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Synthesis of Azaylide-Based Amphiphiles by Staudinger Reaction

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Abstract: Catalyst- and regent-free reactions are powerful tools creating various functional molecules and materials. However, such chemical bonds are usually hydrolysable or require specific functional groups, which limits their use in aqueous media. Here we report that the development of new amphiphiles through the Staudinger reaction. Simple mixing of chlorinated aryl azide with a hydrophilic moiety and various triarylphosphines (PAr3) gave rise to azaylide-based amphiphiles NPAr3, rapidly and quantitatively. The obtained NPAr3 formed ca. 2 nm-sized spherical aggregates (NPAr3)_n in water. The hydrolysis of NPAr3 was significantly suppressed as compared with those of non-chlorinated amphiphiles nNPAr3. Computational study revealed the stability is mainly governed by the LUMO modulation around phosphorus atom due to the o-substituted halogen groups. Furthermore, hydrophobic dyes such as Nile red and BODIPY were encapsulated by the spherical aggregates (NPAr3)_n in water.

An amphiphilic molecule, which can self-aggregate and encapsulate quest molecules in water, has been greatly attracted in the various fields.^[1,2] Such amphiphiles have wellsegregated hydrophobic and hydrophilic parts in the molecule. The synthesis of amphiphiles is usually conducted in two steps: (i) construction of hydrophobic parts, (ii) introduction of hydrophilic chains^[3-5] (Figure 1a). Inspired by the green chemistry,^[6] we envisioned that in situ preparation by simple mixing of the hydrophilic/hydrophobic part and hydrophobic part with optimum hydrophilic-lipophilic balance would be a powerful strategy for development of various supramolecular materials^{[7-} ^{11]}. Actually, amphiphiles produced by this concept has been studied by using noncovalent interactions such as host-guest system^[12,13] and coordination bonding^[14,15] (Figure 1b). In the case of covalent bonding, dynamic covalent bonds $^{\left[16\right] }$ and copper-free click reaction^[17] are well studied as catalyst- and reagent-free reactions. However, despite the micellization is only performed in water, these covalent bonds are very fragile toward water or require specific functional groups. Therefore, in situ method for the development of facile, versatile, and reliable synthetic methodology for aqueous materials is still challenging task, so far.

The Staudinger reaction is a click-type reaction of an azide with a phosphine to form an iminophosphorane, so-called azaylide, which is first reported by Staudinger and Meyer in 1919 (Figure 1c).^[18] This reaction rapidly undergoes in high yield

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under mild conditions. However, the formed azaylide is readily hydrolyzed to primary amine and phosphine oxide once existing water. Taking advantage of this hydrolysis, the Staudinger reaction has been usually employed in the biochemical field,[19-^{22]} as initiated by Bertozzi and co-workers^[23]. Recently, Yan,^[24] Xi,^[25] Yoshida, and Hosoya^[26] found the "non-hydrolysis" Staudinger reaction for a biolabeling, in which halogen atoms on the aryl azide significantly improve the hydrolytic stability of azaylide.[27,28] This unusual stability prompted us to utilize the azaylide formation for various supramolecular materials in water. The advantage of this method is: (i) a variety of trisubstituted phosphine compounds are commercially available^[29] and reported,[30] and (ii) their triaryl moieties can be utilized for hydrophobic parts directly. Here we for the first time present the development of azaylide-based amphiphiles through the Staudinger reaction, as it is simple mixing of the hydrophilic subcomponent and various hydrophobic triarylphosphines (PAr3), and their self-assembly behaviors in water (Figure 1d).



Figure 1. Schematic representations of (a) typical synthetic methodology of amphiphiles, (b) amphiphiles prepared by noncovalent bonding and coordination bonding, (c) the Staudinger reaction, and (d) azaylide-based amphiphiles prepared through the Staudinger reaction.

We first prepared the hydrophilic subcomponent **1** based on the previous literatures (Figures S1-S10).^[26,31] Subcomponent **1** (0.060 mmol) was then mixed with 0.060 mmol of tris(*p*tolyl)phosphine (**PToI3**) in CH₃CN (5.0 mL) at 60 °C for 3 min to form **NPToI3** in over 99% yield (Figure 2a, Figures. S19-S24). The azaylide-based molecules can be switched by combination of hydrophilic and hydrophobic parts. Indeed, mixing of **1** with triphenylphosphine (**PPh3**) and tris(*p*-anisyl)phosphine (**PAni3**) gave rise to corresponding azaylides (**NPAr3**) such as **NPPh3** and **NPAni3**, respectively (Figures S25-S32). Non-chlorinated

nNPAr3 was also synthesized from non-chlorinated **2** and **PAr3** (Figure 2a, Figures S11-18, S33-44). Colorless single crystals of **NPPh3**^I, in which the counter anion was replaced with I[–], were obtained by slow diffusion of Et₂O into a solution of **NPPh3**^I in CH₃CN. In the crystalline state, two phenyl groups were disordered due to their rotation, and one phenyl group was placed perpendicular to be *trans*-stilbene-like structure (Figures 2b, S45). **NPPh3**^I and phenyl azaylide^[32] indicated similar dihedral angles in the *C*_A-*P*-*N*-*C*_B as 160° and 177°, respectively (Figure S46). As a result of the similar corn angle with triphenylphosphine, trident **NPAr3** would be suitable to form finite aggregates (Figure S46). The optimized structure of **NPAr3**['] with a methyl ester group instead of the hydrophilic chain was also supported the chemical structure of azaylide (Figures 2c and S47).



Figure 2. (a) Formation of azaylide through phosphazide in acetonitrile. (b) The ORTEP drawing of **NPPh3**^I which is an analogue of **NPPh3**. Counter anions are omitted for clarity. (c) The optimized structure of **NPTol3**['] at the B3LYP/6-31G (d,p) level with the conductor-like polarizable continuum model (CPCM; H₂O) (red ball: phosphorus, blue ball: nitrogen, green and light blue: carbon, yellow: chlorine, pink: oxygen)

The supramolecular behavior of azaylide-based amphiphiles was investigated in water. When NPTol3 (4.0 mmol) was dissolved into water (1.0 mL), azaylide-based aggregate (NPTol3)_n was formed spontaneously (Figure. 3a). In the ¹H NMR spectrum of NPTol3 (4.0 mM in D₂O), a set of signals for tolyl moiety was considerably broadened and shifted upfield ($\varDelta\delta$ = ~0.6 ppm) (Figure 3b, Figure S48). The ³¹P NMR signal significantly defused in stark contrast to a sharp and strong signal in CD₃CN (Figure S49). These characteristic spectrum changes arise from existing of dynamic behavior. The diffusionordered spectroscopy (DOSY) NMR spectrum showed a single band with $D = 9.79 \times 10^{-11} \text{ m}^2 \text{ s}^{-1}$ (Figure 3b, Figures S51-52). Dynamic light scattering (DLS) measurement provided relatively small aggregates ~2 nm with a narrow distribution (Figure 3c, Figure S60), which is comparable to a diameter of spherical aggregates (NPToI3)10 (Figure 3d, Figures S61-62).



Figure 3. (a) Schematic representations of the self-assembly of (**NPToI3**)_n in water. (b) ¹H NMR and DOSY spectra (500 MHz, 298 K, D₂O, 4.0 mM based on **NPToI3**) of (**NPToI3**)_n. (c) DLS chart of (**NPToI3**)_n (298 K, H₂O, 4.0 mM based on **NPToI3**). (d) Molecular modeling of the azaylide-based aggregate (**NPToI3**)₁₀.

The aggregation is further supported by nuclear Overhauser effect spectroscopy (NOESY), where correlations are obviously obtained between the terminal methyl and the interior phenyl protons (Hc-HA) (Figure S50). In addition, the concentrationdependent ¹H NMR spectra clearly showed the gradual upfield shifting of $H_{\rm C}$ and $H_{\rm B}$ ($\Delta\delta$ = up to 0.61 ppm), as the concertation increased from 0.40 to 8.0 mM (Figures S64,65, Table S3). Interestingly, in the range of 0.020 to 0.20 mM, all of the proton signals became sharp without shifting because of the monodispersity of NPToI3 (Figures S64,65). Thus, the critical micelle concentration (CMC) of NPToI3 is estimated to be ~0.40 mM, which is relatively low as compared with those of classical amphiphile (e.g., sodium dodecyl sulfonate (SDS) for 8.0 mM^[33]). In the same manner as NPToI3, the formation of (NPPh3)n and (NPAni3)n was confirmed (Figures S53-62). It is noteworthy that the CMC of NPPh3 shows ~2.0 mM which is 5-fold higher than those of NPTol3 and NPAni3. (Figures S66-69, Tables S4,5). This outcome implies that the presence of aromatic-alkyl CH- π interaction leads to enforce the strong aggregation. In addition, the in situ preparation of (NPAr3)n was successfully demonstrated by mixing of 1 and corresponding PAr3 in pure water even in heterogeneous system (Figure S63). Two chlorine atoms probably contribute increasing the hydrophobicity since the broad ¹H NMR signal was not observed in non-chlorinated nNPAr3 (Figures S73-75). (n)NPAr3 displayed absorption

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bands around 320 nm and indicated a weak blue emission in water or acetonitrile (Figures S70-72, Table S6).

Encouraged by the finding of those supramolecular aggregations, we next investigated the stability of the azaylidebased amphiphile toward water. An azaylide moiety is generally sensitive to water and organic solvents such as CS₂.^[24] Indeed, the non-chlorinated nNPToI3 was rapidly hydrolyzed in water at 293 K affording the corresponding primary amine and phosphine oxide after a few hours (Figure 4a bottom and 4b, Figure S73). The half-life of nNPTol3 in water at 293 K was calculated to be ~12 h by monitoring the ¹H NMR signals (Figures S73,79, Table S7). In contrast, the chlorinated NPTol3 came out to be consistently tolerated under the same conditions (Figure. 4a top and 4c, Figure S76). NPPh3 and NPAni3 were more stable than the corresponding non-chlorinated derivatives nNPPh3 and nNPAni3, respectively (Figures, S74,75,77-79, Table S7). This unique stability was also observed under the monodispersity state (Figures S80.81). These results clearly revealed that two chlorine atoms remarkably enhanced the tolerance of azavlide for hydrolysis. The water content ratio in the solvent is strongly related to the rate of hydrolysis, where the reaction accelerated as the water ratio increased in the solvent. To the best of our knowledge, this is first example in which azaylides successfully demonstrated the tolerance test in pure water.^[24,26]



Figure 4. (a) Reaction scheme of the hydrolysis of NPToI3 and nNPToI3 in water. ¹H NMR spectra (500 MHz, 298 K, D₂O) before and after 60 h of (b) 4.0 mM of nNPToI3 and (c) 4.0 mM of NPToI3. (d) Time-course of the decomposition of NPToI3 and nNPToI3 in water for 60 h. (e) Visualization and (f) fragment composition of the LUMOs for nNPToI3' and NPToI3'.

To obtain further insight into the unusual hydrolytic stability of the azaylide, we carried out density functional theory (DFT) calculation in water for chlorinated NPToI3' and non-chlorinated nNPTol3' without the hydrophilic chain as model compounds. The calculated HOMO of NPTol3' shows low energy level in comparison with nNPToI3' (Figure S82). This fact indicated that two chlorine atoms stabilized the HOMO leading to a decrease of basicity of nitrogen atom. Visualization of the molecular orbitals (MOs) is given in Figure S82. The LUMOs of aryl ester (Hyd) and triarylphosphine (Aryl) moieties between NPTol3' and nNPToI3' show precisely different distribution; the fragment composition of Hyd to Aryl was 66% : 24% for NPTol3' and 25% : 64% for nNPTol3', estimated by MO analysis (Figure 4f, Figures S82-83).³⁴ As a result of these differences, the LUMO around phosphorus atom in NPTol3' was remarkably reduced by 40% compared to nNPTol3' (Figure S84). Likewise, other azaylides NPPh3' and NPAni3' show similar tendencies in comparison with those of non-chlorinated ones (Figures S82-84). Thus, the LUMO modulation around phosphorus atom arose electron withdrawing chlorine atoms stabilized the azavlide mojety because nucleophilic addition of water on phosphorus atom has become a rate-determined step (Scheme S1).[35,36] Steric protection effect may be related in this system as well.

Finally, we carried out experiments for the guest encapsulation within the azaylide-based aggregates in water. Whereas polyaromatic amphiphiles capable of uptaking hydrophobic molecules have been reported,[37] amphiphiles containing a triarylphosphine core are rarely reported so far. [38, 39] Thus, the host capability of triarylphosphine core is obscure. When Nile red (NR, excess) as hydrophobic organic dye was mixed with 8.0 mM aqueous solution of NPTol3 (0.5 mL) overnight at room temperature, the solution color changed from colorless to a bright pink (Figure 5a). After removal the excess NR by filtration, the formation of the host-guest complex was confirmed by NMR, UV-vis, fluorescence, and DLS measurements. As a result of NMR analysis, ~0.1 mM of NR was found in 4.0 mM of NPTol3 aqueous solution (Figure S85). The UV-vis spectrum shows a broad absorption band around 493 and 562 nm, which indicates that hydrophobic NR is dissolved in water through encapsulation (Figure 5b). In the fluorescence spectrum, intense fluorescence of NR significantly decreased (Φ_F = 0.1%) because of the tight guest-guest aggregation (Figure 5b). DLS measurement revealed relatively large aggregates with narrow size distribution which is approximately 4.5 nm as a diameter (Figure S88). This size of nanoparticle most probably corresponds to NR15•(NPToI3)40 as estimated by molecular mechanics calculation (Figure 5c). The increased the size of encapsulated micelle was the consequence of strong guest-guest aggregation.[40] Similarly, (NPTol3)n successfully encapsulated pyrromethene 546 (BP) in water. The characteristic absorption band around 455 and 501 nm arose encapsulated (BP)n and significant low fluorescence quantum yield (~0.1%) were confirmed as well as NR (Figure S87). The encapsulated particle was displayed approximately 6.6 nm with a broad distribution (Figures S88,90), which is larger than the encapsulated NR.[40] In the same manner, we succeeded in encapsulation of NR and BP by using NPPh3 and NPAni3, respectively (Figures S86,87,89). Interestingly, NPPh3

has low uptake ability toward both **NR** and **BP**, although **NPAni3** has strong affinity for **NR** as well as **NPToI3** observed by UV-vis

absorption bands. Thus, this result also indicates that $CH-\pi$

interactions between host and guest molecules play an

important role in forming a robust host-guest complex in water.



Figure 5. (a) Schematic representation of the encapsulation of Nile red (NR) by NPToI3 in water. (b) UV-vis and fluorescence spectra of NRn•(NPToI3)m (H₂O, 298 K, 2.0 mM based on NPToI3). (c) Molecular modeling of the azaylide-based aggregate NR₁₅•(NPToI3)₄₀.

In conclusion, we have developed facile synthesis methodology for amphiphiles through the Staudinger reaction. Simple mixing of the hydrophilic subcomponent and hydrophobic triarylphosphines gave rise to corresponding azaylide-based amphiphiles rapidly and quantitatively. The amphiphiles form spherical aggregates which are ~2.0 nm as a diameter in water through π - π and CH- π interactions. Hydrolysable azaylide moiety is significantly stabilized in water because of the LUMO modulation around phosphorus atom arose electron withdrawing chlorine atoms. Furthermore, the obtained amphiphiles provide host capability toward hydrophobic organic dye in water. The present azaylide formation serves as a new type of preparation technique without catalysts and reagents, which will enable to create further functional materials in aqueous media.

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Here we report new type of amphiphiles through the click-type Staudinger reaction, which is *in situ* preparation by mixing of hydrophilic azide and hydrophobic triarylphosphine. These azaylide-based amphiphiles are stable against hydrolysis even in water and form ~2 nm spherical aggregates through self-assembly. Furthermore, the obtained amphiphiles by our method are capable of encapsulation of hydrophobic organic dves in water.

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