



# Development of fermented ice cream in combination with *Lactocaseibacillus rhamnosus* GG and persimmon puree and determination of physicochemical, rheological, textural and bioactive properties

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**ABSTRACT.** This work aimed to produce six distinct ice creams: IC: control ice cream (without any probiotic or persimmon puree), ICP: ice cream with 20% persimmon puree, PIC: probiotic ice cream, FIC: fermented ice cream, ICFP: ice cream with 20% fermented persimmon puree, FICP: co-fermented ice cream with 20% persimmon puree. *Lactocaseibacillus rhamnosus* GG was used as the probiotic and fermentation agent. The addition of persimmon puree, probiotic inoculation and fermentation resulted in a reduction in the total dry matter content of the ice cream ( $P \leq 0.05$ ). Co-fermentation led to a notable reduction in the pH value, with the lowest pH observed in the FICP ice cream sample at 5.33 ( $P \leq 0.05$ ). The addition of persimmon puree resulted in a decrease in the  $L^*$  values and an increase in both the  $a^*$  and  $b^*$  values of the ice cream samples. All ice cream samples exhibited pseudoplastic flow, and fermentation, persimmon puree addition and probiotic inoculation resulted in a reduction in  $K$  and hardness values. Co-fermentation had a protective effect on probiotic viability, with approximately  $8.95 \pm 0.01 \log \text{CFU g}^{-1}$  of probiotic viability detected in the FICP ice cream sample after 120 days of storage. Furthermore, co-fermentation markedly enhanced the bioactive characteristics of the samples, with the FICP sample exhibiting the highest TPC, CUPRAC, and DPPH values ( $242.57 \pm 11.52 \text{mg GAE } 100\text{g}^{-1}$ ,  $22.95 \pm 0.29 \text{mg TE } 100\text{g}^{-1}$ , and  $48.02 \pm 3.27\%$ , respectively). The study demonstrated that persimmon can be employed in the production of ice cream, with the co-fermentation of the ice cream mix promoting the viability of probiotics and enhancing the bioactive characteristics of the ice cream samples.

**Keywords:** Fermented ice cream; probiotic; *L. rhamnosus*; persimmon puree; probiotic viability; antioxidant activity.

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## Introduction

Nowadays, there is notable interest in the demand for functional foods, particularly as a consequence of technological developments and the trend towards healthier eating (Sun-Waterhouse, 2011). Ice cream is a dairy product that is enjoyed across the globe. Its characteristic taste and aroma, cooling effect, and especially its nutritional value are all reasons for its popularity (Goktas et al., 2022). Nevertheless, this dairy-based foodstuff is regarded as deficient in functional components such as phenolic compounds, carotenoids and vitamin C (dos Santos Cruxen et al., 2017). In this context, ice cream has the potential to be employed in studies on functional food development, through the incorporation of specific functional ingredients to meet high consumer demand. Additionally, ice cream could be considered an optimal carrier for probiotic cultures (Goktas et al., 2022).

The persimmon (*Diospyros kaki*) fruit is a rich source of glucose and fructose, as well as bioactive components, carotenoids, and minerals (Santos et al., 2018). The antioxidant, cytotoxic, and antidiabetic properties of persimmon fruit are attributed to the presence of carbohydrates, organic acids, bioactive components, carotenoids, and tannins. These compounds confer high bioactivity and sensory appeal to the fruit (Santos et al., 2018). Therefore, persimmon is a promising candidate for developing functional foods.

A multitude of biochemical alterations occur during the fermentation process, leading to altered proportions of both nutritional and non-nutritional components. These changes subsequently influence the

characteristics of the crop, including its bioactivity and digestibility (Zhang et al., 2012). The fermentation process has been shown to enhance the antioxidant properties and bioavailability of foodstuffs (Gao et al., 2022). Furthermore, fermentation can improve the probiotic viability (Golestani & Pourahmad, 2017), and also influence the physicochemical, textural, sensory, and rheological attributes of ice cream (Soukoulis et al., 2014).

The existing literature on the subject has concentrated on ice cream mix or milk fermentation. A review of the literature reveals no previous studies investigating the co-fermentation of fruit and ice cream mix. In this study, a series of fermented ice creams were produced through the implementation of the following fermentation processes: ice cream mix fermentation with probiotic *L. rhamnosus*, persimmon puree fermentation with *L. rhamnosus* and its subsequent addition to the mix, and the co-fermentation of ice cream mix and persimmon puree with *L. rhamnosus*. Furthermore, ice cream mixes comprising solely probiotics and persimmon puree, as well as control ice cream mixes without probiotics and persimmon puree, were produced. The physicochemical, textural, rheological, sensory and antioxidant characteristics of six distinct ice cream samples were subsequently evaluated. For probiotic ice creams, the impact of the fermentation process or persimmon puree addition on probiotic viability was elucidated by enumerating live probiotics over the storage period.

## Material and methods

### Persimmon puree and ice cream production

Fresh persimmon fruits were procured from a local greengrocer in Istanbul. Once the persimmon fruits had been peeled, they were pureed using a hand blender set to 10,000 rpm for ten minutes. Given that *L. rhamnosus* is regarded as a model probiotic, it was selected as the probiotic and starter culture for this study. Subsequently, persimmon puree was incorporated into the ice cream mixes at a ratio of 20% (g 100g<sup>-1</sup>). Furthermore, *L. rhamnosus* was inoculated as a probiotic bacterium and/or starter culture in the fermentation process (Goktas, 2023). Ice cream production was conducted in accordance with the methodology outlined by Goktas (2023), and the specifics of the production process are illustrated in Figure 1. The overnight probiotic culture was subjected to centrifugation at 7500 rpm for ten minutes, following which it was washed on two occasions with sterile PBS. The resulting pellet was then inoculated into ice cream mixes as a starter culture or as a probiotic agent. The ice cream samples were placed in 10 cm cube-shaped plastic containers for subsequent analysis. All analyses, with the exception of probiotic viability, were conducted within a seven-day period. Probiotic viability was determined at monthly intervals over the course of the 120-day storage period. The ice cream production was conducted in two replicates.

### Physicochemical properties

Dry matter and acidity of the ice creams were determined according to the procedure specified by Goktas et al. (2022). The pH values of the ice creams were measured from melted samples at room temperature using a pH meter (Hanna HI2002-02). The Gerber methodology was employed to ascertain the total fat content of the ice cream samples, while the Kjeldahl methodology was utilized to determine the total protein content. The analyses were conducted in duplicates.

### Colour properties

The colour measurement of the samples was assessed at four distinct points using a colourimeter (3nh NR200, (Shenzhen, China)). *L*\*, *a*\* and *b*\* values were presented as the average of four measurements.

### Melting properties

A quantity of ice cream approximately 10-15 mg was weighed and placed in aluminium pans, which were then hermetically sealed. Once the pans were placed in the DSC device (TA Instrument Q100 (New Castle, USA)), their melting properties were determined by applying the methodology previously described by Goktas et al. (2022), in which the temperature was increased by 5°C per minute from -20°C to +20°C. The melting properties were evaluated in duplicates.

### Rheological properties

The flow behaviour of the samples was assessed using a temperature-controlled rheometer (Anton Paar, MCR 302, Austria) (Goktas et al., 2022). A flow diagram was constructed using shear stress values corresponding to the shear rate at twenty-five points. The consistency coefficients (*K*, (Pa.s<sup>*n*</sup>)) and flow behaviour index (*n*) values were

obtained by fitting the shear rate and shear stress values to the Ostwald de Waele model ( $\sigma = K(\dot{\gamma})^n$ , (where  $\sigma$ = shear stress (Pa),  $\dot{\gamma}$ = shear rate ( $1\text{ s}^{-1}$ )). Rheological measurements were determined in two repetitions.

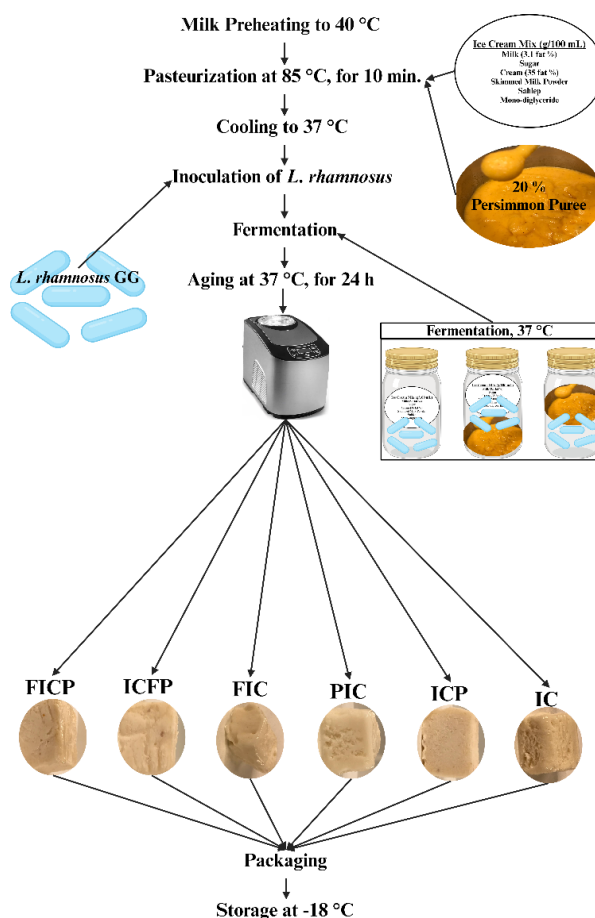


Figure 1. Ice cream production flow chart.

### Textural properties

The ice cream samples were cut into 2 cm cubes at 16°C and stored at a temperature of -18°C overnight. A texturing device (TA-XT Express Stable Micro Systems), equipped with a 10 kg load cell and a P/36R cylindrical aluminium probe, was utilized to ascertain the hardness (positive peak force, g) and cohesiveness (negative peak force, g) values (Hwang et al., 2009). The analysis was conducted according to the following procedure: initial test speed: 2.00 mm s<sup>-1</sup>, test speed: 2.00 mm s<sup>-1</sup>, final test speed: 10.00 mm s<sup>-1</sup>, distance: 40.00 mm and force: 0.1 N. Textural parameters were determined in two repetitions.

### Probiotic viability

The probiotic viability was determined on a monthly basis over the course of the 120-day storage period. The requisite dilutions were prepared and seeded onto 25 µL MRS agar medium according to the spot seeding technique, and incubated at 37°C for 24 hours. The results for the number of colonies formed following incubation are presented in log CFU g<sup>-1</sup>.

### Bioactive properties

The following extraction method was employed to ascertain the total phenolic content (TPC) and antioxidant properties (AP) of the ice cream samples. Briefly, 10 grams of ice cream was combined with 90mL of 80% methanol. The mixture was maintained at 60°C in a shaking water bath for two hours. Subsequently, the sample was subjected to an ultrasonic water bath for 30 min. The mixture was then filtered through filter paper and mixed with 20 mL of hexane. The methanolic fraction was obtained using a separatory funnel and stored at -18°C to determine its phenolic and antioxidant properties.

The following methodology was employed to ascertain the TPC (Singleton & Rossi, 1965) of the ice cream samples: 0.5 mL of extract was treated with 2.5 mL of tenfold diluted Folin reagent. Then, 2 mL of 7.5% sodium

carbonate was added, and the mixture was incubated in the dark for 30 minutes. Finally, the absorbance values of the mixture were recorded at 765 nm (Genesys 10S UV-Vis, Thermo Scientific, USA), and the TPC values were calculated using a previously obtained calibration chart and expressed in mg GAE<sup>-1</sup>.

The AP of the samples was identified using the DPPH (Singh et al., 2002) and CUPRAC (Apak et al., 2004) methods. A volume of 0.1 mL of the ice cream extract was treated with 3.9 mL of a DPPH solution (0.1 mM) and incubated in the dark for 20 minutes. Following the incubation period, absorbance values were recorded at 517 nm and the AP values of the samples were calculated using the following formula:

$$AP(\%) = (\text{Absorbance}_{\text{blank}} - \text{Absorbance}_{\text{sample}}) / \text{Absorbance}_{\text{blank}} * 100$$

The CUPRAC method was applied according to the following procedure: A solution of 0.1 mL of the ice cream extract was prepared by mixing it with 1 mL of CuCl<sub>2</sub>, 1 mL of Neocuproine and 1 mL of ammonium acetate. Subsequently, 1 mL of deionised water was added to the mixture and incubated in the dark for one hour. Finally, the absorbance values were recorded at 450 nm. The AP values were calculated using the calibration chart and results presented in mg TE 100g<sup>-1</sup>.

### Consumer acceptance

In order to ascertain consumer acceptance, the sensory parameters of appearance, flavour, odour, strange taste and overall acceptability were selected for analysis (Goktas, 2023). The consumer acceptance test was conducted by the academic staff of Istinye University. A total of 26 panellists, comprising 16 females and 10 males, participated in the evaluation. Prior to the evaluation, the panellists were provided with comprehensive information regarding the objectives and content of the study. Throughout the evaluation, the panellists were instructed to rinse their mouths with water between samples. The ice cream samples were presented to the panellists in approximately 20 grams in transparent plastic containers, each bearing one of three randomly generated codes. Finally, the panellists were requested to provide a score for the sensory evaluation on a scale of 1 (never liked) to 5 (very liked).

### Statistical evaluation

The results of the analysis are presented as the mean of two replicates. The JMP statistical programme was utilized to ascertain the statistical significance of the differences between the replicates. The 95% significance level between the differences was determined using the Tukey test.

## Results and discussion

### Physicochemical results

The results of the physicochemical analysis of the samples are presented in Table 1. The total dry matter (TDM, %) content exhibited a range of values between 38.06 and 30.20%. The addition of persimmon or probiotic inoculation to ice cream samples resulted in a reduction in the TDM content of the samples ( $P \leq 0.05$ ). The lowest TDM value was determined for FICP, where co-fermentation of the ice cream mix and 20% persimmon puree occurred. Persimmon fruit has a high water content, and the reduction in TDM in ice creams containing persimmon is associated with the high water content of the fruit. Furthermore, the lower TDM content of fermented ice cream samples is attributable to the utilization of carbohydrates as substrates by *L. rhamnosus* as a consequence of the fermentation process. Similarly, Karaman et al. (2014) found that the TDM content of ice cream samples decreased with increasing persimmon fruit concentration. However, Goktas (2023) reported that the addition of rowanberry pulp had no significant effect on the TDM values of the samples. This may be related to the different water content of the fruits.

The titratable acidity (TA) of the samples exhibited a notable alteration solely for FICP ( $P \leq 0.05$ ), wherein the co-fermentation of ice cream mix and 20% persimmon puree resulted in an increase in the TA value. On the other hand, significant alterations were observed in the pH values of the PIC (probiotic ice cream), FIC (fermented ice cream mix), and FICP (co-fermentation of ice cream and persimmon puree) ( $P \leq 0.05$ ). The differences in TA and pH values can be attributed to the growth of *L. rhamnosus* and the production of lactic acid during the fermentation process. Similarly, Homayouni & Norouzi (2016) and Golestani & Pourahmad (2017) reported lower acidity values for fermented ice cream samples, which they attributed to the fermentation process. Additionally, the protein and fat content values of the ice cream samples were found to be similar to those of the control group ( $P > 0.05$ ).

**Table 1.** Physicochemical and colour properties of ice cream samples.

Ice Cream Samples	Physicochemical properties					Colour properties		
	TDM (%)	TA (%)	pH	Protein (g 100g <sup>-1</sup> )	Fat (g 100g <sup>-1</sup> )	L*	a*	b*
IC	38.06 ± 0.01 <sup>a</sup>	3.10 ± 0.05 <sup>b,c</sup>	6.29 ± 0.01 <sup>a</sup>	4.63 ± 0.17 <sup>a,b</sup>	10.70 ± 0.71 <sup>a</sup>	91.22 ± 1.77 <sup>a</sup>	0.39 ± 0.39 <sup>b</sup>	9.76 ± 0.97 <sup>b,c</sup>
ICP	32.59 ± 0.19 <sup>c</sup>	2.95 ± 0.15 <sup>c</sup>	6.23 ± 0.01 <sup>a,b</sup>	4.51 ± 0.12 <sup>a,b</sup>	8.80 ± 0.85 <sup>a</sup>	85.44 ± 2.45 <sup>b,c,d</sup>	2.57 ± 0.26 <sup>a</sup>	13.05 ± 0.73 <sup>a</sup>
PIC	35.16 ± 0.23 <sup>b</sup>	3.19 ± 0.04 <sup>b,c</sup>	6.12 ± 0.03 <sup>b</sup>	4.83 ± 0.23 <sup>a</sup>	9.60 ± 0.28 <sup>a</sup>	88.31 ± 1.87 <sup>a,b,c</sup>	0.22 ± 0.19 <sup>b</sup>	10.48 ± 1.11 <sup>b</sup>
FIC	34.82 ± 0.07 <sup>b</sup>	3.33 ± 0.09 <sup>b</sup>	5.87 ± 0.01 <sup>c</sup>	4.55 ± 0.06 <sup>a,b</sup>	9.85 ± 0.49 <sup>a</sup>	89.46 ± 2.49 <sup>a,b</sup>	0.19 ± 0.21 <sup>b</sup>	8.83 ± 1.01 <sup>c</sup>
ICFP	32.12 ± 0.28 <sup>c</sup>	3.00 ± 0.07 <sup>c</sup>	6.20 ± 0.02 <sup>a,b</sup>	4.12 ± 0.09 <sup>b</sup>	8.60 ± 0.57 <sup>a</sup>	81.54 ± 5.97 <sup>d</sup>	2.93 ± 0.54 <sup>a</sup>	13.33 ± 0.69 <sup>a</sup>
FICP	30.20 ± 0.45 <sup>d</sup>	5.09 ± 0.01 <sup>a</sup>	5.33 ± 0.07 <sup>d</sup>	4.26 ± 0.12 <sup>b</sup>	9.05 ± 0.35 <sup>a</sup>	83.98 ± 3.55 <sup>c,d</sup>	2.62 ± 0.49 <sup>a</sup>	12.83 ± 0.97 <sup>a</sup>

IC: Control ice cream (without any probiotic or persimmon puree), ICP: Ice cream with 20% persimmon puree, PIC: Probiotic ice cream, FIC: Fermented ice cream, ICFP: Ice cream with 20% fermented persimmon puree, FICP: Co-fermentation of ice cream and 20% persimmon puree. TDM: Total dry matter, TA: Titratable acidity. The presence of different letters in the same column indicates a statistically significant difference ( $P \leq 0.05$ ).

### Colour properties

The brightness ( $L^*$ ) values varied between  $91.22 \pm 1.77$  and  $81.54 \pm 5.97$  (Table 1). The lowest  $L^*$  value was identified for the ice cream sample designated ICFP (ice cream with 20% fermented persimmon puree), while the highest  $L^*$  value was observed for IC. Although the  $L^*$  values of the probiotic (PIC) and fermented ice cream (FIC) samples were found to be inferior to those of the control ice cream sample, no statistically significant discrepancy was identified ( $P > 0.05$ ). Nevertheless, the  $L^*$  values of ice cream samples with added persimmon were found to be markedly lower ( $P \leq 0.05$ ). Similarly, Karaman et al. (2014) and Goktas (2023) reported that the addition of persimmon and rowanberry pulp reduced the  $L^*$  value of ice cream samples. In contrast to the  $L^*$  value, the  $a^*$  and  $b^*$  values of ice cream samples increased with the addition of persimmon puree, and the incorporation of persimmon puree significantly elevated the  $a^*$  and  $b^*$  values of the ice cream samples ( $P \leq 0.05$ ). However, the fermentation process did not exert a notable effect on the colour values of the ice cream samples ( $P > 0.05$ ).

### Melting properties

One of the key factors influencing the storage stability and consumer perception of products is the melting properties (Goktas et al., 2022). The melting values ( $T_{onset}$ ,  $T_{peak}$ ,  $T_{end}$  and  $\Delta h$ ) of the samples are given in Table 2. In general, the fermentation process and/or the addition of persimmon puree did not have a significant impact on the melting properties of the ice cream samples ( $P > 0.05$ ). Nevertheless, only the PIC (probiotic ice cream) exhibited a statistically significant difference compared to IC (control ice cream) ( $P \leq 0.05$ ). In a study, Goktas et al. (2022) reported that the addition of *L. rhamnosus* as a probiotic to ice cream significantly affected the melting properties of the samples. In line with this study, Homayouni & Norouzi (2016) reported that the fermentation of soy had no effect on the melting resistance of the ice cream. Conversely, Aboulfazli et al. (2015) indicated that the fermentation process resulted in a reduction in the melting rate of the ice cream samples. Hwang et al. (2009) reported that the addition of grape wine lees decreased the melting enthalpy of ice cream samples. The melting characteristics of ice cream depend on the freezable water content of the ice cream itself (Vittadini & Vodovotz, 2003), as well as the ability of the added ingredients to absorb water (Karaman et al., 2014). Furthermore, the melting properties of ice cream may be affected by lactic acid fermentation (Aboulfazli et al., 2015).

### Rheological properties

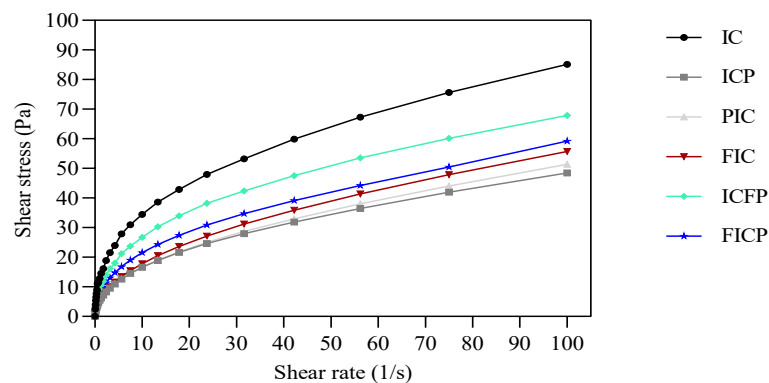
The flow behaviour diagram and flow parameters ( $K$  and  $n$ ) of the ice cream samples are presented in Figure 2 and Table 2, respectively. All samples exhibited shear thinning behaviour, with the shear rate increasing more than the shear stress; the highest shear stress value was observed for IC. The shear stress and shear rate values were fitted to the Ostwald de Waele model, with  $R^2$  values of 0.99 indicating the suitability of this model. All samples demonstrated pseudoplastic behaviour, as the  $n$  values were less than 1. The fermentation process, along with the inoculation of probiotics and/or the addition of persimmon puree, slightly increased the  $n$  values of the samples ( $P > 0.05$ ). However, this same process resulted in a reduction in the  $K$  values ( $P \leq 0.05$ ). The  $K$  values of the samples ranged from  $11.77 \pm 2.89$  to  $5.02 \pm 0.26$  (Pa.s) for IC and ICP, respectively. Similarly, Goktas (2023) and Sayar et al. (2022) reported that the addition of rowanberry pulp and blueberry decreased the  $K$  values of ice cream samples in a manner analogous to our study. However, Mehdiatabar et al. (2020) and Tsevdou et al. (2019) found that the incorporation of pumpkin puree and grape pulp into ice cream samples resulted in increased  $K$  values. Additionally, Homayouni & Norouzi (2016)

indicated that the fermentation of soy led to a reduction in the viscosity of the ice cream sample. In contrast, Aboulfazli et al. (2015) observed that the fermentation process resulted in an increase in  $K$  values for their samples. The reduction in the  $K$  values of the ice creams of PIC, FIC, and FICP may be attributed to a decline in pH (Goktas et al., 2022). Conversely, the decline in  $K$  values for the ice creams of ICP and ICFP may be linked to the composition of persimmon puree, which affects the water-binding capacity and subsequently, the rheological properties of the ice cream samples (Mehditabar et al., 2020; Tsevdou et al., 2019).

**Table 2.** Melting, rheological and textural properties of ice cream samples.

Ice Cream Samples	Melting Properties			Rheological Properties				Textural Properties	
	$T_{\text{onset}}$ (°C)	$T_{\text{peak}}$ (°C)	$T_{\text{end}}$ (°C)	$\Delta h$ (J/g)	$K$ (Pa.s <sup>n</sup> )	$n$	$R^2$	Hardness (g)	Adhesiveness
IC	-15.41 ± 0.69 <sup>a</sup>	-1.93 ± 0.20 <sup>a</sup>	4.84 ± 0.05 <sup>a,b</sup>	97.04 ± 4.47 <sup>a</sup>	11.77 ± 2.89 <sup>a</sup>	0.45 ± 0.03 <sup>a</sup>	0.99	498.12 ± 123.47 <sup>a</sup>	-85.80 ± 23.62 <sup>a</sup>
ICP	-15.30 ± 0.79 <sup>a</sup>	-2.04 ± 0.44 <sup>a</sup>	3.77 ± 0.13 <sup>b,c</sup>	116.45 ± 21.71 <sup>a</sup>	5.02 ± 0.26 <sup>b</sup>	0.51 ± 0.01 <sup>a</sup>	0.99	320.24 ± 23.19 <sup>a</sup>	-61.81 ± 1.08 <sup>a</sup>
PIC	-15.21 ± 0.35 <sup>a</sup>	-2.47 ± 0.07 <sup>a</sup>	3.23 ± 0.02 <sup>c</sup>	107.40 ± 3.68 <sup>a</sup>	5.12 ± 0.12 <sup>b</sup>	0.51 ± 0.01 <sup>a</sup>	0.99	397.66 ± 91.03 <sup>a</sup>	-61.10 ± 11.31 <sup>a</sup>
FIC	-14.93 ± 0.35 <sup>a</sup>	-1.69 ± 0.59 <sup>a</sup>	4.91 ± 0.40 <sup>a,b</sup>	109.20 ± 1.69 <sup>a</sup>	5.23 ± 0.47 <sup>b</sup>	0.53 ± 0.00 <sup>a</sup>	0.99	371.31 ± 121.87 <sup>a</sup>	-57.39 ± 9.94 <sup>a</sup>
ICFP	-14.69 ± 0.19 <sup>a</sup>	-1.43 ± 0.12 <sup>a</sup>	4.53 ± 0.35 <sup>a,b,c</sup>	116.00 ± 1.41 <sup>a</sup>	9.09 ± 0.90 <sup>a,b</sup>	0.46 ± 0.05 <sup>a</sup>	0.99	349.49 ± 49.56 <sup>a</sup>	-64.79 ± 7.10 <sup>a</sup>
FICP	-14.20 ± 0.63 <sup>a</sup>	-1.33 ± 0.52 <sup>a</sup>	5.22 ± 0.62 <sup>a</sup>	121.35 ± 14.78 <sup>a</sup>	6.68 ± 0.45 <sup>b</sup>	0.49 ± 0.02 <sup>a</sup>	0.99	287.54 ± 23.18 <sup>a</sup>	-55.02 ± 9.27 <sup>a</sup>

IC: Control ice cream (without any probiotic or persimmon puree), ICP: Ice cream with 20% persimmon puree, PIC: Probiotic ice cream, FIC: Fermented ice cream, ICFP: Ice cream with 20% fermented persimmon puree, FICP: Co-fermentation of ice cream and 20% persimmon puree. The presence of different letters in the same column indicates a statistically significant difference ( $P \leq 0.05$ ).



**Figure 2.** Flow behaviour of ice cream samples.

### Textural properties

Table 2 presents the textural parameters (hardness and adhesiveness) of the ice cream samples. The highest and lowest textural parameters were identified for IC and FICP, respectively. The fermentation process, probiotic inoculation and persimmon puree addition resulted in a reduction in the hardness values ( $P > 0.05$ ). Furthermore, similar outcomes were observed regarding the adhesiveness values. Concurrently, Karaman et al. (2014) and Hwang et al. (2009) demonstrated that the incorporation of persimmon puree and grape wine lees into the ice cream production process led to a decrease in the hardness values of the resulting ice cream samples. Additionally, Karaman et al. (2014) noted that the addition of persimmon puree elevated the adhesiveness values of the ice cream samples, whereas Hwang et al. (2009) found that the use of grape wine lees did not significantly affect the adhesiveness values. Conversely, Goktas (2023), Mehdiatabar et al. (2020), and Tsevdou et al. (2019) reported that the incorporation of rowanberry pulp, pumpkin puree, and grape pulp resulted in an increase in the hardness values of the ice cream samples. It is anticipated that the fermentation process will lead to an increase in the hardness of the ice cream (Sezer et al., 2022). However, the results of this study indicate that the hardness of the fermented ice creams was lower than that of the control group. This may be associated with total dry matter, which influences the viscosity and hardness values of the samples. Furthermore, the textural characteristics of the ice creams may have been influenced by the fermentation parameters, including inoculation, conditions, pH, and strains (Soukoulis et al., 2014).

### Probiotic viability

The viability of the probiotic was ascertained over a 120-day storage period (Figure 3). The initial inoculation comprised approximately  $8.77 \pm 0.06 \log \text{CFU g}^{-1}$  of the probiotic *L. rhamnosus* GG into the persimmon puree and/or ice cream mixes. The PIC ice cream sample contained only *L. rhamnosus* and did not undergo any fermentation process. The FIC ice cream sample was subjected to fermentation with *L. rhamnosus*. In the ICFP ice cream sample, the persimmon puree was fermented with *L. rhamnosus*, after which the resulting persimmon puree (20%) was mixed with the ice cream mix. In the FICP ice cream sample, the persimmon puree (20%) and ice cream mix were co-fermented with *L. rhamnosus*. All samples were aged for 24 h at 4°C. Following the ageing process, an increase in probiotic count was observed, with the probiotic counts for the PIC, FIC, and FICP ice cream samples determined to be  $8.85 \pm 0.00$ ,  $9.06 \pm 0.02$  and  $9.12 \pm 0.02 \log \text{CFU/g}$ , respectively. However, the reduction in the probiotic count of ICFP was related to the addition of fermented persimmon puree to the ice cream mix. The viability of the probiotics was found to decline in all samples during storage, as a consequence of cold storage effects. The fermentation process enhanced probiotic viability. The fermented ice cream samples FIC and FICP exhibited higher probiotic viability than PIC. Furthermore, the co-fermentation of persimmon puree and ice cream mix demonstrated a protective effect on probiotic viability. In this context, at the end of the 120-day storage period, the FICP ice cream sample exhibited the highest probiotic viability, with a count of approximately  $8.95 \pm 0.01 \log \text{CFU g}^{-1}$ . Notably, the FICP ice cream sample demonstrated probiotic viability that exceeded that of the initial inoculation. Many studies have demonstrated that the addition of various fruits enhances the viability of probiotics (Akca & Akpinar, 2021; dos Santos Cruken et al., 2017; Goktas, 2023; Öztürk et al., 2018). Furthermore, it has been stated that the fermentation process enhances the viability of probiotics (Akca & Akpinar, 2021; Golestani & Pourahmad, 2017; Homayouni & Norouzi, 2016).

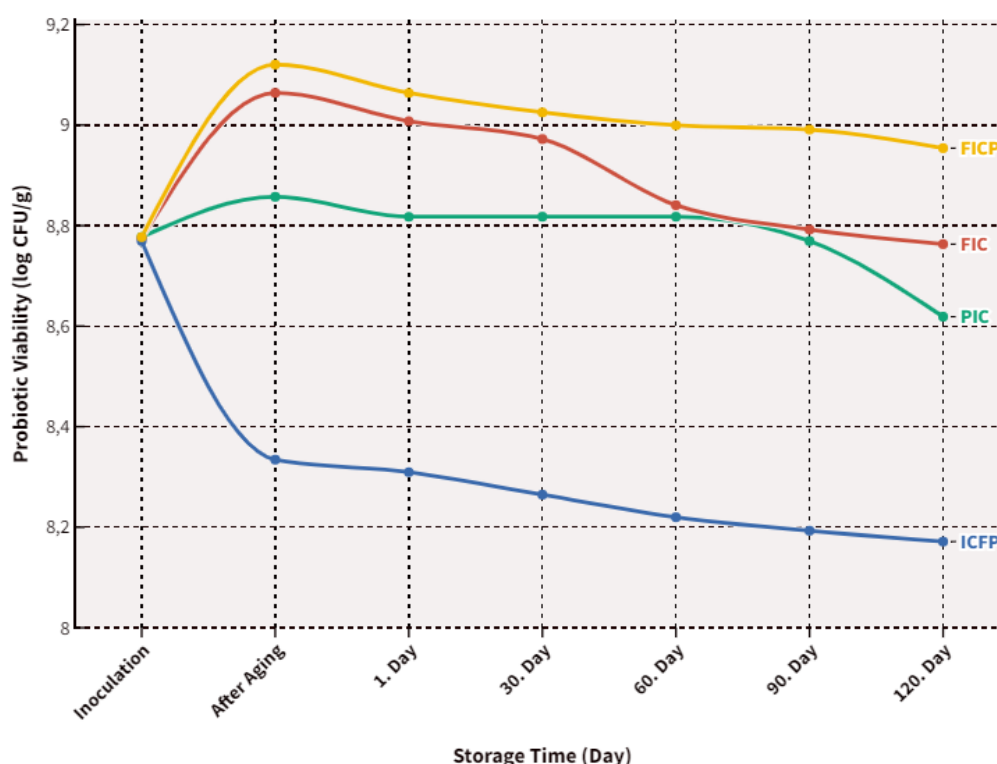


Figure 3. Probiotic viability during storage period.

### Bioactive properties

The Figure 4 illustrates the TPC and AP of the ice cream samples. The incorporation of persimmon puree resulted in a notable enhancement in the TPC and AP values of the samples ( $P \leq 0.05$ ). The addition of persimmon puree led to an approximately 2.5-fold increase in TPC values. Furthermore, the CUPRAC and DPPH results demonstrated that the AP of the samples increased with the addition of persimmon puree. Many studies have reported an increase in the TPC and AP values of ice cream with the addition of various fruits (Akca & Akpinar, 2021; Goktas, 2023; Hwang et al., 2009; Karaman et al., 2014; Öztürk et al., 2018; Sayar



et al., 2022). These results are consistent with the findings of the present work. This study has demonstrated that persimmon puree can be an important ingredient for enhancing the TPC and AP of ice cream. The inoculation of probiotics resulted in an increase in the TPC ( $P > 0.05$ ) and AP ( $P \leq 0.05$ ) of the ice cream sample (PIC). The fermentation process led to a notable increase in the TPC and AP of the samples (FIC, ICFP, FICP) ( $P \leq 0.05$ ). The TPC and AP values of fermented ice cream (FIC) exhibited a significant increase compared to the unfermented sample (IC) ( $P \leq 0.05$ ). The TPC and AP values of the samples in which persimmon puree was fermented and incorporated into the ice cream mix (ICFP) demonstrated an increase; however, no statistically significant difference was observed when compared to the ice cream sample in which persimmon puree was added (ICP) ( $P > 0.05$ ). The highest TPC and AP values were recorded for the FICP sample, in which the ice cream mix and persimmon puree were co-fermented. The TPC, CUPRAC, and DPPH results were determined to be  $242.57 \pm 11.52$  mg GAE  $100\text{g}^{-1}$ ,  $22.95 \pm 0.29$  mg TE  $100\text{g}^{-1}$  and  $48.02 \pm 3.27$  (%), respectively. During the fermentation process, several biochemical changes occur, resulting in the release of certain compounds. Fermentation can convert some compounds into other metabolites that enhance the antioxidant activity of foods (Gao et al., 2022). Consequently, enhancements are observed in the physicochemical, rheological, textural, sensory and bioactive properties of fermented products. For example, Donkor et al. (2007) reported that fermentation results in the release of certain bioactive components. Djadouni & Kihal (2012) observed that the antioxidant properties of soy milk products increased as a result of fermentation. In a separate study, Tsangalis et al. (2005) observed that the bioavailability of soy milk isoflavones was enhanced by the fermentation process. The findings of this study suggest that fermentation may be an effective tool for enhancing the bioactive characteristics of ice cream. Moreover, the bioactive characteristics of ice cream may be enhanced through the co-fermentation of fruits and the ice cream mix.

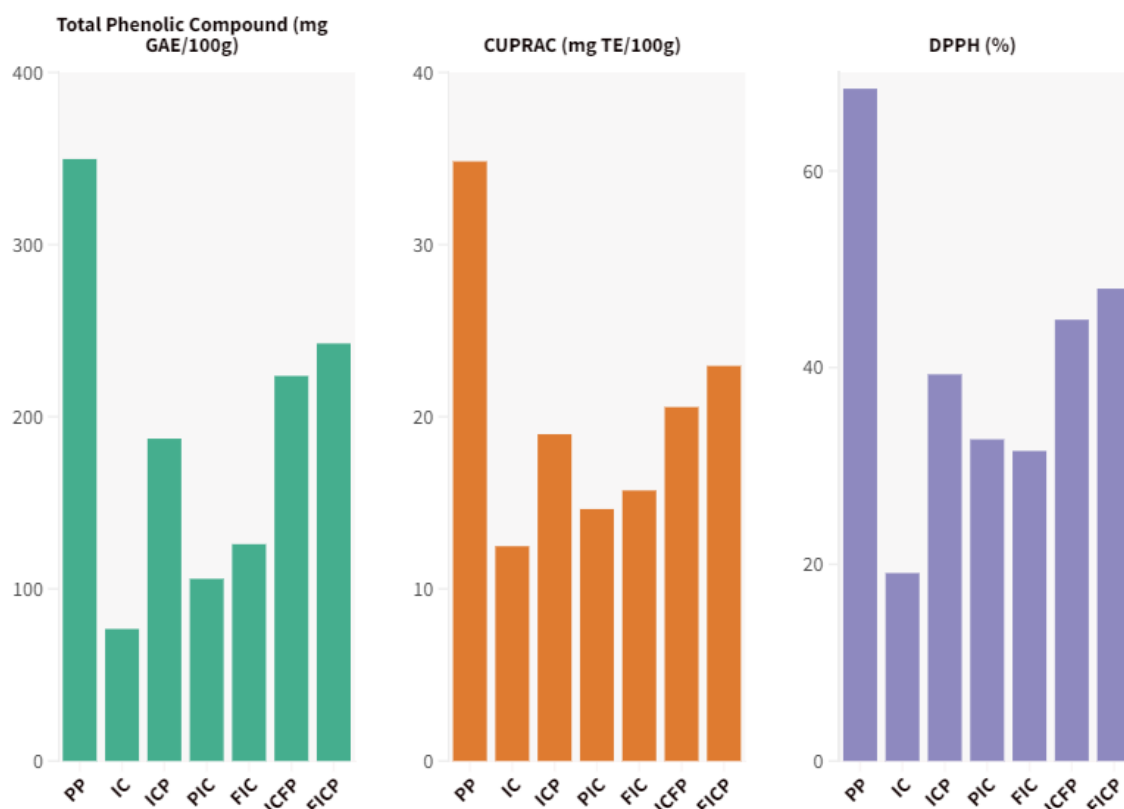


Figure 4. Bioactive properties of ice cream samples.

### Consumer acceptance

The samples were evaluated in accordance with the established criteria for sensory parameters, including appearance, odour, taste, strange taste, and general acceptability (Figure 5). The control ice cream sample (IC) was received the highest score among the sensory parameters. Nevertheless, no notable difference was identified concerning the parameters of appearance and odour ( $P > 0.05$ ). On the other hand, regarding the remaining parameters, statistically significant differences were identified exclusively between the control ice cream (IC) and the FICP ice cream sample ( $P \leq 0.05$ ), in which the ice cream mix and persimmon puree were



co-fermented. The FICP ice cream sample was rated less favorably in terms of taste and general acceptability, with a slightly higher incidence of strange taste. This phenomenon can be attributed to the reduction in pH levels that occurs as a consequence of fermentation. The co-fermentation of ice cream mix and persimmon puree resulted in a notable decrease in pH, accompanied by the formation of an acidic taste that was less well-received by consumers. However, fermentation led to the development of a multitude of the ice cream's distinctive characteristics. Therefore, production of fermented ice cream can be conducted by terminating the fermentation at the desired pH values for consumer preference. Similarly, Aboulfazli et al. (2015), Golestani & Pourahmad (2017), and Öztürk et al. (2018) reported that the panelists assigned lower scores to the samples of fermented ice cream, which was associated with a decrease in pH.

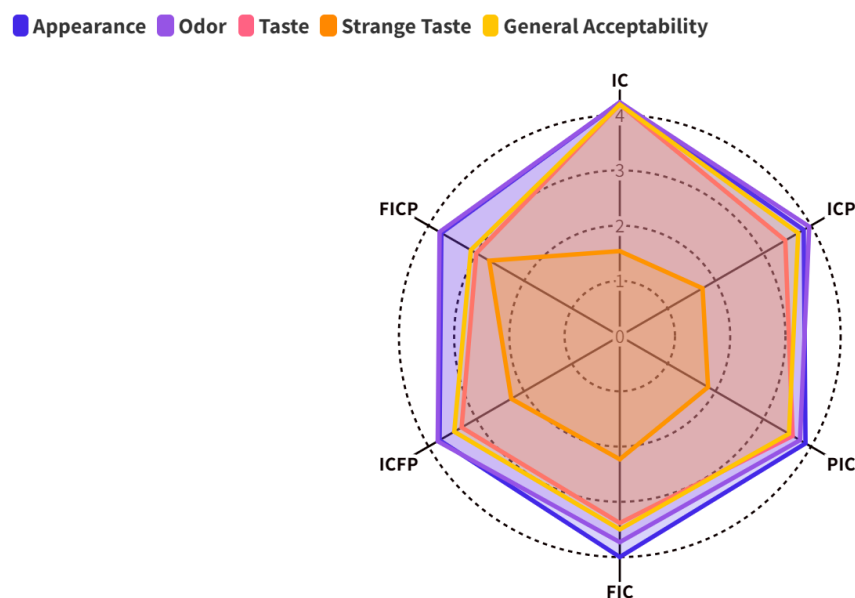


Figure 5. Consumer acceptance of ice cream samples.

## Conclusion

The addition of persimmon may prove an effective means of enhancing the bioactive properties of ice cream. The fermentation process was found to have a beneficial impact on the viability of the probiotics, with the highest levels of bioactive characteristics being observed in the co-fermentation. The addition of persimmon, probiotic inoculation and fermentation process, resulted in notable alterations to the rheological and textural attributes of the ice cream. These changes led to a reduction in the consistency coefficient and hardness values. The process of fermentation resulted in a reduction in pH values, which in turn led to a decline in consumer appreciation due to the formation of an acidic taste. Finally, the findings of this study emphasize that co-fermentation has the potential to enhance the bioactive and probiotic attributes of ice cream. Further studies may focus on aspects such as the impact of fermentation time and the use of different fruits on consumer enjoyment.

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