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The effect of the use of salep powder obtained from different wild orchid species in Turkey on the rheological, thermal, and sensory properties of ice cream

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Abstract

The aim of this study is to investigate the effect of salep species grown in different regions of Turkey on the rheological and microstructural properties of ice cream mix and the thermal and sensory properties of ice cream. For this purpose, ten different types of salep grown in different regions (Mersin, Yozgat, Muğla, Kahramanmaraş, Adana, Van, Muş) in the natural microflora of Turkey were used in a formulation of an ice cream mix. The flow behavior, frequency sweep, and 3-ITT rheological properties of ice cream mixes were studied. All samples showed shear thinning, viscoelastic solid-like, and recoverable character. The K and n values for the ice cream mixes were determined as 0.03-35.08 Pas^n and 0.33-0.80, respectively, and significantly differed according to salep varieties ($p < 0.05$). The zeta potential values of ice cream mix samples ranged from -25.87 mV to -33.95 mV and were significantly affected by salep varieties ($p < 0.05$). The use of different salep varieties significantly affected the thermal properties of ice cream such as freezing point temperature (Tf), temperature range (ΔT), and enthalpy of fusion (ΔH_f). In conclusion, the results of this study showed that the use of different salep varieties can significantly affect the rheological, thermal, and sensory properties of ice cream, and the selection of salep varieties may be vital for the desired quality of ice cream.

Keywords: salep; ice cream; rheology; zeta potential; DSC.

Practical Application: The effect of different salep types on ice cream.

1 Introduction

Salep is the dried tubers of wild orchid species, the largest flowering plant family of the *Orchidaceae* family. Wild orchid species are mostly grown in tropical and subtropical regions. Turkey is a country rich in plant diversity in Europe and the Middle East, with many different orchid species and genera on its fertile soil (Hürkan et al., 2019). In Turkey, there are at least 30 species of orchids belonging to 8-10 different genera and 154 different types of wild orchids (Durmuşkahya et al., 2015; Elife et al., 2017).

The physicochemical composition of salep varies according to the genus and species, and the chemical composition of salep is not standard (Tekinşen & Güner, 2010). Salep contains 1-36% starch, 8-12% moisture, 2-10% ash, 1-4% sugars, and 0.5-1.5% nitrogenous substances (Elife et al., 2017; Tekinşen & Güner, 2010). The main component of salep is a complex sugar called glucomannan, which consists of a combination of glucose and mannose and with frequent application in the food industry as a thickener and an emulsifier (Çalişkan, 2019). Glucomannan is salep's most important polysaccharide component (Farhoosh & Riazi, 2007). The salep tubers may have varying quantities of glucomannan (7-61%), starch (1-36%), moisture (6-12%), fat (2%), protein (3.5-7%), and ash (0.2-7%) based on the species (Acemi et al., 2019). Thanks to glucomannan content, salep is used as a gelling and emulsifying agent and improves taste and flavor in the food industry, especially for traditional beverages and desserts such as hot salep drinks and ice cream in Turkey (Elife et al., 2017; Kagan et al., 2014).

Ice cream has a very complex physicochemical system which consists of milk proteins, sugar, fat globules, ice crystals, air bubbles, and other substances in the ice cream mixture. The stability of this system is an important factor in the production of quality and suitable ice cream because ice cream is a homogenized emulsion (Shahsavari et al., 2022). In order to manufacture high-quality ice cream, the ice cream mix must be balanced in mass and thoroughly processed, with emulsifier and stabilizer components added to the content (Atik et al., 2021). Stabilizers in ice cream are used to increase the stability of the mix, increase its viscosity, prevent serum separation, prevent ice and lactose separation due to temperature fluctuations during storage, prevent moisture transfer from the product by binding water, create a good texture, and contribute to the melting properties of during production (Al et al., 2020).

Salep is an important component in the production of the necessary smooth and homogeneous structure in ice cream as a stabilizer. Furthermore, it is critical in manufacturing for delaying melting and reducing the creation of big ice crystals during freezing and storage of the mix. The rheological and

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thermal properties of the ice cream are affected from the glucomannan content of salep, which is significantly changed according to salep species (Georgiadis et al., 2012). Rheological qualities play an important role in determining ice cream quality. The rheological characteristics and behaviors of ice cream provide information on the product's physicochemical characteristics as well as determining the sensory properties of the product (Atik et al., 2021). Therefore, the selection of salep species is a critical issue in production of high-quality ice cream. There are some studies on the use of salep as an ice cream stabilizer in the literature (Farhoosh & Riazi, 2007; Kaya & Tekin, 2001; Kuş et al., 2005; Sen et al., 2019). However, there are not many detailed comparisons of the rheological (flow, dynamic, and 3-itt) behavior, zeta potential, and melting profile of the ice cream samples produced with different salep species.

This study aimed to investigate the rheological characteristics of ice creams produced using various types of salep. In addition, the effect on the sensory properties of the melting profile of ice cream was evaluated. The comparison of the different salep species of ice cream in terms of physicochemical, overrun, color, melting features, particle size and rheological properties, volatile compounds, and sensorial properties were also analyzed. Thus, the most suitable type of salep for ice cream formulation is determined based on species, and the cultivation of that species and its use as a stabilizer are supported.

2 Materials and methods

10 different species of salep samples used in the study were obtained from different regions of Turkey as dried. A sample of commercial salep was obtained from herbalists. 11 different types of salep used in the study were coded as shown in Table 1. Pasteurized cow's milk (3% milkfat), powdered sugar, milk cream (35% milkfat), salep (as a stabilizer), and egg yolk (as an emulsifier) were used for ice cream production. Xanthan gum (XG) was used instead of salep for the control ice cream sample. Xanthan gum and egg yolk were obtained from Sigma-Aldrich (Sigma Chemical Co., St. Louis, MO, USA), and pasteurized cow's milk, sugar, milk cream were purchased from the local market.

2.1 Sugar composition

The monosaccharide (glucose and mannose) contents of the salep samples were determined by the modified methods determined by Jung et al. (2022). Powder salep samples were weighed 0.5 g and hydrolyzed with 5 mL of 72% H_2SO_4 for 2 h at 30 °C. After completing the first hydrolysis step, the acid was diluted by adding 50 mL of distilled water, and the second hydrolysis step was completed in an autoclave at approximately 121 °C for 1 h. At the end of the process, the samples were cooled to room temperature, then the pH of the acid hydrolysates was adjusted to pH 7 with barium hydroxide buffer solution. Before HPLC, samples were purified from impurities by passing them through 0.45 µm syringe filters. Glucose and mannose contents of hydrolyzed salep samples were determined with a HPLC system (Agilent 12630). Measurements were carried out in the METU Central Laboratory. Metacarb-67C (300 × 7.8 mm) column RI detector was used in the analysis and dH_2O with a flow rate of 0.5 mL/min was used as the mobile phase. The injection volume is 10µ. Measurements were taken by comparing them with the standardized sugars.

2.2 Preparation of ice cream

11 species of salep samples (Table 1) and xanthan gum (used for a control ice cream mix) were used as stabilizers in ice cream production. Generally, 0.1-0.5% stabilizer is used in the ice cream mix. Bahramparvar & Tehrani (2011) used Salep in higher amounts than other stabilizers in ice cream formulation. In this study, based on the rheological data with different ratios of salep varieties, it was deemed appropriate to use 2% of the salep samples. The ice cream mixes are prepared by using pasteurized cow's milk (3% milkfat), 25% powdered sugar, 20% cream (35% fat), 2% salep (stabilizer), 3% egg yolk (emulsifier) in the product recipe. Ice cream samples contain 100 g of cow's milk, 25 grams of sugar, 20 g of milk cream, 3 g of egg yolk, 2 grams of salep, and 0.7 grams of XG for the control sample. Ice creams with 10% fat content were produced. During the ice cream production phase, cow's milk was brought to a constant temperature of 60 °C in a water bath. Powdered salep samples were added to milk at a rate of 2%. With the help of a magnetic stirrer, the mixture was stirred for 15 minutes at a temperature of 60 °C at 1000 rpm. Afterward, sugar, cream, and emulsifier were added and the mixture was pasteurized at 80 °C. Ultra turax Daihan at room temperature. It was homogenized for 3 minutes at 10,000 rpm with the HG-15D device. The resulting ice cream mix was left to rest for 24 hours at (0-4 °C). The rested ice cream samples were ice cream making process with DeLonghi IL Gelataio ICK5000, Treviso, Italy ice-

Table 1. Glucose and mannose contents of salep samples.

Sample code	Species	Regions	Glucose (g/100 g)	Mannose (g/100 g)	Total (g/100 g)
S-1	*Anacamptis pyramidalis*	Yozgat	22.9	15.8	38.7
S-2	*Orchis isaura*	Mersin	0.7	0.1	0.80
S-3	*Anacamptis palustris subsp. palustris*	Muş	7.9	3.8	11.7
S-4	*Orchis morio*	Adana	31.1	15.0	46.1
S-5	*Serapias vomeracea subsp. artemisiae*	Muğla	0.2	0.1	0.30
S-6	*Orchis italica*	Muğla	5.9	1.7	7.60
S-7	*Ophrys mammosa*	Adana	7.3	4.4	11.7
S-8	*Orchis sancta L.*	Kahramanmaraş	13.2	3.2	16.4
S-9	*Dactylorhiza euxina*	Van	0.1	0.02	0.12
S-10	*Ranunculus ficaria subsp. calthifolius*	Kahramanmaraş	0.07	0.01	0.08
S-11	Commercial Salep		16.9	10.5	27.4

cream device. The ice creams were put in a clean, sealable plastic container, covered with cling film, and stored at -18 °C. Ice cream mixes containing different types of salep and control ice cream mix containing xanthan gum are shown in Table 2.

2.3 Analysis of the ice cream mixes

Rheological analyzes

Flow behavior, dynamic, and 3-ITT rheological properties of ice cream mixes were determined using stress and temperature-controlled rheometer (MCR 302, Anton Paar, Australia). A parallel plate probe (PP50, Anton Paar, Australia) was used for rheological measurement. All measurements were performed at 25 °C and duplicated for the accuracy of the results.

Flow behavior rheological properties of ice cream samples were determined using a parallel plate probe (plate diameter 50 mm, gap size 0.5 mm) with the shear rate in the range 0.1-100 (s^{-1}). The measurement was carried out at a constant temperature of 25 °C, and 3 parallel studies were carried out for each sample. The data obtained from the rheological analysis were fitted to the power-law model, and nonlinear regression was used to calculate model parameters;

$$\tau = K \times \gamma^{n} \quad (1)$$

In Equation 1, the τ value represents the shear stress (Pa), K the consistency coefficient (Pas^n), γ the shear rate (s^{-1}), and n the flow behavior index. These parameters are shown in Table 3.

Parallel plate configuration was used for the dynamic rheological analysis of ice cream samples. Initially, the amplitude sweep test was performed between 0.1% and 100% strain to determine the linear viscoelastic region, and according to the result, the frequency sweep test was studied in the frequency range of 0.1-10 Hz and angular velocity of 0.1-64 s^{-1} (ω). Elastic modulus (G') and viscose modulus (G'') corresponding to angular velocity and frequency values were determined. The parameters for dynamic rheological properties were found using the power-law model and nonlinear regression;

$$G' = K'(\omega)^{n'} \quad (2)$$

$$G'' = K''(\omega)^{n''} \quad (3)$$

In Equations 2-3, the G′ value represents storage modulus (Pa), G″ value loss modulus (Pa), ω angular velocity value (s^{-1}), K′,

Table 2. Steady shear power-law parameters and zeta potential values of the ice-cream samples.

Sample code	K ($Pa.s^n$)[1]	n	R^2	Zeta Potential (mV)
IC-S1	35.08 ± 0.02	0.33 ± 0.00	0.99 ± 0.00	-26.63 ± 0.77
IC-S2	7.89 ± 0.00	0.51 ± 0.01	1.00 ± 0.00	-27.63 ± 1.73
IC-S3	3.34 ± 0.00	0.53 ± 0.00	1.00 ± 0.00	-30.25 ± 0.25
IC-S4	19.57 ± 0.03	0.36 ± 0.00	1.00 ± 0.00	-26.74 ± 1.12
IC-S5	6.43 ± 0.00	0.50 ± 0.00	1.00 ± 0.00	-28.95 ± 0.15
IC-S6	0.29 ± 0.00	0.74 ± 0.02	1.00 ± 0.00	-29.78 ± 2.62
IC-S7	7.19 ± 0.00	0.48 ± 0.00	1.00 ± 0.00	-29.45 ± 0.72
IC-S8	2.10 ± 0.00	0.59 ± 0.03	1.00 ± 0.00	-31.43 ± 1.00
IC-S9	3.95 ± 0.00	0.55 ± 0.00	1.00 ± 0.00	-30.50 ± 0.10
IC-S10	0.03 ± 0.00	0.80 ± 0.02	1.00 ± 0.00	-33.95 ± 0.15
IC-S11	11.35 ± 0.00	0.34 ± 0.01	1.00 ± 0.00	-27.02 ± 0.95
IC-C	3.39 ± 0.00	0.46 ± 0.00	1.00 ± 0.00	-25.87 ± 1.27

[1]K ($Pa.s^n$): consistency index values. n: the flow behavior index values.

Table 3. Power Law parameters are defining the dynamic rheological properties of the ice-cream samples.

	$G' = K'(\omega)^{n'}$ [1,2]			$G'' = K''(\omega)^{n''}$ [1,2]		
	K'	n'	R^2	K"	n"	R^2
IC-S1	25.97 ± 0.09	0.51 ± 0.01	1.00 ± 0.00	36.95 ± 0.10	0.27 ± 0.00	0.98 ± 0.00
IC-S2	2.50 ± 0.00	0.74 ± 0.02	1.00 ± 0.01	5.33 ± 0.01	0.45 ± 0.00	1.00 ± 0.03
IC-S3	4.29 ± 0.00	0.69 ± 0.01	1.00 ± 0.00	5.93 ± 0.00	0.13 ± 0.00	0.78 ± 0.03
IC-S4	17.02 ± 0.02	0.51 ± 0.00	1.00 ± 0.00	23.00 ± 0.06	0.28 ± 0.01	0.99 ± 0.02
IC-S5	6.63 ± 0.01	0.67 ± 0.01	1.00 ± 0.00	13.15 ± 0.02	0.41 ± 0.01	1.00 ± 0.00
IC-S6	3.76 ± 0.03	0.70 ± 0.01	1.00 ± 0.00	7.58 ± 0.01	0.44 ± 0.02	1.00 ± 0.02
IC-S7	6.27 ± 0.01	0.63 ± 0.02	1.00 ± 0.01	9.18 ± 0.01	0.22 ± 0.01	0.96 ± 0.01
IC-S8	7.20 ± 0.06	0.43 ± 0.02	0.99 ± 0.00	9.28 ± 0.01	0.40 ± 0.00	0.99 ± 0.00
IC-S9	0.88 ± 0.00	0.91 ± 0.03	1.00 ± 0.00	2.90 ± 0.00	0.54 ± 0.00	1.00 ± 0.01
IC-S10	0.29 ± 0.00	0.99 ± 0.00	1.00 ± 0.01	0.47 ± 0.00	0.74 ± 0.01	0.94 ± 0.00
IC-S11	9.89 ± 0.02	0.44 ± 0.00	1.00 ± 0.00	11.24 ± 0.02	0.38 ± 0.02	0.99 ± 0.00
IC-C	3.58 ± 0.00	0.56 ± 0.02	0.99 ± 0.00	1.92 ± 0.00	0.46 ± 0.04	0.98 ± 0.00

[1]G'(Pa): the storage modulus, G"(Pa): the loss modulus, [2]K' & K": consistency index values. n' & n": the flow behavior index values; ω: angular frequency (1/s): the angular velocity value.

K″ consistency coefficient values (Pas^n) and n′, n″ flow behavior index values.

3-ITT rheological properties of ice cream samples were determined as 0.5 s^{-1} as constant shear rate value and 150 s^{-1} as variable shear rate value. The linear viscoelastic region has been taken into consideration in the selection of the values, and the linear viscoelastic region of the samples ends at 50 s^{-1}. The ice cream samples were subjected to a very low shear rate (0.5 s^{-1}) for 100 s during the first-time interval. In the second time interval, 150 s^{-1} was exposed to the determined cutting force for 40 s. In the third time interval, the dynamic rheological behavior of the ice cream in the second time interval was tested by subjecting the samples to the low shear rate level in the first-time interval. For this purpose, the change in the viscoelastic solid structure (G′) of the samples was observed. The behavior of samples produced using salep in the third time interval was modelled using a second-order structural kinetic model (Equation 4);

$$\left[\frac{G' - G_e}{G_0 - G_e}\right]^{1-n} = (n-1)k * t - 1 \quad (4)$$

In the model, the G′ value indicates the change in the storage module (Pa), G_0 indicates the initial storage module value (Pa) in the 3rd time interval, G_e represents the storage module at the moment when the product fully recovered, in other words, the storage module (Pa) at the moment when the product is fully balanced, and k is the thixotropic velocity constant. The parameters are shown in Table 4.

Zeta potential

The zeta potential value of the samples was determined by the particle size measuring device (Zetasizer, Malvern Instruments, Worcestershire, UK). The samples were diluted 500-fold with ultrapure water before homogenization by stirring in an ultrasonic water bath for 1 min. The zeta potential of the samples was determined according to the dynamic light scattering technique (Tekin et al., 2020).

2.4 Analysis of the ice cream

Color

In the study, the color analyzes of the ice cream samples were calibrated with the CR-400 Chroma Meter, Konica, Minolta, Japan color measuring device, and the measurements were carried out in parallel from three different points. L* value indicates brightness (0-100), a* value indicates color change value from red (+) to green (-), and b* value indicates a color change from yellow (+) to blue (-). Standard deviations of L*, a*, b* values of ice creams made with different salep types are given in Table 5.

Table 4. Second-order structural kinetic model parameters for 3-ITT[1] of ice-cream samples.

	G_e[2]	G_0[2]	k × 1000[2]	G_e/G_o[2]	R^2
IC-S1	108.92 ± 0.11	72.20 ± 0.14	13.63	1.51	1.00 ± 0.00
IC-S2	34.10 ± 0.03	15.18 ± 0.00	20.13	2.25	0.99 ± 0.00
IC-S3	22.94 ± 0.09	10.14 ± 0.01	14.15	2.26	1.00 ± 0.00
IC-S4	79.66 ± 0.02	50.95 ± 0.10	13.44	1.56	1.00 ± 0.00
IC-S5	46.35 ± 0.05	21.32 ± 0.04	12.11	2.17	1.00 ± 0.00
IC-S6	24.87 ± 0.01	6.49 ± 0.00	20.85	3.83	1.00 ± 0.00
IC-S7	27.88 ± 0.02	13.82 ± 0.03	11.51	2.02	1.00 ± 0.00
IC-S8	29.72 ± 0.02	9.66 ± 0.01	18.33	3.08	1.00 ± 0.00
IC-S9	33.01 ± 0.01	11.09 ± 0.01	19.55	2.98	1.00 ± 0.00
IC-S10	2.46 ± 0.00	0.57 ± 0.00	8.07	4.32	1.00 ± 0.00
IC-S11	34.83 ± 0.03	24.33 ± 0.02	13.43	1.43	1.00 ± 0.00
IC-C	26.36 ± 0.02	9.17 ± 0.00	18.41	2.87	1.00 ± 0.00

[1]3-ITT: Three interval thixotropy test; [2]k: the thixotropic rate constant. G_0: the initial storage modulus in the third time interval; G_e: the equilibrium storage modulus.

Table 5. Thermal properties, overrun and color parameters of the ice cream samples.

Sample	T_f (°C)	T_0 (°C)	T_{end} (°C)	T_{peak} (°C)	ΔT (°C)	$ΔH_f$ (J/g)	Overrun (%)	L*	a*	b*
IC-S1	-4.72	-10.04	3.28	-0.79	13.75	134.3	38.71	77.41 ± 0.06^{bcd}	-1.60 ± 0.01^{cd}	19.28 ± 0.06^{abc}
IC-S2	-5.48	-11.99	1.60	-1.64	14.91	127.3	37.46	80.81 ± 0.04^{abc}	-1.46 ± 0.22^{cd}	19.84 ± 0.06^{a}
IC-S3	-5.18	-10.09	2.34	-1.2	13.49	125.3	35.44	76.75 ± 0.83^{bcd}	-1.20 ± 0.06^{bc}	19.67 ± 0.18^{ab}
IC-S4	-4.50	-8.35	2.65	-0.96	11.38	133.8	38.22	84.35 ± 0.80^{a}	-1.76 ± 0.11^{d}	20.10 ± 0.51^{a}
IC-S5	-4.94	-11.77	2.34	-1.06	16.6	137.3	33.15	79.52 ± 0.31^{abc}	-0.86 ± 0.02^{b}	20.25 ± 0.56^{a}
IC-S6	-6.47	-13.76	1.72	-1.63	15.42	101.6	33.23	77.63 ± 0.25^{bcd}	-1.22 ± 0.04^{bc}	17.59 ± 0.69^{bc}
IC-S7	-5.17	-12.45	2.09	-1.29	15.35	140.1	36.80	77.66 ± 0.33^{bcd}	-1.17 ± 0.03^{bc}	19.83 ± 0.33^{ab}
IC-S8	-4.84	-8.35	2.34	-1.26	10.94	115	34.67	75.27 ± 0.51^{cd}	-0.88 ± 0.04^{ab}	16.85 ± 0.95^{c}
IC-S9	-5.14	-11.52	1.78	-1.33	14.86	133.3	29.83	81.31 ± 0.36^{ab}	-1.77 ± 0.03^{d}	19.98 ± 0.30^{ab}
IC-S10	-5.28	-10.65	2.65	-1.56	13.43	118.9	30.62	74.20 ± 0.37^{d}	-0.49 ± 0.05^{a}	13.12 ± 0.03^{d}
IC-S11	-5.19	-11.58	2.47	-0.91	14.48	130.5	37.49	78.33 ± 0.46^{bcd}	-1.41 ± 0.08^{cd}	18.39 ± 0.36^{abc}
IC-C	-5.63	-11.46	5.08	-1.59	14.42	123.3	33.26	84.43 ± 0.60^{a}	-1.80 ± 0.14^{d}	20.20 ± 0.60^{a}

Different superscript letters show significant differences ($P < 0.05$) in a column.

Overrun

In order to determine the volume increase of the ice creams, ice cream samples were placed in the measuring cylinder, which was tared, in such a way that there was no space up to a certain volume. The same process was applied to each ice cream and weighed on a precision scale. Afterward, the ice creams were allowed to melt at room temperature. Melted ice creams were filled to the same volume in the same cylinder measuring cup. Afterward, the melted ice creams were weighed with precision scales. The equation used in the calculation of the volume increase of ice creams is indicated in Equation 5.

$$Overrun\ (\%) = \frac{W_2 - W_1}{W_1} \times 100 \quad (5)$$

Thermal properties

The thermal properties of ice cream samples were analyzed by a differential scanning calorimeter (DSC) by A DTA-DSC (differential scanning calorimetry) operating at atmospheric pressure (STA44gf3, Netzsch, Germany) according to the method reported by Hwang et al. (2009). Ice cream samples of 10 mg were placed in a pre-weighed aluminum sample pan, the pan was sealed using a Quick Press pan crimper (Tzero), and the thermal data were recorded from -20 to +50 °C in a nitrogen atmosphere with a heating rate of 1 °C/min. An empty pan was used as the reference. The flow rates of nitrogen gas for cooling were 50 mL/min. The onset temperatures (T_0), T_{end}, and T_f were determined. T_0 is considered as the intersection of the tangent and baseline to the left side of the melting peak. Freezing points were determined by using the temperature of the steepest slope. The enthalpy of fusion was calculated by extrapolating the baseline under the peak by connecting the flat baseline before and after the melting peak and integrating the peak above the baseline.

Sensory properties

Panelists consist of graduate students, doctoral students, and academics. After the panelists were informed about the product, samples were prepared for sensory evaluation. A table was used as a sensory evaluation form. The taste criteria of the ice cream samples were determined, such as aroma, cream taste, aftertaste, gummy structure, icy structure, roughness, foreign taste, color, melting in the mouth, general acceptability. In sensory evaluations, they were asked to score on a scale ranging from 1 to 5 points. It was evaluated with a scale of liking (1: very bad, 2: dislike, 3: not very bad, 4: good, and 5: very good).

2.5 Statistical analysis

The statistical analysis was performed with SPSS software (version 16; SPSS Inc., Chicago, IL, USA). All experiments were conducted at least in triplicate and the data were presented as means and standard deviations of each experiment. Duncan's multiple range test was used as the multiple comparison test for determining the statistical difference between means at the significance level of 0.05. Rheological parameters of fitted models and goodness of fit (coefficients of determination (R^2) were obtained using STATISTICA software (version 12; Statsoft, Tulsa, OK, USA). Curves were drawn by Microsoft Excel spreadsheet (version 2016; MicrosoftOffice, Redmond, WA, USA).

3 Results and discussion

3.1 Salep sugar

Table 1 indicated 11 different types of salep and their glucose and mannose content. The most important component of salep is glucomannan, which consists of glucose and mannose and acts as a stabilizer, and is found at glucomannan 7-61% in salep (Acemi et al., 2019). As seen in Table 1, glucose contents of salep samples were between 0.07 and 31.1 g/100 g salep whereas mannose contents of samples were between 0.01 and 15.8 g/100 g salep. The regions of salep samples were collected were presented with their sample numbers and the species names in Table 1.

As seen, the specie name of S1 samples is *Anacamptis pyramidalis* and has the highest mannose content (15.8 g/100 g) while the specie name of S4 is *Orchis morio* salep and has the highest glucose content (31.1 g/100 g). Also, the highest glucomannan content belongs to *Orchis morio* and followed by *Anacamptis pyramidalis*. Tekinşen & Güner (2010) determined *Orchis morio* salep as having the 2nd highest glucomannan amount among the various salep species that they studied glucomannan amounts of different salep types.

3.2 Rheological properties and zeta potential of ice cream mixes

Steady shear rheological properties of ice cream mixes

In this study, the data of the steady shear rheological properties were used to evaluate the effect of salep species on the flow curves of the ice cream mixes (Figure 1). Figure 1A showed that the slope of the shear rate versus shear stress graphs of the ice cream mixes decreased with increasing shear rate, demonstrating that the viscosity of all samples reduced with increasing shear rate (Figure 1B). As seen in Figure 1B, all ice cream mixes showed a clearly pseudoplastic (shear-thinning) behaviour (Carvalho et al., 2022). The highest viscosity value was the ice cream sample prepared with S1 salep (IC-S1). Figure 1 was consistent with previous studies about ice cream mixes (Dogan et al., 2013; Yazdi et al., 2020; Sharma et al., 2017). Shear-thinning flow properties were seen in the ice cream mixes, which is the expected flow behavior for an ice cream mix, and this behavior is associated with the breakdown of aggregate structures during the increase of shear rate (Zagorska et al., 2022). Kaya & Tekin (2001) determined the shear-thinning flow behavior for ice cream mixes with different salep content which is attributed to a complex involvement of partially broken-down micellar casein at the droplet surface in the homogenized ice-cream mix.

Power-law model parameters (K and n values) and determination coefficients (R^2) were calculated for all ice cream mixes containing 11 different species of salep and xanthan gum (Table 2). The Power-law model adequately described the flow behavior properties of ice cream mixes for all ice cream mixes ($R^2 > 0.99$). Table 2 showed that K and n values were 0.03-35.08 Pas^n and 0.33-0.80, respectively. n values lower than 1 indicated that all ice cream mixtures exhibited the non-Newtonian pseudoplastic

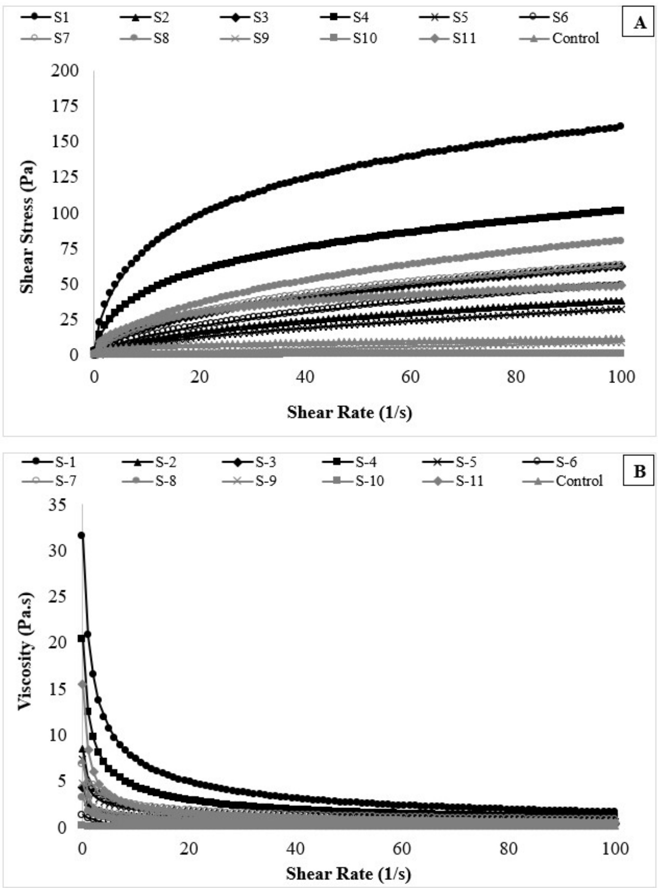

Figure 1. Steady shear rheological properties of the ice-cream samples contained with a different type of salep [flow curves (A), viscosity curves (B)].

flow behavior (Table 2). Ice cream mixes generally exhibit shear-thinning (pseudoplastic) behavior with flow behavior indexes of $0 < n < 1$, indicating the stability of the system characteristics under lower shear rate processing conditions and easier pumping of mix and the desired texture and mouthfeel of the end product (Javidi et al., 2016; Karaca et al., 2009; Kuş et al., 2005). The K value of the ice cream mixes were the most important parameter indicating melting resistant and texture properties of ice cream and significantly differed according to samples ($p < 0.05$). As seen in Table 1, the IC-S10 showed the lowest K value (0.03 Pas^n) and the highest n value (0.80), while the IC-S1 exhibited the highest K value (35.08 Pas^n) and the lowest n value (0.33). IC-S1 showed a strongest pseudoplastic character than that of other samples. The K value showed high correlation with total glucomannan content ($p = 0.78$). The samples containing high glucomannan content showed higher K value. Tekinşen & Güner (2010) reported that the viscosity of the samples containing higher glucomannan content showed higher viscosity.

The K values of the ice cream mix changed with salep species. These results can be explained by the higher K value of ice cream mixes prepared with S1, S4, and S11 compared to the control ice cream mix, which can improve the shear thinning properties of ice cream mixes of these salep varieties. This result could be due to the interaction between the glucomannan content of salep species and milk proteins (Şen et al., 2018). Thus, some salep species (S1, S4, S8, and S11) can be used for improving rheological properties in ice cream. When Turkmen et al. (2021) evaluated the rheological results of the ice creams they produced using different types of salep, they found that the K values of ice cream mixes produced with salep containing high glucomannan were higher.

Table 2 also shows the Zeta potential values used to estimate the stability of ice cream emulsions by measuring the level of electrical repulsion between particles. The zeta potential values of the ice cream mixes are between -25.87 mV and -33.95 mV. Due to the low repulsive force between the droplets, flocculation and close contact are avoided by a high zeta potential, whereas a low zeta potential implies low stability (Liu et al., 2011). All ice cream mixes with different type of salep indicated the high emulsion stability. The zeta potential analysis could be used for determining the surface charge density and interactions between proteins and polysaccharides in emulsions containing both charged biopolymers. In this study, the ice cream mixes containing high glucose and mannose content had also high emulsion stability, explaining with the interaction of milk protein and salep polysaccharides. The complexation of lactoferrin with pectin was investigated by Bengoechea et al. (2011), who discovered that the addition of polysaccharide caused the zeta potential of the protein dispersions to become more negative, which they attributed to the complexation of the protein and polysaccharide.

Dynamic rheological properties of ice cream mixes

The dynamic rheological properties of ice cream mixes containing different type of salep were investigated. Figure 2 showed the viscoelastic properties including storage and loss modulus as a function of angular frequency. The frequency sweep test can simulate the viscoelastic behavior of ice cream samples during chewing in the mouth (Zhang et al., 2018), which helps to comprehensively determine the effect of different salep types with different glucose and mannose content on ice cream quality. Increasing G' and G" values of samples with increasing frequency is evidence of gel-like behavior in ice cream samples (Kurt & Kahyaoglu, 2015). As seen in Figure 2, the value of G' of all samples was higher than the value of G'', indicating that the solid character of all ice cream samples is more dominant than the liquid character. Kurt et al. (2016) reported that the gel-like behavior of salep-based ice cream mixtures is evidenced by primarily elastic behavior ($G' > G''$) and no crossover point throughout the entire frequency range. Also, S1 and S4 samples had the highest G' values related to the highest glucose and mannose.

The dynamic rheological parameters (K', K", n', and n" values) were also calculated by using the power-law model (Table 3). The R^2 values of the model were found in the range of 0.97-0.98. As can be seen in Table 3, the K' and K" values of the samples were in the range of 0.29-25.97 Pas^n and 0.47-36.95 Pas^n, respectively; the values of n' and n" were found in the range of 0.43-0.99 and 0.13-0.74, respectively. K' values were lower than that of K" values of the ice cream mixes prepared with salep while an inverse result was the case in the ice cream mix

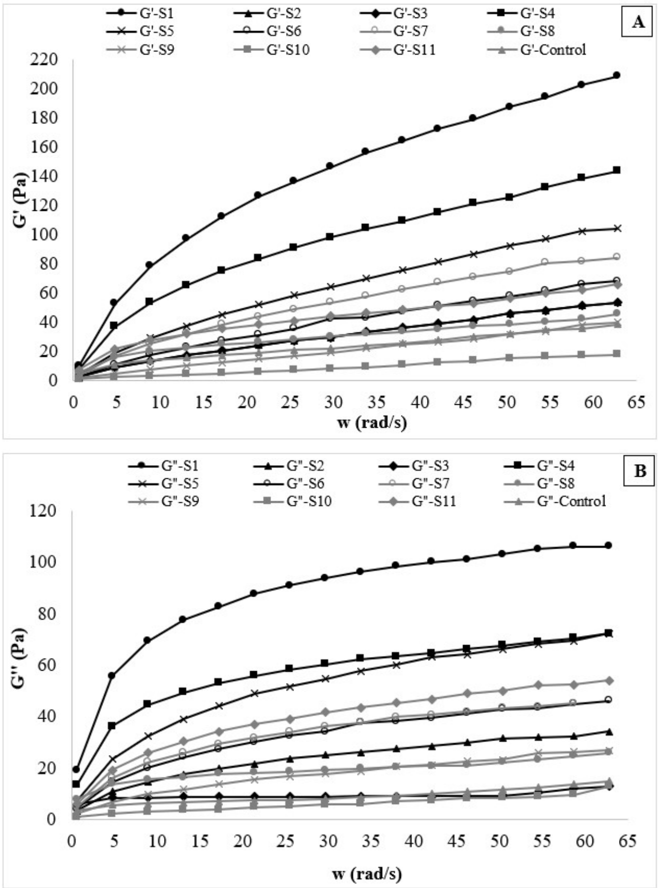

Figure 2. G′ and G′′ values versus angular velocity values of ice-cream samples contained with a different type of salep [G' (Pa): storage modulus (A), G"(Pa): loss modulus (B)].

(control) prepared with 0.4% xanthan gum, that is, it was seen that K' value was higher than K" value. All samples have different K' and K" values. The sample with the highest K' and K" values is S1 (glucose: 22.9 g/100 g and mannose:15.8 g/100 g) while the sample with the lowest is S10. The reason for this has been attributed to their different chemical compositions, mainly glucomannan and starch contents (Farhoosh & Riazi, 2007).

Thixotropic behavior of ice cream mixes

The 3-ITT simulates sudden and non-linear deformation of ice cream mixes. Therefore, this test gives information about the structural recovery of ice cream mixes. Figure 3 showed that all ice cream mixes in the third interval exhibited thixotropic behavior. As shown in Figure 3, the structural recovery tendency of that all ice cream mixes changed with the salep varieties. As we mentioned in the thixotropic properties of the ice cream mixes, the IC-S10 demonstrated the lowest structural recovery while IC-S1 indicated the highest recovery. The lowest structural recovery means that after deformation during food processing, such as homogenization or pumping, it could not be returned to the original structure fast due to high viscosity and strength structural molecular interactions (Razmkhah et al., 2016; Wang et al., 2020). Our ice cream samples stabilized by different

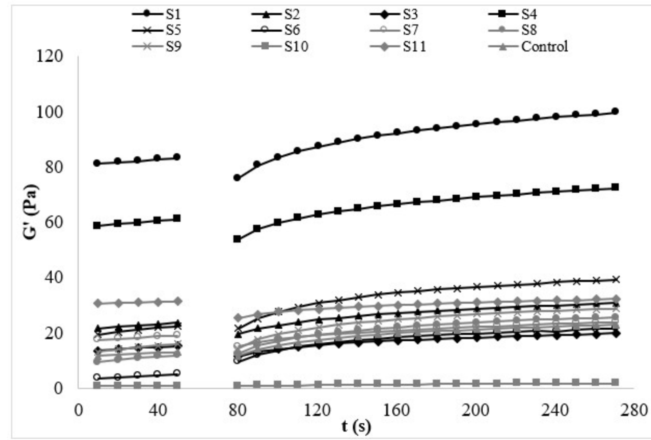

Figure 3. 3-ITT rheological properties of ice-cream samples contained with a different type of salep [G' (Pa): storage modulus)].

salep types and found that recoverable characteristics in the third interval are similar to previous findings by Atik et al. (2021) about the enhancement of thixotropic behavior of ice cream samples stabilized with cold-pressed chia oil by-products. The current investigation found that salep enhanced the thixotropic behavior of ice cream samples following rapid deformation.

Table 4 indicated the parameters (G_0, G_e, k × 1000, G_e/G_0) obtained with the second-order structural kinetic model. G_0, G_e, k×1000, G_e/G_0, and R^2 values were 0.57-72.20, 2.46-108.92, 8.07-20.85, 1.43-4.32, and 0.99-1.00. The greater the number of the Ge/G_0, the faster the recovery tendency. Sample S1 showed the highest G_0, G_e, G_e/G_0, and k × 1000 values, indicating that the sample S1 showed the highest thixotropic behavior. Because of the fact that the higher the thixotropic rate constant value means samples, the higher the tendency is to recover. These results indicated that salep could be applied to improve the recoverable properties of the ice cream mixes, however, the type of salep is important to affect the recoverable properties.

3.3 Analysis of the ice cream

Overrun

Overrun is the increase of the volume of the ice cream mixture generated by air entering it during partial freezing by mixing. There must be some air present so that the ice cream does not get too hard. Excess air decreases the quality, makes the granular structure visible in the mouth, and causes the ice cream to melt faster. Ice creams with less volume expansion have a tougher structure. The overrun values of the ice cream samples were shown in Table 5. The overrun values of ice cream samples S9 (29.83%) and S10 (30.62%) were found to be lower than the other samples. This is probably due to the lower sugar content of these two samples. Camelo-Silva et al. (2022) produced ice cream using by skimmed milk (ice cream 1) and concentrated milk (ice cream 2) and found that the overrun value of ice cream 2 had a higher than ice cream 1. This difference can be explained with the physicochemical composition of the ice cream 2 due to the protein content of ice cream 2. Barros et al. (2022) studied on the impact of substitution of milk by different proportions of concentrated whey and reported that the overrun values of ice cream samples were between 27% and 44%, and ice cream with a partial substitution of milk by concentrated whey demonstrated improved melting resistance.

Thermal properties

Thermal properties of ice cream is one of the vital properties affected overall ice cream quality. Thermal properties of ice cream samples were determined by DSC and showed in Table 5. T_f values of the samples ranged from -6.47 to -4.72. A statistical difference was observed between the T_f values of the samples. The T_f value is related to the solute concentration in the aqueous phase and the molecular weight of the solute in ice cream. T_f value decreased as the solute concentration increased and the molecular weight of the solutes decreased (Fuangpaiboon & Kijroongrojana, 2017; Hartel, 2002). The different T_f values of the samples can be explained by the different glucomannan contents. Different glucomannan content affected the solubility and molecular weight of the salep samples. In this case, different T_f values may have been obtained.

ΔT and ΔH values of the samples were found between 10.94-16.61 and 101.6-140.11 values, respectively. The ΔT values of the samples give an idea about the homogeneity of the ice cream samples. ΔH values show the energy value required for the melting of frozen water (Pintor-Jardines et al., 2018). Different ΔH values can be explained by the different interactions between water and macromolecules in the aqueous phase of ice cream of salep varieties (Atik et al., 2021; Pintor-Jardines et al., 2018). In samples with low ΔH value, salep may have interacted more tightly with water in ice cream and caused less water to freeze. Therefore, more frozen water form and less ΔH value emerged. These results showed that the difference in salep variety significantly affects the thermal properties of freezing, which is one of the main characteristics.

Color

The color parameters (L*, a*, b*) of ice cream samples were shown in Table 5. The highest L* value of the ice cream was observed in the Control sample (83.43) and the lowest L* value in the S10 sample (74.20). Although the L* values of the samples prepared with salep were lower than the control sample prepared with xanthan gum, the S4 sample and the control sample were not statistically different ($P > 0.05$). The highest a* value of the ice cream mixes was observed with -0.49 in S10 and the lowest a* value of the control sample was observed. A* values of ice cream samples below zero indicate that green color is more dominant than red in these samples. The effect of the some salep type used on the a* values of the mixes was found to be significant ($P < 0.05$). The b* values of the ice cream samples were found to be between 13.12-20.25. The highest b* value was observed in S5 and the lowest b* value was observed in S10 sample. The effects of the use of some different salep species as stabilizers on the b* values of the samples were found to be statistically significant ($P < 0.05$). It is thought that the difference between the color values of the samples, especially the L*, a* and b* values, is due to the type of wild orchid from which salep is obtained and the region where these plants are collected. When Ürkek (2021) examined the color values of ice cream samples containing salep and chai powder in different ratios, L*, a* and b* values were found to be 76.18-86.84, (-3.64)-(-2.71), 8.58-12.10, respectively.

Sensory properties

In Table 6, aroma, taste, creamy, gummy, icy, roughness, foreign taste, color, melting, and general acceptance of all ice cream samples were evaluated. Sensory parameters may vary according to the product studied and its ingredients. Yazdi et al. (2020) studied that the sensory properties (flavour, aftertaste, colour, texture and overall acceptability) of ice cream samples enriched with microencapsulated pistachiopeel extract (MPPE) and revealed that the addition of MPPE had no significant effect on the scores for colour, flavour and overall acceptability. On the other hand, Cais-Sokolińska et al. (2021) studied on the sensory properties of fried ripened curd cheese such as flavour (cooked, whey, sulfur, free fatty acid, cowy/phenolic, animal-like, and waxy/crayon aromatics), aroma (cream, butter and old milk), texture and mouthfeel, hardness, surface film, stickiness, friability, solubility, firmness, and graininess while Los et al. (2021) investigated the sensory properties (white

Table 6. The scores[1] relating to the sensory attributes of the ice cream samples.

	Aroma	Taste	Creamy	Gummy	Icy	Roughness	Foreign Taste	Color	Melting	General acceptance
IC-S1	4.4 ± 0.5	4.4 ± 0.5	4.6 ± 0.4	4.8 ± 0.1	4.6 ± 0.4	3.6 ± 1.1	4.6 ± 0.2	4.6 ± 0.5	4.8 ± 0.1	4.4 ± 0.1
IC-S2	4.2 ± 0.4	4.2 ± 0.7	4.2 ± 0.7	4.3 ± 0.6	3.6 ± 0.8	4.1 ± 0.6	4.5 ± 0.3	4.5 ± 0.8	4.3 ± 0.5	3.8 ± 0.7
IC-S3	4.0 ± 0.8	4.5 ± 0.8	4.3 ± 0.4	3.3 ± 1.2	4.5 ± 0.5	4.5 ± 0.5	4.8 ± 0.1	4.5 ± 0.8	4.3 ± 0.2	4.2 ± 0.7
IC-S4	4.8 ± 0.1	4.3 ± 0.5	4.3 ± 0.5	4.7 ± 0.5	4.2 ± 0.2	4.5 ± 0.5	4.7 ± 0.1	4.5 ± 0.5	4.3 ± 0.6	4.7 ± 0.2
IC-S5	4.0 ± 1.0	4.7 ± 0.1	4.0 ± 1.0	4.2 ± 0.7	4.0 ± 0.8	4.0 ± 0.9	4.5 ± 0.4	4.2 ± 0.7	4.0 ± 0.7	3.8 ± 0.9
IC-S6	4.2 ± 0.7	4.5 ± 0.3	4.3 ± 0.5	3.7 ± 0.8	4.3 ± 0.5	4.0 ± 1.1	4.3 ± 0.6	4.3 ± 1.0	4.2 ± 0.3	3.5 ± 1.6
IC-S7	3.8 ± 0.7	3.7 ± 1.4	3.8 ± 1.3	3.3 ± 1.3	3.7 ± 1.0	3.8 ± 1.1	3.8 ± 1.3	4.3 ± 1.2	3.8 ± 0.7	3.6 ± 1.0
IC-S8	4.0 ± 0.8	4.2 ± 0.5	4.5 ± 0.5	4.3 ± 0.2	3.5 ± 1.3	4.2 ± 0.2	3.7 ± 1.6	4.1 ± 1.1	3.7 ± 1.0	3.8 ± 0.9
IC-S9	4.7 ± 0.1	4.5 ± 0.4	3.7 ± 1.0	4.2 ± 0.7	3.7 ± 1.0	3.5 ± 1.2	3.5 ± 1.0	4.2 ± 0.7	4.2 ± 0.7	3.2 ± 0.7
IC-S10	3.5 ± 1.0	4.0 ± 0.9	3.8 ± 1.1	1.7 ± 0.5	2.2 ± 0.7	2.3 ± 0.5	3.5 ± 1.2	4.0 ± 1.1	2.5 ± 0.8	2.7 ± 0.8
IC-S11	3.3 ± 1.2	3.3 ± 1.4	3.5 ± 1.2	3.7 ± 1.7	3.0 ± 1.4	3.8 ± 1.3	3.8 ± 1.6	4.2 ± 1.1	3.5 ± 0.8	3.7 ± 1.0
IC-C	3.5 ± 0.8	3.7 ± 1.2	3.2 ± 0.6	3.0 ± 0.5	4.0 ± 1.0	4.3 ± 0.2	3.8 ± 0.3	4.5 ± 0.7	3.2 ± 0.4	4.0 ± 0.8

[1]Scores were on a scale of 1-5; values are the mean ± SD; different superscript letters show significant differences ($P < 0.05$) in a column.

colour, mycelium density, yellow colour, consistency at 23 °C, ammonia odour, butter odour, yoghurt odour and mould odour, rind hardness, velvety rind, creaminess, greasiness, bitter taste, acid taste, ammonia taste, bitter after taste, ammonia after taste and mould after taste) of Camembert-type cheeses.

As seen in Table 6, sensory evaluation of all ice cream samples showed that all ice cream sample are generally acceptable except for S-10. While S-4 could be preferred with respect to the aroma and roughness, S-1 could be preferred based on creamy, gummy, icy, color, and melting and S-8 had generally the high scores. Therefore, the content of glucose and mannose of S-1, S-4 and S-8 samples were higher than the other ice cream samples, indicating that sugar content is important parameter of consumer acceptance.

4 Conclusion

The salep varieties contained different amounts of glucomannan. The rheological properties of the ice cream mix and the thermal and sensory properties of the ice cream were significantly affected by the difference in salep variety. All ice cream samples exhibited pseudoplastic, viscoelastic liquid and recoverable characters. The samples containing high glucomannan content showed higher K, K', K" value than those of ice cream samples formulating with lower glucomannan content salep. Samples with higher K value had higher overall sensory score. The results of this study indicated that the difference in salep variety can significantly affect the quality characteristics of ice cream and it is essential to determine the right salep variety in the production of quality ice cream.

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