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Development and shelf-life determination of pasteurized, microfiltered, lactose hydrolyzed skim milk

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ABSTRACT

The segment of the world population showing permanent or temporary lactose intolerance is quite significant. Because milk is a widely consumed food with an high nutritional value, technological alternatives have been sought to overcome this dilemma. Microfiltration combined with pasteurization can not only extend the shelf life of milk but can also maintain the sensory, functional, and nutritional properties of the product. This studied developed a pasteurized, microfiltered, lactose hydrolyzed (delactosed) skim milk (PMLHSM). Hydrolysis was performed using β-galactosidase at a concentration of 0.4 mL/L and incubation for approximately 21 h at 10 ± 1°C. During these procedures, the degree of hydrolysis obtained (>90%) was accompanied by evaluation of freezing point depression, and the remaining quantity of lactose was confirmed by HPLC. Milk was processed using a microfiltration pilot unit equipped with uniform transmembrane pressure (UTP) ceramic membranes with a mean pore size of 1.4 μm and UTP of 60 kPa. The product was submitted to physicochemical, microbiological, and sensory evaluations, and its shelf life was estimated. Microfiltration reduced the aerobic mesophilic count by more than 4 log cycles. We were able to produce high-quality PMLHSM with a shelf life of 21 to 27 d when stored at 5 ± 1°C in terms of sensory analysis and proteolysis index and a shelf life of 50 d in regard to total aerobic mesophile count and titratable acidity.
Key words: lactose hydrolyzed milk, microfiltration, shelf life

INTRODUCTION

It has been estimated that at least 65% of the adult world population manifests signs and symptoms of lactose intolerance, and the prevalence of this inability to digest lactose varies considerably between different races and age ranges. The distribution of different lactase phenotypes in human populations is highly variable, an observation that has long been a source of interest in relation to evolutionary genetics. Family studies suggest that adults who are lactase nonpersistent (lactose intolerant) are homozygous for an autosomal recessive allele that causes the postweaning decline in lactase activity, whereas people who are lactase persistent (lactose tolerant) are either heterozygous or homozygous for a dominant allele that allows lactase to persist (Swallow, 2003).

The recommendation for individuals with hypolactasia or alactasia is the reduction or exclusion of lactose-containing foods from the diet (Rusynyk and Still, 2001). However, with the exclusion of dairy products from the diet, lactose-intolerant individuals generally show low ingestion of calcium and other nutrients provided by milk (Batista et al., 2008). One option in these cases is the consumption of lactose hydrolyzed products. The conversion of lactose to its constituent monosaccharides by hydrolysis has been practiced industrially for almost 20 yr and recent research has focused on the determination of residual lactose in delactosed milk by calculating its freezing point (Colinas et al., 2006). Lactose hydrolyzed milk is milk with a reduced lactose content (normally reduced by 90%), this reduction being obtained by the action of the enzyme β-galactosidase, which promotes enzymatic hydrolysis of the lactose. Although lactose-free milks have addressed the needs of lactose-intolerant consumers, such products need to have strong similarity to regular milk for the consumer to purchase and be satisfied with the products (Adhikari et al., 2010).

One significant barrier to extending the shelf life of dairy products is the difficulty in achieving the removal or destruction of spoilage microorganisms and spores present in raw milk while limiting product color changes, vitamin destruction, and milk protein denaturation (García and Rodríguez, 2014). The application

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of a combination of pasteurization and microfiltration to milk has the potential to yield high-quality milk with an extended shelf life (Schmidt et al., 2012) and it improves the keeping quality of cheese made from the milk due to removal of the spores (Pafylias et al., 1996). Microfiltration is the technology used to separate the components of a liquid medium, the separation occurring when the medium is forced to flow, under low pressure (~100 kPa), over the surface of a semipermeable membrane with pore sizes ranging from 0.2 to 5 μm (Dziezak, 1990; Fernández García et al., 2013). This technique can be used to reduce the microbial load of milk by mechanical separation, without causing heat-induced chemical alterations, thus conserving the sensory, functional, and nutritional properties of the milk (Hoffmann et al., 2006). Bacteria generally range from 0.4 to 2 μm in size; therefore, under certain circumstances, microfiltration should be able to completely remove bacteria from the fluid permeate (Fernández García et al., 2013). Madec et al (1992) observed decimal reductions in milk close to 1.9 units for *Listeria* and 2.5 units for *Salmonella* using a multichannel microfiltration membrane with a 1.4-μm pore size. In milk processing, a pore size of about 1.4 μm is normally used to achieve the right balance between rejection of bacteria and little or no rejection of milk nutrients (García and Rodríguez, 2014).

Pasteurized, microfiltered, lactose hydrolyzed skim milk (of which the microbiological, physicochemical, and sensory characteristics are presented in this paper) is an innovative product, and scientific literature on this type of product is scarce. However, milk processed using microfiltration (**MF**) and HTST (72°C for 15s) pasteurization (without lactose hydrolysis) is commercially available in Canada and northern European countries, it has a mean shelf life of 35 d at 6°C (Saboya and Maubois, 2000; Elwell and Barbano, 2006; Mintel, 2013).

Thus, the objectives of the present work were to (1) develop a pasteurized, microfiltered, lactose hydrolyzed skim milk, (2) evaluate its physicochemical, microbiological and sensory characteristics, and (3) estimate its shelf life.

MATERIALS AND METHODS

Milk Samples and Enzyme

About 240 L of skim milk produced on only one farm was pasteurized at 72 to 75°C for 15 to 20 s and bottled on the farm premises according to Brasil (2011). β-Galactosidase (EC 3.2.1.23) isolated from *Kluyveromyces lactis* (50,000 U/mL) was donated by Prozyn SP/Brazil (Sao Paulo, Brazil).

Product Processing and Storage

Two batches of the pasteurized, microfiltered, lactose hydrolyzed skim milk (**PMLHSM**) were produced, each using 120 L of milk. The final products were stored for 60 d at 5 ± 1°C and evaluated for their microbiological, physicochemical, and sensory parameters every 7 d.

Hydrolysis of Milk Lactose

For each processing, β-galactosidase was initially added to the pasteurized milk at a concentration of 0.4 mL/L, and incubated for about 21 h at 10 ± 1°C (lactose hydrolysis condition selected in preliminary tests). During these procedures, the degree of hydrolysis obtained (above 90%) was accompanied by measurement of freezing point depression, according to Ramet et al. (1979) and the remaining quantity of lactose was confirmed by HPLC according to the methodology proposed by Burgner and Feinberg (1992). The detection limit of the HPLC method was 0.2 g of lactose per 100 mL of milk.

Microfiltration of the Milk

After hydrolysis of the lactose, the milk was submitted to MF in a microfiltration MFS-1 pilot unit (Tetra Laval, Paris, France), equipped with uniform transmembrane pressure (**UTP**) ceramic membranes (Membralox, Societe des Céramiques, Bazet, France). The MFS-1 unit was equipped with a 1P19-40 filter module containing a ceramic element with 0.24-m^2 membrane area, which allows a capacity of approximately 150 L of skim milk/h, and with a mean pore size of 1.4 μm. The parameters of the process were as follows: permeate flux of 120 L/h, retentate flux of 6.3 L/h, volumetric concentration factor (VCF) of 20, and temperature of 48 ± 1°C. To minimize membrane fouling, a UTP of 60 kPa was used. Sterile 1-L glass bottles were filled with the MF milk using an automatic doser inside a laminar flow chamber.

Microbiological Analyses of the Milk After MF and During Storage

Before and immediately after MF, counts were made of total mesophilic aerobes, total psychrotrophic aerobes, coliforms at 30°C and 45°C, coagulase-positive staphylococci, *Salmonella* spp., and yeasts and molds. The following microbiological analyses were carried out every 7 d: total mesophilic count, coliforms at 30°C, coliforms at 45°C, and yeasts and molds.

The procedures recommended by the American Public Health Association (Wehr and Frank, 2004) were followed for decimal dilutions and for microbiological analyses of milk samples. Total aerobic mesophilic count was done in plate count agar (Difco, Detroit, MI) with triphenyltetrazol chloride (Merck, Whitehouse Station, NJ) incubated at 32 ± 1°C for 48 h (Frank and Yousef, 2004). Coliforms at 30°C and 45°C were determined using the most-probable-number (**MPN**) method with lauryl sulfate tryptose (Acumedia, Sao Paulo, Brazil) broth and brilliant green bile lactose broth (Difco), incubated at 35 ± 1°C for 24 to 48 h for coliforms at 30°C (ISO, 2006; ISO 4831), and EC broth (*Escherichia coli*) incubated at 44 ± 1°C for 24 h (ISO, 2005; ISO 7251) for heat-tolerant coliforms. The yeast and mold count was done in chloramphenicol bengal rose dichloran agar (Difco), incubated at 25 ± 1°C for 5 d (ISO-IDF, 2004; ISO 6611-IDF 94). The psychrotrophic aerobes were counted in plate count agar (Difco) incubated at 7 ± 1°C for 7 d (Frank and Yousef, 2004). The presence of coagulase-positive staphylococci was detected using Baird Parker agar (Difco), incubated at 36 ± 1°C for 48 h according to Henning et al. (2004). Typical colonies were confirmed using coagulase, thermonuclease, catalase, and Gram tests. The presence of *Salmonella* was detected according to Henning et al. (2004) using a pre-enrichment medium, followed by a tetrathionate broth selective enrichment medium (Difco) and cystine selenite broth (Oxoid, Basingstoke, UK). The media were incubated at 35°C for 24 h. Differential plating was done in Hektoen enteric agar (Difco), bismuth sulfate agar (Difco), and desoxycholate lysine xylose agar (Difco). The media were incubated at 35°C for 24 h and evaluated for the occurrence of a typical reaction for *Salmonella* spp. A small amount of cell mass from one colony was inoculated into lysine iron agar (Difco) and into triple sugar iron agar (Difco). The inoculated media were incubated at 35°C for 24 h and evaluated for the presence of typical *Salmonella* spp. colonies. The following tests were applied to typical colonies: serological test, urease, dulcitol fermentation, malonate test, and indol test. The results of the microbial counts were expressed in log colony-forming units per milliliter, with the exception of coliforms, which were expressed in MPN per milliliter.

Physicochemical Analyses of PMLHSM After Processing and During Storage

The following were determined on d 1 of storage: pH, titratable acidity, TS, fat, nonfat solids (**NFS**), total nitrogen (**TN**), noncasein nitrogen (**NCN**), NPN, total protein, ash, and the freezing point of the PMLHSM. The pH was measured using a digital pH meter (Micronal B-375, Micronal SA, Santo Amaro, Brazil). Density and total titratable acidity were determined according to the standard methods of the Adolfo Lutz Institute (2005). The percentages of TS and fat (Gerber method) and the depression of the freezing point were determined according to the methodologies in Brasil (2006). The NFS content was obtained from the relationship NFS = (TS − fat). The fixed mineral residue was obtained according to AOAC International (2000; method 985.35). The TN content was obtained using the Kjeldahl method (AOAC International, 1995), and the total protein content calculated using a conversion factor of 6.38. The NCN content was obtained by measuring the TN in the supernatant after isoelectric precipitation of the caseins (AOAC International, 1995). The NPN content of the samples was determined by dosing the TN in the supernatant after the total precipitation of the proteins in the presence of 12% TCA as described by Aschaffenburg and Drewry (1959).

During the 60 d of storage, the PMLHSM were evaluated weekly for their titratable acidity and proteolysis values. The proteolysis index (**PI**) was used to evaluate proteolysis. This index corresponds to the decrease in casein (CN) as a percentage of the true protein (**TP**) (CN%TP), where

$$CN = (TN - NCN) \times 6.38, \text{ and}$$

$$TP = (TN - NPN) \times 6.38,$$

where the values for TN, NPN, and NCN were obtained as described above.

Sensory Test for Storage Stability

The difference from the control test was used to determine the stability of PMLHSM during refrigerated storage, according to Meilgaard et al. (2006). The sample used as the control corresponded to pasteurized lactose hydrolyzed skim milk obtained weekly by the addition of β-galactosidase (incubated for 21 h at 10°C ± 1) to commercial skim milk pasteurized (but not microfiltered). The PMLHSM was compared with the control to evaluate the effect of storage. After processing, sensory tests were applied after 7, 14, 21, 28, 35, 43, and 50 d of storage. The MF milk samples were maintained under refrigeration and the sensory test applied with respect to the attributes taste, astringency, bitterness, and acidity. These evaluations were carried out by a panel of 24 judges selected for their sensory acuity, using the following difference from control scales: (a) color and sweetness: 7-point scale, where 7 = much more intense/sweet than control; 4 = equal to control; 1 = much less intense/sweet than control; (b) taste:

7-point scale, where 7 = better than control; 6 = equal to control; 5 to 1 = inferior to control, with increasing levels of the intensity of off-flavors (no off-flavor to very strong off-flavor); (c) astringency, bitterness, and acidity: 6-point scales, where 6 = less astringent/bitter/acid than control; 5 = equal to control; 4 to 1 = increasing intensities of astringency/bitterness/acidity (from slightly to extremely more astringent/bitter/acid than control).

The research plan was approved by the Research Ethics Committee [protocol 223/2007, Pontifícia Universidade Católica de Campinas (PUCCamp), Campinas, Brazil].

Statistical Analysis

For the sensory tests, judgments were submitted to ANOVA and means were compared by the Dunnett test, which is used for comparing samples to a standard (ABNT, 1995).

RESULTS AND DISCUSSION

Microbiological Analyses

Microbiological analyses were carried out on control and PMLHSM milks just after MF for the 2 processing procedures. After MF, we observed a reduction in count of aerobic mesophiles of 4 log cycles (from 4.23 to >1 log cfu/mL; Table 1). Elwell and Barbano (2006) reported an average 3.79 log reduction by microfiltration of milk, and García and Rodríguez (2014) reported that microfiltration led to a logarithmic reduction in bacteria of 4.

Counts below detection limits were recorded for total aerobic psychrotrophs, yeasts and molds, coliforms at 30°C, coliforms at 45°C, and coagulase-positive staphylococci before and after MF; *Salmonella* spp. were absent (Table 1).

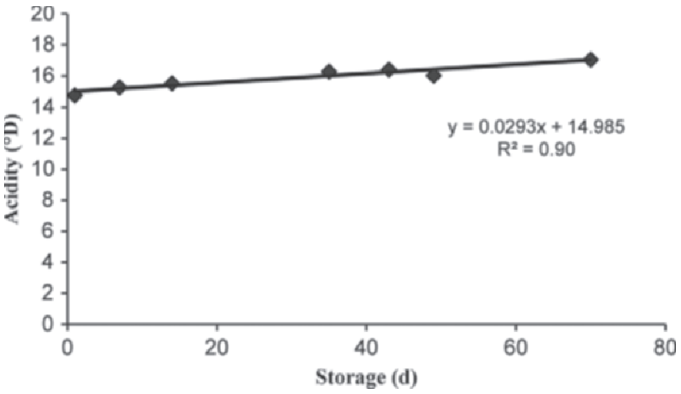

Figure 1. Mean values (n = 2) for titratable acidity (°Dornic, °D) obtained for the pasteurized, microfiltered, lactose hydrolyzed skim milk samples during refrigerated storage.

Until 50 d of storage, the PMLHSM samples showed counts for total plate counts (total aerobic mesophiles; Table 2) that complied with microbiological standards for pasteurized milk in the literature. Elwell and Barbano (2006) used an aerobic mesophilic bacterial count of >20,000 cfu/mL (corresponding to 4.30 log cfu/mL) as the criterion to determine the end of shelf life of microfiltered milk. They estimated the shelf life of the product as a function of storage temperature (0.1, 2.0, 4.2, and 6.1°C), varying from 16 d (at 6.1°C) to 66 d (at 0.1°C). García and Rodríguez (2014) used an aerobic mesophilic bacterial count of >30,000 cfu/mL (corresponding to 4.47 log cfu/mL) as the end of milk shelf life. They used a combination of MF and subsequent pasteurization treatment (73°C for 15 s) and obtained extended-shelf-life milk with a shelf life close to 30 d (70% longer than regular pasteurized milk). Based on this criterion, the PMLHSM developed in the current study maintained its aerobic mesophilic bacterial count below 4.30 log cfu/ for up to 50 d of storage at 5 ± 1°C.

Counts below the detection limits were observed for coliforms and yeasts and molds for up to 57 d of

Table 1. Mean values (n = 2) for the microbiological analyses of the pasteurized skim milks before and after microfiltration for the 2 processing procedures

Microorganism group	Pasteurized skim milk	Pasteurized microfiltered skim milk
Total aerobic mesophiles (log cfu/mL)	4.23	<1
Total aerobic psychrotrophs (log cfu/mL)	<1	<1
Coliforms at 30°C (MPN[1]/mL)	<0.3	<0.3
Coliforms at 45°C (MPN/mL)	<0.3	<0.3
Salmonella spp. (log cfu/mL)	Absent[2]	Absent
Coagulase-positive staphylococci (log cfu/mL)	<1	<1
Yeasts and molds (log cfu/mL)	<1	<1

[1]Most probable number.

[2]Absent in 25 mL.

Table 2. Microbial counts (mean ± SD) for batches (n = 2) of pasteurized, microfiltered, lactose hydrolyzed skim milk (PMLHSM) during storage at 5 ± 1°C

	Refrigerated storage time (d)								
Microorganism group	0	7	14	21	28	35	43	50	57
Total aerobic mesophiles (log cfu/mL)	<1	<1	<1	<1	<1	1.77 ± 0.7	2.59 ± 0.9	3.91 ± 2.9	6.87 ± 0.8
Coliforms at 30°C (MPN[1]/mL)	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
Coliforms at 45°C (MPN/mL)	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
Yeasts and molds (log cfu/mL)	<1	<1	<1	<1	<1	<1	<1	<1	<1

[1]Most probable number.

refrigerated storage. These results indicate good microbiological stability of the PMLHSM samples, which was reinforced by the maintenance of values for acidity similar to that of just-pasteurized milk throughout the whole storage period (Figure 1).

Physicochemical Composition of PMLHSM on Day 1 of Storage

Table 3 shows the mean values obtained (n = 3) for the physicochemical composition of the PMLHSM samples on d 1 of refrigerated storage. The results in Table 3 showed that the PMLHSM samples complied with the physical and chemical requisites established in the literature for pasteurized skim milk (Walstra et al., 1999; Brasil, 2011), except for lactose, which was present at a very low concentration (<0.2 g/100 mL) due to hydrolysis. Before the MF step, the batches used in the 2 processing procedures had a fat content of 0.5%. However, MF removed 100% of the fat because most fat globules in milk are similar in size to bacteria, according to García and Rodríguez (2014).

Cryoscopy is a measurement of the freezing point of milk related to the substances dissolved in it; that is, mainly lactose and mineral salts. The freezing point depression observed in milk depends mainly on the lactose and mineral salts present in solution; fats and proteins do not significantly influence this property (Colinas et al., 2006). When lactose is hydrolyzed, the number of free molecules increases, thus increasing the amount of substances dissolved in the milk, which alters (decreases) the freezing point. This can be observed in Table 3, where the mean freezing point index of the hydrolyzed lactose milk was much smaller (−0.805 ± 0.002°H, where °H are degrees Hortvet, a modification of degrees Celsius) than that reported (Brasil, 2011) for nonhydrolyzed milk, which ranges from −0.530 to −0.550°H.

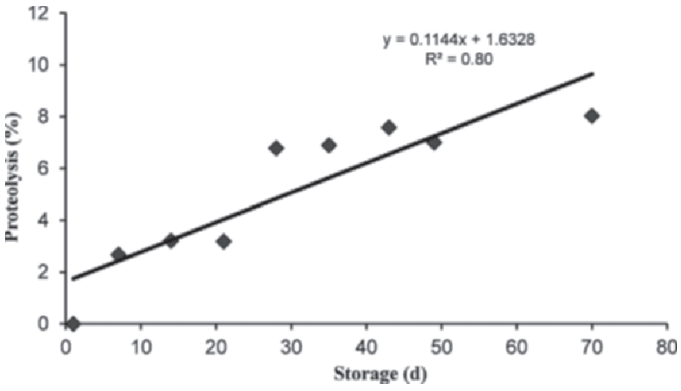

Figure 2. Mean values (n = 2) for proteolysis index (%) obtained for the pasteurized, microfiltered, lactose hydrolyzed skim milk samples during refrigerated storage.

Physicochemical Composition of PMLHSM During Refrigerated Storage

Figures 1 and 2 show the mean results (n = 2) obtained in evaluations of titratable acidity and PI, respectively, during refrigerated storage.

The results obtained for titratable acidity were within Brazilian limits for pasteurized milk (14 to 18°D, where °D are degrees Dornic; Brasil, 2011). Titratable acidity did not represent a determinant factor in the sensory rejection of PMLHSM samples during 60 d of refrigerated storage.

Santos et al. (2003) produced pasteurized MF milk and determined that the end of the useful life of this product corresponded to a decrease in the percentage of casein in relation to the total true protein equal or greater to 4.76% (PI ≥4.76). This level was established

Table 3. Mean (n = 2) physicochemical composition of the pasteurized, microfiltered, lactose hydrolyzed skim milk samples on d 1 of storage

Composition	Mean ± SD
Density (g/mL)	1.032 ± 0.000
pH	6.76 ± 0.02
Titratable acidity (°Dornic)	14.76 ± 0.00
Total solids (%)	8.56 ± 0.06
Fat (%)	0.00 ± 0.00
Nonfat solids (%)	8.56 ± 0.06
Lactose (g/100 mL)	<0.2
Ash (%)	0.71 ± 0.01
Total protein (%)	3.28 ± 0.01
Freezing point (°H)[1]	−0.805 ± 0.002

[1]°H = degrees Hortvet, a modification of degrees Celsius.

Table 4. Mean values obtained in the evaluations of pasteurized, microfiltered, lactose hydrolyzed skim milk (PMLHSM) with respect to color, sweetness, taste, astringency, bitterness, and acidity (at 7 d) and for flavor, astringency, bitterness, and acidity (the other periods) in relation to the control sample

Evaluation time	Attribute	Sample		MSD[2]
		Control[1]	PMLHSM	
7 d	Color	4.2 (0.5)[a]	4.2 (1.0)[a]	0.43
	Sweetness	3.9 (0.6)[a]	4.3 (0.9)[a]	0.71
	Taste	6.0 (0.6)[a]	5.0 (1.6)[b]	0.83
	Astringency	4.9 (0.3)[a]	4.6 (0.8)[a]	0.50
	Bitterness	4.9 (0.3)[a]	4.9 (0.6)[a]	0.45
	Acidity	5.0 (0.2)[a]	4.9 (0.4)[a]	0.45
14 d	Taste	6.0 (0.7)[a]	4.8 (1.5)[b]	0.79
	Astringency	5.0 (0.6)[a]	4.5 (0.8)[a]	0.54
	Bitterness	5.0 (0.7)[a]	4.5 (0.8)[a]	0.49
	Acidity	5.1 (0.5)[a]	4.8 (0.6)[a]	0.39
21 d	Taste	5.9 (0.6)[a]	3.0 (1.4)[b]	0.76
	Astringency	4.9 (0.3)[a]	3.9 (0.7)[b]	0.41
	Bitterness	4.9 (0.3)[a]	4.1 (1.0)[b]	0.52
	Acidity	4.9 (0.3)[a]	3.6 (1.1)[b]	0.57
28 d	Taste	6.1 (0.5)[a]	4.6 (1.2)[b]	0.66
	Astringency	5.1 (0.4)[a]	4.7 (0.8)[b]	0.34
	Bitterness	5.1 (0.3)[a]	4.6 (0.8)[b]	0.42
	Acidity	5.1 (0.4)[a]	4.7 (0.8)[b]	0.37
35 d	Taste	6.2 (0.5)[a]	4.1 (1.6)[b]	0.8
	Astringency	5.1 (0.4)[a]	4.3 (1.2)[b]	0.65
	Bitterness	5.1 (0.4)[a]	4.5 (1.0)[b]	0.51
	Acidity	5.1 (0.3)[a]	4.6 (0.8)[b]	0.38
43 d	Taste	6.2 (0.5)[a]	4.9 (1.1)[b]	0.65
	Astringency	5.1 (0.5)[a]	4.7 (0.6)[b]	0.37
	Bitterness	5.1 (0.3)[a]	4.6 (0.6)[b]	0.38
	Acidity	5.1 (0.2)[a]	4.7 (0.7)[b]	0.41
50 d	Taste	5.9 (0.5)[a]	4.9 (1.8)[b]	0.89
	Astringency	4.9 (0.3)[a]	4.5 (1.1)[a]	0.51
	Bitterness	4.9 (0.4)[a]	4.8 (1.0)[a]	0.55
	Acidity	4.9 (0.4)[a]	4.7 (1.0)[a]	0.48

[a,b]For each period and attribute (row), means followed by letters different from that of the codified control differ significantly from the control at a 5% error level.

[1]Commercial pasteurized (but not microfiltered) skim milk obtained weekly with β-galactose added to hydrolyze the lactose. Values are expressed as the mean (SD in parentheses) of the correct judgments.

[2]MSD = minimum significant difference by the Dunnett test at a 5% error level.

because it was the point at which flavor defects (bitterness and astringency) caused by proteolysis were detected by 50% of the sensory panel. By adopting the end of the useful life as being when the PI was 4.76, the shelf life of the samples of PMLHSM in the current study would be approximately 27 d at 5 ± 1°C. This useful life was close to that obtained by Elwell and Barbano (2006), whose samples of MF pasteurized skim milk, stored at 6.1°C, reached a PI of 4.76 after 32 d of storage.

Sensory Analysis

For each period evaluated, Table 4 shows the mean values obtained from the correct judgments; that is, those where the control was correctly identified in the sensory evaluations of the samples. Panelists found no difference between control samples and PMLHSM samples for the attributes color, sweetness, astringency, bitterness, or acidity when samples were evaluated 7 d after processing the PMLHSM. Similarly, after 14 d of storage, control samples and PMLHSM samples did not differ in astringency, bitterness, or acidity. These samples only differed significantly for taste.

From d 21 of storage, PMLHSM samples differed ($P < 0.05$) from control samples for all attributes. However, the PMLHSM samples presented means close to "slightly more astringent, bitter, and acid than controls" during the 50 d of storage for which sensory evaluations were made, which is not indicative of important sensory alterations during the evaluation period. These observations were reinforced by the means obtained for astringency, bitterness, and acidity of the PMLHSM samples, which, after 50 d of storage, did not differ from the control at the 5% error level.

The results obtained in the sensory evaluations showed that none of the main sensory attributes of PMLHSM were compromised compared with the control, which is important for good acceptance of extended-shelf-life delactosed milk.

Lactose-free or lactose hydrolyzed milk is normally sold as an ultrapasteurized (**UP**) product, as can be seen in Adhikari et al. (2010). In that work, after conducting a descriptive analysis of UP lactose-free milks and regular commercial milks (skim, reduced-fat, and whole milks), the authors concluded that UP lactose-free milks had higher intensities of 4 negative attributes: chalky texture, lack of freshness flavor (change in the overall rounded dairy notes, commonly associated with fresh milk), light oxidized flavor, and processed flavor. The authors emphasized that these differences may be caused by the UP process, by the enzymatic reactions that result in lactose-free milk, or by a combination of these factors. In lactose-free milks, the increase in the amount of reducing monosaccharides (glucose and galactose), which are more reactive in Maillard reactions, may affect the color and lack of freshness of the product.

To better understand the influence of MF and lactose hydrolysis processes on milk characteristics, further sensory studies should be performed. Examples of these studies were presented by Pimentel et al. (2013) and Cadena et al. (2012). Those authors applied methods of sensory profiling and consumer tests for dairy products.

CONCLUSIONS

It is possible to obtain PMLHSM with a shelf life, in terms of microbiological stability and acidity, of up to 50 d at 5°C. Microfiltration technology, in combination with pasteurization, could be used by the dairy industry to produce extended-shelf-life delactosed milk. In the sensory evaluations, PMLHSM samples not differ, at least in the initial stages of storage, from freshly pasteurized control samples of lactose hydrolyzed skim milk. From d 21 of storage, the PMLHSM sample differed significantly from the control with respect to taste, astringency, bitterness, and acidity. However, during 50 d of storage, the PMLHSM samples were described by sensory panelists as only slightly more astringent, bitter, and acid than controls, indicating that sensory changes during the evaluation period were not extensive. These variations in sensory analysis may be due to inherent characteristics of the milk used in manufacture and may be unrelated to treatment (MF); microbial counts remained low during 50 d of refrigerated storage.

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