



Functional properties of ultrasonically generated flaxseed oil-dairy emulsions



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ABSTRACT

This study reports on the functional properties of 7% flaxseed oil/milk emulsion obtained by sonication (OM) using 20 kHz ultrasound (US) at 176 W for 1–8 min in two different delivery formulae, viz., ready-to-drink (RTD) and lactic acid gel. The RTD emulsions showed no change in viscosity after sonication for up to 8 min followed by storage up to a minimum of 9 days at 4 ± 2 °C. Similarly, the oxidative stability of the RTD emulsion was studied by measuring the conjugated diene hydroperoxides (CD). The CD was unaffected after 8 min of ultrasonic processing. The safety aspect of US processing was evaluated by measuring the formation of CD at different power levels. The functional properties of OM gels were evaluated by small and large scale deformation studies. The sonication process improved the gelation characteristics, viz., decreased gelation time, increased elastic nature, decreased syneresis and increased gel strength. The presence of finer sono-emulsified oil globules, stabilized by partially denatured whey proteins, contributed to the improvements in the gel structure in comparison to sonicated and unsonicated pasteurized homogenized skim milk (PHSM) gels. A sono-emulsification process of 5 min followed by gelation for about 11 min can produce gels of highest textural attributes.

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1. Introduction

In today's consumer market, dairy systems such as liquid milk and yoghurt are considered as common vehicles for the delivery of high-value nutrients [1,2]. Carotenes, fish oil, flaxseed oil and phytosterols are some popular non-polar bioactives/nutrients [3]. In both delivery formulae, the bioactives had to be initially dispersed as a stable oil-in-water emulsion and later down-streamed into a variety of end products. Generally, conventional emulsification techniques (e.g., piston homogenizer and high shear mixer) are widely employed to deliver the bioactives in liquid foods and in a variety of dairy systems [4,5]. Very recently, low frequency high intensity US has gained popularity among food researchers in order to deliver high-value nutrients [6]. Few review papers have summarised the advantages of US for the production of food

grade emulsions [7–9]. The physical effects of US, viz., mechanical vibration, microstreaming and intense shear forces generated by acoustic cavitation are considered to be the main reason for the generation of emulsions [10–12]. In a previous study, we reported physical characterization and stability of US-generated flaxseed oil emulsions in a dairy system, viz., PHSM under various operating conditions [13]. In this article, we discuss the functional properties of ultrasonically generated 7% OM at different power levels (132 and 176 W) and different sonication time (1–8 min). The choices of the above variables are based on the properties of OMs, which showed highest cream stability against phase separation (until 9 days at 4 ± 2 °C) at lowest processing times and optimum power levels, as previously reported [13].

The selection of liquid milk for the delivery of nutrients is based on two reasons: (1) frequency of consumption, ease of processing, storage and packaging conditions [5], and (2) ease of conversion into a variety of consumer products such as RTD [14] and yoghurt (lactic acid gel). Similarly, the choice of flaxseed oil is due to its novelty to be dispersed as an emulsion in the dairy system using a non-conventional emulsification technique at ambient temperature. Flaxseed oil, a vegetarian source of omega-3 fatty acids is rich in α -linolenic acid and is used in the diets targeting heart health benefits, neural growth and cognitive skills development [15–17].

Abbreviations: OM, flaxseed oil/milk emulsion by sonication; US, ultrasound; RTD, ready-to-drink; CD, conjugated diene hydroperoxides; PHSM, pasteurized homogenized skim milk; PUFA, poly unsaturated fatty acid; ALA, alpha-linolenic acid; NEM, N-ethylmaleimide; GDL, glucono-delta-lactone; NAP, nominal applied powers; ANOVA, analysis of variance; DWP, denatured whey proteins.

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There are no previous studies reported on the functionality of flaxseed oil enriched dairy formulations, viz., RTD (beverage emulsions) and acid gels prepared using ultrasound. In the current study, the functionality of a food formulation is determined from the rheological/flow behaviour studies and shelf life/storage studies.

The commercial feasibility of milk RTD is based on their functional properties during and after production. Industrially, the rheology of beverage emulsions is important as it influences the ease of mixing, flow characteristics, and packaging. This requires careful control of the total emulsion droplet concentration in the system [18]. In addition, oxidation of beverage emulsions is a main concern as there are many technical difficulties in incorporating PUFA rich flaxseed oil due to their high susceptibility to oxidation, which are primarily caused by environmental stress such as exposure to light and oxygen. The oxidation of a non-polar bioactive could lead to loss of ingredient functionality and development of off-flavours, making the product undesirable and inconsumable. In complex emulsion systems, many factors, such as transition metals, interfacial area, processing conditions, type of emulsifier and droplet size, can affect the initiation and propagation of oxidation. For example, radical scavenging properties provided by the DWP (emulsifier) can reduce oxidation of omega 3 fatty acids of fish oil [19]. In the present study, US induced changes to milk proteins could possibly bring protective effect to the emulsions [20].

The functional properties of milk gels, especially rheological and textural properties determine the sensory perception and consumer acceptability of the product [21]. These properties are monitored by small and large scale deformation measurements in the current study in order to predict consumer acceptability. Pereira et al. [22] have described the correlation between non-oral and instrumental analyses of acid gel texture using gel strength, syneresis and microstructural studies. Most of the instrumental studies explain the rheology of heat-treated milk system and the mechanism of gelation in comparison to unheated-native milk system [23–26]. Few of these studies discuss the improvements in the gelation characteristics and textural attributes/advantages of acid gels by altering the amounts of fat, solids not fat content and fat membrane surfaces [21,27–29].

As mentioned earlier, no previous study is available on the functionality of sono-emulsified flaxseed oil rich dairy systems, viz., RTD and acid gel. A few preliminary studies on sonication and thermosonication highlight the changes to the individual components of OM emulsions, such as flaxseed or a PUFA rich oil, cheese milk and skim milk. Studies on the oxidative deterioration of vegetable oils, such as, unsaturated fatty acid-rich flaxseed oil, sunflower oil and olive oil upon sonication at 20 and 24 kHz have shown contrasting results. A few studies have indicated the generation of oxidative end products, such as, lipid peroxides, conjugated dienes, malonaldehyde are negligible/minor [30,31], while some reported prevalence of oxidative damages [32,33]. These variations in oxidation could be due to varying experimental and environmental conditions prevailed during the study. Ashokkumar et al. [34] have shown that an unwanted reaction like oxidation between ultrasonically generated radicals and food ingredients could be minimised by selecting lower ultrasonic frequencies for processing.

Only a handful of studies exist highlighting the gelation properties of ultrasonicated milk. Most of these studies involve the effect of heat and/or pressure along with US [35–37], but studies relating to sole effect of US are very few. Wu et al. [38] have shown that homogenization of fresh raw milk at 15 °C using 20 kHz US (applied power 200–500 W) improved the gel characteristics of yoghurt in comparison to conventional homogenization process (12410 kPa at 60 °C). In a similar study, ultrasonication of skim milk using 22.5 kHz US resulted in slight improvements in storage modulus (G') of acid gels in comparison to unheated skim milk

[39]. A different study by Bermudez-Aguirre and Canovas [40] has reported the incorporation of flaxseed oil (1% w/w) in cheese milk and increase in cheese yield by thermosonication process at 24 kHz, 63 ± 0.5 °C for 5–30 min. However, the effect of thermal treatment and its consequences on denaturation of whey proteins [41] and yield of curd has not been uncoupled from the effect of US. The current study reports on the viscosity, gelation, gel strength, storage and oxidative changes of OM prepared in two different product formulations, viz., RTD and acid gel. The findings of the study may help food manufacturers to develop new products and processes using bioactives and US.

2. Materials and methods

2.1. Materials

Fresh PHSM was purchased from a local supermarket and immediately stored at 4 °C until further use. The composition of the milk was 3.5% protein, 0.1% fat, and 4.9% lactose as labelled by the manufacturer. The manufacturer's specification was cross-checked in our lab for the proteins. The protein content was 3.46% by Bradford Assay. Also, we did not observe any creaming-off in the PHSM sample for about 10 days of storage at 4 ± 2 °C indicating that fat content was very low. Ultra pure (MilliQ) water was used in all experiments. Unrefined organically grown cold pressed flaxseed oil with 59.9% of ALA was a gift sample from Stoney Creek Oil Products Pty Ltd., Australia. NEM, GDL, iso-octane, 2-propanol were purchased from Sigma-Aldrich Chemicals, Australia.

2.2. Emulsification by US

Milk (93% v/v) and oil (7% v/v) phases were added sequentially to a water jacketed glass vessel and the sonicator horn was positioned at a depth of 0.3 ± 0.1 cm. Emulsions were obtained as 50 ml aliquots using a 20 kHz, 450 W ultrasonic unit (12 mm diameter horn, Branson Sonifier, Model 102 (CE)) at 132 and 176 W of NAP for different processing times (1–8 min). During sonication, thermostated water was circulated continuously through a jacket surrounding the sonication cell and the water temperature was maintained at 22.5 ± 2 °C. The emulsified samples were stored in a refrigerator for about 9 days at 4 ± 2 °C. The analysis and storage studies were performed on both fresh and stored samples.

2.3. Viscosity

The apparent viscosity of the PHSM and OM was determined using Rheometrics ARG2 rheometer (TA Instruments). A steady stress sweep experiment was carried out using a 40 mm parallel plate geometry and 1 mm gap at 22 °C. The stress was varied to obtain the viscosity data between the shear rates of 10 and 200 s^{-1} .

2.4. Conjugated Dienes

Lipid oxidation in the emulsions was evaluated from the formation of CD. An emulsion sample of 200 μl was mixed with 10 ml iso-octane/2-propanol (2:1, v/v), vortexed immediately for 1 min and centrifuged (5000g, 5 min, Allegra 25R). The organic phase was isolated and absorbance was measured at 232 nm using a UV-vis spectrophotometer (Carey Bio 50, Varian) after filtering through 0.25 μm syringe filter. The filtration was carried out just before the measurement to remove the proteins from the sample, which might contribute to spectral interference in the same region [42].

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