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Efficient Synthesis of Amine-Functional Diblock Copolymer Nanoparticles via RAFT Dispersion Polymerization of Benzyl Methacrylate in Alcoholic Media

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Supporting Information

ABSTRACT: Benzyl methacrylate (BzMA) is polymerized via reversible addition—fragmentation chain transfer (RAFT) chemistry under alcoholic dispersion polymerization conditions in ethanol using a poly(2-(dimethylamino)ethyl methacrylate) (PDMA) chain transfer agent (CTA) at 70 °C. In principle, polymerization-induced self-assembly can lead to the formation of either spherical micelles, worm-like micelles, or vesicles, with the preferred morphology being dictated by the hydrophilic—hydrophobic balance of the PDMA–PBzMA diblock copolymer chains. Very high monomer conversions (>99%) are routinely obtained within 24 h as judged by ¹H NMR



studies. Moreover, THF GPC analyses confirmed that relatively low polydispersities ($M_w/M_n < 1.30$) are achieved, indicating reasonably good pseudoliving character. A detailed phase diagram was constructed using a PDMA₃₁ macro-CTA by systematically varying both the target degree of polymerization of the PBzMA block and the total solids concentration of the reaction solution. This phase diagram can be used to reliably predict the synthesis conditions required to produce pure phases, rather than merely mixed phases (e.g., spheres plus worms or worms plus vesicles). Finally, these PDMA–PBzMA diblock copolymer nanoparticles remain colloidally stable when transferred from ethanol into water; aqueous electrophoresis studies confirmed that the particles acquire appreciable cationic character below pH 7 due to protonation of the PDMA stabilizer chains.

■ INTRODUCTION

Over the past decade or so, reversible addition—fragmentation chain transfer (RAFT) polymerization¹ has been utilized by many research groups to prepare a wide range of diblock copolymers with relatively low polydispersities. Amphiphilic diblock copolymers are of particular interest since they can self-assemble in aqueous solution to form various morphologies.^{2–6} They are already widely used as polymeric surfactants (e.g., Pluronics) and have many other potential applications, such as drug delivery,^{7–9} MRI imaging,¹⁰ water purification membranes,¹¹ nanoreactors,^{12,13} and inorganic templates for photonics.¹⁴

Traditionally, amphiphilic block copolymers were synthesized and isolated, with a subsequent processing step being used to induce self-assembly. Typical processing techniques include a solvent switch, a pH switch, or thin film rehydration.^{15–17} However, such self-assembly is usually only achievable at relatively low copolymer concentration, which makes the scaled-up production of diblock copolymer-based "nano-objects" rather problematic. Recently, a polymerizationinduced self-assembly approach has been investigated, which enables bespoke organic nanoparticles to be prepared directly at much higher concentrations.^{18,19} For example, Charleux et al. have optimized various RAFT aqueous emulsion formulations in order to prepare various "nano-objects" based on waterimmiscible commodity monomers such as styrene or *n*-butyl acrylate, with relatively high monomer conversions being

achieved.²⁰⁻²² Alternatively, we have shown that RAFT aqueous dispersion polymerization formulations based on 2hydroxypropyl methacrylate (HPMA) appear to have considerable commercial potential, since this approach allows efficient syntheses of pure block copolymer phases comprising spheres, worms, or vesicles.^{4,23,24} Moreover, a *predictive* phase diagram for one particular formulation has been recently published,²³ while careful sampling of the reaction solution of a second formulation has shed new light on our understanding of the mechanism of the worm-to-vesicle transition during the in situ evolution of diblock copolymer morphologies.²⁴ Finally, Pan and co-workers reported that various copolymer morphologies can be obtained by RAFT alcoholic dispersion polymerization using polystyrene as the core-forming (i.e., structure-directing) block.^{25–27} However, in this case monomer conversions are typically substantially incomplete (ranging from 30 to 70%)^{28,29} due to the relatively slow polymerization of styrene. This disadvantage almost certainly makes such formulations unsuitable for industrial applications, since the cost of removing the unreacted styrene monomer would be prohibitive.

For surfactant amphiphiles, Israelachvili and co-workers³⁰ have demonstrated that the final morphology depends on the dimensionless "packing parameter", p, as defined by eq 1.³¹

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^aHere only a spherical morphology is depicted, but systematic variation of the reaction conditions allows either PDMA–PBzMA worms or vesicles to be targeted (see main text).

$$p = \frac{\nu}{a_0 l_c} \tag{1}$$

Here v, a_0 , and l_c represent the volume of the hydrophobic chains, the optimal area of the headgroup, and the length of the hydrophobic tail, respectively. If $p \le 0.33$, the self-assembly of amphiphiles in aqueous solution produces spherical micelles, whereas anisotropic worm-like micelles are obtained when $0.33 \le p \le 0.50$ and vesicles are formed when p > 0.50.³¹ It is generally believed that this geometric argument also holds for amphiphilic diblock copolymers as well as conventional surfactants.³⁰⁻³³

Recently, Charleux and co-workers have reported^{18,22} that the polymerization-induced self-assembly of diblock copolymers via RAFT aqueous emulsion polymerization is much more difficult to achieve if the stabilizer chain has polyelectrolyte character. This is because strong lateral electrostatic repulsive forces prevent the efficient packing of highly charged chains within a coronal layer. In principle, this problem can be avoided by addition of salt to screen the electrostatics or by diluting the stabilizer charge density by copolymerizing the ionic monomer with a nonionic monomer.²² We have also recently explored a third possibility in the context of RAFT aqueous dispersion polymerization, which is the judicious use of binary mixtures of polyelectrolyte and nonionic macro-CTAs in order to modulate the charge density within the coronal layer via an entropic mixing mechanism.³⁴

Herein we report the polymerization-induced self-assembly of an all-methacrylic diblock copolymer via RAFT alcoholic dispersion polymerization. Unlike the styrene-based formulations reported by Pan and co-workers,^{26,27} we show that this new formulation enables very high monomer conversions to be achieved within 24 h at 70 °C. A poly(2-(dimethylamino)ethyl methacrylate) (PDMA) macro-CTA with a fixed mean degree of polymerization (DP, or x) is chain-extended using benzyl methacrylate (BzMA) so as to systematically vary the packing parameter, p, and hence the morphology of the diblock copolymer nanoparticles (see Scheme 1). A detailed phase diagram for this formulation is constructed by varying the total solids concentration of the reaction solution, using the approach described by Sugihara et al.²³ One motivation for this study was the realization that the polyelectrolytic block copolymer self-assembly problem described above for aqueous formulations might be addressed by such an alcoholic formulation. More specifically, the synthesis of a weak polyelectrolyte-based diblock copolymer via RAFT dispersion polymerization in alcohol ensures that the polyelectrolyte chains remain uncharged during their self-assembly, while the subsequent transfer of these neutral "nano-objects" into

aqueous solution at an appropriate solution pH allows the PDMA stabilizer chains to acquire polyelectrolytic character.

EXPERIMENTAL SECTION

Materials. All reagents were purchased from Sigma-Aldrich (UK) and used as received unless otherwise noted. 4,4'-Azobis(4-cyanovaleric acid) (ACVA)) or 2,2'-azobis(isobutyronitrile) (AIBN) was used as an initiator. Benzyl methacrylate (96%) was passed through a column of inhibitor remover (also purchased from Sigma) prior to use.

Synthesis of 4-Cyano-4-(2-phenylethanesulfanylthiocarbonyl)sulfanylpentanoic Acid (PETTC). 2-Phenylethanethiol (10.5 g, 76 mmol) was gradually added over 10 min to a stirred suspension of sodium hydride (60% in oil) (3.15 g, 79 mmol) in diethyl ether (150 mL) at between 5 and 10 °C. Vigorous evolution of hydrogen gas was observed, and the grayish suspension was slowly transformed into a white viscous slurry of sodium phenylethanethiolate over 30 min. The reaction mixture was cooled to 0 °C, and carbon disulfide (6.0 g, 79 mmol) was gradually added to produce a thick yellow precipitate of sodium 2-phenylethanetrithiocarbonate, which was collected by filtration after 30 min and subsequently used in the next step without further purification. Solid iodine (6.3 g, 0.025 mol) was gradually added to a suspension of sodium 2-phenylethanetrithiocarbonate (11.6 g, 0.049 mol) in diethyl ether (100 mL). This reaction mixture was then stirred at room temperature for 1 h, and the insoluble white precipitate of sodium iodide was removed by filtration. The yellow-brown filtrate was washed with an aqueous solution of sodium thiosulfate to remove excess iodine, dried over sodium sulfate, and then evaporated to yield bis(2-phenylethanesulfanylthiocarbonyl) disulfide (~100% yield). A solution of 4,4'-azobis(4-cyanopentanoic acid) (ACVA) (2.10 g, 0.0075 mol) and bis(2-phenylethane sulfanylthiocarbonyl) disulfide (2.13 g, 0.005 mol) in ethyl acetate (50 mL) was degassed by nitrogen bubbling and heated at reflux under a dry nitrogen atmosphere for 18 h. After removal of the volatiles under vacuum, the crude product was washed with water (five 100 mL portions). The organic phase was concentrated and purified by silica chromatography using a mixed eluent (petroleum ether:ethyl acetate = 7:3, gradually increasing to 4:6) to afford 4-cyano-4-(2phenylethanesulfanylthiocarbonyl)sulfanylpentanoic acid as a yellow oil. ¹H NMR (400.13 MHz, CD₂Cl₂, 298 K): δ (ppm) = 1.89 (3H, -CH₃), 2.34-2.62 (m, 2H, -CH₂), 2.7 (t, 2H, -CH₂), 3.0 (t, 2H, -CH₂), 3.6 (t, 2H, -CH₂), 7.2-7.4 (m, 5H, aromatic). ¹³C NMR (400.13 MHz, CD_2Cl_2 , 298 K): δ (ppm) = 24.2 (CH₃), 29.6 (CH₂CH₂COOH), 30.1 (CH₂Ph), 33.1 (CH₂CH₂COOH), 39.9 (SCH₂CH₂Ph), 45.7 (SCCH₂), 118.6 (CN), 127.4, 128.8, 129.2, 144.3 (Ph), 177.4 (C=O), 222.2 (C=S).

Synthesis of Poly(2-(dimethylamino)ethyl methacrylate) (PDMA) Macro-CTA Agent. A round-bottomed flask was charged with 2-(dimethylamino)ethyl methacrylate (DMA; 10.0 g, 63 mmol), PETTC (0.432 g, 1.27 mmol), ACVA (36 mg, 0.127 mmol), and THF (10.0 g) (target DP = 50). The sealed reaction vessel was purged with nitrogen and placed in a preheated oil bath at 70 °C for 6 h. The resulting polymer (monomer conversion = 67%; M_n = 5500 g mol⁻¹,

 $M_w/M_n = 1.22$) was purified by precipitation into excess petroleum ether. The mean degree of polymerization (DP) of this PDMA macro-CTA was calculated to be 31 using ¹H NMR spectroscopy by comparing the integrated signals corresponding to the aromatic protons at 7.2-7.4 ppm with those due to the methacrylic polymer backbone at 0.4-2.5 ppm. This was repeated with an increased amount of DMA to synthesize a longer PDMA macro-CTA; in this case the amounts of reagents used were DMA (30.0 g, 190 mmol), PETTC (0.648 g, 1.9 mmol), ACVA (54.0 g, 0.19 mmol), and THF (30 g) (target $\overrightarrow{DP} = 100$). Again, these were sealed in a reaction vessel, purged with nitrogen, and then placed in an oil bath at 70 °C for 8 h 10 min. The resulting polymer (monomer conversion = 68%; M_n = 11 300 g mol⁻¹, $M_w/M_n = 1.19$) was purified by precipitation into excess petroleum ether. The mean DP of this PDMA macro-CTA was calculated to be 74 using ¹H NMR spectroscopy by comparing the integrated signals corresponding to the aromatic protons at 7.2-7.4 ppm with those due to the methacrylic polymer backbone at 0.4-2.5ppm.

Synthesis of Poly(2-(dimethylamino)ethyl methacrylate)– Poly(benzyl methacrylate) (PDMA–PBzMA) Diblock Copolymer Particles via Dispersion Polymerization in Ethanol. In a typical RAFT dispersion polymerization synthesis conducted at 17% w/w total solids, BzMA (2.50 g, 14.2 mmol), AIBN (0.90 mg, 0.006 mmol), and PDMA₃₁ macro-CTA (144 mg, 0.028 mmol) were dissolved in ethanol (13.23 g). The reaction mixture was sealed in a round-bottomed flask, purged with nitrogen gas for 15 min, and then placed in a preheated oil bath at 70 °C for 24 h. The final monomer conversion was determined by ¹H NMR analysis by integrating the PBzMA peak (CH₂) at 4.9 ppm to BzMA monomer vinyl peaks (CH₂) at 5.2 and 5.4 ppm.

In further PDMA–PBzMA diblock copolymer syntheses, the mean DP of the PBzMA block was systematically varied by adjusting the amount of BzMA monomer. Finally, syntheses were also conducted at varying total solids concentrations (either 10, 13, 20, 25, or 29% w/w) simply by adjusting the amount of ethanol added to the formulation.

Copolymer Characterization. Diblock copolymer molecular weight distributions were assessed using gel permeation chromatography (GPC). The GPC setup comprised two 5 μ m (30 cm) "Mixed C" columns and a WellChrom K-2301 refractive index detector operating at 950 ± 30 nm. THF eluent contained 2.0% v/v triethylamine and 0.05% w/v butylhydroxytoluene (BHT) was used at a flow rate of 1.0 mL min⁻¹. A series of ten near-monodisperse linear poly(methyl methacrylate) standards (M_p ranging from 1280 to 330 000 g mol⁻¹) were purchased from Polymer Laboratories (Church Stretton, UK) and employed for calibration using the above refractive index detector.

¹H NMR spectra were acquired on a Bruker 400 MHz spectrometer in either CDCl₃ or CD₂Cl₂. All chemical shifts are reported in ppm (δ). TEM studies were conducted using a Philips CM 100 instrument operating at 100 kV. To prepare TEM samples, 5.0 μ L of a dilute aqueous copolymer solution was placed onto a carbon-coated copper grid, stained with uranyl formate, and dried under ambient conditions. DLS measurements were conducted using a Malvern Instruments Zetasizer Nano series instrument equipped with a 4 mW He–Ne laser operating at 633 nm, an avalanche photodiode detector with high quantum efficiency, and an ALV/LSE-5003 multiple tau digital correlator electronics system. Aqueous electrophoresis measurements were performed on 0.01% w/v aqueous copolymer solutions in 0.01 M NaCl using the same Malvern Instruments Zetasizer Nano series instrument. The solution pH was adjusted by the addition of 0.01 M HCl or 0.01 M KOH using an autotitrator.

Transmission electron microscopy (TEM) imaging was performed at 100 kV on a Phillips CM100 instrument equipped with a Gatan 1 k CCD camera. Aggregate solutions were diluted with ethanol at 20 °C to generate 0.20% w/w dispersions. Copper/palladium TEM grids (Agar Scientific, UK) were surface-coated in-house to yield a thin film of amorphous carbon. The grids were then plasma glow-discharged for 30 s to create a hydrophilic surface. Each aqueous diblock copolymer dispersion (0.20% w/w, 10 μ L) was placed onto a freshly glowdischarged grid for 1 min and then blotted with filter paper to remove excess solution. To stain the deposited nanoparticles, a 0.75% w/w aqueous solution of uranyl formate (10 μL) was placed via micropipet on the sample-loaded grid for 20 s and then carefully blotted to remove excess stain. Each grid was then carefully dried using a vacuum hose.

RESULTS AND DISCUSSION

In a recent communication, we reported that simply replacing styrene with BzMA as the core-forming monomer in RAFT



Figure 1. Kinetic data derived from ¹H NMR spectroscopy studies of the RAFT dispersion polymerization of benzyl methacrylate at 70 °C in ethanol using a PDMA₃₁ macro-CTA and AIBN initiator at a total solids concentration of 17%. Target diblock composition was PDMA₃₁-PBzMA₅₀₀; [macro-CTA]/[AIBN] = 5.0.



Figure 2. Evolution of the number-average molecular weight and polydispersity (M_w/M_n) with conversion for the RAFT dispersion polymerization of benzyl methacrylate at 70 °C in ethanol using a PDMA₃₁ macro-CTA and AIBN initiator at a total solids concentration of 17%. The target diblock composition was PDMA₃₁–PBzMA₅₀₀, and the [macro-CTA]/[AIBN] molar ratio was 5.0.

alcoholic dispersion polymerization syntheses leads to much higher monomer conversions being attained in each case when using four different methacrylate-based RAFT macro-CTAs.³⁵ Such all-methacrylic formulations represent a potentially important breakthrough, since they are much more amenable to industrial scale-up. Previously, the use of a weak polyacid macro-CTA was emphasized.³⁵ In the present study, we focus on a complementary weak polybase macro-CTA, as exemplified by PDMA. Thus, a PDMA macro-CTA was synthesized by solution polymerization in THF at 70 °C (see Scheme 1).



Figure 3. Gel permeation chromatography traces obtained for the RAFT dispersion polymerization of BzMA in ethanol at 70 °C using a PDMA₃₁ macro-CTA and AIBN initiator at a total solids concentration of 17%. The target composition was PDMA₃₁– PBzMA₅₀₀, and the [macro-CTA]/[AIBN] molar ratio was 5.0.



Figure 4. Phase diagram constructed for the PDMA₃₁–PBzMA_x RAFT dispersion polymerization formulation by systematic variation of the mean target degree of polymerization of PBzMA (x) and the total solids concentration using a fixed mean degree of polymerization of 31 for the PDMA macro-CTA. The mean DP of the PBzMA block in each case was determined by ¹H NMR spectroscopy (in either CD₂Cl₂ or CDCl₃), by comparing the integrated benzyl protons at 4.9 ppm with the methylene vinyl protons due to the BzMA monomer at 5.2 and 5.4 ppm.

After purification by precipitation into excess petroleum ether, its mean DP was determined to be 31 as judged by ¹H NMR spectroscopy, while THF GPC analysis indicated a final polydispersity of 1.22. This PDMA₃₁ macro-CTA was then chain-extended with BzMA via RAFT dispersion polymerization in ethanol at 70 °C to produce a series of PDMA₃₁– PBzMA_x diblock copolymers. Since the PBzMA chains are insoluble in ethanol, a range of copolymer morphologies can be generated via *in situ* self-assembly by varying the DP of the PBzMA chains and hence adjusting v and l_c for a fixed a_0 according to eq 1. In each case the alcohol-soluble PDMA chains act as an effective steric stabilizer for the diblock copolymer "nano-object".

A kinetic study of the BzMA polymerization was conducted when targeting a DP of 500 for the core-forming block (Figure 1). 1 H NMR analysis indicated that a BzMA conversion of 89%



Figure 5. TEM images obtained for $PDMA_{31}-PBzMA_x$ diblock copolymer particles synthesized at a total solids concentration of 17% via RAFT dispersion polymerization in ethanol at 70 °C using a PDMA₃₁ macro-CTA, AIBN initiator, and a [macro-CTA]/[AIBN] molar ratio of 5.0. Systematic variation of the target degree of polymerization, *x*, of the core-forming PBzMA block enables the particle morphology to be adjusted from (a) spheres (*x* = 60) to (b) worms (*x* = 80) to (c) a worm/vesicle mixed phase (*x* = 90) to (d) pure vesicles (*x* = 100).

was obtained after 12 h, with essentially full conversion being achieved after 24 h. After just 2 h, there is a discernible increase in the rate of polymerization, which is consistent with previously reported observations for a RAFT aqueous dispersion polymerization formulation.²⁴ This time period corresponds to the onset of micellar nucleation, since visual inspection confirms a concomitant increase in the turbidity of the reaction solution. This suggests that the enhanced rate of polymerization is due to the solubilization of unreacted BzMA monomer within the growing PBzMA nanoparticles. We believe that such monomer partitioning leads to a relatively high local monomer concentration and that the nascent growing nanoparticles effectively act as "nanoreactors". However, this hypothesis is rather difficult to confirm experimentally. Nevertheless, analysis of the kinetic data shown in Figure 1 suggests that micellar nucleation leads to an apparent rate enhancement by at least a factor of 4.5, which compares quite well with that recently reported by Blanazs et al. for the RAFT aqueous dispersion polymerization of HPMA. For the RAFT synthesis of a PBzMA homopolymer in ethanol, macroscopic precipitation is observed when targeting a mean DP as low as 20. However, the enhanced rate of polymerization observed after 2 h in Figure 1 occurs at a monomer conversion of around 10%, which corresponds to a PBzMA block DP of \sim 50 (since the final target DP is 500). Visual inspection confirms that this rate enhancement coincides with the onset of nucleation, since the reaction solution becomes distinctly turbid at around 2 h. This critical DP is significantly higher than that required for precipitation of PBzMA homopolymer because the solvent mixture is not pure ethanol, but a mixture of ethanol and BzMA monomer. The unreacted monomer acts as a cosolvent for the PBzMA chains and hence delays the onset of nucleation. It is also likely that conjugation of the solvatophobic PBzMA chains to the solvatophilic PDMA stabilizer chains

Table 1. Monomer Conversions, Intensity-Average Particle Diameters, and GPC Molecular Weights and Polydispersities Obtained for PDMA₃₁–PBzMA_x Diblock Copolymer Particle Syntheses Conducted at a Total Solids Concentration of 10-29%w/w by RAFT Alcoholic Dispersion Polymerization in Ethanol at 70 °C

				GPC		DLS ^a		TEM	
solids (%)	target composition	BzMA % conv	actual BzMA DP	M _n	$M_{\rm w}/M_{\rm n}$	diameter	PDI	morphology	
10	PDMA31-PBzMA40	92	37	10 300	1.31	65	0.29	spheres	
	PDMA31-PBzMA60	97	58	12 500	1.36	96	0.13	spheres + worms	
	PDMA31-PBzMA20	92	64	15 000	1.25	179	0.29	spheres + worms	
	PDMA31-PBzMA80	89	71	15 100	1.31	330	0.39	worms	
	PDMA31-PBzMA90	84	76	16 500	1.24	979	0.63	worms + vesicles	
	PDMA31-PBzMA100	91	91	17 300	1.33	450	0.25	vesicles + worms	
	PDMA31-PBzMA120	85	102	22 000	1.25	411	0.21	vesicles	
13	PDMA31-PBzMA60	100	60	14 000	1.27	75	0.21	worms + spheres	
	PDMA31-PBzMA20	95	67	17 000	1.19	334	0.81	worms + spheres	
	PDMA31-PBzMA500	6^b	30	12 500	1.28	58	0.2	spheres	
	PDMA31-PBzMA500	12^{b}	60	15 000	1.27	73	0.07	spheres	
	PDMA31-PBzMA65	95	62	16 100	1.19	154	0.34	worms + spheres	
17	PDMA31-PBzMA500	6^b	30	12 500	1.28	58	0.20	spheres	
	PDMA31-PBzMA500	12^{b}	60	15 000	1.27	73	0.07	spheres	
	PDMA31-PBzMA65	95	62	16 100	1.19	154	0.34	worms + spheres	
	PDMA31-PBzMA70	100	70	15 000	1.30	310	0.32	worms	
	PDMA31-PBzMA80	100	80	16 300	1.29	1274	0.89	worms	
	PDMA31-PBzMA90	100	90	22 000	1.19	1056	0.81	worms + vesicles	
	PDMA31-PBzMA100	98	98	22 300	1.22	642	0.3	vesicles	
20	PDMA31-PBzMA20	97	68	16 400	1.22	333	0.85	worms + spheres	
	PDMA31-PBzMA90	98	88	19 300	1.21	693	0.89	worms + vesicles	
23	PDMA ₃₁ -PBzMA ₄₀	97	39	11 250	1.24	48	0.14	spheres	
	PDMA31-PBzMA60	100	60	14 800	1.21	301	0.28	worms	
	PDMA31-PBzMA80	97	78	16 300	1.28	541	0.56	worms	
	PDMA31-PBzMA85	100	85	17 900	1.25	631	0.52	worms	
	PDMA31-PBzMA100	98	98	19 150	1.31	563	0.2	worms + vesicles	
	PDMA31-PBzMA120	98	118	24 800	1.18	529	0.26	vesicles	
	PDMA31-PBzMA500	7^b	35	14 000	1.23	30	0.21	spheres	
	PDMA31-PBzMA500	14^b	75	20 000	1.22	57	0.13	spheres	
29	PDMA ₃₁ -PBzMA ₈₀	100	80	18 800	1.21	510	0.76	spheres + worms	
	PDMA ₃₁ -PBzMA ₉₀	100	90	19 700	1.24	542	0.61	worms	
	PDMA ₃₁ -PBzMA ₁₀₀	100	100	20 000	1.30	256	0.26	worms + vesicles	
	PDMA ₃₁ -PBzMA ₁₁₀	100	110	22 900	1.23	490	0.41	vesicles	

^{*a*}Intensity-average diameter. PDI is the polydispersity calculated using cumulants analysis software provided by the instrument manufacturer (Malvern). ^{*b*}Indicates data obtained from the analysis of multiple samples extracted from an *in situ* polymerization in which higher values were targeted for the final DP of the PBzMA block.



Figure 6. Zeta potential vs pH curves obtained for $PDMA_{31}$ – $PBzMA_{40}$ spheres (\blacksquare), $PDMA_{31}$ – $PBzMA_{80}$ worms (O), and $PDMA_{31}$ – $PBzMA_{190}$ vesicles (\blacktriangle).

delays precipitation of the former block. Very recently, similar kinetic data have been reported by Charleux et al. for RAFT dispersion polymerization of BzMA conducted in various ethanol/water mixtures.³⁶ In this case, the addition of water worsens the solvency for the growing PBzMA chains, which leads to much faster rates of polymerization (~100% conversion within 2 h at 80 °C). In contrast, herein we focus on a purely alcoholic formulation to ensure that the PDMA stabilizer chains (p $K_a \sim 7.0-7.5$) remain nonprotonated.

The evolution of molecular weight with conversion was also monitored to assess the character of the BzMA polymerization (see Figure 2). The observed linear relationship indicates a well-controlled pseudoliving RAFT polymerization. Polydispersities remained between 1.20 and 1.30 throughout the reaction, with the final PDMA₃₁–PBzMA₅₀₀ diblock copolymer having an M_w/M_n of 1.25. GPC traces were invariably unimodal with little or no tailing, which indicated a relatively high blocking efficiency and suggested that relatively few polymer chains were terminated prematurely (see Figure 3).

The PDMA₃₁ macro-CTA was prepared on a relatively large scale so that a detailed phase diagram could be constructed using a *single batch* of macro-CTA. This is an important point,

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Figure 7. TEM images obtained for (top row) $PDMA_{31}-PBzMA_{40}$ spheres, (middle row) $PDMA_{31}-PBzMA_{80}$ worms, and (bottom row) $PDMA_{31}-PBzMA_{120}$ vesicles. The continuous phase in which these nano-objects were dispersed prior to TEM grid preparation was either ethanol (a–c), an acidic aqueous solution at pH 3 (d–f), or an alkaline aqueous solution at pH 10 (g–i).

because it is not trivial to precisely target a particular mean DP for a macro-CTA, and ideally this parameter should be held constant. The phase diagram shown in Figure 4 was generated by systematic variation of the total solids concentration for the BzMA polymerization from 10 to 29% w/w and the mean DP of the core-forming PBzMA block. For a given DP of the macro-CTA, the latter parameter dictates the molecular curvature (or packing parameter p) of the diblock copolymer chains, which in turn determines the final copolymer morphology (as judged by *post mortem* TEM studies).

For a given total solids content, there is a gradual evolution from spheres to worms to vesicles as the target DP of the PBzMA chains is increased, with mixed phases always being observed between the three pure phases. This important observation is illustrated in Figure 5, which depicts a series of TEM images recorded for BzMA polymerizations conducted at 17% w/w solids. Well-defined spheres are obtained at a mean PBzMA DP of 60, whereas a pure worm phase is identified for a DP of 80. A "worm plus vesicle" mixed phase is observed at a mean PBzMA DP of 90, while the sole phase produced when targeting a PDMA₃₁-PBzMA₁₀₀ diblock composition is polydisperse vesicles. The same general behavior is observed at each of the concentrations investigated. It is interesting to compare this phase diagram with that recently published by our group for a related RAFT aqueous dispersion polymerization formulation using poly(2-hydroxypropyl methacrylate) (PHPMA) as the core-forming block.²³ In both cases, the worm phase space is relatively narrow. However, there are important qualitative differences: diblock copolymer vesicles could only be generated in aqueous solution by targeting a high DP for the core-forming block and also by performing the polymerization of 2-hydroxypropyl methacrylate at relatively high solids (22.5-25%). In contrast, the present alcoholic formulation allows vesicles to be generated at just 10% solids, which suggests that the copolymer concentration has a much weaker influence on particle morphology in this case. In this sense the data shown in Figures 4 and 5 are consistent with the observations made by Blanazs and co-workers, who also found that spheres, worms, or vesicles could be obtained at 10% solids using an alternative RAFT aqueous dispersion polymerization formulation.²⁴

Hence it seems likely that both the precise chemical nature and mean DP of the stabilizer chains may also play an important role in determining the final copolymer morphology. In addition, differences in the interaction of the core-forming block with the continuous phase may be important. PHPMA is only weakly hydrophobic in the presence of water: although this homopolymer is water-insoluble, it is known to exhibit thermoresponsive behavior when conjugated to a more hydrophilic block if its DP is relatively low.³⁷ In contrast, ethanol is a strong non-solvent even for relatively short PBzMA chains (DP ~ 20).

All of the experimental results (including DLS particle diameters and THF GPC data) associated with the phase diagram shown in Figure 4 are summarized in Table 1. TEM studies confirm that the spherical diblock copolymer nanoparticles can exhibit relatively narrow size distributions (see Figure 5a), whereas worms or vesicles (or mixed phases) invariably possess significantly higher polydispersities. However, the Stokes-Einstein equation only reports a "sphereequivalent" diameter, so the DLS technique should be treated with some caution when characterizing the worm phase. The relatively high vesicle polydispersities indicated by DLS studies are consistent with the corresponding TEM images obtained for these dispersions. Regardless of the final copolymer morphology, the diblock copolymer chains had relatively low polydispersities $(M_w/M_n = 1.18 - 1.36)$; vs poly(methyl methacrylate) calibration standards) as judged by THF GPC, which suggests reasonably good control. Perhaps more importantly,

Table 2. Monomer Conversions, GPC Molecular Weights and Polydispersities, Intensity-Average Particle Diameters and Morphologies Obtained for $PDMA_{74}$ –PBzMA_x Diblock Copolymer Particles Prepared at 10% Solids by RAFT Alcoholic Dispersion Polymerization of Benzyl Methacrylate in Ethanol at 70 °C

			GPC		DLS ^a		TEM
target composition	BzMA % conv	actual DP of PBzMA	$M_{\rm n}$	$M_{\rm w}/M_{\rm n}$	diameter	PDI	morphology
PDMA74-PBzMA100	88	88	19 200	1.40	36	0.06	spheres
PDMA74-PBzMA250	92	230	40 400	1.22	50	0.04	spheres
PDMA74-PBzMA500	96	480	68 800	1.25	73	0.05	spheres
PDMA74-PBzMA800	97	776	104 000	1.23	95	0.01	spheres
PDMA74-PBzMA1000	93	930	116 000	1.34	108	0.04	spheres

^aIntensity-average diameter. PDI is the polydispersity calculated using cumulants analysis software provided by the manufacturer (Malvern).



Figure 8. TEM images obtained for PDMA₇₄–PBzMA_x diblock copolymer particles synthesized at a total solids concentration of 10% using RAFT dispersion polymerization in ethanol at 70 °C with a PDMA₇₄ macro-CTA, AIBN initiator and using a [macro-CTA]/[AIBN] molar ratio of 5.0. (a– e) Systematic variation of the target degree of polymerization, *x*, of the core-forming PBzMA block enables the particle diameter to be adjusted from 36 to 108 nm (as judged by DLS in ethanol). (f) Intensity-average diameter distributions for the same five samples of PDMA₇₄–PBzMA_x spherical nanoparticles in ethanol.

¹H NMR studies indicated that most BzMA conversions ranged from 90 to 100%, which is significantly higher than those reported by Pan and co-workers for polystyrene core-forming chains via RAFT dispersion polymerizations in alcohol.^{26,28} THF GPC data for all copolymers gave monomodal curves with little or no tailing, indicating good blocking efficiency and a fairly well controlled reaction with little evidence of premature termination.

Diluting these ethanol-dispersed "nano-objects" 1000-fold with water leads to protonation of the PDMA stabilizer chains (pK_a 7.0–7.5); hence the particles acquire cationic character. Aqueous electrophoresis studies were conducted on each of the three "pure phase" copolymer morphologies (i.e., spheres, worms, and vesicles) in order to assess their surface charge (zeta potential) and relative colloidal stabilities (see Figure 6). At low pH these dispersions each possess positive zeta potentials ranging from +34 to +47 mV, confirming protonation of the weakly polybasic PDMA chains. Isoelectric points were observed at around pH 7.6, pH 8.8, and pH 9.3 for the vesicles, spheres, and worms, respectively. The physical explanation for these subtle variations in isoelectric point is not clear and will be the subject of future work.

Dilute aqueous dispersions of spheres, worms and vesicles were prepared in turn at both pH 3 and pH 10 and subsequently analyzed to investigate whether their transfer from ethanol to water had any effect on copolymer morphology (see Figure 7). TEM studies indicated that the sphere and vesicle particle morphologies are not dramatically altered at either low or high pH. In the case of the worm-like morphology, some vesicles are generated at pH 10. This is believed to be due to the minimal cationic charge on the PDMA chains since this pH significantly exceeds their pK_{a} . However, pH-dependent DLS studies using an autotitrator indicate a significant increase in apparent size above pH 7 for all three copolymer morphologies. This is interpreted as evidence for flocculation at around the isoelectric point, rather than due to a significant change in the particle dimensions.

A second PDMA macro-CTA with a somewhat higher mean DP of 74 ($M_w/M_n = 1.19$) was also evaluated. Given the higher

copolymer curvature conferred by this longer PDMA₇₄ macro-CTA, it was anticipated that solely spherical nanoparticles would be produced over a relatively wide size range. Thus the target DP for the core-forming PBzMA block was systematically varied between 100 and 1000 (see Table 2). Although the final BzMA conversions for this series are slightly lower than previously observed (88-97%), the intensity-average particle diameter increased monotonically as longer PBzMA chains were targeted, while size distributions remained relatively narrow in all cases (DLS polydispersities were below 0.07 in each case). TEM images and DLS size distributions for each of these alcoholic dispersions are shown in Figure 8. Recent work by Zhang et al.³⁸ on diblock copolymer nanoparticles prepared via RAFT aqueous emulsion polymerization of styrene suggests that some chains may be buried within the particle cores; it has not been determined whether this is also true in the present study.

CONCLUSIONS

In summary, RAFT dispersion polymerization of benzyl methacrylate (BzMA) using a poly(2-(dimethylamino)ethyl methacrylate) (PDMA) macro-CTA in ethanol at 70 °C leads to the efficient production of near-monodisperse diblock copolymers, which self-assemble in situ to form spheres, worms, or vesicles depending on the precise reaction conditions. Near-monodisperse sterically stabilized spherical nanoparticles can be reproducibly synthesized with mean diameters that can be readily adjusted from 36 to 108 nm simply by varying the degree of polymerization of the coreforming PBzMA block. A diblock copolymer worm phase can also be accessed; such worms are relatively polydisperse in terms of their lengths but have well-defined widths ($\sim 10-20$ nm). Relatively polydisperse vesicles with intensity-average diameters ranging from 411 to 642 nm can also be obtained. For a fixed PDMA stabilizer DP of 31, a detailed phase diagram has been constructed. The observed particle morphology is very sensitive to the target degree of polymerization of the coreforming PBzMA block but is somewhat less sensitive to the total solids concentration of the reaction solution, which ranged

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from 5 to 29% w/w. Thus, this phase diagram differs qualitatively from that previously reported for a RAFT aqueous dispersion polymerization formulation, which probably reflects the much lower degree of solvation of the core-forming PBzMA chains in the present case. The phase diagram can be used to reliably predict the particle morphology for a given set of reaction conditions and also enables mixed phase regions to be identified (and hence avoided). Compared to previous RAFT alcoholic dispersion polymerization syntheses reported in the literature, our formulation leads to very high monomer conversions within 12-24 h at 70 °C and also relatively high blocking efficiencies for the RAFT macro-CTA. When transferred from ethanol to aqueous solution, these PDMA-PBzMA "nano-objects" acquired a high cationic surface charge due to protonation of the PDMA chains, as judged by aqueous electrophoresis. However, electron microscopy studies confirmed that the original particle morphology was retained after this solvent exchange. This is presumably related to the relatively high glass transition temperature of the PBzMA chains, which leads to kinetically frozen morphologies at ambient temperature.

ASSOCIATED CONTENT

Supporting Information

Zeta potential and hydrodynamic diameter data as a function of solution pH. 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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REFERENCES

(1) Chiefari, J.; Chong, Y. K.; Ercole, F.; Krstina, J.; Jeffery, J.; Le, T. P. T.; Mayadunne, R. T. A.; Meijs, G. F.; Moad, C. L.; Moad, G.; Rizzardo, E.; Thang, S. H. *Macromolecules* **1998**, *31*, 5559.

- (2) Jain, S.; Bates, F. S. Science 2003, 300, 460.
- (3) Howse, J. R.; Jones, R. A. L.; Battaglia, G.; Ducker, R. E.; Leggett, G. J.; Ryan, A. J. *Nat. Mater.* **2009**, *8*, 507.
- (4) Li, Y.; Armes, S. P. Angew. Chem., Int. Ed. 2010, 49, 4042.
- (5) Zhang, L.; Eisenberg, A. J. Am. Chem. Soc. 1996, 118, 3168.
- (6) Zhang, L.; Eisenberg, A. Science 1995, 268, 1728.
- (7) Ahmed, F.; Discher, D. E. J. Controlled Release 2004, 96, 37.
- (8) Savic, R.; Luo, L. B.; Eisenberg, A.; Maysinger, D. Science 2003, 300, 615.

(9) Geng, Y.; Dalhaimer, P.; Cai, S. S.; Tsai, R.; Tewari, M.; Minko, T.; Discher, D. E. *Nat. Nanotechnol.* **2007**, *2*, 249.

(10) Ghoroghchian, P. P.; Frail, P. R.; Susumu, K.; Park, T. H.; Wu, S. P.; Uyeda, H. T.; Hammer, D. A.; Therien, M. J. *J. Am. Chem. Soc.* **2005**, *127*, 15388.

(11) Phillip, W. A.; O'Neill, B.; Rodwogin, M.; Hillmyer, M. A.; Cussler, E. L. ACS Appl. Mater. Interfaces 2010, 2, 847.

(12) Vriezema, D. M.; Garcia, P. M. L.; Oltra, N. S.; Hatzakis, N. S.; Kuiper, S. M.; Nolte, R. J. M.; Rowan, A. E.; van Hest, J. C. M. Angew. Chem., Int. Ed. **2007**, 46, 7378.

- Article
- (13) Broz, P.; Driamov, S.; Ziegler, J.; Ben-Haim, N.; Marsch, S.; Meier, W.; Hunziker, P. Nano Lett. **2006**, *6*, 2349.
- (14) Caruso, R. A.; Giersig, M.; Willig, F.; Antonietti, M. Langmuir 1998, 14, 6333.
- (15) Hayward, R. C.; Pochan, D. J. Macromolecules 2010, 43, 3577.
- (16) Wang, X.; Guerin, G.; Wang, H.; Wang, Y.; Manners, I.; Winnik, M. A. Science **2007**, *317*, 644.
- (17) Cui, H.; Chen, Z.; Zhong, S.; Wooley, K. L.; Pochan, D. J. Science 2007, 317, 647.
- (18) Boissé, S.; Rieger, J.; Belal, K.; Di-Cicco, A.; Beaunier, P.; Li, M.-H.; Charleux, B. *Chem. Commun.* **2010**, *46*, 1950.
- (19) Zhang, X.; Boissé, S.; Zhang, W.; Beaunier, P.; D'Agosto, F.; Rieger, J.; Charleux, B. *Macromolecules* **2011**, *44*, 4149.
- (20) Rieger, J.; Zhang, W.; Stoffelbach, F.; Charleux, B. Macromolecules 2010, 43, 6302.
- (21) Charleux, B.; D'Agosto, F.; Delaittre, G. Adv. Polym. Sci. 2010, 233, 125.
- (22) Boissé, S.; Rieger, J.; Pembouong, G.; Beaunier, P.; Charleux, B. J. Polym. Sci., Part A: Polym. Chem. 2011, 49, 3346.
- (23) Sugihara, S.; Blanazs, A.; Armes, S. P.; Ryan, A. J.; Lewis, A. L. J. Am. Chem. Soc. **2011**, 133, 15707.
- (24) Blanazs, A.; Madsen, J.; Battaglia, G.; Ryan, A. J.; Armes, S. P. J. Am. Chem. Soc. **2011**, 133, 16581.
- (25) He, T.; Zou, Y.-F.; Pan, C.-Y. Polym. J. (Tokyo, Jpn.) 2002, 34, 138.
- (26) Wan, W.-M.; Pan, C.-Y. Polym. Chem. 2010, 1, 1475.
- (27) Huang, C.-Q.; Pan, C.-Y. Polymer 2010, 51, 5115.
- (28) Cai, W.; Wan, W.; Hong, C.; Huang, C.; Pan, C. Soft Matter 2010, 6, 5554.
- (29) Wan, W. M.; Sun, X. L.; Pan, C. Y. Macromol. Rapid Commun. 2010, 31, 399.
- (30) Israelachvili, J. N.; Mitchell, D. J.; Ninham, B. W. J. Chem. Soc., Faraday Trans. 2 1976, 72, 1525.
- (31) Israelachvili, J. Intermolecular & Surface Forces, 2nd ed.; Academic Press: London, 1991.
- (32) Antonietti, M.; Förster, S. Adv. Mater. 2003, 15, 1323.
- (33) Blanazs, A.; Armes, S. P.; Ryan, A. J. Macromol. Rapid Commun. 2009, 30, 267.
- (34) Semsarilar, M.; Ladmiral, V.; Blanazs, A.; Armes, S. P. Langmuir 2012, 28, 914.
- (35) Semsarilar, M.; Jones, E. R.; Blanazs, A.; Armes, S. P. Adv. Mater. **2012**, DOI: 10.1002/adma.201200925.
- (36) Zhang, X.; Rieger, J.; Charleux, B. Polym. Chem. 2012, 3, 1502.
 (37) Madsen, J.; Armes, S. P.; Bertal, K.; MacNeil, S.; Lewis, A. L. Biomacromolecules 2009, 10, 1875.
- (38) Zhang, W. J.; D'Agosto, F.; Boyron, O.; Rieger, J.; Charleux, B. *Macromolecules* **2011**, *44*, 7584.