Research Article

Progress in Drug Discovery & Biomedical Science

HH PUBLISHER

Factors Affecting the Stability and Physical Properties of Pickering Emulsions Stabilized by Fe_3O_4@CNC Nanocomposites

Liang Ee Low[1,2], Boon Hoong Ong[3], Beng Ti Tey[4,5], Siah Ying Tang[4,5,*]

[1]Institute of Pharmaceutics, College of Pharmaceutical Sciences, Zhejiang University, 866 Yuhangtang Road, Hangzhou 310058, P. R. China.
[2]Key Laboratory of Biomedical Engineering of the Ministry of Education, College of Biomedical Engineering & Instrument Science, Zhejiang University, Hangzhou 310058, P. R. China.
[3]Nanotechnology & Catalysis Research Centre, University of Malaya, Jalan Universiti, 50603 Kuala Lumpur, Wilayah Persekutuan Kuala Lumpur, Malaysia.
[4]Chemical Engineering Discipline, School of Engineering, Monash University Malaysia, Jalan Lagoon Selatan, Bandar Sunway, 47500 Subang Jaya, Selangor, Malaysia.
[5]Advanced Engineering Platform, Monash University Malaysia, Jalan Lagoon Selatan, Bandar Sunway, 47500 Subang Jaya, Selangor, Malaysia.

Abstract: Particle-stabilized emulsion systems, namely Pickering emulsions have recently emerged as novel yet attractive dosage form for controlled delivery of biologically active compounds. The stability and properties of emulsions are influenced by formulation and processing factors. In this study, oil-in-water Pickering emulsions were prepared using Fe_3O_4@cellulose nanocrystal (MCNC) nanocomposites and the effects of CNC/Fe_3O_4 (CNC/MNP) ratios, MCNC particle concentration C_{mcnc}, oil volume fraction φ_{oil} and ionic strength on the colloidal stability were evaluated. The results showed that stable emulsions could be attained using MCNC particles with CNC/MNP ratio ≤ 1. Increasing CNC/MNP ratio > 1 resulted in different degree of phase separation of emulsions. The average droplet size of MCNC-stabilized Pickering emulsions (MCNC-PE) decreased from 17.34 to 3.58 µm with an increase in C_{mcnc} as a result of improved coalescence stability due to higher droplet surface coverage S_{mcnc} by MCNC particles. An increase in φ_{oil} led to pronounced increase in droplet size from 2.82 to 17.00 µm. Interestingly, the ionic strength showed little or no impact on the emulsion droplet size and creaming. The storage stability study revealed that most emulsions remained fairly stable with no apparent change in droplet size. In conclusion, the physical stability of MCNC-PE was found to be greatly influenced by particulate emulsifier concentration and oil loadings. These two parameters must be well controlled in the development of Pickering emulsions for potential food and pharmaceutical applications.

Keywords: Pickering emulsions; CNC/Fe_3O_4 ratio; particle concentration; oil loading; surface coverage; ionic strength.

***Correspondence**: Siah Ying Tang, School of Engineering, Monash University Malaysia, Jalan Lagoon Selatan, Bandar Sunway, Subang Jaya, Selangor, Malaysia; patrick.tang@monash.edu

Received: 10th March 2020
Accepted: 12th April 2020
Published Online: 21th April 2020

Citation: Low, L. E., Ong, B. H., Tey B. T., & Tang, S.Y. Factors affecting the stability and physical properties of pickering emulsions stabilized by Fe_3O_4@cnc nanocomposites. Prog Drug Discov Biomed Sci 2020; 3(1): a0000073. https://doi.org/10.3687/pddbs.a0000073

Highlights:

Stability of Pickering emulsions can be influenced by MCNC loading with different CNC/MNP ratios.

Formation of stable emulsions was achieved using MCNC particles with CNC/MNP ratio ≤ 1

Increasing MCNC loading resulted in smaller droplets with comparable emulsion stability.

The oil volume fraction has significant impact on the emulsion droplet sizes.

Changes in ionic strength exhibited negligible impact on emulsion stability.

Introduction

An emulsion is a system consisting two immiscible liquids where one of the liquids is dispersed in the other. Emulsion system without a stabilizer is thermodynamically unstable and tends to experience serious inter-droplet coalescence and creaming[1–3]. Thus, the stabilizer has been an important ingredient

for the preparation of stable emulsions that resist coalescence. Conventionally, emulsions are stabilized by chemical surfactants. However, excessive usage of surfactants often results in drawbacks including cost, toxicity, and tissue irritation[4]. To address such issues, particle-stabilized emulsions or Pickering emulsions with surfactant-free nature has been proposed[5–8]. Pickering stabilization occurs through the irreversible attachment of partial wettable particles onto the interface between the two immiscible liquids, which reduces the interfacial energy, a dominant driving force for the emulsion deformation[9–10]. Since its introduction by Ramsden[11] and Pickering[12], Pickering emulsions have been receiving substantial attention by academic and industrial researchers owing to their lower cost, low-toxicity and astonishing stability against coalescence compared to those stabilized by surfactants[11–14]. To date, Pickering emulsion can be considered as one of the researchers-preferred topics that have been extensively studied in the food[15–17], biomedical[4,18–19] and pharmaceutical sectors[13,20–21].

With the increasing awareness in sustainability, "green" bio-based or hybrid nano-/micro-particles have received growing interest for Pickering emulsion preparation. Amongst, cellulose nanocrystal (CNC) that is biocompatible, biodegradable, non-toxic and sustainable can be recognized as a promising candidate for Pickering stabilization[22–24]. CNCs were previously confirmed to yield highly stable Pickering emulsion[23–26]. Additionally, the reactive functional group-rich nature of CNC further inspired researchers to incorporate stimuli-responsive ligands to CNC to extend its usage in biomedical fields[4,27–28]. In past decades, various stimuli-responsiveness, namely thermal-, fluorescent-, pH- and magnetic- responsive materials have been reported for preparing CNC-based stimuli-controllable smart nanomaterials[27–30]. In fact, our research group has also prepared the Pickering emulsion stabilized by Fe_3O_4@CNC (MCNC) nanocomposites previously, and demonstrated its potential as drug delivery carrier for colon cancer therapy[29–31].

Despite the intensive efforts made on MCNC-stabilized Pickering emulsion (MCNC-PE), the influence of the compositional parameters and ionic strength on the properties and stability of MCNC-PE have not been systematically investigated. Herein, the current work aimed to study the impacts of CNC/Fe_3O_4 (CNCN/MNP) ratio, MCNC particle concentration C_{mcnc} and oil volume fraction, φ_{oil} on the characteristics and physical stability of MCNC-PE. As part of this study, the effect of physiological variable i.e. ionic strength on the physical and creaming stability of the Pickering emulsion were also evaluated. The findings of this parametric study provided further knowledge and understanding into the key factors affecting the colloidal stability and properties of magnetic Pickering emulsions stabilized by MCNC particles, which could be beneficial for the future formulation development of colloidal drug delivery platform for controlled delivery of therapeutic compounds.

Materials and Methods

Materials

Iron (II) chloride tetrahydrate ($FeCl_2.4H_2O$, ≥ 99%), iron (III) chloride hexahydrate ($FeCl_3.6H_2O$, 99%), ammonium hydroxide (28% NH_3 in H_2O), calcofluor white (for microbiology) and potassium hydroxide were procured from Sigma-Aldrich Chemicals Company (Malaysia). CNC (freeze dried, 0.96 wt% sulphur content) was obtained from the University of Maine. Red palm superolein (275 ppm β-carotene, melting point 19°C,) was purchased from Sime Darby Jomalina Sdn Bhd (Malaysia). All water used in this experiment is ultrapure and obtained from Milli-Q® Plus apparatus (Millipore, Billerica, USA). Ethanol (AR standard), Hydrochloric acid (HCl, 1 M), and sodium hydroxide (NaOH) were received from R & M Chemical (Syarikat Saintifik Jaya, Malaysia). All chemicals used in this study are of analytical grade.

Synthesis of MCNC nanocomposites

MCNC composites were prepared using the ultrasound-assisted in situ co-precipitation method[32]. First, CNCs with varying concentration (0.01, 0.05, 0.1, 0.5 and 1 wt%) were dispersed in 80 ml of water under sonication for 2 minutes. Subsequently, 0.054 g of iron (III) and 0.027 g iron (II) chloride (1.5/ 1 Fe^{3+}/ Fe^{2+} mol ratio) were added to the CNC dispersion to obtain a CNC/MNP ratio of 0.2, 1, 2, 10 and 20, respectively. The mixtures were then stirred and heated to 45°C. Next, the mixtures were treated with ultrasound (60 w, 5 min) in the presence of ammonium hydroxide (2.2 ml) to yield the MCNC nanocomposites. The resultant MCNCs were then precipitated, magnetically separated and washed 3 times with ethanol to remove all residual ammonium hydroxide. The recovered MCNCs were centrifuged at 4500 rpm for 8 minutes, followed by drying in an oven for 24 hours, prior to its storage characterization. The prepared MCNC samples with different CNC/MNP ratio are denoted as MCNC02, MCNC1, MCNC2, MCNC10 and MCNC20.

Characterization of MCNC nanocomposites

The size and surface morphology of the MCNC composites were analyzed by Hitachi SU8010 field emission scanning electron microscope (FE-SEM) (Hitachi, Japan) via scanning transmission electron microscopy (STEM) mode at 15 kV. Available functional groups were examined using a Fourier transform infrared (FTIR) spectrophotometer equipped with a diamond probe (Nicolet iS10, Thermo Scientific, USA) in a frequency range of 550–4000 cm^{-1}. The magnetization of MCNC was measured via vibrating sample magnetometry (VSM) (Lakeshore 7400 Series). The thermal stability was examined using a thermogravimetric analyzer (Q50 TGA, TA instrument, USA) over a temperature range of 25 to 900°C at 10°C/min.

Preparation of MCNC-PE

The MCNC-PE was prepared using the as-synthesized MCNC nanocomposites as the stabilizers and β-carotene

rich palm olein as the oil phase. All emulsion samples were transferred to glass vials and stored at room temperature for further characterization. The preparation procedure of MCNC-PE with different formulations is as follow.

MCNC-PE with different CNC/MNP ratio

Emulsions with a fixed φ_{oil} (0.3) and a fixed C_{mcnc} (0.1 wt%) were prepared by using MCNC with different CNC to MNP ratio (0.2, 1, 2, 10, 20). The mixture was emulsified for 3 minutes using an ultrasound probe sonicator (20 kHz, Lab750, NexTgen ultrasonic platform, Sinaptec, France) under pulse mode (15 s pulse on, 10 s pulse off).

MCNC-PE with various MCNC particle concentrations

Emulsions were produced using the MCNC1 sample under a fixed oil content (φ_{oil} = 0.3) with varying aqueous suspension of different C_{mcnc} (0.025, 0.050, 0.100, 0.200, and 0.300 wt%). The mixture was then emulsified for 3 minutes using pulse mode sonication.

MCNC-PE with different oil volume fraction

Emulsions with various φ_{oil} were prepared by combining C_{mcnc} 0.1 wt% (MCNC1 sample) and different oil volume fractions (φ_{oil} = 0.1, 0.2, 0.3, 0.4, and 0.5). The mixture was emulsified for 3 minutes under pulse-mode sonication.

Effects of NaCl on MCNC-PE stability

The effect of salt on the properties of MCNC-PE was evaluated by adding various quantities of NaCl (0, 100, 200, 300, 400, 500 mM) into the freshly made Pickering emulsions with the following formulation (φ_{oil} = 0.3, C_{mcnc} = 0.1 wt%, MCNC1 sample). The MCNC-PEs were stirred at 200 rpm for 5 min before storage.

Characterization of MCNC-PE

The hydrodynamic diameter of the MCNC-PE was measured via laser diffraction using a Mastersizer (Mastersizer 3000, Malvern Instruments, UK) equipped with a Hydro EV wet dispersion unit. The images of MCNC-PE were obtained using an inverted fluorescent/optical microscope (Nikon Eclipse TS100, Nikon Instruments Inc., USA) at 10x magnification. The adsorption of MCNC particle at the oil/water interface was determined by chemically staining a representative emulsion sample (MCNC-PE stabilized by 0.05 wt% MCNC particles) with calcofluor white prior to the fluorescence microscopy. The staining procedure was carried out as described in our previous work[29].

Storage stability of MCNC-PE

The MCNC-PEs were stored at room temperature (25°C) for 14 days. The stability of emulsions against coalescence and creaming was monitored after 0, 7, and 14 days of storage in term of droplet diameter and creaming profile via the standard laser diffraction and emulsion storage properties analysis[33].

Statistical analysis

Analysis of variance (ANOVA) were conducted using Prism software and $p < 0.05$ was considered as statistically significant.

Results and discussion

Characterization of MCNC nanocomposites

MCNC nanocomposites were synthesized using the previously developed method[32]. The STEM micrograph of MCNC1 sample revealed the uniform deposition of MNPs onto the rod-like CNCs (Figure 1a). The MCNC1 sample was observed to exhibited superparamagnetic properties with a saturation magnetization (M_s) of 28.31 emu/g (Figure 1b), comparable to those previously reported[34]. However, the FTIR curves showed that the characteristics C—OH bonding of CNCs were prominent in MCNC10 and MCNC20 samples (Figure 1c), suggesting the presence of abundant surface hydroxyl groups of CNCs as compared to the insufficient iron precursors for a complete reaction. The ash content of MCNC was found to decrease with increasing CNC concentration, which was primarily attributed to the decreased MNP content in MCNC nanocomposites as CNC/MNP ratio increased. Interestingly, the typical weight loss induced by the removal of MNP-bonded oxygen atoms (at approximately 780°C) only existed when the CNC/MNP ratio is ≤ 1 (Figure 1d)[34]. This demonstrated the decrease in the thermal stability of MNPs upon reaction with the hydroxyl groups of CNC. This is most likely owing to the crystalline structure of MNP in each MCNC sample, as reported by Kalska-Szostko *et al.* where supreme thermal stability was observed on monocrystalline MNP as compared to the less dense polycrystalline MNP[35]. Hence, the reduction in thermal stability in MCNC01, MCNC1, and MCNC2 simply indicated that a less dense crystalline structured MNP is presence in the mentioned samples while a monocrystalline structured MNPs are found in MCNC10 and MCNC20. Apart from that, the TGA curves of both MCNC02, and MCNC2 also revealed a lower second weight loss than the MCNC1. This phenomenon may be due to the presence of monocrystalline structured MNPs with a higher thermal stability[35]. Using the weight loss curve, the MNP content in the as-prepared MCNC nanocomposite are determined at approximately 80, 55, 45, 30 and 20 wt% for MCNC02, MCNC1, MCNC2, MCNC10 and MCNC20 respectively.

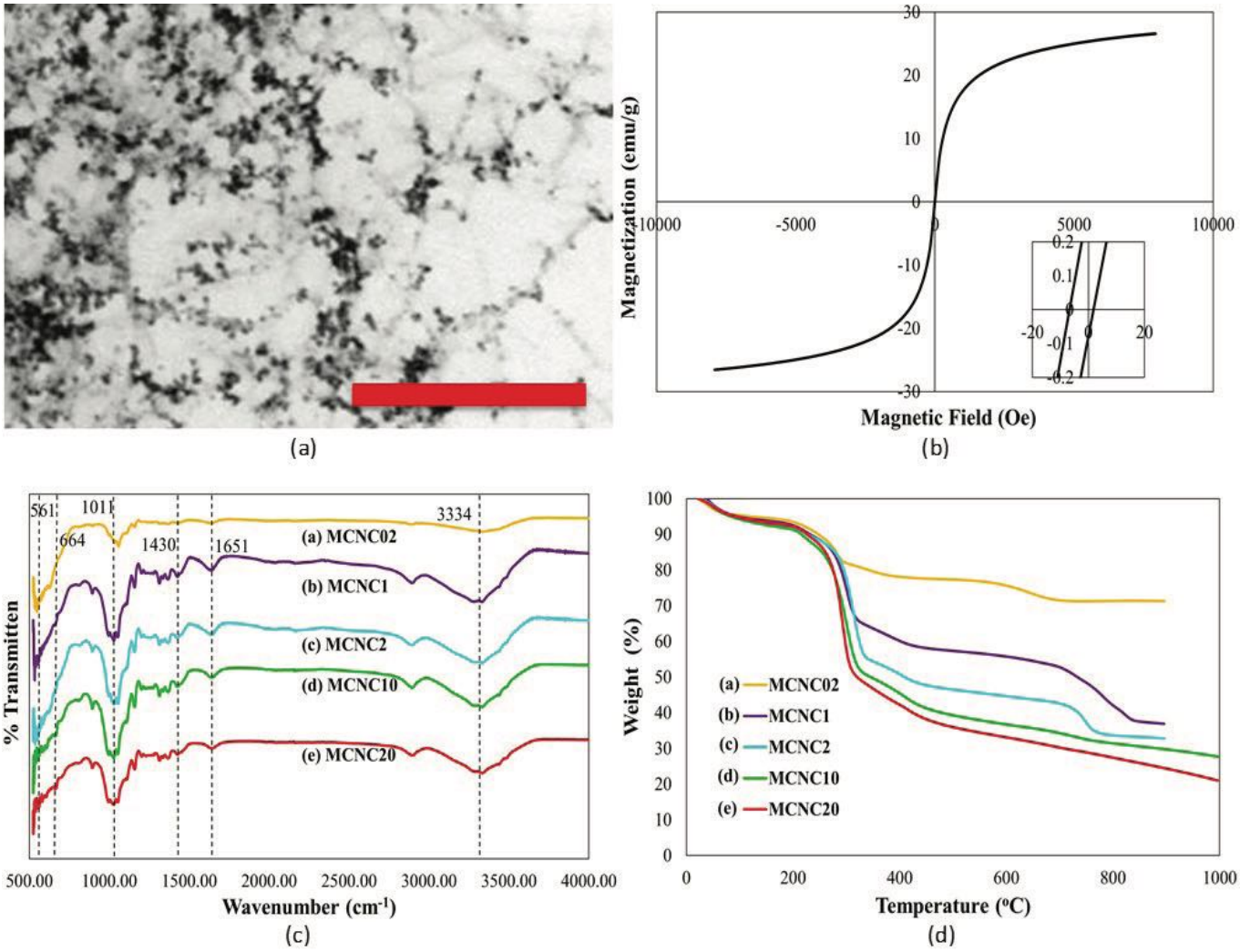

Figure 1. (a) FESEM image and (b) VSM data for MCNC1 sample. (c) FTIR spectra and (d) TGA data of MCNC nanocomposite with CNC/MNP ratio of (i) 0.2, (ii) 1, (iii) 2, (iv) 10 and (v) 20. Scale bar represents 200 nm.

Properties of MCNC-PE

Varying CNC/MNP ratio

MCNC nanocomposites with different CNC/MNP ratio were used to prepare the MCNC-PE. The laser diffraction results show the emulsions with monomodal-sized distribution when the CNC/MNP ratio was ≤ 1. Contrarily, bimodal distribution with multiple peaks was noticed at emulsion samples prepared using MCNC2, MCNC10 and MCNC20 (Figure 2a). Similar observation was noticed in the optical microscopy analysis (Figure 2c). Evidently, the optical images of Pickering emulsion stabilized by MCNC2, MCNC10 revealed emulsion droplets with a large size ranges, demonstrating the possible coalescence of MCNC-PE prepared using MCNC with CNC/MNP ratio of ≥ 2. This shows that the successful stabilization of MCNC-PE is only possible at CNC/MNP ratio is ≤ 1, and the finest emulsion droplet (≈ 7 µm) was obtained using MCNC1 as stabilizer (Figure 2b). This indicated that the MCNC1 is likely to be the most desired CNC/MNP ratio for the preparation of stable MCNC-PE. To verify the claim further, the emulsion samples were subjected to storage stability analysis to monitor the changes in size and creaming index. It was noticeable that only the size of emulsions fabricated using MCNC1 remained the same throughout the storage period at room temperature (Figure 2b). In term of creaming profile, the extent of droplet creaming was measured by the change in height of the bottom serum phase with storage time. The creaming index (CI) was determined according to Equation 1.

$$CI = (h_{total} - h_{emul}) / h_{total} \times 100\% \quad (1)$$

where CI represents the creaming index, is the height of emulsion, and is the total height of all solutions.

Based on Figures 3b and c, lowest CI was recorded at CNC/MNP ratio = 1 (Figure 3c). Since creaming is known to occurs as a result of the density difference between the dispersed (oil) and the continuous (water) phases, and larger oil globule usually experienced a greater influences

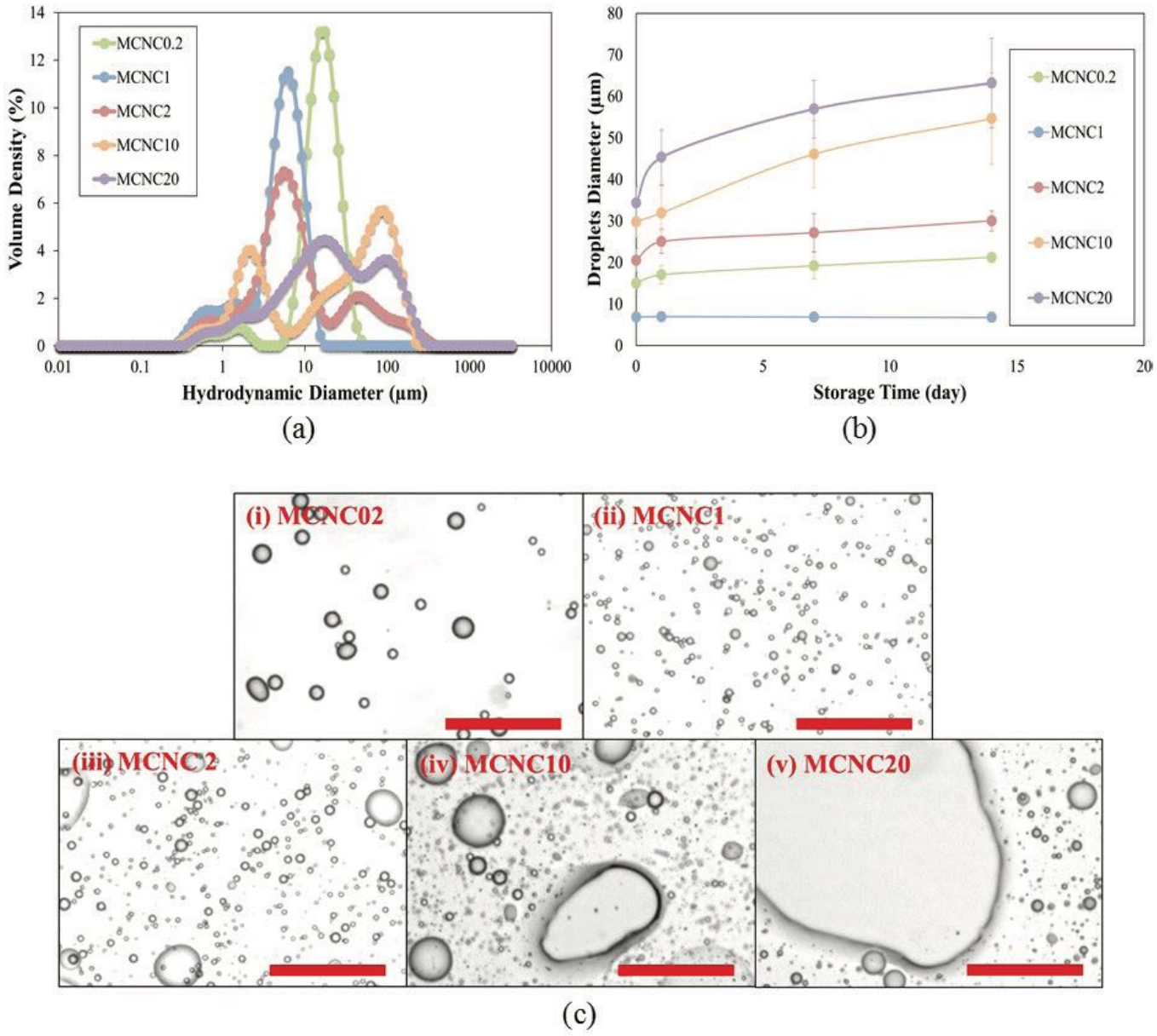

Figure 2. (a) Droplet size distributions, (b) emulsion diameter changes over 14 days and (c) microscopic images of MCNC-PE as a function of CNC/MNP ratio. All formulations contain a fixed C_{mcnc} of 0.1 wt% and φ_{oil} of 0.3. The standard error of the mean of triplicate readings was represented by the error bars in each graph, and different alphabetic letters were significantly different at $P \leq 0.05$ using Bonferroni's multiple comparison test. All scale bars represent 100 µm.

by the density difference between the continuous medium (water) and droplets (oil)[36], The latter outcome may be due to the fact that the emulsion coalescence is at minimal when MCNC-PE is prepared using MCNC1. The results confirmed the aforementioned claims and therefore, the MCNC1 will be focused for further experimentation.

MCNC particle concentration

The MCNC-PE was subsequently prepared using the φ_{oil} of 0.3 at varies C_{mcnc}. The fluorescence microscopy showed the locations of MCNCs (blue fluorescent) around the surface of oil droplets (Figure 4c[iii]). Based on Figure 4a, all MCNC-PE samples displayed monomodal-sized distributions regardless of the C_{mcnc}. This is in good agreement with the qualitative data obtained from optical microscopy where the captured emulsion droplets were of uniform spherical size (Figure 4c). The emulsion mean droplet diameter was observed to gradually decrease from 17.34 to 3.58 µm upon increment of C_{mcnc} from 0.025 to 0.300 wt% (Figure 4b).

This is mainly attributed to the increased surface coverage by MCNC particles around the emulsion droplets due to the increase in C_{mcnc}. The surface coverage S_{mcnc} of the emulsion droplets by MCNC was calculated as follow[24]:

$$S_{mcnc} = m_p D_{3,2} / 6hpV_{oil} \quad (2)$$

where m_p is the mass of MCNC, h is the MCNC thickness, ρ is the MCNC density, and V_{oil} is the volume of oil in the emulsion.

The calculated C_{mcnc} of MCNC-PE prepared at different are tabulated in Table 1.

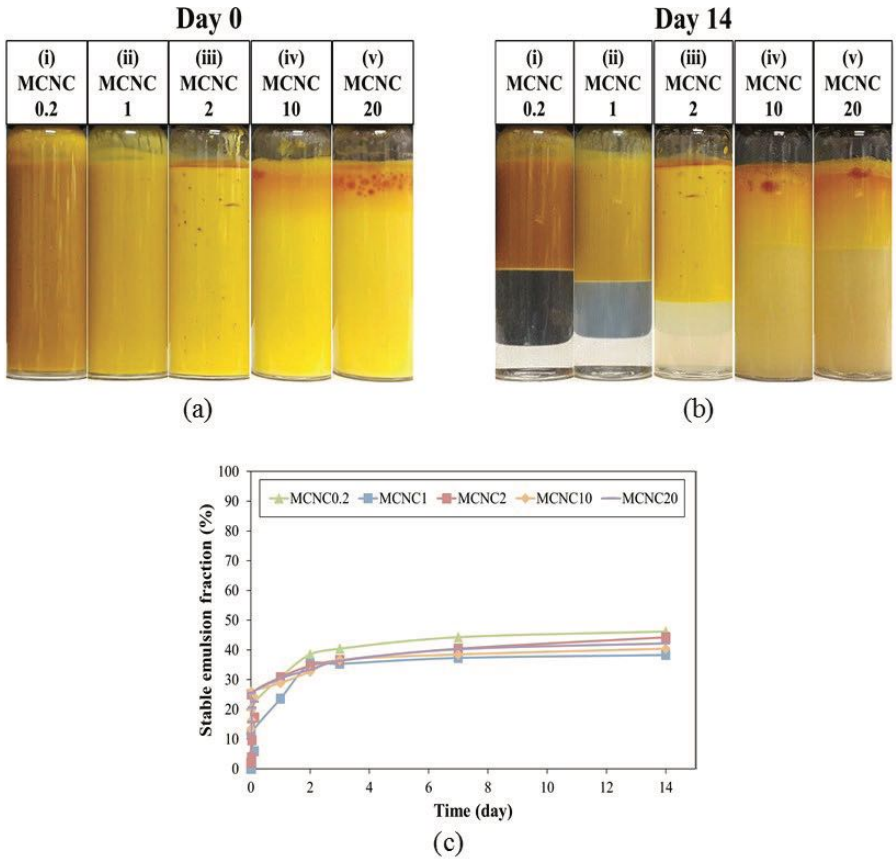

Figure 3. (a) Day 0 and (b) day 14 of photographs of MCNC-PE with CNC/MNP ratio of (i) 0.2, (ii) 1, (iii) 2, (iv) 10 and (v) 20 and (c) creaming indexes.

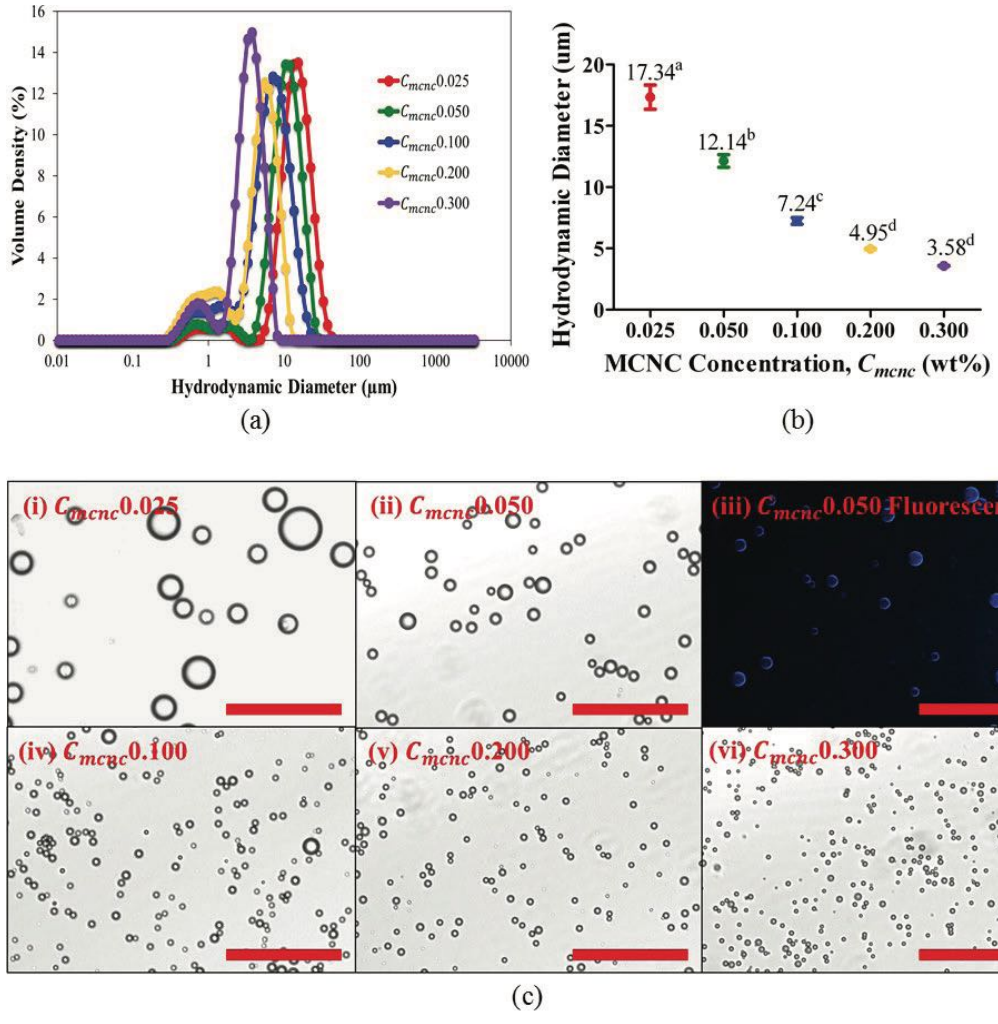

Figure 4. (a) Droplet size distributions, (b) mean droplet diameters and (c) microscopic images of MCNC-PE as a function of . All formulations contain fixed of 0.3. The standard error of mean of triplicate readings were represented by the error bars in each graph, and different alphabetic letters were significantly different at $P \leq 0.05$ by Bonferroni's Multiple Comparison Test. All scale bar represents 100 μm..

Table 1. Surface coverage, S_{mcnc} of O/W Pickering emulsion prepared under different MCNC Concentration, C_{mcnc}.

MCNC Concentration, (wt%) C_{mcnc}	Surface Coverage, (%) S_{mcnc}
0.025	8.496
0.050	11.896
0.100	15.028
0.200	19.410
0.300	21.025

As the C_{mcnc} grew from 0.025 to 0.300 wt%, the S_{mcnc} of the MCNC-PE markedly increased from 8.496 to 21.025%, thereby demonstrating that higher MCNC content leads to higher of the oil globules and thus finer emulsions with enhanced stability against coalescence. The MCNC-PE size changes were then recorded for storage duration of 14 days.

As shown in Figure 5, except for those prepared with 0.025 wt% of particles, the rest of the MCNC-PEs did not experienced change in the mean droplet diameter throughout the storage at room temperature. At day 1, a slight increase in emulsion diameter (17 to 20 μm) has been observed in those prepared by 0.025 wt% MCNC. This is because of the limiting coalescence where the oil globules coalesce to reduce the effective interfacial area between the oil and water phase, which renders a lower energy requirement to remain stable[1–2].

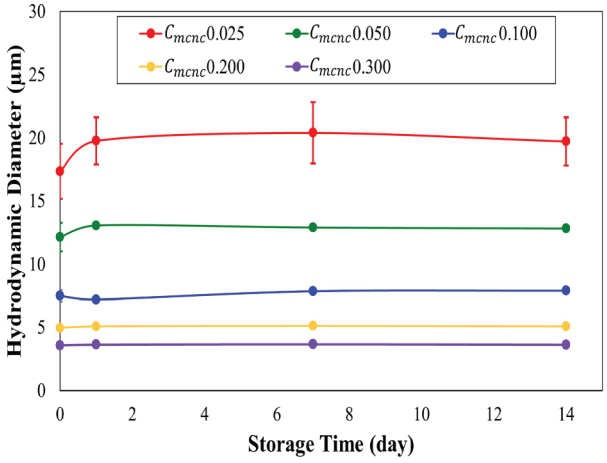

Figure 5. Changes in diameter of MCNC-PE with ranging from 0.025 to 0.300 wt% during 14 days of storage.

Figures 6a and b show photographs of the MCNC-PE prepared using various C_{mcnc} over 14 days storage. Based on Figures 6b and c, the CI gradually decreased with increasing C_{mcnc}. This is not only because of the reduced MCNC-PE diameter, but also due to the presence of abundant MCNC particles around the aqueous phase, which results in the formation of particles network that hinder the upward movement of emulsion droplets[36]. Overall, it is reasonable to consider that MCNC-PE with smaller droplets diameter (stabilized at higher C_{mcnc}) possess enhanced creaming and coalescence stability as compared to those with the larger ones (stabilized by lower C_{mcnc}).

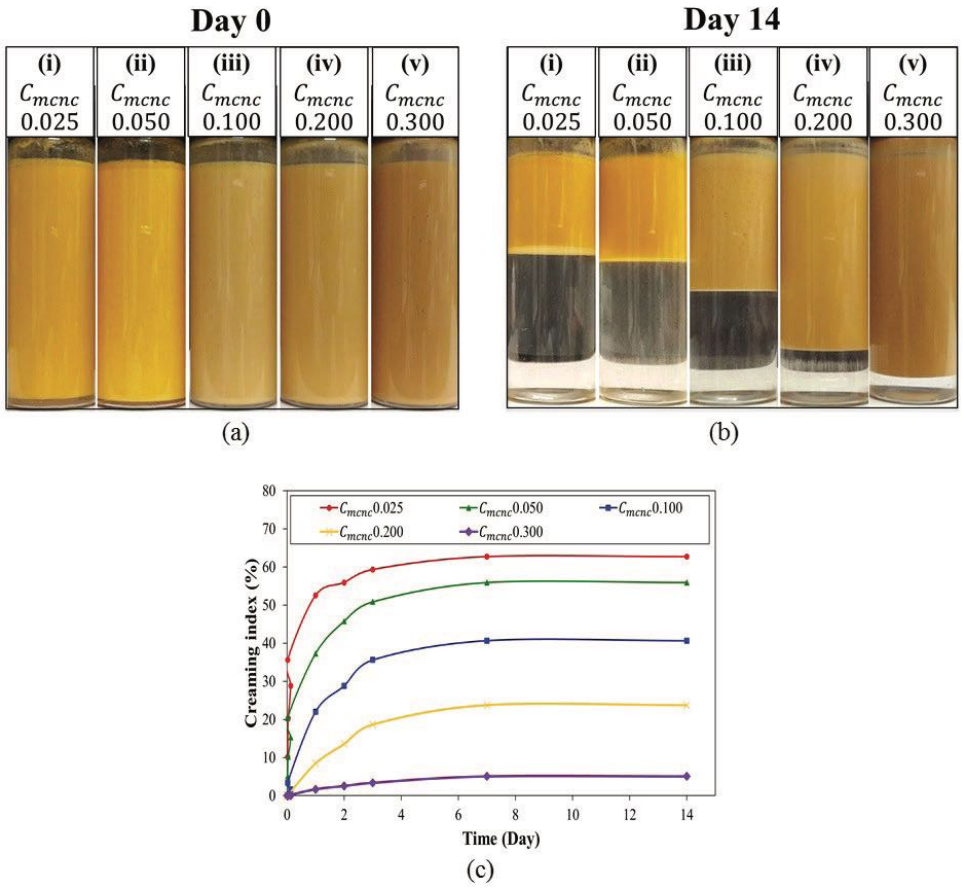

Figure 6. (a) Day 0 and (b) day 14 of photograph of O/W Pickering emulsion stabilized by MCNCs at (i) 0.025, (ii) 0.050, (iii) 0.100, (iv) 0.200, and (v) 0.300 wt%. (c) creaming indexes.

Oil volume fraction

The effect of oil content is another important parameter needing attentions. The size, S_{mcnc}, coalescence stability and creaming of MCNC-PE were determined as the function of φ_{oil}. As shown in Table 2, the of S_{mcnc} emulsion initially decreased from 16.556 to 12.070% when the φ_{oi} increased from 0.1 to 0.2. The amount of MCNC covering the MCNC-PE then increased from 12.070 to 19.872% upon further increment of the φ_{oil} to 0.5. To examine the relationship between the φ_{oil} and S_{mcnc} we express the Equation 2 in term of $D_{3,2}/V_{oil}$ " representing Equation 3 (Equation 3) and the derived terms were calculated as shown in Table 2. As expected, the calculated values showed a similar trend to that of S_{mcnc}, suggesting the increasing φ_{oil} to have a significant impact on the surface properties and droplet sizes for a constant quantity of the particle stabilizer.

$$S_{mcnc} = f\left(D_{3,2}/6hpV_{oil}\right) \quad (3)$$

Table 2. Surface coverage, S_{mcnc} and calculated values of $D_{3,2}/V_{oil}$ of MCNC-PE prepared at different oil volume fraction, φ_{oil}. .

Oil volume fraction, φ_{oil}	Surface Coverage, S_{mcnc} (%)	(µm/ml) $D_{3,2}/V_{oil}$
0.1	16.556	0.704
0.2	12.070	0.513
0.3	13.088	0.557
0.4	15.698	0.668
0.5	19.872	0.845

Based on Figure 7b, the MCNC-PE diameter progressively increases with increasing φ_{oil} (Figure 7b). This is due to the limiting quantity of MCNC particles available for Pickering stabilization at increasing φ_{oil}. In this study, the minimum MCNC-PE size of 2.82 µm with a narrow monomodal distribution was attained at φ_{oil} = 0.1. Optical microscopy images of MCNC-PEs showed spherical shaped droplets that are uniformly dispersed in the continuous phase regardless of the φ_{oil}, with the oil (Figure 7), which is consistent with particle size measurements (Figure 7a).

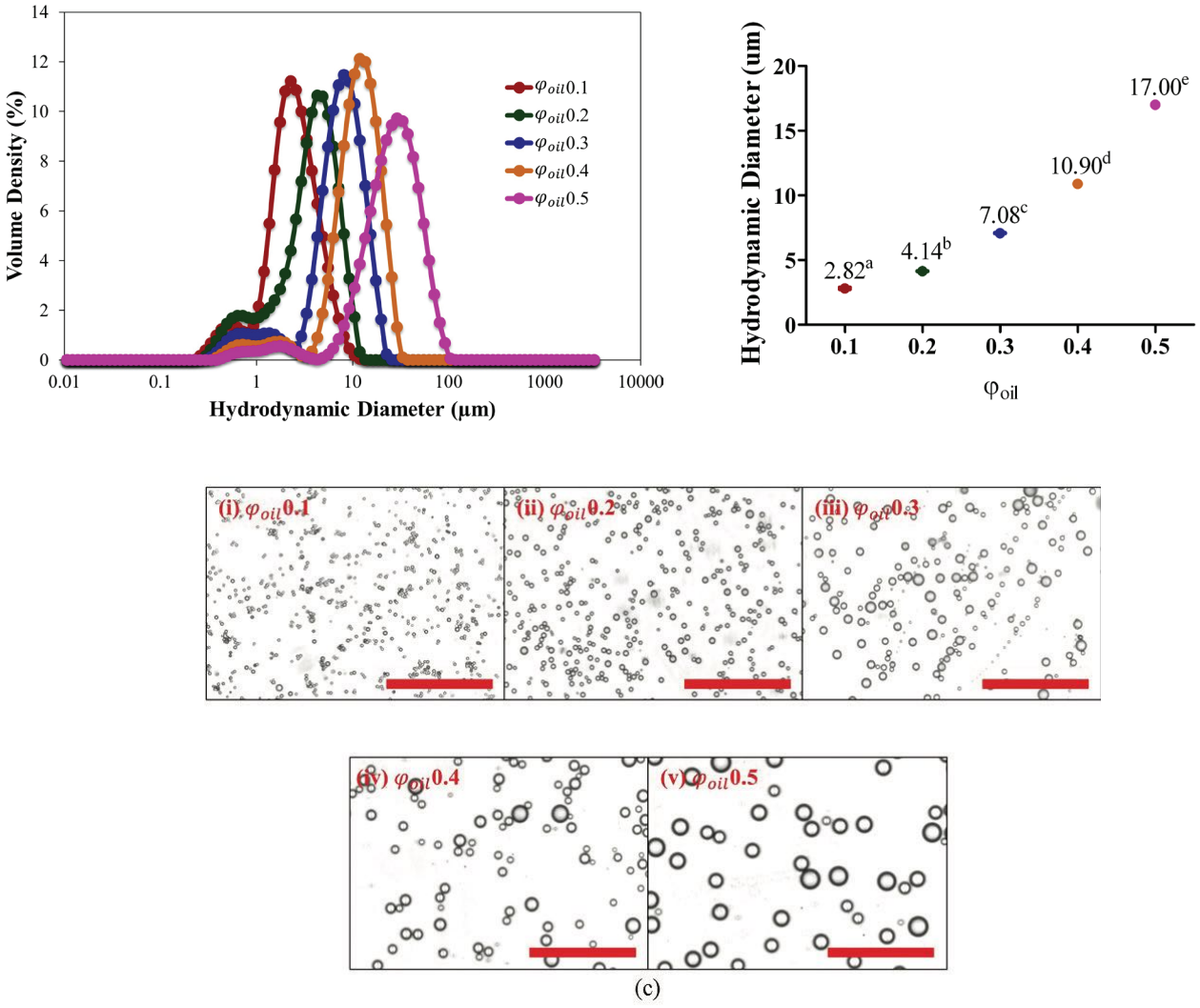

Figure 7. (a) Droplet size distributions, (b) mean droplet diameters, and (c) optical micrographs of MCNC-PE as a function of φ_{oil}. All MCNC-PEs were prepared with fixed C_{mcnc} of 0.1 wt%. The standard error of the mean of triplicate measurements was represented by the error bars in each graph, and different alphabetic letters were significantly different at $P \leq 0.05$ using Bonferroni's multiple comparison test. All scale bars represents 100 µm.

Figure 8a depicts the change in the size of MCNC-PE upon storage. Note that all the emulsions prepared at varying φ_{oil} from 0.2 to 0.5 showed no significant change in droplet diameter after 14 days of storage. However, a mild increment in droplet size observed at φ_{oil} = 0.1 and that was most likely due the particles bridging effects as a result of an excessive content of MCNC particles. In this regard, fluorescence microscopy has been performed to visualize the size and morphology of MCNC-PE prepared at φ_{oil} of 0.1. As shown in Figure 8b, the emulsion droplets were terribly agglomerated, promoting the formation of a highly entangled network of MCNC particles in the continuous phase, which rendered larger emulsion sizes.

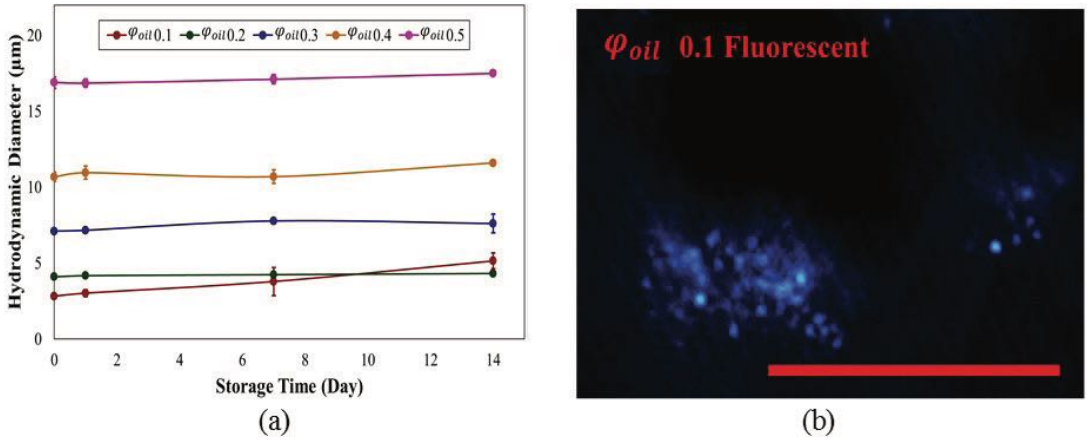

Figure 8. (a) Changes of MCNC-PE diameter at different , throughout 14 days of storage. (b) Fluorescent microscopic image of MCNC-PE prepared at = 0.1 φ_{oil} (40x magnification). All formulations contained a fixed C_{mcnc} of 0.1 wt%. The scale bar represents 50 μm.

Figures 9a and b show the photographs of MCNC-PE produced with various φoil at day 0 and day 14, respectively. The CI decreased as the was increased from 0.1 to 0.5. It is apparent that a φ_{oil} Pickering emulsion prepared at of 0.5 displayed the lowest degree of creaming among φ_{oil} the emulsion samples. This is because the emulsion CI is directly related to the initial φoil during emulsion preparation. Since creaming occurred in all samples, and emulsion sample with = 0.1 holds the lowest oil loading, despite its relatively small φ_{oil} oil droplets diameter, the CI eventually reached a point that corresponded to its . A similar trend was reported by He *et al.*[10] who studied φ_{oil} the stability of emulsion systems stabilized by graphene oxide.

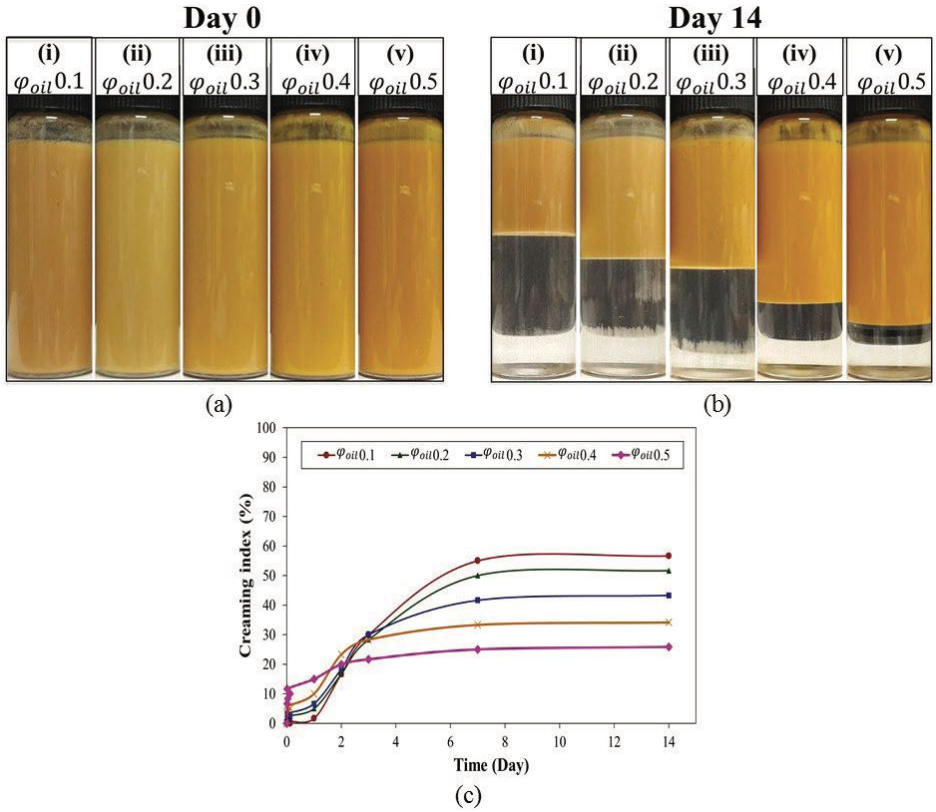

Figure 9. Photographs of MCNC-PE at (a) day 0, and (b) day 14. (c) Creaming profile of the Pickering emulsions prepared using different φ_{oil}. All formulations contain fixed C_{mcnc} = 0.100 wt%.

Effect of ionic strength on the MCNC-PE properties

One of the normally encountered stresses in the human body is the ionic content. Here, the influence of ionic strength was evaluated by varying the NaCl concentration from 0 to 500 mM. All MCNC-PE samples were stored at room temperature for 14 days prior to analysis. Figures 10a–c revealed that all MCNC-PEs show similar mean droplet diameter around 7 to 8 µm with a narrow size distribution regardless of the salt concentrations (Figure 10a). Conversely, minor increment in emulsion creaming (< 10% of CI) was observed with increasing ionic content (Figure 10b).

It should be noticed that the obtained optical microscopic images displayed the aggregated bound droplets at all NaCl concentrations (Figure 10c). The observed agglomeration could be owing to the electrostatic screening effects by an increase in the ionic strength of the continuous phase[28]. Noteworthy, the outcomes, as supplemented by mastersizer analysis demonstrated that the MCNC-PEs were highly stable against coalescence for 2 weeks despite a pronounced aggregation upon the addition of NaCl. A possible explanation for this scenario is that under conditions of high particle charge and weak screening, the coalescence of the vast majority of particles is severely impeded by the presence of a fixed quantity of MCNC particles.

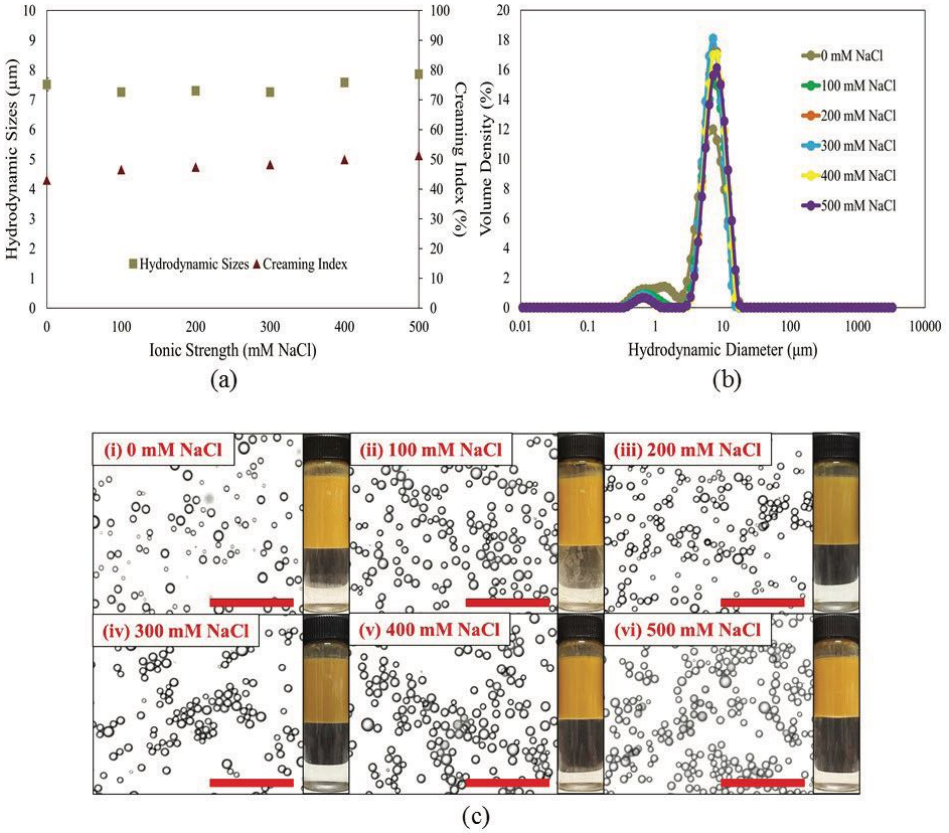

Figure 10. (a) Mean droplet diameter and creaming profiles, (b) Droplet size distribution and (c) optical micrographs of MCNC-PE as a function of ionic strength. All formulations contain fixed φ_{oil} of 0.3 and C_{mcnc} of 0.100 wt%. The samples were stored at room temperature for 14 days prior to measurement. All scale bar represents 100 µm.

To verify this further, we prepared two MCNC dispersions containing 0.1 wt% MCNC particles. The addition of 500 mM NaCl into the dispersion triggered the partial sedimentation of the MCNC particles (Figure 11).

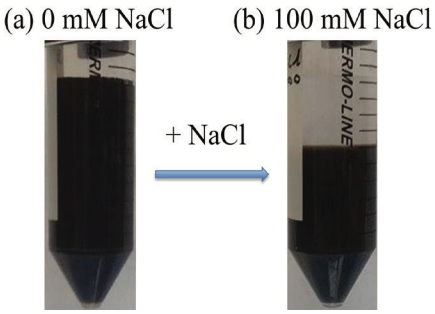

Figure 11. MCNC suspensions (a) without NaCl and (b) with 500 mM NaCl.

The latter confirmed the hypothesis that described the correlation between charge effects and emulsion aggregation. The results of current study suggest that the stability of MCNC-stabilized Pickering emulsions against creaming and coalescence was unaffected by ionic strength (NaCl) from 100 to 500 mM. In comparison to other existing Pickering emulsions in the literature where none of them remained physically unchanged (in size and surface coverage) when subjected to ionic strength variations, our results have shown significant advances on the stability of the green MCNC-PE against the environmental ionic strength[37–40].

Conclusion

The present study investigated the effects of CNC/MNP ratios, C_{mcnc} and ionic strength on the emulsion stability and φ_{oil} physical properties of MCNC-PE. At fixed φ_{oil}

of 0.3, extremely stable emulsions with minimum droplet sizes of about 7 μm could be prepared using 0.1 wt% C_{mcnc} with CNC/MNP ratio of ≤ 1. It was found that the emulsion droplet sizes decreased gradually with improved creaming stability with the increase in S_{mcnc}. This could be mainly attributed to the increased droplet S_{mcnc} when the C_{mcnc} was increased from 0.025 to 0.300 wt%. An increase in the φ_{oil} led to formation of larger emulsion droplet sizes. Our study showed that MCNC-PE were stable against droplet coalescence with the S_{mcnc} > 11%. No instability in term of emulsion droplet size and phase separation were observed upon changes in ionic strength from 0 to 500 mM. These findings are useful in the development of Pickering emulsion-based delivery system with predictable emulsion stability and droplet size variation.

Conflict of Interest

The authors declare that there is no conflict of interest.

Acknowledgements

Authors thank the Ministry of Higher Education Malaysia who funded this study through Fundamental Research Grant Scheme (FRGS/1/2017/STG07/MUSM/03/1). The authors also gratefully acknowledge the support from Graduate Merit Research Scholarship and Advanced Engineering Platform (AEP), Monash University Malaysia.

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