

7. Appendix

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How fat quantity and composition affect plum cake characteristics:
a response surface methodology.

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Running Head: Optimizing fat quantity and quality in cake

Abstract: Few studies have been addressed to investigate how the fat quantity and composition affect the characteristics of a sweet chemically leavened baked product. In this study, these aspects were modelled and optimized by means of an experimental design. Fats were blends of palm oil and palm olein with different slip melting points (from 36.3 °C to 11.3 °C, in dependence of increasing olein content). Fat content (from 5.3% to 30.7% on batter) and percentage of olein in the fat blend (from 42.7% to 92.2%) were varied. A plum cake containing 18% butter was the reference. Fat quantity mainly affected cake consistency and volume; olein content only influenced cakes texture. An optimized formulation containing 19.7% fat with 92% olein in the fat blend was identified, demonstrating the possibility to replace butter or shortening with a vegetable fat blend of low melting point, thus increasing unsaturated fat content and reducing total cholesterol in the product.

Keywords: cake; palm oil; palm olein; butter; fat blend; melting point; texture, experimental design; response surface methodology; desirability function.
The Technological Role of Fats in a Baked Product

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The aim of this PhD thesis is to study the technological role of fats in bakery products; plum cake was chosen as model system. After having modelled the effects of quantity and type of fat on cakes, an optimized formulation (19.7% fat in the batter, made up of 92% palm olein and 8% palm oil) was compared with references containing butter and anhydrous butter. Rheology of batters and fats was studied, as well as cake performances and shelf life. Preliminary results on organogels rheological characterization and their baking properties are shown.

Ruolo Tecnologico dei Grassi in un Prodotto da Forno

L'obiettivo di questo progetto di dottorato è lo studio del ruolo tecnologico dei grassi nei prodotti da forno tipo plum cake. In questa fase, due formulazioni con burro e burro anidro sono state confrontate con una formulazione ottimizzata (19.7% grasso nell’impasto, composto da 92% oleina di palma e 8% olio di palma), ottenuta modellando gli effetti di quantità e tipo di grasso. Sono state valutate le proprietà reologiche dei tre grassi impiegati e dei rispettivi impasti, le caratteristiche del prodotto finito e la sua conservabilità. Sono inoltre riportati dati preliminari riguardanti la caratterizzazione reologica di organogel e la loro applicazione nel cake.

Keywords: fat, cake, rheology, batter, storage.

1. Introduction

Vegetable fats are commonly used in bakery industry, as cheaper and zero-cholesterol alternatives to butter; their crystallization and melting properties can influence quality and structure of the baked product. Among vegetable fats, palm oil is one of the most common fats used in the food industry, especially in confectionery and bakery products, because of its low cost, stability to oxidation and versatility. At room temperature, it is a semi-solid material (melting point between 33 and 41°C). It can be separated in different fractions with different melting points: palm stearin, a high melting fraction mainly composed of trisaturated triacylglycerols (TAG), and palm olein, a low melting fraction mainly composed of monosaturated TAG (Tarabukina et al., 2009).

The characteristics of a baked cake, such as volume and texture, can be correlated with the rheological properties of the batter (Sakiyan et al., 2004). In particular, inclusion of air - a key aspect of batter preparation - not only determines the structure of the baked product but also affects batter rheology, which governs the expansion of the material before the continuous phase sets (Chesterton et al., 2011). In fact, changes in batter viscosity can be correlated with variations in cake volume, as an increase in batter viscosity can aid air incorporation and enhance air bubbles retention during the first stages of baking (Gomez et al., 2008). Fats play an important role in this, as they contribute to the incorporation of air into the batter and enhance
heat transfer during cooking, bringing to finished products characterized by a moist mouthfeel and a softer texture (Conforti, 2006). The evaluation of cake quality covers not only the fresh product, but also its behaviour throughout storage. Smith and Johansson (2004) observed that fat melting characteristics affect texture and volume of bread, as an increase in solid fat leads to a softening of bread and a decrease in the rate of staling, suggesting an interaction between saturated TAG and amylopectin. Thus, raising the concentration of solid fat could improve bread quality, although increasing saturated fat content. Nowadays, unsaturated fats are preferred for their nutritional properties, even though they are more susceptible to oxidation. In fact, lipid oxidation reactions occurring during the storage of bakery products are the main deteriorative event affecting their quality (Calligaris et al., 2007). Römer et al. (2008) reported that oxidised lipids could even react with protein by hydroperoxides or secondary lipid peroxidation products, bringing to a nutritional loss in the product. In order to evaluate the degree of oxidation, peroxide value (PV) is commonly used; Calligaris et al. (2007) considered it as a representative index of the quality depletion of biscuits during their shelf life, linearly related to consumer acceptability. In this phase of the PhD research project, three plum cake formulations containing different types of fat were studied: butter, anhydrous butter and a fat blend composed of 92% palm olein and 8% palm oil, formulated according to the results of the optimization process carried out in the first phase of this PhD. Fats and batters rheology was studied, as well as baked product characteristics and shelf life.

2. Materials and Methods

2.1 Cakes preparation

Fats used were butter (B), anhydrous butter (AB, prepared from butter as reported in Ferrari, 2010) and a blend of 92% bi-fractionated palm olein and 8% palm oil (O), prepared as previously reported (Ferrari, 2010). B and AB cakes had 18% butter and anhydrous butter, respectively, while O cake had 19.7% fat blend; all percentages are expressed on batter (w/w).

2.2 Analytical methods

Official method Cc 3-25 (AOCS, 2009) was used to measure fat slip melting point (SMP, °C). Solid fat content (SFC, %) of fats was measured following the Cd 16b-93 official method (AOCS, 2009), by using a Minispec NMS 120 (Bruker BioSpin MRI GmbH, Ettlingen, D). Before SFC analysis, butter was melted and filtered on a filter paper containing anhydrous sodium sulphate to remove water. Apparent specific gravity of batters was measured by weighing a standard cup (n=10) filled with batter or with water and dividing the weights (Dogan et al., 2007). Plum cakes were analyzed for moisture content (whole slice and crumb) and specific volume, as reported by Mariotti et al. (2006). Texture analyses (fresh and stored samples) were performed using an Instron Universal Testing Machine 3365 (Instron Division of ITW Test and Measurement Italia S.r.l., Trezzano sul Naviglio, I) equipped with a 100 N load cell and supported by BlueHill software (BlueHill2, v2.9, 2005, Instron Corp., UK). Two cakes were evenly sliced obtaining four 25±1 mm thick slices for each cake. Each slice was penetrated with a 27 mm diameter plate. Analyses were carried out at room temperature, obtaining Young modulus (slope of the stress-strain curve of penetration tests) and load at 25% strain (index of sample hardness). Regarding shelf life tests, cakes were packed and sealed in OPP bags (previously sprayed with ethanol, in order to prevent microbial growth). Packed cakes were stored at 25°C for 35 days; PV and texture analysis were performed on 0 - 2 - 5 - 7 -10 -14 - 21 - 28 - 35 days from production.
PV was measured following the EU official method (Reg. CE 2568/91, 1991). Fat was extracted by milling two slices of cake using a rotating blade mixer; 12 g of the milled cake (in duplicate) were weighted, 100 mL chloroform was added and the suspension was stirred for 45 min. The extract was then filtered with Whatman filter paper n.4 (Whatman Int. Ltd, UK) and subjected to PV analysis.

2.3 Rheological determinations
Fat and batter rheological behaviour was evaluated using a Physica MCR 300 rheometer (Anton Paar, Graz, A), supported by Rheoplus/32 software (v3.00, Physica Messtechnik GmbH, Ostfildern, D), and equipped with a plate-plate geometry (d=25 mm, rough surface for batter analyses; d=50 mm, plain surface for fat analyses), gapped to 1 mm. All tests were run at least in duplicate.

For fats, temperature sweep (TS) tests simulating the temperature profile of crystallization (70°C to 0°C for O, 70°C to 15°C for AB; cooling rate, 2.5°C/min) were performed applying a strain (\(\gamma\)) of 0.1% and an oscillation frequency (f) of 1 Hz. Frequency sweep (FS) tests were performed at 25°C, with \(\gamma=0.1\%\) and \(f=0.01-100\) Hz. These test conditions were within the linear viscoelastic range, previously determined by strain sweep (SS) tests carried out at 25°C, with \(f=1\) Hz and \(\gamma=0.01-100\%\).

For batters, TS tests (25°C to 70°C; heating rate, 2.5°C/min) were performed with \(\gamma=0.01\%\) and \(f=1\) Hz. FS tests were performed at 25°C and 60°C, with \(\gamma=0.01\%\) and \(f=0.01-100\) Hz. The test conditions were within the linear viscoelastic range, previously determined by SS tests performed at both 25°C and 60°C, with \(f=1\) Hz and \(\gamma=0.001-100\%\). The exposed edges of the sample were covered with a thin layer of paraffin oil to prevent dehydration during measurements.

3. Results and Discussion
As reported in the previous part of the study (Ferrari, 2010), the effects of fat melting characteristics and fat quantity in plum cake formulations were studied and modelled through the Design of Experiment (DoE) methodology. It resulted that cake texture was significantly affected (p<0.01) by both fat content and percentage of olein in the fat blend, while volume was influenced (p<0.01) only by fat content. In order to identify a cake formulation optimized for texture and volume, a desirability function was developed. Optimization was based on both minimization of load at 25% strain (between 5.5 and 10 N) and achievement of a targeted specific volume of 2.23 cm³/g, resembling the one obtained with B recipe. The desirability function corresponded to an optimized formulation (O) containing 19.7% fat in the batter and 92% olein in the fat blend. Validation values were fairly consistent with those predicted, showing a full correspondence for specific volume (2.25±0.04 cm³/g vs. 2.23 cm³/g predicted) and a minor discrepancy for load (7.68±0.27 N vs. 7.09 N predicted).

3.1 Cake characteristics
The three formulations (B, AB, O) showed different performances (Table 1), being B significantly (p<0.05) softer and moister than the other two samples, which showed similar texture and water content, both in slice and crumb. This difference is probably related to the lower batter moisture of AB and O (B=25.7%, AB=22.5%, O=21.7%), since butter, containing 18% water, adds around 3% moisture to batter. O and B samples showed the same specific volume, thus the slightly higher fat content in O seemed not to affect negatively the cake volume. This result highlights the role of fat slip melting point (SMP) on cake specific volume, SMP of O fat blend (11°C) being definitely lower than that of AB (33°C). This is in agreement
with the trend observed in DoE experiments for a positive (although not significant) effect of lower SMP on cake volume.

Table 1 Values of selected responses for B, AB and O formulations. Numbers with different letters within the same column are statistically different (p < 0.05).

<table>
<thead>
<tr>
<th>Cake</th>
<th>SMP (°C)</th>
<th>Slice Moisture (%)</th>
<th>Crumb Moisture (%)</th>
<th>Specific Volume (cm³/g)</th>
<th>Young Modulus (kPa)</th>
<th>Load at 25% Strain (N)</th>
</tr>
</thead>
<tbody>
<tr>
<td>B</td>
<td>32.8</td>
<td>20.33±0.07 a</td>
<td>23.97±0.04 a</td>
<td>2.23±0.03 a</td>
<td>54.08±0.70 a</td>
<td>5.63±0.03 a</td>
</tr>
<tr>
<td>AB</td>
<td>33.0</td>
<td>17.54±0.03 b</td>
<td>20.79±0.14 b</td>
<td>2.01±0.04 b</td>
<td>80.51±4.37 b</td>
<td>7.86±0.91 b</td>
</tr>
<tr>
<td>O</td>
<td>11.3</td>
<td>17.47±0.31 b</td>
<td>20.63±0.02 b</td>
<td>2.25±0.04 a</td>
<td>83.61±2.91 b</td>
<td>7.68±0.27 b</td>
</tr>
</tbody>
</table>

O was compared with B and AB recipes also in terms of apparent specific gravity (SG) of batters. This parameter is strictly related to the creaming performance of fat: the lower the SG, the higher the air content in the batter (Dogan et al., 2007). B formulation showed the lowest SG (0.97±0.02) compared to those of O (1.19±0.02) and AB (1.19±0.03). The better creaming performance of B is probably related with its physical state, i.e. a water in oil emulsion, which helped air incorporation and air bubble stabilisation, thanks to the naturally present emulsifiers, represented by the residue of the milk fat globule membranes. Furthermore, the creaming performances seemed not to be affected by SMP, as indicated by the similar values of SG found for AB and O.

To further investigate this point, Fig. 1 presents SFC values of the different fats used (B and AB have the same profile because B is dehydrated before the analysis).

Considering a working temperature of 20°C, it is possible to observe SFC values of about 25% for palm oil, 19% for B/AB and 1% for O, while olein is liquid at T>10°C. Consequently, our findings suggest that in a plum cake formulation a well aerated structure can be achieved with a minimal solid fat content, directly in contrast with previous results that indicated a minimum content of 5% crystalline fat at the working temperature for cake production (Podmore, 2002).

3.3 Rheological determinations
Considering the differences noticed among the three formulations, fats and batters were examined for their rheological behaviour.

Observing the TS curves of AB and O fats (Fig. 2A), an expected and similar slow increasing of moduli with the decrease of temperature can be noticed. At temperatures around 45°C, a first
crossover can be observed for both samples, with $G''$ exceeding $G'$. A second clear crossover of $G'$ and $G''$, indicating the initial solidification of the fat, was observed around 21°C for AB only. In the FS tests, carried out at 25°C, the three fat samples showed $G'$ values higher than $G''$; only for O a clear dependence of $G'$ and $G''$ from frequency was observed, with both moduli increasing with the increasing of frequency (Fig. 2B). $G'$ and $G''$ values of O blend were substantially lower than the values of B and AB, due to its lowest SMP.

Regarding batter rheology, the mechanical spectra of the three samples, measured at 25°C (Fig. 2C) and at 60°C (data not shown), presented a similar trend, but with definitely lower values for O recipe. All the batters showed a solid-like behaviour, with the elastic modulus ($G'$) higher than the viscous modulus ($G''$). To simulate structural changes in cakes during the first stages of baking, TS tests from 25 to 70°C were done. Unfortunately, it was not possible to study temperature values higher than 70°C, thus the real structure modifications due to cooking were not observed. A steep decrease of the complex viscosity (Fig. 2D) with the increase in temperature was observed for B and AB batters, mainly due to the fat melting. The viscosity decrease was, in fact, definitely less evident for O batter, in which the fat is already liquid at room temperature.

A clear influence of the fat type on batter structural characteristics has been demonstrated, even if rheological behaviour of batters did not always reflect differences evidenced among baked cakes. For instance, the highest $G'$ and complex viscosity values were found for B batter, which also resulted in more aerated batters (B had the lowest SG) and cakes with high specific volume and the lowest hardness. Thus, it seems that the more structured batter helped in the retention of air in cakes, confirming previous findings reported by Kalinga and Mishra (2009). Nevertheless, for the other two samples (AB and O) no clear relation between viscoelastic properties of batter and baked product characteristics could be found.
Figure 2  A: $G'$ and $G''$ in TS for AB and O (fats); B: $G'$ and $G''$ in FS for all samples (25°C, fats); C: $G'$ and $G''$ in FS for all samples (25°C, batters); D: Complex viscosity in TS for all samples (batters).
3.4 Shelf life study
The increasing trend of cake hardness during the 35 days of storage was similar for the three formulations; in particular, after 8 days of storage, load values almost tripled. The increasing in hardness can be partially explained by the variations in moisture, which decreased both in crumb (data not shown) and in the whole slice (Fig. 3). Moisture decrease showed a similar trend for O, B and AB, indicating that water loss is not influenced by the type of fat used. The different moisture values of the three formulations at the beginning of storage were due to a different moisture content in cake formulations. Observing the oxidation index (PV), O resulted more stable to oxidation (although starting from a higher PV); B and AB showed similar trends, with a steady increase in the first 2 days of storage. AB sample was characterized by the lowest values.

Figure 3 Cakes characteristics during storage at 25°C: hardness and slice moisture (left) and peroxide value (right).

4. Conclusion and Perspectives
In conclusion, it appears that the optimized cake formulation attained good structural properties, revealing in the meantime the best oxidation stability. Fat properties had an impact on batter viscoelastic behaviour, also affecting creaming performances. A higher content of unsaturated fatty acids, such as in the optimized formulation, resulted to be positive for baking performances, if adequately balanced in the formulation. On the basis of the results obtained so far, the research is going on studying the effects of the use of structured fats, i.e. organogels (OG), in the cake formulation. An OG can be defined as an organic liquid entrapped within a thermo-reversible, three-dimensional gel network. This gel network is formed by the self-assembly of a relatively low concentration of organogelator molecules, which are most often low molecular weight compounds that are capable of gelling organic solvents. OG can be formed from liquid organic solvents (like benzene, hexane, etc) or liquid oils, at organogelator concentrations as low as 0.5% wt depending upon chemical properties of the organogelator (Hughes et al., 2009). At this stage of the research, OG were prepared in sunflower oil, using β-sitosterol and γ-oryzanol (2:3, w/w) as organogelators. These molecules are phytosterols, known for having antioxidants properties and lowering blood cholesterol. Preliminary rheological evaluations were done on gels with different gelator concentrations (Fig. 4). These first trials showed that G’ of OG increased with increasing gelators concentration, as reported in the literature (Sawalha et al., 2011). Considering the results obtained, more speculation on the topic is required to understand if rheological differences in structured fats have an influence on batter rheology and
baking performances. Thus, the study of the influence of OG characteristics on the final product, through DoE methodology, is now in progress.

![Figure 4](image)

Figure 4 FS (γ=0.01%) for organogels at different concentrations.

5. References


EATING CAKES AND BEING HEALTHY: WILL THIS BE TRUE?

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Cereal products are at the basis of nutritional habits all over the world. Butter is traditionally used in bakery formulations, as it guarantees good performances in terms of volume, softness and sensory outcomes; on the other hand, it contains saturated fats and cholesterol, the two dietary ingredients that increase LDL cholesterol (commonly known as "bad cholesterol"). Therefore, finding alternatives to traditional animal fats for bakery products is an attractive challenge.

In a first phase of the research, the effects of vegetable fat melting characteristics (palm oil and palm olein blends) and fat quantity in plum cake formulations were studied and modelled by applying a Central Composite Design. Cakes produced with butter or anhydrous butter were used as references. The models obtained demonstrated that cake texture was significantly affected (p<0.01) both by the fat content and percentage of olein in the fat blend, while volume was influenced (p<0.01) only by the fat content. An optimized cake was obtained, corresponding to a formulation containing 19.7% fat in the batter and 92% olein in the fat blend, similar to the one obtained with butter for texture and volume characteristics, which are important quality indexes of a baked product. Furthermore, the comparison of the optimized formulation with reference cakes highlighted the role of fat slip melting point on cake volume, suggesting that a well aerated structure can be achieved even with a minimal solid fat content.

Fats and batters rheology was also studied, as well as cake characteristics and shelf life, comparing the formulations made with the optimized fat blend, butter and anhydrous butter. A clear influence of the fat type on batter structural characteristics was demonstrated, even if the rheological behaviour of batters did not always reflect differences observed among cakes. In particular, fat properties had an impact on batter viscoelastic behaviour, also affecting creaming performances. A higher content of unsaturated fatty acids, such as in the optimized formulation, resulted to be positive for baking performances, if adequately balanced in the formulation. In fact, the optimized cake attained good structural properties, revealing in the meantime the best oxidation stability, probably due to the antioxidant action of tocopherols and tocotrienols, naturally present in palm derived oils.

In a second phase, the effects of the use of structured fats, such as organogels (OG), in the cake formulation were studied. An OG can be defined as an organic liquid entrapped within a thermo-reversible, three-dimensional gel network, formed by the self-assembly of organogelator molecules. In this research, OG were prepared in sunflower oil, using as organogelators two phytosterols, β-sitosterol and γ-oryzanol (2:3, w/w), known for having antioxidant properties and lowering blood cholesterol. Preliminary rheological evaluations on gels with different gelator concentrations showed that $G'$ of OG increased with increasing gelator concentration. Considering the results obtained, more speculation on the topic is required; thus, the study on the influence of OG characteristics on baking performances is now in progress.
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MODELLING CAKE PERFORMANCE AS A FUNCTION OF FAT QUANTITY AND QUALITY
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INTRODUCTION

Fat is a key ingredient in cake formulations, as it contributes to textural, nutritional and sensory properties of the product. In the literature, impact of lipid on cake characteristics by a systematic approach has not yet been studied. The aim of this PhD thesis is to study the technological role of fats in bakery products, developing the possibility of enhancing healthy properties without compromising quality. The first part of the final product (Ferreri, 2010) in this first phase, the effects of the quantity and the quality of fat on cake performances were modelled and the optimisation was chosen. Furthermore, the relation between palm olein content and slip melting point of fat blends was studied.

MATERIALS AND METHODS

Palm oil is one of the most common fats used in industrial formulations. It must be refined before use and can also be fractionated, obtaining fats (stearin, olein, bi-fractionated olein) with different melting points.

Fat blends used in the experimental design were composed of different proportions of refined palm oil and bi-fractionated palm olein, in order to obtain fat mixtures with different physical characteristics, starting from the same raw material.

EXPERIMENTAL DESIGN

The experimental design chosen was a full factorial central composite design (CCD). The two factors considered were quantity of fat in the batter and percentage of olein in the fat blend.

RESULTS AND DISCUSSION

From the experimental design outcomes, it can be noticed that fat quantity had a more significant effect on cake performances than olein content in the fat blend. Both fat quantity and quality reduced cake consistency in particular, a higher fat quantity resulted in a softer cake but with lower specific volume, whereas olein content had no significant influence on cake volume. Image analysis and colour parameters were not significant for the design.

CONCLUSIONS

- A non linear dependence of slip melting point on olein content is found.
- The increase of fat content reduces the cake's softness without lowering its volume.
- Higher olein content in fat blends, corresponding to lower slip melting points, improves the cake texture.
- The optimisation of cake structural properties also leads to an improvement of nutritional properties (higher olein content in the fat blend, corresponding to lower slip melting points, improves the cake texture).

NEXT STEPS...
- Biological characteristics of fat blends and batter (in progress)
- Shelf life studies and texture modification; in proposed and sensory evaluation
- Use of organoleptic, as partial fat replacements

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14th Workshop on the Developments in the Italian PhD Research on Food Science, Technology and Biotechnology
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Study of the Technological and Functional Role of Fats in Bakery Products Using a Modellistic Approach

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**Aims of the PhD Thesis**
This thesis is going to study the technological role of fats in bakery products, developing the possibility of enhancing the healthy properties without compromising the quality and shelf life of the final product. Through the application of appropriate experimental design, mathematical models describing the optimization of product formulation will be calculated for different experimental hypothesis, including also the use of innovative ingredients.

**State of the Art**
Bakery products contribute to satisfy the nutritional needs of human beings. Fats are one of the key ingredients of this type of foods as they perform major functions to entrap air during creaming process, to interact with the continuity of starch and protein particles, and to emulsify the liquid parts of the formulation, contributing to volume, texture, and overall appearance of the baked products. However, from a nutritional point of view, the current presence of saturated fatty acids (SFA) and trans fatty acids (TFA) in bakery fats tends to raise the level of blood cholesterol and increases the risk of cardiovascular disease. Removing trans fatty acids and lowering saturated fat contents would improve the nutritional quality of bakery products but can lead to adverse consequences on the quality of the baked goods, as positive performances of fats in bakery are mainly a consequence of their melting characteristics that provide the desired solid fat content at processing temperature. Another issue concerning fats in bakery's shelf life of the final product. Fats in fact help delaying staling reactions, limiting moisture migration in the meantime, they are also subjected to degradation (i.e. oxidation) developing off-flavours and leading to nutritional losses.

**Aims and Expected Results**
In order to find a healthier alternative to traditional shortenings, without affecting technological properties and final product quality, this PhD thesis following topics will be studied:
- role of fat in traditional cake formulation and influence on the final product, through the application of an experimental design (CCD)
- role of high melting fats (HMF) with edible emulsifiers, a differentiated class of materials in which a network of self-assembled molecules immobilize an organic liquid core, forming thermally reversible gels upon cooling
- partial or total replacement of HMF with unsaturated oils through microencapsulation

During the whole project, product characteristics will be monitored by chemical, micro/nano structural and sensory analysis.

**3. Shelf Life Studies**
- Monitoring of the functional activity of emulsifiers through the analysis of water and oil partitioning of emulsions with different storage conditions
- Comparison of different storage conditions

**4. High Melting Fats**
- Importance of the use of high melting fats in bakery applications
- Improving the shelf life of bakery products through the use of HMF

**5. Microencapsulated Oils**
- Importance of the use of microencapsulated oils in bakery applications
- Improving the storage stability of bakery products through the use of microencapsulated oils

**References**