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Original Article

Properties and stability of Pickering emulsions stabilized by nanofibrillated mangosteen cellulose: Impact of oil type and emulsifier concentration

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Abstract

We aimed to examine the influence of three types of oil and the concentrations of nanofibrillated cellulose (NFC) extracted from mangosteen rind on the properties and stability of 10% oil-in-water (O/W) Pickering emulsions. The properties and stability of the emulsions were influenced by the NFC concentration rather than the oil type. Microscopic observations revealed that NFC stabilized the emulsions by adsorbing at the oil-water interface and forming a steric barrier that provided electrostatic repulsion. Furthermore, a three-dimensional network in the continuous phase was formed which retarded droplet mobility and collision, thereby preventing coalescence. NFC stabilized emulsions exhibited a gel-like behavior and an increase in the NFC concentration led to an increase in the strength and stability of the emulsion. The emulsions with NFC concentrations ≥0.5% were found stable to coalescence for a period of 90 days.

Keywords: Pickering emulsion, oil type, nanofibrillated cellulose, mangosteen rind, phase separation

1. Introduction

An emulsion is a mixture of two immiscible liquids mainly oil and water where one liquid is dispersed in the other in the form of small spherical droplets. They can be classified according to the spatial distribution of the oil and water phases into three types. First are oil-in-water (O/W) emulsions where the oil droplets are dispersed in the aqueous phase. Second are water-in-oil (W/O) emulsions where water is dispersed as droplets in the oil phase and third are multiple emulsions that are more complex where the dispersed phase is itself an emulsion (McClements, 2005; McClements & Decker, 2000). These emulsions systems are known to have a wide range of applications in the cosmetic, food, and pharmaceutical Industries (Dickinson, 2010; Yang *et al.*, 2017). However, they are thermodynamically unstable and are unable to remain stable for prolonged periods. Therefore, emulsifiers are incorporated that help increase their stability by reducing the interfacial tension between the two phases and postpone phase separation (McClements & Decker, 2000; McClements, 2005). In dif-ferent food emulsions, the droplets are stabilized by food grade emulsifiers, such as phospholipids, proteins, polysac-charides, and particles. Recently, however, there has been considerable interest in the utilization of solid particles as emulsifiers over the proteins and other synthetic emulsifiers.

Solid particles can form and stabilize oil-in-water emulsions through the "Pickering mechanism" to form Pickering emulsions (Chevalier & Bolzinger, 2013; McClements & Decker, 2000; Wu *et al.*, 2015). These emulsions have benefits of being extremely stable to coalescence in comparison to emulsions stabilized by other surfactants, such as proteins and small molecule emulsifiers because the Pickering emulsifiers follow an irreversible adsorption mechanism and

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require very large free energy for their spontaneous desorption from the interface (Gong, Wang, & Chen, 2017; Gould, Garcia, & Wolf, 2016; Yang *et al.*, 2017). The theory that describes the stabilization mechanism of solid particles is the formation of a densely packed particle layer at the oil-water interface that provides a steric barrier to prevent droplet coalscence and retard creaming and sedimentation (Gestranius, Stenius, Kontturi, Sjöblom, & Tammelin, 2017; Hu, Ballinger, Pelton, & Cranston, 2015; Yang *et al.*, 2017). Many inorganic and petrochemical-based particles, such as silica, clay, calcium carbonate, hematite, and polystyrene, can be used to stabilize Pickering emulsions. However, Pickering emulsifiers of biological origins, such as cellulose derivatives and starch granules, are preferred since they are biocompatible, biodegradable, and environmentally friendly (Gong *et al.*, 2017; Hu *et al.*, 2015).

In the past decade, cellulose materials derived either from plant sources or bacterial sources are used more often as Pickering emulsifiers because cellulose can pack densely at the interface as a monolayer between the two phases or can form a three-dimensional network that stabilizes the emulsion droplets (Gestranius *et al.*, 2017; Paximada, Koutinas, Scholten, & Mandala, 2016; Rein, Khalfin, & Cohen, 2012; Winuprasith & Suphantharika, 2015). Nanofibrillated cellulose (NFC) is potentially available as a Pickering emulsifier which is derived from natural fibers by chemical pre-treatment and mechanical treatment for fibrillation. The fibrillation process causes irreversible changes in the physical properties of the fiber by breaking the cell wall and reducing fiber size. In addition, NFC exhibits many interesting properties such as a high area/volume ratio which is suitable for nanocomposite production and a high specific surface area that provides an efficient interaction between the NFC and matrix (Ardanuy *et al.*, 2012; Correia, Santos, Telxeria, & Junior, 2018; Gómez *et al.*, 2016; Naderi *et al.*, 2017; Tuzzin, Godinho, Dettmer, & Zattera, 2016). Furthermore, due to the hydrophilic nature, NFC is wetted better by water than oil, thereby solubilizing in the continuous phase of the emulsion system and better stabilizing the oil-in-water emulsions as suggested by the Bancroft's theory. Also, NFC is an anionic carbohydrate biopolymer that stabilizes emulsions mainly through a steric (mechanical) barrier (Winuprasith & Suphantharika, 2013, 2015). Moreover, since it has small fibers, it is more effective in covering the surface of the emulsion droplets which provides greater emulsion stability (Capron, Rojas, & Bordes, 2017; Carrillo, Nypelö, & Rojas, 2015; Gómez *et al.*, 2016; Rein *et al.*, 2012).

NFC is generally isolated from various sources such as cellulosic residues and wastes generated from agricultural and industrial activities, such as areca nut husk (Chandra, George, & Narayanankutty, 2016), agave and barley husk (Espino *et al.*, 2014), wheat straw (Kaushik & Singh, 2011), banana fiber (Deepa *et al.*, 2011), jute bast (Thomas *et al.*, 2015), sugarcane bagasse (Li *et al.*, 2012; Mandal & Chakrabarty, 2011), and pineapple leaf (Cherian *et al.*, 2010, 2011; Santos *et al.*, 2013). In this study, NFC was extracted from mangosteen (*Garcinia mangostana* L.) rind which is an agricultural waste by combining alkali pre-treatment with high pressure homogenization. The emulsion stabilizing properties of NFC were evaluated by investigating major factors such as NFC concentration (0.3–0.7% [w/w]) and oil type (soybean oil, coconut oil, and rice bran oil) on the initial oil droplet size, ζ-potential, color, microstructure, and rheological properties of NFC stabilized O/W Pickering emulsions.

2. Materials and Methods

2.1 Materials

Dried mangosteen rind (*Garcinia mangostana* L.) rind, a byproduct of the mangosteen canning process, was provided by a local manufacturer (Chanthaburi, Thailand). Three types of oil that included soybean oil, coconut oil, and rice bran oil were purchased from a local supermarket. All chemicals and reagents used in this study were of analytical grade and double distilled water was used for the preparation of all solutions and emulsions. Vitamin D_3 in medium chain triglyceride oil was obtained from BASF (Thai) Ltd. Whey protein isolate (Provon 292) was obtained from Glanbia Nutritionals (NA), Inc.

2.2 Nanofibrillated cellulose (NFC) preparation

Nanofibrillated cellulose (NFC) was prepared using an alkaline extraction method according to a previously described protocol (Winuprasith *et al.*, 2018). Briefly, the dried mangosteen rind powder was extracted for cellulose using hot (90 °C) aqueous sodium hydroxide (NaOH) solution at pH 12, washed, neutralized, and then bleached using hot hydrogen peroxide solution. The purified cellulose was then used for the preparation of NFC by re-dispersing in double distilled water to a final concentration of 1% (w/w) and then passing it through a high pressure homogenizer (APV-2000, SPX Co., Charlotte, NC, USA) at a pressure of 500 bar and 20 passes at room temperature (25 °C). The NFC sample had fiber diameters of around 57 nm with several micrometers in length.

2.3 Emulsion preparation

The aqueous phase was prepared by dispersing NFC at different concentrations (0.3%, 0.5%, and 0.7% [w/w]) in an aqueous buffer solution (10 mM potassium phosphate buffer, pH 7). NFC stabilized O/W emulsions were then prepared by blending 10% (w/w) oil each of coconut oil, soybean oil, and rice bran oil with 90% (w/w) aqueous NFC suspensions using a rotor-stator (Ultra Turrax T25, IKA Works, Inc., Wilmington, NC, USA) at 12,000 rpm for 2 min. These coarse emulsions were then passed through a two-stage high pressure homogenizer (APV-1000, SPX Co., Charlotte, NC, USA) at 300/30 bar and 3 passes to get fine emulsions. Sodium azide (NaN_3, 0.01% [w/w]) was added to the emulsions as an antimicrobial agent.

2.4 Particle size determination

The particle size and particle size distribution of freshly prepared O/W emulsions were measured using a laser diffraction particle size distribution analyzer (Mastersizer 3000, Malvern Instruments Ltd., Worcestershire, UK). To avoid multiple scattering effects, the emulsions were diluted with the same phosphate buffer as the continuous phase. Optical properties of the sample were defined as follows: refractive indices of oil and water were 1.46 and 1.33, respectively, and the absorption was assumed to be 0.

2.5 ζ-potential determination

The ζ-potential of NFC stabilized emulsions was determined using a particle electrophoresis instrument (Zetasizer Nano ZS, Malvern Instruments Ltd., Malvern, Worcestershire, UK). Prior to measurement, the emulsion samples were diluted using a phosphate buffer solution to prevent multiple scattering effects.

2.6 Color measurement

The fresh emulsions were measured for color in the L^*, a^*, and b^* system using a colorimeter (ColorFlex EZ, Hunter Associates Laboratory, Inc., Reston, VA, USA), where L^*, a^*, and b^* represent lightness, redness (+) to greenness (-), and yellowness (+) to blueness (-) respectively.

2.7 Scanning electron microscopy

The microstructure of the emulsions was visualized under a field emission scanning electron microscope (FE-SEM) (JSM-7610F, JEOL, Ltd., Oxford, UK).

2.8 Rheological measurement

Rheological properties of the emulsions were measured using a controlled-strain rheometer (HAAKE MARS 40 Rheometer, Thermo Scientific, GmbH, Germany) equipped with a cone and plate sensor (1° cone angle, 50 mm diameter, and 0.05 mm gap). For dynamic viscoelastic measurements, dynamic frequency sweep tests were conducted in the linear viscoelastic region by applying a constant strain of 0.5% within a frequency range of 0.1–100 rad/s. The steady flow tests were performed by continuously increasing the shear rate from 0.1–300 s^{-1} followed by a decrease from 300 s^{-1} to 0.1 s^{-1} in 6 min.

2.9 Creaming stability

The fresh emulsions were transferred to glass vials (20 mm diameter and 70 mm height), sealed with screw caps. The emulsions were kept in an air conditioned storage room and the temperature was controlled at 25 °C for 90 days. Visual assessment of creaming was then carried out to measure the phase separation of emulsions into a cream phase on top and a serum phase at the bottom. The extent of creaming was then reported as creaming index (*CI*) which is defined by the following equation:

$$CI(\%) = \frac{H_s}{H_T} \times 100$$

where H_T is the total height of the emulsions and H_S is the serum phase height of the emulsions.

2.10 Statistical analysis

All measurements were performed in triplicates. The results are expressed as mean±standard deviation. A two-way analysis of variance (ANOVA) and Duncan's multiple range test were used to indicate the significant differences ($P \leq 0.05$) among the mean values. The statistical analysis used SPSS version 18.0 Windows program (SPSS Inc., Chicago, IL, USA).

3. Results and Discussion

3.1 Particle size distribution

The droplet size and size distribution of the emulsion droplets following the homogenization process helps to determine the stability and final appearance of the emulsion product. Typically, stable emulsions are known to exhibit a homogeneous droplet distribution and their stability is also known to be influenced by particle size. Small droplets produced during the homogenization process can help to retard gravitational separation. Hence, it is possible to reduce the rate of droplet aggregation, flocculation, and coalescence by decreasing the oil droplet size (Degner, Chung, Schlegel, Hutkins, & McClements, 2014; Mikulcová, Bordes, & Kašpárková, 2016). The influence of different oil types (coconut, soybean, and rice bran) and NFC concentrations (0.3%, 0.5%, and 0.7% [w/w]) on the particle size distribution of 10% O/W emulsions is shown in Figure 1A. Bimodal distributions with

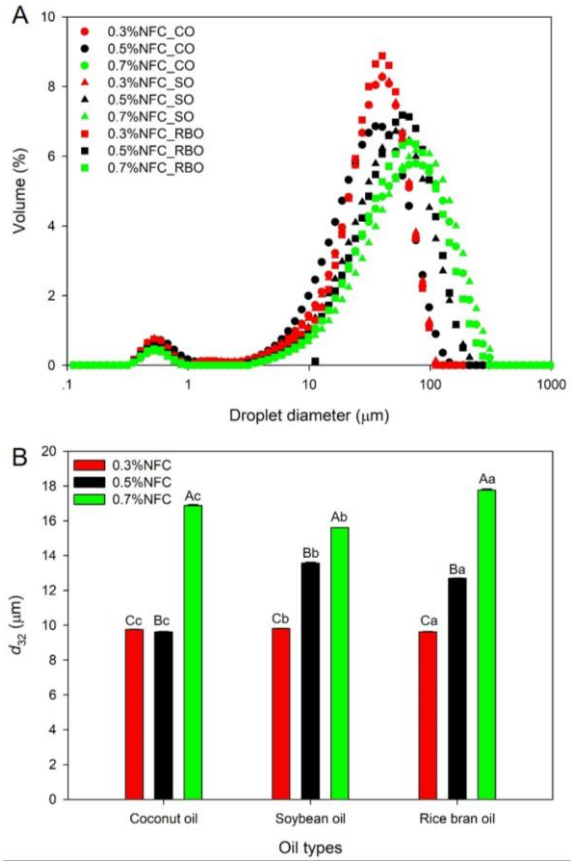

Figure 1. Influence of oil type and NFC concentration on particle size distribution and surface-weighted mean particle diameter (d_{32}) of 10% O/W emulsions stabilized by NFC. (Capital letters indicate significant difference among NFC concentrations and lowercase letters indicate significant difference among the oil types within the NFC concentration.)

two peaks were observed for all NFC stabilized emulsions and the average droplet diameter ranged from 10 to 100 μm which is a typical size range of conventional emulsions (McClements & Li, 2010). Further, the surface-weighted mean particle diameters (d_{32}) of the NFC-stabilized emulsions are presented in Figure 1B. A significant increase in particle size (d_{32}) was observed for all emulsions as the NFC concentration increased from 0.3% to 0.7% (w/w). The d_{32} values increased from 9.75 to 16.87 μm for the coconut oil emulsions and from 9.81 to 15.60 μm and 9.62 to 17.17 μm, for the soybean oil and rice bran oil emulsions, respectively. The results also revealed that the particle size distribution and d_{32} values were influenced by the NFC concentration rather than the type of oil. This can be attributed to the fact that NFC exhibits a gel like structure due to hydrogen bonding between the nanofibrils. Therefore, the NFC strongly affects the viscosity of the continuous phase (Missoum, Belgacem, & Bras, 2013; Winuprasith & Suphantharika, 2015). When the NFC concentration was low (0.3% [w/w]), the emulsion droplet size was the smallest which increased with increasing concentration with the largest particle size at 0.7% (w/w) having a broad distribution peak. Since, NFC increases the viscosity of the continuous phase, it can decrease the homogenization efficiency during the emulsification process. Therefore, a higher concentration of NFC for emulsion stabilization can result in a larger emulsion droplet size. It has been reported by Hayati, Man, Tan, and Aini (2009) that the presence of a polysaccharide can lead to an increased droplet size due to the ability to suppress the formation of small eddies during turbulence flow of the homogenization process. The results obtained were also in accordance with Ni *et al*. (2016) who stated that emulsion droplet size increased and their uniformity reduced when the concentration of konjac glucomannan increased. This occurred because of the higher viscosity of the continuous phase by konjac glucomannan that resulted in an inhomogeneous emulsified effect during emulsification. On the other hand, droplet size and distribution were not found to be affected by the type of oil. It was reported that the size of emulsion droplets were mainly influenced by emulsifier type, emulsifier concentration, homogenizer type, homogenization condition, and the physicochemical properties of the component phase (McClements, 2005). In addition, Winuprasith *et al.* (2018) demonstrated that whey protein isolate was a much more effective emulsifier than NFC at producing small droplets during homogenization which can be attributed to differences in the molecular and physicochemical properties of the two types of emulsifier.

3.2 ζ-potential

ζ-potential represents the surface charge of the oil droplets which is expressed in terms of the electrokinetic potential of the colloidal systems that helps to determine emulsion stability. A higher magnitude of ζ-potential indicates higher electrostatic repulsion between the oil droplets that provides a greater stability to the emulsion systems (Mitri, Shegokar, Gohla, Anselmi, & Müller, 2011). The ζ-potential value of all NFC stabilized emulsions indicated that NFC accumulated around the oil droplets (Figure 2). A previous study by Winuprasith and Suphantharika (2013) reported that NFC is an anionic polysaccharide that exhibits a negative charge which influences the ζ-potential of emulsion systems because a negative charge leads to a repulsive force that helps prevent droplet coalescence.

From the results obtained, it was also observed that the higher NFC concentrations resulted in larger magnitudes of ζ-potential values that indicated higher stability against coalescence. The highest emulsion stability was observed for the soybean oil emulsions containing 0.7% (w/w) NFC (−35. 92 mV) and the least emulsion stability was observed for the coconut oil emulsions containing 0.3% (w/w) NFC (−27.40 mV). Similar to the particle size data, it was observed that the oil type did not significantly affect the ζ-potential values of the prepared emulsions. According to the DLVO (Derjaguin-Landau-Verwey-Overbeek) theory, an emulsion system with a ζ-potential of 30 mV with either a positive or negative charge is considered a suitable value for providing emulsion stability (Sharma, Shukla, Misra, & Mishra, 2014). Hence, it can be concluded that NFC stabilized emulsions were stable to coalescence and at least 0.5% (w/w) NFC is required for stabilizing the emulsions effectively.

3.3 Color

The colors of the emulsions stabilized by NFC at different concentrations and with different types of oil are shown in Table 2 in the form of *L**, *a**, and *b** tristimulus values. All emulsions were found to exhibit an opaque appearance that can be attributed to the fact that their dimensions

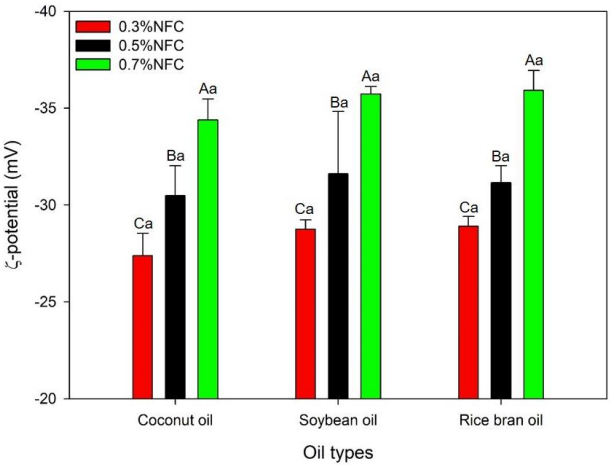

Figure 2. Influence of oil type and NFC concentration on the ζ-potential of 10% O/W emulsions stabilized by NFC. (Capital letters indicate significant difference among the NFC concentrations and lowercase letters indicate significant difference among oil types within the NFC concentration.)

Table 1. Fatty acid composition of coconut oil, soybean oil, and rice bran oil.

Oil type	Fatty acid composition (per 100 g)		
	Saturated fatty acid	Monounsaturated fatty acid	Polyunsaturated fatty acid
Coconut oil	92.86	7.14	0.00
Soybean oil	14.29	21.43	64.29
Rice bran oil	17.86	44.29	32.86

Table 2. Influence of oil type and NFC concentration on the color (L^*, a^*, and b^*) of 10% O/W emulsions stabilized by NFC.

Sample		L^*	a^*	b^*
NFC concentration	Oil type			
0.3%	coconut oil	59.19 ± 0.10^{Ab}	5.08 ± 0.02^{Ca}	15.50 ± 0.06^{Ca}
	soybean oil	61.55 ± 0.36^{Aa}	4.51 ± 0.06^{Cc}	14.44 ± 0.02^{Cc}
	rice bran oil	61.44 ± 0.09^{Aa}	4.62 ± 0.03^{Cb}	14.52 ± 0.04^{Cb}
0.5%	coconut oil	56.57 ± 0.09^{Bb}	5.79 ± 0.03^{Ba}	17.12 ± 0.05^{Ba}
	soybean oil	59.50 ± 0.13^{Ba}	5.19 ± 0.01^{Bc}	15.89 ± 0.07^{Bc}
	rice bran oil	59.11 ± 0.31^{Ba}	5.27 ± 0.05^{Bb}	16.03 ± 0.14^{Bb}
0.7%	coconut oil	53.74 ± 0.05^{Cb}	6.41 ± 0.01^{Aa}	18.55 ± 0.03^{Aa}
	soybean oil	57.85 ± 0.13^{Ca}	5.46 ± 0.04^{Ac}	16.59 ± 0.03^{Ac}
	rice bran oil	58.23 ± 0.27^{Ca}	5.46 ± 0.05^{Ab}	16.73 ± 0.10^{Ab}

Results are presented as mean±SD. In the same column, the values with the different letters were significantly different from the others ($P \leq 0.05$). Capital letters indicate significant difference among the NFC concentrations and lowercase letters indicate significant difference among oil types within the NFC concentrations.

were in the micrometer scale which is of the same order as the wavelength of light since the light scattering was relatively strong (McClements & Li, 2010). Also, since NFC itself exhibited a yellow-brown color (L^*=18.72±0.03, a^*=0.95±0.03, and b^*=1.44±0.02), an increase in the NFC concentration resulted in darker emulsions. The darkest emulsion was the coconut oil emulsion containing 0.7% (w/w) NFC (L^*=53.74±0.05, a^*=6.41±0.01, and b^*=18.55±0.03), whereas the lightest emulsion was the soybean oil emulsion containing 0.3% (w/w) NFC (L^*=61.55±0.36, a^*=4.51±0.06, and b^*=14.44±0.02) (Table 2).

According to Winuprasith and Suphantharika (2015), the color of the emulsions was affected by light scattering and absorption which in turn was influenced by the emulsion droplet diameter and concentration of the emulsifier (NFC). From our results, it can be observed that the color parameters of the emulsions were in agreement with the droplet size data where higher NFC concentrations led to lower lightness values due to their larger droplets (Figure 1) which tended to absorb more light than the small droplets due to their decreased light scattering efficiency. Further, the emulsions with different oil types were found to have significantly different color parameters; however, these differences were not noticeable by visual observation.

3.4 Microstructure

The SEM micrographs of the NFC-stabilized emulsions are shown in Figure3. From the micrographs, it is easily noticeable that the NFC stabilized the emulsions by adsorbing at the oil-water interface. The NFC can cause aggregation of oil droplets by trapping them in the NFC network; however, it was observed that NFC provided a thick layer at the interface that prevented the oil droplets from coalescence. Moreover, the NFC was found in the continuous phase where it formed a solid three-dimensional network. This network stabilization and the adsorption of NFC at the interface resulted in forming Pickering emulsions. Further, entanglement between the nanofibers in the continuous phase increased the viscosity of the emulsion system which in turn retarded droplet movement, thereby reducing the rate of phase separation. An increase in the NFC concentration resulted in higher accumulation of NFC at the oil surface and the formation of a more solid NFC network in the continuous phase. From the micrographs, it can also be clearly seen that the emulsions formed using coconut oil exhibited a different morphology in comparison to the emulsions formed using soybean oil or rice bran oil. This was possibly due to the fact that coconut oil is an oil that contains more saturated fatty acids than the other oils. The fat crystal of coconut oil can be present below their melting point (23–26 °C) (Firestone, 2006) which can penetrate through the interfacial membranes surrounding the droplets since the oil droplets collapse as seen in Figures 3A & 3D. This phenomenon may occur during sample preparation for FE-SEM measurement or during storage.

3.5 Rheological properties

The rheological properties are important characteristics of emulsions because they help determine their appearance, sensory attributes, processing conditions, and stability. They are known to be influenced by different factors such as volume fraction of the dispersed phase, droplet size, interacttion between emulsion droplets, and the structure of the oil-water interface (Carrillo *et al.*, 2015; Zou, Yang, & Scholten, 2018). The rheological properties (viscoelastic and flow behavior) of the NFC-stabilized emulsions are presented in Figure 4. It was observed that the emulsions containing different NFC concentrations and different types of oil exhibited G' (storage modulus) higher than G'' (loss modulus) without any crossing over for the entire frequency range tested (Figure 5A). This phenomenon indicated that all emulsions exhibited a typical gel-like behavior. Moreover, the emulsions stabilized by NFC at a concentration of 0.7% (w/w) exhibited the strongest emulsion structure which was indicated by their high G' values whereas the emulsions stabilized by NFC at a concentration of 0.3% (w/w) exhibited the weakest structure. The increase in G' value with an increasing NFC concentration indicated an increased strength in the emulsion structure and the emulsions to exhibit a more solid-like behavior. Similar re-

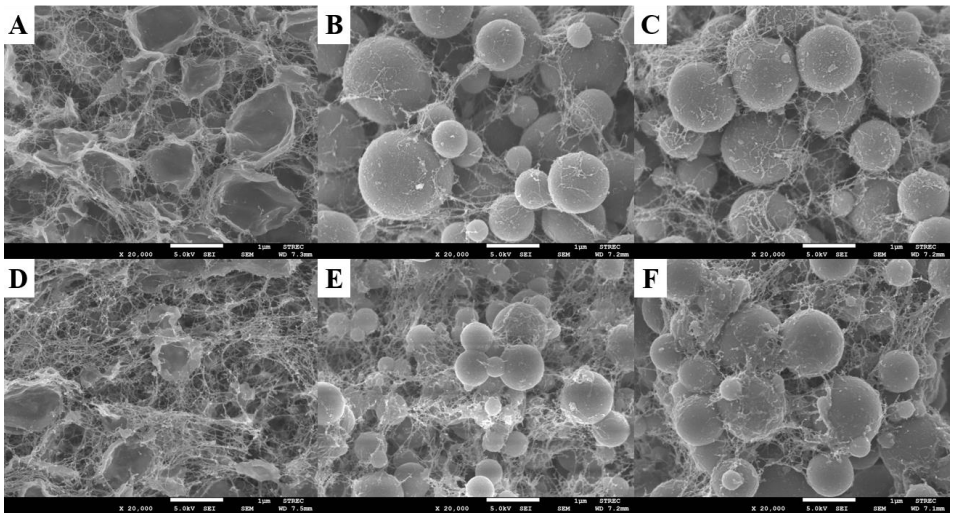

Figure 3. FE-SEM micrographs of 10% O/W emulsion stabilized by NFC at various concentrations with different oil types: (A) 0.3% NFC with coconut oil; (B) 0.3% NFC with soybean oil; (C) 0.3% NFC with rice bran oil; (D) 0.7% NFC with coconut oil; (E) 0.7% NFC with soybean oil; and (F) 0.7% NFC with rice bran oil.

sults have also been reported by other studies where the gel-like behavior and the solid-like behavior of the emulsions was dominated by the cellulose concentration as cellulose is involved in the formation of a network in the continuous phase which becomes more compact and stronger with an increasing cellulose concentration (Li *et al.*, 2018; Lu, Zhang, Li, & Huang, 2018; Xiao, Wang, Gonzalez, & Huang, 2016; Zou *et al.*, 2018).

For the flow behavior, all the emulsions exhibited a shear-thinning behavior where there was a decrease in their apparent viscosity as the shear rate increased (Figure 5B). Also, an increase in the NFC concentration was found to increase the viscosity of the emulsion system. This is because excessive NFC adsorbs around the oil droplets located in the continuous phase and forms a three-dimensional network that provides a greater solid-like characteristic and increased emulsion stability. Another study also reported the shear thinning behavior of multiple emulsions stabilized by cellulose nanofibrils (Carrillo *et al.*, 2015). Our results were also in agreement with other studies that utilized cellulose as a Pickering emulsifier and found that it significantly increased the viscosity of the continuous phase and formed a polymer network. Hence, the stability of an emulsion containing cellulose depends mainly on the cellulose concentration (Li *et al.*, 2018; Paukkon, Ukkonen, Szilvay, Yilperttula, & Laaksonen, 2017; Sun, Sun, Wei, Liu, & Zhang, 2007). Hence, it can be stated that the major factor influencing emulsion strength was the NFC concentration rather than the type of oil that did not much affect the viscoelastic properties and flow behavior.

3.6 Creaming index

Creaming index helps to determine phase separation in the emulsions caused by gravitational forces which gives an idea of the emulsion creaming behavior. Figure 4 shows the creaming behavior of O/W emulsions stabilized by NFC using different types of oil. At a low NFC concentration (0.3% [w/w]), the emulsions with different types of oil began to cream from the first measurement time point at 3 h and

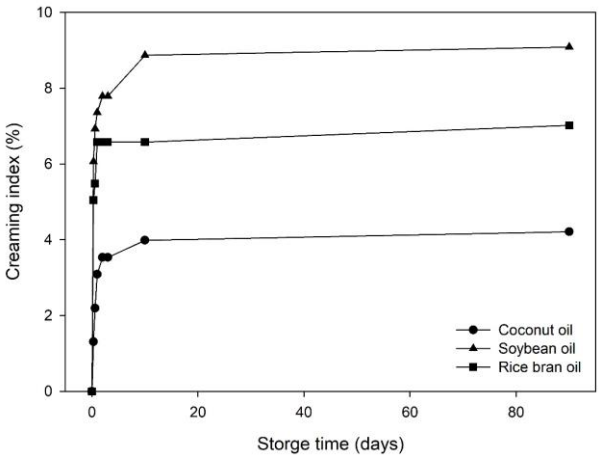

Figure 4. Influence of oil type on the creaming index of 10% O/W emulsions at a NFC concentration of 0.3% (w/w).

creamed readily within 24 h of emulsion preparation. These emulsions reached their final extent of creaming at day 20 after which no further creaming was observed until the end of the storage period of 90 days. Although 0.3% (w/w) NFC stabilized emulsions were found to exhibit the lowest droplet size, they were still prone to phase separation due to their low viscosity and low repulsive forces which caused the droplets to come close together and coalesce. On the contrary, the emulsions stabilized by NFC at concentrations of 0.5% and 0.7% (w/w) using the three different oil types did not cream throughout the entire storage period of 90 days (data not shown) because an increased concentration of NFC in the continuous phase caused an increase in viscosity which retarded droplet movement. Therefore, a high NFC concentration (≥ 0.5% [w/w]) is needed to efficiently emulsify and stabilize the emulsion system. Likewise, Li *et al.* (2018) also reported that regenerated cellulose at a concentration above 0.5% (w/w) led to the production of more stable O/W emulsions because the adsorption of regenerated cellulose at the oil droplet surface

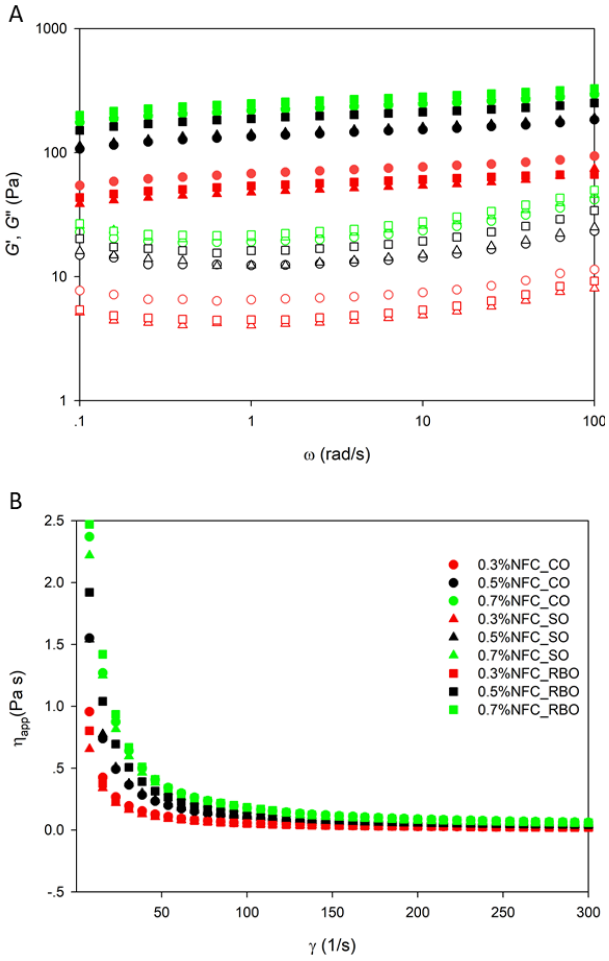

Figure 5. Influence of oil type and NFC concentration on the rheological properties of 10% O/W emulsions stabilized by NFC: (A) mechanical spectra − storage modulus, *G*', (closed symbols) and loss modulus, *G*", (open symbols) as a function of angular frequency; (B) apparent viscosity as a function of shear rate. All measurements were performed at 25 °C.

minimizes the density differences between the oil and continuous phase which helps improve the emulsion stability. From our results, it is clearly visible that emulsion stability was mainly influenced by the NFC concentration. Although emulsions stabilized by 0.3% (w/w) NFC with different types of oil had different creaming index values during the initial storage period, an increase in the NFC concentration resulted in a significant increase in the stability of the emulsions against phase separation.

4. Conclusions

The properties of emulsions were found to be influenced by the NFC concentration rather than the type of oil. NFC is a polysaccharide obtained by fibrillation of cellulose that adsorbs at the oil-water interface and entraps the oil droplets in a spherical shape. They are known to stabilize the emulsions by forming a steric barrier of NFC fibers that adsorb around the oil droplets and provide electrostatic repulsion against droplet coalescence. This was confirmed by the increasing ζ-potential value, viscosity, and viscoelastic properties with increasing NFC concentration. The formation of a three-dimensional network of NFC in the continuous phase could help to reduce the mobility of the oil droplets which retarded droplet coalescence and phase separation due to gravitational forces. In addition, it was observed that at least 0.5% (w/w) of NFC concentration was required to effectively stabilize the emulsions. However, it was clearly seen that the increase in NFC concentration affected the homogenization efficiency because of larger oil droplet formation. Nevertheless, this is beneficial as it is known that a large droplet size provides the emulsions with the capacity to load high concentration of lipophilic active ingredients, nutrients, or drugs for encapsulation applications. It can be suggested that nanofibrillated mangosteen cellulose is a useful emulsifier and stabilizer for producing and stabilizing O/W emulsions. Also, since NFC is a plant-based natural Pickering emulsifier, it is biodegradable and has low toxicity. Furthermore, it can be used in different food, pharmaceutical, and environmental applications such as encapsulation of bioactive ingredients or nutrients, bio-based drug delivery systems, and scaffold materials for wastewater management.

Acknowledgements

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