**Oxidative stability and characterization of oleogels made from safflower oil-based beeswax and rice bran wax and their utilization in cake production**

Running title

**Role of oleogels in cake properties and quality**

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**Abstract**

In this study, oleogels based on safflower oil were produced from beeswax and rice bran wax at different ratios. It was aimed to produce cakes with high level of unsaturated fatty acids by using these oleogels as a shortening replacer. The characterization and oxidative stability of oleogels were investigated. Oil binding capacity (OBC), solid fat content (SFC) and crystallization time (CT) were determined in oleogels. Moisture content, pH, texture and sensory analysis were performed in the cakes. In addition, fatty acid composition, free fatty acidity, peroxide value, conjugated diene-triene and 3-monochloropropane-1,2-diol (3-MCPD) and glycidyl analyzes were performed pre- and post-cooking in oleogels and shortening. SFC increased as gelator concentration increased. Beeswax showed the highest OBC. The shortest CT was determined in rice bran wax. No changes were observed in the fatty acid composition of safflower oil following oleogelation. The change in major fatty acids post-cooking was also not significant. Cakes made with oleogel were acceptable in terms of texture and sensory properties compared to cake produced using shortening. Sensory results showed that some cakes produced with oleogels more acceptable than control. This study revealed that oleogels produced with safflower oil-based beeswax and rice bran wax with high unsaturated fatty acid content can be used in cakes rather than commercial shortening.

**KEYWORDS:** 3-MCPD, beeswax, rice bran wax, oleogel, cake, solid fat content

**INTRODUCTION**

Fats in solid form are preferred to provide desired structural properties in bakery products. However, solid fats pose a risk to human health because of their high saturated and trans-fat content. Vegetable oils with high unsaturated fat content are converted into solid form by hydrogenation technique. However, it has been reported that trans and saturated fats produced during the hydrogenation process cause many health problems such as cardiovascular diseases, oxidative stress, endothelial dysfunction, obesity, increased insulin resistance, diabetes and cancer (Micha and Mozaffarian, 2009). In recent years, techniques such as interesterification and fractionation have been used to reduce and prevent these problems. In addition to these, organogelation is one of the alternative methods. Organogelation is a promising alternative to provide structure and texture to vegetable oils with no trans and/or saturated fatty acid formation. Organogels, if the liquid phase is oil, are called as oleogel. Compounds capable of gelling the liquid phase in oleogels are called oleogelators. The ability of oleogelators to gel oils even at low concentrations has increased their involvement in the food industry. Oleogels are used as breakfast spreads, margarines, animal fat alternatives, ice cream and cocoa butter alternative in the food industry. Oleogel is a continuous, thermo-reversible and three-dimensional networked gel formed by asymmetric crystallization or self-agglomeration of low molecular weight and limited solubility oleogelator in a consumable vegetable oil (Mert and Demirkesen, 2016b). In the oleogelation technique, the liquid phase is either an organic solvent or crude/vegetable oil, and the gel agent (oleogelator) is low molecular weight organic gelators or polymeric gelators. The immobilization of the liquid phase in the gel network is achieved by the polymeric gelators forming cross or mixed bonds as a result of physical and chemical interactions, and the low molecular weight organic gelators forming large clusters as a result of physical interactions. The gel structure is then strengthened by weak interchain interactions such as hydrogen bonds, Van der Waals forces, and π-π interactions (Shapiro, 2011).

Although the oleogel preparation process varies according to the oleogelator and vegetable oil, it is generally carried out at high temperature and in batch system by mixing the oleogelator and vegetable oil. While the oleogelator is melted in vegetable oil at high temperatures, it is important to consider the sensitivity of unsaturated fatty acids to oxidation at this stage (Jimenez-Colmenero et al., 2015). Different organogelators such as triacylglycerol, diacylglycerol, monoacylglycerol, fatty acids, fatty alcohols, lecithin, sorbitan tristearate, phytosterols, waxes, ceramides and γ-oryzanol are used in the food industry to provide liquid oils a three-dimensional gel structure (Demirkesen, 2017). It is known that the liquid phase in the oleogel technique consists of consumable vegetable oils such as canola oil, sunflower oil, olive oil, hazelnut oil, soybean oil, coconut oil and safflower oil. Safflower oil is used in the food industry as frying oil, as well as in the production of salad dressings and margarines. Safflower oil, obtained from safflower, a spiny plant from the daisy family (Asteraceae), is similar to sunflower oil in that it is tasteless and colorless. The fact that it is rich in linoleic acid, a polyunsaturated essential fatty acid, makes safflower oil important for human consumption both nutritionally and therapeutically (Ny and Rah, 2015).

Utilization of oleogels as a shortening substitute, an important component of bakery products, is currently under investigation. Shortening is a fat traditionally used to make bakery products such as pastries, cakes and cookies. It prevents gluten and starch granules from sticking together during mixing. Thus, the shortening ensures the lubrication of the gluten particles and the products turn into a soft and smooth shape after cooking (Mert and Demirkesen, 2016b). However, because of the presence of high levels of saturated fatty acids and possible trans fatty acids, the use of shortening has been reduced. It has been observed that vegetable oil-based products can be used instead of shortening in bakery products (Jang et al., 2015). The fatty acid profile of the gelled oils is preserved, as is the functionality and texture of the final products. Edible oleogels can be used effectively to help improve nutrient profiles and improve functional properties of food products (Stortz et al., 2012).

Beeswax is a multi-component wax with a melting temperature of 49.9 °C and the ability to form gels at least 1% concentration, mainly composed of hydrocarbons, wax esters, fatty acids and fatty alcohols (Alizadeh et al., 2020). Rice (*Oryza sativa*) bran wax is a natural plant wax obtained from rice bran, which is a by-product after rice milling. Rice bran wax consists of polycosanol, which are bioactive compounds. Policosanol is a long chain alcohol group with a carbon length of 20-36 atoms. Policosanol can self-assemble and form the three-dimensional structure of organogels. Many studies have demonstrated the potential application of long-chain fatty alcohols or policosanol as organogelators (Pandolsook and Kupongsak, 2020).

In this study, safflower oil based oleogels were obtained by using rice bran wax and beeswax at different concentrations. Properties such as CT, OBC, color, free fatty acidity (FFA), fatty acid composition, SFC, conjugated diene (K232) -triene (K270) and peroxide number (PV) of the obtained oleogels were determined. Cakes were produced with oleogels. Cake produced with Shortening was considered a control. The effects of mentioned oleogels on the quality of cakes were investigated by textural properties and sensory analyzes in the produced cakes. In addition, oil was extracted from the cakes with the levels of oxidation products, 3-MCPD and glycidyl formation in these oils were investigated. In short, it is aimed to obtain healthier cakes in terms of nutrition by using oleogels rather than shortening.

**MATERIALS AND METHODS**

**Material**

Beeswax, rice bran wax and commercial shortening were obtained from commercial company (Aven Chemistry Co., Van, Türkiye). Safflower oil was obtained from the Turkish Ministry of Food, Agriculture and Livestock, Field Crops Central Research Institute. 3-MCPD and 3-chloro-1,2-propane-1,1,2,3,3-d5-diol (3-MCPD-d5), glycidyl stearate, diethyl ether, methanol, hexane, sodium hydroxide, sodium bromide, ethyl acetate, phenylboronic acid (PBA), acetone and toluene were obtained from Sigma-Aldrich (Steinheim, Germany). The purity of the chemicals was ensured.

**Preparation of oleogels**

Beeswax was added to the safflower oil at various concentrations as 3%, 5% and 10%, and rice bran wax at 5%, 7% and 10% by weight. These were heated with agitation until they were completely dissolved and then cooled to room temperature to form oleogels. The codes of the prepared oleogels and the cakes are given in Table 1.

**Preparation of cakes**

Cake samples were prepared based on the AACC approved method 10-90 with slight modification (AACC, 2000). The formulation was achieved as 100 g flour, 100 mL of water, 80 g sugar, 50 g shortening/oleogel, 12 g non-fat dry milk, 8 g dried egg white powder, 3 g baking powder, 1.5 g salt and 1 g vanilla. Cake batters were prepared by mixing these ingredients in the KitchenAid Mixer (5K45SS; Kitchen Aid, Elk Grove Village, IL). For the oleogel cakes, shortening was replaced with each oleogel. The cake batter was weighed 225 g into baking pans (Ø: 15 cm, height: 3.5 cm) and baked in an electric oven (GN SCC102E, Öztiryakiler, İstanbul, Türkiye) for 24 min at 175 °C. Following the baking, cakes were removed from pans and cooled at room temperature for 2 h. Baking experiment was carried out in triplicate.

**Crystallization time determination**

For the determination of CT, the oleogels added in the tubes were completely melted in a water bath at 90 ºC, and the temperature was stabilized by keeping them in the water bath for 2 h. Then, the exact time of gelation in the tubes, which were removed from the water bath to room temperature, was determined as a result of observation. The CT was recorded when the flow stopped when the tubes were turned into 90º and parallel (Dassanayake et al., 2009).

**Oil binding capacity (OBC) of oleogels**

Oleogel samples were completely melted in a water bath at 90 ºC and transferred to tared Eppendorf tubes (a) in an amount of approximately 1 mL. Eppendorf tubes containing oleogel were kept at +4 °C for an hour for gelation. After the gelation was completed, the eppendorf tubes (b), (epp. tube + sample) were weighed again, centrifuged at 20 ºC for 15 minutes at 10.000 rpm. After the tubes were removed from the centrifuge, they were turned inside out and drained for 3 minutes. Then the samples were reweighted and (c) %OBC was calculated by using Equation 1 (Da Pieve et al., 2011).

*%* *free oil =  %OBC = 100 - %* *free oil* (1)

**Solid fat content measurement by NMR**

The SFC of the oleogel samples was determined by Minispec mq-20 NMR Analyzer (Bruker, Rheinstetten, Germany) with small modifications in the TS EN ISO 8292 method (ISO, 2012). The oleogel samples were completely melted in a stirred water bath at 75 °C and filled into NMR tubes at 4-5 cm intervals. To form melted oleogels, the tubes were kept in a water bath at 0 °C for 30 minutes. The tubes were then kept at 30 °C and 35 °C for 30 minutes for measurement. At the end of the time, the tubes, which were removed from the water bath, were read in the NMR device. The measurements were performed in triplicate.

**Specific gravity**

The weights of the cake batter produced with a container of known volume were determined. Then, the density (g mL-1) was calculated by dividing the dough weight by the weight of water in the same container (1 g water = 1 ml water) (AACC, 2000).

**Specific volume**

The volumes of the cakes baked in the study were determined according to the AACC (Method 55-50.01) using the principle of substitution with rapeseed seeds (AACC, 2010). The specific volumes of the cakes were calculated by weighing the cakes following an hour baking and dividing the determined cake volumes by their weights.

**Moisture content**

The moisture content of the cake was determined using a moisture analyzer (MX-50, A&D, Tokyo, Japan). The measurements were performed in triplicate.

**pH determination**

The basis of the measurement is based on the principle of measuring the potential difference between the liquid in the pH meter electrode and the homogenized cake liquid. After weighing 25 g of sample into a 150 mL beaker and making up to 100 mL with distilled water, the mixture of cake and pure water was homogenized with Ultraturrax. Then, pH measurement was performed by immersing the pH meter electrode in the solution (AOAC, 2000).

**Sensory analysis**

The sensory parameters (appearance, pore structure, moisture, taste and aroma, mouthfulness and general acceptance) of the cakes produced were evaluated by 15 panelists, most of whom were faculty members of the Department of Food Engineering. Panelists were trained prior to evaluation to become familiar with quality attributes of cake samples. Sensory evaluation was carried out in the research laboratory under controlled temperature and lighting conditions. Cakes were presented in 2 cm cubes for testing on white plastic plates coded with random numbers and served in random order. Drinking water was used for rinsing. A hedonic scale was used in the evaluation and each feature was evaluated over 5 points. The scale of values ranged from "extremely dislike" to "extremely like", with the highest and lowest scores corresponding to ‘1’ and ‘5’ respectively.

**Textural measurement**

Texture profile analysis was (TPA) test was carried out by using a texture analyzer (TA-XT2, Texture Technologies, Hamilton, MA, USA) to determine the textural properties of cakes. The textural properties of the samples were determined by the 'Measure Force in Compression' method (Test speed: 3.0mm sec-1, Post-Test velocity: 10 mm s-1 and Distance: 23 mm) using a 45° acrylic conical probe. From the graphics obtained, the textural properties of the samples such as firmness, adhesiveness, flexibility, cohesiveness, gumminess, chewiness and elasticity were calculated with the help of the software program of the device.

**Color measurement**

Color measurements of oleogels and cakes produced in the study were made using color meter (PCE Instruments, PCE-CSM1, Southampton, UK). Before the measurement, the device was calibrated with its standard and measurements were made directly from the sample surface in three different areas. Color determinations were made on the crust and crumb in the cakes. Results were expressed in the CIE L\*a\*b\* color space. In this standard, L\* stands for brightness/darkness, a\* stands for redness (+)/greenness (-), b\* stands for yellowness (+)/blueness (-) (Öğütçü et al., 2008).

**Determination of fatty acid composition**

After the cakes were ground using a laboratory grinder, the oils in the cake powder were extracted with hexane as solvent in a soxhlet extractor. First, fatty acids were derivatized to fatty acid methyl esters. For analysis (IUPAC, 1992) GC/MS (Shimadzu GC/MS QP 2010, Japan) instrument was used. The device was set to the conditions as column: DB-23 (60m x 0.25mm, 0.25 μm), carrier gas: helium, total flow:36.6 mL min-1, column flow:0.66 mL min-1, linear velocity:21.2 cm sec-1, split ratio:50, initial temperature: 80 ºC, temperature program: 10 ºC min-1, final temperature: 220 ºC, injection temperature: 250 ºC, detector temperature: 250 ºC, ion source temperature: 200 ºC, total analysis time: 34 min.

**Determination of free fatty acidity**

Percentage of FFA of oil samples extracted from oleogels and cakes was determined according to AOCS Official Method Ca 5a-40 (AOCS, 2004).

**Peroxide value measurement**

Detection of PV in oil samples extracted from oleogel and produced cakes was determined according to AOCS Official Method Cd 8-53 (AOCS, 1989a).

**Conjugated-diene and conjugate-triene analysis**

Conjugate-diene (K232) and conjugate-triene (K270) analysis was performed according to AOCS Official Method Ch 5-91 (AOCS, 1989b).

**Analysis of 3-MCPD and glycidyl**

3-MCPD and glycidyl levels were determined according to the DGF C VI 18 (10) (DGF, 2011) standard method with some modifications detailed in our previous article (Gündüz et al., 2023).

**Statistical analysis**

SPSS (version 20.0 for Windows, SPSS Inc., Chicago, Illinois) package program was used to evaluate the results obtained in the study. A one-way analysis of variance (One-Way ANOVA) was used to determine whether there was a statistically significant difference between the group means. The significance of the difference was determined using the Duncan multiple comparison test.

**RESULTS AND DISCUSSION**

**Some physical properties of oleogels**

The OBC, SFC, CT and color values of oleogels are given in Table 2. OBC was lower than the others in the oleogel made with 5% rice bran wax (p<0.05). In other oleogels, this value was close to each other in the range of 98.03-99.98%. OBC values in beeswax oleogels did not change significantly with wax concentration. (p>0.05). On the other hand, OBC values in oleogels other than OR5 were close to that of shortening (S) (p<0.05). These results are consistent with the findings in other studies (Yi et al., 2017; Yildiz et al., 2015; Yilmaz et al., 2015). It is recommended that oleogels have a high fat binding capacity in order to prevent the release of fat, which causes quality losses in many products such as chocolate, spreadable breakfast products, soft candies and halva (Patel et al., 2014).

SFC is an important parameter for the quality and technological properties of the final product in oils used in chocolate and bakery products. The SFC values of OB ranged from 1.00% to 6.30% at 30 °C and from 0.80% to 5.00% at 35 °C. On the other hand, the SFC values of OR oleogels varied between 4.40% and 8.10% at 30 °C, and between 4.10% and 7.70% at 35 °C (Table 2). In both oleogels, the SFC increased as the wax content increased (p<0.05). SFC was slightly lower at 35°C than at 30°C as a matter of course. It was noteworthy that the SFC ratios were higher in OR oleogels. It was noted that SFC was lower in oleogels compared to shortening. However, it is well known that oleogelation does not increase SFC in oils (Co and Marangoni, 2012). Grape seed oil based oleogels containing 5%, 10% and 15% beeswax had SFCs of 0.54, 1.82, and 3.78%, respectively, at 40°C (Yi et al., 2017). Öğütçü et al. (2015) found SFC ratios at 35°C to be 2, 6 and 8 %, respectively, in oleogels obtained from cod liver oil containing 3%, 7% and 10% beeswax. Our findings were mostly consistent with these data.

The CT was changed from 3 to 8 minutes in ORs and from 7 to 11 minutes in OBs (Table 2). There was no significant difference in CT at 7% and 10% concentrations of OR oleogel. It was determined that the CT decreased as the oleogel concentrations increased. KTs were shorter in ORs than in OBs. In other words, it was determined that rice bran wax formed crystals in a shorter time than beeswax. Yilmaz and Öğütçü (2014) determined the CT's for olive oil-based oleogels containing 3%, 7% and 10% beeswax to be 10.0, 7.0, and 6.0 min, and for those containing sunflower wax at the same concentrations to 5.0, 3.5, and 4.0 min, respectively. Dassanayake et al. (2009) determined the CT between 4.0-11.0, 9.0-14.0 and 12-14 minutes in olive oil-based oleogels prepared with rice bran wax, candelilla wax and carnauba wax at concentrations between 0.2-4%, respectively. In virgin olive oil-based oleogels containing 7% and 10% carnauba wax, 3%, 7% and 10% monoglyceride, CT was reported as 10.5, 7.50, 23.5, 8.00 and 7.50 min, respectively (Öǧütcü and Yılmaz, 2014). Our results were largely consistent with these data. The CT can give information about the gelling ability of the gelator molecule.

Hunter color values of oleogels are given in Table 2. L\* values expressing lightness-darkness in oleogels were found in the range of 35.61-50.76 (p<0.05). The L value of the shortening was found to be higher than the oleogels (81.12). In general, the L value also increased due to the increase in gelator concentration. In the study conducted by Yilmaz and Öğütçü (2014), L values of beeswax-hazelnut oil oleogels were found to be between 39.03 and 56.01, while L values of sunflower wax-hazelnut oleogels were found to be between 48.70 and 65.78. When the a\* values are examined, it is seen that all samples are on the red side of the scale and vary between 1.94-6.46 (p<0.05). A change in a\* value was found inversely proportional to the gelator concentration. In addition, the a\* value was higher than the control (S) at high concentrations in both oleogels. While a\* values were found in the range of -5.60 to -6.37 in oleogels containing virgin olive oil-based carnauba wax, it was determined in the range of -3.48 to -5.98 in oleogels containing monoglycerides (Öǧütcü and Yılmaz, 2014). In the same study, b\* values were determined in the range of 16.48 to 25.22 in those containing virgin olive oil-based carnauba wax, and in the range of 10.43 to 16.06 in those containing monoglyceride. The reason why a\* values differed from our values on the negative side may be due to the fact that it is based on virgin olive oil. The b\* value, indicating blue-yellowness, was on the yellow side, and b\* increased as gelator concentration increased. While b\* values were not affected by wax types, a\* values were partially affected. In both oleogels, b\* values were higher than shortening at high concentrations.

**Some physical properties of cakes**

The pH, moisture content, specific volume, specific gravity and color values of the cakes using oleogels are given in Table 3. In cakes, the highest pH was determined in CS, the control sample, and the lowest in COB3. Except for the cakes with these extreme values, there was no significant difference between the pH values.

The moisture content of the cakes was 19.68% in the cake produced with shortening and between 22.69-28.12% in the cakes produced with oleogels, and there was no statistically significant difference between them. This shows that there is a similarity between oleogels and shortening in terms of water holding capacity. Giacomozzi et al. (2018) determined moisture content in the range of 20.72% to 22.49% in the muffins they made with sunflower oil-based monoglycerol containing oleogels. These data are consistent with our values. Yılmaz and Öğütçü (2015) determined the moisture content of the cookies they used oleogels based on hazelnut oil, formed with 5% beeswax and 5% sunflower wax, as 2.63% and 2.32%, respectively. In commercial shortening, which is the control sample, they reported the moisture content as 5.72%. However, it is expected that the moisture contents of cookies and cakes are different.

The specific volume was determined as 0.61 in the cake produced with shortening, in the range of 0.52 to 0.56 in those produced with ORs, and in the range of 0.50 to 0.51 in those produced with OBs (Table 3). There was no significant difference between the specific volumes of OBs and the cakes produced. The difference between the specific volumes of the cakes produced with the ORs was statistically significant (p<0.05). Lim et al. (2017) found specific volumes in the range of 1.53 to 1.87 mL g-1 in muffins in which oleogels were used at different rates. The low specific volume of our cakes can be attributed to the low air holding capacity of the oleogels.

The specific gravity of the cake batter produced varied between 0.87-1.24 g mL-1. The lowest specific gravity was determined in dough made with shortening (0.87 g mL-1). In oleogels, this value varied between 1.18-1.24 g mL-1. However, the difference between the specific gravity was not statistically significant (p>0.05). Lower specific gravity is required for larger final cake volume and height. Willett and Akoh (2019) found specific gravity in the range of 0.72-0.97 in cake batters made with different fat fractions. Specific gravity in our cake batters was higher than those findings. This is likely due to the inability of these oleogels to stabilize and retain air bubbles in the dough during mixing. In another study, the specific gravity of muffins using hydroxypropyl methylcellulose and oleogellin made with sunflower oil varied between 0.87-1.12 g mL-1 (Oh and Lee, 2018). Lim et al. (2017) determined specific weights in the range of 0.81-1.10 g mL-1 in muffin doughs they prepared with different ratios of shortening/oleogel. Our values were largely consistent with these findings.

The crust (external, ‘e’ subscript) and crumb (internal, ‘i’ subscript) color values of the cakes prepared with shortening and oleogels are presented in Table 3. In addition, the images of oleogels and the cakes produced from them can be seen in Fig 2. Li values expressing the lightness-darkness of the cake crumb were determined to be between 60.66-70.77 (p<0.05). The highest Li value was observed in the CS cake. This was followed by COB10 and COR10. The height of this value indicates that the color is light. There was no significant difference between COR5, COR7 and COB5 in their Li values. Li values tended to increase depending on the increase in oleogelator concentration. Cake crust Le values ranged from 37.86 to 51.68 (p<0.05). The Le value was highest COR5, lowest COR7 and COB10.

ae\* values ranged from 15.60 to 21.75 (p<0.05). The highest value was CS. This means that the CS cake is closer to red. There was no significant difference between COB3 and COB5 related to ae\* values. The ai\* values were found to be between 1.40 and 2.62. The highest ae\* value was observed in CS. CS was closer to red in crust and crumb compared to cakes containing oleogel.

be\* values varied between 18.41-35.16 and bi\* values varied between 13.90-19.51. The highest be\* value was determined in COR5 and bi\* value was determined in COB10. So, these were closer to the yellow color. The crust and clumb (be\* and bi\*) values closer to blue color in COR5 and COB10 cakes, respectively. Oh et al. (2017) found that the L values in cakes using oleogels containing rice bran wax, beeswax and candelilla wax were significantly higher than that produced by shortening. It has been stated that the cakes made with shortening as the crust color have a darker color. It was determined that b\* values in oleogel cakes were lower than the control. This means that the oleogel cakes are less yellow. Oh and Lee (2018) showed that the L values of the cakes made by mixing HPMC oleogel with different ratios of shortening decreased significantly as the HPMC oleogel concentration in the crust increased. While a\* values increased in the crust of oleogel cakes, b\* values also increased inside and outside the cake. Giacomozzi et al. (2018) determined the L, a and b values in muffins made with commercial margarine, high oleic sunflower oil and different oleogels in the range of 48.39-50.40, 6.13-9.66 and 30.56-32.14, respectively. L, a\* and b\* values were determined in the ranges of 66.57-72.25, 5.89-8.31 and 21.20-25.87, respectively, in cakes prepared with oleogels containing carnauba wax and shortening (Pehlivanoglu et al., 2018). Color values are affected by many factors such as cake formulation, oil or substitute, and baking conditions. The important thing here is consumer acceptability of the product. By looking from this point of view, it has been revealed that cakes containing oleogel with high unsaturated fat content can be used in the bakery industry with similar color characteristics to cakes containing shortening.

**Textural properties of cakes**

The textural properties of the cakes such as hardness, adhesiveness, springiness, cohesiveness and chewiness are exhibited in Table 4. Hardness is known as the maximum force at initial compression. The cakes exhibited hardness values in the range of 790.3 g to 1098.4 g. There was no significant difference between COB10 and CS in their hardness values. This is probably due to the COB10 cake having higher volume and more air incorporation. COR5, COR7 and COB5 hardness values were also close to each other. It was noteworthy that as the wax ratio increased, the hardness decreased. Willet and Akoh (2019) found hardness values in the range of 5 to 8 N (509.8 to 815.7 g) in cakes using different oleogels. The hardness values in muffins using different ratios of shortening and HPMC oleogels were between 10.78 and 18.71 N (1099.2 and 1907.8 g) (Oh and Lee, 2018). In another study, Oh et al. (2017) determined the hardness values of cakes prepared with beeswax, rice bran wax and candelilla wax as 8.14, 11.38 and 11.25 N (830.0, 1160.4 and 1147.1 g), respectively. Alvarez-Ramirez et al. (2020) reported that total butter substitution with oleogel reduced cake hardness from 29.61 to 13.37 N (3019.3 to 1363.3 g). It was determined by Pehlivanoglu et al. (2018) that the hardness values in cakes prepared with different oleogels ranged from 2.34 to 7.62 N (238.6 to 777.0 g).

Adhesiveness is defined as the work required to overcome the attractive force between the food and the outer surface. This value is the negative force field during the first compression (Antoniou et al., 2000). Adhesiveness values varied from 0.342 to 0.687 in cakes containing OB and from 0.337 to 1.213 in cakes containing OR (p<0.05). In CS (control), this value was higher (1.708). As the OR content increased, the adhesiveness value increased. COB cakes exhibited lower adhesiveness than others and control. On the other hand, COR5, COB3 and COB10 were close to each other and had the lowest adhesiveness value. Adhesiveness values in cakes prepared by replacing candelilla wax/canola oil oleogel/butter mixtures with oleogel and butter at 0, 25, 50, 75 and 100% were determined in the range of 1.81 to 6.67 (Alvarez-Ramirez et al., 2020).

The springiness was 0.779 mm in CS, 0.643 mm in COB10, and 0.902 to 0.921 mm in other cakes (p<0.05). Cakes other than COB10 had higher springiness values than CS (control). Springiness is the cake's ability to regain its height after compression and is related to the number of air bubbles. Oh and Lee (2018) reported springing values from 0.77 to 0.88 mm in muffins using different proportions of fat and HPMC oleogels. Pehlivanoglu et al. (2018) found the springiness values in the range of 0.92-1.14 mm in the cakes they prepared with different ratios of oleogel.

Cohesiveness is defined as the degree of deformation of the sample before it breaks in the mouth, which is the ratio of the positive force area during the second compression to that of the first compression. It also indicates the ability of the product to hold together (Cierach et al., 2009).Cohesiveness values of the cakes ranged from 0.614 to 0.701. However, these variations were not statistically significant. The cohesiveness values of the cakes using different ratios of oil and HPMC oleogels were found to be between 0.56 and 0.64 (Oh and Lee, 2018). Cohesiveness values in cakes prepared with rice bran wax, beeswax and candelilla wax were reported as 0.68, 0.64 and 0.67, respectively (Oh et al., 2017). Pehlivanoglu et al. (2018) found the cohesiveness values in the range of 0.76-0.79 in the cakes they prepared with different ratios of oleogels.

Scales of chewiness value of the cake samples were varied between 372.6 and 664.8 g. Chewiness values decreased as OB ratios increased (p<0.05). There was no significant difference between COR7 and COB5, COB10 with CS, and between COR5 and COR10. Pehlivanoglu et al. (2018) determined the values in the range of 216.1 to 407.8 g in cakes prepared with different ratios of oleogel. On the other hand, Willet and Akoh (2019) found the values in the range of 407.8 to 611.8 g.

**Some physicochemical properties of the shortening and oleogels**

Fatty acid composition, PV, FFA, K232, K270, 3-MCPD and glycidyl values of shortening and oleogels before and after cooking are displayed in Table 5. Safflower oil contains approximately 89.4% unsaturated fatty acids (UFA) and 10.6% saturated fatty acids (SFA). Safflower oil is one of the richest oils in linoleic acid, which is among the essential fatty acids and is important for the nutritional value of oils (Peker and Baştürk, 2019). Polyunsaturated fatty acids (PUFA) have been reported to support the human immune system against viruses such as Sars-COv-2 (Baştürk et al., 2021). On the other hand, shortening contains 46.7% UFA and 51.3% SFA. The aim of this study was to use fats with high UFA content (as oleogel) as a shortening substitute with high SFA content in cakes. However, since oils with high UFA content are particularly sensitive to thermal oxidation, some deterioration parameters should be taken into account. For this, these parameters were compared before (pre-cooking) and after cake cooking (post-cooking). In general, no significant changes were observed in fatty acids except linolenic acid (p<0.05) in oleogels during post-cooking. Interestingly, the linolenic acid content increased at post-cooking (p<0.05). While there was a decrease (p<0.05) in stearic acid content at post-cooking, there was no significant change in other fatty acids in shortening. After conversion to oleogel, there was no significant change in the ratio of dominant fatty acids in safflower oil. The predominant linoleic acid in the oleogels varied between 67.34% and 71.56%. Although this rate decreased on a small-scale post-cooking, these variations were insignificant (p>0.05). Oleic acid content decreased from 18.67% to 16.70%. While this ratio increased at post-cooking in ORs, it decreased in OBs. These changes were not significant. The change in palmitic acid ratio in oleogels was also insignificant. Palmitic acid (37.11-37.81%), oleic acid (32.05-31.69%) and linoleic acid (14.09-15.37%) contents changed in the shortening before and after cooking (p<0.05). The changes in SFA, MUFA, PUFA, UFA, PUFA/SFA and UFA/SFA indices were not significant before and after cooking in oleogels (p>0.05). These values were different from shortening. PUFA/SFA ratio is a good indicator of nutritional values of dietary oils. This ratio ranged from 5.97 to 6.71 in SO and oleogels. These variations were not significant. In shortening, this rate was 0.28 and 0.31 at pre-cooking and post-cooking, respectively. While UFA/SFA ratios were 0.91-0.93 in shortening, this ratio ranged from 7.44 to 8.39 in oleogels. Javidipour et al. (2015) found the UFA/SFA ratio in refined cottonseed oil and extra virgin olive oil in the range of 2.66–2.75 and 5.47–5.67, respectively. In short, it can be said that cake baking did not have a significant effect on the fatty acid composition of oleogels. Jang et al. (2015) reported that there was no significant change at post-cooking fatty acid profiles in cookies made with oleogels prepared using canola oil and candelilla wax at different concentrations.

PV was initially 3.76 meqO2 kg-1 in safflower oil and 27.7 meqO2 kg-1 in shortening. It is thought that the possible reason for the high rate of shortening may be unsuitable storage conditions in the market. This value was decreased sharply to 4.8 meqO2 kg-1 with post-cooking shortening. This can be explained by the breakdown of hydroperoxides into secondary oxidation products. The PV ranged from 13.21 to 20.45 meqO2 kg-1 in the rice bran wax-containing oleogels and from 4.3 to 7.72 meqO2 kg-1 in the beeswax-containing oleogels. The higher PV in ORs can be explained by the high polyunsaturated fatty acid content of rice bran waxes. The PV change in OR5 and OR7 with the cooking process was insignificant. However, this value decreased in OR10. At post-cooking, PV decreased in OB3 (p<0.05), increased in OB5 (p<0.05), but did not change significantly in OB10. The reason why this value is higher in oleogels compared to the PV of safflower oil at the beginning may be the temperature applied during the preparation of the oleogel. Bozkurt and Baştürk (2018) determined the highest PV in margarines stored at 4 and 25 °C as 11.08 and 70.97 meqO2 kg-1, subsequently. Willett and Akoh (2019) determined PV in the range of 4.50 to 43.63 meqO2 kg-1 and 14.36 to 68.25 meqO2 kg-1 at 0 and 7 days, respectively, in the cakes they made with oleogels, which were obtained from β-sitosterol/γ-oryzanol and sucrose stearate/ascorbyl palmitate mixtures as oleogelators and different lipid phases.

FFA showed an increase after cooking in samples except OB3 and OB5 samples. The increase of FFA was more in ORs compared to OBs. The FFA in the samples ranged from 0.04 to 0.22%. The highest increase was in S, followed by OR5. In all samples, the FFA was at the acceptable level specified in the codex. Yılmaz and Ögütçü (2015) found FFA in the hazelnut oil and virgin olive oil oleogels in the range of 0.57 to 1.47%. K232 was initially 1.35 in safflower oil and 5.27 in shortening. In oleogels, K232 was in the range of 2.91-5.39 in ORs and 1.73-2.34 in OBs before cooking. K232 increased to 5.61-5.71 in ORs and 4.37-6.12 in OBs after cooking. In S, this value decreased slightly after cooking. This was the case for PV as well. Because primary oxidation products may be broken down and secondary oxidation products may have started to form. K270 was not detected in samples other than S and OR10 pre-cooking. However, it was detected in all samples after cooking. K270 determined in oleogels after cooking was lower than that determined in shortening. It was determined in the range of 1.78-1.97 in ORs and in the range of 1.12-1.69 in OBs. The highest rate was determined as 2.46 in shortening.

3-MCPD and glycidyl analyzes were performed on oleogels and shortening before and after cake cooking. 3-MCPD was determined to be 1.89 in S, range from 0.93 to 1.06 mg kg-1 in ORs, and range from 0.37 to 0.52 mg kg-1 in OBs. The fact that safflower oil-based oleogels have lower 3-MCPD values than shortenings can be explained by the fact that safflower oil is unrefined. In addition, it is thought that the sugar used in cake making may trigger the formation of 3-MCPD. At least 3-MCPD occurred in the beeswax oleogels. The effect of gelator concentration on 3-MCPD formation was not significant. It did not occur in glycidyl S and OB3. It was detected in the range of 0.32 to 0.81 mg kg-1 in ORs, and 0.27 and 1.01 mg kg-1 in OB5 and OB10, respectively. In contrast to 3-MCPD, the glycidyl content increased as the oleogelator concentration increased. In our previous study (Gündüz et al., 2023), we determined the 3-MCPD and glycidyl values of three different shortening brands that we obtained from the market in Türkiye as 0.12, 0.68, 0.32 mg/kg and 1.98, 6.46, 2.65 mg kg-1, respectively. 3-MCPD concentrations were found in the range of 0.57 to 4.54 mg kg-1 in margarines obtained from the market in Türkiye (Deniz Şirinyıldız, 2019). 3-MCPD concentrations in margarine and shortenings in the Netherlands have been reported as 0.16 to 1.8 mg kg-1 (Boon and te Biesebeek, 2016). 3-MCPD and glycidyl concentrations in hazelnut oil were determined as 1.476 and 0.747 mg kg-1, respectively (Ceylan and Baştürk, 2022). To the best of our knowledge, no study on 3-MCPD and glycidyl concentrations in oleogels in the literature.

**Sensory evaluation**

Sensory evaluation of cake samples is given with radar plot and bar graph (Fig 1). Features such as appearance, pore structure, moisture, taste and aroma, mouthfulness and general acceptance were rated out of 5 points by the panelists. In the appearance of the cakes, COB3 samples showed the highest (4.20) and CS showed the lowest values (3.20). No significant change was observed in appearance scores (3.53-3.87) in cakes other than these (Fig 1/A, G). There was no significant difference between the scores given by the panelists in terms of pore structure in the cake samples. Scores ranged from 3.13 to 3.67 (p>0.05). The humidity sensation scores of the cakes ranged from 3.07 to 3.78 (p<0.05). There was no significant difference between the mean values of the cakes except for the cakes with the highest (3.78) and lowest (3.07) humidity scores. Humidity sensation scores in the cakes of COBs increased as the oleogel concentration increased. In terms of taste-aroma, while the most liked one was COB3 (3.67), the least liked was COR10 (2.80). The scores of the others were close to each other (p>0.05). Cakes made from beeswax oleogels were close to control in terms of mouth fulness. Those containing rice bran wax showed lower scores compared to those containing beeswax (p<0.05). In overall acceptability, while the highest scores were observed for COB10 (3.87) and COB3 (3.80) cakes, the lowest scores were determined for COR5 (3.07) and COR10 (2.87) cakes. In terms of general acceptability, cakes made from beeswax oleogels were more appreciated. There was no significant difference between COR7 (3.33), COB5 (3.53) and CS (3.53) scores (Fig 1/F).

**CONCLUSION**

Solid fats, as an important component of bakery products, play an important role in extending the shelf life, swelling of the products and providing the desired taste and texture. However, because of the high amount of saturated fat used in bakery products, they cause many diseases such as cardiovascular diseases, diabetes, obesity, increased insulin resistance. Currently, there is an increasing interest to use oleogels with high unsaturated fat content and no trans fatty acids. In this study, cold-pressed safflower oil-based oleogels were obtained by using different ratios of beeswax and rice bran wax. It was used to make oleogels and commercial shortening cakes. Then, selected quality analyzes were done for oleogel and cakes and compared with commercial shortening. Some physical, textural and sensory properties of the cakes using oleogels were acceptable when compared to the cakes using shortening. In fact, the overall acceptability of the cakes containing beeswax was higher than the control. In addition, the deterioration parameters (Fatty acid composition, PV, FFA, K232, K270, 3-MCPD and glycidyl values) of oleogels and shortening used in cakes pre- and post-cooking were compared. In general, there was no significant change was determined in fatty acid composition following cooking process. In oleogels and shortening, 3-MCPD was formed post-cooking. This value was determined less for oleogels compare to shortening. Glycidyl formed in oleogels (except OB3) post-cooking. As the oleogelator concentration increased, the glycidyl increased. The results obtained with the study showed that oleogels can be used instead of shortening in cakes. Thus, preferable cakes by consumers with low saturated fat content can be produced.

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**AUTHORSHIP CONTRIBUTIONS**

Ş.B. designed the study, performed the experiments, analyzed the data, wrote the first draft of the manuscript, A.B. designed and supervised the study, analyzed the data, carried out the research. All authors contributed to reviewing and editing the manuscript and approved the final draft of the manuscript.

**CONFLICT OF INTEREST**

The authors declare that they have no conflict of interest.

**ETHICS STATEMENT**

This article does not contain any studies with human participants or animals performed by any of the authors

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**Figure Legends**

**Fig. 1 Sensory scores of cake samples in a radar plot and bar graph**

**Fig. 2 The imaging appearances of oleogels and cakes**

**Tables**

**Table 1. Oleogel and cake codes**

**Table 2. Some physical properties of oleogels**

**Table 3. Some physical properties of cakes**

**Table 4. Textural properties of cakes**

**Table 6.** **Some physico-chemical properties of the shortening and oleogels before and after cooking**