The influence of sweeteners in probiotic Petit Suisse cheese in concentrations equivalent to that of sucrose

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ABSTRACT

As in the case of probiotic functional foods in recent years, demand has increased notably for light or diet foods with added sweeteners. However, little is known about the effect of different sweeteners on the microorganisms present. Thus, the objective of the current study was to establish the ideal sucrose concentration and equivalent concentrations of different sweeteners and to determine, by microbiological analyses, the influence of these compounds on the viability of the starter and probiotic cultures used in the production of strawberry-flavored Petit Suisse cheese during its shelf life. The ideal sucrose concentration was determined using the just-about-right (JAR) scale, and the equivalent concentrations of the sweeteners were subsequently determined by the magnitude estimation method. Microbiological analyses were also carried out to check the viability of the cultures during the product's shelf life. The results showed that the compounds Neotame (NutraSweet, Chicago, IL) and stevia presented, respectively, the greatest and least sweetening power of the sweeteners tested. None of the sweeteners used in this study exerted a negative effect on the viability of the starter or probiotic cultures, and thus we were able to obtain a probiotic, functional food with reduced calorie content.

Key words: Petit Suisse cheese, probiotic, sweetener, magnitude estimation

INTRODUCTION

Demand for functional foods has increased notably in recent years (Granato et al., 2010). When probiotics or prebiotics are added to these foods as biologically active components, they produce metabolic and physiological health benefits in addition to their basic nutritional properties (Siró et al., 2008). Petit Suisse cheese has a

high moisture content and a creamy consistency. It is produced from a mass obtained using a mixed coagulation process, and can be added to sweet and savory condiments to be consumed fresh (Brasil Ministério da Agricultura, Pecuária e Abastecimento, 1996). Because of its characteristics, it is an appropriate vehicle for probiotic bacteria because it requires no maturation period and has a relatively short shelf life (Helle et al., 2003).

In parallel, low- or reduced-caloric food products with sweeteners added (as sucrose substitutes) are becoming increasingly prominent in the marketplace. According to the Brazilian Association of Industries for Diet and Special Needs Foods (**ABIAD**), the light or diet food product market grew about 800% in the last 10 yr (ABIAD, 2011). When planning to substitute sucrose with a sweetening substance, it is important to know the sweetener well and to determine the concentration that best portrays an equivalent product sweetened with sucrose, in terms of intensity and sensory profile (Souza et al., 2011). First, the ideal concentration of sucrose for the specific product should be determined and then the equivalent sweetness of each sweetener measured. Of the existing sensory techniques, the just-about-right (**JAR**) scale is one of the effective methods most used (Gagula et al., 2007) because of both the reliability and validity of its results and its simplicity in use by the sensory panel (Meilgaard et al., 2006). Various methodologies can be used to obtain information about the equivalence in sweetness of a sweetener. However, the magnitude estimation method stands out, with normalized graphical representation of the results using Steven's Law and the Power Function (Stone and Oliver, 1969), which, by staggering the ratio, makes it possible to get a direct measurement of the sweetness intensity.

The introduction of sweeteners can result in significant sensory alterations in a product, particularly in appearance, texture, and flavor (Lawless and Heymann, 2010). However, little is known about the influence of these compounds on the viability of the probiotic bacteria used in functional foods, such as probiotic lactic products. Thus, the objectives of the present study

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were to establish the ideal sucrose concentration and equivalent concentrations of the sweeteners stevia, sucralose, aspartame, and Neotame (NutraSweet Corp., Chicago, IL), and to verify, by microbiological analyses, the viability of the starter and probiotic cultures used in strawberry-flavored Petit Suisse cheeses during the product's shelf life.

MATERIALS AND METHODS

Ingredients

The Petit Suisse probiotic cheese was manufactured using the following ingredients: pasteurized milk type A (Ati Latte, Atatiba, São Paulo, Brazil), calcium chloride (0.25 g/L, ECIBRA, Santo Amaro, São Paulo, Brazil), starter culture (3% wt/vol, *Streptococcus thermophilus* TA 040, Danisco, São Paulo, São Paulo, Brazil), probiotic cultures (5% wt/vol *Lactobacillus acidophilus* LA 14, *Bifidobacterium lactis* BL 04, Danisco), rennet (Chr. Hansen, Valinhos, São Paulo, Brazil), sterilized milk cream (25% wt/wt fat, Nestlé, Araçatuba, São Paulo, Brazil), strawberry pulp (Icefruit, Tatuí, São Paulo, Brazil), artificial strawberry colorant (Arcolor, Arco-Íris Brasil Ltda, São Paulo, São Paulo, Brazil), strawberry aroma (Duas Rodas Industrial Ltda, Campinas, São Paulo, Brazil), xanthan gum (Keltrol, CPKelco, Limeira, São Paulo, Brazil), carrageenan gum (LRA 50, CPKelco, Limeira), and guar gum (Tec Pharma, São Paulo, São Paulo, Brazil).

Sweeteners

Four sweeteners were used in Petit Suisse probiotic cheese production, all of which are allowed by the current Brazilian Legislation (Brasil Agência Nacional de Vigilância Sanitária, 2008), beyond that of sucrose: aspartame (NutraSweet, Chicago, IL), Neotame (NutraSweet), sucralose (NutraMax, Catanduva, São Paulo, Brazil), stevia (Estévia com 90% de rebaudio-side, Clariant, São Paulo, São Paulo, Brazil), and refined sugar (Açúcar Refinado, União, Sertãozinho, São Paulo, Brazil).

Probiotic Petit Suisse Cheese Processing

The base mass (quark cheese) and Petit Suisse cheese were prepared according to the methodology described by Cardarelli et al. (2008) with some changes, including the probiotic culture used, using cultures of *L. acidophilus* and *B. lactis*. The base mass (quark cheese) was obtained by heating pasteurized milk to 37 to 38°C, transferring it to isothermal containers and then adding the starter and probiotic cultures by direct inoculation

(direct vat starter), followed by homogenization. The containers with the inoculated milk were maintained at 37°C until pH reached 6.3 to 6.5, and then the rennet was added, and the mixture rehomogenized and maintained at 37°C until a curd was formed with pH of about 5.6 to 5.8. The curd was then carefully cut into cubes, which were placed in sterile cloth sacks to drain off the whey at a temperature of 10°C for 15 h. After drainage, the quark cheese was placed in sterile containers and stored at 4°C until mixed with the other ingredients to make the Petit Suisse cheese. All the remaining ingredients were then added and homogenized in a multi-processor for 30 min (Estephan rpm 1750/3500, Geiger, Pinhais, Paraná, Brazil), until a consistent mass was obtained. After homogenization, appropriate containers were filled with cheese and maintained under refrigeration at 4°C for 28 d. The ideal test and magnitude estimation test were applied the day after filling. The processing was performed twice, on 2 different days.

Determination of the Ideal Sucrose Concentration

Five formulations of probiotic Petit Suisse cheese were prepared, varying only in sucrose concentration: 7.5, 11, 14.5, 18, and 21.5% (wt/wt). The central point was defined according to estimates of the amounts of sucrose normally used in commercial dairy products and considering a previous study with Petit Suisse cheese (Souza et al., 2011). Fifty-two panelists were used, all Petit Suisse consumers, recruited among students and employees on the university campus, aged between 18 and 45 yr, and the majority (70%) being women. The panelists evaluated the samples using a 9-cm nonstructured scale anchored at the extreme left by "extremely less sweet than the ideal" and at the extreme right by "extremely sweeter than the ideal," the central point being the ideal concentration to add to the product (Meilgaard et al., 2004). The panelists were requested to indicate on the scale the intensity of the perceived sweet stimulus.

The probiotic Petit Suisse cheese was presented monadically to the panelists according to a complete block design (MacFie et al., 1989), 5 d after the processing. Samples were presented in disposable 50-mL white plastic cups, coded with 3-digit numerals chosen at random, so as not to influence the panelists. Water and water biscuits were also provided to rinse out the mouth and neutralize the flavors between samples. The samples were served in individual booths in the Sensory Analysis Laboratory of the Department of Food and Nutrition (DEPAN) of the State University of Campinas (UNICAMP), Brazil. The booths were equipped with the FIZZ Network Sensory Software (Biosystemes, Couteron, France).

Preselection of Panelists

To efficiently analyze the data of some sensory tests, the taste panel must have adequate discriminatory power. Thus, a preselection was carried out of the candidate panelists for the magnitude estimation test of the different sweeteners used in the strawberry-flavored probiotic Petit Suisse cheeses. A total of 20 candidates were recruited, all consumers of the product having no restrictions in consuming the product and showing interest in taking part in the test.

In the tests, the candidates were instructed to evaluate the samples from left to right, and identify, according to the coding, which sample was different from the others. The discriminative power of was evaluated by Wald’s sequential analysis (Amerina et al., 1965), using triangular difference tests with a significant difference at the 1% level with respect to sweetness, the objective being to select the candidates best able to discriminate the samples. Each candidate performed 3 triangular tests per day to preserve their sensory capacity. The parameters used to analyze the discriminatory capacity were prefixed at $\rho_0 = 0.45$ (maximum acceptable lack of ability), $\rho_1 = 0.70$ (minimum acceptable ability), and for the risks $\alpha = 0.05$ (probability of accepting a candidate without acuity) and $\beta = 0.05$ (probability of rejecting a candidate without acuity) (Moraes and Bolini, 2010). Having defined the parameters, 2 lines of equation were obtained and used to construct the Wald analysis figure. For these parameters, a graph with 3 defined areas was obtained: acceptance area, indecisive area, and rejection area of the panelists. Thus, the panelists were selected or rejected according to the number of correct replies in the triangular tests applied. In the end, 15 panelists were selected for the determination of sweetness equivalence of the Petit Suisse cheeses sweetened with different sweeteners.

Sweetness Equivalence

Fifteen panelists, selected using Wald sequential analysis and trained in the use of the magnitude scale for samples with different sweetness intensities, were used to determine sweetness equivalence. Training consisted of an explanation of the methodology used, the

correct use of the magnitude scale and an explanation of the existence of samples with intensities different to that of the reference sample. After training, the panelists were directed to the booths of the sensory analysis laboratory of DEPAN/UNICAMP, where they carried out affective analyses of samples sweetened with the different sweeteners and sucrose, on different days. A series of concentrations was used to evaluate each sweetener in the strawberry-flavored probiotic Petit Suisse cheeses. Table 1 shows the concentrations used in the magnitude estimation test. The central concentrations used in the determination of sweetness equivalence were based on data from the literature (Cardoso and Bolini, 2007; Palazzo et al., 2011), and the other concentrations were calculated using a multiplication factor of 1.6 (Cardoso et al., 2004). Five concentrations were determined for each sweetener used, and having defined these concentrations, the amounts were weighed, added to the probiotic Petit Suisse cheeses, and homogenized.

The samples were served to the panelists at $4 \pm 2^\circ\text{C}$ in individual booths, placed in disposable 50-mL plastic cups, coded at random with 3 digits, using balanced complete blocks (MacFie et al., 1989). They were accompanied by a reference strawberry-flavored probiotic Petit Suisse cheese, coded with the letter R, which was sweetened with the ideal concentration of sucrose as found using the JAR scale. The selected and trained panelists received the reference sample with its intensity designated by an arbitrary sweetness score of 100, followed by various coded samples at random, with higher and lower intensities compared with the reference, as already experienced during training. The panelists were requested to designate sweetness intensities to the coded samples in relation to the reference sample. For example, if a sample presented half the sweetness intensity of the reference sample, it should receive a score of 50, if it presented twice the sweetness, it should receive a score of 200, and so on, not being permitted to award scores of 0 to the samples.

Microbiological Analyses

Because no preservatives were included in the Petit Suisse cheeses produced here, which could affect the

Table 1. Concentrations of sucrose, sucralose, stevia, Neotame, and aspartame to determine sweetness equivalence, using the magnitude scale, of a concentration of 15.2% sucrose in probiotic Petit Suisse cheese

Sweetener	Concentration (% wt/wt)				
Sucrose	5.94	9.50	15.20	24.32	38.91
Sucralose	0.0095	0.0152	0.0243	0.0389	0.0622
Stevia	0.0594	0.0950	0.1520	0.2432	0.3891
Aspartame	0.0415	0.0523	0.0836	0.1338	0.2140
Neotame ¹	0.0244	0.0016	0.0025	0.0040	0.0064

¹NutraSweet Corp. (Chicago, IL).

probiotic cultures, shelf life was only 28 d, shorter than that of the conventional commercial product (Pereira et al., 2010). Thus, the viabilities of the cultures of *Streptococcus thermophilus*, *Lactobacillus acidophilus*, and *Bifidobacterium lactis* were monitored throughout the shelf life of the Petit Suisse cheeses, in triplicate, at 1, 14, and 28 d. The *Streptococcus thermophilus* count was determined by pour plating 1 mL of each dilution in M17 agar (Oxoid, Basingstoke, UK) and incubating aerobically at 37°C for 2 d (Pereira et al., 2010). The *Lactobacillus acidophilus* count was determined by pour plating 1 mL of each dilution in de Man, Rogosa, Sharpe (MRS) agar with the addition of 0.15% (wt/vol) bile salts and anaerobic incubation (Anaerogen anaerobic system, Oxoid) at 37°C for 3 d (Cruz et al., 2012a,b, 2013; Karimi et al., 2012). *Bifidobacterium lactis* was counted by pour plating 1 mL of each dilution in MRS agar (Oxoid) with the addition of sodium propionate (0.3 g/100 mL) and lithium chloride (0.2 g/100 mL) and incubating anaerobically (Anaerogen anaerobic system, Oxoid) at 37°C for 3 d (Cruz et al., 2010; Castro et al., 2013a,b). All microbiological analyses were carried out in triplicate, and the viable cells were counted after the determined incubation period and expressed as the log colony-forming units per gram (log cfu/g).

Statistical Analysis

The results of the sensory analysis of the probiotic Petit Suisse cheese samples using the JAR scale were obtained by way of the FIZZ Network Sensory Software (Biosystemes), and analyzed by a simple linear regression analysis and using the bar histogram analysis as suggested by Vickers (1998), using Excel 2007 for Windows (Microsoft Corp., Redmond, WA). For the magnitude estimation method, the estimated sweetness magnitude values were converted into logarithmic values and expressed as the geometric mean. The curves for concentration versus sensory response for each sweetener corresponded to a power function with the following characteristics: $S = a \cdot C^n$, where S is the perceived sensation, C is the concentration of the stimulus, a is the antilog of the value for y on the intercept, and n is the angular coefficient of the straight line obtained (Moskowitz, 1974; Stone et al., 2012).

RESULTS AND DISCUSSION

Determination of the Ideal Sucrose Concentration

After obtaining the data from the FIZZ Network Sensory Software (Biosystemes), the means of the scores awarded by the panelists for each sucrose concentra-

tion added to the product were calculated. A straight line was obtained from the calculation of the means, intercept, and slope, as shown in Figure 1. From the equation of the straight line, the sucrose concentration necessary to obtain the sweetness considered ideal for the strawberry-flavored Petit Suisse cheese could be determined, and was established as 15.17%, which was rounded to 15.2% to facilitate subsequent experiments.

The JAR scale has been shown to be effective in the optimization of numerous foods, including lemonades (Epler and Kemp, 1998), apples (Hampson et al., 2000), rice (Suwansri et al., 2002), tortilla chips (Meullenet et al., 2002), milk puddings (Ares et al., 2009), and a symbiotic soymilk beverage (Mondragón-Bernal et al., 2010). Other studies have aimed to specifically elucidate the ideal sucrose concentration to add to products such as mate tea (Cardoso et al., 2004), pineapple juice (Marcellini et al., 2005), peach nectar (Cardoso and Bolini, 2007), and mango nectar (Cadena and Bolini, 2012). In a study with conventional Petit Suisse cheese (Souza et al., 2011), a concentration of 17% (wt/vol) sucrose was determined as ideal for the product, slightly higher than the value found in the present study, which could be explained by the use of different ingredients and distinct cultures. It was possible to show that the ideal sucrose concentration varied according to the type of product. Such variations are expected and likely exist because of interactions between components in the product.

Figure 2 gives a general view of the distribution of scores among the different sucrose concentrations. Considering a 9-point scale, where -4.5 corresponded to extremely less sweet than the ideal, $+4.5$ to extremely sweeter than the ideal, and the central point to the ideal sweetness, it can be seen that the samples with 14.5% and 18% sucrose presented a sweet taste closest to that considered ideal. The magnitude estimation method was used to provide the ideal sweetness in the probiotic Petit Suisse cheese and, in addition to the concentrations to be added to provide a sweetness equivalent to 15.2% sucrose, it was used to determine the sweetening power of the sweeteners used.

Determination of Sweetness Equivalence

For the data analysis, the geometric mean of the values awarded by each of the 15 panelists was calculated. The results for each panelist were then normalized, dividing the value given by each panelist by his or her respective geometric mean. Using the normalized data, the results of all the panelists were used to calculate the geometric mean of each sucrose concentration and of the different sweeteners. After obtaining these means, the logarithm of each concentration and the geometric

Figure 1. Ideal sucrose concentration to be added to probiotic Petit Suisse cheese, obtained with the use of a just-about-right (JAR) scale. Color version available in the online PDF.

mean derived from the normalized data for sucrose and for the other sweeteners were calculated. The linear regression between Log sweetener concentration (x) and Log geometric mean (y) for sucrose and the sweeteners was calculated with these values, obtaining the linearized results shown in Figure 3.

The values for the angular coefficient, intercept on the ordinate, and coefficient of linear correlation could then be determined from these graphs, as shown in Table 2. The straight line found was the linearized result of the simple power function $S = a \cdot C^n$, known as Steven's Law

or the Power Function (Moskowitz, 1974), where S is the sensation perceived by the panelists, C is the concentration of the stimulus (sweetener), a is the antilog of the value y on the intercept, and n is the angular coefficient of the straight line obtained. By substituting the variables of the function, the equivalent concentration of each sweetener was determined for the probiotic Petit Suisse cheese, as shown in Table 3.

The power of a sweetener is defined as the number of times a compound is sweeter than sucrose, based on its sweetness (Cadena and Bolini, 2012). Various

Figure 2. Histograms with the distribution of responses in acceptance test for probiotic Petit Suisse cheese samples with 7.5, 11, 14.5, 18, and 21.5% sucrose. Color version available in the online PDF.

Figure 3. Results of linearized power functions for probiotic Petit Suisse cheeses sweetened with sucrose, stevia, sucralose, aspartame, and Neotame (NutraSweet Corp., Chicago, IL). Color version available in the online PDF.

studies have aimed to determine the sweetening power of sweeteners (Cardoso et al., 2004; Marcellini et al., 2005; Cardoso and Bolini, 2007; De Marchi et al., 2009; Moraes and Bolini, 2010) but no previous studies have been done with probiotic Petit Suisse cheese and the sweeteners used in this study. Thus, the equivalent concentrations found for the sweeteners in this study divided by the sucrose concentration considered ideal

(15.2% wt/vol) for the probiotic Petit Suisse cheese determined their sweetening powers, as shown in Table 4. According to the data obtained in the present study, the sweetener sucralose was 625 times sweeter in the Petit Suisse cheese than 15.2% sucrose. This value was identical to that found by De Marchi et al. (2009) in a study on passion fruit juices sweetened with 10% sucrose. The value found in the present study was close to

Table 2. Values of slope, intercept on ordinate, linear correlation coefficient (r), and power function, obtained in tests using magnitude scale to determine the sweetness equivalence of sweeteners in probiotic Petit Suisse cheese¹

Sweetener	Slope	Intercept	r	Power function ²
Sucrose	1.2045	-0.7118	0.99	$S = 0.038 \cdot C^{1.2045}$
Sucralose	1.0127	0.8173	0.98	$S = 43.112 \cdot C^{1.0127}$
Stevia	0.6613	0.2705	0.99	$S = 3.475 \cdot C^{0.6613}$
Aspartame	1.0703	0.5657	0.99	$S = 13.533 \cdot C^{1.0703}$
Neotame ³	0.9456	1.2303	0.98	$S = 288.802 \cdot C^{0.9456}$

¹Equivalent sweetness to 15.2% (wt/vol) of sucrose.

² $S = a \cdot C^n$, where S is the perceived sensation, C the concentration of the stimulus, a is the antilog of the value for y on the intercept, and n is the angular coefficient of the straight line obtained (Moskowitz, 1974; Stone et al., 2012).

³NutraSweet Corp. (Chicago, IL).

Table 3. Concentrations of sucralose, stevia, aspartame, and Neotame in probiotic Petit Suisse cheese determined by magnitude estimation method

Sweetener	Concentration of sweetener ¹ (%)
Sucralose	0.0243
Stevia	0.1520
Aspartame	0.0877
Neotame ²	0.0025

¹Sweetness equivalent to 15.2% (wt/vol) of sucrose.

²NutraSweet Corp. (Chicago, IL).

that found by Cardoso and Bolini (2007) for equivalence in peach nectar sweetened with 10% sucrose. Cardoso et al. (2004) found powers of 679 and 554 in their study with hot and iced mate tea, respectively, as well as Marcellini et al. (2005) in pineapple juice and Moraes and Bolini (2010) in strawberry-flavored yogurts. However, in a study carried out with conventional Petit Suisse cheese, Souza et al. (2011) found a sweetening power of 262 for sucralose, a value much lower than that found in the present study, which can be explained by the use of different cultures with distinct metabolic profiles. More recently, using the time intensity technique, Palazzo et al. (2011) determined the sweetening power of sucralose in the formulation of diet chocolates. They found that sucralose was 570 times sweeter than sucrose in chocolates sweetened with 43% sucrose.

The sweetener stevia was only 100 times sweeter than sucrose in the probiotic Petit Suisse cheese, a value identical to that found for the same compound in instant coffees at 9.5% by Moraes and Bolini (2010). However, Cardoso and Bolini (2007) found a slightly higher value for stevia, whereas Cardoso et al. (2004) determined a value of 116 in hot mate tea. According to Palazzo et al. (2011), stevia showed a sweetening power 200 times greater in chocolates with 43% sucrose. The value determined in the present study was close to that found by Cavallini et al. (2005) of 94 in mango juice, equivalent to 8% sucrose.

In the current study, aspartame was shown to be 173 times sweeter than sucrose. Reis et al. (2012), analyzing strawberry-flavored yogurts sweetened with 11.5% su-

crose, found a sweetening power of 160 for aspartame. Values similar to those found in the present study were also encountered by Moraes and Bolini (2010) for equivalence in instant coffees sweetened with 9.5% sucrose (187 times sweeter) and by Cardoso and Bolini (2007), who determined a power of times 185 for aspartame in peach nectar sweetened with 10% sucrose. Marcellini et al. (2005) observed that aspartame presented a sweetening power of 144 times in pineapple juice sweetened with 8.5% sucrose, and Cavallini et al. (2005) observed a power of 146 for aspartame in mango juice sweetened with 8% sucrose. In hot mate tea, the power was 163 and was even greater in iced mate tea, with a power of 277 times (Cardoso et al., 2004). Neotame, the most recently discovered artificial sweetener, can present a sweetening power varying from 6,000 to 13,000 times higher than sucrose (Satyavathi et al., 2010). In the present study, this sweetener showed a sweetening power 6,082 times higher than sucrose when evaluated in the probiotic Petit Suisse cheese sweetened with 15.2% sucrose. Corroborating these data, in a recent study with diet chocolates, evaluating sweetness equivalence by the time intensity data collection method, the sweetening power of Neotame was established as 8,600 (Palazzo et al., 2011). It is worth mentioning the lack of studies on the behavior of this sweetener when added to various types of reduced-calorie foods.

Viability of the Starter and Probiotic Cultures

Figure 4 shows the viable cell counts for *Streptococcus thermophilus*, *L. acidophilus*, and *Bifidobacterium lactis* in the 5 samples of strawberry-flavored probiotic Petit Suisse cheeses. Initially, the population of the starter culture *Streptococcus thermophilus* showed good viability in all the samples, with values above 9 log cfu/g in nearly all the samples. Around d 14, all cultures showed a decrease in numbers compared with the initial counts, but maintained values >8.5 log cfu/g until the end of the shelf life, with the exception of the sample containing sucrose, which had counts slightly lower than the others.

The viability of *L. acidophilus* showed little change between the initial and final counts for the majority of samples, with almost all initial values above 7.5 log cfu/g.

The *Bifidobacterium lactis* counts showed curious results. Whereas the other cultures analyzed showed a slight loss in viability during storage, the *Bifidobacterium lactis* counts increased during the first half of the shelf life, reaching values >7.0 log cfu/g in all samples, and then remaining unaltered until the end of storage, with the exception of samples containing aspartame, which showed a slight reduction.

Table 4. Values of sweetening power (number of times that the compound is sweeter than 15.2% sucrose) determined for sweeteners in probiotic Petit Suisse cheese

Sweetener	Sweetening power
Sucralose	625
Stevia	100
Aspartame	173
Neotame ¹	6,082

¹NutraSweet Corp. (Chicago, IL).

Figure 4. Viability of *Streptococcus thermophilus*, *Lactobacillus acidophilus*, and *Bifidobacterium animalis* ssp. *lactis* obtained in probiotic Petit Suisse cheese during storage at 4°C for 28 d. Color version available in the online PDF.

As in other studies, the cheeses showed satisfactory probiotic bacterial counts (Gomes et al., 2011; Escobar et al., 2012; Lollo et al., 2012; Minervini et al., 2012), because of the protective effects of the food matrix (higher pH values and greater compactness; Cruz et al., 2009; Granato et al., 2010; Pereira et al., 2010). The viability of *Bifidobacterium* spp. appears to increase

in the presence of other probiotic strains, resulting in elevated values for all microorganisms evaluated (Vinderola et al., 2000; Corrêa et al., 2008). On the other hand, our results suggest the sweeteners had no influence on the metabolism of the probiotic bacteria, which maintained their viability, in accordance with previous studies performed in other foods, such as a wheat-based beverage (Angelov et al., 2006), chocolates (Nebesny et al., 2007), and dairy-based processed foods (Vinderola et al., 2002; Basyigit et al., 2006). However, it is important to emphasize that the sweeteners were added after the fermentation of the base cheese, not during it, which could have resulted in different viability values of the probiotic bacteria. So, this statement should be viewed with caution and additional research should be performed to further investigate this observation.

Based on the results presented, the addition of different sweeteners—0.024% sucralose, 0.152% stevia, 0.088% aspartame, 0.0025% Neotame, or 15.2% sucrose (wt/wt)—to probiotic Petit Suisse cheeses presented no negative effects on the viability of the probiotic bacteria during shelf life, established as 28 d, confirming the potential of these sweeteners for use in low-calorie functional foods. However, future studies should establish the sensory profiles and evaluate consumer acceptability of these products.

CONCLUSIONS

Petit Suisse cheese is a viable matrix for the application of probiotic microorganisms that allows viable cell counts (required for health beneficial effects) to be maintained throughout the shelf life. The sweeteners used, when added in the concentrations applied here, had no negative effect on the starter and probiotic cultures. Additional studies should be carried out to determine the viability of these microorganisms during a longer storage period and the acceptability of the product by affective and sensory descriptive tests.

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