Temperature-directed micellar morphological transformation using CABC-block copolymers and its applications in encapsulation and hidden segment

Zheng, Jie; Chen, Chen; Goto, Atsushi

2020

Zheng, J., Chen, C., & Goto, A. (2019). Temperature-directed micellar morphological transformation using CABC-block copolymers and its applications in encapsulation and hidden segment. *Angewandte Chemie (International edition)*, 59(5), 1941-1949.
doi:10.1002/anie.201913988

<https://hdl.handle.net/10356/137713>

<https://doi.org/10.1002/anie.201913988>

© 2020 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim. All rights reserved. This paper was published in *Angewandte Chemie (International edition)* and is made available with permission of Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim.

Downloaded on 26 Apr 2023 22:17:11 SGT

Temperature-Directed Micellar Morphological Transformation Using CABC-Block Copolymers and its Applications in Encapsulation and Hidden Segment

Jie Zheng, Chen Chen, Atsushi Goto*

Abstract: We developed a temperature-directed micellar morphological transformation using CABC multi-block copolymers with a hydrophobic block A, a hydrophilic block B, and a thermally responsive block C with a lower critical solution temperature (LCST). The micellar structure was switched from a star (below LCST) to a flower (above LCST). The transition-temperature was tuneable in a wide range (11-90 °C) by varying the C monomer composition. The large difference in the loading capacity between the star and flower enabled efficient encapsulation and controlled release of external molecules. Unlike conventional systems, the present star-to-flower transformation keeps micellar structures and hence does not liberate polymers but only external molecules selectively. Another application is a hidden functional segment. A functional segment is hidden (shielded) below LCST and exposed to interact with external molecules or surfaces above LCST, which may serve as a new temperature-directed interface for, e.g., biological and sensing applications.

Introduction

Morphological transformation of polymer nanoparticles, where the particle structures reversibly change in response to external stimuli such as temperature, light, and pH, has attracted great attention for diversifying smart responsive materials in, e.g., bio-sensor, fluorescence thermometer, and drug delivery applications.^[1-7] Such morphological transformation enables the control of physicochemical properties of the nanoparticles such as rheological, optical, encapsulation, and bio-interaction properties.^[8-11]

Block copolymers are of growing interest for preparing self-assembled nanostructures such as micelles, rods, and vesicles.^[12] Their morphological transformation such as micellar inversion,^[13] micelle-to-vesicle transformation,^[14,15] and assembly-dissolution conversion^[16-22] are widely studied using di- and tri-block copolymers.^[12,23,24] However, there are scarce examples studying morphological transformation of nanostructures prepared with more complex multi-block copolymers such as CABC tetra-block copolymers.^[25-27]

Living (or reversible-deactivation) radical polymerization^[28-36] is a useful method for synthesizing block copolymers. Our research group developed an organocatalyzed living radical polymerization using an alkyl iodide (R-I) as an initiator and an organic molecule as a catalyst.^[37-41] In this system, polymer-iodide dormant species (Polymer-I) reversibly generates the propagating radical (Polymer*) by the work of the organic catalyst (Scheme 1a). The catalysts include organic salts such as tetrabutylammonium iodide (Bu₄N⁺I⁻).^[38] Previously, we studied a temperature-selective radical generation from a newly designed initiator, i.e., alkyl diiodide (I-R²-R¹-I) (I-MEPE-I in Scheme 1b).^[42] Because of the largely different reactivities of I-R² and R¹-I, this initiator can selectively initiate from the R¹-I site at a mild temperature (60 °C) and from the I-R² site at an elevated temperature (110 °C), opening up the synthesis of unique CABC asymmetric multi-block copolymers (Scheme 1b).

(a) Organocatalyzed living radical polymerization

(b) Synthesis of CABC multi-block copolymer

Scheme 1. (a) Reversible activation in organocatalyzed living radical polymerization. (b) Temperature-selective initiation for the synthesis of CABC multi-block copolymer.

[*] J. Zheng, C. Chen, Prof. Dr. A. Goto
Division of Chemistry and Biological Chemistry, School of Physical and Mathematical Sciences, Nanyang Technological University, 21 Nanyang Link, 637371 Singapore
E-mail: agoto@ntu.edu.sg

Scheme 2. (a) Reversible morphological transformation between star micelle (left) and flower micelle (right) self-assembled by CABC multi-block copolymer. (b) Reversible encapsulation and release of external hydrophobic molecules. (c) Hidden and exposed functional segment.

The present work aims to use CABC block copolymers to create novel micellar morphological transformation systems, which is a new topic. The nanostructures are reversibly switched from Janus-type star micelles to symmetric flower-like micelles directed by temperature (Scheme 2a). Block A is a hydrophobic segment (coloured in blue in Scheme 2a), block B is a hydrophilic segment (red), and block C is a thermosensitive segment (green) having a lower critical solution temperature (LCST) in water. Below LCST, block C is soluble in water. Hence, only block A is insoluble to form a core, which is expected to give a Janus-star micelle with blocks B and C in the shell (Scheme 2a). Above LCST, block C is insoluble in water. Blocks A and C are insoluble to form a core, which is expected to give a flower micelle with block B in the shell (petal) (Scheme 2a). To form the flower as expected, blocks A and C should show good compatibility. This type of star-flower transformation is achievable by combining a hydrophobic block A to retain the core, a hydrophilic block B to retain the shell, and a thermosensitive block C to switch the morphology, and hence is unique to CABC block copolymers. ABC tri-block copolymers might be an alternative but can not give an asymmetric star.

The star-flower transformation can tune physicochemical properties of the nanostructures. The large difference in the core size between the star and flower (Scheme 2a) can bring a large difference in the loading capacity in the core (Scheme 2b) and is expected to give the reversible encapsulation and release of external molecules. This system is unique in that, in the release process, the system is expected to liberate only external molecules but not to liberate the polymer in the solvent, because the polymer can remain forming the nanostructure. This system is not a mere aggregation-dissolution system, where the polymer is liberated (dissolved in the solvent) as a contaminant in the release process, but a novel system that retains the nanostructure, where only external molecules are liberated.

Another interesting application is a hidden functional segment. In the star, block B (red segment) is expected to be shielded (hidden) by block C (green segment) (Scheme 2c). Upon heating, because of the transformation to the flower, block B is expected to be exposed (Scheme 2c). Recently, a few elegant works were reported on hidden segments on nanostructures by mixing two different diblock copolymers.^[43,44] In the present work, only a single polymer (CABC block copolymer) is used. The hidden segment can interact with external molecules or surfaces, providing a novel interface on the nanostructure. The present paper describes the encapsulation-release and the hidden segment as unique applications of CABC block copolymers.

Figure 1. (a) The deprotection of PEEA-PMMA-PSMA-PEEA to afford PEEA-PMMA-PGLMMA-PEEA. (b) GPC (DMF eluent) curves for the synthesis of PEEA-PMMA-PGLMMA-PEEA (Table 1 entries 1, 2, and 3-1). For I-PMMA-I, the chromatogram of polymer overlapped with those of small molecules or LiBr-cluster in the DMF eluent in the low-molecular-weight region. To avoid the overlap, I-PMMA-I was also analyzed with THF-GPC, giving accurate M_n ($= 3000$) and $\bar{D}$ ($= 1.10$) values, and these values are given in Figure 1b and Table 1 (entry 1). For the GPC curve comparison (Figure 1b), the DMF-GPC curves are given for I-PMMA-I and the three other polymers.

Results and Discussion

Synthesis of CABC Block Copolymer. Figure 1a shows the synthesis of a CABC block copolymer. The precursor CAB'C block copolymer was prepared by using methyl methacrylate (MMA) (block A), solketal methacrylate (SMA) (block B'), and 2-(2-ethoxyethoxy)ethyl acrylate (EEA) (block C) (Table 1 entries 1, 2, and 3-1) and Figure 1b). A mixture of MMA (50 equiv), I-MEPE-I (1 equiv) (alkyl di-iodide initiator), and tetraoctylammonium iodide (ONI) (0.5 equiv) (catalyst) was heated at 60 °C, yielding an I-ME-PMMA-I polymer with $M_n = 3000$ (25 monomer units of MMA) and dispersity $\bar{D}$ ($= M_w/M_n$) = 1.10 after the purification by reprecipitation, where PMMA is poly(methyl methacrylate) and M_n and M_w are the number- and weight-average molecular weights, respectively. The M_n and M_w values were obtained with PMMA-calibrated GPC, and the number of the monomer units was obtained by ¹H NMR (from the signal areas of the monomer units and the initiator (phenyl group)) (Supporting Information) throughout the paper. ¹H NMR showed that 100% of the PE-I site initiated and 97% of the I-ME site remained unreacted, demonstrating the high selectivity of the initiation from the two sites (Supporting Information, Figure

S1b). The purified I-ME-PMMA-I was used as a macroinitiator in the polymerization of SMA at 60 °C to obtain a diblock copolymer I-ME-PMMA-PSMA-I with $M_n = 9300$ (27 monomer units of SMA) and $\bar{D} = 1.24$ after the purification, where PSMA is poly(solketal methacrylate). The GPC traces (Figure 1b) show that a large fraction of the macroinitiator was extended to the diblock copolymer, confirming the high block-efficiency of the PMMA-I site of the macroinitiator. 95% of the I-ME site remained unreacted after the two polymerizations (Supporting Information, Figure S1c). In the third step, the purified I-ME-PMMA-PSMA-I was used as a macroinitiator in the polymerization of EEA at an elevated temperature 110 °C, yielding an I-PEEA-PMMA-PSMA-

PEEA-I tetrablock copolymer with $M_n = 48000$ (82 monomer units in each EEA segment) and $\bar{D} = 1.52$ after purification, where PEEA is poly(2-(2-ethoxyethoxy)ethyl acrylate) (Supporting Information, Figure S1d). Subsequently, the obtained precursor CAB'C copolymer was hydrolyzed to remove the acetal group in the B' segment to produce the desired CAB'C block copolymer (PEEA-PMMA-PGLMMA-PEEA) with $M_n = 26000$ and $\bar{D} = 1.48$ after purification, where PGLMMA is poly(glycerol monomethacrylate) (Supporting Information, Figure S1e). PMMA (block A) is hydrophobic, and PGLMMA (block B) is hydrophilic. PEEA (block C) is temperature-responsive.^[45]

Table 1. Synthesis of CAB'C multi-block copolymers.

Entry	Monomer	I-R-I	$[M]_0/[I-R-I]_0/[ON]_0$ (mM) ^[a]	T (°C)	t (h)	conv (%)	M_n (GPC) ^[b] ($M_{n,theo}$ ^[c])	DP (¹ H NMR) ^[d]	$\bar{D}$ (GPC) ^[b]
1	MMA	I-MEPE-I	8000/160/80	60	2	57	2900 (3000)		1.11
						After purification	3000	25	1.10
2	SMA	I-PMMA-I	8000/80/80	60	4	30	8800 (8400)		1.28
						After purification	9300	+ 27	1.24
3-1	EEA	I-PMMA-PSMA-I	8000/13/52	110	19	29	48000 (39000)		1.53
						After purification	48000	+ 164	1.52
						After deprotection	26000		1.48
3-2	EEA/PEGA	I-PMMA-PSMA-I	(7600/400)/11/44	110	22	23	46000 (40000)		1.61
						After purification	49000	+ 146/8	1.57
						After deprotection	28000		1.48
3-3	EEA/PEGA	I-PMMA-PSMA-I	(7200/800)/9/36	110	22	17	46000 (42000)		1.56
						After purification	47000	+139/15	1.52
						After deprotection	29000		1.54
3-4	EEA/PEGA	I-PMMA-PSMA-I	(6400/1600)/9/36	110	24	15	45000 (46000)		1.61
						After purification	50000	+122/30	1.56
						After deprotection	31000		1.48
3-5	EEA/PEGA	I-PMMA-PSMA-I	(5600/2400)/8/32	110	24	15	49000 (43000)		1.44
						After purification	50000	+88/37	1.53
						After deprotection	28000		1.43
3-6	EEA/PEGA	I-PMMA-PSMA-I	(4800/3200)/7/28	110	24	13	50000 (44000)		1.51
						After purification	49000	+70/47	1.54
						After deprotection	29000		1.50
3-7	EEA/PEGA	I-PMMA-PSMA-I	(4000/4000)/7/28	110	24	12	37000 (47000)		1.60
						After purification	40000	+58/58	1.59
						After deprotection	28000		1.52
3-8	PEGA	I-PMMA-PSMA-I	8000/7/28	110	36	8	33000 (52000)		1.53
						After purification	35000	+90	1.51
						After deprotection	28000		1.53

^[a]M = monomer. ^[b]PMMA-calibrated value by THF-GPC for entry 1 and DMF-GPC for all other entries. ^[c]Theoretical M_n calculated with $M_n = ([M]_0/[I-R-I]_0) \times \text{conversion} \times (\text{molecular weight of monomer}) + (\text{molecular weight of I-R-I})$. ^[d]DP obtained from the peak areas of the initiating phenyl group and the monomer units (with $\pm 10\%$ experimental error).

Figure 2. (a and b) DLS analysis of PEEA-PMMA-PGLMMA-PEEA in an aqueous solution at different temperatures. TEM image of the particles generated at (c) 4 °C and (d) 21 °C (1 wt% polymer in water/DMF = 10/1 (v/v)). For (c), PMMA and PEEA domains are not able to be clearly distinguished with TEM and the blue-coloured domain given in the figure is just for the guide for the eyes.

Temperature-Directed Micellar Morphological Transformation. The self-assemblies of the obtained block copolymer were studied. We used *N,N*-dimethyl formamide (DMF) to dissolve all of the three segments and water to aggregate the hydrophobic segments to generate the assemblies. The polymer was first homogeneously dissolved in DMF, to which water was added to generate the assemblies (1 wt% polymer in water/DMF = 10/1 (v/v)). If PEEA (block C) is hydrophilic, the star is expected to form (Scheme 2a and Figure 2a). The dynamic light scattering (DLS) analysis (Figure 2a) showed that the hydrodynamic size (diameter) of the assembly was 21 nm (peak top) at a low temperature 4 °C. PEEA is expected to be hydrophilic at 4 °C, because the LCST of PEEA in the studied solution is 11 °C as shown below. The contour length of the CABC copolymer (216 units) is 54 nm (= 0.25 nm × 216 units). The particle size (21 nm) was reasonably smaller than the contour length, indicating that the assembly was a micelle (sphere) (not vesicle) and that the

aggregation among the micelles was negligible. Figure 2c shows the TEM image of the micelle. PGLMMA (block B) was stained with FeCl_2 through a strong affinity between 1,2-diol groups in PGLMMA and Fe^{2+} (Experimental Section below). The dark domain in the TEM image (PGLMMA segment) was surrounded by the light grey domain (PMMA and PEEA segments), which is consistent with the expected star structure (Figure 2a).

Above LCST, PEEA is hydrophobic, and the nanostructure is expected to be the flower (Figure 2a). Arising the temperature to 24 °C (above LCST), the hydrodynamic size of the assembly changed (from 21 nm) to 48 nm, indicating the change in the morphology. The TEM image (Figure 2d) of the assembly prepared at 21 °C (above LCST) shows that the dark domain (PGLMMA segment) covers the whole surface in each sphere, which is consistent with the expected flower structure (Figure 2a).

Figure 2b shows the change in the hydrodynamic size with temperature. A sharp change in the size was observed between 9 °C (24 nm) and 12 °C (51 nm), showing the morphological change by the LCST transition. We determined the LCST to be 11 °C at a half change in the size (38 nm = (24+51)/2 nm), which is consistent with the LCST (11 °C) of the PEEA homopolymer (not CABC block copolymer) as determined below. Importantly, the change in the micellar size was reversible upon the heating (square in Figure 2b) and cooling (triangle) processes, demonstrating the reversible morphological transformation. The sizes of the flower (48–51 nm) and the star (21–24 nm) were considerably different, suggesting that the morphological transition may accompany with an inter-particle re-organization. The flower size (48–51 nm) was still within the contour length of the polymer (54 nm), indicating negligible vesicle formation.

The formation of the star and flower was further rationalized in two ways using ^1H NMR and fluorescence. Figure 3a shows the variable-temperature (VT) ^1H NMR spectra of the CABC block copolymer in D_2O /dimethyl sulfoxide ($\text{DMSO}-d_6$) (v/v = 10/1). At 4 °C (below LCST), we clearly observed the characteristic proton peaks of PEEA (a methine proton **a** at 2.25–2.05 ppm and methylene protons **b** at 4.04–3.82 ppm), meaning that PEEA was dissolved in D_2O and located in a hydrated shell. This result matches the expected star structure (Scheme 2a). The two characteristic peaks became smaller with an increase in temperature from 4 °C (below LCST) to 24 °C (above LCST), meaning that PEEA was undissolved in D_2O and formed the core. This result also agrees with the expected flower structure (Scheme 2a).

The other way is to use a special fluorophore, i.e., 4-(*N,N*-dimethylaminosulfonyl)-7-(2-aminoethylamino)-2,1,3-benzoxadiazole (DBDED), whose fluorescence is quenched in an aqueous environment (quenched by hydrogen bonding).^[46] The iodides at the CABC polymer chain ends were converted to DBDED (Figure 3b and Supporting Information).^[47] At 24 °C (above LCST), the micellar solution exhibited a strong fluorescence (Figure 3b (square)), since the DBDED chain ends and hence PEEA (block C) segments were in the hydrophobic core of the flower. At 4 °C (below LCST), the fluorescence intensity significantly dropped, suggesting that the DBDED chain ends and hence PEEA segments extended to the aqueous environment and formed the hydrated shell of the star. This change in the fluorescence intensity clearly demonstrates the change in the location of the PEEA segments, supporting the expected flower and star structures (Scheme 2a). A sharp change in the fluorescence intensity was observed between

10 °C and 15 °C (Figure 3b (square) (left vertical axis)), which is also consistent with the micellar size transition observed with DLS between 11 °C and 14 °C (Figure 3b (triangle) (right vertical axis)). The assembly size at 24 °C (above LCST) increased from 48 nm without DBDED (Figure 2b) to 68 nm with DBDED (Figure 3b) in the solution. We suppose that the assembly with DBDED was still the flower but that the core tended to be swollen because DBDED is hydrophilic. The TEM image in the dry state (Supporting Information, Figure S18) showed similar assembly sizes with and without DBDED, suggesting that the attachment of DBDED did not significantly impact the actual assembly size (and hence the morphology) at 24 °C. The assembly size at 4 °C (below LCST) was unchanged with and without DBDED (21 nm (Figures 2b and 3b)), suggesting no impact of the attachment of DBDED on the morphology (star) at 4 °C. In the star, DBDED is located in the already fully hydrated shell and would not further swell the shell. The observed increase in the flower size and no change in the star size in the solution would also be consistent with the locations of DBDED. The mentioned DLS, TEM, ^1H NMR, and fluorescence results proved the successful star-flower transformation directed by temperature.

Figure 3. (a) Variable-temperature (VT) ^1H NMR spectra (1 wt% polymer in $\text{D}_2\text{O}/\text{DMSO-}d_6 = 10/1$ (v/v)) of PEEA-PMMA-PGLMMA-PEEA. (b) Fluorescence intensity and particle size of DBDED-PEEA-PMMA-PGLMMA-PEEA-DBDED in an aqueous solution at different temperatures ($\lambda_{\text{ex}} = 450$ nm for fluorescence) (1 wt% polymer in water/DMF = 10/1 (v/v)).

The mentioned assemblies were studied in the water/DMF mixture (10/1 (v/v)). By removing DMF through dialysis, we also studied the assemblies in pure water. We observed a sharp change in the size between 11 °C and 14 °C with DLS (Supporting Information, Figure S19). The assembly sizes in pure water (14–18 nm (star) and 28–36 nm (flower) (Figure S19)) were smaller than those in the water/DMF mixture (21–24 nm (star) and 48–51 nm (flower) (Figure 2b)), because the hydrophobic cores would shrink more in the absence of DMF (pure water). The assemblies described below were studied in the water/DMF mixture.

Tailoring of Transition Temperature. The transition temperature was able to tailor by changing the monomer composition of block C. We used poly(ethylene glycol) methyl ether acrylate (PEGA) as a co-monomer of EEA to provide EEA/PEGA random copolymers in block C. The EEA/PEGA composition was varied from 100/0 to 50/50. With an increase in the hydrophilic PEGA composition, LCST increased from 11 °C (EEA/PEGA = 100/0) to 81 °C (EEA/PEGA = 50/50) (Figure 4b (circle) and Supporting Information, Figure S16) for the EEA/PEGA random copolymers (not CABC multi-block but only block C polymers). The LCST of the CABC multi-block copolymer was slightly higher than that of the only block C polymer due to the presence of the other segments (A and B blocks). With an increase in the hydrophilic PEGA composition, the LCST of the CABC multi-block copolymer increased from 11 °C (EEA/PEGA = 100/0) to 90 °C (EEA/PEGA = 60/40), and no transition was observed at EEA/PEGA = 50/50 (Figures 4a and 4b (square) and Table 1 (entries 3–1 to 3–7)). Therefore, we were able to tune the morphological transition in a wide range of temperature (between 11 °C and 90 °C).

Figure 4. (a) DLS analysis of $\text{PEEA}_m/\text{PPEGA}_n\text{-PMMA-PGLMMA-PEEA}_m/\text{PPEGA}_n$ ($m/n = 100/0, 95/5, 90/10, 80/20, 70/30, 60/40$, and $50/50$) at different temperatures. (b) LCST of $\text{PEEA}_m/\text{PPEGA}_n\text{-PMMA-PGLMMA-PEEA}_m/\text{PPEGA}_n$ (CABC) block copolymers and $\text{PEEA}_m/\text{PPEGA}_n$ (only block C) random copolymers (1 wt% polymer in water/DMF = 10/1 (v/v)).

Figure 5. (a) Reversible encapsulation and release of hydrophobic Nile Red through the morphological transformation. (b) Fluorescence intensity of Nile Red at different temperatures, (c) fluorescence spectra of Nile Red at 24 °C and 4 °C, and (d) fluorescence intensity of Nile Red at 24 °C and 4 °C altered in a recycled manner, using PEEA-PMMA-PGLMMA-PEEA (1 wt% polymer in water/DMF = 10/1 (v/v), λ_{ex} = 550 nm).

Reversible Encapsulation and Release of External Molecule. An interesting application of this morphological transformation is the reversible encapsulation and release of

external molecules. As mentioned above, in the release process, unlike mere micellization-dissolution systems, this system does not liberate the polymer but liberates only external molecules, which is a unique and useful aspect in applications. We studied the encapsulation and release of a hydrophobic fluorescence dye Nile Red (Figure 5a). To a DMF solution of Nile Red and CABC block copolymer (PEEA-PMMA-PGLMMA-PEEA with LCST of 11 °C), we added water (1 wt% polymer in water/DMF = 10/1 (v/v)), vigorously stirred for 1 h, and removed a precipitated excess amount of Nile Red using a filter (pore size = 450 nm) at room temperature (21 °C) (above LCST). At room temperature (above LCST), both PMMA (block A) (25 units) and PEEA (block C) (164 units) segments formed the core and encapsulated the hydrophobic Nile Red (Figure 5a), and hence a strong fluorescence was observed (Figure 5b). By cooling down (below LCST), PEEA (block C) became hydrophilic and released the hydrophobic Nile Red (Figure 5a). Because the fluorescence of the released Nile Red was partially quenched in an aqueous environment, the fluorescence intensity was reduced (Figure 5b). This result demonstrates the release of Nile Red. Additionally, the maximum fluorescence peak shifted from 627 nm (more hydrophobic environment) at 24 °C to 639 nm (more aqueous environment) at 4 °C (Figure 5c).

Importantly, the morphological change was reversible. Upon heating from 4 °C to 24 °C, the flower (large loading capacity) was regenerated, recovering the high fluorescence intensity (Figure 5d). After 7 repeating cycles, approximately 73% of Nile Red was still re-encapsulated successfully (629 arbitrary intensity (eighth run) / 857 arbitrary intensity (first run) = 73%) (Figure 5d). Thus, we successfully demonstrated the reversible encapsulation and release of external molecules in a controlled manner using this thermal responsive morphological change.

We also prepared a Nile-Red-containing purified star (i.e., we removed extra Nile Red at 0–4 °C (below LCST)). The fluorescence intensity for the Nile-Red-containing purified star (Figure 5b (triangle) at 4 °C) was 27% of that for the Nile-Red-containing flower (Figure 5b (circle) at 24 °C) (230 arbitrary intensity / 857 arbitrary intensity = 27%), suggesting that approximately 27% of Nile Red was retained in the PMMA core in the release process in the first run.

Hidden Segment and Interaction with External Substrate.

Another interesting application is a hidden functional segment. The PGLMMA segment (block B) with a diol functionality was used as the hidden segment (colored in red in Scheme 2c). Below LCST, PGLMMA is shielded (hidden) by hydrophilic block C (colored in green in Scheme 2c), because block C forms the outermost surface of the particle. PGLMMA is difficult to contact external substrates because of the steric hindrance of block C. In contrast, above LCST, block C becomes hydrophobic and forms the core. PGLMMA is exposed to the outermost surface (Scheme 2c) and is able to interact with external substrates.

We studied CABC block copolymers with four different EEA/PEGA compositions in block C (100/0 (polymer 1 (LCST = 11 °C)), 95/5 (polymer 2 (LCST = 20 °C)), 80/20 (polymer 3 (LCST = 64 °C)), and 0/100 (polymer 4 (no LCST)) (Table 1 (entries 3-1, 3-2, 3-4, and 3-8)). The self-assembly was prepared in an aqueous buffer solution at pH ~10 (1 wt% polymer in buffer water/DMF (10/1)). At a low temperature 8 °C, all of the four polymers formed the star with shielded PGLMMA. At a higher temperature 24 °C, polymers 1 and 2 turned to form the flower with exposed PGLMMA, while polymers 3 and 4 kept forming the star with shielded PGLMMA.

Figure 6. (a) No attachment of the star to the QCM sensor due to the hidden block B (left), attachment of the flower to the QCM sensor due to the exposed block B (middle), and release of the flower from the QCM sensor in an acidic condition (right). (b and c) QCM frequency change for PEEA_m/PPEGA_n-PMMA-PGLMMA-PEEA_m/PPEGA_n block copolymers with $m/n = 100/0$, 95/5, 80/20, and 0/100 at (b) 8 °C and (c) 24 °C (1 wt% polymer in buffer water/DMF = 10/1 (v/v)) at pH ~10. The pH was decreased to ~5 after 2000 s for $m/n = 100/0$ in (c).

The interaction of the micelle with an external substrate was studied with quartz crystal microbalance (QCM). We studied the interaction between the micelle and the QCM sensor surface. A gold-coated QCM sensor was fabricated with a boronic acid (Figure 6a) by immersing the sensor in an ethanol solution of 4-mercaptophenylboronic acid (Experimental Section). Upon the exposure, PGLMMA segment can chemically attach on the sensor surface through a condensation of the diol in PGLMMA and the boronic acid on the sensor surface (Figure 6a). Figures 6b and 6c show the QCM frequency change at 8 °C and 24 °C, respectively, where time zero is a time when the micelle solution was injected to the QCM cell. At 8 °C, PGLMMA was shielded for all of the four polymers, giving no change in the frequency (no attachment of the micelle on the QCM sensor) (Figure 6b). No obvious change was observed even after 2000 s, meaning that PGLMMA was shielded in a stable manner. By heating to 24 °C, the frequency gradually changed with time for polymers 1 and 2 (Figure 6c (red and orange lines)), meaning that PGLMMA was exposed and reacted with the boronic acid to attach on the sensor surface. After 2000 s, the frequency change (ΔF) was 15.1 and 10.1 Hz for polymers 1 and 2, respectively, which correspond to 89 and 59 ng/cm² mass changes according to the Sauerbrey equation^[48] (Experimental Section). The theoretically possible maximum mass change is calculated to be 1500–2900 and 1400–2700 ng/cm², respectively, assuming that the micelle is a swollen sphere (density = 0.5–1 g/cm³) and is attached on the sensor surface in a close packing manner (Experimental Section). The experimental surface occupancy was thus 3–6% (89/2900 to 89/1500) and 2–4% (59/2700 to 59/1400),

respectively. The value of 2–6% would be in a reasonable range for actual micellar attachment. For polymers 3 and 4, PGLMMA was shielded even at 24 °C, giving no change in the frequency (Figure 6c (blue and green lines)). This result confirms that the micellar attachment of polymers 1 and 2 at 24 °C is ascribed to the morphological change but not the temperature change in the same star morphology. Thus, we successfully demonstrated the concept of the hidden functional segment, its controlled exposure by temperature, and its interaction with the external substrate upon the exposure. Additionally, the attached micelle (polymer 1 at 24 °C) was able to release from the sensor surface by decreasing the pH from 10 to 5 (Figure 6c (red line after 2000 s)) through an acid-catalyzed hydrolysis of the boronic ester to the original diol and boronic acid.

Conclusion

As highlighted in this paper, we developed a reversible temperature-directed micellar morphological transformation using CABC block copolymers with a hydrophobic block A, a hydrophilic block B, and a thermally responsive block C. The assembly structure was reversibly switchable between the star (below LCST) and the flower (above LCST) (Scheme 2a). The LCST of the CABC block copolymer was tuneable in a wide range from 11 °C to 90 °C by varying the EEA/PEGA composition in block C.

The morphological transformation enabled us to tune the physical property (loading capacity) and the chemical property

(surface reactivity) of the micelle. The different loading capacity in the star and flower allowed the encapsulation and release of external molecules in a recycled manner. Unlike mere aggregation-dissolution systems, the present star-flower transformation keeps micellar structures and liberates only external molecules but not the polymer. This advantage charms for drug delivery and separation applications. Another interesting application was a hidden functional segment. Block B (PGLMMA) was hidden below LCST and exposed to interact with external substrates above LCST, serving as a new temperature-directed interface. The functional block B can possess zwitterionic, electron donor/acceptor, and ligand groups for biological, electronic, and sensing applications. Also interestingly, block B forms a loop structure (petal) in the flower, and such topology can provide ultra-low friction and high surface permeability and may find applications in lubrication and biological fields.

Acknowledgements

This work was supported by National Research Foundation (NRF) Investigatorship in Singapore (NRF-NRFI05-2019-0001) and Academic Research Fund (AcRF) Tier 2 from Ministry of Education in Singapore (MOE2017-T2-1-018).

Keywords: polymerizations • block copolymers • micellar morphological transformation • encapsulation • hidden segment

References

- [1] M. Motornov, Y. Roiter, I. Tokarev, S. Minko, *Prog. Polym. Sci.* **2010**, *35*, 174-211.
- [2] C. Boyer, N. A. Corrigan, K. Jung, D. Nguyen, T. K. Nguyen, N. N. M. Adnan, S. Oliver, S. Shanmugam, J. Yeow, *Chem. Rev.* **2016**, *116*, 1803-1949.
- [3] J. Hu, S. Liu, *Macromolecules* **2010**, *43*, 8315-8330.
- [4] Z. Yang, J. Cao, Y. He, J. H. Yang, T. Kim, X. Peng, J. S. Kim, *Chem. Soc. Rev.* **2014**, *43*, 4563-4601.
- [5] C. J. Kim, X. Hu, S. J. Park, *J. Am. Chem. Soc.* **2016**, *138*, 14941-14947.
- [6] H. Liu, R. Wang, J. Wei, C. Cheng, Y. Zheng, Y. Pan, X. He, M. Ding, H. Tan, Q. Fu, *J. Am. Chem. Soc.* **2018**, *140*, 6604-6610.
- [7] J. Lee, K. H. Ku, C. H. Park, Y. J. Lee, H. Yun, B. J. Kim, *ACS Nano* **2019**, *13*, 4230-4237.
- [8] J. Gaitzsch, X. Huang, B. Voit, *Chem. Rev.* **2016**, *116*, 1053-1093.
- [9] A. Walther, A. H. E. Müller, *Chem. Rev.* **2013**, *113*, 5194-5261.
- [10] S. M. Lee, S. T. Nguyen, *Macromolecules* **2013**, *46*, 9169-9180.
- [11] M. Karimi, P. S. Zangabad, S. Baghaee-Ravari, M. Ghazadeh, H. Mirshekari, M. R. Hamblin, *J. Am. Chem. Soc.* **2017**, *139*, 4584-4610.
- [12] U. Tritschler, S. Pearce, J. Gwyther, G. R. Whittell, I. Manners, *Macromolecules* **2017**, *50*, 3439-3463.
- [13] D. Wang, J. Yin, Z. Zhu, Z. Ge, H. Liu, S. P. Armes, S. Liu, *Macromolecules* **2006**, *39*, 7378-7385.
- [14] H. Ren, D. Chen, Y. Shi, H. Yu, Z. Fu, W. Yang, *Macromolecules* **2018**, *51*, 3290-3298.
- [15] R. Takahashi, T. Sato, K. Terao, S. I. Yusa, *Macromolecules* **2016**, *49*, 3091-3099.
- [16] Y. Li, T. Zhao, C. Wang, Z. Lin, G. Huang, B. D. Sumer, J. Gao, *Nat. Commun.* **2016**, *7*, 13214.
- [17] P. G. Falireas, M. Vamvakaki, *Macromolecules* **2018**, *51*, 6848-6858.
- [18] V. K. Kotharangannagari, A. Sanchez-Ferrer, J. Ruokolainen, R. Mezzenga, *Macromolecules* **2011**, *44*, 4569-4573.
- [19] H. I. Lee, W. Wu, J. K. Oh, L. Mueller, G. Sherwood, L. Peteanu, T. Kowalewski, K. Matyjaszewski, *Angew. Chem.* **2007**, *119*, 2505-2509; *Angew. Chem. Int. Ed.* **2007**, *46*, 2453-2457.
- [20] R. Deng, M. J. Derry, C. J. Mable, Y. Ning, S. P. Armes, *J. Am. Chem. Soc.* **2017**, *139*, 7616-7623.
- [21] X. Jiang, B. Zhao, *Macromolecules* **2008**, *41*, 9366-9375.
- [22] K. Feng, N. Xie, B. Chen, L. P. Zhang, C. H. Tung, L. Z. Wu, *Macromolecules* **2012**, *45*, 5596-5603.
- [23] M. F. Cunningham, P. G. Jessop, *Macromolecules* **2019**, <https://doi.org/10.1021/acs.macromol.9b00914>.
- [24] Y. Zhao, *Macromolecules* **2012**, *45*, 3647-3657.
- [25] A. K. Brannan, F. S. Bates, *Macromolecules* **2004**, *37*, 8816-8819.
- [26] E. D. Gomez, T. J. Rappl, V. Agarwal, A. Bose, M. Schmutz, C. M. Marques, N. P. Balsara, *Macromolecules* **2005**, *38*, 3567-3570.
- [27] R. Hoogenboom, F. Wiesbrock, M. A. M. Leenen, H. M. L. Thijs, H. Huang, C. A. Fustin, P. Guillet, J. F. Gohy, U. S. Schubert, *Macromolecules* **2007**, *40*, 2837-2843.
- [28] K. Matyjaszewski, *Adv. Mater.* **2018**, *30*, 1706441.
- [29] P. B. Zetterlund, S. C. Thickett, S. Perrier, E. Bourgeat-Lami, M. Lansalot, *Chem. Rev.* **2015**, *115*, 9745-9800.
- [30] J. Nicolas, Y. Guillauneuf, C. Lefay, D. Bertin, D. Gigmes, B. Charleux, *Prog. Polym. Sci.* **2013**, *38*, 63-235.
- [31] K. Matyjaszewski, N. V. Tsarevsky, *J. Am. Chem. Soc.* **2014**, *136*, 6513-6533.
- [32] M. Ouchi, M. Sawamoto, *Macromolecules* **2017**, *50*, 2603-2614.
- [33] D. J. Keddie, G. Moad, E. Rizzardo, S. H. Thang, *Macromolecules* **2012**, *45*, 5321-5342.
- [34] G. David, C. Boyer, J. Tonnar, B. Ameduri, P. Lacroix-Desmazes, B. Boutevin, *Chem. Rev.* **2006**, *106*, 3936-3962.
- [35] S. Yamago, *Chem. Rev.* **2009**, *109*, 5051-5068.
- [36] A. Goto, T. Fukuda, *Prog. Polym. Sci.* **2004**, *29*, 329-385.
- [37] A. Goto, T. Suzuki, H. Ohfujii, M. Tanishima, T. Fukuda, Y. Tsujii, H. Kaji, *Macromolecules* **2011**, *44*, 8709-8715.
- [38] A. Goto, A. Ohtsuki, H. Ohfujii, M. Tanishima, H. Kaji, *J. Am. Chem. Soc.* **2013**, *135*, 11131-11139.
- [39] A. Ohtsuki, L. Lei, M. Tanishima, A. Goto, H. Kaji, *J. Am. Chem. Soc.* **2015**, *137*, 5610-5617.
- [40] C. G. Wang, A. Goto, *J. Am. Chem. Soc.* **2017**, *139*, 10551-10560.
- [41] X. Liu, C. G. Wang, A. Goto, *Angew. Chem.* **2019**, *131*, 5654-5659; *Angew. Chem. Int. Ed.* **2019**, *58*, 5598-5603.
- [42] J. Zheng, C. G. Wang, Y. Yamaguchi, M. Miyamoto, A. Goto, *Angew. Chem.* **2018**, *130*, 1568-1572; *Angew. Chem. Int. Ed.* **2018**, *57*, 1552-1556.
- [43] H. Gao, T. Cheng, J. Liu, J. Liu, C. Yang, L. Chu, Y. Zhang, R. Ma, L. Shi, *Biomacromolecules* **2014**, *15*, 3634-3642.
- [44] F. H. Ma, C. Li, Y. Liu, L. Shi, *Adv. Mater.* **2019**, 1805945.
- [45] K. Skrabania, J. Kristen, A. Laschewsky, O. Akdemir, A. Hoth, J. F. Lutz, *Langmuir* **2007**, *23*, 84-93.
- [46] K. Okabe, N. Inada, C. Gota, Y. Harada, T. Funatsu, S. Uchiyama, *Nat. Commun.* **2012**, *3*, 705.
- [47] C. Chen, L. Xiao, A. Goto, *Macromolecules* **2016**, *49*, 9425-9440.
- [48] G. Sauerbrey, *Z. Phys.* **1959**, *155*, 206-222.

Entry for the Table of Contents

RESEARCH ARTICLE

Designed CABC asymmetric multi-block copolymers were used to attain unique temperature-directed reversible micellar morphological transformation. This transformation opened up conceptually new controlled encapsulation-release system and hidden-segment interfacial system.

Jie Zheng, Chen Chen, Atsushi Goto*

Page No. – Page No.

Temperature-Directed Micellar Morphological Transformation Using CABC-Block Copolymers and its Applications in Encapsulation and Hidden Segment