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Microfluidic Fabrication of Monodisperse Polylactide Microcapsules with Tunable Structures through Rapid Precipitation

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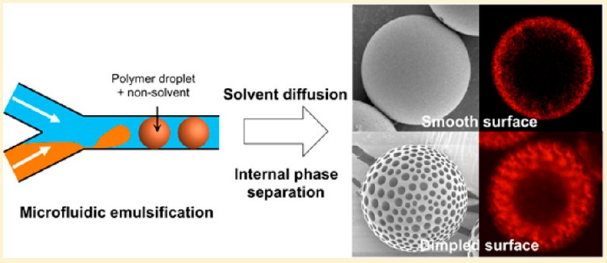
Supporting Information

ABSTRACT: We describe a versatile and facile route to the continuous production of monodisperse polylactide (PLA) microcapsules with controllable structures. With the combination of microfluidic emulsification, solvent diffusion, and internal phase separation, uniform PLA microcapsules with a perfluorooctyl bromide (PFOB) core were successfully obtained by simply diluting monodisperse ethyl acetate (EA)-in-water emulsion with pure water. Rapid extraction of EA from the droplets into the aqueous phase enabled the solidification of the polymer droplets in a nonequilibrium state during internal phase separation between a concentrated PLA/EA phase and a PFOB phase. Higher-molecular-weight PLA generated structural complexity of the microcapsules, yielding core−shell microcapsules with covered with small PFOB droplets. Removal of the PFOB via freeze drying gave hollow microcapsules with dimpled surfaces. The core−shell ratios and the diameter of these microcapsules could be finely tuned by just adjusting the concentration of PFOB and flow rates on emulsification, respectively. These biocompatible microcapsules with controllable size and structures are potentially applicable in biomedical fields such as drug delivery carriers of many functional molecules.



1. INTRODUCTION

Biodegradable polymeric microparticles have attracted a great deal of attention in applications for medical and pharmaceutical fields such as sustained release carriers for drugs,[1] agrochemicals,[2] medical imaging agents,[3] personal care ingredients, and cell scaffolds for tissue engineering.[4] In these ubiquitous applications, among many kinds of biodegradable polymers, polylactide (PLA) and poly(lactide-*co*-glycolide) (PLGA) are the most commonly used in the world because they both have good biocompatibility and high mechanical strength.[5,6]

Generally, these microparticles are produced through a top-down emulsification approach known as emulsion−solvent evaporation.[7−11] In this technique, an oil-in-water (O/W) emulsion is formed by the emulsification of a polymer/volatile organic solvent mixture in an aqueous surfactant solution, after which the organic solvent is removed by evaporation, yielding polymeric microparticles dispersed in the medium. This technique can also be used to fabricate liquid-filled microcapsules by either adding nonsolvent to the organic phase prior to emulsification[12−14] or preparing water-in-oil-in-water (W/O/W) emulsions used as a template.[15−18] These techniques have been shown to be a promising way to produce core−shell polymeric microcapsules encapsulating lipophilic or hydrophilic compounds in the core and to control their release behavior.

In terms of microcapsule size, monodisperse microcapsules are preferable in several applications, especially in drug delivery carriers because they can decrease undesirable side effects and exhibit controlled drug release kinetics and encapsulation efficiency. Because monodisperse microcapsules can be produced only from precision emulsion droplets, the generation of uniform droplets is of utmost importance in fabricating microcapsules using top-down techniques. To date, several methods have been developed to produce monodisperse polymeric microcapsules using flow dynamics, such as membrane emulsification,[19] microfluidic emulsification,[20−25] and jet acoustic excitation.[26] Among them, the droplet-based microfluidic system is considered to be one of the most effective ways to prepare monodisperse microcapsules. Pioneering work by Utada et al. has been carried out to fabricate a monodisperse W/O/W double emulsion using a coaxial microcapillary fluidic device.[27] Since then, there have been many reports regarding the fabrication of monodisperse microcapsules using a method of combining microfluidics with solvent evaporation. For example, Liu et al. have successfully controlled the stability and size of monodisperse PLGA microcapsules by tuning the osmotic pressure between the internal and the external aqueous phases during W/O/W double emulsion formation.[28] Amstad et al. have developed monodisperse polymersomes that are thermo- and photo-responsive by introducing thermoresponsive diblock copolymer

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poly(*N*-isopropylacrylamide)-*b*-poly(lactide-*co*-glycolide) and photosensitive gold naoparticles into the shell of the polymersomes.[29] For oil-filled microcapsules, Lensen et al. have prepared monodisperse dodecane-filled poly(L-lactide) (PLLA) microcapsules using a combination of O/W emulsion−solvent evaporation and internal phase separation.[30] However, these methods require toxic organic solvents and much time to form microcapsules, and it is difficult to tailor microcapsule morphologies. In emulsion preparation, dichloromethane or chloroform is commonly used as the organic solvent because each is a good solvent for biodegradable polyesters and because of the high volatility that facilitates the evaporation of the solvent from the emulsion droplets, although the vapor or residue is considered to be harmful to the environment and the human body. In addition, even when using volatile organic solvents as the dispersed phase, the rate-determining step for microcapsule formation is a solvent-evaporation process, which actually takes a few minutes to several hours, depending on the volume of the continuous phase. That is why the emulsion is required to be stored in a vessel until the solvent evaporates from the system and the microcapsules are formed. Moreover, despite the fact that the morphology of microparticles plays a crucial role in determining their behavior in the fluid, controlling the loading efficiency and release kinetics of the ingredients and tuning the structure of polymeric microparticles on the micrometer scale are still challenging. Hence, developing a simple and environmentally friendly process for the production of monodisperse microcapsules with controllable size and tailored architecture is in great demand in expanding their functionalities.

Herein, we present a facile, straightforward method of continuously producing monodisperse PLA microcapsules with controlled structures by a modified droplet-to-particle technology[31] that includes microfluidic emulsification, emulsion−solvent diffusion, and internal phase separation. In our process, for a disperse organic phase, PLA and perfluorooctyl bromide (PFOB), a nonsolvent for PLA, were dissolved in ethyl acetate (EA), which is a nontoxic organic solvent approved by the U.S. Food and Drug Administration and has relatively higher solubility in water (8.3 wt % at 20 °C). Monodisperse O/W emulsion droplets produced by using a commercial Y-shaped microfluidic device were poured directly into an excess amount of water, which induced the rapid extraction of EA from the droplets and internal phase separation between the PFOB phase and the concentrated PLA/EA phase. Within a few seconds, PLA precipitated at the surface of each droplet, resulting in the formation of monodisperse PLA microcapsules encapsulating liquid PFOB. Different from conventional techniques, our approach to forming robust microcapsules takes only a few seconds to precipitate the polymer after the onset of solvent diffusion, which has enabled us not only to produce uniform microcapsules in a continuous manner but also to solidify microcapsules in a nonequilibrium state and design their structures. In addition, all components that we used in microcapsule formation are biocompatible, which is very advantageous to applications in biomedical fields. To the best of our knowledge, this is the first report of to continuously fabricating monodisperse core−shell PLA microcapsules with tunable size and structures using solvent diffusion from simple O/W emulsion droplets.

2. EXPERIMENTAL SECTION

2.1. Materials. Poly(D,L-lactide) (PLA) and poly(ethylene glycol)-*b*-poly(D,L-lactide) (PEG-*b*-PLA) were synthesized by the ring-opening polymerization of D,L-lactide in the presence of tin(II) 2-ethylhexanoate as a catalyst using lauryl alcohol and poly(ethylene glycol) monomethyl ether (PEG, M_n = 4000, M_w/M_n = 1.06) as initiators, respectively, as previously reported.[32] The D,L-lactide was purchased from Purac (Netherland). The PEG was kindly supplied by NOF (Japan). Tin(II) 2-ethylhexanoate, ethyl acetate (EA), and perfluorooctyl bromide (PFOB) were obtained from Wako Pure Chemical Industries, Ltd. (Japan). The porphyrin derivative was kindly supplied by a porphyrin laboratory (Japan). Ultrapure water was produced by a Millipore Milli-Q purification system (EMD Millipore Corporation, USA).

2.2. Preparation of Monodisperse PLA Microcapsules. A schematic illustration of the preparation procedure of monodisperse PLA microcapsules is shown in Figure 1. The microfluidic device that

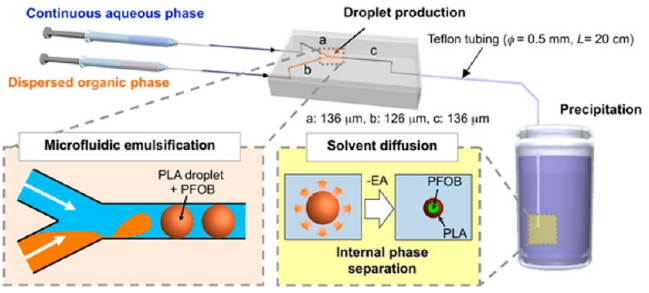

Figure 1. Schematic illustration of the process used to continuously produce monodisperse PLA microcapsules encapsulating liquid PFOB in the core through microfluidic emulsification and solvent diffusion, coupled by internal phase separation.

was used for microcapsule fabrication consisted of a Y-shaped channel (126- and 136-μm-wide channels with 75 μm depth) made of an SUS basement and a glass cover plate, fabricated by Kasen Nozzle Mfg. Co., Ltd., Japan. The continuous aqueous phase and the dispersed organic phase were pumped independently at adjustable flow rates using syringe pumps connected to the device via Teflon tubing. An aqueous solution saturated with EA containing 1 wt % water-soluble PEG-*b*-PLA (w-PEG-*b*-PLA, M_n = 4,400, M_w/M_n = 1.05, hydrophile−lipophile balance (HLB)= 18.2) was used as the continuous phase, and an EA solution composed of 25 mg mL^{-1} PLA (M_n = 13 600, M_w/M_n = 1.14 or *Mn* = 52 000, M_w/M_n = 1.29) and 1.25−15 μL mL^{-1} PFOB was used as the disperse phase. For confocal microscopy, a trace amount of a porphyrin derivative was added to the disperse phase before the feeding. The obtained O/W emulsion was transferred to a bath filled with 100 mL of ultrapure water through Teflon tubing (Φ = 0.5 mm, L = 20 cm) whose exit tip was submerged in the water. The EA was then rapidly removed from the droplet into a large amount of pure water by solvent diffusion with or without gentle stirring, leading to the precipitation of PLA microcapsules. The microcapsules were washed with ultrapure water three times by centrifugation (himac CF 15R, Hitachi, Japan) (3000 rpm, 3 min) to remove the surfactant, followed by freeze drying overnight to yield dried PLA microcapsules.

2.3. Interfacial Tension Measurements. An interfacial tension measurement was carried out by the pendant drop method using a DSA10 (Kruss, Germany). An EA droplet containing 25 mg mL^{-1} PLA and a pure PFOB droplet were formed in an aqueous solution of 1 wt % w-PEG-*b*-PLA saturated with EA. The interfacial tensions were calculated from the droplet images using image analysis software. The measurements were carried out at 20 °C.

2.4. Sample Characterization. Emulsions and microcapsules were observed using an optical microscope (Olympus BX50, Japan) equipped with a digital camera (Olympus CS 230). A porphyrin-derivative-labeled microcapsule dispersion was observed with a confocal laser scanning microscope (CLSM) equipped with a 1 mW

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helium−neon laser (Zeiss LSM-510, Japan). The red fluorescence was observed with a long-pass 560 nm emission filter under 543 nm laser illumination. The morphology of the microcapsules after freeze drying was observed with a scanning electron microscope (SEM, S-4700, Hitachi Ltd., Japan) at an intensity of 1 kV under various magnifications. A sputter coater (E-1030 ion sputterer, Hitachi Ltd. Japan) was used to coat the samples with Pd−Pt to prevent the samples from being charged. Before the observation, the freeze-dried samples were stored in a desiccator. We evaluated the microcapsule size and the size distribution on the microscopy images by using image analysis software (Winroof, Mitanishoji Co., Ltd., Japan). In the analysis, the size distribution was expressed by the coefficient of variation (CV) that is defined as the ratio of the standard deviation to the mean diameter. We used 200 microcapsules in each calculation.

3. RESULTS AND DISCUSSION

The continuous preparation of monodisperse microcapsules with a liquid oil core and a biocompatible polymeric shell of PLA via emulsion−solvent diffusion has been performed in this study. The process follows our previous technique of preparing monodisperse compact PLA microparticles using a combined method of microfluidic emulsification and subsequent solvent diffusion.[31] This method does not require toxic materials and a time-consuming solidification process. The disperse phase is a mixture of PLA, EA, and PFOB. EA is a good solvent for PLA with high solubility in water, whereas PFOB that is used as a model-encapsulated reagent acts as a nonsolvent for PLA. The continuous phase is an aqueous solution saturated with EA containing biocompatible w-PEG-*b*-PLA as a surfactant. The diblock copolymer enhances the stability of the EA/water emulsion.

In detail, the organic phase and the aqueous phase are first separately introduced into the Y-shaped microfluidic device by using syringe pumps to produce monodisperse polymer droplets. Then, the polymer droplets travel downstream from the channel and are finally poured into a solidification bath filled with a sufficient amount of ultrapure water via Teflon tubing. Because of the higher solubility of EA in water, once the polymer droplets touch pure water, rapid diffusion of EA to the outer aqueous phase begins. Solvent diffusion leads to a rapid reduction of the droplet sizes and a steep increase in the volume fraction of PLA and PFOB within the droplets. As a result of the concentration increase, the droplets readily reach the bimodal boundary, which induces internal phase separation and results in the formation of the PFOB phase as tiny droplets and a concentrated PLA/EA phase within each droplet. At this stage, we can see two interfaces; one is between the PFOB phase and the concentrated PLA/EA phase within the droplets and the other is between the dispersed organic phase and the continuous aqueous phase as we recognize emulsion. As the diffusion of EA proceeds, tiny PFOB droplets within each polymer droplet tend to coalesce to minimize the interfacial area and eventually form a single large core within the emulsion droplets.

The equilibrium structure of the microcapsules can be predicted by using a spreading coefficient theory established by Torza and Mason, which is based on the interfacial energy of the O/W interface.[33] According to the theory, if droplets of two immiscible liquids (phases 1 and 3) are brought into contact in the third mutually immiscible liquid (phase 2), then the final equilibrium morphology can be predicted by calculating spreading coefficient values using each interfacial tension (γ_{12}, γ_{23}, and γ_{31}). The spreading coefficients S_i for each phase are defined as

$$S_i = \gamma_{jk} - (\gamma_{ij} + \gamma_{ik})$$

$$S_j = \gamma_{ik} - (\gamma_{jk} + \gamma_{ij})$$

$$S_k = \gamma_{ij} - (\gamma_{ik} + \gamma_{jk})$$

In our system, two immiscible drops correspond to pure liquid PFOB (phase 1) and EA dissolving 25 mg mL^{-1} PLA (phase 3), and the third immiscible phase is an aqueous solution of 1 wt % w-PEG-*b*-PLA saturated with EA (phase 2) as shown in Figure 2A. We measured two initial interfacial tension values:

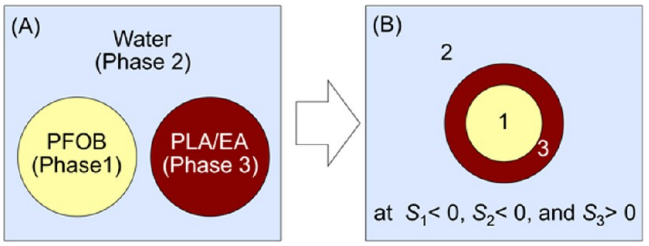

Figure 2. (A) Designation of each phase for the calculation of spreading coefficients. (B) Expected equilibrium core−shell configuration obtained at $S_1 < 0$, $S_2 < 0$, and $S_3 > 0$.

(1) between phases 1 and 2 and (2) between phases 2 and 3. It should be noted that we could not measure the interfacial tension between pure liquid PFOB (phase 1) and the EA solution dissolving PLA (phase 3) because these solutions were completely miscible under the initial experimental condition and there was therefore no interface, which is considered with respect to the fact that the interfacial tension would be considerably smaller than that of the other two interfaces. The measurement results were γ_{12} = 8.51 mN m^{-1} and γ_{23} = 2.53 mN m^{-1} (Table 1). If we assume that the interfacial tension

Table 1. Interfacial Tensions Measured by the Wilhelmy Plate Method

entry	interfacial tension (mN m^{-1})
PFOB−water (γ_{12})	8.51
PLA/EA−water (γ_{23})	2.53
PFOB−PLA/EA (γ_{13})	NA[a]

[a]The interfacial tension was assumed to be almost 0 mN m^{-1} when calculating spreading coefficients.

value between phases 1 and 3, corresponding to γ_{13}, is close to 0 mN m^{-1}, then the spreading coefficient values of our system will be $S_1 < 0$, $S_2 < 0$, and $S_3 > 0$, indicating that the equilibrium configuration of the microcapsules is the core−shell morphology (Figure 2B).

With the aid of microfluidics, we obtained a monodisperse O/W emulsion without any satellite droplets as shown in Figure 3a. This indicates that the droplets are stabilized by w-PEG-*b*-PLA at the interface of the emulsion droplets. We then carried out the precipitation of the polymer in a water bath with stirring at 120 rpm. The resultant microcapsules after solvent diffusion are shown in Figure 3b. Highly monodisperse microcapsules (d = 35.0 μm, CV = 4.0%) with core−shell structure were observed, indicating that internal phase separation occurred in the course of solvent diffusion. Understanding the detailed internal microcapsule structure, we prepared microcapsules with a trace amount of porphyrin

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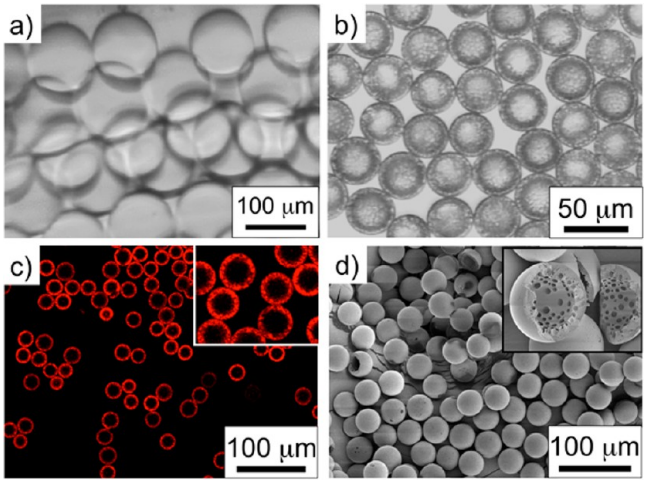

Figure 3. Optical micrographs of (a) monodisperse oil-in-water emulsion droplets in Teflon tubing and (b) monodisperse PLA microcapsules containing PFOB prepared with stirring at 120 rpm during solvent diffusion (Q_d = 60 μL h^{-1}, Q_c = 1200 μL h^{-1}). (c) Confocal micrograph of porphyrin-derivative-labeled monodisperse PLA microcapsules. The inset is the magnified image (Q_d = 60 μL h^{-1}, Q_c = 6000 μL h^{-1}). (d) SEM image of the microcapsules after freeze drying. The inset image shows the magnified cross-sectional view of the microcapsule (Q_d = 60 μL h^{-1}, Q_c = 1200 μL h^{-1}).

derivative as a fluorescent marker. This fluorescent marker colors hydrophobic PLA red but does not color PFOB, and it also does not affect the stability of the emulsion. The CLSM image has proven that each microcapsule possesses a relatively uniform shell thickness and a large cavity located at the radial center of the microcapsules (Figure 3c). It is also important to note that the shell of microcapsules has small pores although the configuration roughly corresponds to the theoretical prediction. The small cavities would be derived from small droplets of the PFOB phase before coalescence. The diffusion of EA to the outer aqueous phase is a much faster process than the evaporation of dichloromethane reported by other groups. Because of the higher solubility of EA in water, the microcapsules are obtained in a nonequilibrium configuration. The precipitation starts from the surface of EA droplets; therefore, some of the phase-separated small PFOB droplets before coalescence where it is located near the surface would be entrapped in the polymer matrix, forming the shell of the microcapsules during the precipitation process. SEM observation after freeze-drying clearly shows that most of the microcapsules maintain a spherical shape with a smooth surface (Figure 3d). In addition, we found that the microcapsules had hollow internal structure as a result of the evaporation of the PFOB core during freeze drying and many dimples at the core–shell interface (Figure 3d, inset). The dimples would be another clue that the rapid precipitation of polymer provides a nonequilibrium structure of microcapsules. These results show that our simple process is capable of the continuous production of well-defined monodisperse PLA microcapsules with a hydrophobic oil core and hollow microcapsules.

We achieved robust control over the microcapsule size by varying the flow rates upon emulsification. The flow rate of the continuous phase (Q_c) was varied between 1200 and 6000 μL h^{-1} while keeping the disperse-phase flow rate (Q_d) constant at 60 μL h^{-1}. As shown in Figure 4a–c, the diameter of the resultant microcapsules decreased with increasing Q_c, which

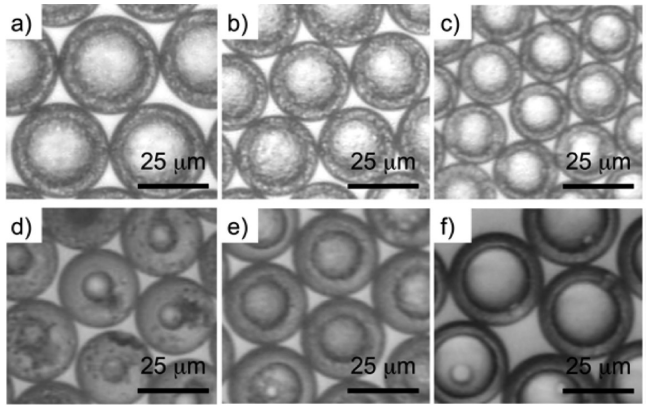

Figure 4. (a–c) Optical micrographs of monodisperse PLA microcapsules prepared by changing Q_c at a fixed Q_d (60 μL h^{-1}). Q_c was (a) 1200, (b) 3000, and (c) 6000 μL h^{-1}. C_{PFOB} was constant at 15 μL mL^{-1}. (d–f) Optical micrographs of monodisperse PLA microcapsules prepared by varying the concentration of PFOB in the disperse phase while keeping each flow rate constant (C_{PFOB} = (d) 1.25, (e) 5, and (f) 15 μL mL^{-1} at Q_d = 60 μL h^{-1} and Q_c = 1200 μL h^{-1}).

was controlled from 20.8 to 34.6 μm, in which the CV values of the core and the microcapsules size remained below 7%. Moreover, it was found that the shell thickness to radius (T/R) ratio was approximately constant at 0.28 regardless of the microcapsule size (Table 2). These results show that our system can produce monodisperse PLA microcapsules with tunable size without any effect on the T/R ratio of the core–shell structure.

Table 2. Effect of Q_c on the Microcapsule Size, the Inner Core Size, and the Shell Thickness to Radius Ratio

Q_c (μL h^{-1})	1200	3000	6000
microcapsule size (μm) (CV)	34.6 (4.0%)	26.8 (6.8%)	20.8 (4.0%)
inner core size (μm) (CV)	25.0 (4.4%)	19.2 (6.1%)	15.0 (5.2%)
shell thickness to radius ratio (T/R (−))	0.28	0.28	0.28

We have also demonstrated that the ratio of the core and shell of the microcapsules can be modulated by changing the concentration of PFOB from 1.25 to 15 μL mL^{-1} while keeping the other compositions fixed. As shown in Figure 4d–f, independent of the PFOB concentration, monodisperse microcapsules with core–shell structure were successfully obtained. The images show that the T/R ratio decreases with increasing PFOB concentration and that it can be controlled from 0.28 to 0.61 (Table 3). In addition, each shell thickness showed good agreement with the theoretical calculation results (Table S1).

Table 3. Effect of the PFOB Concentration in the Dispersed Phase on the Microcapsule Size, the Inner Core Size, and the Shell Thickness to Radius Ratio

C_{PFOB} (μL mL^{-1})	1.25	5	15
microcapsule size (μm) (CV)	33.8 (3.1%)	35.5 (3.3%)	38.4 (3.1%)
inner core size (μm) (CV)	13.1 (9.2%)	20.5 (3.4%)	27.6 (4.6%)
shell thickness to radius ratio (T/R (−))	0.61	0.42	0.28

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As an alternative to lower-molecular-weight PLA (M_n = 13 600, M_w/M_n = 1.14), in the case of PLA having a relatively higher molecular weight (M_n = 52,000, M_w/M_n = 1.29) used as a shell-forming material, we found that the structural complexity of the microcapsules occurred as a result of solvent diffusion. As shown in Figure 5a, monodisperse microcapsules

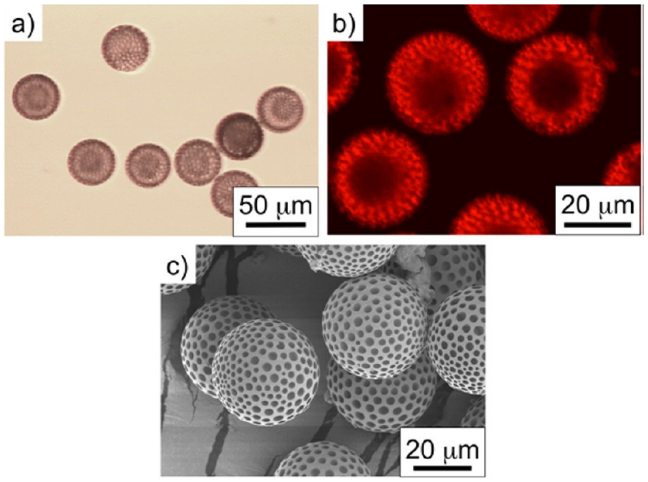
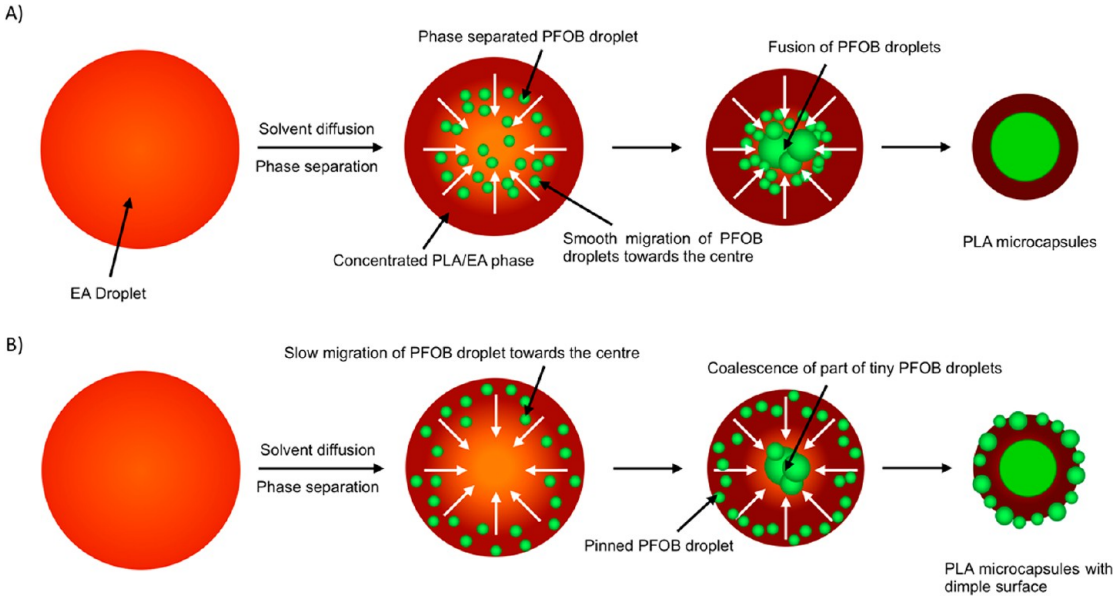


Figure 5. (a) Optical and (b) confocal micrographs of porphyrin-derivative-labeled monodisperse PLA microcapsules covered with small PFOB droplets after solvent diffusion. (c) SEM image of the microcapsules after freeze drying. The microcapsules were produced by using higher-molecular-weight PLA (M_n = 52 000, Mw/M_n = 1.29).

whose surfaces were covered with many small droplets were obtained, which looks like a Pickering emulsion.[34] The CLSM image revealed that the microparticles also had core–shell structure (Figure 5b). In the image, the microcapsules have a dark, rough surface, indicating the existence of the PFOB phase forming small droplets. Moreover, after freeze drying, we obtained monodisperse hollow PLA microcapsules with dimpled surfaces as shown in Figure 5c. The size of the dimples was apparently polydisperse, and the dimples did not penetrate the shell of the microcapsules. The distinct surface morphology would be caused by the increase in the viscosity of the PLA solution. Scheme 1 shows the proposed formation mechanism of these microcapsules. Under the preparation condition in which monodisperse PLA microcapsules with smooth surfaces were obtained, the viscosity of the disperse phase was low. That is why tiny PFOB droplets that stem from the emulsion droplet during internal phase separation can be easily moved in the polymer droplet and smoothly migrate toward the center as a result of the higher interfacial tension between the PFOB phase and the continuous phase whereas small PFOB droplets coalesce with each other before polymer precipitation (Scheme 1A). However, in the case of higher-molecular-weight PLA, the viscosity of the solution is relatively high as a result of the increase in the degree of polymer entanglement in the solution. By increasing the viscosity, the small PFOB droplets formed by internal phase separation become difficult to move and to migrate toward the center. In addition, it is much easier for the emulsion composition to reach the bimodal boundary in the droplets as a result of solvent diffusion, which leads to rapid solidification of the polymer droplets from the surface. Because the volume shrinkage of the polymer droplets is much faster and PFOB is immiscible with water and a nonsolvent for PLA, partially phase-separated small PFOB droplets before coalescence would be left on the surface of the microcapsules as a spherical form and the other PFOB droplets that escaped from the precipitation front would migrate to the center of the polymer droplet with coalescence prior to complete solidification. Consequently, monodisperse core–shell microcapsules covered with a number of small PFOB droplets are formed in the collection bath (Scheme 1B).

To confirm the formation mechanism of the microcapsules with a dimpled surface, we monitored the time course of microcapsule formation during solvent diffusion. However, because of several limitations on the volatility of EA, the quick diffusion of EA into the aqueous phase, and the rapid change in

Scheme 1. Schematic Illustration of the Proposed Formation Mechanism of PLA Microcapsules with (A) a Smooth Surface and (B) a Dimpled Surface



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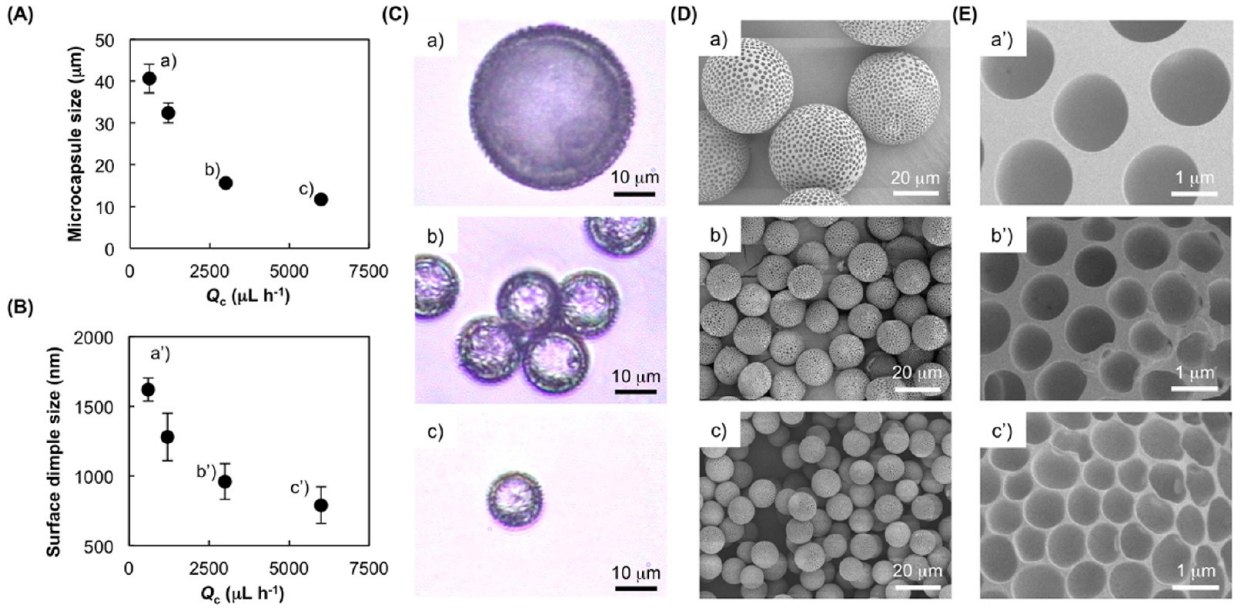

Figure 6. Effect of Q_c on (A) the diameter and (B) the surface dimple size of the microcapsules. The microcapsules were produced by varying Q_c while keeping Q_d constant. Q_c = (a, a′) 600, (b, b′) 3000, and (c, c′) 6000 μL h^{-1} at Q_d = 60 μL h^{-1}. C_{PFOB} was 15 μL mL^{-1}. (C) Optical micrographs of the microcapsules before freeze drying. SEM images of (D) monodisperse PLA microcapsules after freeze-drying and (E) the magnified surface morphology.

the density of the droplets within a few seconds, it was difficult to observe the microcapsule formation process under typical experimental conditions. Therefore, we carried out the model experiment using the emulsion (prepared with a homogenizer) on a slide glass covered with a coverslip on a microscope stage. In the experiment, we induced the solvent diffusion of EA by adding pure water to the system from the gap between the slide glass and the coverslip. Because of the step-by-step addition of pure water from one side of the sample, the diffusion time scale was longer than that of our typical experimental process and the diffusion was spread heterogeneously in the sample, which gave the resultant microcapsules with irregular shapes. However, we confirmed that in the middle stage the surface roughness was formed as a result of pinning a portion of tiny PFOB droplets at the surface, which indicated that the proposed mechanism would be plausible (Figure S1).

The diameter of the microcapsules with a dimpled surface can be tuned by changing Q_c on the microfluidic emulsification. We obtained monodisperse dimpled PLA microcapsules with 11.7 to 40.6 μm diameter (Figure 6A,C,D). It was also found that the dimple size decreased with decreasing microcapsule size (Figure 6B,E). It should be noted that the dimple size prepared with gentle stirring is smaller than that prepared without stirring, although there is no difference in the microcapsule size. This result indicates that the stirring during solvent diffusion facilitates polymer precipitation and suppresses the fusion of small PFOB droplets until some of them are stabilized on the surface.

4. CONCLUSIONS

Monodisperse PLA microcapsules encapsulating a liquid PFOB core were successfully produced by simply diluting a monodisperse O/W emulsion with pure water under stirring. The rapid extraction of EA from the droplets into outer aqueous phase led to internal phase separation between the concentrated PLA/EA phase and the PFOB phase and rapid solidification of the droplets, resulting in core−shell microcapsules with nonequilibrium structures. The core−shell ratios and the diameter of the microcapsules could be modulated by varying the compositions of the dispersed phase and flow rates upon emulsification. Our process has enabled us to prepare monodisperse PLA microcapsules having a hydrophobic oil core and a hollow structure with either smooth or dimpled surfaces. We believe that such PLA microcapsules with controllable structures have great potential for carriers of functional molecules used in biomedical fields such as cosmetics, pharmaceutics, and contrast imaging.

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Equation for estimating the theoretical shell thickness of the microcapsules. Calculation results and optical microscopy images illustrating the formation of the microcapsules with dimpled surfaces. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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