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Innovative *dulce de leche* made by sheep's milk with and without the addition of sheep's milk cream

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Abstract

The objective of this work was to develop innovative formulations of *dulce de leche* made with sheep's milk with and without the addition of sheep's milk cream and evaluate the shelf life of these products. Physicochemical and microbiological evaluations were performed on sheep's pasteurized milk and cream, as well as on *dulce de leche* right after production and during the storage time. Addition of cream to sheep *dulce de leche* did not change the physicochemical and microbiological parameters between formulations. The *dulce de leche* showed dark color and adequate results of protein, fat, total and microbiological counts (thermotolerant coliforms, *Staphylococcus aureus*, *Salmonella* sp. in all evaluations, and total mesophilic aerobic up to 150 days of storage). Innovative formulations of *dulce de leche* made by sheep's milk were developed, differentiated from those existing in the Brazilian market, but a specific legislation is required in Brazil focused on the specific characteristics and quality parameters of sheep's milk and its derivatives.

Keywords: sheep dairy products; shelf life; physicochemical and microbiological evaluation.

Practical Application: The development of sheep's *dulce de leche* formulations with and without the addition of sheep's milk cream contributes to the diversification and innovation of production in dairy, especially from milk of small ruminants. The definition of conditions of processing and verification of quality during storage demonstrate diversification potential for small-scale production.

1 Introduction

Sheep farming has grown in recent years in Brazil and the Brazilian sheep herd represents 22.11% of the sheep in the American continent in 2017 (Food and Agriculture Organization of the United Nations, 2019). Climatic conditions in Southern Brazil favor the production and adaptation of several dairy sheep breeds and, consequently, their productivity, presenting a high economic potential for this sector (Munieweg et al., 2017). Sheep milk has higher macronutrient contents than bovine milk (Balthazar et al., 2017; Munieweg et al., 2017), but its composition might be influenced by animal's race and health status, milking techniques, environment and processing technology, among other factors (Balthazar et al., 2017). The average composition of raw sheep milk produced in Southern Brazil ranges from 5.5 to 6.0% of protein, 5.9 to 7.3% of fat, 0.7 to 0.9% of ashes and 17.1 to 17.5% of total solids (Munieweg et al., 2017).

Sheep milk production in Brazil is concentrated in one company being responsible for all stages of production (Santos et al., 2016). This raises the cost of milk production, reduces its commercialization and leads to the production of higher value added to derivatives such as cheese, yogurt and *dulce de leche*, allowing the maintenance of the farm and the expansion of dairy sheep farming (Santos et al., 2016; Balthazar et al. 2017; Munieweg et al., 2017).

Dulce de leche is a traditional South American product prepared by slowly heating milk and sugar, with or without the addition of flavoring, until it reaches a concentration of approximately 68-70 °Brix (Brasil, 1997; Gaze et al., 2015; Francisquini et al., 2019). Brazilian law defines the characteristics and classification of *dulce de leche* (Brasil, 1996b, 1997, 2001), focused on products of bovine origin, and this dairy product from sheep milk could represent an economic alternative for producers and a differentiated option for consumers.

Derivatives made of sheep milk have been recently studied, including Ricotta cheese (Fusaro et al., 2019), ice cream with probiotic (Balthazar et al., 2018b), beverage with strawberry (Balthazar et al., 2018a) and milk-juice beverage with fermented milk (Balthazar et al., 2019). Studies with sheep milk and derivatives have demonstrated that they are good sources of proteins and minerals (Balthazar et al., 2019), beneficial fatty acids such as α-linoleic acid and conjugated linoleic acid (Renes et al., 2018; Fusaro et al., 2019), in addition to their unique sensorial characteristics and functional properties (Balthazar et al., 2018a, b, 2019; Fusaro et al., 2019).

The innovative character of sheep dairy products in Brazil occurs due to the use of raw sheep milk, which is unconventional

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and with a limited number of commercial products. The production of *dulce de leche* from this raw material and with the addition of sheep's milk cream presents an innovative product to meet a strong demand for the gourmet and premium market. Thus, the objective of this work was to develop two innovative formulations of *dulce de leche* using sheep milk, with and without the addition of sheep milk cream, and to monitor the shelf life of these products through physical, chemical and microbiological evaluations.

2 Materials and methods

Sheep milk and cream were supplied by a dairy industry located in Southern Brazil that works with animals of the Lacaune breed. Sheep milk was previously skimmed to 3% of fat. Milk and cream were pasteurized and transported in plastic buckets for food use, placed in thermal boxes and transported under refrigeration to the laboratories. The ingredients such as crystallized sugar, glucose syrup and sodium bicarbonate were purchased on a local market.

Preliminary tests were performed to define the proportions of the raw materials and ingredients, as well as to the cooking times and inclusion of each component. The temperature during the preparation of the formulations was checked using a skewer thermometer CE F1007 (Incoterm®) and aliquots were collected to evaluate the total soluble solids (TSS) content in a model DR201-95 digital bench refractometer (A KrussOptronic®). Two formulations of *dulce de leche* were used: one without (DOC) and another with the addition (DWC) of sheep's milk cream. The DWC formulation contained 3% of cream in partial substitution to the sheep milk. The other ingredients added were crystallized sugar, glucose syrup and sodium bicarbonate. The product was packaged while warm in sterilized glass jars (120 mL) with lids and stored at room temperature for 180 days, with random collection of three jars every 30 days for further evaluations.

Pasteurized milk and cream were evaluated upon arrival and the physicochemical analyzes included: protein, titratable acidity and pH analysis, as well as total dry extract, fat and cryoscopic index for pasteurized milk only. In the *dulce de leche*, water activity (Aw) and titratable acidity were evaluated, with periodic collections every 30 days, in addition to fat, protein and pH at 0, 90 and 180 days after production. Assessments of fat, protein, titratable acidity and cryoscopic index followed official methodologies (Brasil, 2017). The Aw was performed on an Aqualab 4TE meter (Decagon®) and the pH was determined on a pg1800 benchtop potentiometer (GGHAKA®) (Brasil, 2017). The values used as references were those established by the current Brazilian legislation (Brasil, 1996a, b, 1997, 2018).

Microbiological analyzes on milk and cream were performed upon arrival and on *dulce de leche* every 30 days up to six months. Microbiological analyzes in all samples followed standardized methodologies and included: total mesophilic aerobic counts, total and thermotolerant coliforms (Brasil, 2017), *S. aureus* and *Salmonella* sp. (Silva et al., 2017). Psychrotrophic microorganisms (Silva et al., 2017) were determined only in milk and cream and the quantification of mold and yeast (Silva et al., 2017) was performed in the *dulce de leche*. The values were compared with the limits established by the current Brazilian legislation (Brasil, 1996a, b, 1997, 2001, 2018).

Color analyzes were performed with a portable measuring Chroma Meter CR-400 (Konica Minolta®) colorimeter, which works with three scales: L*, a* and b*. L* measures brightness and ranges from 0 for black to 100 for perfectly white surfaces. The a* scale measures the intensity of green (-) and red (+) color, while b* evaluates the transition from blue (-) to yellow (+) color (Ahmed et al., 2019; Gaze et al., 2015). The colorimeter was previously calibrated and the analyzes were performed in triplicate. The total color difference (ΔE) between the two formulations was calculated as a way of assessing the differences noticeable to the human eye, considering the formula without cream (DOC) and with cream (DWC), using the formula that considers the variations of L*, a* and b*: $\Delta E^* = [\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2}]^{1/2}$ (Gaze et al., 2015; Ahmed et al., 2019).

Results were tabulated using a Microsoft Excel 2013 program, obtaining mean and standard deviation from the mean, and microbiological counts values converted to logarithms (log). Data were evaluated by the ASSISTAT 7.7 beta program, applying the analysis of variance followed by the Tukey test.

3 Results and discussion

The raw materials used in the production of *dulce de leche* were evaluated and pasteurized sheep milk showed 3.04 ± 0.03% of protein; 3.35 ± 0.05% of fat; 17.64 ± 2.35% of total dry extract; titratable acidity of 0.19 ± 0.01% in lactic acid; pH of 6.66 ± 0.05 and cryoscopic index of -0.518 ± 0.02 °H. The fat content in pasteurized milk was lower than whole milk of Lacaune sheep, which has average contents above 5.8% (Munieweg et al., 2017; Tribst et al., 2020), due to previous fat removal. Protein values of pasteurized sheep milk were lower than those observed in other studies with raw sheep milk, with averages of 4.8 to 6.0% (Balthazar et al., 2017; Munieweg et al., 2017; Renes et al., 2018), probably due to differences in the processing with removal of components during skimming and heating in pasteurization. Both percentages of fat and protein in sheep milk evaluated were in accordance with the Brazilian legislation (Brasil, 2018). The protein content of sheep's milk cream was lower than those found in bovine cream marketed in Brazil, with an average of 2.39% (Stephani et al., 2011) and the low efficiency of the skimming equipment may be the reason for this reduced percentage. Pasteurized sheep milk showed acidity above 0.14 to 0.18% of lactic acid defined by legislation (Brasil, 2018), according to other authors (Balthazar et al., 2017; Munieweg et al., 2017; Tribst et al., 2020). For sheep's milk cream, the average acidity observed was below the maximum value allowed in legislation (0.20% of lactic acid) (Brasil, 1996a). The pH values of sheep raw milk and cream were similar to those found in bovine milk cream (Stephani et al., 2011) and lower than those observed in raw sheep milk (Munieweg et al., 2017; Tribst et al., 2020) and sheep milk ice cream made with skimmed sheep milk added to sheep milk cream (Balthazar et al., 2018b). The cryoscopic index observed in pasteurized sheep milk was below the range recommended by law (-0.530 ° H to -0.555 °H) (Brasil, 2018), the same reported in another study with raw sheep milk (Munieweg et al., 2017), and these differences may be due to the higher milk solids content of sheep as opposed to the legal limits set up for bovine milk (Brasil, 2018).

Microbiological evaluation of pasteurized sheep milk resulted in the following counts: 9.92 ± 0.01 log CFU/mL for total mesophilic aerobic; 1.18 ± 0.01 log NMP/mL total coliforms; 0.96 ± 0.01 log NMP/mL of thermotolerant coliforms; 9.24 ± 0.17 log CFU/mL for psychrotrophs; absence of *S. aureus* at dilution 10^{-1} and *Salmonella* sp. in 25 mL of milk. The pasteurized sheep's milk cream contained 0.17 ± 0.06% of protein, titratable acidity of 0.08 ± 0.01% in lactic acid, and pH 6.75 ± 0.01. The mesophilic aerobic count in cream was 5.56 ± 0.01 log CFU/g; less than 0.48 log NMP/g for both total and thermotolerant coliforms; 5.83 ± 0.07 log CFU/g for psychrotrophs; absence of *S. aureus* at dilution 10^{-1} and *Salmonella* sp. in 25g. The mesophilic aerobic count in the cream and pasteurized milk exceeded the legal limits (Brasil, 1996a), as well as in other studies with raw milk of goats and sheep in Brazil (Agibert, 2013; Munieweg et al., 2017; Jiménez-Sobrino et al., 2018), a fact not observed in European studies using sheep milk (Renes et al., 2018). Brazilian legislation establishes maximum limits for pasteurized milk of 0.69 log NMP/mL for *Enterobacteriaceae* (Brasil, 2018) and 0.60 log NMP/mL for thermotolerant coliforms (Brasil, 2001), so pasteurized sheep's milk was above the allowed limit for this last evaluated agent. For pasteurized sour cream, the maximum limit for total coliforms is 1.0 log MPN/g (Brasil, 1996a) and for thermotolerant coliforms ranges from 0.48 log MPN/g (Brasil, 1996a) to 0.60 log MPN/g (Brasil, 2001), therefore the raw material evaluated in the present study was in accordance with the established values. Several pathogenic and deteriorating microorganisms are mesophilic aerobics and coliform group is an indicator of hygienic-sanitary quality (Forsythe, 2013), and high counts may indicate hygiene failures during milking, transport and/or refrigerated milk storage (Munieweg et al., 2017; Jiménez-Sobrino et al., 2018). The absence of *S. aureus* and *Salmonella* sp. in the evaluated raw materials is in accordance with the legal parameters (Brasil, 1996a, 2001) and this monitoring is important since they are pathogenic bacteria and are associated with contamination in dairy products (Forsythe, 2013). Psychrotrophic counts were high in pasteurized milk and sheep's milk cream, but there is no limit set by the Brazilian legislation. This group of microorganisms is common in products stored under refrigeration and has also been found in pasteurized bovine milk, raw sheep and goat milk (Agibert, 2013; Munieweg et al., 2017; Jiménez-Sobrino et al., 2018). Sheep milk and sour cream were pasteurized to reduce microbial contamination (Forsythe, 2013), but this process was not enough in pasteurized sheep milk because high total and psychrotrophic mesophilic aerobic counts were observed. Brazilian sheep milk producers indicate the need for specific legislation for sheep milk, such as that defined for goat milk, due to the specificities of this raw material and the difficulties in adapting to the requirements established for bovine milk.

The results of protein, fat and pH contents of the *dulce de leche* are shown in Table 1.

Based on fat contents (Table 1) and the legislation (Brasil, 1997), *dulce de leche* without cream (DOC) was classified as a pure *dulce de leche* and the formulation with cream (DWC) was classified as *dulce de leche* with cream milk, compatible with the composition of each formulation. Other authors report average fat content in *dulce de leche* made of goat milk ranging from 16 to 20% (Agibert, 2013), 7.9% for sweetened condensed milk (Renhe et al., 2017) and 3.6 to 7% in *dulce de leche* of bovine milk (Gaze et al., 2015; Silva et al., 2015). The structure and distribution of fat globules can be affected by some processes such as heat treatment, agitation and centrifugation employed to obtain milk cream (Balthazar et al., 2017). The DOC showed higher amounts ($p < 0.05$) of protein compared to DWC at 180 days of storage (Table 1) and formulations with sheep's milk cream contained protein levels above the minimum required by the legislation (Brasil, 1997). In the DWC formulation, there was a partial replacement of the pasteurized sheep's milk by pasteurized sheep's milk cream, which resulted on a lower protein content due to lower amount of protein in the cream compared to the milk (0.17% and 3.04%, respectively). Protein percentages in the DOC and DWC formulations were similar to those found in other studies with *dulce de leche* from goat milk (Agibert, 2013), from goat milk with chia flour (Chaves et al., 2018) and from bovine milk (Silva et al., 2015). Mean pH values ranged from 6.34 to 6.53, with no significant differences between formulations or sampling times (Table 1). These values were higher than those found in bovine *dulce de leche*, with the addition of goat's whey and with different concentrations of umbu pulp, whose pH ranged from 4.40 to 5.49 (Silva et al., 2011), similar to those observed in commercial bovine *dulce de leche* (6.14 to 6.37) (Gaze et al., 2015) and lower than those of buffalo *dulce de leche* (7.02 and 6.87) (Jacob et al., 2018). Considering the observed pH values, the formulations of the present work can be classified as low acid foods. which increases the risk of contamination by pathogenic microorganisms (Forsythe, 2013).

The evaluation of water activity and titratable acidity in *dulce de leche* made by sheep's milk are presented in Figure 1. The Aw results of *dulce de leche* ranged from 0.71 to 0.82 (Figure 1A), with significantly ($p < 0.05$) higher values at times 0 and 90 days

Table 1. Fat, protein and pH contents of two formulations of *dulce de leche* with and without the addition of sheep's milk cream during storage.

Period (days)	Fat (%)		Protein (%)		pH	
	DOC	DWC	DOC	DWC	DOC	DWC
0	5.2 ± 3.5^{ns}	6.4 ± 2.0^{ns}	10.3 ± 1.3^{ab}	7.8 ± 0.2^{bc}	6.52 ± 0.03^{ns}	6.52 ± 0.05^{ns}
90	7.7 ± 1.2^{ns}	11.0 ± 3.1^{ns}	10.2 ± 0.4^{ab}	9.9 ± 0.2^{ab}	6.41 ± 0.02^{ns}	6.37 ± 0.04^{ns}
180	8.2 ± 2.6^{ns}	10.8 ± 0.1^{ns}	11.4 ± 2.5^{a}	7.0 ± 0.8^{c}	6.34 ± 0.1^{ns}	6.53 ± 0.05^{ns}
Standard*	6 to 9%	Min. 9%	Min. 5%		-	

DOC: *Dulce de leche* without sheep's milk cream; DWC: *Dulce de leche* with sheep's milk cream. Mean values ± standard deviation (n = 2 for fat; n = 3 for the others). Different letters in the same column indicate statistically significant difference ($p < 0.05$) and ^{ns} not significant. *Brasil (1997).

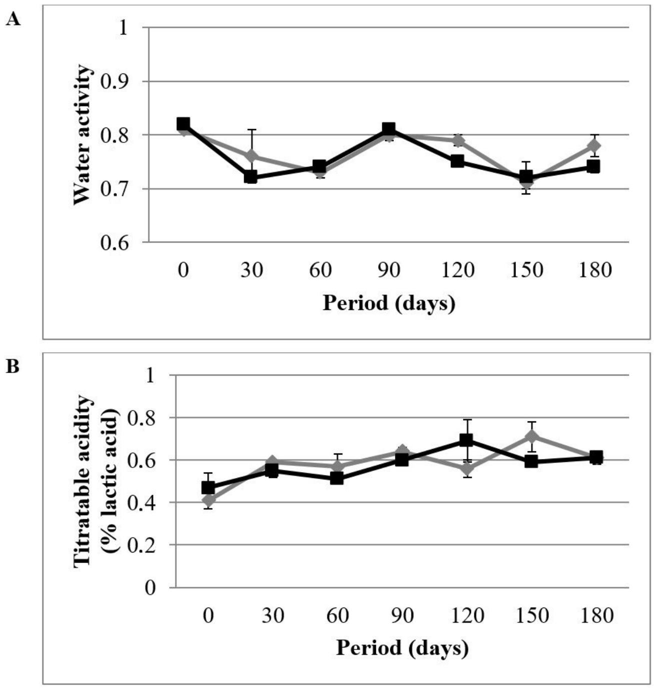

Figure 1. Evaluation of (A) water activity and (B) titratable acidity in two formulations of *dulce de leche* with and without the addition of sheep's milk cream during storage. The gray line represents the formulation DOC (dairy without cream) and the black line indicates the DWC (*dulce de leche* with cream) (n = 3).

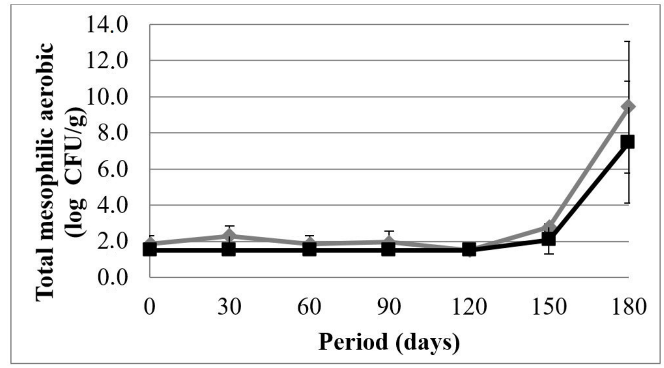

Figure 2. Total mesophilic aerobic count in both formulations of sheep's milk with and without the addition of sheep's milk cream during storage. The gray line represents the formulation DOC (dairy without cream) and black indicates the formulation DWC (dairy with cream) (n = 3).

Table 2. Mold and yeast count of two formulations of *dulce de leche* with and without the addition of sheep's milk during storage.

Period (days)	Mold and Yeast (log UFC/g)	
	DOC	DWC
0	3.35 ± 0.38^{b}	nd*
30	3.51 ± 0.43^{b}	2.14 ± 0.83^{b}
60	2.52 ± 0.19^{b}	nd
90	3.39 ± 0.18^{b}	3.48 ± 0.09^{b}
120	3.75 ± 0.20^{b}	3.24 ± 0.30^{b}
150	3.47 ± 0.26^{b}	3.25 ± 0.27^{b}
180	3.86 ± 0.23^{a}	4.29 ± 0.21^{a}
Standard**	Maximum 2.00	

DOC: *Dulce de leche* without sheep's milk cream; DWC: *Dulce de leche* with sheep's milk cream. Mean values ± standard deviation (n = 3). Different letters in the same column indicate a statistically significant difference ($p < 0.05$). *nd: not detected at dilution 10^{-1}; **Brasil (1996b, 1997).

for both formulations. Studies with bovine *dulce de leche* showed Aw values between 0.85 and 0.91 for industrialized formulations and condensed sweetened milk (Ferreira et al., 2012; Renhe et al., 2017) and these variations may occur with *dulce de leche*, due to heating that can concentrate the ingredients and increase gradually the amount of insoluble solids (Francisquini et al., 2019). The titratable acidity in the products ranged from 0.41 to 0.71% of lactic acid in the DOC formulation and from 0.47 to 0.69% lactic acid in the DWC (Figure 1B). There was a gradual increase in acidity in both formulations during storage, with significant differences ($p < 0.05$) on day 180 of storage compared to the initial time, but without variation between DOC and DWC at any of the sampling times. Studies report 3.02% acidity in lactic acid of fresh milk of buffalo and 3.6% in *dulce de leche* stored for nine months (Jacob et al., 2018), much higher values than those found in fresh *dulce de leche* of this study. The acidity values in seven brands of *dulce de leche* marketed in Brazil showed values of 0.23 to 0.50% acidity in lactic acid (Gaze et al., 2015), which are much lower than those observed in the present study.

The results of water activity of *dulce de leche* in the present study showed values below the minimum limit for multiplication of pathogenic bacteria, mainly due to the addition of sugar that acts to reduce the amount of free water in the food, however, the titratable acidity was low for microbial control to occur (Forsythe, 2013).

The quantification of the total mesophilic aerobic group over the storage period can be seen in Figure 2 and mold and yeast counts are shown in Table 2. The DOC formulation had counts between 1.52 to 9.42 log CFU/g, while DWC ranged from 1.52 to 7.49 log CFU/g, with significant differences ($p < 0.05$) at 180 days for both formulations compared to the other sampling times (Figure 2). The increase in the counting of this group after 150 days of storage might be associated with a process of deterioration, since these products were stored at room temperature, with favorable conditions for the growth of mesophilic microorganisms (Forsythe, 2013). High counts of these microorganisms were also found in bovine *dulce de leche* in Lavras, MG (Southeast of Brazil), with an average count of 5.86 log CFU/g (Oliveira et al., 2012) as well as for sweetened condensed milk samples, ranging from 1.0 and 6.2 log CFU/g (Renhe et al., 2017). Although Brazilian legislation does not set limits for this microbial class in *dulce de leche*, monitoring of total mesophilic aerobes provides information on product contamination and may assist in defining the shelf life of a new product (Forsythe, 2013).

Beside this, both formulations of *dulce de leche* had counts of less than 0.48 log NMP/g for total and thermotolerant coliforms and absence of *Salmonella* sp. and *S. aureus*, which indicates that the samples are appropriate to the legislation considering these microbiological parameters (Brasil, 1996b, 1997, 2001).

The low values quantified for the coliform group indicate that the processing and handling conditions were adequate, and the non-detection of *Salmonella* sp. and *S. aureus* emphasizes the safety of the food produced (Forsythe, 2013). Milk pasteurization and the cooking process while making the *dulce de leche* contribute to the destruction of microorganisms (Forsythe, 2013), and these steps were effective for the control of these microorganisms.

Mold and yeast counts presented values of 3.86 and 4.29 log CFU/g for the DOC and DWC formulations, respectively, at 180 days of storage (Table 2). In only two DWC sampling, the maximum limit for mold and yeast was not exceeded, while in DOC the counts were above the limit allowed by law (Brasil, 1996b, 1997) during the whole storage period. Previous work on goat *dulce de leche*, with or without whey (Agibert, 2013), have found mold and yeast counts lower than 2.0 log CFU/g allowed. On the other hand, counts above 3.0 log CFU/g were reported in bovine *dulce de leche* marketed in Lavras, MG (Southeast of Brazil) (Oliveira et al., 2012). In some of these formulations, the low counts for mold and yeast may have been influenced by the addition of preservatives allowed in the preparation of *dulce de leche* (Brasil, 2010), considered the factor responsible for controlling the presence of fungi in *dulce de leche* formulated with or without sucrose (Milagres et al., 2010). However, preservatives were not used in the formulations of the present study. Water activity results in sheep *dulce de leche* (Figure 1A) showed values from 0.71 to 0.82, which are limiting for pathogenic and some spoilage microorganisms, but the increase in sugar concentration would only inactivate microbial multiplication in Aw below 0.70 (Forsythe, 2013). The quantification of molds and yeasts, above the values allowed by legislation may indicate flaws in the processing and demonstrated how easy it is for fungi to grow in foods with low water activity, with potential risk to consumer health, since many fungi species produce mycotoxins (Oliveira et al., 2012; Forsythe, 2013). The preparation of DOC and DWC involved pasteurization and cooling of raw materials, concentration by addition of sugar and heat, and the use of good processing practices. All these processes help in the prevention and conservation of food, with destruction of pathogenic bacteria and reduction of microbial load (Oliveira et al., 2012; Forsythe, 2013). under storage at room temperature. However, the presence of molds and yeasts above the levels allowed by the legislation reveals failures that occurred in any of the production or sealing steps of the packaging, allowing the passage of oxygen and contaminants, although the non-use of chemical preservatives in sheep's milk *dulce de leche*.

Color results of the formulations can be seen in Table 3, without significant differences ($p < 0.05$) between formulations or storage periods.

The L* values in the DOC and DWC were below 50 (Table 3), the midpoint of this scale, indicating a tendency of dark staining of the samples. In studies with *dulce de leche*, the brightness values were between 48 and 68 (Ferreira et al., 2012; Agibert, 2013; Silva et al., 2015; Gaze et al., 2015), higher than those found in the present work. In *dulce de leche* containing umbu pulp, the measurements ranged from 39.0 to 47.9 (Silva et al., 2011) and the values ranged between 41.8 and 43.3 for *dulce de leche* from goat milk and with chia flour (Chaves et al., 2018), close to the *dulce de leche* of the present study. The results of the a* scale were all positives, with no significant differences between formulations ($p < 0.05$), demonstrating a tendency to red (Table 3). The trend for this color was also observed in goat *dulce de leche* (Agibert, 2013; Chaves et al., 2018), commercial bovine *dulce de leche* (Gaze et al., 2015) and bovine *dulce de leche* with different concentrations of umbu pulp (Silva et al., 2011). The b* values indicated that both formulations of *dulce de leche* showed higher wavelength reflection associated with the yellow color (Table 3). These positive results were also identified by other authors (Agibert, 2013; Chaves et al., 2018; Gaze et al., 2015; Ferreira et al., 2012; Silva et al., 2011, 2015). The total color difference (ΔE) between the formulations indicated values of 0.96 at initial time, 2.13 at 90 days, and 1.88 at 180 days of storage. According to Gaze et al. (2015), total color differences up to 1.5 are considered small, as observed at baseline, and ΔE of 1.5 to 3.0 between formulations indicate that they are distinct, which occurred at 90 and 180 days of storage. The brightness difference (ΔL*) showed negative results in all periods evaluated, indicating that the DWC formulation was slightly brighter than the DOC, but without significant variations ($p < 0.05$). The fact that there were no significant variations between both formulations of *dulce de leche* elaborated in the present study regarding their color, demonstrates that the technology used in the production of both was similar and that the cream added to the *dulce de leche* (DWC) did not interfere in this parameter. The initial acidity of milk, the amount and timing of the addition of sodium bicarbonate, and the initial and final soluble solids content may be some of the triggering factors for differences in the coloration of *dulce de leche* (Ferreira et al., 2012), a fact not observed in the present study.

In the next step of the study, we will conduct the sensory analysis of the *dulce de leche* formulations to verify the acceptance of these differentiated products in relation to those existing on the market. Acceptance sensory tests can evaluate parameters by a numerical scale (Balthazar et al., 2018a; Fusaro et al., 2019) combined with relevance coefficients for each sensory parameter (Heydari et al., 2018) and descriptive analysis can be used to identify the relationship between products and their sensory attributes, which must be performed by consumers with high

Table 3. Color analysis on the L*, a* and b* scales for both formulations of *dulce de leche* with and without the addition of sheep's milk during storage.

Period (days)	L* ^{ns}		a*^{ns}		b*^{ns}	
	DOC	DWC	DOC	DWC	DOC	DWC
0	41.18 ± 0.30	40.94 ± 0.88	3.71 ± 0.93	3.97 ± 0.62	12.43 ± 1.64	12.87 ± 1.07
90	41.81 ± 1.55	40.83 ± 0.38	3.07 ± 0.58	2.52 ± 0.28	11.20 ± 1.10	10.46 ± 0.50
180	42.07 ± 0.25	41.60 ± 0.29	3.25 ± 0.33	2.91 ± 0.22	11.59 ± 0.81	10.64 ± 0.44

DOC: *Dulce de leche* without sheep's milk cream; DWC: *Dulce de leche* with sheep's milk cream. Mean values ± standard deviation (n = 3). ^{ns} indicates that the result of the statistical evaluation was not significant ($p < 0.05$) for these parameters.

sensory acuity in dairy assessment (Balthazar et al., 2018a; Chetachukwu et al., 2019; Fusaro et al., 2019). The use of the projective techniques has been applied to evaluate the sensorial perception of Brazilian consumers about healthy, differentiated or unconventional products in the market. These techniques, such as free word association and Haire's shopping list methods, can be useful to comprehend the perceptions and beliefs about food products and can be applied in the development or reformulation of products and marketing strategies (Judacewski et al., 2019; Pinto et al., 2018). For similar purpose, consumer-based methodologies are gaining popularity in the sensorial science to improve the novel dairy development and to expand efficient marketing strategies of commercialization of these products. The preferred attribute elicitation methodology demonstrated to be useful to assess the perceptions of consumers of dairy product samples, and it did not need training, resulting in suitable reliability with a low number of consumers (Soares et al., 2019). The check-all-that-apply questionnaire allows assessors to select all appropriate sensory characteristics for describing a product and contributes to optimizing the different dairy foods with similar or complex sensory characteristics (Torres et al., 2017). All of these methods can contribute so that the *dulce de leche* developed in the present study have consumption potential in the Brazilian market.

4 Conclusions

Addition of cream to sheep *dulce de leche* did not change the physicochemical and microbiological parameters between formulations, that showed dark color, and sheep *dulce de leche* were stable up to 150 days of storage for most of the evaluated parameters, except for mold and yeast over the evaluated period. Innovative formulations of *dulce de leche* made by sheep's milk were developed, differentiated from those existing in the Brazilian market, but a specific legislation for sheep's milk and its derivatives is necessary in Brazil to establish the identity and quality of these products.

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