Morphological Behavior for Micelle from Polystyrene-b-poly[2-(β-D-glucopyranosyloxy)ethyl acrylate] upon Changing the Copolymer Concentration

Yu-Zeng Liang, Zi-Chen Li, and Fu-Mian Li*
Department of Polymer Science and Engineering, College of Chemistry and Molecular Engineering
Peking University, Beijing 100871, China

(Received December 7, 1999, CL-991031)

Multiple morphologies of molecular assemblies of polystyrene-b-poly[2-(β-D-glucopyranosyloxy)ethyl acrylate] (PSt-b-PGEA) were obtained by initially dissolving it in DMF and then adding water to the organic solution. With increasing the initial copolymer concentration (ICC) from 0.1 wt% to 2.0 wt% in DMF, the molecular assemblies of PSt_{77}-b-$PGEA_6$ were transformed from spheres, to rods, to vesicles, to tubules, and again to vesicles. *Concanavalin* (*Con A*) was used to characterize the surface properties of the resulting aggregates.

It has been known that when the length of hydrophilic blocks of amphiphilic block copolymers is far shorter than that of hydrophobic blocks, as the result of the association of hydrophobic blocks, multiple morphologies of molecular assemblies (so called crew-cut aggregates) are formed in water.[1-3] These morphologies include spheres, rods, vesicles, and tubules, etc.. Several factors, like the nature and composition of the block copolymers, the nature of the common solvent, and the initial copolymer concentration (ICC), etc., have been found to influence the morphologies of aggregates.[3] So far, several block copolymers have been investigated for their multiple morphologies of crew-cut aggregates in water.[4-6]

Glycopolymer in which saccharide residues are incorporated to polymer backbones as pendent groups have been studied as important materials for the study on the carbohydrate-protein interactions.[7] Amphiphilic diblock copolymers with glycopolymer segment as the hydrophilic block have also been found to form ordered aggregates under certain conditions.[8] We have reported that a new kind of highly asymmetric amphiphilic block copolymer, polystyrene-b-poly[2-(β-D-glycopyranosyloxyl)ethyl acrylate] (PSt-b-PGEA), exhibits multiple morphologies of molecular assemblies in water by initially dissolving it in a common solvent, e.g. DMF, THF, etc., at room temperature.[9]

In this article, we reported that the aggregates transformed from spheres, to rods, to vesicles, to tubules, and again to vesicles with the increase of ICC from 0.1 wt% to 2.0 wt% in DMF. The specific interaction between PSt-b-PGEA aggregates and *Con A* was investigated in order to characterize the saccharide groups in the surface of aggregates.

PSt-b-PGEA diblock copolymer was synthesized by atom transfer radical polymerization (ATRP). The fractionated diblock copolymer used in the present study is PSt_{77}-b-$PGEA_6$ with a polydispersity of 1.27. The subscripts of PSt_{77}-b-$PGEA_6$ represent numbers of the repeat units of PSt blocks and PGEA blocks. The procedure for preparation of aggregates and the characterization with transmission electron microscopy (TEM) can be found in the previous paper.[9] The solvent used was DMF and a series of copolymer solutions with the concentration from 0.1 wt% to 2.0 wt% were investigated.

Figure 1 shows the morphological transitions of the crew-cut aggregates of PSt_{77}-b-$PGEA_6$ by changing ICC in DMF. When ICC was 0.1 wt%, PSt_{77}-b-$PGEA_6$ yielded a mixture of spheres and short rods (Figure 1A). The outer diameter of

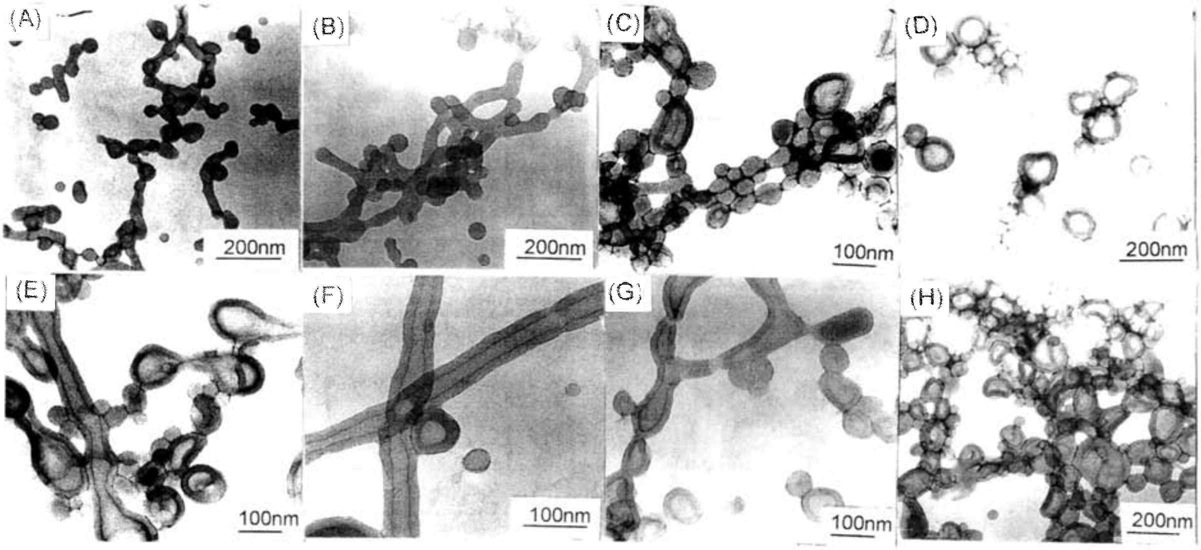

Figure 1. Morphological effect of the initial copolymer concentration (wt%) in DMF on the crew-cut aggregates of PSt_{77}-b-$PGEA_6$ at room temperature: (A) 0.1; (B) 0.2; (C) 0.4; (D) 0.6; (E) 0.8; (F) 1.0; (G) 1.5; (H) 2.0.

spheres is from 30 nm to 50 nm. The outer diameter of rods is ca. 50 nm. When ICC was increased to 0.2 wt%, rods were dominantly aggregates (Figure 1B). The length of the rod increases appreciably compared to that in Figure 1A, but the diameters do not change obviously. As ICC was increased to 0.4 wt%, most of the rod-like aggregates disappeared and vesicles were formed (Figure 1C), the diameters of which range from 80 to 120 nm. The vesicular nature is evidenced from their higher transmission in the core of aggregates compared to skin. Spheres still exist, but the diameters increase obviously as compared with those in Figure 1A and Figure 1B. As ICC was raised to 0.6 wt%, most aggregates formed from PSt$_{77}$-b-PGEA$_6$ were vesicles (Figure 1D), coexisting with some spheres. The vesicles are in regular shape. The average diameter of the vesicle increases compared to that in Figure 1C. With the increase of ICC to 0.8 wt%, the copolymer started to form short and irregular tubules (Figure 1E), coexisting with vesicles. When ICC was increased to 1.0 wt%, the copolymer formed long and regular tubules (Figure 1F). The tubular nature is evidenced from their higher transmission in the core compared to skin. The length of tubules ranges from 6 μm to 10 μm. The diameter of tubules is ca. 55 nm. Some small spheres could still be seen. As ICC was further increased to 1.5 wt%, the copolymer formed again irregular and short tubules, coexisting with vesicles (Figure 1G). When ICC reached 2.0 wt%, the copolymer formed dominantly large compound vesicles (Figure 1H). The transition from spheres to tubules can be explained from that the aggregation numbers of micelle increase as the increase of ICC.[10] The transition from tubules to vesicles is still being studied.

It is well known that glucose moieties have a strong and specific interaction with *Concanavalin A* (*Con A*).[11] In order to characterize the surface properties of the crew-cut aggregates we discussed above, the interaction of *Con A* with them was studied. Thus, an equal volume of aggregate solution prepared from 2.0 wt% of PSt$_{77}$-b-PGEA$_6$ in DMF and aqueous *Con A* buffer solution was mixed. The concentration of *Con A* in the mixture was 400 μg/mL. The interaction was monitored by following the increase of turbidity at 360 nm (Shimadzu 2100 UV-visible spectrophotometer).[12] Figure 2 shows the results. The solution has a absorption of 0.4 after equal volume of deioned water was added. Soon after *Con A* solution was added, the absorption increased very quickly, and reached a maximum at about 45 min. At this moment, even though the solution is very turbid, it is homogenous, and no precipitation can be observed. This state continued for about 40 min, then the precipitation was gradually formed. Until about 120 min, all the aggregates have been precipitated. This result indicates the secondary aggregation of crew-cut aggregates, which further confirms that the surface of the aggregates is covered with glucose moiety. After complexation with *Con A*, the aggregates are connected together to give large aggregates whose hydrophilicity are not strong enough to keep it in water as suspension. Other aggregates gave similar results. The only difference is the maximum value of turbidity and the time to reach it. To confirm that the original aggregates are stable enough and this complexation is reversible, an aliquot of concentrated glucose solution was added to the precipitated sample. After shaking the solution, it was observed that the turbidity gradually decreases and almost returns to the original value after 2 hours (data not shown). The interaction between *Con A* and glucose has replaced the interaction of *Con A* with aggregates, due to higher concentration of glucose.

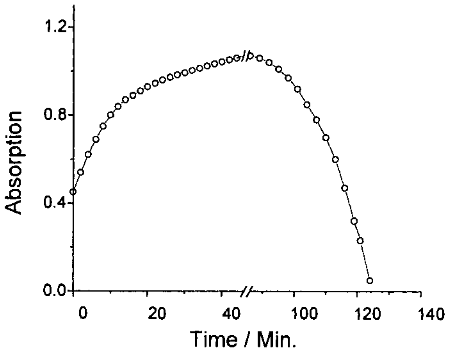

Figure 2. Con A-induced secondary aggregation of PSt$_{77}$-b-PGEA$_6$ vesicular aggregates at room temperature.

It is thus concluded that by changing ICC of PSt$_{77}$-b-PGEA$_6$ in DMF, different morphologies of aggregates can be obtained. At lower ICC, the aggregates are mainly micelle-like spheres, while bilayer aggregates are formed by increasing ICC. All the outer surface of the aggregates was covered with hydrophilic glycopolymer segments.

We would like to thank the National Natural Science Foundation of China (Nos: 59603004; 29995648-4) for financial support of this research.

References and Notes

1 L. Zhang and A. Eisenberg, *Science*, **268**, 1728 (1995).

2 A. Halperin, M. Tirrel, and T. P. Lodge, *Adv. Polym. Sci.*, **100**, 31 (1992).

3 L. Zhang and A. Eisenberg, *Polym. Adv. Technol.*, **9**, 677 (1998).

4 L. Zhang and A. Eisenberg, *Macromolecules*, **31**, 1144 (1998).

5 a) K. Yu and A. Eisenberg, *Macromolecules*, **31**, 3509 (1998). b) K. Yu, C. Bartels, and A. Eisenberg, *Langmuir*, **15**, 7157 (1999).

6 H. Shen, L. Zhang, and A. Eisenberg, *J. Am. Chem. Soc.*, **121**, 2728 (1999).

7 a) K. Aoi, K. Ito, and M. Okada, *Macromolecules*, **28**, 5391 (1995). b) K. Kobayashi, T. Tsuchida, T. Usui, and T. Akaike, *Macromolecules*, **30**, 2016 (1997). c) K. Yamada, K Minoda, and T. Miyamoto, *Macromolecules*, **32**, 3553 (1999).

8 a) K. Ohno, Y. Izu, S. Yamamoto, T. Miyamoto, and T. Fukuda, *Macromol. Chem. Phys.*, **200**, 1619 (1999). b) T. Miyamoto, M. Minoda and Y. Tsujii, *Chinese J. Polym. Sci.*, **17**, 21 (1999).

9 Z. C. Li, Y. Z. Liang, and F. M. Li, *Chem. Commun.*, **16**, 1557 (1999).
The preparation and TEM observation of aggregates: the copolymer was first disolved in DMF. Then, water was added dropwise to the solution with stirring to induce the micellization of the PS blocks. After the water content reached ca. 95 wt%, the solutions were placed in dialysis bags to get rid of DMF and to obtain the aqueous solutions of copolymer aggregates. One drop of diluted solution was put onto a carbon-coated copper grid, and was negatively stained with uranyl acetate. The observation was conducted with a JEOL-100-CXII TEM operated at 100 kV.

10 L. Zhang and A. Eisenberg, *Macromol. Symp.*, **113**, 221 (1997).

11 a) D. Page and R. Roy, *Bioconjugate Chem.*, **8**, 714 (1997). b) S. Takeoka, K. Sou, T. Ohgushi, and E. Tsuchida, *Supramol. Sci.*, **5**, 159 (1998).

12 Measured in 10-mm quartz cell filled with 3 mL of sample solution. [*Con A*] = 400 μg/mL. Aggregates solution is made from PSt$_{77}$-b-PGEA$_6$ with the ICC of 2.0 wt% in DMF and diluted by a factor of 10.