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The effects of inulin combined with oligofructose and goat cheese whey on the physicochemical properties and sensory acceptance of a probiotic chocolate goat dairy beverage

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ABSTRACT

The aim of this study was to produce chocolate goat dairy beverages with the probiotic *Bifidobacterium lactis* and to evaluate the effects of goat cheese whey and prebiotics (inulin and oligofructose) on the physicochemical parameters and sensory features of the beverages. All of the formulations ($n = 7$) exhibited decreased pH values and a concomitant increase in acidity during refrigerated storage. Beverages made with the lowest amounts of whey (F1 and F3) exhibited a greater decrease in pH after 14 days of storage. The apparent viscosity increased for up to 21 days for all formulations and up to 28 days for F4 (6 g 100 mL^{-1} prebiotics and 45 mL 100 mL^{-1} whey). *B. lactis* exhibited counts between 6 and 8 log CFU mL^{-1}. F4 presented the highest median sensory attributes for flavor and aroma, which may be related to the larger amounts of prebiotics and whey in this formulation. Thus, F4 is considered to be the formulation that best represents the desirability profile chosen for the probiotic chocolate goat dairy beverage as defined as probiotic viability above 7 log CFU mL^{-1} and improved viscosity and sensory features.

1. Introduction

The consumption of foods such as probiotics and prebiotics that promote wellness, health, and a reduced risk of diseases has grown worldwide. During the past decade, more than 500 new products were introduced to the market (Ashraf & Shah, 2011). Among probiotic microorganisms, bifidobacteria have primarily been used in bovine dairy products, especially in fermented milks, yogurts and dairy beverages (Castro et al., 2013; Ranadheera, Evans, Adams, & Baines, 2013a). Bifidobacteria have low viability at pH values below 4.0 (Saarela, Alakomi, Mättö, Ahonen, & Tynkkynen, 2011), and their multiplication can be affected by oxygen and hydrogen peroxide (Roy, 2005). Therefore, one strategy for promoting the high viability of these bacteria in products would involve the use of a separately fermented inoculum containing a high number of viable cells before incorporating it into milk formulations (Kailasapathy & Rybka, 1997).

Dairy products can help bifidobacteria survive in gastric juice because of their buffering effect. Studies involving *Bifidobacterium lactis* species have demonstrated excellent viability maintenance in fermented milk until the time of consumption (Gueimonde et al., 2004; Ross, Desmond, & Stanton, 2005). The maintenance of *B. lactis* viability in dairy products may be improved by adding prebiotic ingredients such as inulin and oligosaccharides, which have bifidogenic properties and do not interfere with the flavor of the final product (Roberfroid, 2007).

Dairy beverages formulated with cheese whey have gained prominence in the global dairy market because they are produced by using simple technologies and are widely accepted by consumers of different age groups (Krešić, Herceg, Lelas, & Jambrak, 2010). These products have an interesting nutritional value because of their protein content and are an important alternative for reusing whey generated during cheese production, which is a large source of environmental pollution when improperly disposed

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(Hernández-Ledesma, Ramos, & Gomez-Ruiz, 2011; Sanmartín, Díaz, Rodríguez-Turienzo, & Cobos, 2011).

The consumption of goat dairy products has increased worldwide with a consequent increase in the demand for goat milk, which is stimulating production in several countries (Queiroga et al., 2013). Formulations of mixed goat and cow milk beverages have been studied (Gomes et al., 2013); however, there is a lack of information regarding dairy beverage formulations with only goat milk, particularly when formulated with probiotics and prebiotics. In addition, most of the existing studies about dairy beverages have reported formulations with fruits or fruit jams, and there are no reports of goat dairy beverages containing chocolate.

Therefore, the aim of this study was to produce chocolate goat dairy beverages containing the probiotic *B. lactis* and to evaluate the effects of goat cheese whey and prebiotics (inulin and oligofructose) on the physicochemical parameters and sensory features of the beverages.

2. Materials and methods

2.1. Materials

Dairy beverage formulations were prepared by using the following ingredients: *B. lactis* culture (BLC 1, Sacco Brazil, São Paulo, Brazil), Synergy 1 prebiotic (a mixture of inulin and oligofructose) (Beneo-Orafti, Oreye, Belgium), pasteurized goat cheese whey from the production of rennet-type cheese (Laboratory of Research and Development of Dairy Products, Center for Humanities, Social and Agrarian Sciences - Federal University of Paraíba, Bananeiras, Brazil), UHT goat milk (Caprilat, Paraná, Brazil), sucrose (União, São Paulo, Brazil), powdered chocolate (50 g 100 g^{-1} cocoa) (Nestlé, São Paulo, Brazil) and xanthan, sodium carboxymethyl cellulose and carrageenan gums (Genkorlac CM 130) (São Paulo, Brazil). The compositions of the milk and whey used in the formulations are shown in Table 1.

2.2. Experimental design and statistical analysis

Seven formulations were prepared according to the central composite design to obtain a model that represents the behavior of the independent variables goat cheese whey (X1) and Synergy 1 prebiotic (oligofructose and inulin) (X2) that were added at different proportions to the formulations (Table 2). The formulations were prepared randomly. Analyses of the residues, coefficients of determination (adjusted R^2) and lack of fit were used to verify the model adequacy. The regression coefficients of the Scheffé canonical polynomial equation for the adjusted model were used to evaluate the effects on the dependent variables (*B. lactis* viability, apparent viscosity, pH and total solids). Following model adjustment, the results were expressed as the mean ± the standard deviation and submitted to analysis of variance (ANOVA) and Tukey's test at $P < 0.05$. For the sensory analysis data, the results were expressed as the median [25% quartile −75% quartile], and a comparison of medians was performed by Mann–Whitney *U* test at $P < 0.05$. All analyses were performed with Statistica 7.0 software (Statsoft Inc., USA).

Table 1
The physicochemical parameters of the goat milk and goat cheese whey employed in the production of chocolate goat dairy beverages (mean ± standard deviation).

Physical-chemical parameters	Goat milk	Goat cheese whey
pH	6.78 ± 0.01	6.23 ± 0.05
Titratable acidity (g lactic acid 100 g^{-1})	0.16 ± 0.02	0.15 ± 0.01
Lactose (g 100 g^{-1})	4.35 ± 0.03	5.04 ± 0.07
Protein (g 100 g^{-1})	3.38 ± 0.04	0.85 ± 0.02
Fat (g 100 g^{-1})	3.36 ± 0.05	0.60 ± 0.01
Ash (g 100 g^{-1})	0.80 ± 0.01	0.51 ± 0.01
Non-fat solids (g 100 g^{-1})	8.11 ± 0.02	6.21 ± 0.01
Total solids (g 100 g^{-1})	11.47 ± 0.03	6.81 ± 0.02

Table 2
The central composite design for the independent variables, namely goat cheese whey (X1) and prebiotic (Synergy 1) (X2), in the chocolate goat dairy beverages.

Formulation	Coded variables		Uncoded variables[a]	
	Whey (X1)	Prebiotic (X2)	Whey (mL 100 mL^{-1})	Prebiotic (g 100 mL^{-1})
F1	−1	−1	15	0
F2	1	−1	45	0
F3	−1	1	15	6
F4	1	1	45	6
F5	0	0	30	3
F6	0	0	30	3
F7	0	0	30	3

[a] Expressed in whole matter.

2.3. Producing the inoculum and dairy beverages

The *B. lactis* inoculum was prepared by adding 1 g of culture to 100 mL of UHT goat milk and statically incubating the mixture at 35 °C for 12 h. The viability of the *B. lactis* was determined according to the procedure detailed in Section 2.6 over 24 h, at 2 h time intervals. For this analysis, bacterial suspensions were serially diluted at each time interval in peptone water ($10^{-5} - 10^{-9}$), and the viable cell counts were determined and expressed as the logarithm of colony forming units per mL of product (log CFU mL^{-1}) (data not shown). At 12 h of incubation, the counts were approximately 10^{11} CFU mL^{-1}, and they were stable or decreasing until 24 h.

The production of these dairy beverages consisted of the homogenization and pasteurization (65 °C 30 min^{-1}) of sucrose (70 g L^{-1}), powdered chocolate (28 g L^{-1}), gums (2 g L^{-1}) and goat milk. Pasteurized goat cheese whey and/or prebiotics were added and homogenized according to each formulation (Table 2). During the last step, 20 mL L^{-1} of the inoculum that was prepared as described above and that contained approximately 10^{11} CFU mL^{-1} was added to the beverage that was stored in plastic bottles (150 mL) at 5 °C ± 2 °C for 28 days. The same goat milk and goat cheese whey batches were used.

2.4. The physicochemical analysis of dairy beverages

Physicochemical analyses were determined on the first day of storage according to AOAC methods (2005) for the following components: fat (g 100 g^{-1}) (method # 2000.18), protein (g 100 g^{-1}) (method # 939.02), lactose (g 100 g^{-1}) (method # 923.09), total solids (g 100 g^{-1}) (method # 990.19), ash (g 100 g^{-1}) (method # 930.30), titratable acidity (g of lactic acid 100 g^{-1}) (method # 920.124). Physicochemical analyses of the pH, titratable acidity, total solids, apparent viscosity and syneresis were performed after 1, 7, 14, 21 and 28 days of storage.

The apparent viscosity was measured with a Brookfield-type viscometer (FUNGILAB, Italy) at 5 ± 2 °C and a rotation speed of 60 rpm. The results are given in millipascal seconds (mPa s). The syneresis was analyzed by centrifugation (Gauche, Tomazi, Barreto, Ogliari, & Bordignon-Luiz, 2009).

2.5. The microbiological analysis of dairy beverages

The microbiological analysis was performed according to the methodology recommended by the American Public Health

Association (APHA, 2001, chap. 7) as follows: the determination of the most probable number (MPN) of total coliforms (MPN mL^{-1}) and of thermotolerant coliforms (MPN mL^{-1}); the enumeration of molds and yeasts (CFU mL^{-1}); the enumeration of *Staphylococcus aureus* (CFU mL^{-1}); the detection of *Salmonella* spp, the last two were only performed after 7 days of refrigerated storage to ensure the sanitary quality of the product, and the others were performed after 1, 7, 14, 21 and 28 days of storage.

2.6. B. lactis viability

The viability of *B. lactis* in dairy beverages was determined after 1, 7, 14, 21 and 28 days. The samples were submitted to serial decimal dilutions in peptone water (1 g L^{-1}) and pour-plated in deMan-Rogosa-Sharpe agar (MRS Agar Himedia, India) that was enriched with sodium propionate (3 g L^{-1}), lithium chloride (2 g L^{-1}) and L-cysteine hydrochloride (0.5 g L^{-1}) followed by anaerobic incubation (Anaerobac Probac, São Paulo, Brazil) at 37 °C for 72 h (Vinderola & Reinheimer, 1999). The results were expressed as the logarithm of colony forming units per mL of product (log CFU mL^{-1}).

2.7. Sensory evaluation

The sensory evaluation used in this study was approved by the Ethics Research Committee of the Health Sciences Center at the Federal University of Paraíba, Paraíba State, Brazil (CAAE: 17196513.3.0000.5188; Protocol No: 440.040/2013), as recognized by the Ethics Research National Commission (CONEP).

Acceptability tests were conducted with 50 untrained panelists (consumers), and 50 mL of coded samples were randomly presented in individual booths; they were served in plastic cups and accompanied by a sensory evaluation form (Meilgaard, Civille, & Carr, 2007). The tests were performed after 14 days of storage, and each panelist tested a maximum of two samples of different formulations in a monadic order. The samples were assessed for flavor, color, aroma and texture by using a nine-point hedonic scale (9 = liked extremely, 5 = neither liked nor disliked; and 1 = disliked extremely).

3. Results and discussion

3.1. The microbiological analysis and physicochemical composition of dairy beverages

Microbiological analyses revealed that all of the goat dairy beverage formulations were in accordance with Instruction 16/2005 of Brazilian legislation (Brazil, 2005) during the storage period (28 days). The counts of total and thermotolerant coliforms were lower than 3.0 mL MPN^{-1}, and the mold and yeast counts were lower than 10 CFU mL^{-1}. The *S. aureus* counts were lower than 10^2 CFU mL^{-1}, and *Salmonella* spp was not detected.

The highest levels of lipids, proteins and ash were observed in formulations with the lowest amounts of whey (F1 and F3) (Table 3), which were similar to the results reported by Gerhardt, Monteiro, Gennari, Lehn, and Souza (2013) for bovine and mixed bovine and caprine dairy beverages that were formulated with different amounts of ricotta whey. These results are expected because significantly lower amounts of these nutrients were found in goat whey than in goat milk (Gomes et al. 2013).

The addition of a prebiotic (F3 to F7 formulations) increased the total solid contents of dairy beverages (Table 3). The lactose content varied among formulations. The formulations with higher lactose contents were those with the highest proportion of goat milk (F1 and F3) ($P < 0.05$) (Table 3). Gomes et al. (2013) reported lactose contents (5.03 g 100 g^{-1}) of dairy goat milk beverages that were similar to the F5 to F7 formulations that contained the same proportion of goat cheese whey (30 mL 100 mL^{-1}).

The pH and titratable acidity values of dairy beverages also differed ($P < 0.05$) among the formulations (Table 3), which could be related to the proportion of milk and whey used, the activity of the initial inoculum, the storage time and the interaction of other ingredients present in the formulations (Thamer & Penna, 2006). Although the addition of *B. lactis* inoculum to the formulations requires the adaptation of microorganisms to the new medium, the acidity and pH may have been influenced by the metabolism of ingredients that were available for this adaptation.

3.2. Physicochemical analysis during refrigerated storage

All formulations exhibited a pH decrease ($P < 0.05$) throughout refrigerated storage (Table 4). Beverages formulated with the lowest amounts of whey (F1 and F3) had a higher pH decrease after 14 days of storage ($P < 0.05$). By contrast, the acidity values increased ($P < 0.05$) in all formulations after 14 days of refrigerated storage, except for F2, which only showed an increase in this parameter at 21 days. Although there is a reported pH and acidity stabilization in dairy beverages during storage (Gomes et al. 2013; Wang, Bao, Hendricks, & Guo, 2012), this finding was not observed in the present study. However, it is known that the acidification of fermented milk products may evolve during refrigerated storage, becoming less pronounced because of the effect of the low temperatures used for storage (Rojas-Castro, Villalobos, & Castro, 2007). In this study, the increased acidity of the formulations can be the result of post-acidification because of continued fermentation by *B. lactis* during storage, which is also reported for yogurt that is produced in co-culture with *Streptococcus thermophilus* and

Table 3
The physicochemical compositions of the chocolate goat dairy beverages (mean ± standard deviation).

	pH	Acidity[a]	Total solids[b]	Lactose[b]	Fat[b]	Protein[b]	Ash[b]
F1	6.56 ± 0.01^{e}	0.29 ± 0.01^{a}	17.43 ± 0.01^{a}	5.55 ± 0.02^{d}	3.38 ± 0.01^{e}	2.74 ± 0.02^{e}	0.77 ± 0.01^{c}
F2	6.40 ± 0.01^{b}	0.33 ± 0.01^{b}	17.42 ± 0.03^{a}	4.85 ± 0.01^{a}	3.22 ± 0.02^{c}	2.07 ± 0.02^{a}	0.68 ± 0.01^{b}
F3	6.52 ± 0.01^{d}	0.30 ± 0.01^{a}	21.72 ± 0.03^{c}	5.71 ± 0.01^{e}	3.27 ± 0.02^{d}	2.69 ± 0.03^{d}	0.77 ± 0.01^{c}
F4	6.35 ± 0.01^{a}	0.33 ± 0.02^{b}	21.77 ± 0.02^{c}	4.91 ± 0.01^{b}	3.00 ± 0.01^{a}	2.16 ± 0.01^{b}	0.69 ± 0.02^{b}
F5	6.44 ± 0.01^{c}	0.33 ± 0.01^{b}	20.13 ± 0.05^{b}	5.05 ± 0.01^{c}	3.16 ± 0.02^{b}	2.37 ± 0.01^{c}	0.65 ± 0.01^{a}
F6	6.44 ± 0.02^{c}	0.33 ± 0.01^{b}	19.97 ± 0.08^{b}	5.05 ± 0.01^{c}	3.16 ± 0.04^{b}	2.38 ± 0.02^{c}	0.64 ± 0.01^{a}
F7	6.45 ± 0.01^{c}	0.33 ± 0.02^{b}	19.89 ± 0.03^{b}	5.05 ± 0.01^{c}	3.17 ± 0.01^{b}	2.39 ± 0.02^{c}	0.65 ± 0.01^{a}

F1 (15 mL 100 mL^{-1} whey); F2 (45 mL 100 mL^{-1} whey); F3 (15 mL 100 mL^{-1} whey and 6 g 100 mL^{-1} prebiotic); F4 (45 mL 100 mL^{-1} whey and 6 g 100 mL^{-1} prebiotic); and F5, F6 and F7 (30 mL 100 mL^{-1} whey and 3 g 100 mL^{-1} prebiotic).
Different superscript letters in the same column indicate significant differences between formulations ($P < 0.05$).
[a] g of lactic acid 100 g^{-1}.
[b] g 100 g^{-1}.

Table 4
Physicochemical analyses of the pH, titratable acidity, total solids and apparent viscosity of the chocolate goat dairy beverages during refrigerated storage (mean ± standard deviation).

	Formulation	1 day	7 days	14 days	21 days	28 days
pH	F1	6.56 ± 0.01^{eE}	6.37 ± 0.01^{cD}	4.90 ± 0.01^{aC}	4.60 ± 0.01^{aB}	4.48 ± 0.01^{aA}
	F2	6.40 ± 0.01^{bE}	6.28 ± 0.01^{aD}	5.98 ± 0.01^{eC}	5.15 ± 0.01^{cB}	4.77 ± 0.01^{cA}
	F3	6.52 ± 0.01^{dE}	6.36 ± 0.01^{cD}	5.21 ± 0.01^{bC}	4.82 ± 0.01^{bB}	4.76 ± 0.01^{bA}
	F4	6.35 ± 0.01^{aE}	6.26 ± 0.01^{aD}	5.91 ± 0.01^{cC}	5.52 ± 0.01^{eB}	5.03 ± 0.02^{dA}
	F5	6.44 ± 0.01^{cE}	6.33 ± 0.01^{bD}	5.96 ± 0.01^{dC}	5.42 ± 0.01^{dB}	5.05 ± 0.01^{eA}
	F6	6.44 ± 0.02^{cE}	6.34 ± 0.01^{bD}	5.95 ± 0.01^{dC}	5.43 ± 0.01^{dB}	5.06 ± 0.01^{eA}
	F7	6.45 ± 0.01^{cE}	6.34 ± 0.01^{bD}	5.94 ± 0.01^{dC}	5.41 ± 0.01^{dB}	5.06 ± 0.01^{eA}
Titratable acidity (g lactic acid 100 g^{-1})	F1	0.29 ± 0.01^{aA}	0.34 ± 0.02^{aA}	0.64 ± 0.03^{bB}	1.09 ± 0.04^{cC}	1.29 ± 0.01^{cD}
	F2	0.33 ± 0.01^{bA}	0.34 ± 0.04^{aA}	0.37 ± 0.02^{aA}	0.81 ± 0.02^{bB}	1.22 ± 0.01^{bC}
	F3	0.30 ± 0.01^{aA}	0.35 ± 0.02^{aA}	0.61 ± 0.02^{bB}	1.01 ± 0.03^{cC}	1.30 ± 0.05^{cD}
	F4	0.33 ± 0.02^{bA}	0.35 ± 0.02^{aA}	0.43 ± 0.04^{aB}	0.57 ± 0.04^{aC}	0.80 ± 0.04^{aD}
	F5	0.33 ± 0.01^{bA}	0.37 ± 0.02^{aAB}	0.41 ± 0.02^{aB}	0.63 ± 0.02^{aC}	0.76 ± 0.01^{aD}
	F6	0.33 ± 0.01^{bA}	0.36 ± 0.03^{aAB}	0.40 ± 0.02^{aB}	0.59 ± 0.01^{aC}	0.78 ± 0.02^{aD}
	F7	0.33 ± 0.02^{bA}	0.37 ± 0.02^{aAB}	0.40 ± 0.02^{aB}	0.61 ± 0.04^{aC}	0.80 ± 0.02^{aD}
Total solids (g 100 g^{-1})	F1	17.43 ± 0.01^{aA}	17.45 ± 0.03^{aA}	17.50 ± 0.07^{aA}	17.41 ± 0.04^{aA}	17.29 ± 0.02^{aA}
	F2	17.42 ± 0.03^{aA}	17.22 ± 0.04^{aA}	17.45 ± 0.05^{aA}	17.42 ± 0.05^{aA}	17.36 ± 0.02^{aA}
	F3	21.72 ± 0.03^{cA}	21.65 ± 0.05^{cA}	21.81 ± 0.01^{cA}	21.72 ± 0.02^{cA}	21.08 ± 0.03^{cA}
	F4	21.77 ± 0.02^{cA}	21.16 ± 0.04^{cA}	21.54 ± 0.02^{cA}	22.02 ± 0.03^{cA}	21.09 ± 0.06^{cA}
	F5	20.13 ± 0.05^{bA}	20.15 ± 0.04^{bA}	20.02 ± 0.04^{bA}	19.68 ± 0.05^{bA}	19.79 ± 0.05^{bA}
	F6	19.97 ± 0.08^{bA}	20.01 ± 0.04^{bA}	20.11 ± 0.02^{bA}	19.74 ± 0.06^{bA}	19.54 ± 0.03^{bA}
	F7	19.89 ± 0.03^{bA}	20.04 ± 0.09^{bA}	20.03 ± 0.06^{bA}	19.90 ± 0.05^{bA}	19.43 ± 0.06^{bA}
Apparent viscosity (mPa s)	F1	125.77 ± 0.47^{cA}	135.70 ± 0.65^{cB}	166.80 ± 0.82^{dC}	188.03 ± 0.75^{cD}	126.77 ± 0.55^{bA}
	F2	118.57 ± 0.55^{aA}	126.23 ± 0.68^{aB}	138.13 ± 0.67^{aC}	166.20 ± 0.70^{aD}	116.20 ± 0.50^{aA}
	F3	129.63 ± 0.49^{cA}	139.97 ± 0.25^{dB}	187.63 ± 0.49^{eD}	198.30 ± 0.79^{cE}	176.37 ± 0.55^{dC}
	F4	137.50 ± 0.89^{dA}	140.80 ± 0.53^{dB}	153.17 ± 0.60^{bC}	170.23 ± 0.97^{bD}	176.03 ± 0.51^{dE}
	F5	123.67 ± 0.31^{bA}	130.30 ± 0.56^{bB}	163.97 ± 0.80^{cD}	168.60 ± 0.66^{bE}	150.83 ± 0.70^{cC}
	F6	123.41 ± 0.46^{bA}	129.43 ± 0.50^{bB}	163.20 ± 0.96^{cD}	169.60 ± 0.85^{bE}	150.40 ± 0.75^{cC}
	F7	123.13 ± 0.75^{bA}	130.27 ± 0.32^{bB}	163.87 ± 0.90^{cD}	168.43 ± 0.81^{bE}	149.47 ± 0.59^{cC}

F1 (15 mL 100 mL^{-1} whey); F2 (45 mL 100 mL^{-1} whey); F3 (15 mL 100 mL^{-1} whey and 6 g 100 mL^{-1} prebiotic); F4 (45 mL 100 mL^{-1} whey and 6 g 100 mL^{-1} prebiotic); and F5, F6 and F7 (30 mL 100 mL^{-1} whey and 3 g 100 mL^{-1} prebiotic).
Different superscript lower-case letters in the same column indicate significant differences between formulations ($P < 0.05$). Different superscript capital letters in the same line indicate significant differences between different days of storage ($P < 0.05$).

Lactobacillus delbrueckii subsp. *bulgaricus* (Rojas-Castro et al., 2007).

F3 and F4 formulations (containing 6 g 100 mL^{-1} prebiotic) presented total solid contents that were significantly higher during the 28 days of storage and did not differ from each other ($P < 0.05$), and F1 and F2 (without added prebiotic) showed lower values. Differences in the total solid contents among the formulations can affect viscosity and syneresis because the higher the total solids content, the lower the intensity of attractive forces between casein micelles, increasing water retention (Vargas, Chafer, Albors, Chiralt, & Gonzalez-Martinez, 2008).

No syneresis occurred during the 28 days of storage, showing the positive effect of the mixture of gums employed here. It is known that anionic hydrocolloids (xanthan gum, guar gum, pectin and carrageenan) interact with positive charges on the surface of casein micelles in fermented milks, reinforcing the network that was formed, and consequently reducing syneresis (Everett & Mcleod, 2005). Furthermore, the addition of whey and prebiotics (inulin and oligofructose) did not affect syneresis in the dairy beverages of the present study, although increased syneresis has already been reported in fermented dairy beverages that were enriched with cheese whey and oligofructose (Castro et al., 2009).

The apparent viscosity increased for up to 21 days in all formulations, with a decline between 21 and 28 days, except for F4 ($P < 0.05$) (Table 4). This increase may be related to the solidification of the gel structure and eventual thixotropy of the product (Gomes et al., 2013; Wang et al., 2012). Moreover, the combination of whey and prebiotics most likely influenced the maintenance of the formulation viscosity because F4 had the highest amounts of these ingredients. Inulin shows good stability during the storage of acidic products, such as yogurt and dairy beverages, and it is capable of interacting with water to form microcrystals and of making the mixture more soft and creamy (Pimentel, Garcia, & Prudencio, 2012).

3.3. *B. lactis* viability

There was a reduction in the viability of *B. lactis* after one day of storage in all formulations (with the standardized addition of 9 log CFU mL^{-1}); however, F1 presented a higher population of *B. lactis* than the other formulations ($P < 0.05$) (Table 5). This behavior was possibly a result of the F1 composition, which, among the formulations studied, was the closest to the nutrient composition that was used to obtain the inoculum.

A significant increase ($P < 0.05$) in the viability of *B. lactis* up to 14 and 21 days for formulations F1, F2 and F3, and F4, F5, F6 and F7, respectively, was observed, remaining between 6 and 8 log CFU mL^{-1} throughout the storage period (Table 5). This result should be highlighted because it meets the recommended minimum daily intake of viable probiotic cells per portion of product ready for consumption (Brazil, 2008), indicating the functional potential of these formulated dairy beverages.

Ranadheera, Evans, Adams, and Baines (2013b) reported an increase in the viability of *B. lactis* in probiotic chocolate goat ice cream that was similar to that reported in this study, and other authors have demonstrated stability in the viability of *B. lactis* that was added to dairy products during storage (Cardarelli, Buriti, Castro, & Saad, 2008; Casarotti, Monteiro, Moretti, & Penna, 2014; Raeisi, Ouoba, Farahmand, Sutherland, & Ghoddusi, 2013). The goat milk composition includes minerals such as calcium, zinc and magnesium, which are important components of the enzyme complexes that are involved in lactose fermentation, in addition to a high protein content, favoring the multiplication of bifidobacteria (Slačanac et al. 2010) (Table 1).

Table 5
B. lactis viability (log CFU mL^{--1}) in the chocolate goat dairy beverages during refrigerated storage (mean ± standard deviation).

Formulation	1 day	7 days	14 days	21 days	28 days
F1	6.95 ± 0.30^{bA}	7.27 ± 0.11^{abA}	8.13 ± 0.03^{dC}	8.00 ± 0.01^{abBC}	7.52 ± 0.09^{abAB}
F2	6.46 ± 0.28^{abA}	7.45 ± 0.08^{bB}	7.86 ± 0.03^{cC}	7.82 ± 0.02^{aC}	7.90 ± 0.09^{bcC}
F3	6.42 ± 0.31^{abA}	7.37 ± 0.20^{abB}	8.06 ± 0.02^{dD}	7.99 ± 0.01^{abCD}	7.71 ± 0.10^{bcC}
F4	6.10 ± 0.12^{aA}	7.35 ± 0.10^{abB}	7.72 ± 0.01^{bC}	8.05 ± 0.06^{bD}	7.23 ± 0.11^{aB}
F5	6.12 ± 0.10^{aA}	7.04 ± 0.14^{aB}	7.54 ± 0.01^{aC}	7.85 ± 0.05^{abCD}	7.95 ± 0.05^{bcD}
F6	6.05 ± 0.23^{aA}	7.02 ± 0.07^{aB}	7.53 ± 0.01^{aC}	7.95 ± 0.05^{abD}	7.99 ± 0.07^{bD}
F7	6.13 ± 0.31^{aA}	7.04 ± 0.14^{aB}	7.49 ± 0.01^{aC}	7.89 ± 0.04^{abCD}	7.94 ± 0.08^{bcD}

F1 (15 mL 100 mL^{-1} whey); F2 (45 mL 100 mL^{-1} whey); F3 (15 mL 100 mL^{-1} whey and 6 g 100 mL^{-1} prebiotic); F4 (45 mL 100 mL^{-1} whey and 6 g 100 mL^{-1} prebiotic); and F5, F6 and F7 (30 mL 100 mL^{-1} whey and 3 g 100 mL^{-1} prebiotic).
Different superscript lower-case letters in the same column indicate significant differences between formulations ($P < 0.05$). Different superscript capital letters in the same line indicate significant differences between different days of storage ($P < 0.05$).

3.4. The sensory evaluation of dairy beverages

Formulation F4 (whey and prebiotics at maximum concentrations) (Table 2) had the highest median values for flavor (6 [4, 7]) and aroma attributes (7 [6, 8]), differing significantly from the other formulations in relation to the aroma (Fig. 2) ($P < 0.05$). In a previous study, Montanuci, Garcia, and Prudencio (2010) reported that the addition of inulin did not affect the intensity of sensory attributes for kefir-type fermented dairy beverages; however, it contributed to the acceptance of the product. Thus, the combination of prebiotics with cheese whey was positive for flavor and aroma attributes in the dairy beverages from the present study.

The median flavor attributes ranged from 4 [3, 6] to 6 [4, 7] (Fig. 2). The F2 formulation had the smallest median, which differed ($P < 0.05$) from those of formulations F3 (6 [4, 6]), F4 (6 [4, 7]) and F6 (5 [3, 7]). With regards to the aroma, the medians ranged from 5 [4, 7] and 5 [5, 7] (F1 and F3) to 7 [6, 8] (F4) (Fig. 2). Formulations F2 and F5–F7 had a median of 6 [5, 7] and only F4 differed ($P < 0.05$) from the other formulations, which suggests that the increased whey content improves the flavor perception of the dairy beverage.

The appearance attribute presented medians ranging from 6 [5, 7] (F2) to 7 [6, 8] (F3, F4, F5–F7) (Fig. 2). Formulation F1 presented the highest median of 7 [7, 8], differing ($P < 0.05$) from formulations F2, F4 and F5. Formulation F2 showed the lowest value for this attribute, differing from formulations F1, F3, F6 and F7 ($P < 0.05$).

The texture attribute varied from medians 6 [6, 7] (F2) to 7 [6, 8] (F1, F3, F5–F7), with differences ($P < 0.05$) between formulations F1 and F2 and formulations F3 and F7 (Fig. 2). Formulation F4 had a median of 7 [6,7]; although 6 was the minimum score, all formulations were considered to be approved by panelists with regards to the texture and differences in the total solids or prebiotic contents among formulations and did not affect the sensory evaluation of texture. The values obtained for texture are thought to be directly related to the apparent viscosity on the day of sensory analysis (Table 4) because the formulation with the lowest apparent viscosity value at 14 days (F2) showed the lowest median for the texture attribute.

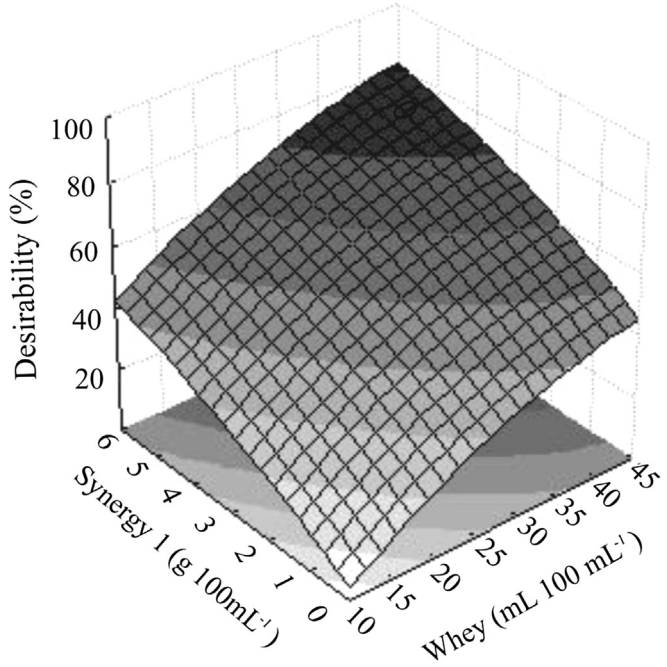

Fig. 1. A contour plot for the multi-response desirability (scale 0–100%) of the chocolate goat dairy beverages as a function of the goat cheese whey (mL 100 mL^{-1}) and prebiotic Synergy 1 [inulin + oligofructose (g 100 mL^{-1})].

3.5. Desirability profile

The linear model and Scheffé equations of the adjusted model were adopted to obtain dairy beverage formulations that would best fulfill the desired results for the selected responses (pH, total solids, *B. lactis* viability and apparent viscosity) at 14 days of storage. The effect of the goat whey (X_1) and prebiotic (X_2) and the interaction of ingredients (X_{12}) used in the formulations were determined according to Equations (1)–(4).

$$pH = 5.698 + 0.446X_1 + 0.059X_2 - 0.059X_{12} \quad (1)$$

$$\text{Total solids} = 19.908 + 1.973X_2 \quad (2)$$

$$\text{Viability of } B.\ lactis = 7.759 - 0.151X_1 - 0.053X_2 - 0.021X_{12} \quad (3)$$

$$\text{Apparent viscosity} = 162.395 + 15.783X_1 + 8.967X_2 + 1.45X_{12} \quad (4)$$

In these equations, the value and the sign (+ or −) of linear coefficients obtained for each response show that both factors contributed to the increased pH and apparent viscosity (positive β_1 and β_2) and decreased the viability of *B. lactis* (negative β_1 and β_2) and that only the prebiotic helped to increase the total solid contents (positive β_2). The low values of the probiotic viability coefficients signify that there was little influence from independent variables on this response. The effect of the whey interaction with the prebiotic ingredient (β_{12}) demonstrates that these factors together contributed to the pH decrease and the viability of *B. lactis* and increased the apparent viscosity. No significant interaction (β_{12}) regarding the total solids content was observed.

The maximum overall desirability was 80% (Fig. 1). This result indicates that the optimal area that was statistically obtained and

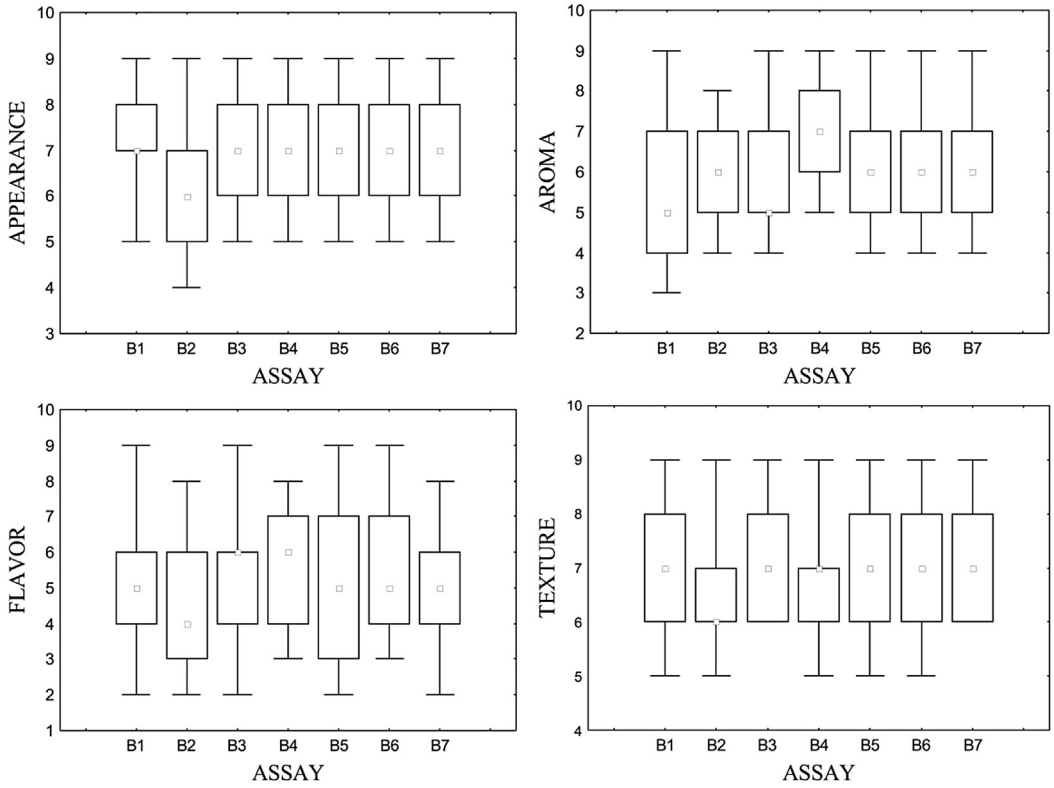

Fig. 2. The sensory attributes of the goat dairy beverages after 14 days of refrigerated storage. Median (small square), quartile 25% – quartile 75% (rectangular box), maximum and minimum values (plus and minus bar) of 50 untrained panelists.

viewed by using the darker area of the response surface corresponded to the addition of 30–45 mL 100 mL^{-1} whey and 4–6 g 100 mL^{-1} prebiotic to the dairy beverage. These ranges include the F4 formulation (45 mL 100 mL^{-1} whey and 6 g 100 mL^{-1} prebiotic), which exhibited the highest scores for the flavor and aroma sensory attributes and, therefore, would be the most suitable formulation according to the studied conditions, when considering a minimum daily intake of 100 mL for the dairy beverage.

In conclusion, the results suggest that goat cheese whey and the combination of inulin with oligofructose may be used as functional ingredients in formulating a probiotic chocolate goat dairy beverage to maintain sufficient probiotic viability and improve its viscosity and sensory features.

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References

AOAC. (2005). *Official methods of analysis* (18th ed.). Gaithersburg, MD: AOAC Intl.

APHA. (2001). *Compendium of methods for the microbiological examination of foods* (4th ed.). Washington: American Public Health Association.

Ashraf, R., & Shah, N. P. (2011). Selective and differential enumeration of Lactobacillus delbrueckii subsp. bulgaricus, Streptococcus thermophilus, Lactobacillus acidophilus, Lactobacillus casei and *Bifidobacterium spp*. in yoghurt—A review. *International Journal of Food Microbiology, 149*, 194–208.

Brasil, Ministério da Agricultura e da Agropecuária e Abastecimento. (2005). Instrução Normativa n°. 16 de 23 de agosto de 2005. In *Regulamento Técnico de Identidade e Qualidade de Bebida Láctea*. Brasília, DF: Diário Oficial da União, 23 ago. Seção 1.

Brasil Ministério da Saúde Agência Nacional de Vigilância Sanitária – ANVISA. (2008). *Alimentos com Alegação de Propriedades Funcionais e, ou, de Saúde, Novos Alimentos/Ingredientes, Substâncias Bioativas e Probióticos*. Comissões e Grupos de Trabalho. Comissão Tecnocientífica de Alimentos, Jul 2008. Available at http://www.anvisa.gov.br/alimentos/comissoes/tecno_lista_alega.htm.

Cardarelli, H. R., Buriti, F. C. A., Castro, I. A., & Saad, S. M. I. (2008). Inulin and oligofructose improve sensory quality and increase the probiotic viable count in potentially synbiotic petit-suisse cheese. *LWT – Food Science and Technology, 41*, 1037–1046.

Casarotti, S. N., Monteiro, D. A., Moretti, M. M. S., & Penna, A. L. B. (2014). Influence of the combination of probiotic cultures during fermentation and storage of fermented milk. *Food Research International, 59*, 67–75.

Castro, W. F., Cruz, A. G., Bisinotto, M. S., Guerreiro, L. M. R., Faria, J. A. F., & Bolini, H. M. A. (2013). Development of probiotic dairy beverages: Rheological properties and application of mathematical models in sensory evaluation. *Journal of Dairy Science, 96*(1), 16–25.

de Castro, F. P., Cunha, T. M., Ogliari, P. J., Teófilo, R. F., Ferreira, M. M. C., & Prudêncio, E. S. (2009). Influence of different content of cheese whey and oligofructose on the properties of fermented lactic beverages: study using response surface methodology. *LWT – Food Science and Technology, 42*(5), 993–997.

Everett, D. W., & Mcleod, R. E. (2005). Interactions of polysaccharide stabilizers with casein aggregates in stirred skim-milk yoghurt. *International Dairy Journal, 15*, 1175–1183.

Gauche, C., Tomazi, T., Barreto, P. L. M., Ogliari, P. J., & Bordignon-Luiz, M. T. (2009). Physical properties of yoghurt manufactured with milk whey and transglutaminase. *LWT – Food Science and Technology, 42*, 239–243.

Gerhardt, A., Monteiro, B. W., Gennari, A., Lehn, D. N., & Souza, C. F. V. (2013). Características físico-químicas e sensoriais de bebidas lácteas fermentadas utilizando soro de ricota e colágeno hidrolisado. *Revista do Instituto de Laticínios Cândido Tostes, 68*(390), 41–50.

Gomes, J. J. L., Duarte, A. M., Batista, A. S. M., Figueiredo, R. M. F., Sousa, E. P., Souza, E. L., et al. (2013). Physicochemical and sensory properties of fermented dairy beverages made with goat's milk, cow's milk and a mixture of the two milks. *LWT – Food Science and Technology, 54*, 18–24.

Gueimonde, M., Delgado, S., Mayo, B., Ruas-Madiedo, P., Margolles, A., & de los Reyes-Gavilın, C. G. (2004). Viability and diversity of probiotic *Lactobacillus* and *Bifidobacterium* populations included in commercial fermented milks. *Food Research International, 37*, 839–850.

Hernández-Ledesma, B., Ramos, M., & Gomez-Ruiz, J. A. (2011). Bioactive components of ovine and caprine cheese whey. *Small Ruminant Research, 101*, 196–204.
Kailasapathy, K., & Rybka, S. L. (1997). *Acidophilus* and *Bifidobacterium spp.*: their therapeutic potential and survival in iogurt. *Australian Journal of Dairy Technology, 52*, 28–35.
Krešić, G., Herceg, Z., Lelas, V., & Jambrak, A. R. (2010). Consumers' behaviour and motives for selection of dairy beverages in Kvarner region: a pilot study. *Mljekarstvo, 60*(1), 50–58.
Meilgaard, M., Civille, G. V., & Carr, B. T. (2007). *Sensory evaluation techniques* (4th ed.). Boca Raton: CRC Press.
Montanuci, F. D., Garcia, S., & Prudencio, S. H. (2010). Sensory characterization and acceptance of sweetened full fat and low fat Kefir with inulin. *Brazilian Journal of Food Technology, 8*, 79–90.
Pimentel, T., Garcia, S., & Prudencio, S. H. (2012). Effect of long-chain inulin on the texture profi le and survival of Lactobacillus paracasei ssp. paracasei in set yoghurts during refrigerated storage. *International Journal of Dairy Technology, 65*(1), 104–110.
Queiroga, R. C. R. E., Santos, B. M., Gomes, A. M. P., Monteiro, M. J., Teixeira, S. M., Souza, E. L. de, et al. (2013). Nutritional, textural and sensory properties of Coalho cheese made of goats', cows' milk and their mixture. *LWT – Food Science and Technology, 50*, 538–544.
Raeisi, S. N., Ouoba, L. I. I., Farahmand, N., Sutherland, J., & Ghoddusi, H. B. (2013). Variation, viability and validity of bifidobacteria in fermented milk products. *Food Control, 34*, 691–697.
Ranadheera, C. S., Evans, C. A., Adams, M. C., & Baines, S. K. (2013a). Probiotic viability and physico-chemical and sensory properties of plain and stirred fruit yogurts made from goat's milk. *Food Chemistry, 135*(3), 1411–1418.
Ranadheera, C. S., Evans, C. A., Adams, M. C., & Baines, S. K. (2013b). Production of probiotic ice cream from goat's milk and effect of packaging materials on product quality. *Small Ruminant Research, 112*, 174–180.
Roberfroid, M. B. (2007). Prebiotics: the concept revisited. *Journal of Nutrition, 137*, 830S–837S.
Rojas-Castro, W., Villalobos, A. C., & Castro, M. L. P. (2007). Características del yogurt batido de fresa derivadas de diferentes proporciones de leche de vaca y cabra. *Agronomía Mesoamericana, 18*(2), 221–237.
Ross, R. P., Desmond, C., & Stanton, C. (2005). Overcoming the technological hurdles in the development of probiotic foods. *Journal of Applied Microbiology, 98*(6), 1410–1417.
Roy, D. (2005). Technological aspects related to the use of bifidobacteria in dairy products. *Le Lait, 85*(1–2), 39–56.
Saarela, M., Alakomi, H. L., Mättö, J., Ahonen, A. M., & Tynkkynen, S. (2011). Acid tolerant mutants of *Bifidobacterium animalis subsp. lactis* with improved stability in fruit juice. *LWT—Food Science and Technology, 44*(4), 1012–1018.
Sanmartín, B., Díaz, O., Rodríguez-Turienzo, L., & Cobos, A. (2011). Composition of caprine whey protein concentrates produced by membrane technology after clarification of cheese whey. *Small Ruminant Research, 105*, 186–192.
Slačanac, V., Božanić, R., Hardi, J., Szabó, J. R., Lučan, M., & Krstanovič, V. (2010). Nutritional and therapeutic value of fermented caprine milk. *International Journal of Dairy Technology, 63*(2), 171–189.
Thamer, K. G., & Penna, A. L. B. (2006). Caracterização de bebidas lácteas funcionais fermentadas por probióticos e acrescidas de prebiótico. *Ciência e Tecnologia de Alimentos, 26*(3), 589–595.
Vargas, M., Chafer, M., Albors, A., Chiralt, A., & Gonzalez-Martinez, C. (2008). Physicochemical and sensory characteristics of yoghurt produced from mixtures of cows' and goats' milk. *International Dairy Journal, 18*, 1146–1152.
Vinderola, C. G., & Reinheimer, J. A. (1999). Culture media for the enumeration of *Bifidobacterium bifidum* and *Lactobacillus acidophilus* in the presence of yoghurt bacteria. *International Dairy Journal, 9*, 497–505.
Wang, W., Bao, Y., Hendricks, G. M., & Guo, M. (2012). Consistency, microstructure and probiotic survivability of goats' milk yoghurt using polymerized whey protein as a co-thickening agent. *International Dairy Journal, 24*, 113–119.