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# Use of Hemp Seed Milk in Ice Cream and Its Effect on Physicochemical, Rheological, Bioactive and Sensorial Properties

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## ABSTRACT

The nutritional and positive effects of plant-based milks on human health are increasingly studied, and their integration into various food formulations has become essential in new product development. In this study, hemp seed milk was added to the ice cream mix as a functional ingredient and a replacement for cow's milk (0-25-50-75-100%) to investigate its effect on the physicochemical, rheological, textural, functional, and sensory properties of ice cream. As the ratio of hemp seed milk with a high protein content increased in the mix, ice cream samples with higher protein content were obtained compared to the control sample. The use of hemp seed milk reduced the first dripping time and increased in the melting rate of ice creams, causing them to melt in a greater mass. However, statistically insignificant changes in the hardness parameter were observed as their textural properties. The apparent viscosity values of ice creams decreased, and the difference between elastic (G') and viscous (G'') properties also reduced compared to the control ice cream. Among ice creams with hemp seed milk, the sample containing 25% hemp seed milk received the highest sensory score. Considering the functional properties, as the hemp seed milk ratio increased, ice creams with higher total phenolic contents and antioxidant activities were obtained compared to the control sample. In conclusion, taking into account its favorable sensory properties, using a 25% hemp seed milk ratio can be suggested as a plant-based milk option that delivers desirable physicochemical and functional characteristics to ice cream.

Keywords: Hemp seed milk, Seed-based milk, Vegetable milk, Ice cream, Functional

# Dondurmada Kenevir Tohumu Sütü Kullanımı ve Fizikokimyasal, Reolojik, Biyoaktif ve Duyusal Özellikleri Üzerine Etkisi

# ÖΖ

Bitki bazlı sütlerin insan sağlığı üzerindeki besleyici ve olumlu etkileri giderek daha fazla araştırılmakta ve bunların çeşitli gida formülasyonlarına entegrasyonu, yeni ürün geliştirmede önemli bir hale gelmektedir. Bu çalışmada, kenevir tohumu sütü, fonksiyonel bir bileşen ve inek sütü yerine (0-25-50-75-100%) kullanılmak üzere dondurma karışımına eklenerek, dondurmanın fizikokimyasal, reolojik, tekstürel, fonksiyonel ve duyusal özellikleri üzerindeki etkileri araştırılmıştır. Dondurma karışımında protein içeriği yüksek kenevir tohumu sütü oranı arttıkça kontrol dondurmaya göre daha yüksek protein içeriğine sahip dondurmalar elde edilmiştir. Kenevir tohumu sütü kullanımı, dondurmaların ilk damlama süresini kısaltıp erime hızını artırarak daha büyük kütlede erimeye neden olmuştur. Ancak tekstürel özelikler incelendiğinde sertlik parametresi üzerinde istatistiksel olarak anlamlı bir değişiklik gözlenmemiştir. Dondurmaların görünür viskozite değerleri azalmış, elastik (G') ve viskoz (G'') özellikleri arasındaki fark da kontrol dondurmasına göre azalmıştır. Kenevir tohumu sütü ilave edilen dondurmalar arasında %25 kenevir tohumu sütü içeren dondurma en yüksek duyusal değerlendirme puanını almıştır. Fonksiyonel özellikler dikkate alındığında ise kenevir tohumu sütü oranı arttıkça kontrol dondurmaya göre daha yüksek toplam fenolik ve antioksidan aktiviteye sahip dondurmalar elde edilmiştir. Sonuç olarak %25 kenevir tohumu sütü oranı dondurmanın hem fizikokimyasal ve

fonksiyonel özelliklerini geliştiren hem de duyusal açıdan beğenilen bitki bazlı süt olarak önerilebilir.

Anahtar Kelimeler: Kenevir tohumu sütü, Tohum bazlı sütler, Bitkisel sütler, Dondurma, Fonksiyonel

# INTRODUCTION

Milk and dairy products constitute an important part of our diet. However, interest in plant-based products is increasing due to reasons such as the adverse effects of animal food production on the environment, the danger of insufficient animal food sources for the increasing population, lactose intolerance, adverse health effects, and new lifestyles including vegetarian and vegan diets [1-3]. Plant-based milk substitutes, or plant extracts, are water-soluble extracts of cereals (oats, rice), legumes (chickpea, soybeans), nuts (almonds, coconut, cashew nut, hazelnuts), seeds (sesame, sunflower, hemp, flaxseed) and pseudocereals (quinoa, teff) [4, 5]. Plantbased milks are not only consumed as beverages but also used as ingredients in food formulations. There are many studies in the literature investigating the use of plant-based milks in a fermented beverage [6, 7], yoghurt [8, 9], pudding [1, 10], cheese [10, 11], and ice cream [12-14] formulations. Since the sugar used in ice cream can mask the unfamiliar taste of plant-based ingredients, ice cream is a suitable food matrix in which plant-based ingredients can be included [15].

The cultivation and production of hemp (Cannabis sativa L.) were under control for years due to the psychoactive substance (delta-9 tetrahydrocannabinol, THC) it contains [16]. However, in the production of hempbased food products, materials obtained from the industrial hemp plant containing 0.3% to 1.5% THC are used. Therefore, unlike Marijuana, which has psychoactive properties including 5%-10% or more THC, industrial hemp can be used legally [17]. Hemp seed is a nutritional valuable seed due to its protein (20-25%), lipids (25-35%), carbohydrate (20-30%), insoluble fiber (10-15%), and ash (4-7%) content, which varies depending on the variety. Hemp seed is also vital for human nutrition, mainly due to its total polyphenols and essential fatty acids (linoleic acid-LA, C18:2,  $\omega$ -6 and  $\alpha$ linolenic acid-ALA, C18:3, ω-3) [18, 19]. Hemp seeds also a rich source of antioxidants are and phytochemicals that positively affect health [20]. Therefore, hemp-based materials such as protein, oil, flour, and milk can be used as functional ingredients in producing functional food products [20, 21].

Hemp seed milk is one of seed-based plant milk and is an oil-in-water emulsion resulting from the extraction of hemp seeds with water [22]. There are studies examining hemp seed milk only as a beverage [23-25] or examining the effects of some technological processes, such as ultrasound [26, 27], homogenization [16, 26], enzyme applications [28] and heat treatment [29], on the stability of hemp seed milk. Studies also investigate its use as an ingredient in different food formulations to develop new products. The possibilities of using hemp seed milk in the production of products such as ayran [9], yoghurt [30], ice cream [31] and kefir [32] was investigated.

In this study, because the ice cream mix already included stabilizers and emulsifiers, it was considered a food matrix that did not require any further processing to stabilize hemp seed milk. Differing from previous research, this study focused on evaluating the impact of varying hemp seed milk to cow's milk ratios, rather than just a single formulation, by analyzing how these ratios influenced the structure of the resulting ice cream. The research investigated the use of hemp seed milk as a replacement for cow's milk in ice cream formulations without the need for additional processing- and examined its effects on the physicochemical, rheological, sensory, and functional properties of the ice cream.

## **MATERIALS and METHODS**

## Materials

The raw materials used in ice cream production were purchased from the following companies in Türkiye: skim milk powder (Enka Süt, Adana), sugar (Doğuş Çay, Afyonkarahisar), vanilla (Dr. Oetker, İzmir), emulsifier and stabilizer mixture (Danisco, İstanbul) and milk cream with 35% milk fat content (Şok Market, Samsun). Hemp seeds were supplied from Kenevir Araştırmaları Enstitüsü (Ondokuz Mayıs University, Samsun). To obtain hemp seed milk, hemp seeds (20%, w/v) were mixed with distilled water and mixed with Ultra Turrax homogenizer (Ika Homogenizer T-25 basic Ultra Turrax, Staufen im Breisgau, Baden-Württemberg, Germany) at 10000 rpm for 10 min. Then, hemp seed milk passed through a 120-mesh sieve.

## Methods

## Ice Cream Production

The ice cream mix was calculated to contain 30% total dry matter and 8% fat. The ingredients used in the ice cream mix were as listed: 15% sugar, 0.75% stabilizer, and the remainder were cow milk (CM) or hemp seed milk (HM) and various combinations of them (CM-HM ratios: 100-0 (Control), 75-25, 50-50, 25-75, 0-100%). Besides, to achieve 8 g/100 g fat content in ice cream formulations, milk cream with 35% milk fat content was used. The milk was heated to approximately 45°C, and powder ingredients were added. The mixture was then heated and homogenized at 10000 rpm for 10 minutes (Ika Homogenizer T-25 basic Ultra Turrax, Staufen im Germany). Breisgau, Baden-Württemberg, The homogenized mix was pasteurized in a water bath at 80°C for 10 minutes. Ice cream mixes were cooled to 4°C and then aged overnight at 4°C.

#### **Physicochemical Analyses of Samples**

The dry matter and ash content of the ice cream samples were determined gravimetrically by keeping them in an oven at 105 °C and a furnace at 550°C, respectively [33]. Kjeldahl method was applied to determine protein content [34]. After the total nitrogen amount was found, the protein amount was calculated by multiplying it by 6.38. Fat content was determined using the Gerber assay [35]. The pH values of the ice cream mixes were measured with a pH meter (Eutech Instruments, pH 700, Waltham, Massachusetts, USA).

$$\Delta \mathsf{E}^* = \sqrt{(L_2 - {L_1}^2) + (a_2 - a_1)^2 + (b_2 - b_1)^2}$$

## **Rheological Properties of Ice Cream Mix**

The rheological properties of ice cream samples were determined using a Haake Mars III (Thermo Scientific, Waltham, Massachusetts, USA) device with conical and plate geometry (35 mm diameter, 0.104 mm gap, 2° angle) according to Kurt and Atalar [35]. The steady and viscoelastic properties of the ice creams were measured at +4°C. In addition, the ice cream samples were loaded onto the geometric probe and waited for 2 minutes for structure recovery and temperature equilibrium before each measurement. Shear rates from 0.1 s<sup>-1</sup> to 100 s<sup>-1</sup> (increasing) were applied to the samples for 3 min to determine the flow behavior graphs and apparent viscosity values (at a 50 s<sup>-1</sup> shear rate). The flow behavior was analyzed by using the Herschel–Bulkley model based on the following Equation 2:

$$\tau = \tau_{\rm Y} + K \gamma^{\cdot n} \tag{2}$$

where  $\tau$  is the shear stress (Pa),  $\tau_y$  is the yield stress (Pa), K is the consistency coefficient (Pa s),  $\gamma$  is the shear rate (s<sup>-1</sup>) and *n* is the flow behavior index (dimensionless).

Dynamic rheological measurements were made to determine ice cream samples' viscoelastic properties, and storage (G') and loss modulus (G") values were obtained. For this purpose, measurements were made at a shear stress of 0.2 Pa (linear viscoelastic area determined by stress sweep analysis) and a frequency range of 0.1-10 Hz.

## **Textural Properties of Ice Cream Mix**

A texture analyzer (TA-XT2 Stable Micro Systems Co., Ltd., Surrey, UK) was used to determine the hardness value of ice creams stored in a plastic container at -18°C for 24 h. The analysis was carried out using a cylindrical probe with a diameter of 5 mm, penetration depth of 5 mm, and penetration speed of 1.0 mm/s. The hardness

Color values of ice cream samples were determined with a color measuring device (Minolta Chroma Meter CR-400, Tokyo, Japan) using CIELAB system. The "L\*" value represents brightness (0: black, 100: white), the "a\*" value represents red/green (+: red, -: green), and the "b\*" value represents yellow/blue (+: yellow, -: blue). The color difference value ( $\Delta E^*$ ) value, which shows the difference of the ice creams with hempseed milk (L<sub>2</sub>, a<sub>2</sub>, b<sub>2</sub>) from the control ice cream (L<sub>1</sub>, a<sub>1</sub>, b<sub>1</sub>) in terms of color, was calculated according to Equation 1 given below;

(1)

is expressed as the peak pressure force (N) during penetration [35].

## **Melting Properties of Ice Cream Mix**

The first dripping time (min) and melting rate (g/min) were determined according to the method reported by Aboulfazli, Baba [36]. The hardened ice cream sample (~30 g) was left to melt on a 1.34 mm sieve on top of a pre-tared beaker in a cabinet set at  $25^{\circ}$ C, and the amount of ice cream melted into the beaker was recorded at a 5-minute interval for 45 min. The percentage of ice cream drip-off was calculated using the following Equation 3 [37]:

Drip-off (%) = 
$$(m_d/m_s) \times 100$$
 (3)

where  $m_d$  is the weight of the ice cream drip-off (g) at the end of 30 min, and  $m_s$  is the weight of the ice cream sample (g) at the beginning of the analysis.

## **Functional Properties of Ice Cream Samples**

Bioactive compound extraction from ice creams was carried out according to the method of Gul, Atalar [8] with some modifications. The samples were mixed with 80% methanol at a ratio of 1:1 and shaken for 1 hour at ambient temperature. Then samples were centrifuged (Nüve-Bench Top Centrifuge, NF 1200R, Ankara, Türkiye) at 8,000xg for 30 min at 4°C. Suspensions were filtered with Whatman filter paper no. 1 and the filtrate was used for total phenolic content (TPC) and 1,1-diphenyl-2-picrylhydrazil (DPPH) analysis.

The filtrates were mixed with Folin reagent to determine total phenolic component determination, and then 2.5 mL of 7.5% (w/v) Na<sub>2</sub>CO<sub>3</sub> was added. After keeping them in a dark condition for 30 min, absorbance was measured at 760 nm using a UV/VIS spectrophotometer (Shimadzu UV-1800, Tokyo, Japan). Results were calculated as Gallic acid equivalent using the calibration curve (R<sup>2</sup>:0.9998) given below (Equation 4);

## TPC (mg GAE/100g) = [(Absorbance - 0.179)/0.0123] $\times$ Dilution factor (4)

To determine DPPH activity, 4.9 mL of DPPH agent was added to 0.1 ml of filtrate, and the absorbance was read at 517 nm using a UV/VIS spectrophotometer

(Shimadzu UV-1800, Japan) after waiting in the dark for 30 min. Results were calculated using the calibration

curve (R<sup>2</sup>:0.9997) below based on the Trolox® equivalent (Equation 5);

Trolox (mM) = [(Absorbance + 0.5998)/(56.608)]×Dilution factor

### **Sensorial Properties of Ice Cream Samples**

Ice cream samples stored at -18°C for 25 days were evaluated using a hedonic scaling method for color/ 5=desired), (1=undesired, appearance texture (1=undesired, 9=desired), taste and aroma (1=undesired, 9=desired) and general acceptability [35]. Thirty panelists (academic staff and graduate students) at Ondokuz Mayis University Food Engineering Department participated in the sensorial evaluation test.

## **Statistical Analysis**

The data was analyzed using IBM SPSS software (Version 20, Chicago, USA). The one-way ANOVA and Duncan's post-hoc test at a 5% significance level were conducted.

## **RESULTS and DISCUSSION**

#### **Physicochemical Properties of Samples**

The physicochemical compositions of cow and hemp seed milk used for ice cream making are given in Table 1.

Table 1. Physicochemical properties of cow and hemp seed milk							
Physicochemical properties	Cow milk	Hemp seed milk					
Dry matter, %	11.94±0.09	12.13±0.04					
Ash, %	1.70±0.28	0.73±0.08					
Protein, %	2.50±0.44	4.67±0.03					
Fat, %	3.45 0.07	9.30±0.14					
рН	6.32±0.01	6.66±0.01					
L* (lightness/ darkness)	90.10±0.20	87.70±0.22					
a *(red/green)	-2 09+0 01	-0 71+0 23					

4.10±0.17

\* L\*: light (100) to dark (0), a\*: red (+a\*) to green (-a\*), b\*: blue (-b\*) to yellow (+b\*)

The dry matter content of hemp seed milk, prepared with 20% hemp seeds to obtain the dry matter similar to cow's milk (11.94%±0.09), was determined as 12.13%. In the study by Gram and Mortas (2023), the dry matter of hemp seed milk prepared at a 1:5 (seed: distilled water) ratio was around 10.3-10.46%. Protein, fat, and pH values were higher in hemp seed milk than cow milk. On the contrary, the ash amount and L\* value were found to be higher in cow's milk. Since the composition of hemp milk varies depending on the amount of hemp seeds used, hemp milk with different compositions has

b\* (blue/yellow)

been obtained in the literature. The protein, fat and ash contents of the hempseed milk samples used were found at 0.83-6.96%, 1.25-18.02% and 0.47-0.664%, respectively in studies [9, 16, 25]. Considering these minimum and maximum values in the literature, the chemical composition of the hemp seed milk produced was within the specified range.

9.28±0.39

The physicochemical properties of ice cream samples are given in Table 2.

Deremetere	Hemp Seed Milk Concentration								
Parameters	Control	25%	50%	75%	100%				
Dry matter, %	30.13±0.08 <sup>bc</sup>	29.89±0.04 <sup>b</sup>	29.52±0.10 <sup>a</sup>	29.98±0.10 <sup>b</sup>	30.42±0.02 <sup>c</sup>				
Ash, %	0.58±0.06 <sup>a</sup>	0.54±0.09 <sup>a</sup>	0.56±0.01ª	0.55±0.04ª	0.70±0.01ª				
Protein, %	2.47±0.37 <sup>a</sup>	2.81±0.07 <sup>ab</sup>	3.10±0.03 <sup>ab</sup>	3.50±0.10 <sup>bc</sup>	3.94±0.04 <sup>c</sup>				
Fat, %	8.40±0.28 <sup>a</sup>	8.50±0.14 <sup>a</sup>	8.30±0.14 <sup>a</sup>	8.60±0.01ª	8.70±0.14 <sup>a</sup>				
pH	6.29±0.01 <sup>a</sup>	6.66±0.01 <sup>b</sup>	6.70±0.01 <sup>bc</sup>	6.72±0.00°	6.78±0.02 <sup>d</sup>				
L* (Lightness)	89.42±0.23°	87.64±0.03 <sup>b</sup>	86.75±0.10 <sup>a</sup>	87.72±0.00 <sup>b</sup>	87.76±0.01 <sup>b</sup>				
a* (redness)	-1.26±0.08ª	-1.40±0.01ª	-1.31±0.01ª	-0.75±0.01 <sup>b</sup>	-0.59±0.01°				
b* (yellowness)	8.75±0.36 <sup>a</sup>	8.40±0.11 <sup>ª</sup>	8.26±0.08 <sup>a</sup>	9.84±0.03 <sup>b</sup>	10.06±0.02 <sup>b</sup>				
$\Delta E^*$ (Color difference)	-	1.82±0.05 <sup>a</sup>	2.08±0.01 <sup>b</sup>	2.21±0.02 <sup>b</sup>	2.71±0.10 <sup>c</sup>				
Hardness (N)	10.32±1.65ª	15.48±1.43ª	13.71±2.47ª	11.48±4.28ª	13.37±2.01ª				
* Values are means ± standard deviation. a-c means within the same line with different letters are significantly different at p<0.05									

The dry matter content of all ice creams was around 30% since the mix formulation was calculated to be 30% solids. Ice creams containing similar dry matter were produced in a study examining quince seeds' effect on ice cream's characteristics [35]. There was no statistical difference between the ash amount of the ice cream

samples (p>0.05). The amount of protein increased as the hemp seed milk ratio increased. This was because the protein content of hemp seed milk was higher than cow's milk. It has been reported that plant milk generally has a higher protein content than cow's milk [38]. Since the fat content in the ice cream mix was calculated as

(5)

8%, the fat percentages were around this. According to the Turkish Food Codex (Number: 2022/13) the ice creams produced were included in the fatty ice cream category. The pH value of the samples increased as the hemp seed milk ratio in the ice cream mix increased since the pH value of hemp seed milk was higher than cow milk. The L\* values of the samples with hemp milk added were statistically different from the control ice cream sample (p<0.05). The L\* value of ice creams with hemp seed milk was lower than control ice cream because of the chlorophyll naturally found in hemp seeds [39]. Some studies also indicate that the L\* value of ice cream prepared from plant milk was lower than ice cream prepared from cow's milk [12, 31, 36].

When the hardness values of the ice cream samples were examined, no statistically significant difference was found between the samples (p>0.05). The texture of ice cream varies according to ingredients and processing conditions [40]. The hardness values of the samples vary between  $10.32\pm1.65$  and  $15.48\pm1.43$ . Some studies in the literature have similar results [40, 41]. The increase in the hardness value of the ice cream containing 25% hemp seed milk compared to the control sample can be attributed to increased protein content with adding hemp seed milk. The rich protein content may have interacted with other ingredients in the ice cream mixture and contributed to increased water immobilization in the ice cream due to its water-binding

properties, resulting in the formation of harder ice cream [42].

However, a decrease was observed in the hardness value of the ice creams containing 50, 75 and 100% hemp milk compared to the ice cream containing 25% hemp milk, although it was not statistically significant. The reason for this decrease may be that as the hemp milk ratio increases, the unsaturated fat ratio increases as well as the protein content. The fat droplets emulsified by the proteins in the colloidal structure of the ice cream may have lost their stability due to the increase in the unsaturated fat ratio. The destabilized oil droplets may have also caused the viscosity to decrease as the shear rate increase [35, 43].

# **Rheological Properties of Ice Cream Mix**

Rheological properties are important in terms of sensory evaluation in ice cream consumption. Furthermore, rheological parameters are important in mechanical transportation and convey the mix through lines during the process [31]. While the viscosity at low shear stress is associated with the consistency felt in the mouth, the viscosity at high shear rate can be understood by the flow characteristics in process operations such as pumping and spraying [44]. Apparent viscosities at 50 s<sup>-1</sup> and Herschel-Bulkley model parameters are given in Table 3.

Table 3. The apparent viscosity and Herschel-Bulkley model parameters of ice creams

	Apparent viscosity at 50 at	Herschel-Bulkley Model Parameters					
Samples	Apparent viscosity at 50 s <sup>-</sup> ,	Consistency index,	Flow behavior index,	<b>D</b> 2			
	1[50 (Fa.S)	K (Pa.s <sup>n</sup> )	n	K-			
Control	0.59±0.01°	9.98±2.28°	0.31±0.05ª	0.9999			
25%	0.39±0.00 <sup>b</sup>	5.94±0.39 <sup>bc</sup>	0.34±0.01 <sup>ab</sup>	0.9998			
50%	0.29 0.02ª	1.19±0.45 <sup>a</sup>	0.65±0.07 <sup>cd</sup>	0.9999			
75%	0.27±0.01ª	1.87±0.04 <sup>ab</sup>	0.52±0.00 <sup>bc</sup>	0.9999			
100%	0.25±0.01ª	0.64±0.14 <sup>a</sup>	0.75±0.05 <sup>d</sup>	0.9998			

\*Values are means ± standard deviation. a-e means within the same column with different letters are significantly different at p<0.05

Herschel-Bulkley model was applied to the data obtained as a result of rheological analysis (R<sup>2</sup>>0.99). Similar to our study, in many types of research in the literature, the flow behavior properties of ice creams produced were found to be suitable for the Herschel-Bulkley model [35, 54]. All ice cream mixes had lower (0.31-0.75) flow behavior index values than 1 (one) and thus exhibited pseudo-plastic or shear-thinning flow, meaning that their viscosity decreased as the shear rate increased. As the hemp seed milk ratio increased, the apparent viscosity (50 s<sup>-1</sup>) and consistency index (K) decreased, while the flow behavior index (n) increased (p<0.05). High K value represents high consistency [45]. The increase in n and the decrease in K may result from the structural rupture of the protein network of the ice cream due to shearing [36]. Samples with a low flow behavior index were reported to be easier to pump and more effective in providing the desired mouthfeel and

texture [31]. At the same time, the apparent viscosity values of the ice cream samples decreased as the hemp seed milk ratio increased. The reason for this may be that the milk fat in cow's milk is of animal origin, while the fat in hemp milk is of vegetable origin. An animal-based partially coalesced fat provides firmness and structure to ice cream [46]. Although an optimum mix viscosity can not be defined, the apparent viscosity range at 50 shear ratio after aging is suggested to be 0.1-0.8 Pas. In this study, the viscosity values of all prepared ice cream mixes were within this range [31].

Dynamic rheological properties of ice cream samples were examined to determine their viscoelastic properties. The storage (G') modulus and loss (G'') modulus and fluid behavior curves of ice cream mixes are given in Figure 1.



Figure 1. (a) Apparent viscosity vs. shear rate for ice cream mixes containing 0-100% hemp seed milk; (b-c-d-e-f-) Plots of storage (*G*') and loss (*G*'') modulus as a function of frequency for ice cream mixes containing 0-0.100% hemp seed milk

The G' values of all ice cream samples were higher than the G" values. This indicates an elastic (solid-like) structure of the ice cream mix rather than a viscous (liquid-like) structure. While the storage modulus (G') is associated with solid body-like behavior and therefore the hardness of ice cream, the loss modulus (G") describes viscous behavior and fluidity and is associated with the sensation of the creaminess of ice cream [47]. As the hemp milk ratio increased, viscous and elastic behavior values became closer. This situation was supported by the fact that as you add hemp milk, the viscosity decreased, and it became more liquid. Moreover, the reason why the loss modulus value of the ice cream mix increased as the hemp milk ratio increased could be shown to be that hemp seeds contain long-chain fatty acids, which are known to be liquid [46]. Similar to this study, ice creams with storage modulus values higher than loss modulus values have been produced in the literature [48, 49]

## **Melting Properties of Ice Cream Samples**

The chemical composition and physical structure of ice cream affect its melting properties [37]. The melting rate (g/min), drip-off (%) and first dripping time (min) of ice cream samples are given in Table 4. Also, images of ice cream samples at 0, 20, 30, and 45 minutes are shown in Figure 2.

As the hemp milk ratio increased, the melting rate and dripping amount of the ice cream samples also increased. This may be because hemp seed milk's fat melted faster than cow's milk fat. Similar results were obtained in a study using plant-based coconut milk [14]. Similar to this study, in a study where walnut milk was used in ice cream making, it was determined that the energy required to melt ice cream made with walnut milk was less than the control sample [13]. Unlike this study, the study conducted by Atalar and Kurt [12] stated that hazelnut milk improved the melting properties of ice cream samples due to its high protein content, which melted later.



Figure 2. Melting behavior of ice cream containing 0-100% hemp seed milk (HM) 0 (a), 20 (b), 30 (c) and 45 (d) min later at room temperature ( $25\pm2$  °C)

Milk fat globules affect the melting properties of ice cream by surrounding air bubbles and contributing to air phase stabilization. It has been reported that the more unsaturated the vegetable oil used in the ice cream mix and the longer the fatty acid chain, the more pronounced the destabilization of the fat globules [50]. The presence of polyunsaturated fatty acids (linoleic acid, C18:2 $\omega$ 6 and  $\alpha$ -linolenic acid, C18:2 $\omega$ 3) in

hempseed milk can explain the negative effect on melting properties as the hemp milk ratio increases in the ice cream mix [51]. Similar to this study, in the study conducted by Güven and Kalender [52], it was observed that the use of vegetable oils such as hazelnut oil and olive oil negatively affected and accelerated the melting of ice cream.

Table 4.	Meltina	properties	of	ice	creams
			••••		

Sample	Melting rate (g/min)	Drip-off (%)	First dripping time (min)
Control	0.27±0.08ª	17.67±5.66 <sup>a</sup>	3.32±0.19 <sup>a</sup>
25%	0.40±0.13 <sup>ab</sup>	26.67±8.48 <sup>ab</sup>	2.85±0.46ª
50%	0.59±0.16 <sup>ab</sup>	39.58±10.48 <sup>ab</sup>	2.74±0.51ª
75%	0.73±0.03 <sup>b</sup>	48.50±2.12 <sup>b</sup>	2.57±0.01ª
100%	0.74±0.06 <sup>b</sup>	49.50±4.00 <sup>b</sup>	2.30±0.04ª
11.1.1			

\*Values are means  $\pm$  standard deviation. a-b means within the same column with different letters are significantly different at p < 0.05.

No statistical difference was observed between the ice cream samples regarding the first dripping times (p: 0.149). A high negative correlation exists between initial drip time and melting (r: -0.93) rate. Additionally, a high negative correlation was also calculated between apparent viscosity values and melting rate (r: -0.94). A high positive correlation was also found between the first drip time and viscosity (r: 0.95). These results revealed the importance of viscosity on the melting properties of ice cream. In the study conducted by [35], it was stated that viscosity is important in ice cream samples with high correlation parameter.

## **Functional Properties of Ice Cream Samples**

Among the functionalization strategies of ice cream, the presence of bioactive components is important [53]. Plant-based milk stand out with their bioactive components (for example, soy-based beverages contain isoflavones and phytosterols; almond-based beverage contains tocopherol and arabinose; oat-based beverage contains glucan; hemp seed milk contains polyunsaturated fatty acids (PUFAs) and essential fatty acids (EFAs), are therefore used to produce functional products [38, 54-56]. Bioactive compounds in ice creams were determined by total phenolic content and antioxidant activity analysis, and the results are shown in Figure 3.



Figure 3. The total phenolic contents (TPC) (a) and antioxidant activity levels of ice cream mixes containing 0-100% hemp seed milk (b)

As the ratio of hemp seed milk in the ice cream mix increased, both the total phenolic content and antioxidant activity of the samples increased. Bioactive compounds such as linoleic acid,  $\gamma$ -tocopherol, cannabidiol acid, lignan-amides, and terpenes contained in hemp seed can be shown as the reason why the phenolic and antioxidant content increased as the hemp seed milk ratio increased [25]. A study conducted by Beşir and Mortaş [9] determined that the antioxidant and phenolic component content of ayran drink with increased hemp milk content was higher than that of ayran prepared entirely from cow's milk. Similar to this

study, phenolic and antioxidant contents increased in ice creams produced from hazelnut milk in a study researched by Atalar and Kurt [12]. In another study, the effect of non-dairy ingredients (coconut milk and coconut sugar) on total phenolic content was found to be highly significant [14].

## **Sensorial Properties of Ice Cream Samples**

Sensorial parameters evaluated for ice cream samples are given in Table 5.

Table 5. Ochooly parameters of control and this added lee of came	Table 5.	Sensory	parameters	of	control	and	ΗM	added	ice	creams
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Table 5. Sensory parameters of control and this added ice creams									
Baramatara	Hemp seed milk concentration								
Parameters	Control	25%	50%	75%	100%				
Appearance	8.59±0.62ª	8.53±0.72 <sup>a</sup>	8.29±0.92ª	7.88±0.99 <sup>a</sup>	7.94±1.03ª				
Taste and aroma	8.58±0.62 <sup>b</sup>	7.88±0.93 <sup>ab</sup>	7.23±0.97 <sup>a</sup>	6.88±1.32 <sup>a</sup>	7.53±1.46 <sup>a</sup>				
Texture	8.12±0.86 <sup>b</sup>	7.59±1.12 <sup>ab</sup>	7.47±0.80 <sup>ab</sup>	7.18±0.88 <sup>ab</sup>	7.12±1.41ª				
General acceptance	8.56±0.55 <sup>b</sup>	7.91±0.94 <sup>ab</sup>	7.56±0.74 <sup>a</sup>	7.23±0.97 <sup>a</sup>	7.44±1.25 <sup>a</sup>				

\* Values are means  $\pm$  standard deviation. (n = 25 panelists for each ice cream type). a-b means within the same line with different letters are significantly different at *p*<0.05. Sensory attributes and their scales: 1 = undesired, 9 = desired. Values are means  $\pm$  standard deviation. a-b means within the same line with different letters are significantly different at p<0.05

There was no statistical difference between the ice cream samples regarding appearance (p: 0.058). For the taste-aroma parameter, ice cream containing 25% hemp milk was scored similarly to control ice cream. Taste and aroma differed statistically from control ice cream, starting from a 50% hemp seed milk ratio. Regarding texture, except 100% hemp seed milk all ratios were evaluated as close to control ice cream (p>0.05). When general acceptability was assessed, the most liked ice cream with hemp seed milk added by the panelists was the ice cream containing 25% hemp seed milk. While the addition of plant-based milk increased the acceptability compared to the control sample in some studies [12, 13], it decreased the acceptability in some studies [56-58]. In a study where ice cream was made using soy and sweet bean milk, as the proportion of plant milk increased, the panelists' approval score decreased, and they determined that the best concentration of plant-based milk was 25% [57]. In the sensory analysis performed on ice creams produced using hemp seed milk and almond milk, ice cream

produced from hemp seed milk was less liked due to the high density and slightly unpleasant aroma specific to hemp seed milk [56]. Like hemp seed milk, ice cream made from soy milk was also disliked due to its intense flavor [58].

### CONCLUSION

This study investigated the usability of hemp milk in ice cream production. Thanks to the high protein content of hemp seed and its rich phenolic-antioxidant content, hemp seed milk-based ice creams with higher protein content and improved functional properties were obtained compared to the control ice cream sample made from cow's milk. Using hemp milk instead of cow's milk did not positively affect viscosity and melting properties. When the hardness value was examined, ice creams similar to the control ice cream were obtained. As the hemp milk ratio increased, the apparent viscosity of the ice cream decreased due to the hemp seeds containing long-chain fatty acids, which are known to be liquid. In addition, the loss modulus increased, and the viscous and elastic behavior values became closer to each other. From a sensory perspective, considering the general acceptability parameter, the acceptable hemp milk rate was determined to be 25%. If the results of this study are supported by future studies examining its effects on health, hemp seed milk can be recommended as an alternative plant-based milk that can be used in the production of functional ice cream, as it increases the total phenolic and antioxidant content of ice cream.

# **CONFLICT of INTEREST**

The authors declare no conflict of interest.

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