Effects of colostrum in milk on the effectiveness of the pasteurization process and cheese milk quality

Byron Herrera-Chávez a,b, Antonio José Trujillo b, Paola Calero a, María Inés Falconi a and Davinia Sánchez-Macías a

aAnimal Production and Industrialization Research Unit, Faculty of Engineering, Universidad Nacional de Chimborazo, Riobamba, Ecuador; bCentre d’Innovació, Recerca i Transferència en Tecnologia dels Aliments (CIRTTA), TECNIO, XIA, Departament de Ciència Animal i del Aliments, Facultat de Veterinària, Universitat Autònoma de Barcelona, Bellaterra, Spain

ABSTRACT
Colostrum in the milk bulk tank could affect cheese milk and the characteristics of the final product, leading to multiple problems in industrial processes. The objective of this study was to evaluate colostrum concentrations (0%, 1, 5% and 10%) in cheese milk subjected to two discontinuous heat treatments (56°C for 1 h or 63°C for 30 min) on the physicochemical characteristics, cheese-making properties, and the effectiveness of pasteurization. All physicochemical parameters were modified by colostrum, and some of them were varied due to the thermal treatment. Milk rennet coagulation characteristics were no different among the thermal treatments applied when colostrum (up to 10%) was added to the milk. Both thermal treatments had the same effectiveness in reducing total plate count in raw milk without colostrum; however, when colostrum was added, the treatment at 63°C for 30 min proved to be more efficient.

Introduction

Microorganisms can be introduced into milk at any stage in the production to consumption chain (Sharma et al. 2014). Recently, Alegbeleye et al. (2018) listed the primary sources and routes of microorganisms in milk, such as commensal microflora, diseases, mastitis, environmental contamination, and post-pasteurization contamination. These authors also summarized the microbiological hazards associated with the consumption of raw milk, such as relatively benign symptoms to more severe complications, even long-term, and chronic complications. These risks can be extended to the consumption of non-well pasteurized milk or low-efficiency pasteurization process.

Colostrum is the mammary secretion obtained after parturition, which provides nutrients and passive immunity to the newborn. Abd El-Fattah et al. (2012), Sánchez-Macías et al. (2014) and Tsouplas et al. (2007), working with bovine, goat, and buffalo colostrum, respectively, reported extreme differences in the physical and technological parameters between colostrum and mature milk. Colostrum is obtained in large quantities. It is considered nutraceutical and contains plenty of physiologically active components, including immunoglobulins (Igs), lactoferrin, lysozyme, cytokines, growth factors, hormones, oligosaccharides, and lipid components (Pakkanen and Aalto 1997; Korhonen 2009; Borad and Singh 2018). The main difference between mature milk and colostrum is the level of IgG concentration (0.72–0.90 versus 32–212 mg mL−1 of bovine immunoglobulin G, respectively) (Gapper et al. 2007; Godden et al. 2019). Moreover, colostrum presents a higher microbial count than mature milk, which could increase a load of bacteria if colostrum is added to the milk bulk tank (Godden et al. 2019).

To address food security concerns, many countries, through governments, regulatory authorities, and public health practitioners, require undergoing processing techniques to treat raw milk to eliminate pathogenic microorganisms. These significantly reduce spoilage microflora and inactivate enzymes, to enhance the shelf-life of dairy products. These techniques include thermization, pasteurization, sterilization, ultraviolet treatment, high pressures, microwave treatment, membrane processing, and microfiltration, among others (Sarkar 2015; Bucci et al. 2018). However, scientific evidence shows that pasteurization is the primary, uncontroversial safeguard for ensuring milk quality (Alegbeleye et al. 2018).

Low-temperature-long-time pasteurization or holder method is a typical batch method where milk is placed in an open vat and heated to 63°C for 30 min (Gedam et al. 2007). This is considered the standard minimum time-temperature combination recognized worldwide. It has less impact on milk than higher temperatures and, therefore better retains of raw milk constituents while inactivating relevant pathogens (Angulo et al. 2009). However, in many countries and regions raw milk is used for dairy products processing and consumers’ increased consumption of raw milk or raw dairy products is the trend (Verraes et al. 2015).
Heat treatment adversely affects the bioactivity of colostrum immunoglobulins. Researchers have found the onset of thermal unfolding of IgGs starting at 62.6°C, depending on the pH of colostrum, and complete denaturation at 89°C (Lindström et al. 1994; Singh and Havea 2003). Moreover, it is established that IgGs are vulnerable to heating above 65°C (Indyk et al. 2008; Borad et al. 2019), causing an increase in viscosity and gelation. The thermo-labile nature of colostrum IgGs and other bioactive compounds is well established and depends on temperature, treatment time, pH, and compositional factors (McGrath et al. 2016; Borad and Singh 2018; Morales-delAñuez et al. 2020). Abd El-Fattah et al. (2014) reported that commercial pasteurization time-temperature combinations (63°C for 30 min or 72°C for 15 s) significantly reduce IgG, IgM, IGF-1 and lactoferrin, and increase the viscosity of colostrum compared with 60°C for 60 min treatment. However, Steinbach et al. (1981) and Trujillo et al. (2007) did not observe differences in IgG concentration in colostrum after pasteurization at 65°C for 30 min or 56°C for 60 min.

There is scarce literature about the effects of colostrum on the effectiveness of milk pasteurization and cheese milk quality. In this study, we used the lowest colostrum heat treatment found in the literature (56°C for 60 min) that reduces the bacterial load but affects minimally IgGs concentration (Argüello et al. 2003; Morales-delAñuez et al. 2020), and conventional milk batch low-temperature-long time pasteurization (LTLT, 63°C for 30 min). McGrath et al. (2016) observed that the duration of mammary secretion classified as colostrum varies according to the literature from immediately after parturition to 5–7 days post-partum, which is considered unmarketable and often excluded from bulk milk collection for milk processing. The introduction of colostrum to the milk bulk tank is also possible due to bad farming practices, intentionally or unintentionally. Its composition and high antimicrobial compounds lead to multiple problems and may affect the industrial and fermentation processes (Marnila and Korhonen 2002; McGrath et al. 2016). This could interfere with the effectiveness of the pasteurization of milk. For this reason, we proposed a study to evaluate the effects of two heat treatments applied to milk containing 0%, 1%, 5%, and 10% of colostrum on physicochemical characteristics, pasteurization effectiveness and cheese-making properties.

### Material and methods

#### Colostrum and milk handling

Colostrum and mature milk were obtained from a Holstein dairy farm (Chimborazo Province, Ecuador) using mechanical milking. Animals had good health, including low somatic cell count, and did not receive veterinary treatment before or during the experimental period. First-milking bovine colostrum of ten multiparous Holstein cattle was procured and frozen (−26°C) to create a colostrum bank. Thawed colostrum at 4°C was pooled to create a unique 80-L batch to ensure uniformity of the colostrum sample used in experimental trials. On the other hand, fresh mature milk (20 L) from a bulk tank milk was collected from the same Holstein dairy farm (with 32 animals in their middle lactation) when needed for each batch. For colostrum and mature milk collection, proximate composition, physicochemical characteristics, colour, total plate count and IgG quantification were estimated to ensure the quality of the samples.

#### Experimental design and processing of milk and colostrum

The present experiment was conducted at the laboratories of the Engineering Faculty of the Universidad Nacional de Chimborazo (Riobamba, Ecuador). To study the influence of different concentrations of colostrum in mature milk on the pasteurization effectiveness and cheese milk quality, colostrum was added to milk in different concentrations (0%, 1%, 5% and 10%) until it became 3L. Each group was subdivided into three subgroups of 1L: the first one was treated at 56°C for 60 min (LT56), the second one at 63°C for 30 min (LT63), and the third one received no thermal treatment (Raw). Thermal treatment was performed in 2 L sterilized bottles in a water bath, using an extra bottle to control the internal temperature of the milk during heating. Time started when the experimental temperature was reached. When the time was reached, milk was cooled down and analysed at 20°C. Samples were taken and frozen (−80°C) for later IgG quantification. Each treatment was carried out in triplicate.

#### Physicochemical characteristics of colostrum and milk

Proximate composition (fat, protein, lactose, total solids, and non-fat solids) of colostrum and milk samples was estimated using a MilkoScan 6000 FT (Foss Electric, Hillerød, Denmark). Titratable acidity (percentage of lactic acid) of the samples was measured according to Sánchez-Macias et al. (2014), and the ethanol stability was determined according to Tsioulpas et al. (2007). The buffering capacity was evaluated by adding 100 μL of 0.5 M HCl every 30 s in a 100-mL of colostrum or milk sample, and the final buffering capacity was counted when a constant pH of 4.5 was reached (Huppertz et al. 2004). Milk density and pH were determined using a lactosensor meter (Alla France, Chemillé, France) and a digital pH-meter (Hach, Gel-filled, USA), respectively. A Minolta colorimeter CR-400 (Illuminant D65, Konica Minolta, Osaka, Japan) was used to determine lightness (L*), yellow index (b*), and red index (a*) for each sample. For determining total colour differences among milk samples, Δɛ* was calculated as √[(ΔL*)² + (Δa*)² + (Δb*)²]. The final values of Δɛ* were compared with a range established by the Zmeškal scale (Nedomová et al. 2017). All these parameters were measured in triplicate.

#### Cheese-making properties

LT56 and LT63 milk samples with 0%, 1%, 5%, and 10% of colostrum, heat-treated or raw were coagulated with commercial recombinant chymosin (CHY-MAXs extra; EC 3.4.23.4, Isozyme B, 600 IMCU/mL Chr. Hansen Inc., Barcelona, Spain) at 0.035% (v/v) and 32°C. Milk coagulation characteristics were analysed for rennet clotting time (RCT, min), rate of curd firming (RCF, Δ%T/min), curd firmness at 30, 45 and 60 min (F30, F45 and...
F60, Δ%T), using a near-infrared optical device (Optigraph Ysebaert SA, Frépillon, France). Also, the potential cheese yield (CY, %) and curd draining capacity (CD, %) were assessed according to Calvo and Balcones (1998) and Calvo and Espinosa (1999), respectively, using the same chymosin concentration described above.

**Microbiological analysis and IgG quantification**

To assess the total plate count, samples were serially diluted in a saline solution (0.9% of NaCl) and then plated on 3M™ Petrifilm Aerobic Count plates (3M Microbiology, St. Paul, MN, USA) for aerobic total plate count (TPC). All plates were incubated at 32°C, and the colonies were enumerated after 48 h. The obtained results were expressed in log colony-forming units per mL of sample (log cfu mL$^{-1}$). The inactivation ratios were expressed as cell survival percentage (N/N0 x 100), where N is the number of cells in the heat-treated sample and N0 is the initial number of cells in the untreated sample.

To determine colostrum and milk IgG concentrations, commercial ELISA sets (Bethyl Laboratories, Montgomery, TX, USA) were used. Purified bovine IgG was used as the standard reference. Samples were individually analysed in duplicate, and results were expressed as milligrams of IgG per millilitre.

**Statistical analysis**

Statistical analyses were performed using the SAS program (V.9, SAS Institute Inc., Cary, NC). The ANOVA procedure compared compositional, physicochemical properties, cheese-making properties and TPC between mature milk and colostrum samples. The ANOVA procedure for repeated measurements evaluated the effect of different concentrations of colostrum in milk and heat treatments on proximate composition, physicochemical characteristics, cheese-making properties, and bacterial inactivation. Significantly different means were identified using the Tukey test ($P < 0.05$).

**Table 1.** Physicochemical and microbiological characteristics of colostrum and milk.

| Item            | Colostrum | Milk | SEM$^1$ |
|-----------------|-----------|------|---------|
| Fat (%)         | 4.56$^a$  | 4.18$^b$ | 0.21    |
| Protein (%)     | 11.57$^a$ | 3.36$^b$ | 4.50    |
| Lactose (%)     | 3.60$^a$  | 4.60$^b$ | 0.55    |
| Non-fat solids (%) | 15.76$^a$ | 9.08$^b$ | 4.22    |
| Total solids (%) | 20.79$^a$ | 13.39$^b$ | 4.05    |
| Acidity (%)     | 0.46$^a$  | 0.17$^b$ | 0.16    |
| pH              | 6.22$^a$  | 6.63$^b$ | 0.23    |
| Density (g mL$^{-1}$) | 1.049     | 1.030$^b$ | 0.01    |
| Buffering capacity (mL) | 46.93$^a$ | 28.67$^b$ | 10.31   |
| Ethanol Stability (%) | 50$^a$    | 72.66$^b$ | 16.46   |
| Total plate count (log cfu mL$^{-1}$) | 5.46$^a$ | 3.67$^b$ | 1.00    |
| Immunoglobulin G (mg mL$^{-1}$) | 61.15$^a$ | 1.32$^b$ | 34.55   |
| Colour parameters |           |       |         |
| $L^*$ | 82.37$^a$ | 86.38$^b$ | 3.35    |
| $a^*$ | −3.98$^a$ | −4.28$^b$ | 0.33    |
| $b^*$ | 24.42$^a$ | 13.73$^b$ | 8.35    |

$^a$Means for the same variable in the same row with different superscript differ significantly ($P < 0.05$).

$^b$SEM: standard error of the mean.

**Results and discussion**

**Raw colostrum and milk characteristics**

Table 1 summarizes the proximate composition, physicochemical parameters, total plate count, and IgG quantification of raw colostrum and mature milk used in the experiment. The results showed that the composition of colostrum differs markedly from mature milk for all analysed parameters. The values of fat, protein, non-fat solids, and total solid as well as density and lactic acid content, were higher in colostrum than in milk, while lactose content and pH were lower. Likewise, a markedly higher buffering capacity was found in colostrum, as seen by other authors (Borad et al. 2019). These results are according to the literature when comparing colostrum to mature milk (Marnila and Korhonen 2002; Madsen et al. 2004; Kehoe et al. 2007; Tsoulos et al. 2007; Jeong et al. 2009; Sánchez-Macias et al. 2014).

The total plate count in colostrum was higher (5.46 log cfu mL$^{-1}$) than in milk (3.67 log cfu mL$^{-1}$). Morrill et al. (2012) found that up to 60% of colostrum produced in USA dairy farms does not meet the minimum bacteriological standards for milk (TPC of <100,000 cfu mL$^{-1}$). The values of TPC found in our study for colostrum are according to or like those found in the literature. For example, Donahue et al. (2012) reported that TPC ranged from 5.0 to 5.9 log cfu mL$^{-1}$ in fresh colostrum from six large commercial dairy farms in the USA. Gelsinger et al. (2014) reported 4.59 log cfu mL$^{-1}$ in raw colostrum, while Elizondo-Salazar et al. (2010) reported 4.60 log cfu mL$^{-1}$.

The higher TPC values in colostrum than in mature milk can represent an increased load of microorganisms if colostrum is added to the milk bulk tank. Elizondo-Salazar et al. (2010) reported the presence of bacteria in naturally contaminated colostrum such as environmental streptococci (4.23 log cfu mL$^{-1}$).

**Table 2.** Composition of raw or pasteurized milk containing 0%, 1%, 5% and 10% of colostrum treated at 56°C for 1 h (LT56) or at 63°C for 30 min (LT63).

| Item            | Treatment | 0 | 1 | 5 | 10 | SEM$^1$ |
|-----------------|-----------|---|---|---|----|---------|
| Fat (%)         | Raw       | 4.18$^a$ | 4.20$^a$ | 4.23$^b$ | 4.28$^b$ | 0.01    |
|                 | LT56      | 4.25$^a$ | 4.28$^b$ | 4.32$^d$ | 4.32$^d$ | 0.01    |
|                 | LT63      | 4.14$^a$ | 4.15$^a$ | 4.34$^d$ | 4.44$^d$ | 0.04    |
|                 | LT63      | SEM     | 0.01 | 0.03 | 0.01 | 0.01    |
| Protein (%)     | Raw       | 3.36$^a$ | 3.53$^a$ | 3.85$^a$ | 4.56$^a$ | 0.17    |
|                 | LT56      | 3.27$^a$ | 3.46$^a$ | 3.84$^a$ | 4.47$^a$ | 0.17    |
|                 | LT63      | 3.36$^a$ | 3.45$^a$ | 3.84$^a$ | 4.25$^a$ | 0.13    |
|                 | SEM       | 0.01 | 0.02 | 0.02 | 0.01 | 0.01    |
| Lactose (%)     | Raw       | 4.60$^a$ | 4.54$^b$ | 4.41$^b$ | 4.31$^b$ | 0.04    |
|                 | LT56      | 4.54$^a$ | 4.46$^b$ | 4.40$^b$ | 4.34$^b$ | 0.03    |
|                 | LT63      | 4.56$^a$ | 4.53$^a$ | 4.41$^b$ | 4.27$^a$ | 0.04    |
|                 | SEM       | 0.02 | 0.02 | 0.01 | 0.01 | 0.01    |
| Non-fat solids (%) | Raw     | 8.06$^a$ | 8.22$^b$ | 8.48$^b$ | 9.32$^b$ | 0.17    |
|                 | LT56      | 7.95$^a$ | 8.12$^b$ | 8.43$^b$ | 9.09$^b$ | 0.16    |
|                 | LT63      | 8.03$^a$ | 8.14$^b$ | 8.42$^b$ | 8.85$^b$ | 0.12    |
|                 | SEM       | 0.03 | 0.02 | 0.01 | 0.01 | 0.01    |
| Total solids (%) | Raw       | 13.39$^a$ | 13.42$^a$ | 13.69$^a$ | 14.40$^a$ | 0.15    |
|                 | LT56      | 13.29$^a$ | 13.59$^a$ | 13.71$^a$ | 14.36$^a$ | 0.15    |
|                 | LT63      | 13.20$^a$ | 13.25$^a$ | 13.46$^a$ | 14.72$^a$ | 0.24    |
|                 | SEM       | 0.01 | 0.02 | 0.02 | 0.02 | 0.02    |

$^a$Means for the same variable in the same row with different superscript differ significantly ($P < 0.05$).

$^b$SEM: standard error of the mean.
mL\(^{-1}\)), *Staphylococcus aureus* (4.31 log cfu mL\(^{-1}\)), and coliform count (4.31 log cfu mL\(^{-1}\)) among others. In addition, Elizondo-Salazar et al. (2010) and Elizondo-Salazar et al. (2010) reported that disease-causing pathogens that may be transmitted to dairy calves through colostral secretions include *Mycobacterium avium subsp. paratuberculosis*, *Salmonella spp.*, *Mycoplasma spp.*, *Listeria monocytogenes*, *Campylobacter spp.*, *Mycobacterium bovis*, and *Escherichia coli* (also pathogenic bacteria relevant to human health risks). For this reason, and considering different types of microorganisms in colostrum, their presence in milk bulk tank can be a contamination factor that requires an adequate milk heat treatment to ensure its sanitation.

The IgG concentration was also significantly higher in colostrum (61.15 mg mL\(^{-1}\)) than in mature milk (1.32 mg mL\(^{-1}\)). Different authors have found that albumins and Iggs represent higher concentration (6 g L\(^{-1}\)) in colostrum than mature milk (0.5 y 0.09 g L\(^{-1}\), respectively) (Smolenski et al. 2007; Zhang et al. 2011), making up to 70–80% of the total protein in colostrum (Larson 2011).

These results reflect the biological differences between colostrum and mature milk, which could influence the technological characteristics of milk for cheese processing if colostrum is added to cheese milk, according to different authors (Rynne et al. 2004; McGrath et al. 2016).

**Physicochemical characteristics of milks containing colostrum**

The proximate composition of the milk containing 0%, 1%, 5% and 10% of colostrum, raw or pasteurized, is summarized in Table 2. As was expected, as colostrum increased in the milk, percentages of fat, protein, total solids, and non-fat solids were increased, while lactose contents decreased. These results are according to those found by other authors in transitional milk from colostrum to milk (Tsioulpas et al. 2007; Romero et al. 2014; Sánchez-Macías et al. 2014). According to Bogahawaththa et al. (2017), a high protein (mainly whey protein) and mineral contents might render milk unsuitable for food processing operations, such as ultra-high temperature processes, and incompatible with the requirements regarding milk composition for processing (Marnila and Korhonen 2002; Tsioulpas et al. 2007; Sánchez-Macías et al. 2014).

The physicochemical characteristics of raw and thermally treated milk samples containing colostrum concentrations up to 10% are detailed in Tables 3 and 4, respectively. As the concentration of colostrum was increased up to 10%, acidity and density also increased in milk. However, pH decreased significantly when the colostrum concentration in milk was 10%. These results are according to those found in transitional milk from colostrum to milk (Tsioulpas et al. 2007; Romero et al. 2014; Sánchez-Macías et al. 2014).

An important characteristic of milk is its buffering capacity, i.e. resistance to changes in pH on addition of acid or base. The salts in mature milk (especially soluble calcium phosphate, citrate, and bicarbonate) and acidic and basic amino acid side-chains of proteins are mainly responsible for the buffering capacity (Fox et al. 2015). The buffering capacity of milk was not affected by colostrum up to 10%. Ethanol stability values (near to 70% or higher) were not affected by colostrum or heat treatment.

After the heat treatment of milk containing 0%, 1%, 5%, or 10% of colostrum, density and pH values decreased in all cases, while acidity values increased. Pasteurization causes some change in pH due to the loss of CO\(_2\) and precipitation of calcium phosphate (Fox et al. 2015). Heat treatments can result in a decrease on pH due to the thermal oxidation of lactose to various organic acids, hydrolysis of organic phosphate, among others, which accounts for 50% and 30% of the pH decrease, respectively (Singh 2004). This fact could explain the acidity increase and pH decrease of milk after heat treatment in this work.

Literature shows that acid-base equilibria in milk are influenced by processing operations, such as pasteurization above 100°C (Fox et al. 2015). However, in this experiment, neither the milk buffering capacity nor the alcohol stability was affected by the heat treatment, possibly because the pasteurization temperatures were low (56°C and 63°C).

L* and a* values were similar in mature milk and colostrum samples, but the yellow index (b*) differed markedly between colostrum (24.42 ± 12.17) and milk (13.73 ± 2.00) (Table 1). Table 4 summarizes the instrumental colour parameters of milk samples containing 0%, 1%, 5% and 10% of colostrum. L* values were not affected by colostrum in milk or the pasteurization process. Red index values (greenish, negative values) were also not affected by colostrum presence; however, the values increased as pasteurization temperature was higher. The b* values of raw milk increased when the colostrum was added up to 5%. However, these differences due to colostrum disappeared when milk was pasteurized. On the other hand, the values of b* in raw milk and with 1% of colostrum were not affected by the pasteurization process, while this parameter decreased after LT56 and LT63 treatments in milks with 5% and 10% of colostrum. According to the criteria of Nedomová et al. (2017), when 1% of colostrum was added to milk, the difference in colour related to the raw milk was considered lightly perceptible, while adding 5–10% colostrum resulted in higher differences for raw milk (middle differences). On the other hand, colour differences decreased with the pasteurization process, and these differences were lower as pasteurization temperature increased.

The probable reason for differences in colour data can be the higher carotenoid content of colostrum than in mature milk (Kehoe et al. 2007), which varies from yellow to orange and deep red-orange as the concentration of carotenoids increases (Antone et al. 2015).

**Cheese-making properties of milk**

As colostrum exhibits some extreme physical properties, this could be an important issue for the dairy industry because it could affect milk processability when it is present in milk bulk tank, as suggested by Sánchez-Macías et al. (2014) and Tsioulpas et al. (2007). As reported in scientific literature, casein content in milk is double than in colostrum, while the latter has reduced proportions of α\(_s\)-caseins and elevated proportions of κ-casein (Sobczuk-Szul et al. 2013). Alteration of protein content and the proportion of the individual proteins...
in milk could alter the quality of the milk after heat treatment, and consequently the cheese-making properties, the effectiveness of pasteurization and the quality of the final product. However, according to the obtained results (Table 5), there were no statistical differences in the rennet coagulation properties for RCT, RCF, F30, F45 and F60 parameters between non-added colostrum milk and those containing 1% and 10% of colostrum treated at 56°C for 1 h or at 63°C for 30 min. Furthermore, CY values were higher in LT63 milk samples than in their LT56 counterparts, while CD values were lower, but there were no statistical differences for these two parameters when colostrum was present in milk up to 10%.

According to Singh and Fox (1985, 1986), heating at a pH value less than 6.7 results in a greater quantity of denatured whey proteins associated with casein micelles. During the rennet coagulation process, a weak curd structure and higher cheese yield could result from the incorporation of denatured whey proteins, mainly due to the interactions of these proteins with casein micelles after pasteurization (Singh and Waungana 2001). On the other hand, according to Bogahawaththa et al. (2017), the main whey proteins (IgG, β-lactoglobulin, α-lactalbumin and bovine serum albumin) are heat sensitive with various thermal denaturation extensions, depending on the

| Table 3. Physicochemical characteristic of raw or pasteurized milk containing 0%, 1%, 5% and 10% of colostrum treated at 56°C for 1 h (LT56) or at 63°C for 30 min (LT63). |
|---------------------------------|------------------|------------------|------------------|------------------|------------------|
| Item                           | Treatment        | 0%   | 1%   | 5%   | 10%   | SEM  |
| pH                             | Raw              | 6.63az| 6.63az| 6.62az| 6.59az| 0.01 |
|                                | LT56             | 6.55az| 6.54ay| 6.51zy| 6.48yz| 0.01 |
|                                | LT63             | 6.52ay| 6.50yz| 6.46yz| 6.46yz| 0.01 |
|                                | SEM              | 0.02 | 0.02 | 0.02 | 0.02  |      |
| Acidity (%)                    | Raw              | 0.17az| 0.19zy| 0.20zy| 0.22b  | 0.01 |
|                                | LT56             | 0.19zy| 0.20aby| 0.21aby| 0.22b  | <0.00 |
|                                | LT63             | 0.20zy| 0.21aby| 0.22b  | 0.24b  | 0.01 |
|                                | SEM              | <0.00| <0.00| <0.00| <0.00  |      |
| Density (g mL⁻¹)               | Raw              | 1.030az| 1.031az| 1.032abz| 1.033bz| <0.00 |
|                                | LT56             | 1.027ay| 1.028by| 1.029by| 1.029by| <0.00 |
|                                | LT63             | 1.026zy| 1.026bzy| 1.027bzy| 1.029bzy| <0.00 |
|                                | SEM              | <0.00| <0.00| <0.00| <0.00  |      |
| RyMeans for the same variable in the same column with different superscript differ significantly (P < 0.05). | 1SEM: Standard Error of the Mean. |

Table 4. Instrumental colour analysis of raw or pasteurized milk containing 0, 1, 5 and 10% of colostrum treated at 56°C for 1 h (LT56) and 63°C for 30 min (LT63). | Colourostrum concentration (%) |
|---------------------------------|------------------|------------------|------------------|------------------|------------------|
| Item                           | Treatment        | 0%   | 1%   | 5%   | 10%   | SEM  |
| Luminosity                      | Raw              | 86.38| 86.28| 86.60| 86.08| 0.79 |
|                                | LT56             | 86.42| 86.22| 86.29| 86.31| 0.64 |
|                                | LT63             | 86.59| 86.38| 86.32| 86.30| 0.68 |
|                                | SEM              | 0.86| 0.83| 0.82| 0.83  |      |
| Red index (a*)                 | Raw              | -4.28az| -4.64zy| -4.48z| -4.61f| 0.10 |
|                                | LT56             | -4.79y| -4.71yz| -4.91z| -4.73f| 0.08 |
|                                | LT63             | -4.90z| -4.93z| -5.00z| -4.91f| 0.07 |
|                                | SEM              | 0.14| 0.08| 0.13| 0.10  |      |
| Yellow index (b*)              | Raw              | 13.73z| 13.82z| 17.94yz| 18.16yz| 0.94 |
|                                | LT56             | 12.62| 11.80| 13.73y| 15.70z| 1.04 |
|                                | LT63             | 12.14| 11.45| 12.71f| 13.33z| 0.76 |
|                                | SEM              | 0.80| 1.06| 1.09| 1.36  |      |
| ΔE*                            | Raw              | 2.31ay| 4.72aby| 5.31byz| 5.10y| 0.59 |
|                                | LT56             | 1.31yz| 1.95yz| 3.66bzy| 3.32z| 0.32 |
|                                | LT63             | 1.21y| 0.78y| 1.56y| 0.18  |      |
|                                | SEM              | 0.44| 0.69| 1.21  |      |

Table 5. Cheese-making properties of raw or pasteurized milk containing 0, 1, 5 and 10% of colostrum treated at 56°C for 1 h (LT56) or at 63°C for 30 min (LT63). | Colourostrum concentration (%) |
|---------------------------------|------------------|------------------|------------------|------------------|------------------|
| Parameters                      | Treatment        | 0%   | 1%   | 5%   | 10%   | SEM  |
| RCT (min)                       | LT56             | 12.8 | 12.94| 12.00| 11.50| 0.68 |
|                                | LT63             | 13.53| 13.25| 12.11| 11.01| 1.15 |
|                                | SEM              | 0.52| 0.22| 0.08| 0.34  |      |
| RCAF (Δ%T/min)                  | LT56             | 1.03| 1.02| 1.06| 1.14| 0.06 |
|                                | LT63             | 1.04| 1.02| 1.14| 1.23| 0.10 |
|                                | SEM              | 0.01| 0.00| 0.06| 0.06  |      |
| F30 (%T)                       | LT56             | 10.43| 10.33| 10.66| 11.1| 0.35 |
|                                | LT63             | 10.84| 10.37| 10.52| 10.9| 0.25 |
|                                | SEM              | 0.29| 0.03| 0.10| 0.14| 0.06 |
| F45 (%T)                       | LT56             | 13.16| 13.02| 13.34| 13.77| 0.33 |
|                                | LT63             | 13.74| 13.1| 13.11| 13.34| 0.30 |
|                                | SEM              | 0.41| 0.05| 0.17| 0.31  |      |
| F60 (Δ%T)                      | LT56             | 14.0| 14.73| 14.98| 15.04| 0.37 |
|                                | LT63             | 15.64| 14.89| 14.83| 15.07| 0.37 |
|                                | SEM              | 0.59| 0.11| 0.12| 0.3  |      |
| CY (%)                         | LT56             | 22.60y| 22.83y| 24.41y| 25.68y| 1.44 |
|                                | LT63             | 35.10yz| 34.96yz| 34.29yz| 34.24yz| 0.45 |
|                                | SEM              | 8.84| 8.58| 6.99| 6.05  |      |
| CD (%)                         | LT56             | 20.22| 20.76| 21.06| 20.86| 0.36 |
|                                | LT63             | 16.60| 16.99| 16.98| 17.05| 0.21 |
|                                | SEM              | 2.56| 2.66| 2.88| 2.70| 0.11 |

a-cMeans for the same variable in the same row with different superscript differ significantly (P < 0.05).

SEM: Standard Error of the Mean.
nature of the heat treatment, the individual physicochemical characteristics of these proteins and the influence of the inter-protein interactions, generally through the interaction reactions between free sulphhydryl groups and disulphide bonds of proteins (Wijayanti et al. 2014; Bogahawaththa et al. 2017). Therefore, increasing the level of whey protein denaturation (by elevating the pasteurization temperature) could impair the rennet coagulation properties of milk when it is pasteurized above 72°C (Guinee 2021).

Pasteurization has either a positive influence (Salwa and Galal 2002; San Martín-González et al. 2007) or no effect on cheese yield (Drake et al. 1997). Cheese yield depends almost entirely on the moisture content of the cheese, fat and casein contents of milk, and the retention of each during cheese making (Guinee et al. 2007; De Marchi et al. 2008). It was expected that the intensity of heat treatment could affect the denaturation of whey proteins, which could be incorporated into cheese curd, resulting in a higher cheese yield when LT63 treatment was applied compared to LT56 treatment. The lower level of CD observed in curds from milk LT63-treated could be explained by the higher water retention of these curds due to the incorporation of a high extent of denatured whey proteins (Guinee 2021) compared to curds from milk LT56-treated.

Microbiological characteristics

Table 6 shows the mean values of TPC of each milk sample and the pasteurization effectiveness through the bacterial reduction in percentage. TPC in raw milk was 3.67 log cfu mL\(^{-1}\). As colostrum concentration was higher in raw milk, TPC was increasing, and it was statistically different when 5% of colostrum was included in milk (4.41 log cfu mL\(^{-1}\)). Both thermal treatments had similar bacterial reduction (99.81–99.90% of reduction) on raw milk (0% colostrum addition). However, the effectiveness of these discontinuous heat treatments diminished when 1, 5 or 10% of colostrum was included in milk (from 99.9% to 94.59% in LT56; and from 99.81% to 98.88% in LT63, when 10% of colostrum was added). These values are similar to others reported in the literature for LTLT making (Guinee et al. 2007; De Marchi et al. 2008). It was expected that the intensity of heat treatment could affect the fermentative components in colostrum could alter the fermentation process (Marnila and Korhonen 2002; McGrath et al. 2016), or even the residual microbial content during the life of the products during storage. For this reason, it is necessary to maintain good hygiene and control, both udders and during the milking process, to reduce the contamination of this pathogen.

Because of these results, when the dairy industry suspects milk fraud with colostrum or when transitional milk is used, the conventional LTLT pasteurization process (63°C for 30 min) must be used to ensure the safety of their products. Various researchers (Boor and Murphy 2002; Barbano et al. 2006) have shown the poor quality of finished dairy products due to a higher bacteria content in raw milk. Current traditional methods of pasteurization have been effective in reducing bacterial load by as much as 5.0 log cfu mL\(^{-1}\) (Guan et al. 2005).

It is also necessary to mention that the high content of antimicrobial components in colostrum could affect the fermentation process (Marnila and Korhonen 2002; McGrath et al. 2016), or even the residual microbial content during the life of the products during storage. For this reason, it is necessary to

| Table 6. Means values of total bacterial counts (log cfu mL\(^{-1}\)) and bacterial reduction (%) of raw or pasteurized milk containing 0%, 1%, 5% and 10% of colostrum treated at 56°C for 1 h (LT56) or at 63°C for 30 min (LT63). |
| Treatment | Colostrum concentration (%) | SEM\(^1\) |
|------------|-----------------------------|-----------|
| Raw        | 3.67\(^{az}\) | 3.75\(^{az}\) | 4.01\(^{ab}\) | 4.41\(^{ac}\) | 0.09 |
| LT56       | 0.84\(^{ay}\) | 2.06\(^{by}\) | 2.77\(^{by}\) | 3.12\(^{by}\) | 0.31 |
| LT63       | 0.52\(^{az}\) | 1.31\(^{ax}\) | 2.00\(^{bx}\) | 2.32\(^{bx}\) | 0.22 |
| SEM        | 0.51 | 0.36 | 0.51 | 0.33 |
| Bacterial reduction (%) | LT56 | 99.90\(^{*}\) | 97.92\(^{az}\) | 95.28\(^{ab}\) | 94.59\(^{bx}\) | 0.08 |
| LT63       | 99.81\(^{*}\) | 99.63\(^{ay}\) | 98.69\(^{by}\) | 98.88\(^{by}\) | 0.09 |
| SEM        | 0.08 | 0.74 | 0.08 | 0.68 |

\(^{a,b}\) Means for the same variable in the same row with different superscript differ significantly (P < 0.05).

\(^{a,b}\) Means for the same variable in the same column with different superscript differ significantly (P < 0.05).

\(^{1}\) SEM: Standard Error of the Mean.

Figure 1. Microbial reduction (log cfu mL\(^{-1}\)) of milk containing 0%, 1%, 5% and 10% of colostrum after heat treatment at 56°C for 1 h (LT56) or at 63°C for 30 min (LT63).
further studies in the future to evaluate the presence of colostrum in milk on processed products during different storage periods.

**Conclusions**

The presence of colostrum up to 10% can modify the proximate composition and physicochemical characteristics of milk, which could impair the milk’s ability to be processed in the cheese industry. Cheese-making properties of milk are not affected by colostrum presence up to 10%, contrary to the expected. However, the heat treatment at 63°C for 30 min results in milk with higher values for CY and lower CD values, respectively, when compared with milk heat-treated at 56°C for 60.

Microbiological analysis results show the effectiveness of the two discontinuous thermal treatments (63°C for 30 min and 56°C for 60 min) reducing the TPC used in milk processing for the cheese industry. However, the first heat treatment proves to be more efficient for reducing the TPC than the second when colostrum is present in milk as low as 1%.

Because of these results, the conventional HTLT treatment of 63°C for 30 min is recommended when the presence of colostrum is suspected to ensure the safety of dairy products. However, it is necessary for further experiments in the future to evaluate the presence of colostrum on processed products during their storage periods.

As a recommendation for milk producers, the incidence of colostrum in milk bulk tank may be minimized by adopting appropriate operation and maintenance of milking equipment and contemporary dairy husbandry practices.

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**ORCID**

Byron Herrera-Chávez [http://orcid.org/0000-0003-1116-9939](http://orcid.org/0000-0003-1116-9939)
Antonio José Trujillo [http://orcid.org/0000-0003-1437-6060](http://orcid.org/0000-0003-1437-6060)
Davinia Sánchez-Macias [http://orcid.org/0000-0001-5626-4249](http://orcid.org/0000-0001-5626-4249)

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