JOURNAL OF Applied Polymer SCIENCE

Tracking the Fate of Seed Particles in Dispersion Polymerization: Preparation and Application of Fluorescent Polymer Particles

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ABSTRACT: The mechanism of seeded dispersion polymerization of methyl methacrylate (MMA) was investigated by employing submicron fluorescent polymer particles as seed. These poly(methyl methacrylate) latex particles, containing fluorescent material, were synthesized by a two-step miniemulsion polymerization process and then applied in the seeded dispersion polymerization of MMA. The seed particles were located by tracking the fluorescent signal in the micron-size final particles. The analysis of the final particles showed that most of them contained more than two seed particles. On average, there were 3.7 seed particles in each final particle as obtained under the given conditions of the seeded dispersion polymerization. The location of the seed within the particles being well-separated from each other was considered to indicate that the aggregation of the particles did not occur immediately, but took place after some particle growth had first taken place. © 2012 Wiley Periodicals, Inc. J. Appl. Polym. Sci. 000: 000–000, 2012

KEYWORDS: dispersions; seeded dispersion polymerization; fluorescence; microencapsulation; miniemulsion polymerization; colloids

Received 25 October 2011; accepted 15 February 2012; published online
DOI: 10.1002/app.37522

INTRODUCTION

Micron-size, monodisperse polymer particles have found various applications in industry, health care, and scientific research areas.[1–5] Among those technologies that can produce such polymer particles, dispersion polymerization is attractive in that it can be used to prepare these particles in a single polymerization step and is suitable for a variety of monomers.[6] It has been defined as a process by which stable colloidal polymer particles are formed in a continuous liquid medium through polymerization of a monomer that is completely miscible with this medium and in which a stabilizer has been initially dissolved.[7] The size of particles prepared via dispersion polymerization is usually between 1 and 15 μ and can be highly monodisperse.

Seeded dispersion polymerization has been utilized to prepare polymer particles with a well-controlled particle size and with specific morphologies.[8–11] Both micron or submicron size particles have been used as seeds in seeded dispersion polymerization.[12–15] In these seeded dispersion polymerization investigations, the reaction kinetics were studied and the dispersion polymerization mechanism discussed. It is of great interest to determine the fate of the seed particles in the final dispersion. This should help in further understanding the mechanism of the polymerization including the particle nucleation and aggregation behaviors during the seeded dispersion polymerization process.

Fluorescence techniques have been widely used to track and detect the motion and location of a species.[16] These techniques work through the chemical or physical binding of a functionalized group onto the species. This functionalized group, termed dye, will absorb the energy of a specific wavelength and re-emit energy at a different (but equally specific) wavelength. By collecting this emitted energy during the process, the location and motion of the dye, as well as the species that the dye is anchored to, can be determined.

The miniemulsion polymerization process[17] has been shown to be a useful technique to encapsulate a liquid or a solid, an inorganic or an organic material within a polymer shell.[18–24] This technique has also been utilized to prepare fluorescent latex particles by means of encapsulating the fluorescent materials inside the polymer particles.[25–27]

In this study, poly(methyl methacrylate) (PMMA) latex particles containing fluorescent material were synthesized by a two-step miniemulsion polymerization process. These fluorescent polymer particles were used as seed in the seeded dispersion polymerization of methyl methacrylate (MMA). The seed particles were

Table I. Miniemulsion Polymerization Recipe for Preparing PMMA Seed Particles Containing Rhodamine B[a]

Ingredient		Mass (g)	Comment
Oil phase	Rhodamine B	0.1038	2.1 wt % on MMA
	AIBN	0.0026	3 m*M*
	MMA	5.0095	
	EGDMA	0.5073	10 wt % on MMA
	HD	0.1979	4 wt % on MMA
Aqueous phase	SLS	0.1446	10 m*M*
	DIW	49.9932	

[a]Reaction temperature is 70°C; reaction time is 24 h; 2-oz bottle.

located by tracking the fluorescent signal in the final particles. Particle aggregation during the seeded dispersion polymerization is discussed based on the findings of this investigation.

EXPERIMENTAL

Materials

The monomer, methyl methacrylate (MMA) (Sigma-Aldrich, Saint Louis, Missouri), was purified via vacuum distillation to remove the inhibitor before use. All other materials were used as received without further purification, including the stabilizer, polyvinylpyrrolidone (PVP) K30 (TCI America, Portland, Oregon), the costabilizer, hexadecane (HD) (Sigma-Aldrich, Saint Louis, Missouri), the crosslinker, ethylene glycol dimethacrylate (EGDMA) (Sigma-Aldrich, Saint Louis, Missouri), the alcohol, methanol (VWR Scientific, Bridgeport, New Jersey), the initiators, 2,2′-azobis(isobutyronitrile) (AIBN) (Sigma-Aldrich, Saint Louis, Missouri) and potassium persulfate (KPS) (Fisher Scientific, Pittsburgh, Pennsylvania), the surfactant, sodium dodecyl sulfate (MP Biochemicals, Solon, Ohio), the buffer salt, sodium bicarbonate (Mallinckrodt, St. Louis, Missouri), and the fluorescent dye, Rhodamine B (Sigma-Aldrich, Saint Louis, Missouri). Deionized water (DIW) was used in all experiments.

Preparation of Fluorescent Seed Particles

Submicron-size PMMA latex particles containing fluorescent agent were prepared by miniemulsion polymerization followed by a semi-batch emulsion polymerization process. The recipe for miniemulsion polymerization is listed in Table I. The aqueous and oil phases were prepared separately. Then, the oil phase was slowly added to the aqueous phase while stirring using a magnetic stirrer. Next, the mixture was sonified using a Branson sonifier (Model 450) at a power level of 7 and a 50% duty cycle for 10 min. After that, the reaction mixture was transferred into a bottle, which was purged with nitrogen, capped, and sealed. The bottle was then placed in a safety basket supported on a rotor, and was tumbled end-over-end at 40 rpm in a constant temperature (70°C) bottle polymerization unit for 24 h. A semi-batch emulsion polymerization of MMA was carried using the latex particles obtained from the miniemulsion polymerization as seed. The recipe is listed in Table II. The final particles were cleaned by serum replacement to wash out surfactant and other water-soluble materials.[28] The particle size was measured by dynamic light scattering (Nicomp 370; Particle Sizing Systems, Port Richey, Florida). The fluorescence of these particles was analyzed by confocal microscopy (vteye, VisiTech International, Sunderland, United Kingdom). These particles were used as seed in seeded dispersion polymerization of MMA.

Seeded Dispersion Polymerization

Seeded dispersion polymerization of MMA using fluorescent particles as seed was carried out via bottle polymerization. The reaction recipe is listed in Table III. All ingredients were mixed in 1-oz bottles, which were purged with nitrogen, capped, and sealed. The bottles were then placed in a safety basket supported on a rotor, and tumbled end-over-end at 40 rpm in a constant temperature (70°C) bottle polymerization unit for 24 h.

The polymerization conversion (x) was determined by gravimetric analysis of the final latex. A given amount of latex was weighed in a tared aluminum pan. Then the latex was dried at 70°C overnight and the remaining solids weighed. The solids content (X) and the conversion (x) were calculated using eqs. (1) and (2):

$$X = \frac{m_2 - m_0}{m_1 - m_0} \tag{1}$$

$$x = \frac{X \times (m_S + m_{PVP} + m_{AIBN} + m_{MMA} + m_{Seed}) - (m_{PVP} + m_{AIBN} + m_{seed})}{m_{MMA}} \tag{2}$$

where m_0 is the mass of the empty aluminum pan, m_1 is the mass of the aluminum pan with the latex, and m_2 is the mass of the aluminum pan after drying. m_S, m_{PVP}, m_{AIBN}, m_{MMA}, and m_{Seed}, are, respectively, the masses of solvents, PVP, AIBN, MMA, and seed prior to the reaction.

Particles obtained by seeded dispersion polymerization were imaged by scanning electron microscopy (HITACHI, Schaumburg, Illinois; 4300). For SEM images, about 1000 particles were counted to obtain the statistical results of particle size and size

Table II. Semi-Batch Emulsion Polymerization Recipe[a]

Ingredient			Amount
Initial ingredients		Seed latex (solids content is 9 wt %)	10 g
		$NaHCO_3$	1 m*M*
		KPS	3 m*M*
Feed	Oil phase	MMA + EGDMA (10 : 1 wt ratio)	5.4 mL
	Aqueous phase	KPS + SLS solution (0.27 wt % SLS, 4.7 wt % KPS)	5.4 mL

[a]Reaction temperature = 70°C; magnetic bar stirring; Feed rate = 0.6 mL/h; Feed time = 9 h; total reaction time = 10 h.

Table III. PMMA Dispersion Polymerization Standard Recipe[a]

Ingredient	Amount
MMA	9.8 wt % on total
Solvent	Methanol/water = 70/30 (wt/wt)
PVP K30	20.0 wt % on monomer
AIBN	0.52 wt % on monomer
Seed	$N_s = 13.05 \times 10^{12}$ L^{-1}

[a]Reaction temperature = 70°C; reaction time = 24 h

distribution. The volume-average diameter, D_v(m), was used as the size of the particles, calculated from eq. (3). Particle number, N_f (L^{-1}), was calculated using eq. (4).

$$D_v = \left[\frac{\Sigma n_i D_i^3}{\Sigma n_i}\right]^{1/3} \quad (3)$$

$$N_f = \frac{6mx}{\rho \pi D_v^3 V} \quad (4)$$

where n_i is the number of particles with diameter D_i (m), N_f is the final particle number concentration, m (kg) is the mass of the initial MMA, ρ (kg·m^{-3}) is the density of the polymer material (PMMA: 1.19 × 10^3), and V (L) is the volume of the latex.

Locating the Fluorescent Seed Particles

The final particles obtained from the seeded dispersion polymerization of MMA using fluorescent seed particles were analyzed by confocal microscopy (vteye, VisiTech International) to determine the location and number of seed particles inside the final particles. A latex sample was placed in a small container which was mounted on a glass slide and the polymer particles were allowed to settle out for 5 days. The microscopic analysis was carried out on the bottom layer of particles.

RESULTS AND DISCUSSION

The size of the particles obtained from each step is listed in Table IV. The fluorescent particle obtained from miniemulsion polymerization was 95.1 nm. The size increased to 178.7 nm after the semi-batch emulsion polymerization process was complete, which indicated that a shell was added to the initial fluorescent particle. The final particle obtained from the seeded dispersion polymerization was 3.36 μm with a narrow particle size distribution.

Fluorescent Seed Particles

The first step in preparing the nanosize PMMA latex particles containing fluorescent agent was the miniemulsion polymerization of MMA that was designed to encapsulate the fluorescent agent (Rhodamine B) into the PMMA particles. The second step was the semi-batch emulsion polymerization process using the latex particles obtained from the first step as seed. This step was designed to produce a protective shell around the fluorescent particles. This process is needed for three reasons:

1. KPS, which is used as initiator, will provide surface charge by means of the residual sulfate end groups present on the particle surface and keep the particles stable even if the surfactants are removed.
2. A PMMA shell can help to encapsulate the fluorescent agent inside the particle.
3. The seed particle size can be adjusted by changing the amount of monomer fed.

The D_v of the latex particles obtained from the semi-batch reaction was 178.7 nm. As described in the first point, the sulfate

Table IV. Size of Particles Obtained from Each Step of the Polymerizations

	Miniemulsion	Semi-batch emulsion	Seeded dispersion
D_v	95.1 nm	178.7 nm	3.36 μm
PDI	2.02	1.64	1.04

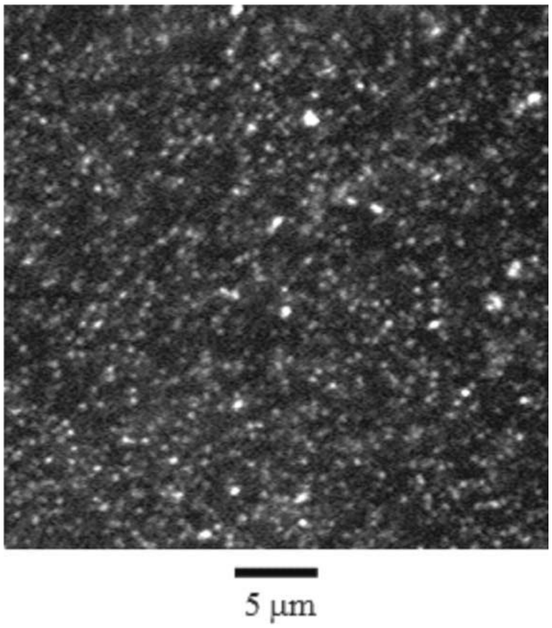

Figure 1. Confocal microscope image of the fluorescent PMMA seed particles prepared by a two-step miniemulsion polymerization.

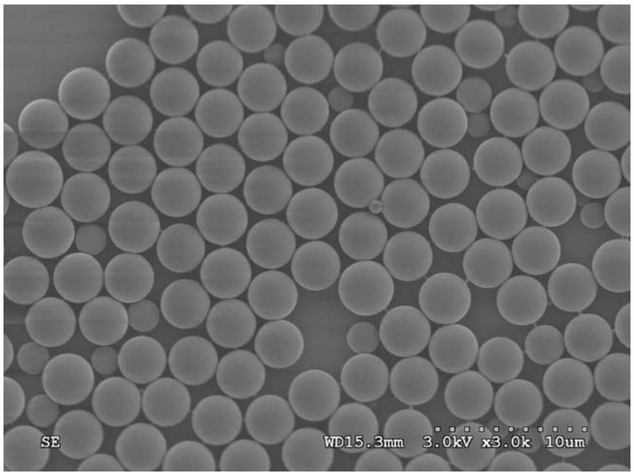

Figure 2. SEM image of particles obtained via seeded dispersion polymerization of MMA using fluorescent PMMA latex particles as seed.

Journal of Applied Polymer Science

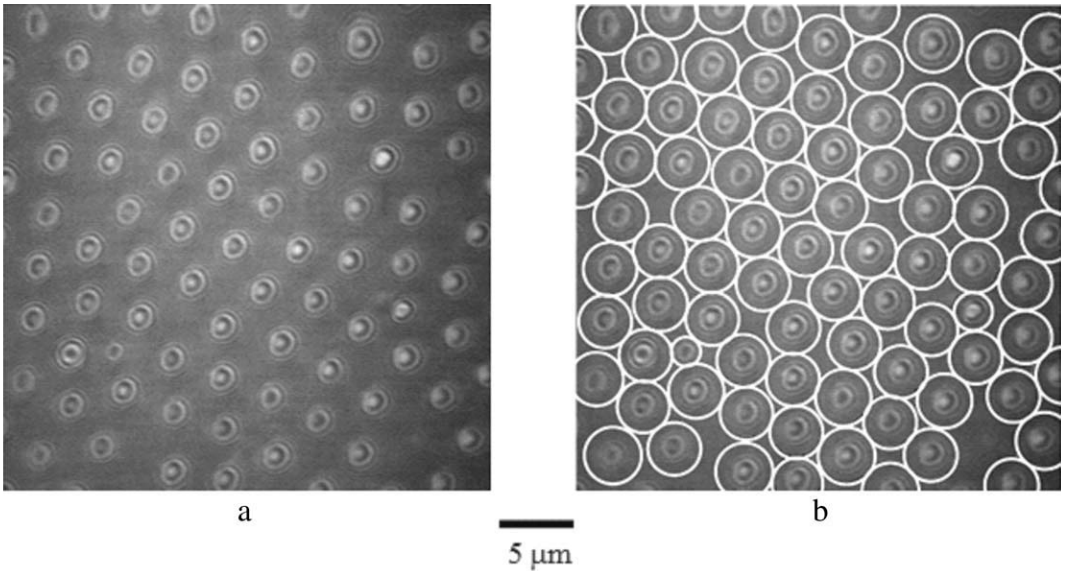

Figure 3. Optical microscopic image of particles obtained via seeded dispersion polymerization of MMA using fluorescent PMMA latex particles as seed. (a) Original image, (b) with particle perimeters outlined.

end groups present on the particle surface provided sufficient stability to the final latex even when the surfactants were removed. Figure 1 shows an image of the particles taken using the confocal microscope. In confocal microscopy, only the fluorescent part of the specimen can be detected and imaged. The bright dots in the confocal microscope image (Figure 1) indicate the fluorescence of the particles prepared by this method.

Seeded Dispersion Polymerization

A SEM image of the particles obtained via the seeded dispersion polymerization of MMA using the fluorescent PMMA latex particles as seed is shown in Figure 2. The particle size (D_v) is 3.36 μm. Some smaller particles can be seen in the SEM image to have been produced outside the main large particle size population.

The number concentration of the final particles was 3.72×10^{12} L^{-1}. Therefore, the average number of seed particles in each final particle can be calculated by eq. (5), indicating that there should be theoretically 3.5 seed particles in each final particle.

$$N = \frac{N_s}{N_f} = \frac{13.05 \times 10^{12} L^{-1}}{3.72 \times 10^{12} L^{-1}} = 3.5 \qquad (5)$$

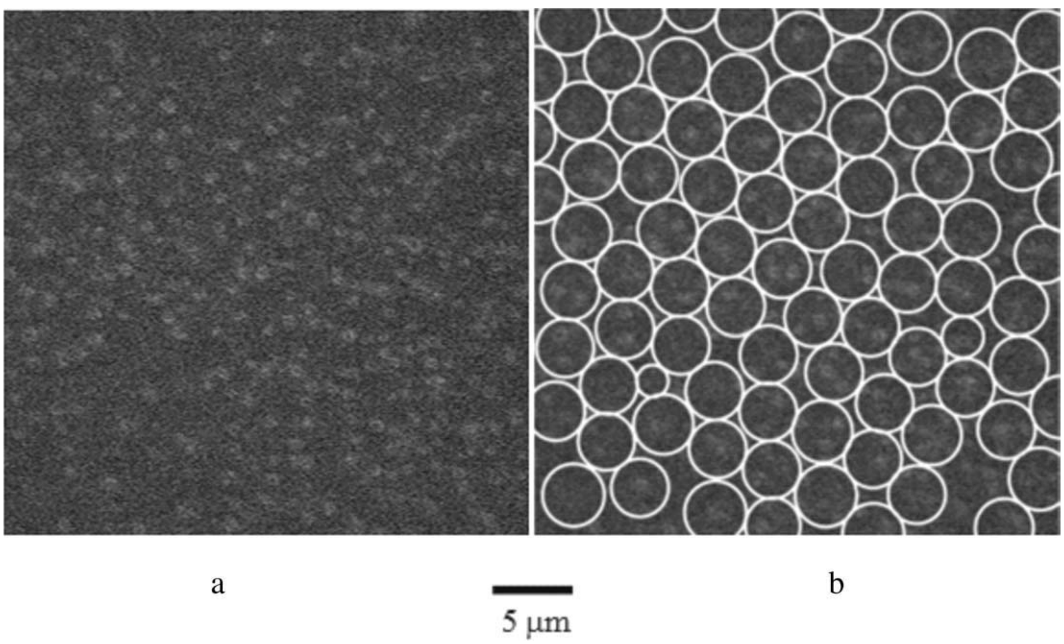

Figure 4. Confocal microscopic image of particles obtained via seeded dispersion polymerization of MMA using fluorescent PMMA latex particles as seed. (a) Original image, (b) with perimeters transposed from Figure 3(b).

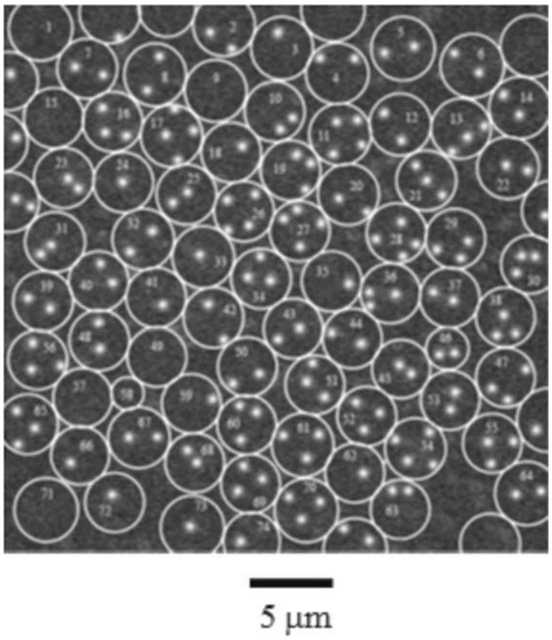

Figure 5. Confocal microscope image with bright spots highlighted and particles labeled.

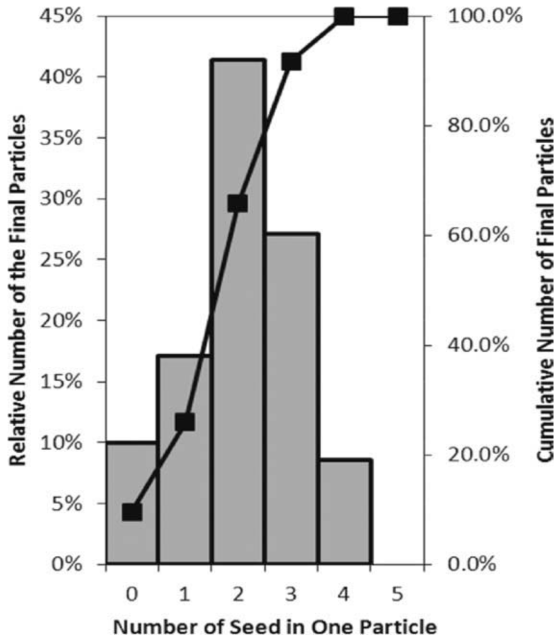

Figure 6. Distribution of the final particles containing different number of seed particles.

Locating the Seed Particles Inside the Final Particles

The location of the fluorescent seed particles inside the final particles was analyzed by confocal microscopy imaging. The confocal microscope can be operated in two different modes: an optical microscope mode and a confocal microscope mode. These two operation modes can be switched easily without moving the sample. The location of the final particles was revealed in the optical microscope mode. In the optical microscopic image [Figure 3(a)], the bright dots indicate the centers of the particles, and the particles are packed in an array. Hence, the outlines of the particles in this picture can be drawn as shown in Figure 3(b). The fluorescent seed particles were imaged and located in the confocal microscope mode. In the confocal microscopic image [Figure 4(a)], only the fluorescent parts of the sample are visible (bright dots in this picture), whereas the nonfluorescent parts remain dark. As shown in Figure 4(a), the confocal microscopic image of the sample at the same location as in Figure 3, the positions of the particles are the same. Therefore, the particle positions in Figure 3(b) can be overlaid onto Figure 4(a) as shown in Figure 4(b). With the fluorescent seed particles highlighted and the final large particles labeled (Figure 5), the location of the seed particles in the final particles could be analyzed. Almost every final particle contained one or more bright spots, which should come from the initial seed particles. The detailed particle counting statistics are summarized in Table V. The distribution of the final particles containing different number of bright spots is shown in Figure 6. Some particles contained two or more bright spots. This indicates that particle aggregation occurred during the seeded dispersion polymerization. Most of the bright spots are separated from each other, which could imply that this kind of aggregation does not take place between the seed particles themselves (i.e., prior to growth), but rather between the growing particles. Some small particles (numbered as 57 and 58 in Figure 5) obviously come from secondary nucleation. However, some large particles (numbers 1, 9, 15, 49, and 71 in Figure 5) also have no bright spots. There could be two reasons for this: (1) confocal microscopy collects only the fluorescent signal from a thin layer, and hence, if there happens to be no fluorescent sites present in this imaged layer of the particle, this particle will contain no bright spots; or (2) these particles form from secondary nucleation and secondary nucleation takes place early in the seeded dispersion polymerization process.

Most of the particles shown in Figure 5 have one to three bright spots, as shown in Figure 6. There are 74 particles and 151 bright spots (not counting the particles on the edge of the image and the bright spots inside these particles). On average, there are two bright spots, attributed to the initial seed particles in each final particle. Theoretically, there should be about 3.5 seed particles in each final particle (N(initial) $= 13.05 \times 10^{12}$ L^{-1}, N(final) $= 3.72 \times 10^{12}$ L^{-1}). This discrepancy is explained by the fact that the confocal microscope collects only the fluorescent signal from a relatively thin layer ($\sim$ 500 nm), whereas the particle size of the final particles is much larger (3.36 μm). Therefore, the real number of the bright spots (seed particles) in each particle should be >2.

Table V. Statistical Data of the Particles and Seeds from Figure 5

Number of bright spots in one particle	Number of the particles
0	7
1	12
2	29
3	19
4	6
>4	0

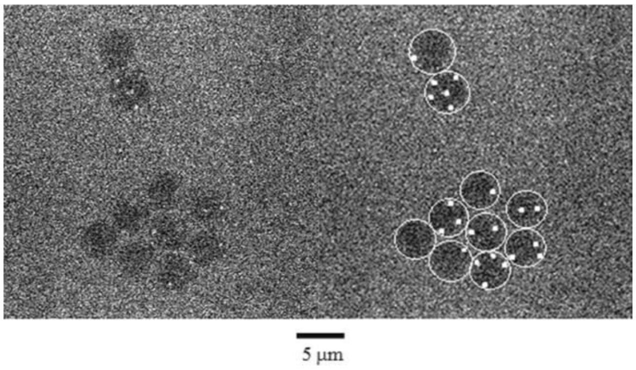

Figure 7. A snapshot of the movie captured by confocal microscopy that shows the profile inside the particles. Left: original image; right: enhanced image with particles circled and seeds highlighted.

To verify the preceding statement, all of the seed particles present within the final particles were imaged by scanning the final particles from the bottom to the top under the confocal microscope at a speed of one frame per 100 nm depth. During this analysis, the particles were immersed in a 0.01-m*M* Rhodamine B aqueous solution so that the particles appear dark with a bright background in the confocal microscope. The seeds inside the particles are bright as well because they are fluorescent. A movie was taken of this sample and the profile inside each particle was recorded. Figure 7 shows a snapshot image taken from the movie. There are 10 particles in the snapshot and 37 seed particles can be counted. On average, there are 3.7 seed particles in each large particle. This agrees well with the theoretical value of 3.5 seeds per particle and is an indication that all of the seed particles participated in the formation of the final particles and ended up inside the final particles. As the seed particles are separated inside the final particles, one could conclude that aggregation took place between growing seed particles rather than the initial seed particles.

CONCLUSIONS

PMMA latex particles containing a fluorescent agent were used to investigate the mechanism of seeded dispersion polymerization. These were first prepared by a two-step miniemulsion polymerization process and were then used as seed in seeded dispersion polymerization of MMA. The seed particles were located in the final particles obtained from seeded dispersion polymerization by means of confocal microscopy analysis. All seed particles were found to end up inside the final particles and more than one seed particle was detected in most of the final particles. These were well-separated within the micron-size particles, indicating that aggregation of the seed particles occurred after some particle growth during the reaction. A small population of final particles containing no seed particles was the result of secondary nucleation.

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