to 32 °Brix. For the ascorbic acid analysis, 2 g of the sample were added to 20 mL of oxalic acid and approximately 0.1 g of Kieselguhr with stirring for 15 min in a horizontal shaker and then filtered onto Whatman grade 40 paper. The ascorbic acid content was determined using the colorimetric method with 2,4-dinitrophenilhydrazine. The results were expressed in mg of ascorbic acid per 100 g of pulp. Anthocyanins were determined using the pH differential method according to the Association of Official Analytical Chemists - AOAC (2005), and the results were expressed as mg of cyanidin-3-glucoside per 100 g of pulp.

3 Results and Discussion

The variable FML presented an increasing linear behaviour in terms of the evaluation days at all temperatures (Figure 1).

The highest FML values, 37 and 33.98%, were found at 5 and 7 °C, respectively, in the last evaluation. The temperature of –1 °C provided low loss of fresh mass during the period

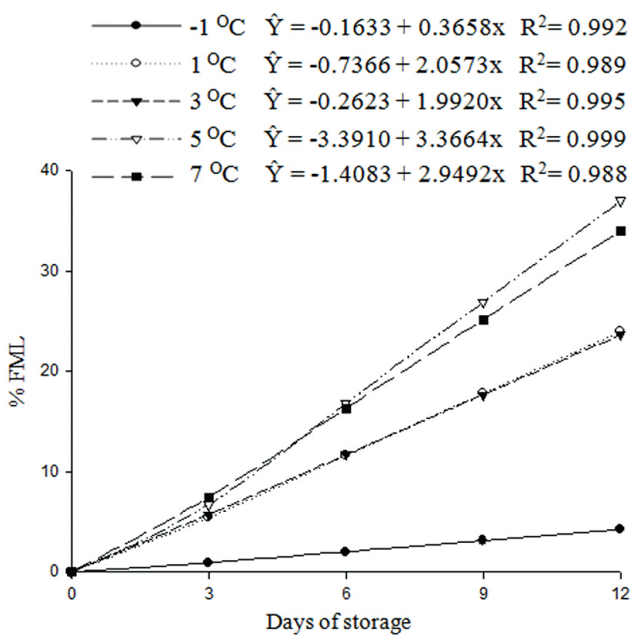

Figure 1. Estimated of fresh mass loss (FML) in black raspberries stored in different temperatures for 12 days.

evaluated with an average FML value of 2.03%. Thus, the black raspberry is more perishable with respect to mass loss at temperatures above 1 °C. Antunes, Duarte Filho and Souza (2003) found only a 14.83% mass loss in blackberries conserved for 12 days at 20 °C and a 7.91% loss when conserved at 2 °C.

The fruit's acceptance by consumers and its useful post-harvest life are drastically reduced with excess water loss and the consequent reduction of the total mass combined with wilting (PALIYATH; MURR, 2008). It is believed that, for most fresh horticultural products, the mass loss limit lies in the range of 5 to 10%. Accordingly, it is important to stress that the lowest temperature utilised in this experiment enabled a mass loss within the acceptable limit for the 12-day period. However, considering the maximum mass loss limit of 10%, this period was reduced to 5.2, 5, 4 and 3.9 days at the temperatures of 1, 3, 5 and 7 °C, respectively.

In the colorimetric analysis, a slight decrease in the luminosity values (L*) was found as storage time increased (Figure 2a).

The average value of L* was initially 36.82. This value decreased to 35.40 on the day 12 of storage. Until the 6th day of storage, similar luminosity values were found among the temperatures studied. On the 9th day of storage, the highest temperatures (5 and 7 °C) showed a significant reduction in the values of L*. These values continued to decrease until the last day of evaluation for all temperatures, with the exception of –1 °C. Based on the chroma values, a reduction in the intensity of colour throughout the storage period was verified (Figure 3). The initial and final average chroma values were 6.73 and 3.99, respectively. These values indicated that the raspberries presented neutral colours (greyish), with few differences found until the end of the experiment.

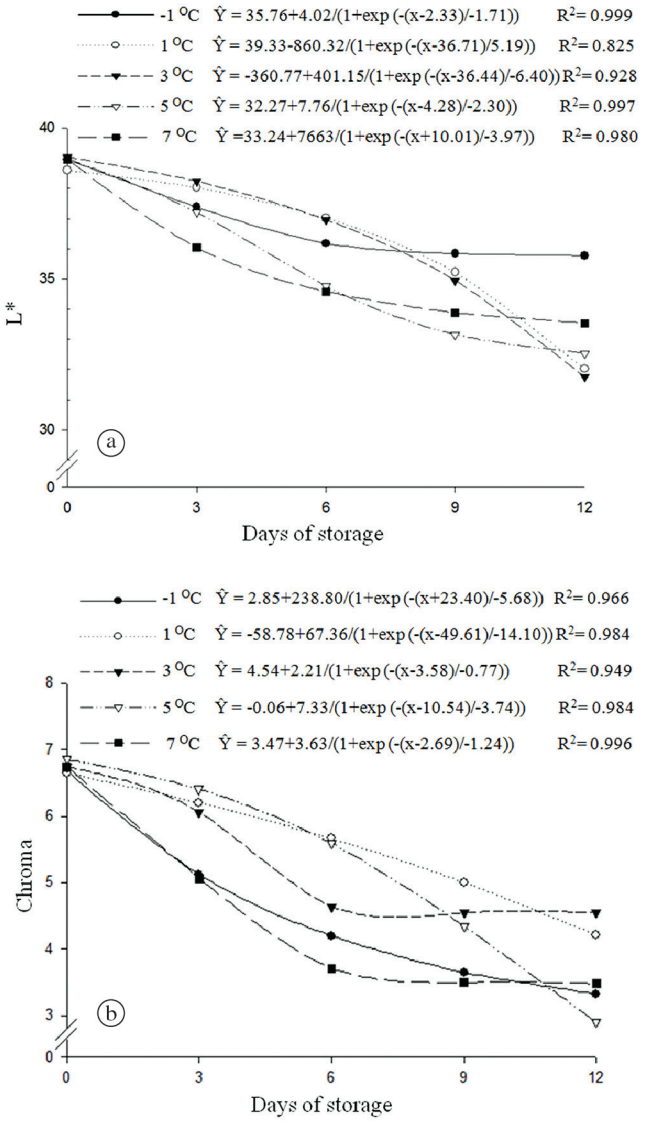

Figure 2. Estimated of luminosity (L*) (a) and chroma (b) in black raspberries stored in different temperatures for 12 days.

Inexpressive variations in the colour coordinates were also detected in blueberry conservation, another small fruit (SPIERS et al., 2004). The authors found L* values of 31.0 at the beginning and 28.5 at the end of the conservation period. The chroma values were situated between 4.23 and 5.46 at the start and at the end of this period, respectively.

The slight alterations found in the colouration of black raspberries can be ascribed to the fact they are non-climacteric fruits and are characterised as presenting few sharp transformations in post-harvest ripening. At harvest, black raspberries appear as small black berries with shades of colour ranging from light to dark. The presence of wax produces the effect responsible for the characteristic colour of the fruits.

During storage, the shades of the black raspberries assumed growing values (Figure 4). This increase in hue was slower at temperatures of –1 and 3 °C, while those stored at higher temperatures experienced a rapid increase in hue from the sixth

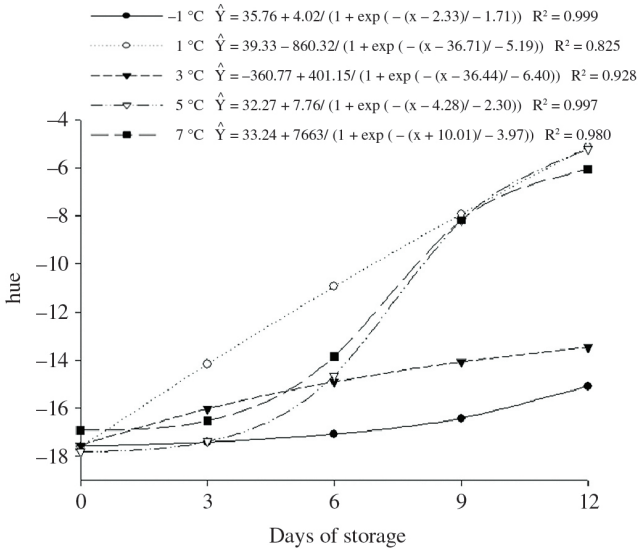

Figure 3. Estimated of hue in black raspberries stored in different temperatures for 12 days.

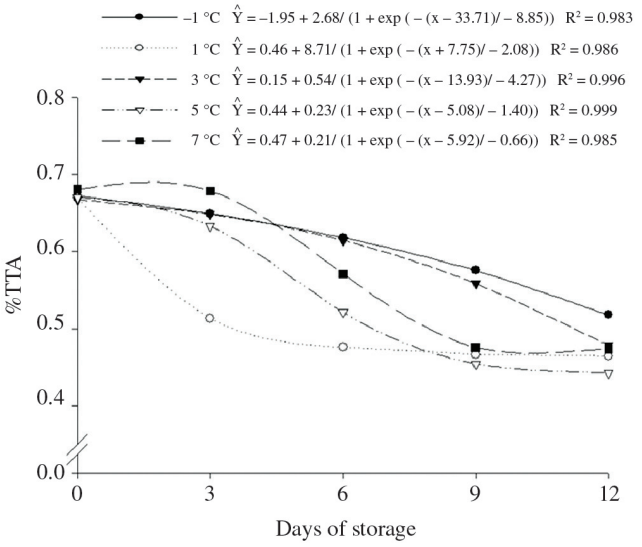

Figure 4. Estimated of total titratable acidity (TTA) in black raspberries stored in different temperatures for 12 days.

day through the final day. The berries presented colouration rangingpassing from bluish tones to slightly reddish. The berries stored at the lowest temperature used in this study suffered the least expressive changes in shades.

The change in the shade of raspberries stored under refrigerated conditions was also found by Haffner et al. (2002). In studying the colouration of five different red raspberry cultivars subjected to a temperature of 1.7 °C, the authors found that the fruits acquired darker red tones.

In every treatment, a decrease in TTA was observed as storage time increased (Figure 4).

The content of citric acid in black raspberries stored at different temperatures decreased by approximately 5% compared to the initial content. Fruits stored at 1 °C first showed a rapid decrease in citric acid content; however, after the initial reduction, these fruits maintained a relatively stable level of TTA. At the other temperatures, the decrease in TTA was more gradual. On the last day of storage, there were no significant differences in citric acid content among the temperatures, with concentrations of approximately 0.47 mg of citric acid/100 g.

The reduction in the citric acid contents may be associated to an increased consumption of organic acids due to the respiratory process. Kays (1991) states that after harvest and during storage, the concentration of organic acids tends to decline in most fruits, as the compounds are used as a respiratory substrate and carbon skeleton for the synthesis of new compounds.

Reports by Haffner et al. (2002) confirmed the decrease in the citric acid contents in raspberries during storage. The authors observed a reduction of 9.05% of the total acidity on the 7th day of storage. Nevertheless, Krüger et al. (2011), who studied the storage of raspberries under refrigerated conditions (2 to 4 °C), found that the TTA is not affected by storage.

The TSSC gradually increased over time and reached a maximum value on the last day of storage (Figure 5).

Until the 9th day of storage, the highest TSSC values were found at a temperature of 5 °C. After day 9, the maximum value was found at 3 °C. Figure 6 shows that a storage temperature of –1 °C was not effective in checking the evolution of TSSC presenting average values on the 12th day of 13.5 °Brix, a value statistically similar to that of the highest temperature.

It was found that 7 °C was the most effective and efficient temperature for analysing the evolution of the TSSCs due to the attack by pathogens in this treatment as early as the third day of storage. The stress caused by the physical damage, including tissue wounding, causes an increase in respiration and attack by microorganisms, which reduces the quality of the fruit to a great extent (CHITARRA; CHITARRA, 2005). The energy demand of the pathogens was satisfied by TSSC consumption and culminated in TSSC values lower than those found in the other treatments.

The evolution in the TSSC contents during storage is a common occurrence in climacteric fruits, which gives continuity to the ripening process even after harvest. This ripening process after harvest is not found in non-climacteric fruits such as the raspberry. Therefore, the increase in the TSSC contents can be ascribed to a merely physical phenomenon, i.e., water loss, which is confirmed by a loss of mass and results in an increased solute concentration.

Krüger et al. (2011) showed that the TSSC content of red raspberries under refrigeration at 2 to 4 °C for three days followed by one day at 20 °C slightly increased, but this increase was not statistically significant.

A decrease in the level of ascorbic acid was found during storage and reached its lowest level of 48.73 mg of ascorbic acid/100 g of pulp at the end of the storage period (Figure 6).

The ascorbic acid from the start of storage through the end of the evaluation period. Reductions in ascorbic acid content of 5 to 10% were also found by Callesen and Hølm (1989) in

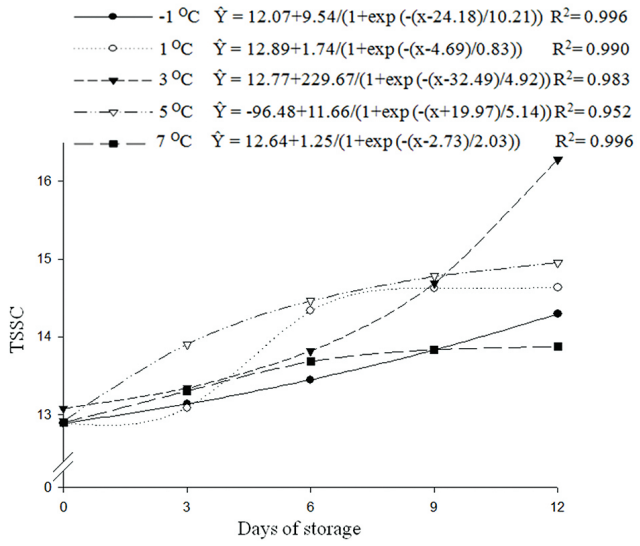

Figure 5. Estimated of total soluble solids content (TSSC) in black raspberries stored in different temperatures for 12 days.

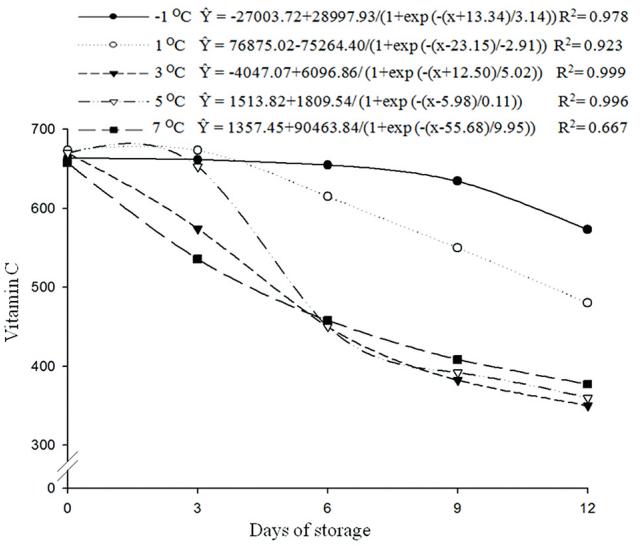

Figure 6. Estimated of vitamin c in black raspberries stored in different temperatures for 12 days.

raspberries subjected to storage 5 °C at different controlled atmospheric conditions for 11 days.

The use of low temperatures after harvest is of great importance for the maintenance of ascorbic acid in fruits. Kalt et al. (1999) verified that the use of temperatures close to zero promoted insignificant losses of ascorbic acid content for raspberries, whereas marked losses were found with increasing temperatures.

Nevertheless, even following a substantial reduction, the ascorbic acid content in black raspberries, as observed in this work, remained high. This finding suggests that these fruits have functional properties that are increasingly desired by a population seeking fruits rich in bioactive compounds that offer a number of health benefits.

The level of anthocyanins present in the black raspberries increased throughout the storage period (Figure 7).

This increase was even greater in the fruits stored at a temperature of 5 °C. Until the 9th day of storage, the anthocyanin content remained similar for fruits stored at –1, 1, 3 and 7 °C. However, after the 9th day, there were clear distinctions in the levels of anthocyanins among these storage temperatures.

The level of anthocyanin in raspberries is strongly affected by the storage conditions (KRÜGER et al., 2011). In a study of raspberry conservation for berries stored at 2 to 4 °C, the anthocyanin contents increased by 25.6% during three days of refrigerated storage followed by one day of storage at room temperature. The increase in anthocyanins in small fruits such as the raspberry and the strawberry during post-harvest was also reported by Kalt et al. (1999).

Kalt et al. (1999) ascribe the increase in anthocyanin content to a decrease in organic acids, which provide carbon skeletons for phenolic synthesis. In this work, in addition to the decrease in total titratable acidity, which provides carbon for the synthesis of new compounds, the increase in anthocyanin contents might be a consequence of increased anthocyanin concentration due to the high loss of fresh mass.

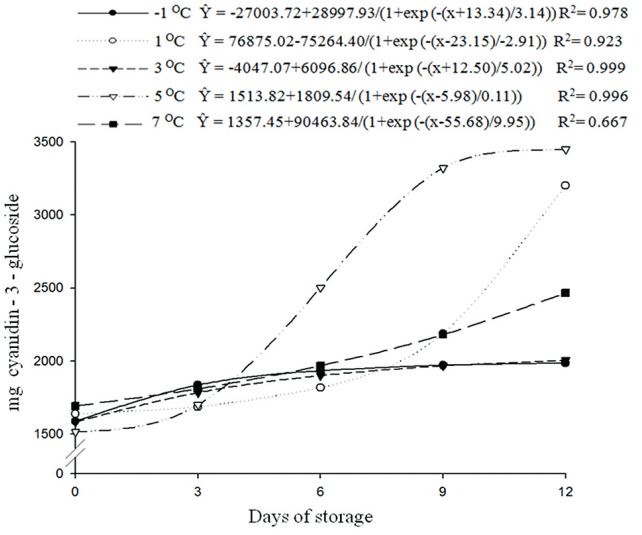

Figure 7. Estimated of anthocyanins in black raspberries stored in different temperatures for 12 days.

4 Conclusions

The fruits demonstrated different conservation properties over the 12-day period as a result of variations in temperature. Black raspberries stored at –1 °C showed good conservation for 12 days, while fruits subjected to temperatures of 1 and 3 °C were fit for consumption until day 3. After this period, the organoleptic quality of the fruits was reduced due to the loss of moisture and the development of an off-flavour. These characteristics, anticipated at 5 and 7 °C, were accompanied by the presence of microorganisms, which was verified at 7 °C.

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