



Design of a high protein, no added sugar pistachio spread using oleogel as fat replacer

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ABSTRACT

The production of multicomponent food items requires a correct ratio among the ingredients when specific quality parameters must be obtained. Recently, there has been an increase in the trend of consumption of healthy products, low in sugar and saturated fat and with increased protein intake. Nut based spreads need a high percentage of sugars and fat to guarantee their spreadability and mouth-melting behaviour. The present work was then aimed to design a pistachio spread without added sugars, low in saturated fat, spreadable and inducing positive sensory attributes to the consumer. An optimal formulation of a no added sugars and high protein pistachio spread was obtained by using olive oil based oleogel as unsaturated fat replacer and increasing the amount of milk originated powders (skimmed milk powder and whey protein concentrate) as structural modifier.

The optimization, done by mixture design, considered technological and sensorial aspects together. The experimental design consisted of twelve different formulations in which oleogel, whey proteins and skimmed milk were set as the ingredients while spreadability, oil binding capacity, and sensory attributes like meltability, mouth adhesiveness and undesirable flavours were used as responses. Polynomial regression models were used to fit the experimental data for each type of investigated response of pistachio spreads. The optimized formulation in terms of the selected quality responses was identified in a spread characterized by 20% of olive oil based-oleogel, 13.16% of whey proteins and 26.84% of skimmed milk, achieving an overall desirability score of 0.56. The optimized spread formulation showed a spreadability of 11.71 N mm, OBC of 95.88% and sensory scores of 5.61, 9.31 and 5.42 for meltability, adhesiveness to mouth and other flavours respectively.

1. Introduction

Sweet nut-based anhydrous creams are one of the most highly demanded and popular products in the confectionery market as they represent a very palatable comfort food used alone or in combination with other foods. However, nut-based spreads are considered unhealthy products due to the large amounts of sugar and saturated fats, which strongly influence the physical and sensory properties.

A key ingredient for the characteristic texture and flavour of spreads is sugar, which is commonly sucrose. Since its excessive consumption negatively affects health (obesity, type 2 diabetes and dental problems) sugar intake should be as low as possible (Turck et al., 2022).

The most common strategy for sugar reduction in food consists in its partial or total replacement by synthetic (aspartame, saccharin, sucralose) or natural low-calorie sweeteners such as polyalcohols (xylitol,

sorbitol, maltitol). However, the use of synthetic sweeteners is not always well accepted by consumers due to controversies about their safety, while the consumption of polyalcohols causes bloating, flatulence, or diarrhea as they are not completely absorbed in the intestinal tract (Castro- Munoz et al., 2022). Among the natural plant-based sweeteners, thanks to its high sweetener power, steviol glycosides present in the *Stevia rebaudiana*, are one of the most popular natural sweeteners currently employed as a total or partial substitute for sugars in many foodstuffs (Schiatti-Sisó et al., 2023).

As regards the fat phase used in nut-based spread, it is mainly composed of saturated fats such as cocoa butter, palm oil and coconut oil whose consumption also leads to negative health effects (Tirgarian et al., 2023). This is because the high intake of saturated fatty acids could be associated with potential health problems such as cardiovascular issues and cholesterol intake (EFSA NDA fats, 2010). The replacement of

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saturated with unsaturated fats represents a challenge for the food industry during the manufacturing of nut-based spreads since the quality parameters of the spreads such as spreadability at room temperature, creamy texture, and the absence of homogeneous structure without oil separation during storage depending on the high saturation degree of the used fat (Aydemir et al., 2021).

Recently, a strategy of great interest for replacing saturated fatty acids has been the structuring of edible unsaturated oils into solid-like systems using a relatively small fraction of the structuring agents (such as natural waxes, monoglycerides or polymeric nature) (Bascuas et al., 2021; Doan et al., 2016; Fayaz et al., 2017; Patel, 2017).

For nut-based spreads several applications of oleogels as healthier alternatives have been developed. Fayaz et al. (2017) studied pomegranate seed oil oleogel, with three different structuring agents (monoacylglycerides, beeswax and propolis wax), as a partial replacement of palm oil in chocolate creams. Hazelnut-based fillings containing oleogel made with rice bran oil and different percentages of beeswax were developed and characterized by Doan et al. (2016). Bascuas et al. (2021) developed different chocolate spreads using an oleogel with hydroxypropyl methylcellulose (HPMC) and xanthan gum as structuring agents while oil phase consists of olive oil and sunflower oil. More recently, Marra et al. (2023) developed cocoa hazelnut cream using oleogel made by olive oil glycerol monostearate as a total replacement for palm oil.

It is known that the rheological properties of anhydrous creams strongly depend on the sugar phase since they are a complex multi-phase system of solids-oil suspensions, in which the oily phase is fat while sweeteners, protein sources, flavourings and emulsifiers represent the dispersed phase (Aydemir et al., 2021; Shakerardekani et al., 2013). Therefore, formulating nut-based spreads low in saturated fats and low in sugars is quite challenging due to the direct impact on the textural and sensory characteristics of spreadable creams.

Considering the above and to the best of our knowledge, no papers are reported in the literature on the simultaneous employment of oleogel and pure stevia as total substitutes of the fat and the added sugar phase respectively in a spreadable cream. Therefore, the present study aims to design a pistachio spread without added sugars, low in saturated fat, spreadable and inducing positive sensory attributes to the consumer. Stevia extract was employed as a sweetener. A mixture design approach was used to optimize the replacement of solids from sucrose with proteins from milk and olive-oil/based oleogel as fat replacer; in particular, oleogel, whey proteins and skimmed milk were set as variables while spreadability, oil binding capacity, and sensory attributes were used as responses.

2. Material and methods

2.1. Materials

100% Pure Pistachio Paste was provided by Brontedolci SRL (Bronte, Catania, Italy). Glycerol Monostearate purified (GMS) was obtained from Thermo Fisher Scientific Inc. (Waltham, Massachusetts, USA) and L- α -Lecithin Soybean from Merck (Darmstadt, Germany). Rebaudioside A 97% (Stevia) was provided by A.C.E.F. s. p.a (Fiorenzuola D'Arda, PC, Italy). Skimmed milk powder, whey protein concentrate and pure vanilla aroma were purchased from Confemix s. r.l (Milano, Italy), ProteroCo GmbH (Germany), and Gioia group s. r.l. (Torino, Italy) respectively.

Olive oil and sucrose were bought from a local retailer.

2.1.1. Oleogel preparation

Oleogel was prepared according to Malvano et al. (2022) with some modifications. Glycerol monostearate (GMS) was dissolved at 75 °C in olive oil reaching a concentration of 5.3% (w/w) under magnetic stirring until the oleogelator was completely melted. The obtained mixture was cooled at room temperature and kept at this temperature for at least 24 h before being utilized.

2.1.2. Preparation and characterization of pistachio spread

The control pistachio spreadable cream (CS) recipe was obtained by preliminary trials focused on the selection of the best ratio among the usual ingredients used for the manufacturing of nut-based spreads. The control pistachio spread formulation included 40% pistachio paste, 34% sucrose, 15% palm oil, 9% skimmed milk, 1% whey protein concentrate, 0.7% soy lecithin and 0.3% vanillin.

Palm oil, pistachio paste and soy lecithin were first blended for 2 min with a 120 rpm stirring rate. Then, sucrose, skimmed milk, whey protein concentrate, and vanillin were added to the fat phase and the mixture was heated at 55 °C for 1.5 h using a food processor (TM21 Thermomix, Vorwerk, Wuppertal, Germany). Finally, a refining phase was carried out for 3 h using a wet grinder (Chocolate Melanger Wet Grinder PG503, Premier Wonder, India) with a speed of 130 rpm. All the pistachio spreads were characterized after 7 days ($t = 0$) and after 40 days ($t = 1$ month) at room temperature in the dark.

2.2. Oil binding capacity

The oil binding capacity (OBC) was calculated according to Fayaz et al. (2019) with some modifications. Briefly, 1 g of each sample was carefully weighted in Eppendorf tube and centrifuged at 13000 rpm for 15 min at 20 °C using a microcentrifuge (MicroCL 21R, Thermo Fisher Scientific Inc, Germany) to evaluate the oil release. The oil released from the samples was removed with a Pasteur pipette and weighed. The samples were run in triplicate and the OBC was calculated using the following equation:

$$OBC (\%) = \left[1 - \left(\frac{\text{mass of released oil}}{\text{mass of spreads sample}} \right) \right] 100 \quad (1)$$

2.3. Texture analysis

The hardness and spreadability of pistachio spreads were performed according to Marra et al. (2023) with a Texture Analyzer (LRX Plus, Lloyd Instruments, Chicago, USA) equipped with a load cell of 50 N.

The spread samples were filled into a female cone (90° angle) avoiding the formation of bubbles and rough head surfaces. The test was performed by penetrating the corresponding male probe (90° angle) into 18 mm depth at 0.5 mm/s speed. The hardness (in N) represents the maximum force used by the probe to penetrate the sample, while the spreadability was calculated as the area (in N mm) of the compression test (Force vs. time) by Nexygen Software 4.1 (Lloyd Instruments, UK). The penetration tests were performed by at least three independent measurements at room temperature.

2.4. Particle size measurement

A Mastersizer laser diffraction particle size analyzer equipped with Hydro 3000 dispersion unit (Malvern Instruments, Worcestershire, UK) was used to evaluate the particle size of spreads. The measurements were performed according to Juzhong and Balu (2017) with some modifications. Pistachio spread samples were mixed with isopropanol alcohol (99% pure) into a suspension solution with a ratio of 1:20 in a 20 mL glass tube. The mixture was then homogenized by a vortex mixer and an ultrasonic bath (ARGOLAB DU-100, MO, Italy) then the mixture was diluted into the dispersant (isopropanol alcohol) until 18–20 % obscuration was obtained. Stirring was also conducted to ensure that particles were independently dispersed.

Size distribution was quantified as the relative volume of particles in size bands and presented as size distribution curves. Particle Size Distribution (PSD) parameters included the volume percentiles $D(v,10)$, $D(v,50)$ and $D(v,90)$. For each sample, 10 measurements were performed.

2.5. Sensory evaluation

Sensory evaluation of pistachio spreads was performed by a panel made of 10 persons trained in the study of nut-based spreads according to Malvano et al. (2022) with some modifications. The judges were selected among professors and students of the Department of Industrial Engineering at the University of Salerno (Italy) based on their ability to identify odours and the 5 basic tastes. The ISO 3972 and ISO 5496 were used to recruit 9 assessors. Triangle tests were performed to evaluate the ability of the judges (Sipos et al., 2021).

Fifteen grams of the 12 different formulations and the control one were served to the panelists in white ceramic pans. The panelists were given a plastic spatula to spread the sample on a toasted bread slice and a plastic spoon to taste the sample. The panelists rated the samples using a nine-point scale. The attributes considered to perform the sensory evaluation, in accordance with Fidaleo et al. (2017) and Di Monaco et al. (2008), are listed in Table 1. The assessors evaluated the control and the 12 different pistachio spreads under study in nine different sessions (four for the first two sessions and five for the last). The sample evaluation order was randomized according to the incomplete block design, with three replications.

All procedures performed in this study involving human participants were carried out in accordance with the 1964 Helsinki Declaration and

Table 1
Attributes assessed in sensory evaluation.

Attributes	Attribute category	Definition	Evaluation techniques
Brightness	Visual	How much light appears to shine from the sample	Evaluate the amount of light reflected from the sample surface. (1 = not shiny; 9 = very shiny).
Adhesiveness to spoon	Non-oral texture	Property of the sample to stick to a spoon surface	Dip the spatula in the pan and evaluate the time of detachment. (1 = very poor; 9 = very good).
Fluidness	Non-oral texture	Force required to mix the sample	Dip the spatula in the cup and evaluate the force required to mix the sample. (1 = very poor; 9 = very good).
Spreadability	Non-oral texture	Property of the sample to be spread over a flat surface	Evaluate the force required to spread the sample. Use the toasted biscuit and the spatula to perform the test. (1 = very poor; 9 = very good).
Pistachio flavour	Gustative	Intensity of pistachio flavour	Taste 1/4 of the spatula content and evaluate the intensity of pistachio flavour. (1 = absent; 9 = very intense).
Other flavour	Gustative	Intensity of other flavour	Taste 1/4 of the spatula content and evaluate the intensity of other flavours. (1 = absent; 9 = very intense).
Meltability	Oral texture	Speed of sample melting	Place the sample between tongue and palate and evaluate its melting rate. (1 = very poor; 9 = very good).
Adhesiveness to mouth	Oral texture	Presence of coarse/big solid crystal that interact with the saliva on the palate	After meltability assessment, evaluate the force required to remove the sample from the palate with the tongue. (1 = very poor; 9 = very good).
Overall Acceptance		The level of liking the sample	Assesses the overall acceptability of the sample. (1 = very poor; 9 = very good).

its later amendments.

2.6. Experimental design and data analysis

The control pistachio spreadable cream was modified by means of the total removal of sugar and palm oil that were replaced with pure stevia and olive oil-based oleogel respectively. The Design of Experiment approach was used to optimize a new no added sugars formulation by changing the amount of whey proteins, skimmed milk and the fat phase. As fat phase, olive oil-based oleogel was used as replacer of palm oil. As sweeteners, pure stevia (0.08/100g_{spread}) was used for all the formulations. A D-optimal mixture design was used to optimize the experiments, which were carried out to evaluate the effect of olive oil-based oleogel (O, x₁), whey proteins concentrate (WP, x₂) and skimmed milk powder (SM, x₃) on the main quality parameters of pistachio spreads, The remaining part of formulation (40%), consisting of pistachio paste, soy lecithin and vanillin, remained unchanged.

The composition of the above ingredients was expressed as a relative mass fraction, defined by the following equation:

$$x_i = \frac{m_i}{m_1 + m_2 + m_3} \tag{4}$$

where m₁, m₂, m₃ represent the mass (g) of the O, the WP and the SM, respectively calculated on 100 g of spread. The sum of the relative mass fractions is 1:

$$\sum_{i=1}^3 x_i = 1 \tag{5}$$

The range of mass of the single ingredients in the mixture was established by performing preliminary tests: 20–30 g/100g for O (x₁), 8–20 g/100g for WP (x₂) and 10–32 g/100g for SM (x₃).

Table 2 presents the experimental design consisting of the twelve different formulations. The responses that were evaluated for each formulation were two physical properties (Spreadability (y₁), OBC (y₂)) and three sensory properties (Meltability (y₃), Adhesiveness to mouth (y₄), Other Flavour (y₅)).

2.7. Statistical analysis

JMP statistical software (SAS Institute. Inc. Cary, NC, USA) was used to constitute the mixture design, to analyze the experimental data and to identify the optimal pistachio spread formulation. The experimental data were fitted with the Scheffé model:

$$y_i = \beta_{11}x_1 + \beta_{12}x_2 + \beta_{13}x_3 + \beta_{112}x_1x_2 + \beta_{113}x_1x_3 + \beta_{123}x_2x_3 + \beta_{1123}x_1x_2x_3 \tag{6}$$

where y_i is the investigated response; β₁₁, β₁₂, β₁₃, β₁₁₂, β₁₁₃, β₁₂₃, β₁₁₂₃ are

Table 2
Pistachio spread formulations returned by the D-optimal mixture design.

Sample	Coded Values			Real Values (g/100g _{spread})		
	x ₁	x ₂	x ₃	O	WP	SM
O ₂₃ WP ₆ SM ₂₉	0.38	0.13	0.49	22.70	8.00	29.30
O ₂₄ WP ₁₁ SM ₂₅	0.40	0.18	0.42	24.00	10.85	25.15
O ₂₇ WP ₆ SM ₂₅	0.45	0.13	0.42	27.00	8.00	25.00
O ₂₅ WP ₂₀ SM ₁₅	0.42	0.33	0.25	25.00	20.00	15.00
O ₃₀ WP ₆ SM ₂₂	0.50	0.13	0.37	30.00	8.00	22.00
O ₂₀ WP ₁₃ SM ₂₇	0.33	0.21	0.45	20.00	12.80	27.20
O ₃₀ WP ₂₀ SM ₁₀	0.50	0.33	0.17	30.00	20.00	10.00
O ₂₀ WP ₆ SM ₃₂	0.33	0.13	0.53	20.00	8.00	32.00
O ₂₀ WP ₂₀ SM ₂₀	0.33	0.33	0.33	20.00	20.00	20.00
O ₂₂ WP ₁₇ SM ₂₁	0.36	0.28	0.35	22.00	16.94	21.06
O ₃₀ WP ₁₃ SM ₁₇	0.50	0.21	0.29	30.00	12.80	17.20
O ₂₈ WP ₁₇ SM ₁₅	0.47	0.28	0.25	27.89	16.83	15.28

O_{xx} (O: oleogel; xx: g/100g_{spread}).
WP_{xxx} (WP: whey proteins; xx: g/100g_{spread}).
SM_{xxx} (SM: skimmed milk; xx: g/100g_{spread}).

the regression estimates parameters and the x_1 , x_2 , x_3 are the investigated independent variables.

Considering desirability as an objective function to maximize, minimize or obtain the target value of the response, the optimal formulation was obtained by maximizing desirability in the range of value of the dependent variables set in the model as reference values. In particular, for spreadability, a maximum value was set using the spreadability of a market-leading nut-based cream as a reference value, while OBC was set at a maximum value. For meltability and adhesiveness to mouth parameters, a maximum value was set while for the other flavour parameter a minimum value. One-way analysis of variance (ANOVA) and multiple comparisons (Duncan's Test) were used to evaluate if differences among the sample were statistically significant (at $\alpha = 0.05$ level) by using the JMP statistical software.

Finally, after a positive control of the KMO (Kaiser-Meyer-Olkin) index and the Bartlett's sphericity test on the data, Principal Component Analysis (PCA) was employed to evaluate the relationships existing between the sensory attributes and the pistachio spread samples investigated (Malvano et al., 2022).

3. Results and discussion

Table 3 shows the number of the investigated spread samples with independent variables and the respective responses.

The experimental data were statistically analyzed to identify the best-fitted model for each independent variable. As reported in Table 4, the model functions showed a coefficient of determination ≥ 0.9 , indicating a good agreement between predicted and observed responses, except for other flavour parameter. Moreover, spreadability, OBC, meltability and adhesiveness to mouth had a $p < \alpha$ ($\alpha = 0.05$) and the lack-of-fit test for these responses is not-significant. This indicates that the model is relevant for spreadability, OBC, adhesiveness to mouth and meltability parameters. In contrast, the model is not relevant for the other flavour response.

To underline the effect of each ingredient and its interactions on the studied responses, the estimated parameters of the prediction models for each response are also reported in Table 5.

Based on the developed models, the results showed that the amount of skimmed milk significantly ($\alpha = 0.05$) influenced all the investigated responses, while whey proteins and olive oil-based oleogel significantly ($\alpha = 0.05$) influenced spreadability, OBC and meltability. Furthermore, physical parameters (spreadability and OBC) were influenced by the interaction among all investigated ingredients (skimmed milk, whey protein and olive oil-base oleogel).

3.1. Oil binding capacity (OBC)

The release of the oil in spreads is perceived as a quality defect by

consumers in terms of unpleasant appearance, taste and texture.

As reported in Table 6, pistachio spreads with an oleogel content in the range 20–24 % showed higher OBC values than pistachio spreads with a higher oleogel content in the range 27–30 %. Anyway, the fat crystalline network produced in all pistachio spreads showed a strong oil immobilizing capability as the registered OBC values are greater than 90%. As reported in the literature (Barroso et al., 2020; Cerqueira et al., 2017; Lopez-Martinez et al., 2015), the capability to entrap the mobile oil phase in a continuous network could be given by the ability of GMS oleogelator to gel vegetable oils with the formation of inverse lamellar phases, which are stabilized by strong hydrogen bonds.

Furthermore, it is interesting to point out that the OBC parameter is also strongly dependent on the interaction between the different fat profiles present in the formulation. In this regard, Ferro et al. (2019) observed that high oleic oils favour the packing of GM crystals in a very cohesive gel, promoting the formation of a well-structured system. In addition, Martins et al. (2016) highlighted the capability of long-chain triglycerides to create a strong oleogel network compared to medium-chain triglycerides.

The main triglycerides (TAGs) present in olive oil are oleic-oleic-linoleic (OOL), palmitic-oleic-linoleic (POL), oleic-oleic-oleic (OOO), and palmitic-oleic-oleic (POO) which account for more than 85% of the total triglycerides (Guerfel et al., 2012). A similar composition was found in pistachio paste, in which the main TAGs are oleic-oleic-oleic (OOO), oleic-linoleic-oleic (OLO), oleic-linoleic-linolenic (OLL), and linoleic-linoleic-linoleic (LLL), which account for more than 80% (Dyszal & Pettit, 1990; Holcápek et al., 2003). Based on these considerations, the fat stability in the pistachio spreads due to the interaction between the two different triglyceride profiles present in olive and pistachio oil was evaluated after a storage period of a month. All spreads showed OBC values higher than those observed at a time 0 confirming the cited previous research about the similarity of fatty acids composition and the presence of long-chain triglycerides that favour the formation of a well-structured fat system.

Finally, according to the prediction model identified for OBC response (y_2), this parameter is influenced not only by the fat phase but also by the whey proteins and skimmed milk content and their interactions. Based on the obtained results, it is possible to assert that as the percentage of non-fat solids increased, higher OBC values were registered. However, all the investigated formulations showed a good ability to avoid liquid/solid phase separation.

3.2. Texture properties

The textural properties of fat-based food products are not only derived from the physico-chemical properties of the fat phase but also its interaction with non-fat ingredients contained within. Hardness and spreadability are two such textural properties that depend heavily on

Table 3
Experimental data for each response (Spreadability, OBC, Meltability, Adhesiveness to Mouth, Other Flavour).

Formulation	Spreadability [N mm]	OBC [%]	Meltability	Adhesiveness to Mouth	Other Flavour
	y_1	y_2	y_3	y_4	y_5
O ₂₃ WP ₈ SM ₂₉	9.37 ± 0.11 ^c	94.43 ± 0.18 ^{bc}	5.90 ± 0.02 ^c	8.60 ± 0.03 ^{ab}	6.20 ± 0.04 ^b
O ₂₄ WP ₁₁ SM ₂₅	8.29 ± 0.29 ^d	94.44 ± 0.03 ^{bc}	5.80 ± 0.03 ^c	8.20 ± 0.02 ^c	6.50 ± 0.01 ^a
O ₂₇ WP ₈ SM ₂₅	7.41 ± 0.00 ^{ef}	93.69 ± 0.21 ^d	6.60 ± 0.01 ^b	6.60 ± 0.05 ^d	6.50 ± 0.03 ^a
O ₂₅ WP ₂₀ SM ₁₅	8.12 ± 0.32 ^{de}	92.40 ± 0.20 ^e	6.70 ± 0.05 ^b	6.00 ± 0.06 ^e	6.30 ± 0.01 ^{ab}
O ₃₀ WP ₈ SM ₂₂	7.36 ± 0.11 ^{ef}	92.00 ± 0.24 ^e	8.00 ± 0.10 ^a	5.70 ± 0.11 ^e	5.50 ± 0.07 ^{cd}
O ₂₀ WP ₁₃ SM ₂₇	11.49 ± 0.16 ^b	95.59 ± 0.18 ^a	5.80 ± 0.02 ^c	8.80 ± 0.03 ^a	5.70 ± 0.04 ^c
O ₃₀ WP ₂₀ SM ₁₀	10.26 ± 0.19 ^{de}	92.16 ± 0.18 ^{ef}	8.20 ± 0.06 ^a	5.30 ± 0.02 ^f	5.30 ± 0.02 ^d
O ₂₀ WP ₈ SM ₃₂	11.52 ± 0.39 ^b	95.04 ± 0.21 ^{ab}	5.30 ± 0.01 ^d	8.30 ± 0.0 ^{bc}	4.80 ± 0.04 ^e
O ₂₀ WP ₂₀ SM ₂₀	11.63 ± 0.06 ^b	94.86 ± 0.04 ^{bc}	5.70 ± 0.05 ^c	8.60 ± 0.11 ^{ab}	5.70 ± 0.01 ^c
O ₂₂ WP ₁₇ SM ₂₁	9.85 ± 0.09 ^b	94.71 ± 0.10 ^{bc}	5.80 ± 0.11 ^c	8.60 ± 0.07 ^{ab}	5.40 ± 0.06 ^d
O ₃₀ WP ₁₃ SM ₁₇	6.82 ± 0.17 ^f	91.72 ± 0.10 ^f	8.30 ± 0.02 ^a	5.30 ± 0.02 ^f	4.50 ± 0.02 ^f
O ₂₈ WP ₁₇ SM ₁₅	6.66 ± 0.05 ^f	92.46 ± 0.01 ^e	6.90 ± 0.01 ^b	6.70 ± 0.02 ^d	6.40 ± 0.05 ^{ab}
CS	14.23 ± 09.02 ^g	97.34 ± 0.01 ^g		8.40 ± 0.02 ^{ab}	1.00 ± 0.00 ^g

Different letters (a-f) in the same column reveal significant differences (at $\alpha = 0.05$ level) between the samples.

Table 4
Statistical parameters of the prediction models for each response.

	Spreadability	OBC	Meltability	Adhesiveness To Mouth	Other Flavour
Model	Significant	Significant	Significant	Significant	Not Significant
R ²	0.9865	0.9831	0.9665	0.9355	0.7657
Adjusted R ²	0.9703	0.9628	0.9264	0.8581	0.4847
p - value	0.00002	0.0003	0.0015	0.0076	0.1455
Lack of Fit	Not significant	Not significant	Not significant	Not significant	Significant
F value	60.8859	48.4624	6.5833	12.0865	2.7245
Standard Deviation	0.3288	0.2677	0.2882	0.5347	0.4794

Table 5
Estimated parameters of the response variables at optimum formulation.

	Spreadability		OBC		Meltability		Adhesiveness To Mouth		Other Flavour	
	Estimated parameter (β _i)	p - value	Estimated parameter (β _i)	p - value	Estimated parameter (β _i)	p - value	Estimated parameter (β _i)	p - value	Estimated parameter (β _i)	p - value
SM	11.449	<0.0001	94.879	<0.0001	5.479	<0.0001	8.353	<0.0001	4.878	0.0001
WP	11.177	0.002	87.919	<0.0001	6.252	0.017	2.211	0.531	4.409	0.196
O	15.178	0.003	82.888	<0.0001	17.699	0.001	-4.650	0.380	-10.376	0.062
O*WP*SM	-58.914	0.004	-45.999	0.006	1.225	0.915	-28.977	0.211	-26.211	0.208
WP*SM	1.818	0.683	14.649	0.008	-0.278	0.944	13.369	0.113	3.631	0.586
O*WP	-11.479	0.225	26.148	0.012	-13.091	0.137	24.831	0.131	31.281	0.052
O*SM	-23.486	0.056	11.065	0.052	-12.266	0.048	12.743	0.205	30.036	0.012

In **bold**, the values of the independent variable that significantly (at α = 0.05 level) affects the response.

Table 6
OBC (%) values of all pistachio spreads at t = 0 and t = 1 month.

Sample Code	t = 0	t = 1 month
O ₂₃ WP ₈ SM ₂₉	94.43 ± 0.18 ^{bcA}	95.05 ± 0.01 ^{bcB}
O ₂₄ WP ₁₁ SM ₂₅	94.44 ± 0.03 ^{bcA}	94.67 ± 0.09 ^{cdB}
O ₂₇ WP ₈ SM ₂₅	93.69 ± 0.21 ^{dA}	93.95 ± 0.03 ^{eB}
O ₂₅ WP ₂₀ SM ₁₅	92.40 ± 0.20 ^{eA}	93.82 ± 0.03 ^{eB}
O ₃₀ WP ₈ SM ₂₂	92.00 ± 0.24 ^{eA}	93.44 ± 0.04 ^{efB}
O ₂₀ WP ₁₃ SM ₂₇	95.59 ± 0.18 ^{aA}	96.73 ± 0.04 ^{aB}
O ₂₀ WP ₂₀ SM ₁₀	92.16 ± 0.18 ^{efA}	93.86 ± 0.24 ^{efB}
O ₂₀ WP ₈ SM ₃₂	95.04 ± 0.21 ^{abA}	95.78 ± 0.02 ^{bb}
O ₂₀ WP ₂₀ SM ₂₀	94.86 ± 0.04 ^{bcA}	94.95 ± 0.02 ^{cdB}
O ₂₂ WP ₁₇ SM ₂₁	94.71 ± 0.10 ^{bcA}	95.18 ± 0.05 ^{bcB}
O ₃₀ WP ₁₃ SM ₁₇	91.72 ± 0.10 ^{fA}	92.65 ± 0.02 ^{fb}
O ₂₈ WP ₁₇ SM ₁₅	92.46 ± 0.01 ^{eA}	94.03 ± 0.06 ^{deB}
CS	94.39 ± 0.10 ^{cA}	94.24 ± 0.17 ^{bcA}

Different letters (a-f) reveal significant differences (at α = 0.05 level) among the samples for each storage time, and different letters (A-B) reveal significant differences (at α = 0.05 level) for each sample during the storage time.

this relationship (West & Rousseau, 2018).

For all investigated pistachio spread formulations, hardness and spreadability parameters were analyzed, which correspond to the maximum force (N) used by the probe to penetrate the sample and the area of the compression test (Force vs time) respectively. Hardness is significantly associated with spreadability: the higher the hardness, the greater the resistance to spread (Abdolmaleki et al., 2022; Lopez-Martinez et al., 2015).

The obtained results (Table 7) highlighted that as the percentage of non-fat solids increased, the resistance to spreading increased and thus higher hardness and spreadability values were recorded compared to the values registered for spreads with lower amounts of non-fat solids. Moreover, the spreads with the highest non-fat solids content showed hardness and spreadability values close to those observed for the control spread.

These results agree with the prediction model identified for spreadability response (γ₁), which highlighted that the amount of non-fat solids and the interactions between solid particles (WP and SM) dispersed in the fat system (O) affect the structuration level of spread samples and thus their spreadability.

The texture results of pistachio spreads agreed with the OBC ones: as

Table 7
Hardness (N) and Spreadability (N mm) values of all pistachio spreads at t = 0 and t = 1 month.

Sample Code	Hardness [N]		Spreadability [N mm]	
	t = 0	t = 1 month	t = 0	t = 1 month
O ₂₃ WP ₈ SM ₂₉	2.26 ± 0.07 ^{bcA}	3.42 ± 0.03 ^{bcB}	9.37 ± 0.11 ^{cA}	11.90 ± 0.04 ^{cB}
O ₂₄ WP ₁₁ SM ₂₅	2.26 ± 0.01 ^{bcA}	2.82 ± 0.07 ^{bcB}	8.29 ± 0.29 ^{dA}	10.14 ± 0.29 ^{dB}
O ₂₇ WP ₈ SM ₂₅	1.90 ± 0.22 ^{cdA}	3.24 ± 0.02 ^{cdB}	7.41 ± 0.00 ^{efA}	10.29 ± 0.38 ^{efB}
O ₂₅ WP ₂₀ SM ₁₅	2.14 ± 0.13 ^{bcdA}	2.72 ± 0.16 ^{bcdA}	8.12 ± 0.32 ^{deA}	9.57 ± 0.07 ^{deB}
O ₃₀ WP ₈ SM ₂₂	1.95 ± 0.05 ^{bcdA}	2.55 ± 0.00 ^{bcdB}	7.36 ± 0.11 ^{efA}	8.72 ± 0.16 ^{efB}
O ₂₀ WP ₁₃ SM ₂₇	3.01 ± 0.08 ^{aA}	4.78 ± 0.10 ^{ab}	11.49 ± 0.16 ^{bA}	15.66 ± 0.09 ^{bb}
O ₃₀ WP ₂₀ SM ₁₀	3.79 ± 0.06 ^{bA}	3.79 ± 0.06 ^{bb}	10.26 ± 0.19 ^{deA}	10.26 ± 0.03 ^{deB}
O ₂₀ WP ₈ SM ₃₂	2.87 ± 0.05 ^{aA}	3.67 ± 0.21 ^{ab}	11.52 ± 0.39 ^{bA}	12.63 ± 0.07 ^{bb}
O ₂₀ WP ₂₀ SM ₂₀	3.14 ± 0.10 ^{aA}	3.47 ± 0.05 ^{aA}	11.63 ± 0.06 ^{bA}	13.23 ± 0.04 ^{bb}
O ₂₂ WP ₁₇ SM ₂₁	2.73 ± 0.33 ^{aA}	3.92 ± 0.17 ^{ab}	9.85 ± 0.09 ^{bA}	13.86 ± 0.11 ^{bb}
O ₃₀ WP ₁₃ SM ₁₇	2.14 ± 0.19 ^{bA}	3.35 ± 0.10 ^{bb}	6.82 ± 0.17 ^{fA}	10.07 ± 0.04 ^{fb}
O ₂₈ WP ₁₇ SM ₁₅	1.72 ± 0.08 ^{dA}	3.06 ± 0.01 ^{db}	6.66 ± 0.05 ^{fA}	9.70 ± 0.17 ^{fb}
CS	3.11 ± 0.11 ^{aA}	3.59 ± 0.19 ^{aA}	12.83 ± 0.20 ^{aA}	13.70 ± 0.21 ^{aA}

Different letters (a, b, c, ...) reveal significant differences (at α = 0.05 level) among the samples for each storage time, and different letters (A, B, C, ...) reveals significant differences (at α = 0.05 level) for each sample during the storage time.

non-fat solids increased, OBC, as well as hardness and spreadability values increased. Additionally, their values increased during the storage period (Table 8). As reported below, this behaviour could be attributed to a similarity in fatty acids composition between fat phases that ensures a strong fat structure but also to a high inter-particulate interaction among particles dispersed in a fat system, producing a strong food matrix (Babin et al., 2005; Palla et al., 2021)

Table 8
The values of D (v, 10), D (v, 50) and D (v, 90) of all pistachio spreads.

Sample Code	D (v, 10) (µm)	D (v, 50) (µm)	D (v, 90) (µm)
O ₂₀ WP ₂₀ SM ₂₀	1.85 ± 0.007 ^f	10.74 ± 0.054 ^g	29.30 ± 0.274 ^{cde}
O ₂₅ WP ₂₀ SM ₁₅	1.91 ± 0.006 ^{de}	10.90 ± 0.082 ^{ef}	29.78 ± 0.394 ^{bcd}
O ₃₀ WP ₁₃ SM ₁₇	1.93 ± 0.008 ^{de}	10.46 ± 0.055 ^h	29.10 ± 0.400 ^{de}
O ₂₈ WP ₁₇ SM ₁₅	2.02 ± 0.008 ^{ab}	11.75 ± 0.058 ^c	30.55 ± 0.341 ^b
O ₂₀ WP ₈ SM ₃₂	2.02 ± 0.008 ^{ab}	12.86 ± 0.114 ^a	33.48 ± 0.756 ^a
O ₂₄ WP ₁₁ SM ₂₅	1.89 ± 0.009 ^e	10.65 ± 0.129 ^{gh}	28.93 ± 0.953 ^e
O ₃₀ WP ₂₀ SM ₁₀	1.95 ± 0.007 ^{cd}	10.55 ± 0.071 ^{gh}	30.60 ± 0.707 ^{bc}
O ₂₇ WP ₈ SM ₂₅	1.72 ± 0.008 ^g	8.45 ± 0.270 ^j	23.34 ± 0.270 ^g
O ₂₀ WP ₁₃ SM ₂₇	1.72 ± 0.011 ^g	12.44 ± 0.311 ^b	30.35 ± 0.129 ^{bc}
O ₃₀ WP ₈ SM ₂₂	1.99 ± 0.011 ^{bc}	11.22 ± 0.503 ^d	30.06 ± 0.503 ^{bcd}
O ₂₃ WP ₈ SM ₂₉	1.85 ± 0.035 ^f	11.33 ± 0.170 ^d	30.08 ± 0.170 ^{bcd}
O ₂₂ WP ₁₇ SM ₂₁	1.66 ± 0.005 ^h	9.34 ± 0.353 ⁱ	26.30 ± 0.353 ^f
CS	2.05 ± 0.043 ^a	11.10 ± 0.194 ^{de}	30.26 ± 0.194 ^{bc}

Different letters (a-g) reveal significant differences (at α = 0.05 level) among the samples.

3.3. Particle size measurement

The particle size distribution is an important technological and quality parameter as it influences both flow property as well as the consumer acceptance of the final product. The production process of nut-based creams requires a step for the reduction of solid particle size called refining which the main aim is to reduce the solids to a specific particle size that depends on the specific product (Fidaleo et al., 2017).

As can be seen from Table 8, after 3 h of refining all the pistachio spreads showed D(v,90) values ranging from 26.30 to 30.60 µm, in accordance with Bolenz et al. (2013) who refined white chocolate in ball mills and estimated D(v, 90) in the range 28.14–30.50 µm. Moreover, Bolenz et al. (2013) and Fidaleo et al. (2017) stated that particles higher than 35 µm create a 'sandy mouthfeel' on the palate in sensory perception that must therefore be avoided as it is considered to have a low level of consumer acceptability.

Fig. 1 reports the Particle Size Distribution (PSD) curves of all pistachio spreads investigated. The distributions appeared almost unimodal, presenting a left shoulder that corresponded to smaller particles with sizes between 0.4 µm and 1 µm. These results agree with Cavella et al. (2020), who recorded a similar PSD for chocolate paste after a mill ball refining process.

3.4. Sensory evaluation

In the current study, principal component analysis (PCA) was used to investigate the relationship among sensory attributes and all pistachio spreads investigated. An important capability of the PCA method is to reduce the total variability of variables in two principal components (PC), PC1 and PC2. According to the biplot graph in Fig. 2, two PCs described 79.84% of the variation of the total variables, with significant contributions of 67.87% and 11.97% for the first and the second PC respectively.

Regarding the influence of sensory attributes on the investigated

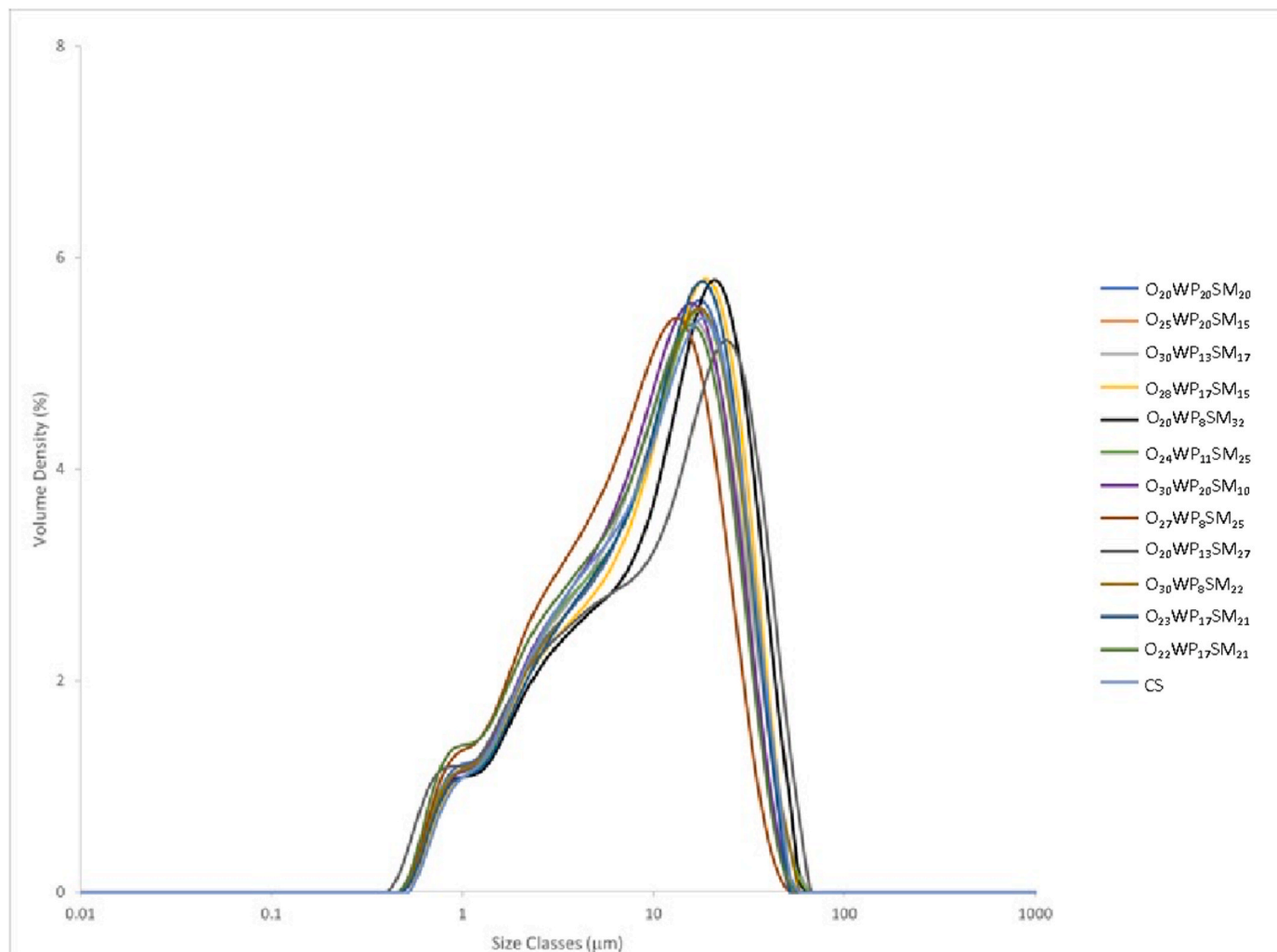


Fig. 1. Particle size distribution of 12 sugar-free pistachio spread formulations.

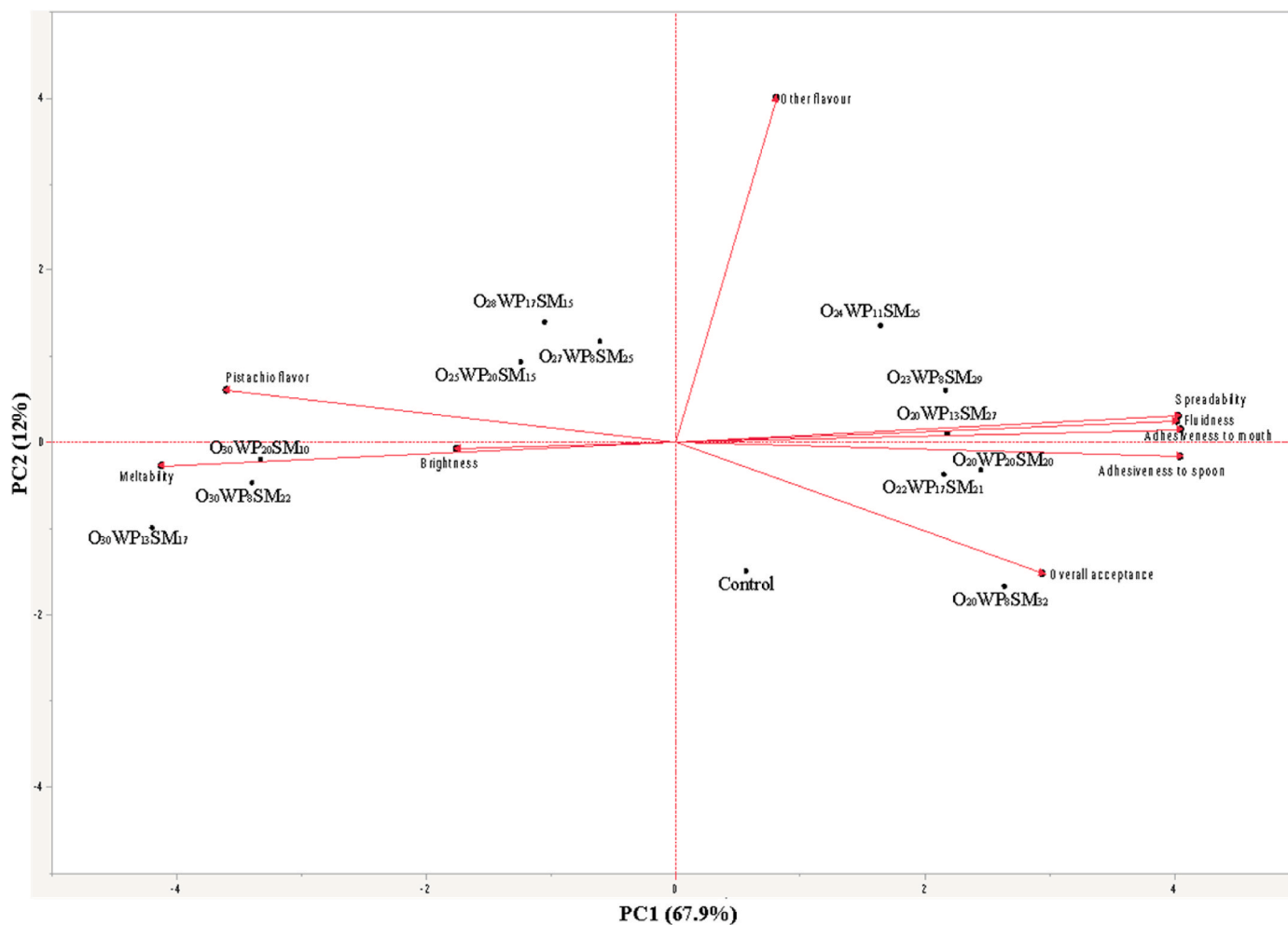


Fig. 2. PCA 2D-plot (score and loadings) of sensory attributes for the pistachio spreads samples.

spreads, it has been possible to observe that all the spreads with the lowest amounts of olive oil oleogel and, thus, the highest amount of non-fat solids, are characterized by high spreadability, adhesiveness to the mouth, adhesiveness to the spoon and fluidness, that are sensory parameters negatively correlated with meltability attribute. Similar results were obtained by Di Monaco et al. (2008) who studied the sensory attributes, and rheological and thermal properties of different hazelnut-based commercial spreads. According to texture and OBC results, these spreads revealed a more cohesive structure, which has been confirmed in the sensory evaluation.

On the contrary, the spreads characterized by a high amount of oleogel present a high meltability. No correlations have been found between pistachio flavour and the other sensory attributes investigated.

3.5. Optimization of the pistachio spreads

The optimization step was carried out to achieve the optimal amounts of each investigated ingredient to produce an optimal pistachio-based spread (palm oil-free and with no added sugars) with appropriate texture and sensory characteristics, taking a market-leading spreadable cream as a reference.

Fig. 3 reports the score of desirability functions (spreadability (Fig. 3a), oil binding capacity (Fig. 3b), meltability (Fig. 3c), adhesiveness to mouth (Fig. 3d) and other flavours (Fig. 3e)) and of total desirability (Fig. 3f) versus the composition of O, WP and SM. In red are reported the values of those functions at the optimal composition. In blue their relative interval of confidence. The total desirability varied

from 0.56 to 0.25, with an optimal value of 0.56 in correspondence of $x_1 = 0.33$, $x_2 = 0.22$ and $x_3 = 0.45$. For values of x_1 higher than 0.33, the desirability decreases. As a function of x_2 , total desirability reaches a maximum plateau for x_2 higher than the optimal value. The total desirability has a clear maximum at $x_3 = 0.45$.

The formulation corresponding to the optimized conditions corresponded to the following % (w/w) of oleogel, WP and SM, respectively 20.00, 13.16, and 26.84% SM.

The resulting combination achieved an overall desirability score of 0.56 which is indicative of the acceptable ability of the models to achieve the desired textural and sensory qualities (Azarbad et al., 2019).

Referred to 100 g of total mass, the optimized formulation presents 3.55 g of saturated fat (64% less than the control), 16.43 g of sugars (vs 41.71 g of sugars referred to the control), and 100% more proteins (24.84 g in the optimized formulation vs 12.46 g of proteins in the control), while the total energy content remains almost unvaried (544.53 kcal and 565.00 kcal respectively for the optimized formulation and the control).

4. Conclusions

In this work, a mixture design approach was used to identify the best formulation with the desired quality parameters of a pistachio-based spread without added sugars and using an unsaturated oleogel as a fat replacer.

Applying the Design of Experiment, 12 different formulations were tested and analyzed in terms of spreadability, oil binding capacity,

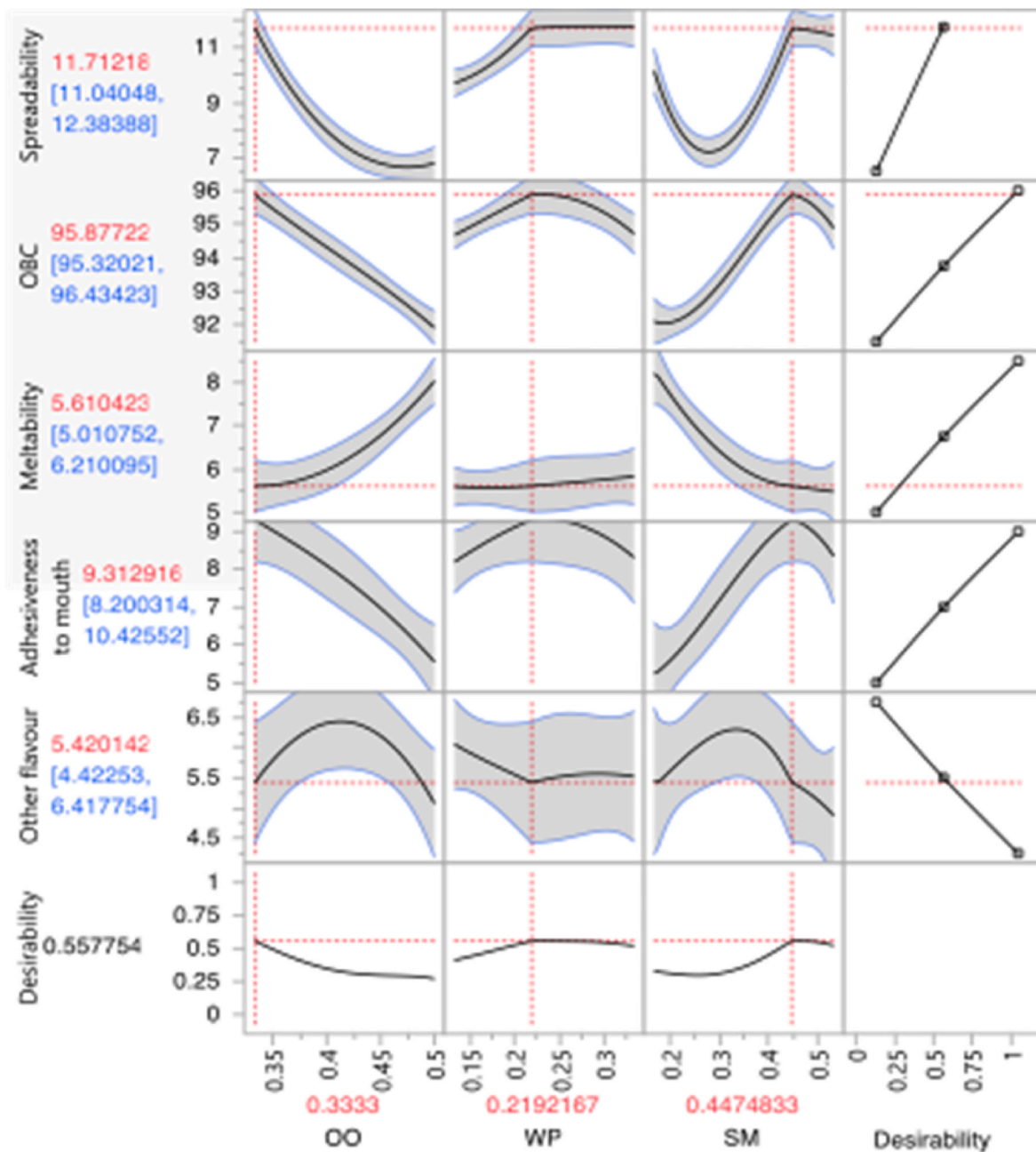


Fig. 3. Desirability functions and total desirability vs olive oil-oleogel, whey proteins, skimmed milk compositions.

meltability, adhesiveness to mouth and other undesirable flavours. Based on experimental results, a mathematical model for each investigated property was identified and used to calculate the optimal formulation to reach the highest overall desirability.

The results highlighted that, as the percentage of non-fat solids in spreads increased, higher oil binding capacity values were registered, as well as higher hardness and spreadability values, compared to the values of spreads with lower percentages of non-fat solids. The increment of non-fat solids resulted, in fact, in a higher inter-particle interaction among dispersed particles, producing a stronger food matrix.

Storage time seems to influence texture properties: hardness and spreadability of pistachio spreads gradually increased during the storage period due to a reorganization of glycerol monostearate crystals in post-production.

As regards the particle size distribution, after 3 h of refining all the pistachio spreads showed $D(v,90)$ values ranging from 26.30 to 30.60

μm , avoiding the sandy sensation on the palate, a sensation considered to have a low level of acceptability by the consumer.

Finally, according to the desirability function approach, the results of the optimization highlighted that the best combination of ingredients was identified as 20.00% oleogel, 13.16% whey proteins, 26.84% skimmed milk.

The forthcoming investigation will be devoted to introducing in the experimental set-up a larger set of data, both in terms of technological and sensory parameters, to tune the desirability function optimization, and to make the same methodology (in this work limited to pistachio spread) applicable to other cream-based products.

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CRedit authorship contribution statement

Francesca Malvano: Writing – original draft, Methodology, Investigation, Data curation. **Eleonora Muccio:** Investigation, Data curation. **Fernanda Galgano:** Writing – review & editing, Investigation, Formal analysis. **Francesco Marra:** Writing – review & editing, Supervision, Methodology, Formal analysis, Conceptualization. **Donatella Albanese:** Writing – review & editing, Supervision, Resources, Methodology, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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