



Açaí extract powder as natural antioxidant on pork patties during the refrigerated storage

Elisa Rafaela Bonadio Bellucci^a, João Marcos dos Santos^a, Larissa Tátero Carvalho^a,
Taís Fernanda Borgonovi^a, José M. Lorenzo^{b,c}, Andrea Carla da Silva-Barretto^{a,*}

^a Department of Food Technology and Engineering, UNESP – São Paulo State University, Street Cristóvão Colombo, 2265, Zip Code 15054-000 São José do Rio Preto, SP, Brazil

^b Centro Tecnológico de la Carne de Galicia, Avda. Galicia nº 4, Parque Tecnológico de Galicia, San Cibrao das Viñas, 32900 Ourense, Spain

^c Área de Tecnología de los Alimentos, Facultad de Ciencias de Ourense, Universidad de Vigo, 32004 Ourense, Spain

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ABSTRACT

The current trends among consumers are pushing for the use of natural antioxidants options. Açaí fruit is rich on polyphenolic components but no studies have been carried out to evaluate their effect in meat products. The objective was to investigate the effect of açaí extract on refrigerated pork patties quality. Five treatments were done: without antioxidant (CON), Sodium Erythorbate 500 mg.kg⁻¹ (ERY), Açaí Extract: 250 (AEL), 500 (AEM), 750 mg.kg⁻¹ (AEH). Açaí extract did not affect the proximate composition, pH and cooking parameters. The concentrations of açaí extract studied increased antioxidant activity and reduced lipid oxidation (0.379, 0.293, and 0.217 vs. 0.889 mg MDA.kg⁻¹ for AEL, AEM, AEH vs. CON, respectively). However, only the AEL treatment did not affect the color parameters, showing the best option for the application on pork patties. Thus, açaí extract at 250 mg.kg⁻¹ can be used as a natural antioxidant replacing sodium erythorbate to preserve the quality of refrigerated pork patties.

1. Introduction

The widely and massive consumption of meat products associated with the increasing world population and concerns about the impact on food components have generated important challenges for the food industry, especially in meat sector to produce healthier meat products (Barone et al., 2021). According to Teixeira and Rodrigues (2021), consumer perceptions in recognizing healthier meat products involve factors like physical and chemical composition, nutritional quality, sensory properties and new ingredients. In addition, consumers are adept at plant-based ingredients and recognize them as more natural (Barone et al., 2021).

The development of healthier meat products is a complex task that involves also the preservation of quality of the meat product. One of the main factors that influencing the quality of meat and meat products is oxidative deterioration (Munekata et al., 2020). Oxidative reactions cause discoloration and affect the flavor and aroma, which lead to consumer rejection. Other important effects are the production of potentially toxic compounds and reduction of nutritional quality

(Amaral, da Silva, & da Lannes, 2018; Lorenzo, Trindade, Ahn, & Barba, 2019). The nutritional quality of meat products is a relevant factor, which can be reduced due to undesirable reactions that occur with lipid oxidation such as losses in vitamins, essential fatty acids and essential amino acids (Domínguez et al., 2019). In meat products like patties, the lipid oxidation can increase because the meat grinding process leads to the rupture of the membrane exposing phospholipids to oxygen, accelerating the development of oxidative rancidity, since one of the main factors for the development of lipid oxidation is the exposure to oxygen (Zamuz et al., 2018). The products of lipid oxidation favor the oxidation of heme-proteins, increasing their pro-oxidant activity, and also induce the oxidation of myoglobin, which alters its color from bright cherry-red color to brownish tones (Tomasevic, Djekic, Font-i-Furnols, Terjung, & Lorenzo, 2021).

Several studies in recent years showed the protective effect of plant-based antioxidants in meat products (Alirezalu et al., 2020). In this regard, Turgut, Işıkçı, and Soyer (2017) evaluated the antioxidant effect of pomegranate peel extract in relation to the oxidation of lipids and proteins in beef meatballs during the frozen storage ($-18 \pm 1^\circ\text{C}$) and

* Corresponding author.

E-mail address: andrea.carla@unesp.br (A.C. Silva-Barretto).

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concluded that the addition of 0.5 and 1% of pomegranate peel extract in meatballs decreased the oxidation of lipids and proteins and increased the sensory scores, demonstrating that the extract delayed the lipid and protein oxidations. In addition, dos Reis et al. (2017) investigated the microencapsulated extract of propolis co-product on the stability of burgers during storage at -15°C and confirmed its efficiency against the lipid oxidation. Moreover, Ferreira et al. (2017) incorporated the extracts from acorns in chicken patties and found that the extract reduced the deterioration of color and texture and delayed the lipid and protein oxidation in patties over the refrigerated storage.

Munekata et al. (2020) noticed that the main bioactive components of plants that can be used in the meat industry are polyphenols (such as anthocyanins, tannins and flavonols) and essential oils (mainly composed of terpenes). Also, the antioxidant and antimicrobial mechanisms exerted by phenolic compounds justify their use as additives in the preservation of meat products (Munekata et al., 2021). In this context, Açai (*Euterpe oleracea*), a native fruit from Amazon region, is widely known for its bioactive compounds (Carvalho, Silveira, Mattietto, de Oliveira, & Godoy, 2017), for its high antioxidant activity and for the great potential as a functional food or ingredient (Garzón, Narváez-Cuenca, Vincken, & Gruppen, 2017; Kang et al., 2010; Silva et al., 2019). The major polyphenolic components in açai pulp include anthocyanins, proanthocyanidins, other flavonoids and lignans (Carvalho et al., 2017; Kang et al., 2011).

Several studies have investigated the antioxidant properties of açai pulp (Garzón et al., 2017; Hogan et al., 2010; Kang et al., 2011; Paz et al., 2015) and its by-products (Brunschwig et al., 2016; Martins et al., 2020; Melo et al., 2021), which support the further investigation of açai as potential natural antioxidant for its application in food products. Although, these studies provide important evidences to explore the natural antioxidants from açai, the effect in a complex system involving components susceptible to oxidative degradation (such as lipids and myoglobin in meat products) remains poorly explored. Therefore, the objective of this work was to obtain an extract from the açai pulp, characterize its antioxidant properties and evaluate the addition of açai extract in pork patties on the physico-chemical properties and oxidative and color stability during the refrigerated storage.

2. Materials and methods

2.1. Preparation of Açai extract (AE) powder and determination of antioxidant activity

2.1.1. Preparation of AE

Açai pulp was obtained from a producer located in São José do Rio Preto (São Paulo, Brazil) on the day of pulp extraction from the ripe açai, showing deep purple color. The pulp was taken to the Department of Food Engineering and Science (UNESP, São José do Rio Preto - São Paulo, Brazil) where the extraction and experiment with the açai extract was carried out. For the extraction, water was added to the pulp in the proportion of 1:1 and then was agitated for 30 min at 150 rpm (under protection from the light) and centrifuged (4500 rpm for 20 min). The supernatant was filtered, frozen and freeze-dried. The resultant powder of açai extract was used to total phenolic compound and antioxidant activity analyses and added to pork patties at three different levels.

2.1.2. Total phenolic compounds (TPC) and antioxidant activity of AE

The açai extract powder was mixed in diluted methanol (1:3 in water) and the total phenolic compound test was carried out following the method described by Singleton, Orthofer, and Lamuela-Raventós (1999) with some modifications. The appropriate dilution of AE with water was made in test tubes (0.5 mL) and mixed with 2.5 mL of diluted reactive Folin-Ciocalteu (1:10 in water). After 2 min, 7.5% Na_2CO_3 solution (2 mL) was added, and the test tubes were incubated at 50°C for 10 min and the absorbance (760 nm) of the solutions was measured. The standard curve was prepared with concentrations in the range of 0–100

mg of gallic acid/L and the TPC was expressed as mg of gallic acid equivalent (GAE)/g sample.

The DPPH assay test was carried out following the protocol described by Brand-Williams, Cuvelier, and Berset (1995), with some modifications. The 100 μL of AE were mixed with 3900 μL of DPPH solution (60 μM in methanol). After incubation for 10 min at 37°C , absorbance (515 nm) was measured. The standard curve was elaborated using Trolox (0–1.2 mM) and the results were expressed as mg Trolox equivalents (TE)/g sample.

2.2. Manufacture of pork patties

The experiment was performed in the pilot plant of the Department of Food Engineering and Technology, Institute of Biosciences, Letters and Exact Sciences - UNESP, São José do Rio Preto, São Paulo, Brazil. A total of 180 pork patties was elaborated: four pork patties for each treatment x five treatments (CON, ERY, AEL, AEM and AEH) x three sampling points (0, 5 and 10 days) x three different replication (on different days with different raw materials and the same ingredients). The treatments were: CON (without antioxidant), Sodium Erythorbate (ERY, 500 $\text{mg}\cdot\text{kg}^{-1}$), Açai Extract at three levels: low (AEL, 250 $\text{mg}\cdot\text{kg}^{-1}$), medium (AEM, 500 $\text{mg}\cdot\text{kg}^{-1}$), and high (AEH, 750 $\text{mg}\cdot\text{kg}^{-1}$).

All treatments were produced with: pork shoulder ground in 8-mm mincing plate (*Triceps brachii*) ($780\text{ g}\cdot\text{kg}^{-1}$), pork backfat ground in 8-mm mincing plate ($150\text{ g}\cdot\text{kg}^{-1}$), salt ($15\text{ g}\cdot\text{kg}^{-1}$) and textured soy protein ($10\text{ g}\cdot\text{kg}^{-1}$). The raw material meat was acquired in local market in the city of São José do Rio Preto, Brazil contemplating the Federal Inspection Service 48 h after slaughter. Water was added to the treatments in order to complete 1 kg. To produce the pork patties, the ingredients were homogenized manually for 10 min and the açai extract was dissolved in water before the addition. The meat mass was shaped in a manual burger former (0.1 m diameter and 0.01 m height). The samples were packed in nylon-polyethylene bags, sealed without vacuum, stored at $2 \pm 1^{\circ}\text{C}$ for 10 days in the dark and evaluated at 1, 5 and 10 days.

2.3. Proximate composition and pH

Proximate composition of pork patties was determined following the official method of the Association of Official Analytical Chemistry (AOAC, 2007) for moisture, protein and ash content. Fat content was determined following the method proposed by Bligh and Dyer (1959). The results were expressed in g/100 g. The pH values were measured directly on pork patties using a digital pH meter mPA210 (Tecnoport, São Paulo, Brazil) equipped with a penetration glasses electrode previously calibrated with two standard solutions (pH 4 and pH 7) at room temperature. In both proximate composition and pH assays, four determinations were made in each treatment.

2.4. Instrumental color parameters

Instrumental color parameters were measured on the surface of uncooked pork patties, avoiding areas with a lot of fat. The blooming time was 20 min before analysis, while the samples were exposed to air. ColorFlex45/0 colorimeter (Hunterlab, Reston, United States) was used and black glasses and white tiles were utilized for calibration. Samples were measured using illuminant with 0.75-in. aperture size and 10° viewing angle geometry. The CIELab system of color specification was used and the parameters obtained were L^* (lightness), a^* (redness) and b^* (yellowness). Chroma (Eq. 1) and hue angle (Eq. 2) were calculated using the parameters a^* and b^* . Three determinations were made in each sampling patty and four samples were used for each treatment.

$$C^* = \sqrt{(a^2 + b^2)} \quad (1)$$

$$h^{\circ} = \arctan\left(\frac{b^{*}}{a^{*}}\right) \quad (2)$$

2.5. Cooking proprieties

In order to measure the loss from cooking process, cooking loss test was carried out and raw pork patties from each treatment were weighed, placed on a baking sheet, covered with aluminum foil and roasted in an electric oven at 100 °C until the internal temperature reached 71 °C. Afterward, the samples were cooled until they reached room temperature and were weighed again. For shrinkage, the diameter was measured three times in each pork patty before and after cooking.

Cooking loss (CL) was calculated as follows:

$$CL (\%) = \frac{(Initial\ weight - Final\ weight)}{Initial\ weight} \times 100$$

The diameter reduction (DR) was calculated as follows:

$$DR (\%) = \frac{(Initial\ diameter - Final\ diameter)}{Initial\ diameter} \times 100$$

2.6. Lipid oxidation

Lipid oxidation (TBARS) was determined by reacting the oxidation products with thiobarbituric acid (TBA) to form compounds that can be measured spectrophotometrically at 532 nm, according to the method described by Vyncke (1975). Briefly, sample (one pork hamburger for each treatment) was completely homogenized, and 5 g were homogenized with 25 mL of 7.5% trichloroacetic acid solution for 2 min with an Ultra-Turrax (IKA®-Werke GmbH & Co. KG, Staufen, Germany). The homogenate was centrifuged (Rotina 420R, Hettich Zentrifugen, Germany) at 3500 rpm for 10 min. The supernatant was obtained after filtration and 5 mL reacted with 5 mL of 0.02 M TBA solution and heated in a water bath (96 °C for 40 min). Açai pigments are soluble in water, so they can be present in supernatant before the addition of thiobarbituric acid solution. Thus, TBARS determination were performed with minor modifications as described by Bellucci et al., 2021, Bellucci, Munekata, Pateiro, Lorenzo, and da Silva Barretto (2021) to avoid pink color interfere. Prommachart et al. (2020) modified this method when studied the black rice water extract in beef patties to avoid the interference of color. The 1,1,3,3 tetraethoxypropane (TEP) was used as standard and the results were expressed as mg of malonaldehyde (MDA)/kg of sample.

2.7. Radical scavenging activity assay (in vitro)

The pork patties extract was obtained as described by Prommachart et al. (2020), with modifications. Briefly, 3 g of grounded pork patty was homogenized with methanol (10 mL) using an IKA T18 digital ultraturax (IKA®-Werke GmbH & Co. KG, Staufen, Germany) (13,000 rpm) for 2 min and the homogenate was centrifuged at 10,000 xg at 4 °C for 10 min (Rotina 420R, Hettich Zentrifugen, Germany) to collect a clear supernatant. The DPPH (Radical scavenging activity assay) was determined as described by Bellucci, Munekata, et al. (2021), where the samples were analyzed using 100 µL of sample solution and 3.9 mL of 60 µM DPPH solution in methanol. The reaction was agitated in vortex and left for 10 min at 37 °C in a water bath. The inhibition percentage of absorbance (at 515 nm) was determined by the following equation:

$$\%Inhibition = \frac{Abs(blank) - Abs(sample)}{Abs(blank)} \times 100$$

The blank was made from the mixture of 100 µL of methanol and 3.9 mL of 60 µM DPPH solution in methanol.

2.8. Statistical analyses

Five treatments (CON, ERY, AEL, AEM, AEH) of pork patties were

Table 1

Proximate composition of pork patties (g/100 g) with sodium erythorbate and different levels of açai extract (AE) during refrigerated storage.

| Parameters | CON | ERY | AEL | AEM | AEH | SEM | Sig. |
|------------|-------|-------|-------|-------|-------|-------|------|
| Moisture | 63.82 | 61.83 | 66.16 | 64.83 | 66.15 | 0.828 | n.s. |
| Protein | 19.13 | 20.47 | 20.39 | 19.83 | 20.33 | 0.372 | n.s. |
| Fat | 11.94 | 12.66 | 10.69 | 11.02 | 10.57 | 0.324 | n.s. |
| Ash | 2.25 | 2.25 | 2.36 | 2.37 | 2.46 | 0.033 | n.s. |

Table 2

Evaluation of pH values of pork patties with sodium erythorbate and different levels of açai extract (AE) during refrigerated storage.

| Days | CON | ERY | AEL | AEM | AEH | SEM | Sig. |
|------|-------|-------|-------|-------|-------|-------|------|
| 1 | 5.74 | 5.75 | 5.88 | 5.73 | 5.78 | 0.029 | n.s. |
| 5 | 5.68 | 5.78 | 5.76 | 5.75 | 5.73 | 0.019 | n.s. |
| 10 | 5.69 | 5.70 | 5.77 | 5.69 | 5.75 | 0.031 | n.s. |
| SEM | 0.058 | 0.024 | 0.022 | 0.019 | 0.008 | | |
| Sig. | n.s. | n.s. | n.s. | n.s. | n.s. | | |

SEM: standard error of the mean; Sig.: Significance; n.s.: Not significant ($P < 0.05$). Treatments: CON: patties prepared without antioxidant; ERY: patties prepared with sodium erythorbate at 500 mg kg⁻¹; AEM: patties prepared with açai extract at 250 mg kg⁻¹; AEH: patties prepared with açai extract at 500 mg kg⁻¹; AEL: patties prepared with açai extract at 750 mg kg⁻¹.

manufactured in triplicate ($n = 3$), and analyzed during the refrigerated storage (at 1, 5 and 10 days). For the statistical analysis of the results, analysis of variance (ANOVA) was carried out using the General Linear Model (GLM). The time of storage and the treatment were considered to be fixed effects, while manufacturing repetition was a random effect. Statistical analyses were carried out using the software STATISTICA version 7 (StatSoft, Inc., 2004).

3. Results and discussion

3.1. Total phenolic compounds (TPC) and antioxidant activity of AE

According to Paz et al. (2015), TPC (determined by Folin-Ciocalteu method) does not measure the amounts of phenolic compounds, but instead measures the chemical reducing capacity of compounds present in the extract relative to gallic acid. So, the TPC found in this study for AE was of 612.54 ± 17.64 mg of GAE/g of extract. This is considerably higher than the value (6.07 ± 2.17 mg GAE/g fresh weight) previously reported by Garzón et al. (2017) in Colombian açai. Lower values also were observed by Paz et al. (2015), who noticed value of 1808 mg of GAE/100 g of açai extract and by Hogan et al. (2010) who studied an anthocyanin-rich extract from açai and reported value of 312 mg of GAE/g of extract. In this study, the level of TPC can be considered high, because it was higher than 2500 mg of GAE/100 g (Vasco, Ruales, & Kamal-Eldin, 2008).

The antioxidant activity, measured using DPPH assay, is a technique based on the reduction of the DPPH radical in the presence of a hydrogen-donating antioxidant. The DPPH values found in this study were of 47.57 ± 0.83 µmol TE/g of AE (or 1190 mg TE/100 g of AE). This result is similar to that found by Paz et al. (2015) (1574 ± 101 mg TE/100 g) and lower than those reported by Hogan et al. (2010) who noticed values of 1208 ± 130 µmoles TE/g of anthocyanin-rich extract from açai. Vasco et al. (2008) analyzed the antioxidant activity of major fruits from Ecuador and found values between 0.3 and 76 µmol TE/g sample (fresh weight) for DPPH assay.

3.2. Proximate composition and pH of pork patties

As expected, no differences were observed for the moisture, protein, fat and ash contents ($P > 0.05$) among the pork patties (Table 1). These results showed that neither açai extract nor sodium erythorbate

Table 3

Cooking loss and shrinkage of pork patties with sodium erythorbate and different levels of açai extract (AE) during refrigerated storage.

| Parameters | CON | ERY | AEL | AEM | AEH | SEM | Sig. |
|-------------------------|--------|--------|--------|--------|--------|-------|------|
| <i>Cooking loss (%)</i> | | | | | | | |
| 1 | 25.152 | 24.720 | 23.089 | 24.662 | 24.307 | 0.513 | n.s. |
| 5 | 25.276 | 24.910 | 22.992 | 23.873 | 23.805 | 0.695 | n.s. |
| 10 | 26.910 | 26.510 | 26.596 | 27.649 | 29.122 | 0.462 | n.s. |
| SEM | 0.738 | 0.896 | 0.679 | 0.735 | 0.907 | | |
| Sig. | n.s. | n.s. | n.s. | n.s. | n.s. | | |
| <i>Shrinkage (%)</i> | | | | | | | |
| 1 | 16.645 | 14.704 | 16.876 | 15.952 | 15.484 | 0.299 | n.s. |
| 5 | 15.158 | 14.755 | 16.385 | 16.160 | 15.759 | 0.364 | n.s. |
| 10 | 16.948 | 16.865 | 16.921 | 18.204 | 17.077 | 0.348 | n.s. |
| SEM | 0.388 | 0.534 | 0.353 | 0.446 | 0.436 | | |
| Sig. | n.s. | n.s. | n.s. | n.s. | n.s. | | |

SEM: standard error of the mean; Sig.: Significance; n.s.: Not significant ($P < 0.05$). Treatments: CON: patties prepared without antioxidant; ERY: patties prepared with sodium erythorbate at 500 mg kg⁻¹; AEM: patties prepared with açai extract at 250 mg kg⁻¹; AEH: patties prepared with açai extract at 500 mg kg⁻¹; AEL: patties prepared with açai extract at 750 mg kg⁻¹.

interfered in the proximate composition of the pork patties. These findings agree with data reported by Bellucci, Barretto, et al., 2021, Bellucci, Munekata, et al. (2021) who did not find significant differences in chemical composition of pork patties elaborated with the addition of a natural extract (from pitaya) and sodium erythorbate. In addition, de Carvalho et al. (2020) also observed that turmeric extract did not affect the proximate composition of fresh lamb sausages.

As for the proximate composition, the pH values (Table 2) did not display significant differences among treatments ($P > 0.05$), showing that the açai extract and sodium erythorbate did not affect the pH values of refrigerated pork patties. In addition, in this study, it was also observed that the pH values also did not differ between sampling times in all analyzed treatments ($P > 0.05$). Likewise, Prommachart et al. (2020) also found no effect on pH values when black rice water extract was added to beef patties.

In this study, the pH values ranged from 5.69 to 5.88 and were similar to those found by Pateiro et al. (2018) in pork patties with addition of guarana seeds extract (from 5.67 to 5.87), also during the

cold storage. In addition, the authors reported differences among treatments from the eleventh day. However, slightly lower pH values (from 5.45 to 5.70) were reported by Bellucci, Barretto, et al., 2021, Bellucci, Munekata, et al. (2021) in pork patties with pitaya extract until 18 days under refrigerated conditions, but this difference can be justified by the differences in packaging conditions and, also, can be inherent to the meat used.

3.3. Cooking parameters

The results of cooking loss and shrinkage tests of pork patties with sodium erythorbate and different concentrations of açai extract during the refrigerated storage are shown in Table 3. The cooking loss of pork patties varied between 22.99% and 29.12% and did not show significant differences ($P > 0.05$) among treatments and storage times. Cooking loss promotes the release of matrix fluids that carry water, water-soluble nutrients and compounds responsible for flavor and aroma, as well as color-forming pigments. Thus, the addition of sodium erythorbate and

Table 4

Color parameters of pork patties with erythorbate and different levels of açai extract (AE) during refrigerated storage.

| Parameters | Days | CON | ERY | AEL | AEM | AEH | SEM | Sig. |
|------------|------|----------------------|----------------------|-----------------------|----------------------|----------------------|-------|------|
| L* | 1 | 55.32 ^{Ba} | 54.83 ^{ab} | 52.34 ^{Bbc} | 49.98 ^{Bcd} | 47.42 ^{Bd} | 0.427 | * |
| | 5 | 57.18 ^{Ba} | 56.50 ^a | 53.86 ^{ABab} | 53.20 ^{Ab} | 50.56 ^{Ab} | 0.437 | * |
| | 10 | 61.58 ^{Aa} | 57.02 ^b | 55.99 ^{Abc} | 52.88 ^{ABc} | 49.95 ^{ABd} | 0.649 | * |
| | SEM | 0.637 | 0.476 | 0.402 | 0.430 | 0.468 | | |
| | Sig. | * | n.s. | * | * | * | | |
| a* | 1 | 9.74 ^{Aa} | 9.91 ^{Aa} | 9.96 ^{Aa} | 8.49 ^{Aab} | 7.15 ^{Ab} | 0.210 | * |
| | 5 | 7.83 ^{Ba} | 8.46 ^{Ba} | 7.98 ^{Ba} | 6.43 ^{Bb} | 6.04 ^{Bb} | 0.183 | * |
| | 10 | 5.24 ^{Cb} | 6.83 ^{Ca} | 6.74 ^{Bab} | 5.88 ^{Bab} | 5.86 ^{Bab} | 0.176 | * |
| | SEM | 0.365 | 0.280 | 0.283 | 0.230 | 0.183 | | |
| | Sig. | * | * | * | * | * | | |
| b* | 1 | 16.15 ^{Aa} | 16.16 ^{Aa} | 14.98 ^{Aa} | 12.33 ^b | 10.66 ^c | 0.243 | * |
| | 5 | 15.33 ^{ABa} | 15.14 ^{Ba} | 13.72 ^{ABb} | 11.90 ^c | 11.10 ^c | 0.196 | * |
| | 10 | 15.19 ^{Ba} | 14.40 ^{Ba} | 13.52 ^{Bab} | 11.75 ^b | 10.43 ^c | 0.296 | * |
| | SEM | 0.161 | 0.168 | 0.189 | 0.188 | 0.183 | | |
| | Sig. | * | * | * | n.s. | n.s. | | |
| C* | 1 | 18.32 ^{Aa} | 18.38 ^{Aa} | 17.43 ^{Aa} | 14.04 ^b | 11.98 ^c | 0.368 | * |
| | 5 | 17.07 ^{ABa} | 17.38 ^{Aa} | 15.01 ^{Bb} | 13.11 ^c | 12.27 ^c | 0.295 | * |
| | 10 | 16.15 ^{Ba} | 15.96 ^{Ba} | 16.99 ^{Aa} | 13.19 ^b | 12.01 ^b | 0.303 | * |
| | SEM | 0.257 | 0.259 | 0.334 | 0.242 | 0.233 | | |
| | Sig. | * | * | * | n.s. | n.s. | | |
| h° | 1 | 60.97 ^{Bab} | 61.26 ^{Ba} | 56.62 ^{Bab} | 56.38 ^{Bb} | 56.62 ^{Bb} | 0.593 | * |
| | 5 | 67.02 ^{Aa} | 62.88 ^{ABb} | 61.96 ^{Ab} | 63.62 ^{Aab} | 61.54 ^{Ab} | 0.481 | * |
| | 10 | 71.02 ^{Aa} | 64.57 ^{Ab} | 60.80 ^{Ab} | 62.42 ^{Ab} | 60.84 ^{Ab} | 0.723 | * |
| | SEM | 1.127 | 0.566 | 0.704 | 0.619 | 0.641 | | |
| | Sig. | * | * | * | * | * | | |

^{a-b} Mean values in the same row (different treatment in the same day) with different letters indicate significant difference ($P < 0.05$); ^{A-B} Mean values in the same column (same treatment in different days) with different letters indicate significant difference ($P < 0.05$); SEM: standard error of the mean; Sig.: Significance; n.s.: Not significant; * $P < 0.05$. Treatments: CON: patties prepared without antioxidant; ERY: patties prepared with sodium erythorbate at 500 mg kg⁻¹; AEM: patties prepared with açai extract at 250 mg kg⁻¹; AEH: patties prepared with açai extract at 500 mg kg⁻¹; AEL: patties prepared with açai extract at 750 mg kg⁻¹.



Fig. 1. Photographs of the A) raw pork patties, B) cooked pork patties. Treatments: CON: patties prepared without antioxidant; ERY: patties prepared with erythorbate at 500 mg kg^{-1} ; AEM: patties prepared with açai extract at 250 mg kg^{-1} ; AEH: patties prepared with açai extract at 500 mg kg^{-1} ; AEL: patties prepared with açai extract at 750 mg kg^{-1} .

different concentrations of açai extract in pork patties did not influence the texture, the sensory and nutritional characteristics of the product, as well as the cooking yield (Pedroso & Demiate, 2008).

The percentage of shrinkage of pork patties varied from 14.70% to 18.20% and did not show significant differences ($P > 0.05$) among treatments over the whole display. Avoiding shrinkage is a main goal to preserve the quality standards of patties, due to the potential negative responses from consumers that resemble such changes with an excess of added water (Sánchez-Zapata, Pérez-Alvarez, & Fernández-López, 2012). The diameter reduction is the consequence of the denaturation of proteins, evaporated water and loss of fat, therefore, the addition of sodium erythorbate and different concentrations of açai extract did not influence the diameter reduction and yield.

3.4. Instrumental color parameters

The color of pork patties was affected by both treatment and storage time ($P < 0.05$) (Table 4). A trend was observed where the luminosity (L^*) increased over the storage time for all treatments. The CON and ERY samples did not differ in any sampling point, however; patties prepared with açai extract showed lower values of L^* . Prommachart et al. (2020) reported that black rice extract decreased lightness in beef patties due to the presence of anthocyanin. Similar results were reported by Lee et al. (2016) in cooked pork patties prepared with brown soybean extract (rich source of various phenolic and anthocyanin compounds).

Regarding a^* values, all treatments presented a decreasing trend during the refrigerated storage (Table 4). This outcome is in agreement with Lorenzo et al. (2018), who also observed the same trend in a^* values in pork burgers prepared with pitanga leaf extract. This result is due to the oxidation of lipids and meat pigments occur simultaneously and one increases the other. Myoglobin and hemoglobin are the main heme-proteins in animal tissue and for the development of rancidity, these heme-proteins are oxidized, compromising the color of the meat (Domínguez et al., 2019), and, in this study, it is possible to compare the reduction of a^* values with the increase of lipid oxidation during the storage period for all treatments. At 1 and 5 days of storage, AEH

presented the lower a^* value ($P < 0.05$), followed by AEM treatment. Up to 10 days of storage, the lower a^* value was found in CON (5.24). It means that açai extract affected a^* value, preserving the redness in comparison with CON group (without antioxidant).

The AE addition also affected b^* values ($P < 0.05$) in all sampling points (Table 4). The decrease in b^* value in all treatments was directly proportional to the increase in AE concentration, where AEH treatment showed the lowest b^* values, followed by AEM and AEL batches, respectively. The same trend was observed by Prommachart et al. (2020) in beef patties with black rice extract, where the higher concentration of the extract presented the lowest b^* value. The authors also affirmed that these samples presented a greater bluish tinge. For both b^* and Chroma values, CON, ERY and AEL treatments presented a decrease in b^* values over the storage time, while AEH and AEM treatments did not show difference among the sampling points.

Hue angle presented lower values at 1 day followed by an increase after 5 days ($P < 0.05$), showing not significant differences on hue angle at 10 days. Prommachart et al. (2020) associated the hue with the discoloration and the authors correlated the increase in hue angle with more discoloration. Therefore, in our study, it was observed that AE treatments showed the lower hue angle after 10 days of storage, whereas the higher angle hue was observed in the control group (without antioxidant) followed by ERY treatment. So, AE was able to reduce discoloration in pork patties during the refrigerated storage. In this regard, Prommachart et al. (2020) also found that 1.2% black rice extract was able to reduce discoloration in beef patties. Concerning color parameters, it is important to highlight that AEL treatment did not differ from sodium erythorbate treatment. Fig. 1 presents the color results obtained instrumentally and it is possible to observe that after cooking AEL and ERY samples remained similar to each other. Therefore, açai extract at 250 mg kg^{-1} can be used in pork patties without affecting its color and consequently its acceptance by the consumer because color is the first impression by the consumer for any meat product and is usually the basis for choosing or rejecting the product (Tomasevic et al., 2019).

Table 5

Evolution of TBARS values of pork patties with sodium erythorbate and different levels of açai extract (AE) during refrigerated storage.

| Day | TBARS (mg of MDA/g sample) | | | | | SEM | Sig |
|-----|----------------------------|---------------------|----------------------|---------------------|---------------------|-------|-----|
| | CON | ERY | AEL | AEM | AEH | | |
| 1 | 0.448 ^{Ba} | 0.100 ^{Bb} | 0.238 ^{Bb} | 0.170 ^{Bb} | 0.151 ^{Bb} | 0.026 | * |
| 5 | 0.677 ^{ABa} | 0.242 ^{Ab} | 0.326 ^{ABb} | 0.276 ^{Ab} | 0.206 ^{Ab} | 0.035 | * |
| 10 | 0.889 ^{Aa} | 0.240 ^{Ab} | 0.379 ^{Ab} | 0.293 ^{Ab} | 0.217 ^{Ab} | 0.049 | * |
| SEM | 0.058 | 0.024 | 0.022 | 0.019 | 0.008 | | |
| Sig | * | * | * | * | * | | |

^{a-b} Mean values in the same row (different treatment in the same day) with different letters indicate significant difference ($P < 0.05$); ^{A-B} Mean values in the same column (same treatment in different days) with different letters indicate significant difference ($P < 0.05$); SEM: standard error of the mean; Sig.: Significance; n.s.: Not significant; * $P < 0.05$. Treatments: CON: patties prepared without antioxidant; ERY: patties prepared with sodium erythorbate at 500 mg kg⁻¹; AEM: patties prepared with açai extract at 250 mg kg⁻¹; AEH: patties prepared with açai extract at 500 mg kg⁻¹; AEL: patties prepared with açai extract at 750 mg kg⁻¹.

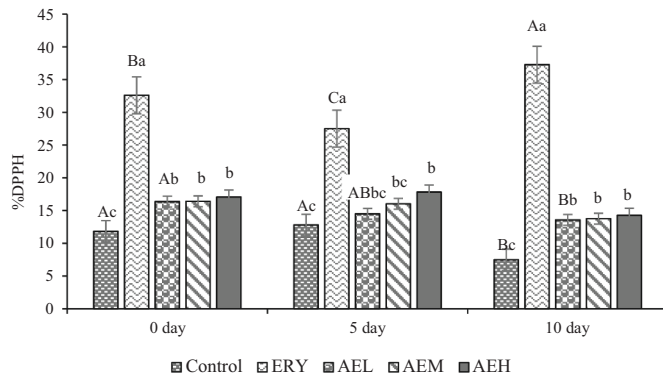


Fig. 2. Antioxidant activity (%DPPH) of pork patties with erythorbate and different levels of açai extract (AE) during 10 days of refrigerated storage. ^{a-c} Mean values in the same day in the different treatment with different letters indicate significant difference ($P < 0.05$); ^{A-C} Mean values in the same treatment in different day with different letters indicate significant difference ($P < 0.05$); Errors bars corresponding to standard error. Treatments: CON: patties prepared without antioxidant; ERY: patties prepared with erythorbate at 500 mg kg⁻¹; AEM: patties prepared with açai extract at 250 mg kg⁻¹; AEH: patties prepared with açai extract at 500 mg kg⁻¹; AEL: patties prepared with açai extract at 750 mg kg⁻¹.

3.5. Lipid oxidation

Lipid oxidation in pork patties is expressed through TBARS values and significant differences were observed among treatments ($P < 0.05$) (Table 5). Sodium erythorbate and AE (all the levels studied) were effective to delay the lipid oxidation, where the samples prepared with antioxidants presented the lowest TBARS values in all sampling points. Consequently, control treatment displayed higher lipid oxidation values than other treatments throughout the storage period (0.448, 0.677 and 0.889 mg MDA/kg of sample for 1, 5 and 10 days, respectively). According to Campo et al. (2006), oxidation is the main factor of deterioration in meat and the limits of perception vary according to the sensitivity and experience of the panelist and can differ according to the origin of the meat (bovine, pork, etc.). It is also important to highlight that only CON treatment reached the limit values for the sensory perception of rancidity (0.5 to 1.0 mg MDA/kg of sample) for pork meat (Tarladgis, Watts, Younathan, & Dugan, 1960).

The TBARS values obtained from lower concentrations of açai extract (250 mg.kg⁻¹) did not differ from ERY samples in all sampling points (1, 5 and 10 days) and this result supports the antioxidant potential of AE extract against the lipid oxidation. This antioxidant effect can be explained by the active compounds present in the extract, as according to Garzón et al. (2017), the high antioxidant activity from açai extract is linked with the presence of flavonoids such as orientin, homoorientin, vitexin, luteolin, chrysoeriol, quercetin, and dihydrokaempferol. Flavonoids are able to scavenge or quench oxygen free radicals or excited

oxygen species. In addition, its action mechanism can inhibit oxidative enzymes to generate these reactive oxygen species (Kang et al., 2011; Paz et al., 2015).

Similarly, Bellucci, Barretto, et al., 2021, Bellucci, Munekata, et al. (2021) also found an effective extract against the lipid oxidation in pork patties. In addition, the authors observed an increase in TBARS values over the storage time for all treatments and this behavior was also observed in this study, where the TBARS values increased from 1 to 10 days of storage ($P < 0.05$). A similar behavior was observed by others authors (Fernandes, Trindade, Lorenzo, & de Melo, 2018; Lorenzo, González-Rodríguez, Sánchez, Amado, & Franco, 2013; Lorenzo, Sineiro, Amado, & Franco, 2014; Pateiro et al., 2018) who added natural extracts into meat products.

3.6. In vitro antioxidant activity of pork patties

In vitro antioxidant activity of pork patties was measured through DPPH assay and the results are shown in Fig. 2. Among all evaluated sampling points, ERY treatments presented the highest antioxidant activity. In this regard, Bellucci, Barretto, et al., 2021, Bellucci, Munekata, et al. (2021) also found the same trend since patties with açai extract showing higher antioxidant activity compared with the control group (without antioxidant). This finding also agrees with data reported by Prommachart et al. (2020), who found lower values of antioxidant activity in control without antioxidant compared with treatments of beef patties added of black rice extract.

4. Conclusion

The addition of açai extract (250, 500 and 750 mg.kg⁻¹) improved the antioxidant status of pork patties, but caused changes on color parameters at medium and high levels. The low level addition (250 mg.kg⁻¹) showed less changes on color parameters than the other levels of açai extract studied. This treatment also displayed a similar protective effect against the lipid oxidation that the samples elaborated with sodium erythorbate. Therefore, at 250 mg.kg⁻¹, açai extract can be used as a natural antioxidant to replace sodium erythorbate to preserve the quality and extend the shelf-life of chilled pork patties.

SEM: standard error of the mean; Sig.: Significance; n.s. Not significant ($P < 0.05$). Treatments: CON: patties prepared without antioxidant; ERY: patties prepared with sodium erythorbate at 500 mg kg⁻¹; AEM: patties prepared with açai extract at 250 mg kg⁻¹; AEH: patties prepared with açai extract at 500 mg kg⁻¹; AEL: patties prepared with açai extract at 750 mg kg⁻¹.

Declaration of Competing Interest

None.

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