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# Production of high-oleic palm oil nanoemulsions by high-shear homogenization (microfluidization)



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#### ABSTRACT

Nanoemulsions present benefits such as an increase in the bioavailability, solubility and targeted delivery of encapsulated substances, and thus, they are a method of incorporating high nutritional value oils, such as high-oleic palm oil (HOPO). In this work, O/W nanoemulsions were obtained by microfluidization using HOPO (1-20% w/w) in the oily phase along with whey (1-20% w/w), Tween 20 (1:1 w/w ratio) and water in the aqueous phase following a surface response design methodology. The response variables were the average drop size (ADS), the polydispersity index (PDI), the zeta potential ( $\zeta$ ), CIELAB color parameters and viscosities of the fresh nanoemulsions (0 days) and nanoemulsions stored at two temperatures (5 and 19 °C) for 4 days. The ADS, PDI and  $\zeta$  values varied between 163 and 2268 nm, 0.2 and 1, and -29.7 and -47.2 mV, respectively. The viscosity was affected by the storage temperature; after 4 days at 19 °C, it increased almost 6-fold compared to the viscosity of the fresh sample. With regards to the color parameters, significant changes were observed based on the concentrations of HOPO and whey. In addition, the prediction equations only presented errors below 7% for the evaluated variables, with R<sup>2</sup> values above 0.85. Finally, the influence of whey protein denaturation at 60 °C on the stabilities of the two most stable nanoemulsions, according to the optimization process, was observed. Industrial relevance: Among its many benefits, nanoencapsulation is characterized by increasing the bioavailability of the encapsulated active compound and by the protection that it grants against environmental and processing effects, as micronutrients (for example vitamins) can be susceptible to chemical, enzymatic and/physical instability prior, during and after the processing of food products that contain. One of the techniques studied in recent decades for obtaining nanoemulsions is microfluidization. Microfluidization is a high-energy method that uses high pressure to force the fluid through microchannels that have a specific configuration, emulsifying the fluid by the combined effects of cavitation, shear and impact, thus showing an excellent emulsifying efficiency. However, in food industries the use of microfluidization is not popular and other kind of high shear homogenization are used. In this work, the development of stable emulsions using microfluidization, calls for the use of other types of materials that can provide emulsifying characteristics, such as whey, a compound that is currently one of the main effluents of dairy processes, depending on the type of product.

Obtaining nanoemulsions for encapsulation purposes has been studied in many functional products, but to the best of our knowledge, it has not been reported with high-oleic palm oil. This oil contains approximately 50% saturated, 10% diunsaturated and 40% monounsaturated fatty acids, with oleic acid in sn-2 position in triacylglycerols. This composition makes palm oil as soluble as olive oil. In addition, high-oleic palm oil (HOPO), in particular, has a high stability because it is an oleic acid-rich oil, which has been introduced to replace trans fats and has presented a healthy alternative to such fats in food formulations and the fried food industry.

It is also important to highlight that oleic acid has a range of health benefits, such as a decrease in the total cholesterol, an increase in HDL (high-density lipoprotein) and a decrease in LDL (low-density lipoprotein). Oleic acid also retards the development of heart diseases, promotes the formation of antioxidants in the body and reinforces the integrity of the cell wall. In addition, red palm oil (crude) contributes significant nutritional value because it is rich in  $\beta$ -carotenes,  $\alpha$ -tocopherol and tocotorienols, supplying vitamins and provitamins that are important for but not produced by the human body.

Finally, this work demonstrates that emulsion drop size does not affect the stability of the nanoemulsion if it formulation is designed. Therefore, the goal of this work was to evaluate the most favorable conditions for the microfluidization, formulation and storage of HOPO nanoemulsions using whey powder to produce stable nanoemulsions.

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#### 1. Introduction

Nanoencapsulation is a process by which substances are packed in a miniature-sized vessel, and bioactive packing is performed at the nanoscale (Quintanilla-Carvajal et al., 2009). Among its many benefits, nanoencapsulation is characterized by increasing the bioavailability of the encapsulated active compound and by the protection that it grants against environmental and processing effects, as micronutrients (for example vitamins) can be susceptible to chemical, enzymatic and/physical instability prior, during and after the processing of food products that contain them (Joye, Davidov-Pardo, & McClements, 2014). In many cases, nanoencapsulation begins with the production of nanoemulsions, which are systems formed by an oily and an aqueous phase, in which they are emulsified through the use of an emulsifier. In addition, the nanoemulsions are formed with small drop sizes and high surface areas (Kim, Oh, Lee, Song, & Min, 2014). Such properties grant them potential advantages over conventional emulsions, such as a good physical stability and higher bioavailability (Tabibiazar et al., 2015). One of the techniques studied in recent decades for obtaining nanoemulsions is microfluidization. Microfluidization is a high-energy method that uses high pressure to force the fluid through microchannels that have a specific configuration, emulsifying the fluid by the combined effects of cavitation, shear and impact, thus showing an excellent emulsifying efficiency (Shen & Tang, 2012). Furthermore, the microfluidization method requires high-energy inputs and dedicated equipment, but could produce ultrafine emulsions at much lower surfactant-to-oil ratio (SOR < 0.1) (Komaiko & McClements, 2015; Yang, Marshall-Breton, Leser, Sher, & McClements, 2012).

It is also important to highlight the role of the surfactant when generating an emulsion that is stable over time. However, the development of stable emulsions calls for the use of other types of materials that can provide this type of emulsifying characteristic, such as whey, a compound that is currently one of the main effluents of dairy processes, depending on the type of product (Das, Sarkar, Sarkar, Bhattacharjee, & Bhattacharjee, 2015). Thus, this residue has been studied in the production of emulsions or as a coating material for encapsulation processes at different scales (Esfanjani, Jafari, Assadpoor, & Mohammadi, 2015; Wang et al., 2016) because it contains approximately 2–18% proteins, 8–14% minerals (ash) and 77–80% carbohydrates (Muangrat, Onwudili, & Williams, 2011).

On the other hand, the characteristics related to the stability of an emulsion are the drop size, the viscosity and the zeta potential  $(\zeta)$ . However, the application of nanoemulsions in diverse food matrices must also ensure the acceptance of parameters such as the color. These parameters are related to the quality and the consumer perception, both visual as well as sensory, towards the product because the color and appearance of foods are important factors that contribute to their selection by the consumer (Silva et al., 2011).

Obtaining nanoemulsions for encapsulation purposes has been studied in many functional products, but to the best of our knowledge, it has not been reported with high-oleic palm oil. This oil contains approximately 50% saturated, 10% di-unsaturated and 40% monounsaturated fatty acids, with oleic acid in sn-2 position in triacylglycerols. This composition makes palm oil as soluble as olive oil (Akoh, 2005). In addition, high-oleic palm oil (HOPO), in particular, has a high stability because it is an oleic acid-rich oil, which has been introduced to replace trans fats and has presented a healthy alternative to such fats in food formulations and the fried food industry (Kodali, 2014).

It is also important to highlight that oleic acid has a range of health benefits, such as a decrease in the total cholesterol, an increase in HDL (high-density lipoprotein) and a decrease in LDL (low-density lipoprotein). Oleic acid also retards the development of heart diseases, promotes the formation of antioxidants in the body and reinforces the integrity of the cell wall (Munévar, 2010). In addition, red palm oil (crude) contributes significant nutritional value because it is rich in  $\beta$ -carotenes,  $\alpha$ -tocopherol and tocotorienols (Akoh, 2005), supplying

vitamins and provitamins that are important for but not produced by the human body.

Therefore, the goal of this work was to evaluate the most favorable conditions for the microfluidization, formulation and storage of HOPO nanoemulsions using whey powder to produce stable nanoemulsions.

#### 2. Methodology and materials

#### 2.1. Materials

The nanoemulsions were prepared with crude high-oleic palm oil donated by the National Federation of Palm Oil Growers (Federación Nacional de Cultivadores de Palma de Aceite) (Colombia), whey powder donated by the Alpina Corporation, Tween 20 (Scharlau, Spain) and Milli-Q water.

#### 2.2. Preparation of coarse emulsions

The coarse emulsions were homogenized in an Ultra-Turrax (IKA, USA) at 9500 rpm, incorporating Tween 20 followed by the sequential addition of whey powder and HOPO to the Milli-Q water over 10 min. Subsequently, such emulsions were processed to obtain the nanoemulsions.

#### 2.3. Nanoemulsion preparation

The nanoemulsions were obtained following the methodology of (Quintanilla-Carvajal et al., 2014), with some modifications. They were homogenized in an LM10 microfluidizer (Microfluidics, England) following a response optimization design obtained from the software Design Expert Version 7.1.0 (Stat-Ease Inc., MN, USA), in which the following three numerical and one categorical factors were varied: HOPO concentration (1–20% w/w), whey concentration (1–20% w/w), and microfluidization pressure (10,000–20,000 psi), as well as the microfluidization cycles as a three-level categorization factor (1, 2 and 3). The Tween 20 concentration was held constant at 1% w/w with respect to the HOPO concentration. This emulsifier was chosen for allowing rapidly adsorb to the surface of the oil droplets, reduce interfacial tension to prevent droplet coalescence (Degner, Chung, Schlegel, Hutkins, & McClements, 2014; Jo & Kwon, 2014; Teo et al., 2016) and has shown good results in small particles for various applications including nanoemulsions (Silva et al., 2011).

Table 1 shows the conditions provided by the response optimization design for the preparation of HOPO nanoemulsions.

### 2.4. Storage of the HOPO nanoemulsions

The nanoemulsions obtained (Table 1) were stored for 4 days (t=4) (Adjonu, Doran, Torley, & Agboola, 2014; Y. Li, Zheng, Xiao, & McClements, 2012). These samples were evaluated at two different conditions: room temperature (19 °C) and refrigeration temperature (5 °C).

#### 2.5. Nanoemulsion characterization

The fresh nanoemulsions were characterized with respect to the average drop size (ADS), the polydispersity index (PDI), the  $\zeta$ , the color according to the CIELAB method (L\*, a\*, b\*, C<sub>ab</sub>\* and h\*<sub>ab</sub>) and the apparent viscosity ( $\eta$ ). This last response variable was measured for the fresh nanoemulsions and t=4 at room and refrigeration temperatures.

## 2.5.1. Analysis of the drop size, PDI and $\zeta$

The ADS, the PDI and the  $\zeta$  were measured in the nanoemulsions reported in Table 1 by the use of a Zetasizer NanoZS (Malvern Instruments, England) dynamic light scattering (DLS) device with a laser diffractometer using a water dilution of 1:100 v/v (Qian, Decker, Xiao, & McClements, 2012; Sessa et al., 2014). The measurements were performed in triplicate with a scattering angle of 173°.

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