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Effect of cooling rate on lipid crystallization in oil-in-water emulsions

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ABSTRACT

The effect of cooling rate on the destabilization of oil-in-water (o/w) emulsions was studied as a function of oil content (20% and 40% o/w), homogenization conditions, and crystallization temperatures (10, 5, 0, −5 and −10 °C). The lipid phase was a mixture of anhydrous milk fat and soybean oil, and whey protein was used as the emulsifier. Differential scanning calorimetry was used to analyze the crystallization and melting behaviors; while a vertical scan macroscopic analyzer measured the physicochemical stability. Slow cooling rate increased the stability of emulsions with 20% oil. In addition, slow cooling promoted the onset of crystallization and delayed crystal growth. These effects were more significant in emulsions formulated with 20% oil and formulated under processing conditions that resulted in bigger droplet sizes (~0.9 μm).

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1. Introduction

Nations around the world (e.g., Denmark, Canada, USA, etc.) have begun to educate the consumer and restrict the use of *trans*-fatty acids (TFA) in foods. They have done this by including TFA information on nutrition labels, restricting the use of TFA in restaurants, and the amount of TFA in oils used for consumption (Barboza, 2007; FDA Consumer Magazine, 2003; Lueck & Severson, 2006; Stender & Dverberg, 2004). A concern with replacing TFA is that desirable properties (e.g., texture, flavor, and shelf-life) are at risk. Currently, TFAs are being replaced by saturated fats such as coconut and palm oil. Saturated fats are a good substitute in as much as they maintain the quality of the product's texture and flavor; however, this can only be a temporary substitute as these saturated fats are high in palmitic and lauric fatty acids, which are known to contribute to heart disease and cholesterol in a similar manner to TFAs (Mensink, Zock, Kester, & Katan, 2003; Simon et al., 1995). Therefore, healthier alternatives to TFA must be sought that will contribute healthy fatty acids to foods while meeting consumers' expectations. Anhydrous milk fat (AMF) has the potential to be used as a TFA replacement since it has a relatively low content of palmitic fatty acids and a very characteristics buttery flavor that consumers prefer.

Several food products such as mayonnaise, spreads, butter, and dressings are composed of an oil and water phase forming emulsions. Processing conditions and ingredients affect the emulsion stability and their destabilization mechanisms. Among the different factors that can affect emulsions stability, protein concentration, type of lipid phase, oil-to-water ratio and the presence of hydrocolloids in the water phase are the most important ones. A myriad of studies have been performed to evaluate the effect of these processing variables on the stability of emulsions (Carrera Sanchez & Rodriguez Patino, 2005; Dickinson, Golding, & Povey, 1997; Elizalde, Pilosof, & Bartholomai, 1991a, 1991b; Sun & Gunasekaran, 2009; Vega, Goff, & Roos, 2007) showing that in general, concentrations of 0.3–2 wt% of dairy proteins are efficient to stabilize oil-in-water emulsions ranging from 10% to 45% oil in water. The most efficient protein concentration will depend on the type of protein used and on the oil-to-water ratio.

Since emulsions are formed by an aqueous and lipid phase, crystal formation in the lipid phase might also affect the stability of these systems. Previous research in bulk lipids has shown that cooling rate affects crystal formation, which can affect the smoothness or graininess of margarine, the snap and gloss of chocolate, and the spreadability of butter and margarine (Campos, Narine, & Marangoni, 2002; Martini, Herrera, & Hartel, 2002). These authors showed that when bulk lipids are cooled quickly (e.g., quenching) many small crystals form, which are considered unstable and yet rigid. If the system is cooled slowly, larger but fewer lipid crystals form having had time for the triacylglycerides (TAGs) to adjust and fit together in a preferable uniform lattice and are in a more stable form (Campos et al., 2002; Martini, Herrera, & Hartel, 2001; Martini et al., 2002; Sato, 2001). In emulsions, it has been suggested that the less stable form is more rigid and therefore unable to bend within its confined barrier (lamella) and thereby puncture the confinement and cause partial coalescence (Coupland, 2002). Thus, emulsion stability is influenced by the crystalline form produced by either a slow or fast cooling rate.

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Crystallization of lipids in oil-in-water emulsions has been well reviewed by Rousseau (2000) and Coupland (2002). These reviews clearly state the importance of lipid crystals in emulsions and how they affect emulsion stability. Lopez et al. (2002) studied the effect of cooling rate on milk fat and cream and found that though crystallization of the lipid state did not change significantly; there was a difference in the melting profiles. These authors showed that the slower the cooling rate the less fractionated the milk-fat fractions. In addition, Vanapalli, Palanuwech, and Coupland (2002) studied the effect of oil type, dispersed volume fraction and cooling rate on the emulsion stability towards freeze–thawing conditions. Their research shows that less saturated lipids and a fast cooling rate increase the stability of the emulsions towards freeze–thawing. Additional contributions in studying the effect of crystallization in emulsions has been done by Hindle, Povey, and Smith (2000, 2002), who have studied the nucleation effect of seed crystals and droplet collision (agitation of emulsion) on cocoa butter and its various forms of crystals. They found that the initial seed crystallization is enhanced when agitation is used. Lopez et al. (2001) studied the effect of combining fats and oils to create a model emulsion, which would maintain liquid status at low temperatures, finding that the blend was more stable than an emulsion with only the solid fat.

The objective of this study is to contribute to the existing knowledge of food emulsions by evaluating the effect of formulation and processing conditions on the crystallization behavior of oil-in-water emulsions and their effect on emulsions' physicochemical stability. Model systems consisted of oil-in-water emulsions formulated with equal parts of AMF and soybean oil (SBO). Two oil-in-water ratios (20% and 40% oil) were used and different processing conditions (cooling rate, homogenization and crystallization temperature) were assayed.

2. Materials and methods

2.1. Emulsion formulation

A 50 wt% blend of soybean oil (SBO) donated by Bunge Limited (St. Louis, MO) and anhydrous milk fat (AMF), donated by Kraft Foods Inc. (Chicago, IL) was used as the oil phase. Both lipids were melted by heating to ~60 °C for at least 30 min prior to mixing.

The water phase was prepared by dispersing 2.0 wt% whey protein isolate (WPI) (Inpro 90 by Vitalus (Abbotsford, B.C., Canada), which consists of ⩾92% whey protein, ⩽3.0% lactose, ⩽5.0% moisture, ⩽1.0% fat, and ⩽3.5% ash) in a 0.01 M ($Na_2HPO_4{\cdot}7H_2O$) aqueous solution (pH 7.28). The solution was then filtered (Whatman #1 filter paper) to eliminate any possible non-dissolved particles that might affect the stability/instability of emulsions. The solution was then heated to ~60 °C for at least 30 min prior to homogenization of the two phases.

Oil phase was added to water phase for a total of 50 g in a 100 mL beaker. Two oil-in-water (o/w) ratios were used: 40:60 and 20:80 (o/w expressed in wt%), which are o/w ratios commonly used in many lipid based foods such as salad dressings.

2.2. Emulsion preparation

The phases were homogenized using two methods: very low pressure homogenization and high pressure homogenization. Very low pressure homogenization (VLPH) was done by first mixing the phases with an Ultra Turrax (IKA T18 basic) at 18,000 rpm for 1 min. The mixture was then quickly (less than 2 min) put through a Microfluidics Microfluidizer Processor (Model M-110S, Newton, MA) at 2530 ± 230 psi (17.4 ± 1.6 MPa). The microfluidizer coil was kept at approximately 60 °C to avoid lipid crystallization during emulsion formation. High pressure homogenization (HPH) was the same as VLPH, except with a pressure of 9430 ± 230 psi (65.0 ± 1.6 MPa).

2.3. Differential scanning calorimetry (DSC)

The crystallization and melting behaviors of the samples were studied by DSC (TA Instruments, 2910, New Castle, DE). Approximately 5–15 mg of a sample was placed in a DSC pan soon after homogenization. The DSC pans were kept at approximately 60 °C to avoid cooling the sample prior to analysis. The DSC was calibrated with Indium at a heating rate of 5 °C/min.

Crystallization and melting enthalpies (expressed in units of J/g), with peak and onset temperatures (given in °C), were calculated for all emulsions. Enthalpy comparisons were based on the oil phase only.

$$\text{Oil, phase enthalpy (J/g)} = \frac{\text{sample enthalpy (J/g)}}{\text{weight fraction of oil}} \quad (1)$$

Samples were placed in the DSC chamber at an initial temperature of 60 °C and then cooled at a rate of 30 °C/min (fast cooling rate) or 0.2 °C/min (slow cooling rate) to T_c (i.e., 10, 5, 0, −5 and −10 °C) and held at T_c for 3 h. Samples were then heated at 5 °C/min to analyze the melting profile of the crystallized fat. The crystallization and melting behavior were analyzed using this technique and compared with the physicochemical stability measured as described below.

2.4. Physicochemical stability

Five to seven mL of the emulsions, formulated as described above, were placed in a test tube designed especially for the TurbiScan 2000 (Sandyhook, CT). The TurbiScan consists of a reading head moving along a flat-bottomed cylindrical cell while scanning the entire sample height. The reading head consists of a pulsed near-infrared light source (λ = 850 nm) and two synchronous detectors. The transmission detector picks up the light transmitted through the product, and the backscattering (BS) detector receives the light backscattered by the product (135° from the incident beam). The emulsions in this study are opaque, therefore only BS profiles were used to evaluate the physicochemical stability of emulsions. The reading head acquires BS data every 40 μm and the profile obtained characterizes the sample's homogeneity, particle concentration, and mean diameter. These parameters are represented by a curve showing the percentage of backscattered as a function of the sample height in mm. The acquisition along the product is then repeated obtaining a superimposition of sample fingerprints, which characterizes the stability or instability of the sample (i.e., the more identical the readings, the more stable the system) (Mengual, Meunier, Cayré, Puech, & Snabre, 1999; Tippetts & Martini, 2009).

Fast cooling rate: An initial reading (i.e., when the sample was still approximately 60 °C) was taken prior to the sample being placed in a water bath thermostatized at a specific T_c. The sample was then placed in a water bath thermostatized at T_c. The physicochemical stability of the emulsions was measured during 3 h at T_c. Measurements were taken every 10 min for the first hour and then every 15 min for the next 2 h. To perform the BS measurement, test tubes with the emulsions were taken from the water bath (set at T_c) and placed in the TurbiScan. After the measurement was taken (40 s) the assay tube was placed again in the thermostatized water bath.

Slow cooling rate: After the initial reading was taken, the sample was placed in a programmable water bath (Ecoline Lauda E300, Westbury, NY), which was initially set at 60 °C. The waterbath

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