Glycerolysis for Reducing Saturated Fats in Ice Cream

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Although high in saturated fatty acids, tropical fats are commonly used in food industries despite their potential health risks. However, consumers are increasingly seeking sustainable, trans-fat-free, and "clean-label" food options. Accordingly, this Ph.D. project aims to decrease the saturated fatty acid content in fat ingredients through the enzymatic glycerolysis process. The obtained structured lipid was used as a fat source in a mixture for the production of ice cream. Particle size and ζ-potential of the mixture, together with physical properties during melting and textural attributes of the ice cream, were analyzed. Results pave an alternative way to design new fat ingredients with enhanced properties.

Glicerolisi per la Riduzione dei Grassi Saturi nel Gelato

Sebbene ricchi di acidi grassi saturi, i grassi tropicali sono comunemente usati nelle industrie alimentari nonostante i loro potenziali rischi per la salute. Ad oggi, i consumatori sono sempre più alla ricerca di prodotti alimentari, “trans-fat-free” e "clean-label". Questo progetto di dottorato si pone come obiettivo di ridurre il contenuto di acidi grassi saturi in prodotti alimentari, attraverso il processo di glicerolisi enzimatica. Il lipide strutturato ottenuto è stato utilizzato come fonte di grassi in una miscela per la produzione di gelato. Sono state analizzate la dimensione delle particelle e il ζ-potenziale della miscela, nonché le proprietà fisiche e strutturali del gelato. I risultati mostrati aprono un modo alternativo per progettare nuovi ingredienti lipidici con proprietà migliorate.

**Keywords**: Glycerolysis, structured lipids, low-saturated fat ice cream, high-melting fat.

# **1. Introduction**

Following the Ph.D. thesis project earlier described (Savchina, 2022), the poster reports the main results of the activities:

(A1) Lipid structuring through enzymatic glycerolysis. Where the production of structured fats and optimization of the process were performed.

(A3) Application of structured lipids in a food formulation. Structured fats were introduced into ice cream with a subsequent characterization; explicitly, particle size distribution, ζ-potential, stability while melting, and texture analysis were studied.

# **2. Materials and Methods**

Glycerolysis reactions were performed according to the optimized method described by Nickolson and Marangoni (2020). The main reaction components were peanut oil (10 g), glycerol (1.04 g), and lipase Novozym® 435 immobilized on the beads (0.40 g) were mixed together and placed onto the controlled temperature magnetic stirrer (IKA, USA) at 65°C and 200 rpm speed. After 16 h, the resulting substance was centrifuged (SL 16R Centrifuge, Thermo Scientific, Waltham, MA, USA) at 20 °C at 5,000 rpm for 5 min, and the supernatant was collected as structured lipid (SL). A cream-based ice cream (CB) was prepared by mixing: pasteurized milk cream (81.1 %), sucrose (14 %), and 0.2 % w/w lecithin and guar gum. Two other formulations were prepared by replacing milk cream with an oil-in-water emulsion based on peanut oil (POB) or structured peanut oil (SLB). Whey powder was added to adjust the milk-non-fat content to 5%. The liquid blends were mixed at 75 °C with a laboratory homogenizer (Digital UltraTurrax T25, IKA, USA) at 11,000 rpm for 2 minutes. The resulting blends were mixed with the dry ingredients, cooled down in an ice bath to 4 °C, and aged for 6 h. Aged ice cream premixes were frozen at –10 °C for 20 min at constant mixing in an ice cream maker (Unold 48816, Montereale Valcellina, Italy). Each batch was then sorted in plastic containers and kept at –25 °C for one week for the hardening stage.

The ice cream mixture particle size distribution was determined by a static light scattering technique using a Mastersizer Hydro 3000 (Malvern Instruments Ltd., Malvern, Worcestershire, UK). The sample was dispersed dropwise in deionized water until obscuration values of around 10 % were reached (refractive index of 1.52, absorption index of 0.01). To determine the ζ-potential, the measurements were conducted at a constant temperature of 25 °C after the ice cream blends were diluted (1:1000) with deionized water using a Malvern Zetasizer Nano ZS (ZEN 3600) instrument (Malvern Instruments Ltd, Malvern, Worcestershire, UK).

Turbiscan™ TOWER (Formulation Inc., France) was used to assess ice cream stability while melting. Ice creams were placed in cylindrical glass cells and stored at –18 °C for 24 hours and then placed into the tower at 25 °C for 3 h at 880 nm wavelength scanning. From the backscattering spectra, the Turbiscan stability index (TSI) was calculated and reported as a function of time.

The meltdown test was performed on 50 g ice cream aliquots placed on a plastic grid (3 holes/cm) fitted to drip into beakers. Ice cream blends were kept undisturbed at +23 °C (± 0.5 °C) for 1 h. The first dripping time was recorded, and the melting rate was calculated as a ratio between the melted mass over 60 min time.

A back extrusion test with a 40 mm rig was done to perform the texture analysis of the ice cream using a Texture Analyzer (TA.XT plus C, Stable Micro Systems, UK) fitted with a 50 kg loading cell. Before the analysis, 100 mL plastic cells with frozen samples (–15 °C) were placed onto the measuring plate at 25 °C for 20 min to soften.

# **3. Results and Discussion**

## **3.1 Ice cream mixtures characterization**

Cream-based and structured lipid-based blends did not have significantly different ζ-potential values (p < 0.05), opposing to significantly higher (p < 0.05) values for peanut oil-based samples demonstrating that the modification in the acylgycerols fractions ratio increased its surface charge. The higher surface charge would, in accordance with prior research, increase the electrostatic repulsion between particles, prevent particle agglomeration, and allow smaller particles to form more stable mixes (Zhu et al., 2019). A similar tendency was observed for the surface mean diameters D[3,2] and the volume mean diameters D[4,3], indicating that the particles of vegetable oil-based ice cream formulation clumped together, increasing their particle size. SLB had a 74 % smaller Dv(90) particle size when compared to POB, which demonstrated that lipid structuring allowed the formation of smaller aggregates due to a minor degree of coalescence among the de-emulsified fat (Roy et al., 2021). The presence of partial acylglycerols decreased the particle size of the SLB sample by 42.53 % compared to the POB, disclosing that the low-polar, non-ionic lipophilic nature of monoglycerides could lucratively inhibit particle aggregation.

## **3.2 Ice cream textural properties and stability**

## According to the findings of the texture analysis, lipid structuring ameliorated the firmness, consistency, and cohesiveness of the produced ice cream compared to the peanut oil-based sample. When compared to the CB formulation, the firmness and consistency values of the SLB were not significantly different (p < 0.05); however, significantly increased for POB (p < 0.05). This denoted that MAG and DAG fractions in SLB samples cross-linked with the water and lipid phase, subsequent in increased matrix stability. The greater firmness values for the POB sample thus could be justified by the development of ice crystals and large aggregates, as also confirmed by the particle size measurements of the mixtures.

Multiple-light scattering technique was used to assess the stability of the ice cream samples over melting at 25 °C. During the 3 h measurement, the TSI values of the POB sample were noticeably higher (up to 48) than those of the other samples, varying between 22–29, implying the progressed stability of SLB ice cream formulations. This could be reasoned by the incidence of MAG crystalline structure formed by the glycerolysis in the SLB sample, which hampered particle aggregation via the creation of a resistant to melting and structural failure system.

Melting is one of the benchmarks when assessing ice cream quality. The melting rate of the PBO ice cream was expressively higher (0.77 ± 0.042 g⋅min‑1) than that of the CB (0.017 ± 0.014 g⋅min-1) and SLB (0.044 ± 0.021 g⋅min-1) ice creams. Bearing the same fat content midst the samples, the ice cream made with the SL was more resistant to melting than the sample with the unstructured oil. The bettered melting resistance was attributed to a higher water-binding capacity, provided by an increased emulsification efficiency of the SLB sample compared to the POB ice cream (Góral et al., 2018). Calligaris et al. (2018) have also reported that partial acylglycerides could counteract the lack of solid fat crystals at the surface of air cells, supporting partial coalescence and leading to a structure more able to hold its shape during ice crystals melting. Thus, creating a more stable matrix with a softer texture due to the entrapped air.

The obtained results demonstrated that the studied properties of the ice cream were significantly diminished when liquid oil was used to substitute milkfat in the formulation. This effect is attributed to the absence of solid fat crystals on the air cell surfaces, which limited partial coalescence and made the structure less stable when ice crystals melted (Calligaris et al., 2018). Thus, the experimental results agree with the expected ones, and the present Ph.D. thesis project can be advanced devoid of any significant modification.

# **4. References**

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