doi:10.1246/cl.2010.1106

Spontaneous Nanoaggregate Formation of Amphiphilic Poly(amide acid)s in Water

Takashi Hamada, Toshio Takayama, and Kazuaki Kudo*
Institute of Industrial Science, The University of Tokyo, 4-6-1 Komaba, Meguro-ku, Tokyo 153-8305

(Received July 14, 2010; CL-100633; E-mail: kkudo@iis.u-tokyo.ac.jp)

Alicyclic poly(amide acid) triethylammonium salts having alkyl ester side chains spontaneously self-assembled to form nanoaggregates in water. The nanoaggregates of poly(amide acid)s encapsulated a hydrophobic guest molecule, Nile red, within their apolar microenvironment. The size of the aggregates and the critical aggregation concentration (CAC) were considerably affected by both side chain length and M_w, of the polymers.

There has been a growing interest in amphiphilic polymers because of their potential to generate assemblies such as micelles, vesicles, and fibers.[1,2] The ability to tailor such nano- to microscale morphologies is very important because of desirable application in areas ranging from material science to biology such as drug delivery.[3]

Most of the amphiphilic polymers reported to date can be classified into AB diblock, ABA triblock, and random copolymers.[4–6] Recently, several groups have demonstrated a new type of aggregate-forming polymers, amphiphilic vinyl homopolymers that have side chain with both hydrophilic and hydrophobic moieties on each repeating unit.[7,8] Because of the limited number of research on amphiphilic homopolymers, their structure–property relationships are still not well understood compared to that of amphiphilic block copolymers. Further studies, especially those on non-vinyl amphipathic homopolymers, are desirable.

Poly(amide acid) (PAA), a synthetic intermediate of polyimide, has two hydrophilic carboxylic acid residues per repeating unit and the rest of the unit is essentially hydrophobic. This means that PAAs can be regarded as a candidate for aggregate-forming amphiphilic homopolymers. Horie et al. have shown that formation of the carboxylate salt of PAA with amines could significantly increase the hydrophilicity resulting in the solubilization of PAAs in water.[9] However, in their work, the aggregate-forming behavior of PAA salts was not referred to. We herein report the first example for the observation of self-assembled nanoaggregates of amphiphilic PAA in water, along with a brief structure–property relationship study.

We thought that the aggregating behavior of amphiphilic PAA salt could be systematically evaluated through changing the molecular structure of PAA. Therefore, alkyl esters of 3,5-diaminobenzoic acid were chosen as diamines because a series of related compounds having different lipophilicity is readily accessible. We used spiroalicyclic **DAn**, which was previously developed in our group, as a sole dianhydride.[10]

We employed propyl, nonyl, and hexadecyl esters of 3,5-diaminobenzoic acid as diamine monomers.[11] The PAAs were synthesized through the reaction of each diamine with **DAn** by a conventional method (Figure 1). It was revealed that the polyaddition occurred in nonselective manner according to ^{1}H NMR spectral analysis of PAAs.[12] M_w of the PAAs was determined by gel permeation chromatography (GPC) after the conversion of the carboxylic acid moieties to the corresponding methyl esters by treatment with diazomethane in order to avoid aggregation during the analysis.

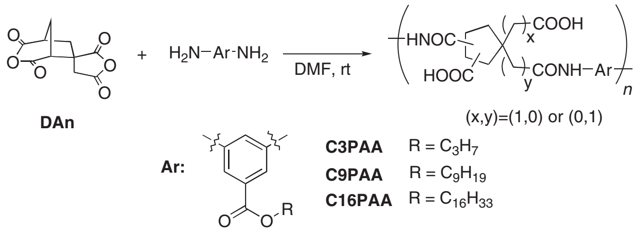

Figure 1. Synthesis of amphiphilic PAAs.

Although the PAAs were not soluble in water, they could be solubilized up to 2.0 g L^{-1} through the derivatization to the corresponding triethylammonium salts according to Horie's report.[9] The solutions were optically transparent and colorless.

Then, we checked the presence of hydrophobic microenvironment in the PAA solutions by using a Nile red (NR) as a polarity-sensitive fluorescent probe.[13,14] Although NR displayed a weak emission in water, a marked increase in the fluorescence intensity was observed in the presence of PAAs (Figure S1).[17] Interestingly, addition of poly(amide acid) prepared from **DAn** and *m*-phenylenediamine did not bring about the increase in the fluorescence intensity of NR. This means that the alkyl ester side chain is essential for providing effective hydrophobic environment.

We then estimated critical aggregation concentration (CAC) of the PAAs. The plot of the fluorescence intensity of NR as a function of the concentration of PAAs showed an inflection point, and this point was assigned to CAC (Figure S2).[17] Smaller CAC values were observed for PAAs with longer alkyl side chains (Table 1). Such a tendency has been recognized for wide variety of low-M_w surfactants, and can be easily explained by considering the difference in the HLB values of the repeating unit of PAAs.[15] The CAC values also varied depending on the M_w of **C9PAAs**: the increase in the M_w of PAA brought about the decrease in the CAC value. This can be attributed to intermolecular hydrophobic interaction, which is larger for

Table 1. Synthesis and aggregating properties of PAAs in 10 mM triethylamine (TEA)

Entry		Yield /%	M_w	M_w/M_n	CAC /mg mL^{-1}	D_H /nm
1	**C3PAA**	97	7600	1.50	0.25	531
2	**C9PAA**	82	7600	1.26	0.05	151
3	**C16PAA**	91	10700	1.46	0.01	11
4	**C9PAA**	70	3300	1.47	0.1	6
5	**C9PAA**	78	5800	1.48	0.05	135
6	**C9PAA**	87	14000	1.93	0.025	172

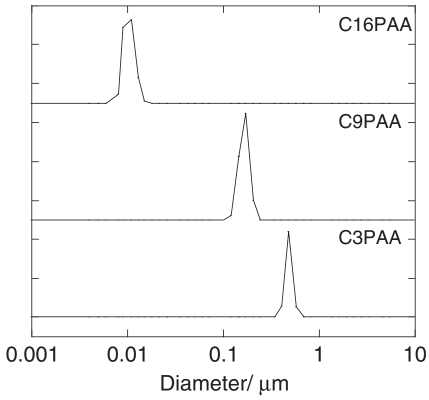

Figure 2. Size distribution for **C3PAA**, **C9PAA**, and **C16PAA** at concentration of 2 mg mL^{-1} in 10 mM TEA solutions.

higher $M_{\rm w}$ PAAs; hence, such PAAs are believed to begin aggregation at lower concentrations.

Dynamic light scattering (DLS) measurement of the PAA solutions at a concentration above CAC was performed. All the PAA solutions gave essentially a unimodal peak in the histogram of hydrodynamic diameter ($D_{\rm H}$) as shown in Figure 2. This is the first example of the observation of spontaneous molecular aggregate formation by amphiphilic polycondensate in water. Mean values of $D_{\rm H}$ considerably varied depending on the chemical structure and $M_{\rm w}$ of PAAs (Table 1). Most of the aggregates were much larger than common polymer micelles, and some of them were even larger than those of the vesicles of amphiphilic vinyl homopolymers reported by the Thayumanavan group.[7]

The size of the aggregate inversely correlated with the length of alkyl chain of the ester in the diamine moieties. Although there has been a report on a similar relationship for the aggregate of poly[*N*-(2-sulfoethyl)maleamic acid-*co*-alkyl vinyl ether] in water, the side chain length effect on the particle size is much more distinct in our case.[16]

The sizes of **C9PAA** aggregates were positively correlated with their $M_{\rm w}$ (Table 1, Entries 2, 4, 5, and 6), and a PAA($M_{\rm w} = 3300$) showed exceptionally small aggregate size compared to those with larger $M_{\rm w}$s. Although the structure of the molecular aggregates is not clear at present, it is obvious that cumulative intermolecular interaction, especially hydrophobic interaction, is required for the formation of particles with a diameter of >100 nm, which is considerably larger than the size of a single polymer chain. **C9PAA** with $M_{\rm w} = 3300$ is calculated to have only five to six side chain nonyl esters per macromolecule, and is believed to have less efficient intermolecular interaction compared to higher-molecular-weight analogs. In addition, this polymer might afford the curvature required for small particle. These might lead to the formation of particles <10 nm in diameter.

The formation of nanosized **C9PAA** particles in water was supported by scanning electron microscopic analysis of a dried solution (Figure 3). Particles with a diameter of 140 to 210 nm were observed and the size was in good agreement with those of the values obtained by DLS.

In conclusion, we have developed novel amphiphilic homopoly(amide acid)s showing spontaneous self-assembly in water. The size and the CAC of the amphiphilic PAAs were affected by both side chain length and $M_{\rm w}$ of the polymers. A study of the structure of the molecular aggregates along with more detailed alkyl chain length dependence is now under investigation.

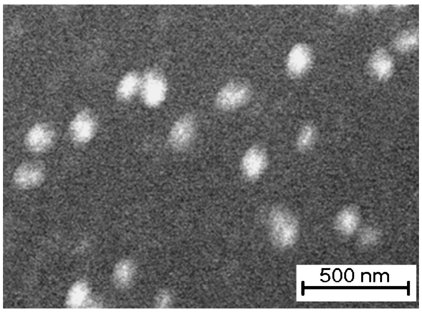

Figure 3. SEM image of the molecular assemblies from **C9PAA** with $M_{\rm w} = 14000$.

The authors thank Prof. Tadashi Sugawara at The University of Tokyo for the use of DLS instrumentation. This work was financially supported by a Grant-in-Aid for Scientific Research (No. 17550113) from the Ministry of Education, Culture, Sports, Science and Technology, Japan.

References and Notes

1 D. E. Discher, A. Eisenberg, *Science* **2002**, *297*, 967.
2 D. Yan, Y. Zhou, J. Hou, *Science* **2004**, *303*, 65.
3 S. Ganta, H. Devalapally, A. Shahiwala, M. Amiji, *J. Controlled Release* **2008**, *126*, 187.
4 S. Förster, V. Abetz, A. H. E. Müller, *Adv. Polym. Sci.* **2004**, *166*, 173.
5 Y. Sato, A. Hashidzume, Y. Morishima, *Macromolecules* **2001**, *34*, 6121.
6 F. Tian, Y. Yu, C. Wang, S. Yang, *Macromolecules* **2008**, *41*, 3385.
7 T. S. Kale, A. Klaikherd, B. Popere, S. Thayumanavan, *Langmuir* **2009**, *25*, 9660; S. Basu, D. R. Vutukuri, S. Shyamroy, B. S. Sandanaraj, S. Thayumanavan, *J. Am. Chem. Soc.* **2004**, *126*, 9890; E. N. Savariar, S. V. Aathimanikandan, S. Thayumanavan, *J. Am. Chem. Soc.* **2006**, *128*, 16224.
8 M. Changez, N.-G. Kang, C. H. Lee, J.-S. Lee, *Small* **2010**, *6*, 63.
9 Q. Li, T. Yamashita, K. Horie, H. Yoshimoto, T. Miwa, Y. Maekawa, *J. Polym. Sci., Part A: Polym. Chem.* **1998**, *36*, 1329.
10 J. Li, J. Kato, K. Kudo, S. Shiraishi, *Macromol. Chem. Phys.* **2000**, *201*, 2289.
11 Y. Tsuda, M. Kojima, J. Oh, *Polym. J.* **2006**, *38*, 1043.
12 K. Kudo, T. Yoshizawa, T. Hamada, J. Li, S. Sakamoto, S. Shiraishi, *Macromol. Rapid Commun.* **2006**, *27*, 1430.
13 D. M. Watkins, Y. Sayed-Sweet, J. W. Klimash, N. J. Turro, D. A. Tomalia, *Langmuir* **1997**, *13*, 3136.
14 G. Chen, Z. Guan, *J. Am. Chem. Soc.* **2004**, *126*, 2662.
15 K. Holmberg, B. Jönsson, B. Kronberg, B. Lindman, *Surfactants and Polymers in Aqueous Solution*, 2nd ed., John Wiley & Sons, Ltd., Göteborg, Lund, and Stockholm, **2002**.
16 A. C. Nieuwkerk, E. J. M. V. Kan, A. Koudijs, A. T. M. Marcelis, E. J. R. Sudhölter, *Langmuir* **1998**, *14*, 5702.
17 Supporting Information is available electronically on the CSJ-Journal Web site, http://www.csj.jp/journals/chem-lett/index.html.