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Effect of process parameters on nanoemulsion droplet size and distribution in SPG membrane emulsification

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ABSTRACT

A Shirasu-porous-glass (SPG) membrane with a mean pore size of 2.5 μm was used to produce an oil/water (O/W) nanoemulsion of flurbiprofen consisting of methylene chloride as the dispersed phase, polyvinyl alcohol (PVA) as the stabilizer and a mixture of Tween 20 and Tween 80 in demineralized water as the continuous phase. Emulsion droplets with a mean droplet size of 25 times smaller than the mean pore size and a narrow droplet size distribution were produced using 5% emulsifier at a feed pressure of 15 kPa. Under these conditions the *z*-average diameter and size distribution of the emulsion droplets formed were influenced by the type of surfactant, agitator speed (150–1200 rpm), feed pressure (15–80 kPa), stabilizer concentration (0–4, w/v) and the temperature of the continuous phase. Increasing the agitator speed and stabilizer concentration increased the *z*-average diameter and decreased the size uniformity. There was a linear relationship between the increased feed pressure and the decreased *z*-average diameter of the emulsion droplets. However, the uniformity of the size distribution decreased with increasing feed pressure. The continuous phase temperature played an important role in particle size and distribution. The nanoemulsion composed of oil, water, PVA and the surfactant mixture at the weight ratio of 10/100/1/5 was prepared using a SPG membrane at an agitator speed of 300 rpm, a feed pressure of 15 kPa and a continuous phase temperature of 25 °C. Our results indicated that these conditions led to relatively uniform emulsion droplets with a narrow size distribution and high zeta potential. This emulsion was stable for at least 13 h. Furthermore, the droplets in the emulsion containing the drug were not smaller but were more uniform with a narrower distribution compared to those without the drug.

1. Introduction

Numerous studies have been done on the preparation of both oil-in-water (O/W) and water-in-oil (W/O) emulsions by utilizing the physicochemical properties of surfactants as emulsifiers, i.e., using the phase inversion technique, the surfactant phase emulsification technique, etc. (Kandori et al., 1991). Furthermore, many pieces of emulsification equipment such as the colloid mill, homogenizer and ultrasonic emulsifier have been developed and improved (Kandori et al., 1991). However, the droplet sizes of emulsions prepared using these emulsification techniques and equipment are not highly monodispersed, because their emulsification conditions cannot be precisely controlled. Membrane emulsification is a process that is used to produce an emulsion, or dispersion, of one liquid phase (such as oil) in a second immiscible liquid phase (such as water). The process usually employs shear at the surface of the membrane in order to detach the dispersed phase liquid drops from the membrane surface, after which they become dispersed in the immiscible continuous phase. In many cases the liquid drops are then polymerized, or otherwise solidified, in order to produce solid particles, usually with a very narrow particle size distribution. Ito and Makino (2004) produced droplets of monodispersed microspheres using the SPG membrane technique, with the result that the size of droplets prepared by membrane emulsification was smaller and narrower than that without membrane emulsification. Examples of such products include calibration materials, food and flavour encapsulates, controlled release depots under the skin and ion exchange resins (Serguei et al., 2008). The first investigation on using membrane emulsification can be traced

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back to the late 1980s when Nakashima et al. (2000) fabricated a particular glass membrane, called Shirasu porous glass (SPG). Highly uniform-sized kerosene-in-water and water-in-kerosene emulsions were successfully produced, and since that time the method has continued to attract attention due to its effectiveness in producing narrow droplet size distributions with low energy consumption (Nakashima et al., 2000). Shirasu porous glass (SPG) membranes are potentially suitable for membrane emulsification due to their uniform-sized pores and wide range of available mean pore diameters (0.05–50 μm) (Kukizaki and Nakashima, 2004).

The objective of this work was to characterize the process variables in the preparation of a uniform-sized nanoemulsion by a conventional direct membrane emulsification technique using standard SPG. In direct membrane emulsification, one immiscible liquid (the disperse phase) is forced through the membrane into the other immiscible liquid (the continuous phase). Fine droplets are then formed at the interface between the membrane surface and the continuous phase. To ensure regular droplet detachment from the pore outlets, shear stress is generated at the membrane–continuous phase interface by re-circulating the continuous phase using a low-shear pump or agitating in a stirring vessel (Vladisavljević et al., 2004). Moreover, it has been reported that in membrane emulsification the speed of the continuous-phase flow and the adsorption kinetics of the surfactant, which is added in the continuous phase, have a significant effect on the size and size distribution of the droplets (Babak et al., 2007; Vladisavljevic and Williams, 2005). During the droplet formation process, the surfactant molecules adsorb onto the newly formed oil–water interface and reduce the interfacial tension and consequently facilitate droplet formation. The rate of transfer of the surfactant molecules from the bulk solution to the newly formed oil–water interface partly depends on the continuous-phase flow. The surfactant type and concentration greatly influence the adsorption kinetics of the surfactant and thus the dynamic interfacial tension (Schröder et al., 1998). In addition, the flow state of the dispersed phase in membrane pores has a great influence on the spontaneous droplet formation behaviour and itself depends on the viscosities of the dispersed and continuous phases and the flow velocity of the dispersed phase through the membrane pore (Sugiura et al., 2002).

Although a considerable amount of work has been carried out in the field of membrane emulsification in the last decade, the influence of process parameters on droplet size distribution has not yet been fully investigated. Furthermore, in some investigations only the mean droplet diameter was given as a parameter of distribution, although the width of the droplet size distribution is a key emulsion property in the case of membrane emulsification. Thus, we investigated the influence of surfactant type, the cross-flow of the continuous phases (agitator speed), the temperature of the continuous phase, the stabilizer concentration (polyvinylalcohol) and the feed pressure (transmembrane pressure) on the size and size distribution of the droplets generated using SPG membranes.

2. Materials and methods

2.1. Materials

Flurbiprofen was supplied by Kolon Life Sciences, Inc. (Kwacheon, Korea). Labrafil M 2125 and Labrafil M 1994 were supplied by Gattefosse (Saint-Priest Cedex, France). Methylene chloride, polysorbate 20 (Tween 20), polysorbate 80 (Tween 80), sorbitan monolaurate 20 (Span 20), sorbitan monooleate 80 (Span 80), polyethylene glycol 4000 and polyethylene glycol 6000 were purchased from Duksan Chemical Co. (Ansan, Korea). Poloxamer 188 and poloxamer 407 were purchased from BASF (Ludwigshafen, Germany). Polyvinylalcohol (PVA) was purchased from Sigma–Aldrich (St. Louis, MO, USA). All other chemicals were of reagent grade and were used without further purification. A miniature kit for emulsification with an MPG module (microporous glass, a brand name of SPG) was purchased from Kiyomoto Iron Works Co. (Miyazaki, Japan).

Table 1
Effect of surfactants on the solubility of flurbiprofen.

Surfactants	Solubility (μg/ml)
Water	5.1 ± 0.2
Polyethylene glycol 6000	71.2 ± 22.1
Polyethylene glycol 4000	46.3 ± 9.4
Poloxamer 188	487.0 ± 251.5
Poloxamer 407	2351.0 ± 548.4
Labrafil M 1994	328.3 ± 1.4
Labrafil M 2125	428.2 ± 262.2
Span 20	496.8 ± 81.7
Span 80	553.5 ± 21.3
Tween 20	8735.1 ± 1220.6
Tween 80	11,193.2 ± 3074.9

Each value means the solubility of flurbiprofen in distilled water containing 10% surfactant. Each value represents the mean ± S.D. ($n = 3$).

2.2. Solubility studies

An excess of flurbiprofen powder (about 100 mg) was added to 10 ml of 10% surfactant as shown in Table 1. An excess of solid dispersions (about 100 mg) was also added to 10 ml of water. They were shaken in a water bath at 25 °C for 7 days, centrifuged at 3000 × *g* for 10 min (Eppendorf, USA) and filtered through a membrane filter (0.45 μm) (Balakrishnan et al., 2009; Choi et al., 1998; Li et al., 2008). The concentration of flurbiprofen in the resulting solution was analysed using an HPLC system (Hitachi, Japan) consisting of Class VP computer software, an L-2130 pump and an L-2400 UV/VIS detector. The column was an Inertsil ODS-3 C18 column (5 μm, 15 cm × 0.46 cm i.d.). The mobile phase consisted of a mixture of phosphate buffer (pH 3.5) and acetonitrile (4:6, v/v). The eluent was monitored at 220 nm with a flow rate of 1 ml/min. The inter- and intra-day variances of this HPLC method were within the acceptable range ($R^2 = 0.999$).

2.3. Pseudo-ternary phase diagram

The existence of emulsion fields that form emulsions under agitation was identified from ternary phase diagrams of systems containing oil–surfactant–water. The effect of surfactant (Tween 20, Tween 80 or a mixture of these two surfactants at 1:1, w/w ratio) and water on the pseudo-ternary phase diagram was systematically observed at room temperature. The surfactant and water were weighed at different ratios (0:1 to 1:3, w/w) in each vial, and were vortexed vigorously for 30 s. Afterwards, the oil phase (methylene chloride) was added in 5% increments to each clear mixture of water and surfactant. This was vortexed for 5 min and kept in a water bath at 25 °C for 30 min. The mixture was then visually examined for emulsion formation. The points from uniform milky solution to separation were designated as the emulsion- and no emulsion-forming regions, respectively (Ping et al., 2005; Yan et al., 2009).

2.4. Experimental set-up and procedure

A schematic diagram of this kit for emulsification with an MPG module (microporous glass, a brand name of SPG) is shown in Fig. 1. The hydrophilic SPG membrane was tube-shaped with an outer diameter of 10 mm, a thickness of 0.75 mm and a pore size of 2.5 μm. Ten grams of dispersed phase (oil phase) were stored in

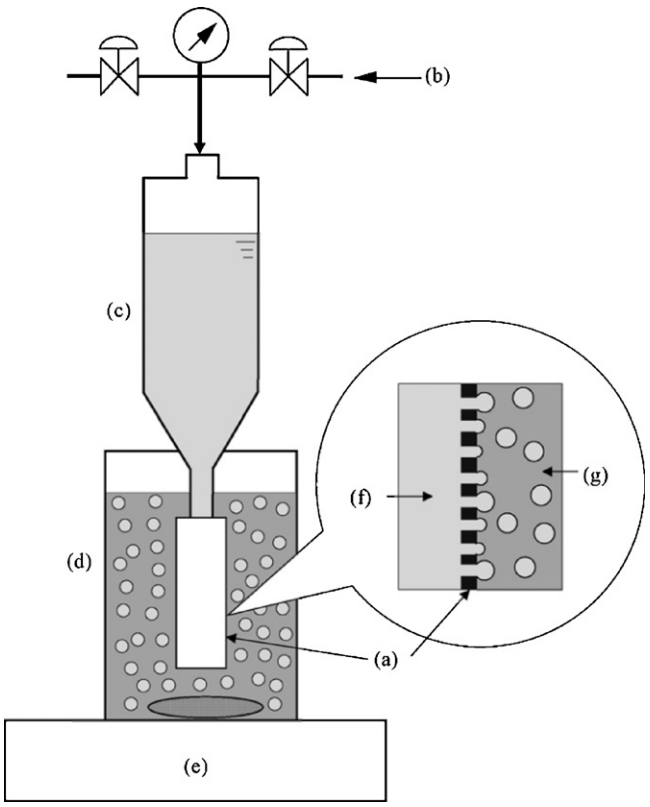

Fig. 1. Schematic diagram of the experimental apparatus used for the preparation of O/W emulsions using an SPG membrane: (a) SPG membrane module; (b) compressed nitrogen gas; (c) dispersed phase container; (d) continuous phase and emulsion container; (e) magnetic stirrer and heater; (f) dispersed phase; (g) continuous phase.

a pressure-tight vessel that was connected to a nitrogen gas inlet attached to a pressure gauge PG-200-163GP-S (COPAL Electronics, Japan). The continuous phase, 100 g of water containing 5% surfactant and 1% PVA, was stirred gently with a magnetic bar (3 cm long) in a beaker to prevent the creaming of the droplets. By applying an adequate pressure of nitrogen gas, the oil phase, 10 g methylene chloride, permeated through the uniform pores of the membrane into the aqueous phase to form the droplets, and was then stabilized by PVA dissolved in the aqueous phase. The experiments were carried out over a wide range of agitator speeds (150–1200 rpm), feed pressures (15–80 kPa), PVA concentrations (0–4%) and continuous phase temperatures (25 or 35 °C).

To prepare the drug-loaded nanoemulsion, 2.5 g of flurbiprofen was dissolved in 10 g of methylene chloride. The flurbiprofen-loaded nanoemulsion was prepared using this methylene chloride containing flurbiprofen as the oil phase as mentioned above.

2.5. Determination of mean droplet size, droplet size distribution and zeta potential

The droplet size distribution and zeta potential of all samples were measured using a NanoZS light scattering particle size analyser (Malvern, UK) with non-invasive back scatter (NIBS®) technology, which allowed the detection of droplets in the range of 0.6–6000 nm. The data on particle-size distribution were collected using the DTS (nano) software (Version 5.0) provided with the instrument. All studies were repeated three times and the values of the *z*-average diameters were used. The *z*-average diameter, also referred to as the harmonic intensity-weighted average hydrodynamic diameter, of the emulsions was derived from cumulated analysis by the Automeasure software (Malvern Instruments, Malvern, UK).

All the experiments in the study were performed at least three times and the data were expressed as the mean ± standard deviation (S.D.). A two-tailed unpaired Student's *t*-test was performed at $p < 0.05$.

2.6. Stability test

To evaluate the stability of the emulsion, an emulsion was kept at 25 °C for 13 h. At the predetermined times, its droplet size and zeta potential were investigated using the methods described above (Ee et al., 2008).

3. Results and discussion

3.1. Solubility studies and pseudo-ternary phase diagrams

The emulsion system consisting of oil, surfactants, cosurfactants and drug should be a stable monophasic liquid at ambient temperature and should have good solvent properties to allow presentation of the drug in solution. The solubility of flurbiprofen in 10% of various vehicles is presented in Table 1. The aqueous solubility of flurbiprofen was about 5.1 μg/ml. Our result indicated that flurbiprofen is poorly water-soluble (Balakrishnan et al., 2009; Choi et al., 1998; Li et al., 2008). The choice of surfactants is important for the production of uniformly sized emulsion droplets via SPG membrane emulsification. The drug was more soluble in all the surfactants compared to its aqueous solubility. The hydrophilic functional groups of the selected surfactants must not carry the opposite charge to that of the membrane surface in order to avoid electrostatic attractions with the membrane surface, which has a negative zeta potential within the pH range of 2–8, due to the dissociation of silanol groups on the membrane surface (Kai et al., 2005; Vladisavljevic and Williams, 2005). Therefore, cationic surfactants must be avoided when using SPG membranes. Among the surfactants tested in this study, Tween 20 (HLB 16.7) and Tween 80 (HLB 15), both non-ionic surfactants, were selected since they gave the highest drug solubility of 8000–14,000 μg/ml. In this study, methylene chloride was used as the oil phase.

The construction of a phase diagram makes it easy to determine the range of concentrations of components in which microemulsions form. Phase diagrams were constructed to determine the individual component ratio for the formulation of an O/W emulsion consisting of methylene chloride as the oil phase, Tween 80 (Fig. 2A) or Tween 20 (Fig. 2B) or a mixture of Tween 20/Tween 80 (1:1) (Fig. 2C) as the surfactants, and water. As shown in Fig. 2, the area in which an O/W emulsion formed varied with the surfactant. Thus, a larger emulsion region was obtained in the system in which the surfactant mixture was used, compared to Tween 20 or Tween 80 alone. This increase was toward the oil–water axis. Our results indicated that the use of the surfactant mixture and the maximum amount of water could let flurbiprofen to be solubilized into the emulsion system. Thus, the system containing the mixture of Tween 20/Tween 80 (1:1) was selected as the surfactant. Furthermore, based on its particle size and low surfactant concentration, the formulation composed of oil, water and the surfactant mixture at the weight ratio of 10/100/5 was selected as the basic component for further studies.

3.2. Influence of process parameters on mean droplet size or distribution

3.2.1. Effect of agitator speed

The influence of agitator speed over a range of 150–1200 rpm on the *z*-average diameter of emulsion droplets and the standard

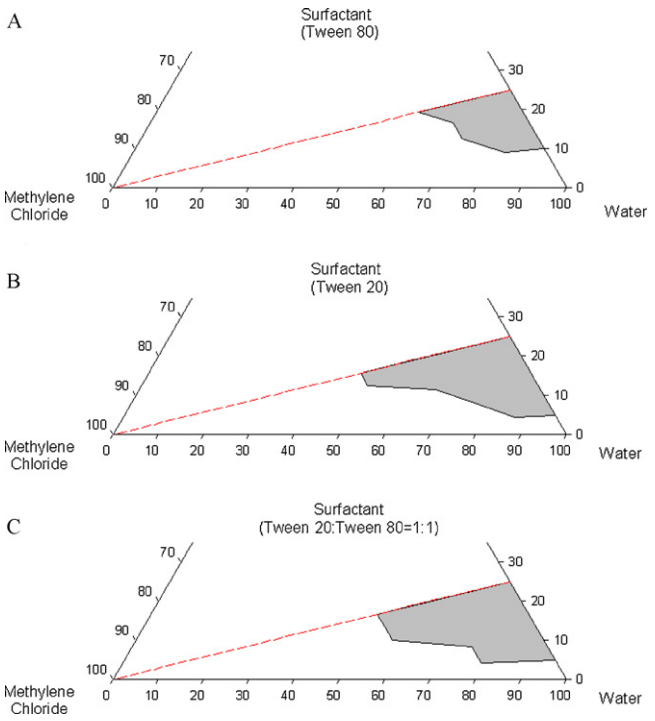

Fig. 2. Pseudo-ternary phase diagrams: (A) methylene chloride, Tween 80 and water; (B) methylene chloride, Tween 20 and water; (C) methylene chloride, Tween 80/Tween 20 (1:1, volume ratio) and water.

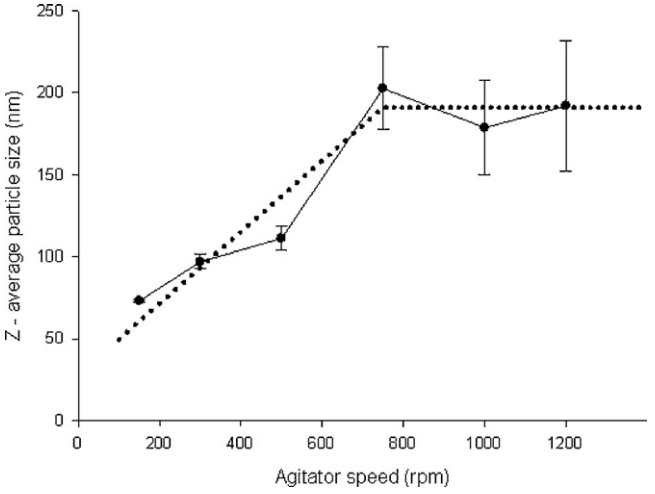

Fig. 3. Effect of agitator speed on the *z*-average diameter of the emulsion droplet. Each value represents the mean ± S.D. ($n = 3$). The dotted line represents the regression line.

deviation of the *z*-average diameter for the 2.5 μm-SPG membrane is shown in Fig. 3. The *z*-average diameter increased sharply as the agitator speed increased to 700 rpm, followed by no significant change in particle size, which was more or less independent of the flow velocity. However, the largest standard deviation of the *z*-average diameter occurred at the higher agitator speeds. Moreover, the polydispersity values were 0.217 ± 0.011, 0.257 ± 0.041, 0.229 ± 0.009, 0.168 ± 0.029, 0.220 ± 0.027 and 0.208 ± 0.011 at 150, 300, 500, 750, 1000 and 1200 rpm, respectively. Our results suggest that an agitator speed of 300 rpm is the optimal speed, since it produced emulsion droplets with a relatively smaller *z*-average diameter and a narrower standard deviation of the *z*-average diameter under this set of conditions.

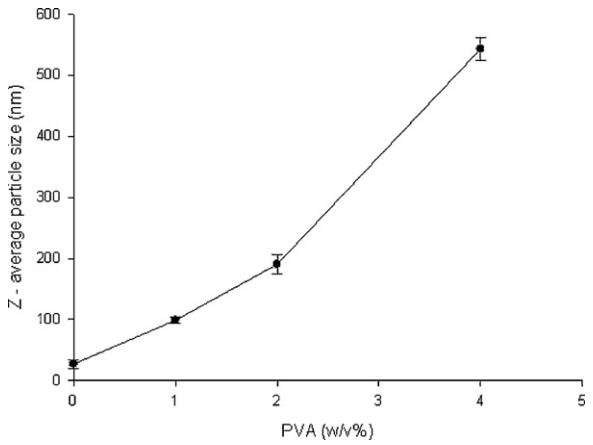

Fig. 4. Effect of polyvinyl alcohol on the *z*-average diameter of the emulsion droplet. Each value represents the mean ± S.D. ($n = 3$).

In contrast to our results, it has previously been reported that the particle sizes in an emulsion decrease with increasing agitation speed (Tsukada et al., 2009). It has also been reported that the effects of agitator speed on the properties of an emulsion have not yet been clarified, even though many mechanisms have been postulated for emulsion formation (Ni et al., 2007; Wei et al., 2009). On penetrating the SPG membranes, droplets that formed at the membrane surface were detached by the flowing continuous phase. During this phase, the capture of entities generated in the continuous phase played an important role in the formation of the emulsions. Thus, as the agitation rate increased, the capture of the monomer surfactants by the growing emulsion droplets increased, and the probability of small droplets contacting to form large droplets in the solution increased, because the surfactants (Tween 20 and Tween 80) predominantly partitioned in the continuous phase (Ni et al., 2007; Wei et al., 2009).

Furthermore, the energy supplied during the emulsification process might produce convective flow in the disperse phase and continuous phases, leading to the deformation and break-up of the disperse phase droplets, as well as the dissipation of mechanical energy (Romeroa et al., 2008). An increase in the agitation speed might give rise to an associated increase in energy consumption. Thus, as the agitation speed increased, the particle size distribution in the emulsion became broader.

3.2.2. *Effect of stabilizer*

The presence of a stabilizer dissolved in the continuous phase also plays a critical role in membrane emulsification. First, it can rapidly decrease the interfacial tension between the dispersed phase and continuous phase by adsorption at the newly formed interface of the droplet, and thus depress the critical transmembrane pressure that arises during the emulsification process. Second, it can restrict the coalescence and aggregation of emulsion droplets. PVA is a typical stabilizer that has often been used in SPG membrane emulsification (Hao et al., 2008). The effect of PVA concentration over the range of 0–4% (w/v) (Table 2) in the continuous phase on the *z*-average diameter of emulsion droplets at 300 rpm agitator speed using an SPG membrane was investigated. As shown in Fig. 4, as the amount of PVA in the continuous phase increased, the *z*-average diameter of the emulsion droplets increased, since the continuous phase viscosity increased, and decreased the interfacial tension between the dispersed phase and continuous phase (Babak et al., 2007; Vladisavljevic and Williams, 2005). The nanoemulsions prepared with 0, 1, 2 and 4% PVA showed the polydispersity values of 0.293 ± 0.019, 0.257 ± 0.041, 0.241 ± 0.015 and 0.333 ± 0.054, respectively. Furthermore, PVA

Table 2
Process variables.

Formulation	Stirring rate (rpm)	Polyvinylalcohol (w/v %)	Nitrogen gas pressure (kPa)	Temperature (°C)
1	150			
2	300			
3	500			
4	750	1	15	25
5	1000			
6	1200			
7		0		
8	300	2	15	25
9		4		
10			10	
11			20	
12	300	1	40	25
13			60	
14			80	
15	300	1	15	35

Each value represents the mean ± S.D. ($n = 3$).

usually increased the standard deviation of the *z*-average diameter. The PVA concentration was subsequently fixed at 1%, as it gave relatively small emulsion droplets and a narrow deviation of *z*-average diameter compared to the other PVA concentrations tested.

3.2.3. Effect of feed pressure of dispersion phase

Another parameter with a strong influence on droplet size distribution is the feed pressure, which can control the flux rate of the dispersed phase across the membrane channel and the detachment of the droplets. An increase in feed pressure induced the formation of smaller *z*-average-diameter emulsion droplets (Fig. 5A). As the dispersed-phase transmembrane pressure increased from 15 kPa to 80 kPa, the uniformity of the emulsion decreased (Fig. 5B). Therefore, it was speculated that a high transmembrane pressure would lead to the detachment of shear-induced droplets and a polydisperse emulsion. The droplet size distribution observed under different emulsification pressures proved the aforementioned speculation (Fig. 5B). In general, the higher the transmembrane pressure, the smaller the particle size in the emulsion (Luca et al., 2004). As pressure increased, the droplet size distribution curve shifted to small droplet diameters and the droplet distributions became narrower. However, at extremely high pressures, the particle size decreased but the droplet distribution became much wider (Vladisavljević, 2003). Furthermore, the nanoemulsions prepared at 15, 20, 40, 60 and 80 kPa gave the polydispersity values of 0.257 ± 0.041, 0.361 ± 0.093, 0.298 ± 0.009, 0.276 ± 0.017 and 0.351 ± 0.045, respectively. Thus, a feed pressure of 15 kPa, a critical pressure for the preparation of this nanoemulsion in this study, was used for subsequent formulation, as it gave a relatively uniform emulsion droplet size compared to higher feed pressures.

3.2.4. Effect of the continuous phase temperature

Temperature can be an important parameter in emulsification, affecting the viscosity of both the dispersed and continuous phases. Furthermore, it can affect the nature and solubility of the emulsifier as a consequence of the phase inversion temperature (Simon and Gun, 2000). However, there have been few reports on the effect of temperature on the uniformity of emulsions during microporous membrane emulsification. For W/O emulsions, heating the continuous phase is needed so that there is a substantial decrease in viscosity which would make it easier to circulate. In this study, methylene chloride was used as the oil phase, and has a very low evaporation temperature of 40 °C. The interfacial tension between the two phases would change at a higher temperature; therefore, it was important to investigate the effect of temperature on the droplet size distribution. Thus, the effect of temperature on the size distribution of the emulsion droplets generated when the continuous phase was at 25 °C or 35 °C was investigated (Fig. 6). The increase in temperature from 25 °C to 35 °C gave a smaller particle size but a broader size distribution (Fig. 6). It might be due to coarsening by Ostwald ripening, and coalescence and evaporation of methylene chloride (Ee et al., 2008; Leal-Calderon and Poulin, 1999; Tadros et al., 2004; Wiácek and Chibowski, 1999). The fluctuation in interfacial tension resulting from the increased temperature might explain the low uniformity of the emulsion droplets formed (Soheil et al., 2008; Babak et al., 2007). Table 3 shows the *z*-

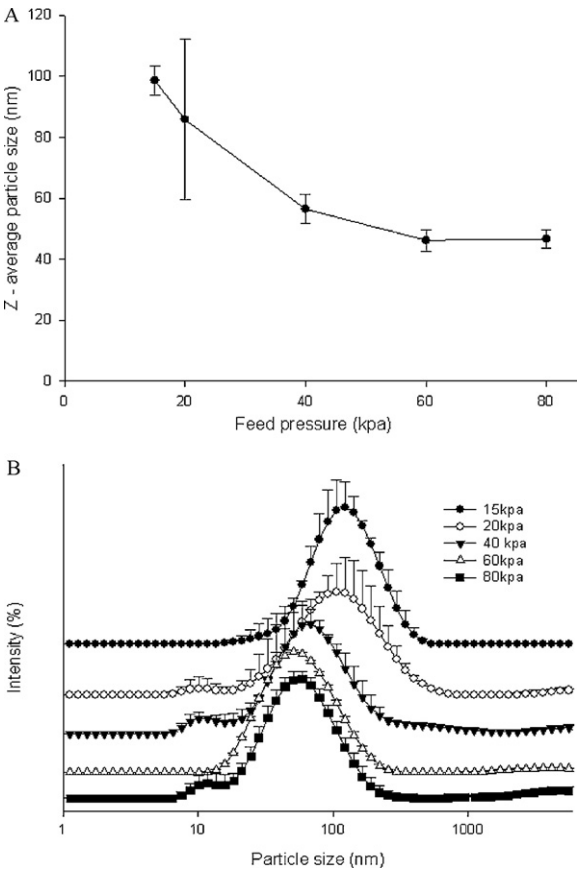

Fig. 5. Effect of the feed pressure of the dispersion phase on the *z*-average diameter of the emulsion droplet (A) and the emulsion droplet size distribution (B). Each value represents the mean ± S.D. ($n = 3$).

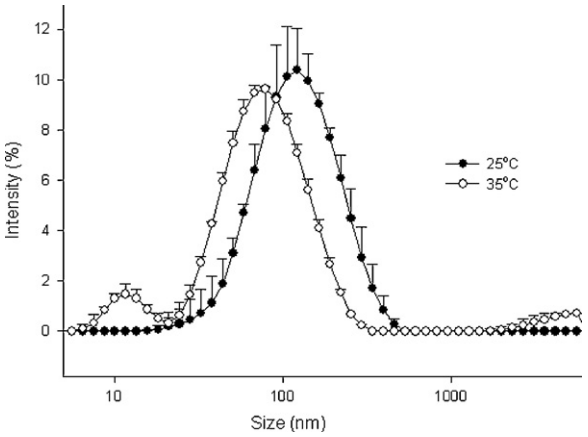

Fig. 6. Effect of continuous phase temperature on the emulsion droplet size distributions. Each value represents the mean ± S.D. (n = 3).

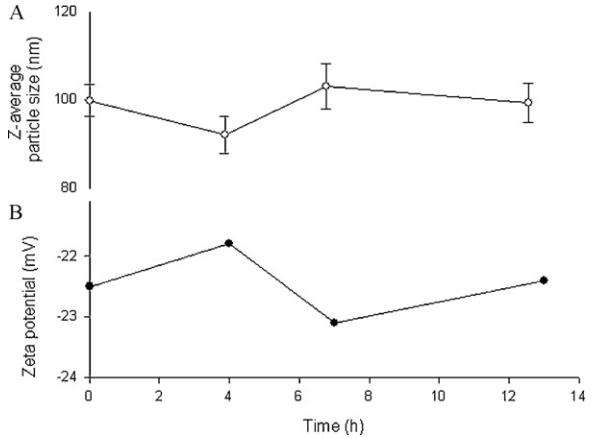

Fig. 7. Stability of emulsion: (A) droplet size and (B) zeta potential. Each value represents the mean ± S.D. (n = 3).

average diameter and zeta potential of emulsion droplets prepared at different temperatures. The continuous phase prepared at 25 °C gave a significantly higher z-average diameter and more negative zeta potentials than that at 35 °C, due to the relatively high viscosity of the continuous phase at 25 °C compared to that at 35 °C in the preparation of nanoemulsion with the same pressure and agitation speed (Capek, 2004; Sinko, 2006). However, after preparation, the nanoemulsion prepared at 35 °C gave no significant viscosity compared to that prepared at 25 °C. Thus, 25 °C was selected as the continuous phase temperature in subsequent parts of the study, since it gave relatively uniform emulsion droplets with a narrow size distribution and high zeta potential.

Based on these findings, the formulation composed of oil, water, PVA and the surfactant mixture at the weight ratio of 10/100/1/5 was selected to produce an optimal uniform nanoemulsion using an SPG membrane at an agitator speed of 300 rpm, a feed pressure of 15 kPa and a continuous phase temperature of 25 °C.

3.3. Stability

To evaluate the stability of this nanoemulsion, it was kept at 25 °C for 13 h and its droplet size and zeta potentials were investigated (Fig. 7). Ultimately, this nanoemulsion is an intermediate product for nanoparticle. To prepare the nanoparticle with the nanoemulsion, an ingredient will be added and mixed in the nanoemulsion followed by spray-drying at the predetermined conditions. Generally, the stability of this nanoemulsion is evaluated at 25 °C for 13 h, since it will take less than 13 h to perform this procedure (Li et al., 2008). In addition, the solvent of methylene chloride in disperse phase may be evaporated at 25 °C for 13 h. However, the property of nanoparticle is not dependent upon the evaporation of organic solvents in the preparation of nanoparticle (Ee et al., 2008). Moreover, the polydispersity values at 0, 4, 7 and 13 h were 0.257 ± 0.041, 0.273 ± 0.033, 0.218 ± 0.024 and 0.258 ± 0.015, respectively. In this study, there were no significant changes in the droplet size, size distribution and zeta potential during the storage period, indicating that it was stable for at least 13 h.

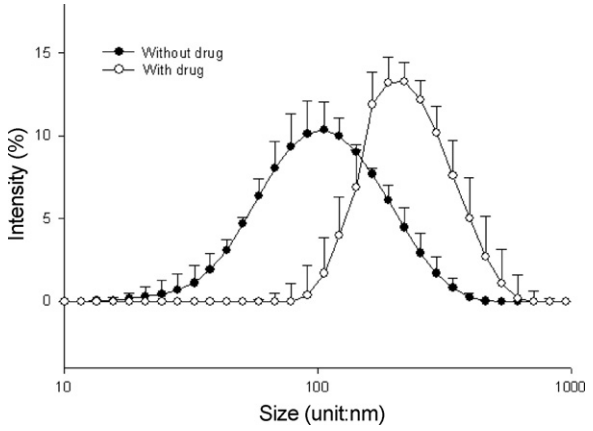

Fig. 8. Effect of drug on the emulsion droplet size and size distributions. Each value represents the mean ± S.D. (n = 3).

3.4. Drug loading test

The flurbiprofen-loaded nanoemulsion was prepared using methylene chloride solution containing flurbiprofen as the oil phase. In brief, the flurbiprofen-loaded nanoemulsion was prepared from a formulation composed of flurbiprofen, oil, water, PVA and surfactant mixture at the weight ratio of 2.5/10/100/1/5 using an SPG membrane at the agitator speed of 300 rpm, a feed pressure of 15 kPa and a continuous phase temperature of 25 °C. Even if flurbiprofen is soluble in the surfactant mixture, the drug was practically entrapped in the dispersed phase, since it was poorly soluble in water and very soluble in methylene chloride. Furthermore, the drug moved to the continuous phase and was entrapped in the surfactant mixture, resulting in complete solublility (Li et al., 2008). Thus, it is important for the drug not to be soluble in the dispersed phase or the continuous phase, but to be soluble entirely in nanoemulsion system. The effect of the drug on the droplet size and size distribution of the emulsion droplets was investigated (Fig. 8). Table 4 presents the z-average diameter and zeta potential of the emulsion with and without the drug. The addition of

Table 3
z-Average diameter and zeta potential of emulsions prepared with a continuous phase temperature of 25 and 35 °C.

Temperature (°C)	z-Average diameter (nm)	Zeta potential (mV)	Polydispersity value
25	98.5 ± 0.26	−22.5 ± 0.53	0.256 ± 0.041
35	60.2 ± 0.39	−5.98 ± 1.76	0.288 ± 0.013

Each value represents the mean ± S.D. (n = 3).

Table 4
z-Average diameter and zeta potential of emulsion with and without drug.

Emulsion	*z*-Average diameter (nm)	Zeta potential (mV)	Polydispersity value
Without drug	98.5 ± 0.26	−22.5 ± 0.53	0.257 ± 0.041
With drug	360.1 ± 14.7	−19.6 ± 1.53	0.251 ± 0.014

Each value represents the mean ± S.D. ($n = 3$).

the drug increased the droplet size of nanoemulsion. However, the emulsion containing the drug had a lower negative zeta potential than that without the drug. It was previously reported that a negative zeta potential higher than 20 mV was sufficient to prevent droplet coalescence (Wiácek and Chibowski, 1999). As the emulsion with the drug had a zeta potential of about −20 mV, the droplets might coalesce in this emulsion. Furthermore, the droplets in the emulsion with the drug were not smaller but were more uniform with a narrower size distribution compared to those without the drug (Fig. 8). Thus, this flurbiprofen-loaded nanoemulsion uniform-sized with about 500 nm was well prepared using SPG membranes. Generally, the emulsions were successfully produced using SPG membranes when they gave the available mean pore diameters of 0.05–50 μm with their uniform-size (Kukizaki and Nakashima, 2004; Nakashima et al., 2000). Further study on the development of flurbiprofen-loaded nanoparticle will be performed with this nanoemulsion.

4. Conclusion

A nanoemulsion composed of oil, water, PVA and surfactant mixture at the weight ratio of 10/100/1/5 was prepared using an SPG membrane at an agitator speed of 300 rpm, a feed pressure of 15 kPa and a continuous phase temperature of 25 °C. It had relatively uniform emulsion droplets, a narrow size distribution and a high zeta potential. Furthermore, it was stable for at least 13 h. The effects of agitator speed, stabilizer concentration, feed pressure, temperature and emulsifier on the uniformity of the droplets indicated that uniform droplets could be prepared when the interfacial tension between the water phase and the thin layer of the oil phase that covered the membrane was high enough; moreover, the interfacial tension between the water droplets and the oil layer must be decreased rapidly by adsorbing emulsifier onto the oil layer. The droplets in the emulsion containing the drug were not smaller but were more uniform with a narrower size distribution compared to those without the drug.

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