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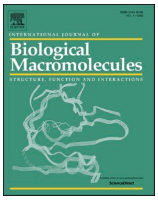

# Bacterial cellulose infusion: A comprehensive investigation into textural, tribological and temporal sensory evaluation of ice creams

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ABSTRACT

The study examines how adding bacterial cellulose also referred to as Symbiotic Culture of Bacteria and Yeast (SCOBY) to ice cream affects the textural, tribological, and sensory attributes, particularly texture and mouthfeel perception. Analytical assessments were performed on three types: SCOBY-added ice cream and two reference samples (control and guar gum-added ice creams). Evaluations included physicochemical properties, textural and tribological characteristics, and dynamic sensory mouthfeel using the temporal dominance of sensation (TDS) methodology. SCOBY ice cream showed higher probiotics content, lower pH, and higher acidity than reference samples. The addition of SCOBY increased hardness and altered the textural properties. TDS analysis highlighted distinct temporal dominance patterns, with guar gum ice cream presenting a pronounced mouth/residual coating pre-swallowing, while SCOBY and control ice cream exhibited a thin/fluid perception. The frictional factor at 37 °C was positively correlated with the melting rate, graininess, and thin/fluid perception while negatively correlated with firmness, smoothness and mouthfeel liking. Additionally, the mouthfeel liking was higher with firm, smooth and mouth/residual coating sensations and lower with grainy and thin/fluid perception. In summary, incorporating SCOBY in ice cream formulations can provide health benefits and meet consumer preferences for natural ingredients, while ensuring careful optimization of mouthfeel.

## 1. Introduction

Ice cream, being a complex colloidal system, comprises partially coalesced fat globules, ice crystals, air bubbles, sugar, proteins, and salts dispersed within a frozen aqueous matrix [1]. It also includes fat and milk solids, which serve as supportive elements for probiotic bacteria. Unlike fermented dairy products like yoghurts, ice cream provides a more conducive environment for probiotic viability throughout the manufacturing and storage conditions [2], making it an ideal medium for incorporating beneficial bacteria and delivering functional healthy food. Additionally, hydrocolloids and dietary fibres used in ice cream formulations act as stabilizers by preventing ice crystal formation and extending the shelf life.

The Symbiotic Culture of Bacteria and Yeast (SCOBY) is a bacterial cellulose produced during the fermentation of Kombucha tea and is a rich source of probiotics. During the fermentation process, a significant amount of SCOBY waste is generated. Upcycling this discarded SCOBY waste into value-added foods such as ice cream presents an opportunity to foster sustainable practices in food production [3,4]. The formation of bacterial cellulose (BC) mat occurs through a symbiotic relationship between acetic acid bacteria and osmophilic yeast species [5]. Bacterial cellulose (BC) is classified as insoluble dietary fiber, is indigestible in the human gut and is considered a “generally recognised safe” food by the U. S. Food and Drug Administration (FDA) [6]. BC is extensively used in the food industry to produce low-fat, low-calorie, and low-cholesterol foods [7–9]. BC is unaffected by pH, temperature and ionic strength changes and is compatible with protein matrices [10]. Additionally, BC functions as a fat droplets stabilizer in oil-in-water emulsions around the air cells, and is a better emulsifier than carboxymethyl cellulose (CMC) due to its compatibility with whey proteins [11], resulting in a strong gel-like structure. The intermolecular interactions between cellulose and protein chains contribute to the textural, rheological and emulsifying properties of BC-enhanced milk products [8,12]. Melting rate is a crucial parameter influencing the stability of ice cream, and addition of

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bacterial cellulose has notably reduced the melting rate [8]. This is primarily due to the enhanced water-holding capability and structural stabilization provided by BC [8,12].

Bacterial cellulose derived from SCOBY exhibits distinct characteristics, such as finer and unbranched structures of pure cellulose devoid of lignin and hemicelluloses, distinguishing it from plant cellulose [13,14]. These properties contribute to its superior water absorption capacity, porosity, biocompatibility, surface area, and mechanical strength [15,16], making it an increasingly sought-after material for various technological applications, including bioplastics, food fortification, and food packaging. Additionally, SCOBY contains probiotic bacteria [17], rendering it a healthy option for incorporating into various food products such as ice cream, yoghurts and desserts [15]. As well as its nutritional benefits, ice cream is cherished for its appealing sensory properties such as creamy texture and sweet taste, making it popular among consumers of all age groups. Oral lubrication is critical in shaping the perceived texture and consumer liking of food products, suggesting its potential application to improve mouthfeel [18]. Limited studies have investigated the effect of BC on the rheological, textural, and sensory properties of frozen products. However, no research has been conducted specifically focusing on the tribological properties of ice cream containing bacterial cellulose.

The present study examines how adding SCOBY to ice cream affects the physicochemical properties (including microbial count, colour, melting rate, pH and acidity), particle size, textural properties and sensory attributes compared to the reference samples. The study investigated the correlation between the dynamics of sensory perception and instrumental measures (tribology) of ice cream formulations to better understand parameters related to perceived textural properties. The insights gained regarding the relationship between sensory perception and instrumental measures could be utilized to optimize quality control processes in ice cream production and potentially lead to the development of novel, value-added products utilizing SCOBY.

## 2. Materials and methods

### 2.1. Sample preparation

The three ice cream formulations, SCOBY-added ice cream and two references (control and guar gum-added ice creams) were prepared for the study (Table 1A). The ingredients were bought from a local Lincoln, New Zealand supermarket. The ingredients were: 25 % (*w*/w) milk (fat 3.3 g/100 ml), 50 % (w/w) cream (fat 36 g/100 ml) and 15 % (w/w) sugar. Additionally, either 10 % (w/w) guar gum or 10 % (w/w) SCOBY and kombucha combined (procured from Get Cultured, Tauranga, New Zealand) were included in the reference samples formulation. In the 10 % SCOBY ice cream sample, the SCOBY mixture made from a semi-solid mass of SCOBY (8 %) and kombucha (2 %) was added. The dry matter content of SCOBY was 2.8 % on a wet basis. The reference sample recipe of guar gum and the control samples were adjusted based on the dry matter content of SCOBY. The experimental scheme is shown in Fig. 1.

The ingredients were weighed and mixed with a hand mixer at room temperature for 15 min. The mixture is then stored at 4 °C for 2 h. Next, the ice cream mixes were frozen in 1000 g batches in a home-style ice cream maker (Cuisinart, Stamford, CT) for 55 min at the temperature of −27 °C [19]. Finally, the ice cream samples were stored at −10 °C for 24 h before sensory and physiochemical analyses.

### 2.2. Microbial count analysis

The microbial cell count of the SCOBY and ice cream samples was done on day 1 of frozen storage. About 5 g of SCOBY and 1 g of ice cream samples were diluted in 45 ml and 9 ml of a Phosphate-buffered solution, respectively. The subsequent serial dilutions were made after homogenizing the dilution in a vortex (Monataro, Japan). Each dilution was plated in De Man, Rogosa and Sharpe (MRS) Agar plate and incubated at 37 °C for 48 h under anaerobic conditions with Thermo Scientific™ AnaeroGen 2.5 L sachets (Auckland, New Zealand). The result was expressed in colony-forming units (log CFU/ml).

**Table 1**
(A) Formulation of the mixes used for ice cream making (g/100 g). (B) Definition of attributes selected for TDS evaluation of ice creams.

| (A) Ingredients | Control | SCOBY | Guar gum |
|---|---|---|---|
| Milk | 27 | 25 | 25 |
| Cream | 50 | 50 | 50 |
| Sugar | 15 | 15 | 15 |
| SCOBY | – | 8 | – |
| Kombucha | – | 2 | – |
| Guar gum | – | – | 0.2 |
| Water | 7.8 | – | 9.8 |

*The ice cream formulations were standardized based on the dry matter content of the SCOBY mass. The dry matter content of SCOBY was 2.8 % on a wet basis.*

| (B) Attributes | Definition |
|---|---|
| Texture | |
| Creamy | Combined perception of smoothness, fat and viscosity |
| Smooth | Evaluate the sample by placing a spoonful on the tongue, sliding it against the palate, and checking for uniform texture without any coarse or rough surfaces |
| Firm | The force required by the tongue to flatten the ice cream against the palate |
| Grainy | Course sensation after detecting ice crystals in the mouth |
| Thin/Fluid | The rate at which the sample melts in the mouth |
| Sticky | The sensation when the product sticks to the palate |
| Thick/Dense | The contrary of fluid or resistance to flow |
| Mouthfeel | |
| Mouth/Residual coating | The sensation caused by the film remaining in the mouth after swallowing the sample. |

### 2.3. Titratable acidity and pH

The titratable acidity of the ice cream samples was measured on day 1 in triplicates. 20 g of the sample was mixed with 20 ml of boiled distilled water for titration. The percentage of titratable acidity was measured by titrating the prepared sample with 0.1 N NaOH, and using phenolphthalein as an indicator [20]. The pH of the ice cream samples was taken with a digital pH meter (Mettler Toledo, Ohio, United States), after calibration with standard pH buffer solutions (pH 7 and pH 4).

### 2.4. Melting rate

The melting rate of the ice cream was measured based on the previously described methodology [21]. Twenty-five grams of ice cream (stored at −10 °C) were left on the wire mesh at 25 ± 1 °C for 80 min in an incubator. The melting rate was measured using the following equation.

$$\text{Melting rate }(\%) = \frac{\text{Weight of ice cream melt}}{\text{Initial Weight}} \times 100$$

### 2.5. Colour

The colour values of ice cream products stored at −10 °C were measured with a CR-400 Chroma Meter (Minolta Camera Co. Ltd., Osaka, Japan) based on the method described. The result was expressed in terms of L*, a* and b* values, which exhibit the light intensity (0−100), red (+) to green (−) and yellow (+) to blue (−) colour characteristics [22]. Delta E (Δe) was calculated using the equation below to measure the change in visual perception of the colours between samples.

$$\Delta e = \sqrt{(\mathrm{L2} - \mathrm{L1})2 + (\mathrm{a2} - \mathrm{a1})2 + (\mathrm{b2} - \mathrm{b1})2}$$

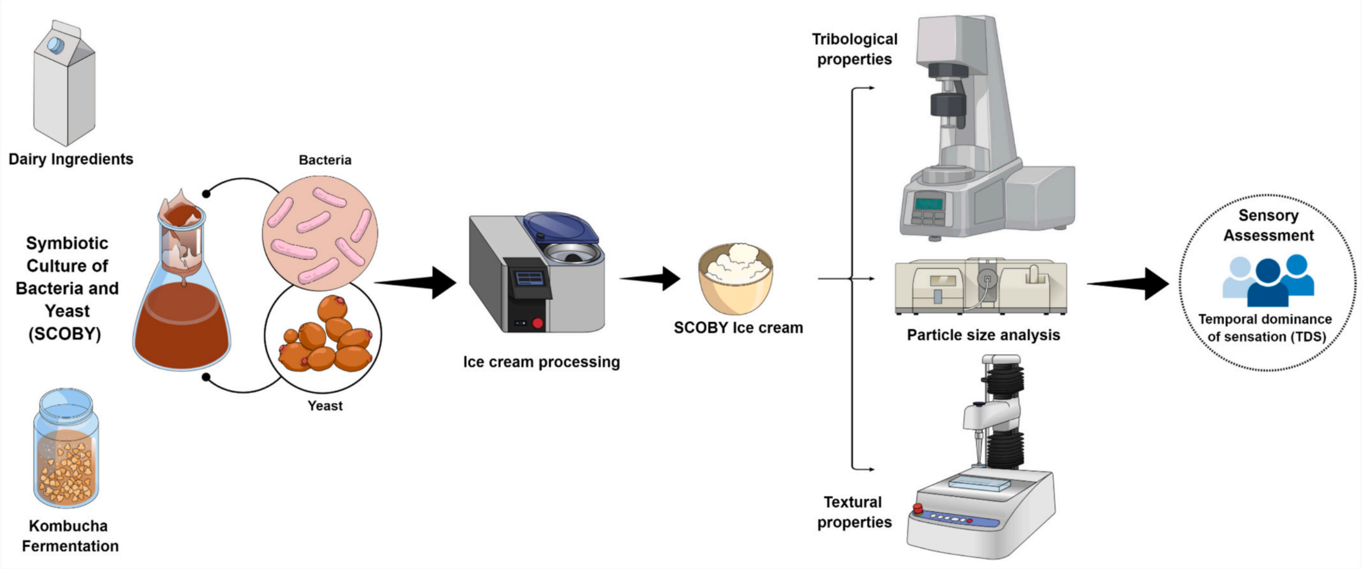

**Fig. 1.** Experimental scheme of the study.

### 2.6. *Particle size*

The particle size of ice cream products was measured in a Mastersizer 3000e Analyzer (Malvern Instruments, Worcestershire, UK) at the refractive index of 1.46 and 1.33 for milk and water. The ice cream mix was directly added to the deionized water until 15 ± 1 % obstruction level [23]. The volume-weighted or De Brouckere mean diameter ($d_{4,3}$) and volume distribution of particles ($d_{10}$, $d_{50}$, and $d_{90}$) were recorded.

### 2.7. *Texture analysis*

The penetration test with TA. XT2i Texture Analyzer (Stable Micro Systems Ltd., Godalming, United Kingdom) with 500 N force measurement cell determined the ice cream hardness. The ice cream samples were stored in a plastic container (30 mm length and 65 mm width) at $-20 \pm 1$ °C before the analysis. The samples were penetrated to a depth of 15 mm at a test speed of 2 mm/s. A cylindrical probe (5 mm diameter) was used to measure the maximum force (N) applied to measure the hardness [24].

### 2.8. *Tribological properties*

The tribological properties of ice cream samples with velocity and temperature sweep were done using an Anton Paar rheometer (MCR 302, Anton Paar, Austria) equipped with a Peltier hood (H-TPD200, measuring shaft (BC12.7), a sample holder (SH-BC6), and a Peltier temperature control hood (T-PTD200). A ball-on-three-pins set-up was used with a glass ball (soda lime glass, 12.7 mm diameter) on PDMS (poly-dimethyl siloxane) pins (Cylinder, 6 mm in height and diameter, 45° deflection angle) to simulate the contact surface configuration. PDMS and glass balls were used because they closely emulate the soft contact surface characteristics of the tongue and palate [25,26]. The tribological behavior of the ice cream samples was measured at 37 °C (average human mouth temperature) at the sliding velocity of $10^{-9}$–0.1 m/s (88 measuring points, 1 s per measuring point). Based on the physiological forces between the palate and the tongue [25,27,28], the normal force of 1 N was used in the experiment. The ice cream samples used for measurement were 1 g, and the sample holder was cleaned regularly with deionized water and dried with compressed air before using a new sample.

Temperature-dependent tribological properties of the ice cream samples were recorded by the temperature sweep from $-5$ °C to 37 °C at a heating rate of 5 °C per minute with the above settings. The relative speed of the tongue to the palate during food intake ranges from 10 to 60 mm/s [26]. After conducting trials in this range, the sliding velocity of 25 mm/s was used for all the measurements.

### 2.9. *Temporal dominance of sensation (TDS)*

#### 2.9.1. *Selection of TDS attributes*

The focus group study was conducted with an expert panel ($n = 5$) to develop TDS attribute vocabulary. Ten attributes describing texture and mouthfeel were chosen for discussion based on the previous studies of ice cream [19,29–31]. Based on the discussion, eight attributes were selected for the final evaluation session (Table 1B).

#### 2.9.2. *Selection and training of participants*

Thirty-six participants (21 females and 15 males) aged 25–30 years were invited to participate in the TDS experiment. The participants were students and faculty at Lincoln University who liked and consumed ice cream at least twice a month. Most participants were students, as youth demographics influence trendsetting and market preferences [32], making them key contributors to sensory studies for new and innovative products. The study was approved by the Human Ethical Committee (HEC2021–08) of Lincoln University, New Zealand. All the participants provided written consent before the experiment.

#### 2.9.3. *Evaluation session*

The participants were briefed about the experiment. The training session was conducted before the experiment to familiarize participants with the procedure and the software. Each participant received a printed copy of the definition of the attribute. The participants were instructed to clean their palate with crackers and water between the samples. The attributes were randomized across the panelists but remained in the same order for each participant across all samples. The participants were instructed to put a sample in their mouths and click the start button to begin the experiment. Participants were asked to select the dominant attribute they perceived at any moment during the tasting. Participants were informed they could choose one attribute multiple times or never select one. Participants were free to choose any attribute and change their selection if the perceived dominant sensation changed. Finally, the stop button was pressed at the swallowing time. The samples' overall and mouthfeel liking were measured on a 9-point hedonic scale.

### 2.10. Data and statistical analysis

The triplicate results from instrumental measurements were analysed using the software Minitab (ver. 19.2020.2) and expressed in mean ± SD values. One-way analysis of variance (ANOVA) was used to determine significant differences between the samples at a 5 % significance level. The Pearson correlation coefficient (*r*) was used to find the relationship between instrumental measurements and sensory attributes. The figure depicting the experimental scheme of the study was created using BioRender.

The TDS data was collected using the Redjade® software (Redjade Sensory Solutions LLC, California Corporation, USA). The attributes chosen as dominant and the time when the dominance started and stopped were collected for each participant. The times for all participants were standardized using XLSTAT® statistical analysis software version 2021.3.1 (Addinsoft, New York, NY, USA, 2021). The chance level ($P_o$) and significance level ($P_s$) were drawn to get additional information about the TDS curve [33]. The chance level ($P_o$) is the dominance rate at which an attribute was obtained by chance and is inversely proportional to the number of attributes ($P_o = 1/\text{p}$, where p is the number of attributes). The significant level is the minimum dominance rate, which is significantly higher than the chance value calculated with the equation below.

$$Ps = Po + 1.645\sqrt{Po\frac{1 - Po}{n}}$$

$Ps$ is the lowest significance ($p < 0.05$) in TDS, and n is the number of panelists multiplied by the number of replications.

## 3. Results and discussion

### 3.1. Microbial count, pH, titratable acidity and melting point

The microbial count, pH, titratable acidity and melting point measurements for the control, SCOBY and guar gum ice creams are shown in Table 2. The probiotic bacteria counts in the SCOBY ice cream was 5.48 $\log_{10}$ CFU/ml, while no bacterial count was found in the control and guar gum ice creams. A significant variation ($p < 0.05$) was observed in pH, titratable acidity and melting point among the samples. SCOBY ice cream had a significantly lower pH (5.50) and higher acidity (3.11 %) compared to the control (pH: 6.76, acidity: 0.99 %) and guar gum ice creams (pH: 7.07, acidity: 0.99 %). Guar gum ice cream had the lowest melting rate (27.07 %) compared to SCOBY (48.44 %) and control (58.0 %) ice creams.

Previous studies [5,34] found that dried bacterial cellulose (SCOBY) has a high amount of crude protein, fiber, lipids and essential amino acids such as lysine, isoleucine and leucine. The high protein content in the SCOBY acts as a nitrogen source for probiotic growth. In addition, the dietary fiber acts as a growth substrate and provides an encapsulation membrane against mechanical stress and freezing injury during the mixing and freezing stage due to air cell bubble incorporation. The pH level also plays a significant role in bacterial growth. Kombucha is a fermented drink prepared from black tea with the help of a symbiotic culture of bacteria and yeast (SCOBY). It has a fruity sour taste due to different organic acids such as acetic, gluconic and glucuronic acid [5]. The probiotic growth is also high in low pHs [21], as seen in SCOBY ice cream compared to control and guar gum ice creams. The low pH in SCOBY ice cream indicated increased lactic acid production, hence a high bacterial count. No such effect was observed in the case of control and guar gum ice cream. The findings align with the previous study of yoghurt made with guar gum [35].

**Table 2**
Microbial count, pH, titratable acidity and melting rate of control, SCOBY, and guar gum ice cream.

| Samples | Control | SCOBY | Guar gum |
|---|---|---|---|
| Microbial count (log CFU/ml) | $0^{a}$ | $5.48 \pm 0.2^{b}$ | $0^{a}$ |
| pH | $6.76 \pm 0.0^{b}$ | $5.50 \pm 0.0^{c}$ | $7.07 \pm 0.0^{a}$ |
| Titratable acidity (%) | $0.99 \pm 0.0^{b}$ | $3.11 \pm 0.0^{a}$ | $0.99 \pm 0.0^{b}$ |
| Melting rate % | $58.0 \pm 0.5^{a}$ | $48.4 \pm 1.2^{b}$ | $27.1 \pm 4.7^{c}$ |

Mean and standard deviation values followed by different lowercase superscripts indicate significant differences among the ice cream samples ($p < 0.05$).

The melting of ice cream is a complex process involving heat and mass transfer. The heat penetrates from exterior to interior, resulting in the melting of the ice cream. The meltdown of the ice cream depends on various parameters such as ingredients, incorporation of air bubbles, size and amount of ice crystals and the fat globule network [36]. Guar gum, commonly used as a stabilizer in ice cream, rapidly hydrates in water, forming a thick solution that increases viscosity and ensures a smooth and creamy texture [37,38]. This enhanced microviscosity allows more time for water to diffuse within the concentrated serum before flowing from the interior to the exterior of the ice cream, consequently reducing the melting rate.

The addition of bacterial cellulose (BC) reduces ice cream's melting rate while improving its viscosity, texture and emulsification properties [6,8]. This effect could be attributed to the high water-holding capacity and large surface area of BC microfibrils, which tightly bind water molecules within the hydroxyl groups of the cellulose polymer [39]. SCOBY has bacterial cellulose made from intracellular 1,4-β-glucan chains by crystallization of cellulose chains [40], which have high porosity, water-holding capacity (up to 100 times its weight) and abundant surface hydroxyl (-OH) groups [41]. The amphiphilic nature of the hydroxyl and amino group makes it a good emulsion stabilizer by retaining water and fat, thereby influencing the melting point of the ice cream [42].

### 3.2. Colour

The appearance of the food is an important sensory property that affects consumer perception. The most commonly used method to quantify food colour is the CIELAB (CIE Lab*) colour space. Which is a three-dimensional model, where L* represents the lightness ranging from 0 (black) to 100 (white), *a** represents the red-green axis (+a* for red and -a* for green) and b* represents the yellow-blue axis (+b* for yellow and -b* for blue) [43,44]. The values of *L**, *a**, *b** and ΔE of control, SCOBY and guar gum ice creams are shown in Table 3.

The addition of SCOBY and guar gum significantly affected the L*, a* and b* values of ice cream compared to control ice cream. The results show that the control ice cream (85.2) was significantly lighter than SCOBY (82.6) and guar gum (81.6) ice creams, while green and yellow colour increased in ice cream with the addition of SCOBY and guar gum. SCOBY ice cream had a* and b* values of −1.40 and 15.4, and guar gum ice cream had −2.64 and 17.8. The reduction in L* of SCOBY and guar gum ice cream samples was related to the opaque gel structure formed by the aggregation of polysaccharides and milk protein caseins [45]. At

**Table 3**
Colour, firmness and particle size values of control, SCOBY, and guar gum ice cream.

| Sample | Control | SCOBY | Guar gum |
|---|---|---|---|
| L* | $85.2 \pm 0.2^{a}$ | $82.6 \pm 0.3^{b}$ | $81.6 \pm 0.2^{c}$ |
| a* | $-2.30 \pm 0.1^{b}$ | $-1.40 \pm 0.1^{a}$ | $-2.64 \pm 0.1^{c}$ |
| b* | $14.8 \pm 0.4^{c}$ | $15.4 \pm 0.2^{b}$ | $17.8 \pm 0.1^{a}$ |
| ΔE | 0 | 2.76 (*recognizable*) | 4.67 (*well visible*) |
| Firmness (N) | $6.3 \pm 0.9^{c}$ | $22.1 \pm 0.9^{a}$ | $16.0 \pm 1.8^{b}$ |
| D (4,3) (Mm) | $112.3 \pm 10.4^{b}$ | $94.3 \pm 15.0^{b}$ | $161.5 \pm 7.8^{a}$ |
| D (10) (Mm) | $1.3 \pm 0.4^{b}$ | $5.7 \pm 1.1^{b}$ | $27.9 \pm 4.8^{a}$ |
| D (50) (Mm) | $73.6 \pm 1.5^{b}$ | $65.2 \pm 11.9^{b}$ | $129.0 \pm 2.8^{a}$ |
| D (90) (Mm) | $263.0 \pm 39.1^{ab}$ | $211.7 \pm 36.9^{b}$ | $320.5 \pm 27.6^{a}$ |

Mean and standard deviation values followed by different lowercase superscripts indicate significant differences among the ice cream samples ($p < 0.05$).

low pH, the interaction between anionic polysaccharide and positively charged milk protein increases significantly, impacting ice cream colour [46], which can be seen in the case of SCOBY ice cream.

The degree of colour change is expressed as ΔE; a value of >3 is considered perceptible by the human eye [47]. A significant colour difference was observed between the control and guar gum ice creams (4.67) compared to the control and SCOBY ice creams (2.76). Guar gum, a light-yellow galactomannan, contains a carotenoid pigment that forms a stable complex between polysaccharide and ice cream emulsion, which reduces the L* value and increases the b* value [35,45]. Additionally, guar gum forms uniform ice crystals, resulting in increased lightness [48]. The influence of guar gum on the colour of products such as yoghurt [35], bread [49], mayonnaise [50] and ice cream [51] have been reported earlier.

### 3.3. *Particle size*

The particle size distribution values of control, SCOBY and guar gum ice creams are presented in Table 3. Particle size and size distribution are the critical factors that impact the textural properties of food products [18]. The $d_{10}$ and $d_{50}$ values of guar gum ice cream (27.9 μm and 129 μm) were significantly higher than those of SCOBY (5.7 μm and 65.2 μm) and control (1.3 μm and 73.6 μm) ice creams.

The addition of SCOBY to ice cream increased its titratable acidity, affecting the particle size of milk fat globules (ranging in size from 0.1 to 15 μm). In this food matrix, colloidal and spherical casein micelles ranged in size from 50 to 600 nm in diameter [52]. The interaction of fat globules and casein micelles due to acidification significantly impacts particle size. Milk fat globules are surrounded by a stabilizing membrane called milk fat globule membrane (MFGM). During acidification, the proteins in the casein micelle dissociate and reassociate and attach with the fat globule through the MFGM, thus increasing the particle size [53].

On the other hand, guar gum consists of galactomannan polysaccharides, which are composed of linear chains of (14)-linked D-mannipyranosyl units with (16)-linked D-galactopyranosyl residues as side chains. These high molecular weight molecules contain hydroxyl groups, which form hydrogen bonds with different food products, impacting viscosity and particle size. The interaction between milk components and galactomannans increases the aggregation rate, viscosity, and gel strength. Milk proteins such as casein and whey interact with galactomannans through electrostatic interactions and hydrogen bonding, facilitating the aggregation of proteins and galactomannans and increasing particle size [54].

The $d_{4,3}$ values of guar gum ice cream (161.5 Mm) were significantly higher than the control (112.3 Mm) and SCOBY ice cream (94.3 Mm). The $d_{4,3}$ values are related to changes in particle size involving destabilization processes and, therefore, more sensitive to large droplets and aggregate phenomena, making them a valuable indicator of polydispersity of samples [55]. The control sample showed a bimodal distribution, while SCOBY and guar gum showed a multimodal distribution (Fig. 2). In overall particle distribution, all the samples have a broad first peak centred around 0.5 to 2 μm. The particle size distribution peak of guar gum exhibits a right-skewed distribution with the largest particle size observed. However, both the control and SCOBY ice cream samples display similar particle size distribution peaks, but the SCOBY ice cream sample presents a trimodal distribution and the control sample demonstrates a bimodal distribution.

### 3.4. *Textural properties*

The hardness of the ice cream samples is shown in Table 3. Hardness was reported as the force required to penetrate the probe into the sample. A significant difference ($p < 0.05$) in hardness was observed among the ice cream samples. SCOBY ice cream (22.1 N) was significantly harder than guar gum (16 N) and control ice creams (6.3 N).

The hardness of ice cream has been affected by various factors such as ice crystals (size, concentration and growth), ingredients used (proteins, hydrocolloids, phospholipids and sugar) and processing conditions (freezing, aeration and homogenization) [56]. The addition of hydrocolloids in the control ice cream augmented the hardness through cryogelation, where hydrocolloids form a gel-like structure contributing to the ice cream's overall firmness. Moreover, hydrocolloids act as cryoprotectants by controlling the water diffusion between the ice crystals through steric hindrance and water holding capacity [57], which helps maintain desirable texture.

The addition of bacterial cellulose in the ice cream further increased the hardness compared to the guar gum. Bacterial cellulose is a high molecular weight linear polymer of ß(1–4)-D-glucose residues linked with glycosidic bonds, which interacts with proteins by hydrogen bonds and van der Waals forces, resulting in a compact 3D network structure. Similar findings were also reported by Guo et al. (2018) [8], where adding bacterial cellulose in the soya protein isolate increased the ice

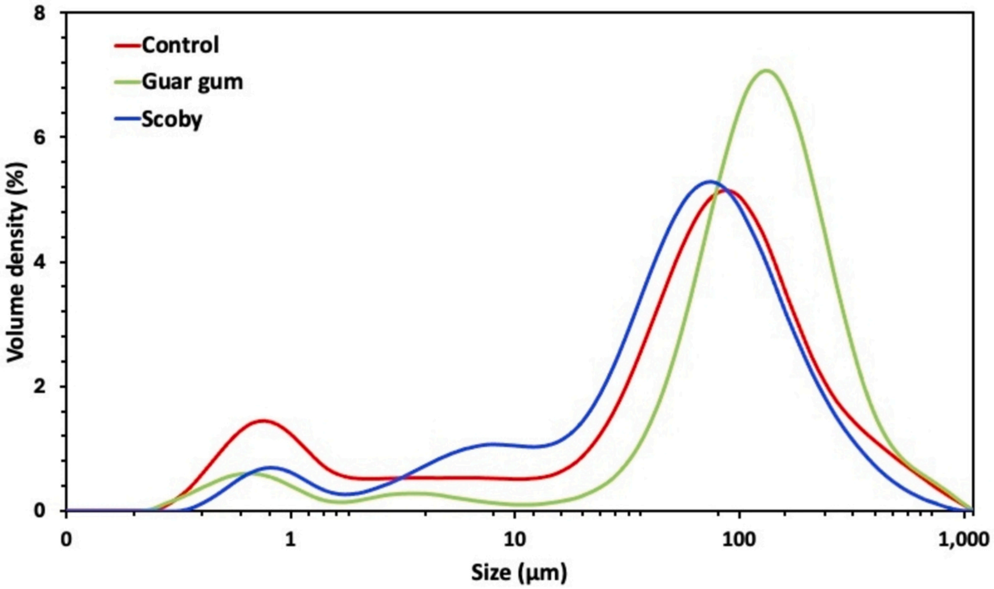

**Fig. 2.** Particle size distribution of control, SCOBY, and guar gum ice cream.

cream's hardness. Panagopoulou et al. (2015) also found that adding bacterial cellulose significantly improved the textural properties and suspension stability of BC/WPI mixtures [11]. An alternative explanation pertains to the water-holding capacity, which varies with pH levels. The decrease in pH leads to protein denaturation, facilitating the binding between whey-whey proteins or whey-casein proteins. This process increases the exposure of hydrophilic groups in an aqueous medium, thereby enhancing the water-holding capacity. An earlier study also noted comparable results [58], showing that substituting milk with yoghurt of lower acidity led to decreased viscosity.

### 3.5. *Tribological properties*

The STRIBECK curve depicts the frictional factor experienced by the ice cream samples as a function of sliding velocity (Fig. 3A). In the extended STRIBECK curve, the static friction regime is included alongside the boundary, mixed and hydrodynamic regimes. The static regime represents the frictional factor resulting from the contact between the surfaces before the onset of macroscopic motion. In this regime, ice cream samples experience elastic and plastic deformation at the contact point.

The frictional factors of guar gum (0.13 and 0.22) and control (0.10 and 0.22) were significantly higher at sliding velocities of 0.001 m/s and 0.01 m/s than SCOBY ice cream (0.07 and 0.15). However, in the hydrodynamic regime of sliding velocity of 0.1 m/s, the frictional factor of SCOBY ice cream increased significantly ($p < 0.05$) compared to guar gum and control ice creams (Table 4A). The extended STRIBECK curve comparison between control, SCOBY and guar gum ice cream reveals interesting differences in the frictional factor across the different regimes. In a static regime, the curves of control, SCOBY and guar gum ice cream exhibited a similar trend, thus showing a comparable frictional resistance at the breakaway point. However, in the boundary regime, the frictional factor differed between the control ice cream on one side and the SCOBY and guar gum ice creams on the other. Control ice cream showed a continuous increase in the frictional factor in the boundary and mixed regime, then decreased in the hydrodynamic regimes. On the other hand, SCOBY and guar gum ice cream exhibited a slight drop in the frictional factor in the boundary regime but increased at the sliding

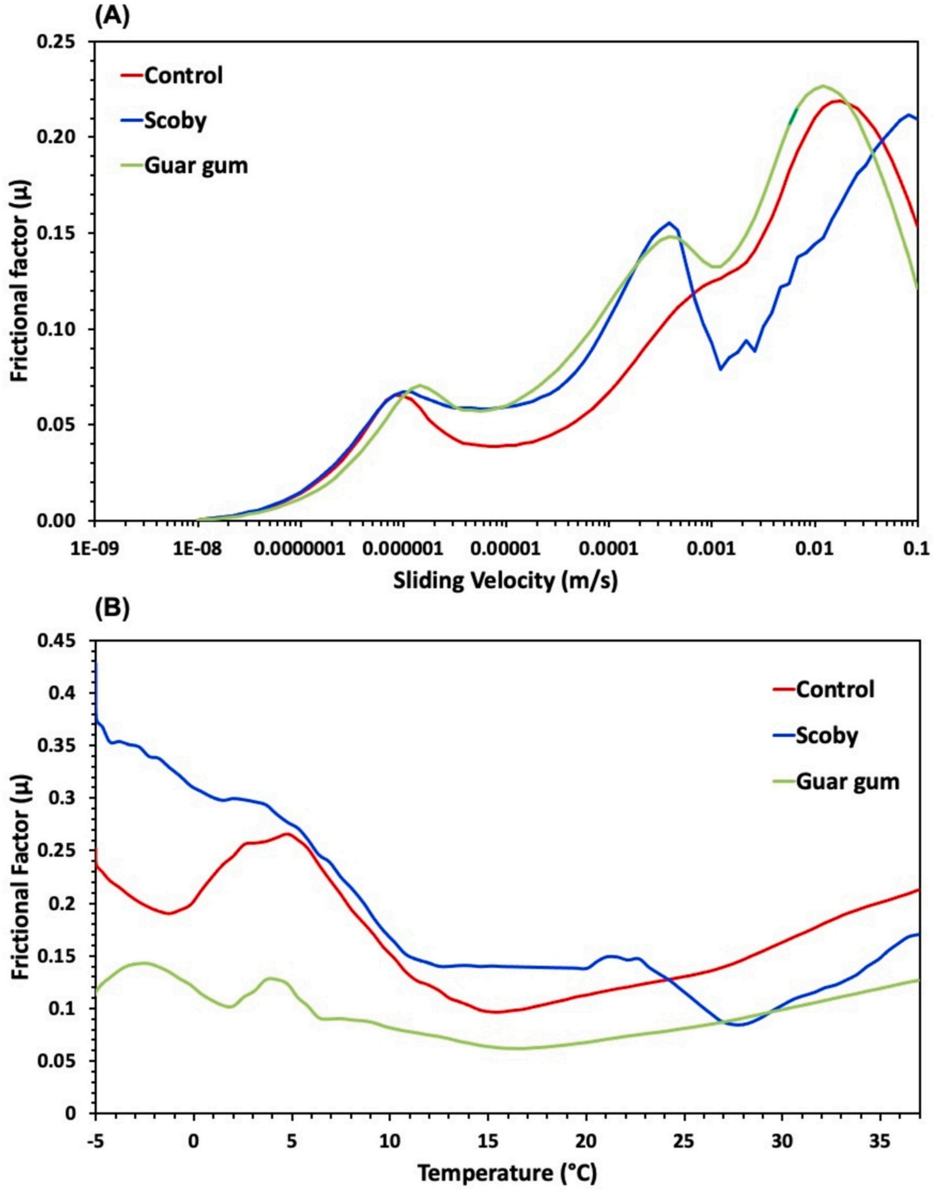

**Fig. 3.** (A) Extended STRIBECK curves of ice cream samples at 37 °C, (B) Frictional factor (μ) analysis of ice cream samples at a constant 25 mm/s sliding velocity with temperature sweep.

**Table 4**

(A) Average frictional factor (μ) of ice cream samples at 37 °C at different sliding velocities. (B) Average frictional factor (μ) of ice cream samples at 25 mm/s sliding velocity with temperature sweep.

| Samples | Control | SCOBY | Guar gum |
|---|---|---|---|
| (A) | | | |
| $\mu_{0.001}$ (m/s) | $0.10 \pm 00^{a}$ | $0.07 \pm 0.01^{b}$ | $0.13 \pm 0.00^{a}$ |
| $\mu_{0.01}$ (m/s) | $0.22 \pm 0.02^{a}$ | $0.15 \pm 0.02^{b}$ | $0.22 \pm 0.01^{a}$ |
| $\mu_{0.025}$ (m/s) | $0.20 \pm 0.01^{a}$ | $0.19 \pm 0.00^{a}$ | $0.21 \pm 0.01^{a}$ |
| $\mu_{0.1}$ (m/s) | $0.13 \pm 0.01^{b}$ | $0.21 \pm 0.00^{a}$ | $0.12 \pm 0.00^{b}$ |
| (B) | | | |
| $\mu_{-5\ ^\circ\mathrm{C}}$ | $0.26 \pm 0.01^{b}$ | $0.42 \pm 0.01^{a}$ | $0.11 \pm 0.01^{c}$ |
| $\mu_{5\ ^\circ\mathrm{C}}$ | $0.26 \pm 0.01^{a}$ | $0.27 \pm 0.00^{a}$ | $0.11 \pm 0.01^{b}$ |
| $\mu_{37\ ^\circ\mathrm{C}}$ | $0.22 \pm 0.01^{a}$ | $0.17 \pm 0.00^{b}$ | $0.13 \pm 0.01^{c}$ |

Mean and standard deviation values followed by different lowercase superscripts indicate significant differences among the ice cream samples ($p < 0.05$).

velocity of 0.001 m/s. In the hydrodynamic regime, a significant drop in the frictional factor of guar gum was observed compared to the SCOBY ice cream.

In a static regime, the frictional factor increased with the increasing sliding velocity due to the elastic deformation of the PDMS pins and ice creams. The increase in the frictional factor resulted from the resistance encountered as the surfaces deform under the applied force. At the breakaway point, the sudden drop in friction is attributed to the displacement of the samples with the measuring shaft [26]. The ice cream samples have experienced boundary (<0.001 m/s), mixed and hydrodynamic regimes (>0.001 m/s), depicting shifts in frictional factor with increasing sliding velocities. In the boundary regime, the fat content in the control ice cream tended to coalesce and form a lubricating film initially, resulting in a low frictional factor, confirming the interpretation of previous studies [18,59]. On the other hand, SCOBY and guar gum have increased the stability of the fat droplet distribution in the emulsion matrix, resulting in a less developed fat film and a high frictional factor.

In the hydrodynamic regime, the frictional factor is governed by the internal friction or viscosity of the sample, and generally, it increases linearly with the sliding velocity [60] due to the continuous film formation between the sliding surfaces. However, in the case of guar gum ice cream, the friction factor decreased in the hydrodynamic regime. The ice cream surface starts to part at high sliding velocity, and galactomannan fills the gaps as a viscous liquid and reduces the frictional factor. The reduction in friction with high-viscosity fluid can be attributed to two factors. Firstly, a polymer layer in the contact zone physically prevents the solid surfaces from coming into direct contact. Secondly, high viscosity prevents turbulent flow within the contact zone [61]. In the case of SCOBY ice cream, the frictional factor increased in the hydrodynamic regime (0.1 m/s), signifying the reduction in lubrication properties and resulting in a high frictional factor at high sliding speeds. Bacterial cellulose has relatively high porosity and water content, which results in more fluid being squeezed into the gap between the surfaces, resulting in a high frictional factor. The previous study confirmed similar findings for bacterial cellulose [62].

A significant variation in the frictional factor at −5, 5 and 37 °C was observed among the samples (Table 4B). At −5 °C, the frictional factor of SCOBY ice cream (0.42) was significantly ($p < 0.05$) higher than that of the control ice cream (0.26). However, at 37 °C, the frictional factor of SCOBY ice cream (0.17) was significantly lower than the control ice cream (0.22), exhibiting improved lubrication properties. The frictional factor of guar gum ice cream was significantly lower at all temperatures than other samples. The variation in the frictional factor of ice cream samples in response to temperature change is depicted in Fig. 3B. SCOBY ice cream displayed a notable decrease in frictional factors as the temperature ascends, while control and guar gum ice creams exhibited more complex variations in their frictional factors in response to temperature changes. Initially, SCOBY ice cream had the highest frictional factor (0.36) at −5 °C compared to control (0.25) and guar gum ice cream (0.11). However, as the temperature increased, SCOBY ice cream's frictional factor dropped significantly from 0.38 at 0 °C to 0.10 at 27 °C. On the other hand, control and guar gum ice creams showed an increase in frictional factor until 5 °C, followed by a drop until 15 °C, and another rise beyond 15 °C was observed until 37 °C. The frictional factor of SCOBY ice cream continued to increase after 27 °C. At 37 °C, guar gum ice cream recorded the lowest frictional factor compared to control and SCOBY ice cream. Both control and SCOBY ice cream exhibited a higher melting rate than guar gum ice cream (refer to Table 2), resulting in increased friction due to reduced lubrication [36].

### 3.6. *Temporal dominance of sensation (TDS)*

Fig. 4 shows the TDS graph of control, SCOBY, and guar gum ice cream. The graph shows the dominant attribute at each moment of tasting. Smooth and grainy sensations were related to the ice crystal size in the ice cream samples [63]. Regand and Goff (2006) stated that the ice crystal size directly affects the texture perception of ice cream [57]. Ice cream with tiny and well-stabilized ice crystals produces smooth and creamy sensory perception. The control was perceived to be grainy in an early stage of consumption, while no grainy perception was reported in the case of SCOBY and guar gum ice cream. The creaminess is sometimes associated with texture and at other times with flavor (vanilla, sweet or high fat content) [64]. In ice cream, creaminess is associated with the soft, melty texture when the ice cream melts swiftly in the mouth without any grainy perception. The dominance rate of creamy sensation was significantly high for all the samples. Creamy perception first appeared for guar gum ice cream (consumption time 30 %), then control (40 % consumption time) and SCOBY ice cream (60 % consumption time). The creamy perception was significantly high (dominance rate up to 40 %) in guar gum ice cream, then in control and SCOBY ice cream (dominance rate up to 30 %). Hydrocolloid affects the ice cream texture in different ways, such as the formation and size of ice crystals, water retention, and the stabilization of oil in water emulsions, which preserves the texture of ice creams by slowing the melting in the mouth, which produces an early appearance of creaminess and a lasting mouth coating sensation [19].

All the samples show some significant dominance of mouth/residue coating at the end of the consumption period, but guar gum ice cream had a high dominance rate of 45 %, and control and SCOBY ice cream had a dominance rate of 23 %. Ice creams made with stabilizers (egg, hydrocolloid and cream) have a viscous mixture, providing a mouth and residual coating sensation during and after swallowing [19]. Thin/fluid was the dominant attribute in the second half of the consumption for control and SCOBY ice creams, which can also be related to the high melting rate (Table 2) and the faster heat transfer through the ice crystals [31].

### 3.7. *Correlations*

Pearson correlation coefficient was employed to investigate the relationships between various measurements, including melting rate, hardness, particle size ($d_{10}$, $d_{50}$, $d_{90}$ and $d_{4,3}$), frictional factor at different sliding velocities (0.001, 0.01, 0.025 and 0.1 m/s) and temperature sweep (−5, 5 and 37 °C) along with TDS sensory and hedonic liking (Fig. 5). A strong correlation between instrumental data and sensory analysis indicated a significant connection between different parameters. The melting rate negatively correlated with smooth perception ($r = -0.99$), indicating that higher melting rates were associated with low perceived smoothness. Friction factors during temperature sweep were negatively correlated with particle size ($r = -0.99$), overall liking ($r = -0.99$), firm ($r = -1.0$) and smooth ($r = -0.97$) perception, while friction factors with velocity sweep were positively correlated with particle size ($r = 1.0$) and overall liking ($r =$

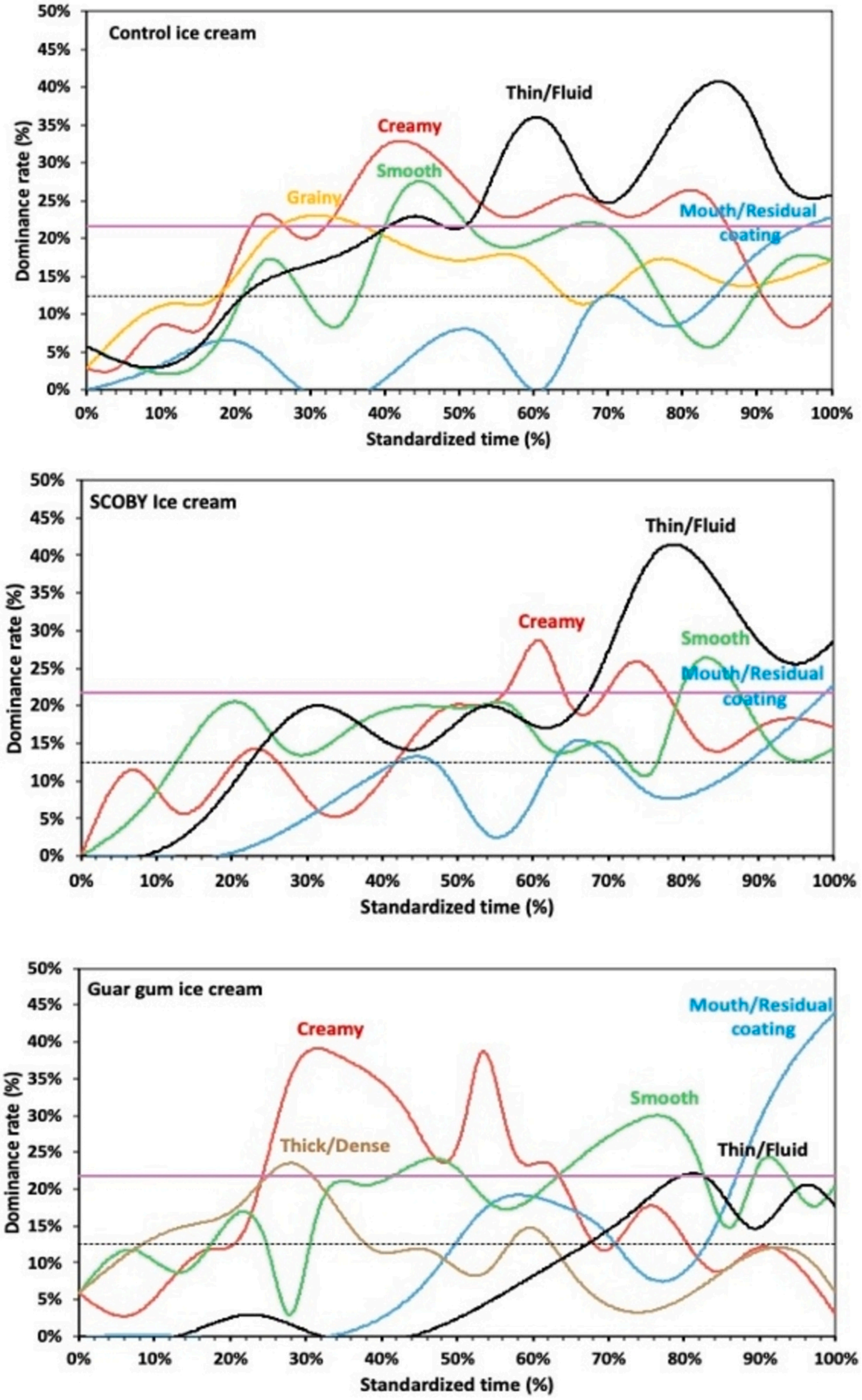

**Fig. 4.** TDS curves of control, SCOBY, and guar gum ice cream.

0.99). A negative relationship between the friction factor and smooth perception has also been reported in other studies [65–68]. Particle size distribution plays an important role in the friction factor of dairy products [69]. Smaller particles tend to entrain between the two rubbing surfaces and prevent dry contact, which, in turn, will result in a low friction factor. However, in the present study, the maximum size diameter below which 90 % of the sample volume exists is substantially larger (100 μm) than the roughness of the contact surface, resulting in a high friction factor at 0.025 m/s sliding velocity.

The mouthfeel liking exhibited positive correlations with mouth/residual coating ($r$ = 1.0) and smooth perception ($r$ = 0.99) but a negative correlation with grainy perception ($r$ = −0.99). Similar findings regarding mouthfeel liking and grainy perception were also reported for yoghurt samples [70]. The mouth/residual coating and

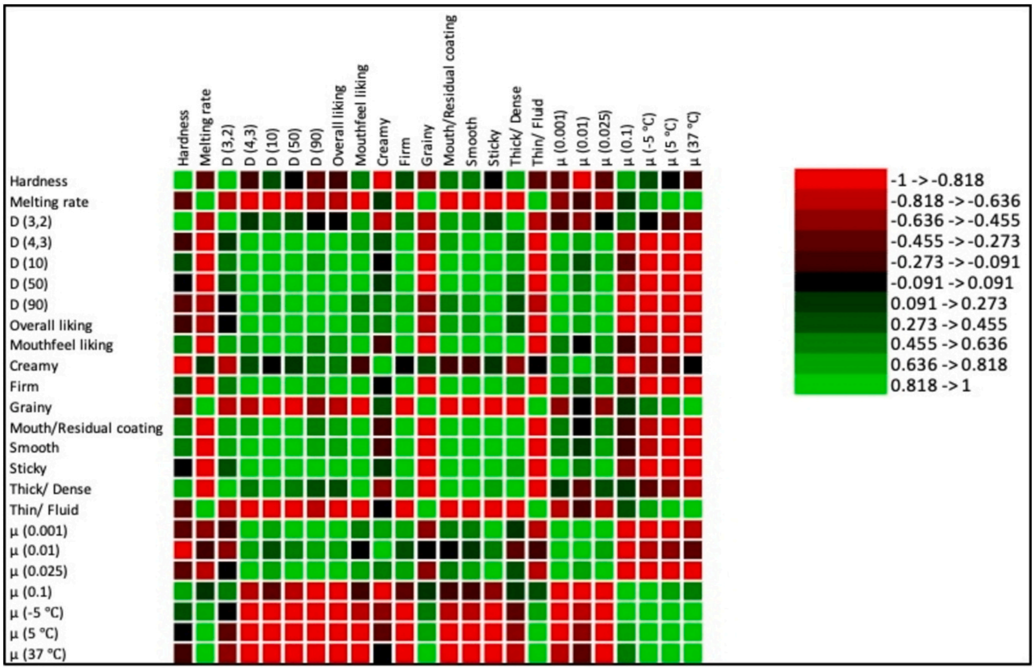

**Fig. 5.** Pearson correlation coefficient (*r*) between measures responses at $p < 0.05$ confidence level (Green = highly positively correlated, Red = highly negatively correlated).

smooth perception are highly correlated in ice cream [71], fermented whey [72], dairy and plant-based yoghurts [18,66], spreads [73] and fermented dairy products [74].

## 4. Conclusion

The present study examined the impact of incorporating SCOBY as an ingredient on the textural properties of ice cream, in comparison to other reference samples. The study highlighted that SCOBY ice cream exhibited distinct characteristics such as high probiotic content, lower pH, and higher titratable acidity than control and guar gum ice cream. The addition of SCOBY resulted in a firmer consistency and reduced melting rate than the control ice cream. The velocity sweep changes the frictional factor across all the samples, with a lower frictional factor in the boundary regime contributing to a heightened perception of creaminess in all variants. In the hydrodynamic regime (temperature sweep), SCOBY ice cream decreases the frictional factor compared to control ice cream, indicating improved lubrication. Correlation analysis unveiled key relationships, indicating smoothness is negatively associated with melting rate and frictional factors at temperature sweep. Moreover, mouthfeel and overall liking exhibit positive associations with attributes such as mouth/residue coating and smoothness and negative associations with thin/fluid perception. The findings enhance our understanding of the textural dynamics of ice cream formulations with the addition of SCOBY as an ingredient and offer valuable insights into the factors influencing consumer preferences and overall liking of SCOBY ice cream, particularly in the context of potential industrial applications. Future studies should investigate the effects of varying SCOBY concentration on ice cream microstructure and explore the impact of long-term storage on textural and mouthfeel properties, particularly for industrial applications.

## CRediT authorship contribution statement

**Annu Mehta:** Writing – review & editing, Writing – original draft, Software, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Lokesh Kumar:** Writing – review & editing, Visualization, Validation, Supervision, Software, Project administration, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Luca Serventi:** Writing – review & editing, Supervision, Resources, Project administration, Conceptualization. **James D. Morton:** Writing – review & editing, Supervision, Conceptualization. **Damir D. Torrico:** Writing – review & editing, Validation, Supervision, Resources, Project administration, Conceptualization.

## Declaration of competing interest

We are pleased to declare that there is no conflict of interest between the authors. This submission has not been submitted for consideration elsewhere.

## Data availability

Data will be made available on request.

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