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## Formulation of a peach ice cream as potential symbiotic food

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### Abstract

Today's population increasingly demands and consumes healthy products. For this reason, the food industry has been developing and marketing food with added bioactive components. The aim of this work was to formulate a peach ice cream reduced in calories with an added probiotic (*Bifidobacterium lactis* Bb-12) and prebiotics (inulin), and to evaluate its sensory quality and acceptability as potential symbiotic food. The moisture content was 76.47%; 7.14% protein; 0.15% fat; 6.37% carbohydrates; 9.87% inulin; 1.22% ash; 0.201% calcium, 0.155% phosphorus and 0.168% sodium. On the first and 21<sup>th</sup> day of storage counts of *B. lactis* Bb – 12 was  $4 \times 10^8$  CFU/mL and  $1.5 \times 10^7$  CFU/mL, respectively. It was possible to formulate a peach ice cream reduced in calories, fat, and sugar and with potential symbiotic effect, by addition of *B. lactis* Bb – 12. A product with suitable organoleptic characteristics, creamy texture, peachy colour, taste and flavour, and no ice crystals was obtained. This ice cream would be a suitable food matrix to incorporate prebiotic and probiotic ingredients as a potential symbiotic food.

**Keywords:** ice cream; inulin; probiotic; prebiotic; symbiotic.

**Practical Application:** Development of an ice cream with healthful properties.

### 1 Introduction

Today's population increasingly demands and consumes healthy products (Rozycki et al., 2011). For this reason, the food industry has been developing and marketing food with added bioactive components (Argentina, 2010), named “functional foods” that not only deliver basic nutrients, but also provide health benefits as they include ingredients that reduce the risk of chronic non-communicable diseases (González Fabre, 2008). Among them are probiotics, defined as live microorganisms that offer health benefits to the consumer beyond basic nutrition when, administered in adequate amounts (Argentina, 2013; Food and Agriculture Organization of the United Nations & World Health Organization, 2001). For probiotics to exert health effects, the recommended minimum level of viable cells has been suggested to be between  $10^6$ - $10^7$  CFU/mL at the moment of consumption (Argentina, 2013; Silva et al., 2015). Therefore, viability and functionality must be maintained throughout all food processing steps, ingestion by consumers, and transit through the gastrointestinal tract (Akin et al., 2007; Cruz et al., 2009). Probiotics are incorporated mainly in dairy products, such as milk, yogurt, cheese, and ice cream (Agrawal, 2005), and specific strains from the genera *Lactobacillus* and *Bifidobacterium* are generally used.

Prebiotics, on the other hand, are considered non-digestible food ingredients that affect the host by selectively and beneficially stimulating the growth and/or activity of one or a limited number of bacteria in the colon, mainly bifidobacteria (Roberfroid et al.,

2010). Inulin is perhaps the best characterized prebiotic and the most used one in food due to its functional, physical, chemical, and organoleptic properties (Charalampopoulos & Rastall, 2012). It has been shown that inulin has a protective effect on different strains of probiotics, increasing their survival and activity during storage (Karimi et al., 2015). Moreover, it is resistant to hydrolysis both in the stomach and the small intestine, exerts a bifidogenic effect and is linked to a variety of beneficial physiological properties (Meyer et al., 2011). It is added primarily to low-fat dairy products, including fermented milk, yogurt, dessert, cheese, and ice cream (Buriti et al., 2010) as it is also a fat replacer.

Symbiotic food contains a combination of probiotics and prebiotics that can act synergistically to modulate the intestinal microbiota and positively impact on people's health (Gotteland, 2010).

Ice cream could be a suitable vehicle to introduce probiotics and prebiotics in the human diet. Compared to fermented dairy products, ice cream is stored at lower temperatures and has a higher pH, which constitutes an advantage because lactic acid can severely affect the survival of probiotic bacteria (Vinderola et al., 2009; Senaka Ranadheera et al., 2012). Ice cream also has pleasant taste and appealing texture (Akalin & Erişir, 2008).

The aim of this work was to formulate a peach ice cream, reduced in calories, with an added probiotic strain (*B. lactis* Bb-12)

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and prebiotics (inulin), and evaluate the survival capacity of the probiotic strain, the sensory quality and acceptability of the product as potential symbiotic food.

## 2 Materials and methods

### 2.1 Preparation of ice cream

The following ingredients were used: skimmed milk powder (La Serenísima, Argentina); inulin powder (Nutralia S.R.L., Argentina); albumin powder (Tecnovo S.A., Argentina); diet peach preserver (PP) (BC La Campagnola, Argentina), peach pulp (pp) (Bahía Coctel, Argentina); sucralose and peach flavoring/aromatizing agents (F/A) (Grupo Saporiti, Argentina); *B. animalis subsp. lactis* Bb-12 (DVS freeze-dried culture, Chr. Hansen, Argentina); yellow colorant (YC) and red colorant (RC) (Circe Laboratories, Argentina). A peach ice cream reduced in calories was developed and standardized based on the formulation of Barrionuevo et al. (2011).

Inulin (100 g/L) was dissolved in boiling water and shaken until complete dissolution. Skimmed milk powder (130 g/L) was reconstituted with water at  $40 \pm 2^\circ\text{C}$ . Sucralose (0.20 g/L), peach pulp (50 g/L), and peach preserver (150 g/L) were drained and incorporated into the mix, along with peach F/A, YC (0.1 mL/L), and RC (0.05 mL/L). The mixture was homogenized with the dissolved inulin ( $47 \pm 2^\circ\text{C}$ ) and pasteurized at  $63 \pm 2^\circ\text{C}$  for 30 min. Subsequently, the mix was cooled to  $37 \pm 2^\circ\text{C}$  and the probiotic culture (0.1 g/L) was added (Akin et al., 2007; Akalin & Erişir, 2008). The mix was cooled in a double boiler by using ice instead of hot water, until the temperature dropped to  $4 \pm 1^\circ\text{C}$  (Di Bartolo, 2005). Rehydrated albumin (80 g/L) was whipped until stiff and incorporated with encircling movements. The mix was matured at  $4 \pm 1^\circ\text{C}$  for 2 h (Jiménez & Herrera de Zelarayán, 2003). Air was incorporated while it was beaten/frozen in an ice cream making machine (Electrolux HICM01), in which the temperature of the ice decreased to  $-21 \pm 2^\circ\text{C}$ . The product was packaged in expanded polystyrene containers and stored at a temperature of  $-18 \pm 2^\circ\text{C}$  (Jiménez & Herrera de Zelarayán, 2003).

### 2.2 Sensory analyses

#### Sensory quality assessment

In order to determine the sensory quality of the ice cream, the samples were analysed by Combined Score Test performed in triplicate. A group of 8 trained professors and graduates from the School of Health Sciences (Universidad Nacional de Salta) evaluated body, flavour, aroma, and colour attributes of the ice cream and assigned it a maximum quality score, taking a flavour peach commercial product as reference (Utset, 2007). The participants were trained on the scoring technique before the sensory test was performed. Fifteen gram samples were served in plastic containers (30 mL) at a temperature of  $-12^\circ\text{C}$ . A glass of rinsing water, sampling spoon, napkins, and instructions were also provided. An evaluation form was used for each assessment (McGhee et al., 2015). Participants were able to write additional comments at the end of the form.

### Acceptability test

The test was performed by 100 untrained evaluators, both male and female, aged 21 to 50, who were regular consumers of sweetener and ice cream. Acceptability was assessed by means of a 9 point verbal Hedonic Scale, categorized from “like extremely” to “dislike extremely”. Each sample (15 g) was served in disposable plastic containers, coded with a random 3 digit number (Pedrero & Pangborn, 1989).

### 2.3. Chemical analysis

The following analyses were performed on the final product: pH (pH – meter Hanna), inulin by HPLC (Zuleta & Sambucetti, 2001); proteins by formol titration (Egan et al., 1991); fat by alkaline hydrolysis; moisture by drying in stove; ash by calcination in muffle; calcium, phosphorus, and sodium (Association of Official Analytical Chemists, 1998) and carbohydrates by difference.

### 2.4 Physical analysis

Overrun was determined by the following formula:  $[(FIV - IMV)/IMV \times 100]$ , where FIV = frozen ice cream volume and IMV = initial mixture volume (Argentina, 2013). First dripping and complete melting were determined according to the methodology proposed by Güven & Karaca (2002), whereby 25 g of the product were placed on a metal mesh with 0.7 mm diameter openings and allowed to stand at  $20 \pm 2^\circ\text{C}$ . The values obtained were recorded with a digital timer and expressed in seconds.

### 2.5 Probiotic bacteria count

Ice cream sample was serially diluted in sterile 0.1 (w/v %) peptone water and pour plated in agar MRS (Man Rogosa Sharpe) supplemented with L-Cysteine-HCl and lithium chloride. The plates were incubated in an anaerobiosis jar with Anaeropack at  $37 \pm 1^\circ\text{C}$  for 72 h (Chr. Hansen, 2013; Vinderola & Reinheimer, 2000). Enumeration was done in duplicate. Viable bifidobacteria cell count was performed on days 1, 7, 14, and 21 during storage.

### 2.6 Data analysis

The values of the physico-chemical determinations, and the sensory quality and acceptability results were expressed in percentages, means, and standard deviation. The viability of probiotic microorganisms was expressed as colony forming units per milliliter (CFU/mL). The bacterial counts were analysed by Analysis of Variance (ANOVA) and Duncan test.

## 3 Results and discussion

### 3.1 Sensory quality

Table 1 shows the mean values obtained from the Combined Score Test of the peach ice cream, according to the sensory attributes evaluated.

Body and colour obtained the highest scores, outstanding characteristics for this product. Following in order of importance, the scores attributed to the flavor and the aroma were found, reaching 75% of adequacy.

The average total score was 85, which is suitable considering that is a product without added fat. The evaluators indicated that this product presented the following attributes: Taste: “soft, sweet, fruity, peachy flavour”; Aroma: “soft”; Colour “uniform, salmon” and Body, “moderate fusion, firm consistency, homogenous texture, perception of fine crystals, presence of fruit pieces”. The technological properties of inulin made it possible to obtain a product that was similar to the conventional one, since inulin replaced fat and maintained ice cream texture. The poor solubility of inulin, linked to its chain length, would be responsible for its ability to form microcrystals upon contact with water or milk, resulting in a creamy texture gel, which provides a smooth fat-like feel to the mouth (Coussement, 1999; Niness, 1999; Charalampopoulos & Rastall, 2012). On the other hand, it would have a synergistic effect with sucralose sweetener, positively influencing the sensory characteristics of the formulated ice cream (Wouters, 2005).

### 3.2. Acceptability test

The results of the acceptability test are shown in Table 2.

The highest frequency of responses was at point 7 of the scale corresponding to “Like moderately” (41%) followed by “Like very much” (25%) and “Like extremely” (11%).

Considering the acceptability percentage, the test yielded 88% for overall acceptability, 7% for indifference and the remaining 5% was for dislike.

During the sensory evaluation, participants had the opportunity to make comments on their forms. The most important comments about the ice cream were: “soft”; “creamy”; “pleasant taste”; “sweet taste” and “shining”.

In general, incorporating prebiotic ingredients into ice cream may have a greater influence on flavor and texture, compared to probiotics, which may affect primarily flavor (Cruz et al., 2010). The metabolism of the probiotic cultures can result in the production of components that might contribute negatively to the taste and aroma of the product (Mohammadi et al., 2011). However, the flavor and aroma were unaffected by the addition of probiotics in this work and the incorporation of inulin produced also no adverse effect on the sensory properties of the peach ice cream at a concentration of 10%.

### 3.3 Chemical analyses

The chemical characteristics of the peach ice cream are displayed in Table 3.

The total carbohydrates values were higher than those obtained by Barrionuevo et al. (2011) (10.51 g) and Sánchez Frías (2009) (15.4 g) lower than those of Asociación Española de Fabricantes de Helados (2008) (23.4 g). Mean protein was higher than that in milk ice cream (3.5 g) (Asociación Española de Fabricantes de Helados, 2008); probiotic ice cream (5.6 g) (Sánchez Frías, 2009) and lower than that obtained by Barrionuevo et al. (2011) (8.4 g). The amount of fat was lower than that of milk ice cream (4.8 g) (Asociación Española de Fabricantes de Helados, 2008) and probiotic ice cream (16.8 g) Sánchez Frías (2009). The final product was characterized by low caloric, glucidic and lipid

**Table 1.** Sensory quality of the peach ice cream.

Characteristic	Maximum Score	Assigned Score	Adequacy Percentage (%)
Flavour	20	15	75
Aroma	20	15	75
Colour	20	20	100
Body	40	35	87.5
Total	100	85 ± 9.46	

**Table 2.** Sensory acceptability of the peach ice cream.

Acceptability	Percentage (%)
Overall acceptability	88
Indifference	7
Dislike	5
Total	100

**Table 3.** Chemical composition of the peach ice cream (100 g).

	Values
Kcal.	70.19
Total carbohydrates*(g)	16.24
Inulin (g)	9.87
Simple Carbohydrates*(g)	6.37
Protein (g)	7.14 ± 0.14
Fat (g)	0.15 ± 0.03
Moisture (g)	76.47 ± 0.53
Ash (g)	1.22 ± 0.06
Sodium (mg)	168.50 ± 11.63
Calcium (mg)	201.40 ± 9.39
Phosphorus (mg)	155 ± 53
pH	6.3 ± 0.05

\*Values obtained by difference; mean ± standard deviation (n = 3); g: grams; mg: milligrams.

values, according to the Argentinian legislation, proteins of high biological value, and good supply of soluble fiber. Moisture content was higher than that reported by Barrionuevo et al. (2011) (68.13 g) content. The concentration of calcium and sodium was higher than that found by Barrionuevo et al. (2011), 148.56 and 133.96 mg, respectively, while the phosphorus value was lower than 167.50 mg (Barrionuevo et al., 2011). According to these data, the calcium/phosphorus ratio was 1.29, the recommended value being equal to or higher than 1. Therefore, the ratio obtained would favour absorption of both minerals (Teegarden et al., 1998)

### 3.4 Physical analyses

The overrun index obtained was 81%. Addition of inulin increased significantly the overrun in the product, indicating its responsibility for increased air incorporation (Akalin & Erişir 2008; Akin et al., 2007). This value is within the standard set by the Argentinian legislation (Argentina, 2013), which states that incorporated air may not exceed 120%, and in agreement with the ideal value of 70 to 100%, cited by Jiménez & Herrera de Zelarayán (2003). In milk ice cream this value falls between 50 to 85% (Di Bartolo, 2005). The overrun obtained would be



**Table 4.** Probiotic microorganism count.

Strain	Cell counts (CFU / mL) along storage (day)			
	1	7	14	21
<i>B. lactis</i> Bb-12	4 x 10 <sup>8</sup> (b)	9 x 10 <sup>7</sup> (a)	2 x 10 <sup>7</sup> (c)	1.5 x 10 <sup>7</sup> (d)

a, b, c, d Mean values with different superscript are significantly different ( $p < 0.05$ ).

influenced by the correct homogenization of the mixture, thereby facilitating the whipping. In addition, egg white and milk proteins would contribute to the stabilization of the foam, and along with the solid mix, to the incorporation of air, thus delivering the sensory characteristics of the final product (Madrid & Cenzano, 2003; Jiménez & Herrera de Zelarayán, 2003).

The first dripping of the peach ice cream occurred at 585 seconds (9 minutes 45 seconds) and total melting was completed at 3099 seconds (51 minutes 39 seconds). These values are lower than those reported by other authors such as Pandiyan et al. (2012); Akin et al. (2007) and Akin (2005), who scored on average higher values than 15 minutes (first dripping) and 60 minutes (total melting). The differences in the melting behaviour of ice cream samples added with probiotic cultures may be attributed to the differences in freezing points and viscosity (Akalin & Erişir, 2008; Favaro-Trindade et al., 2007) and also may be due to the fact that no stabilizers were used in the present investigation. On the other hand, the cited authors used amounts of inulin from 1-3% much lower than those used in this study.

### 3.5 Probiotic bacterial count

Table 4 shows probiotic microorganism counts. The daily values found throughout storage met the probiotics standards of the Argentinian Food Code (Argentina, 2013).

On days 7, 14 and 21, a decrease in the population of microorganisms was observed. However, during storage days, the concentration remained higher than 10<sup>7</sup> CFU/mL. According to the results analysed, although negligible, significant differences in cells counts were found. The decrease in microbial population would be related to various factors either during the production stage or during frozen storage. One of those factors is the freezing temperature during ice cream manufacture. Probiotic cells can be lethally injured by damage to their cell walls or membranes, caused by the mechanical stress of ice crystals, by temperature decrease shock, by condensation of solutes in the extracellular/intracellular medium, or by dehydration (Homayouni et al., 2012; Mohammadi et al., 2011; Cruz et al., 2009). All these factors may cause reduction in cell counts. Furthermore, the oxygen incorporated (overrun) during the beating/freezing step in the ice-cream making machine may result in an additional decrease in viable cell counts, because oxygen content and redox potential are important factors for the viability of bifidobacteria (Mohammadi et al., 2011; Cruz et al., 2009). *Bifidobacterium* are anaerobic bacteria, therefore, molecular oxygen as well as high values of redox potential would be critical factors for their survival (Vasiljevic & Shah, 2008).

Although there was a decrease in the number of viable cells, the ice cream could be considered as a probiotic food during the

storage period, since the number of viable cells remained above the recommended minimum limit of 10<sup>6</sup> CFU/mL (Argentina, 2013; Food and Agriculture Organization of the United Nations & World Health Organization, 2001; Vinderola & Reinheimer, 2000).

Prebiotics such as inuline, are selectively metabolized by probiotics or specific populations of the resident microbiota and enhance their growth and/or activity in the large intestine. This compound can be added to probiotic products as important growth factors and could significantly improve cells viability (especially for bifidobacteria) (Gibson & Roberfroid 1995).

Several studies suggest that the behavior of probiotic populations generally shows good survivability in ice cream up to the end of shelf life. During storage, survival period of probiotic bacteria depends on the strain, the production technology, storage time and temperature and the product formulation (Hagen & Narvhus 1999; Christiansen et al., 1996).

## 4 Conclusions

It was possible to formulate a peach ice cream reduced in calories, fat, and sugar and with potential symbiotic effect, by means of adding inulin and *B. lactis* Bb-12. According to the Argentinian legislation, the product may be labeled as a “diet food with reduced energetic value, modified in its glucidic composition and with high fiber content” and would also be considered “food with probiotics”. A product with suitable organoleptic characteristics, creamy texture, peachy colour, taste and flavour, and no ice crystals was obtained. This ice cream would be a good food matrix to incorporate prebiotic and probiotic ingredients and it would be a potential symbiotic food.

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