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Formed by Thermally Induced pH Changes in Binary Water–
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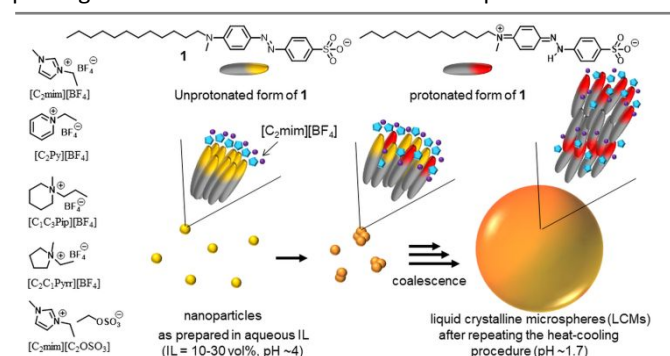
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Anionic azobenzene-containing bilayer membranes dispersed in a binary water-ionic liquid (IL) media undergo proton-responsive transformation into liquid crystalline microspheres (LCMs). This transformation was induced by protons released by the heat-induced hydrolysis of tetrafluoroborate ions in the ILs. This work demonstrates the first beneficial use of hydrolysis-susceptible ILs in chemistry.

Ionic liquids (ILs) have gained prominence in various scientific fields due to their unique physicochemical properties.¹ Despite their smaller cohesive energy as compared to water, ILs have also been established as potential media for self-assembly.² The degree of solvation, i.e., solvophilic interactions at the interface and the simultaneous pursuit of solvophobic interactions are crucial for driving amphiphilic self-assembly in ILs and to secure the dispersion stability.^{2a-c,3} The use of macroscopic interfaces formed between hydrophobic ILs and water as the fluid scaffolds for self-assembling charged nanospheres^{4a} demonstrated the untapped potential of ILs–liquid interfaces,^{4b-d} which came to a recognition with time.^{4e} The presence of nano-sized clusters in ILs and in the binary mixtures with water or organic solvents, meanwhile, has sparked another interest⁵ because they provide distinct environments from each constituent liquid.⁶ In this regard, we have shown that the fluctuating nanoclusters in aqueous ILs exerted significant influences on the molecular self-assembly of azobenzene amphiphiles.^{6b} Despite the unique potential of binary water-IL systems, the selection of ILs has been traditionally made on the premise of their chemical stability, and naturally mixing of hydrolysis susceptible ILs⁷ with water has been avoided.

In this work, we look at the hydrolytic property of these ILs from a totally different angle and view it as a useful means to actively control the pH of the binary water–ILs systems. We herein show that the binary mixtures of water and ILs having tetrafluoroborate (BF_4^-) anions not only provide versatile dispersion media for self-assembly but also display heat-triggered proton release characteristics that induce structural transformation of proton-responsive bilayer membranes to ordered liquid crystalline microspheres (LCMs) (Scheme 1). LCMs are attracting great interest due to their unique properties for photonic applications,⁸ and it is of importance to developing rational methodologies to prepare LCMs carrying photo-functional groups. In this way, the present work provides the first example of positively utilizing hydrolysis prone ILs for promoting the pH-triggered transformation of pre-organized molecular assemblies in the aqueous IL media.



Scheme 1. Molecular structures of amphiphile **1**, ionic liquids and a schematic illustration of the formation of LCMs triggered by thermally induced pH changes in aqueous hydrolytic ILs.

Sodium 4-[4-(*N*-methyl-*N*-dodecylamino)phenylazo]benzene sulfonate (**1**, Scheme 1, and Fig. S1, Electronic Supplementary Information, †ESI) was employed as a proton-responsive amphiphile which shows spectral red-shift upon protonation of the chromophore under acidic pH.⁹ The use of azobenzene chromophores also allows us to gain information on their molecular orientation in ordered self-assemblies, through their electronic spectra that often reveal changes

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† Electronic Supplementary Information (ESI) available: [Experimental methods, DLS, DFOM, UV-Vis Spectra, SEM image and Table S1]. See DOI: 10.1039/x0xx00000x

arising from exciton interactions.^{6b,10} As the hydrolysis-susceptible ILs, we employed those containing BF_4^- anion.⁷ Although **1** was sparingly soluble in water at room temperature due to the considerably low hydrophilicity of the azobenzenesulfonate group, it is fully dispersed in the binary water-IL (2-90 vol%) mixtures employed in this study. This is reasonably explainable by the electrostatic adsorption of $[\text{C}_2\text{mim}]^+$ ions onto **1**, which increased solvophilicity of **1** towards the binary water-IL media.^{6b} We started with 1-ethyl-3-methylimidazolium tetrafluoroborate, $[\text{C}_2\text{mim}][\text{BF}_4]$ as a typical water-miscible hydrolytic IL. Fig. 1A shows dynamic light scattering (DLS) count rates observed for **1** dispersed systematically at varied concentrations in water- $[\text{C}_2\text{mim}][\text{BF}_4]$ mixtures.

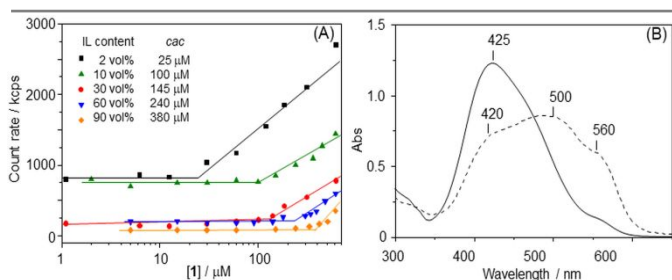


Fig. 1. (A) Count rate variation as a function of concentration of **1** in various water- $[\text{C}_2\text{mim}][\text{BF}_4]$ mixtures and (B) UV-vis absorption spectra of **1** (500 μM, above cac) dispersed in water- $[\text{C}_2\text{mim}][\text{BF}_4]$ (IL; 10 vol%) binary mixtures for as-prepared dispersion (solid line) and after a cycle of heating-cooling treatment (broken line).

Abrupt increases in the DLS count rates were observed at specific concentrations of **1** (Fig. 1A), which shifted towards higher concentration region with elevating the IL content in the binary aqueous media. These changes are typical to the concentration-dependent aggregate formation, and the critical aggregation concentration (*cac*) determined for **1** at varied IL content are also shown in Fig. 1A. In the following studies, the concentration of **1** is kept constant as 500 μM (above *cac*), if not stated otherwise. DLS measurement showed an average hydrodynamic diameter (D_h) of 7~8 nm for freshly prepared dispersions of **1** in water- $[\text{C}_2\text{mim}][\text{BF}_4]$ mixtures (IL content, 2-30 vol%, Fig. S2, †ESI). At higher IL content of 60 vol%, the D_h value further increased to ca. 600 nm (Fig. S2, †ESI). We confirmed that these large scattering intensities were not observed for the binary solvents alone and accordingly, these D_h values reflect the aggregate size of **1**. The absence of appreciable scattering intensity for $[\text{C}_2\text{mim}][\text{BF}_4]$ is quite a contrast to the binary water- $[\text{C}_2\text{mim}][\text{C}_2\text{OSO}_3]$ mixtures which showed the presence of temporary fluctuating nano-domains of $[\text{C}_2\text{mim}][\text{C}_2\text{OSO}_3]$ formed due to the appreciable hydrophobicity of the ion pair containing ethylsulfate ion and intermolecular interactions therein.^{6b}

We then investigated hydrolysis of $[\text{C}_2\text{mim}][\text{BF}_4]$ in aqueous mixtures. Fig. S3 (†ESI) shows the pH of the binary water- $[\text{C}_2\text{mim}][\text{BF}_4]$ mixtures containing **1** at ambient temperature, which was determined just after the preparation and after the indicated periods. The presence of **1** did not affect the pH because of its much lower concentration as compared to that of IL. In aqueous mixtures with IL content

less than 60 vol%, the initially observed pH (3.5~4.5) decreased to 1.7~2 after ca. 5 h to 40 hrs (Fig. S3, †ESI). Meanwhile, the mixture with higher IL content (90 vol%) showed no such significant drop in pH. Thus the hydrolysis of $[\text{C}_2\text{mim}][\text{BF}_4]$ occurs slowly, especially at the higher IL content in the binary aqueous medium. To thermally promote hydrolysis, these aqueous IL dispersions were once heated to 95 °C for 15 min, and then cooled down to room temperature. As a result, the resultant mixtures showed drop of pH below 2 regardless of the volume fraction of IL (Fig. S3B, †ESI), and the hydrolysis of BF_4^- anions is accelerated by the heat treatment. The binary dispersions following this thermal treatment are hereafter referred to as heat-cooled samples. For ease of comprehension, we focus on the aqueous IL mixtures with IL content of 10 vol% ([IL] = 0.56 M) in the following discussion. The comprehensive data obtained for the other IL contents are summarized in ESI.

The effect of heat-treatment-induced pH changes on self-assembly was investigated by DLS and UV-Vis spectroscopy. The heat-cooled dispersions showed larger D_h values as compared to the as-prepared dispersions (Fig. S2, †ESI). Darkfield optical microscopy (DFOM) of dispersions showed scattering dots that support the presence of molecular assemblies for the as-prepared (Fig. S4B, †ESI) and heat-cooled dispersions (Fig. S4F, †ESI). The UV-vis spectra of **1** as-dispersed in binary mixtures (IL: 10 vol%) (Fig. 1B) gave a λ_{max} at 425 nm associated with the $\pi-\pi^*$ transition moment along the long axis of azobenzene chromophore.^{6b,10d} This λ_{max} is blue-shifted as compared to that of a homologous monomeric dye, Methyl Orange (MO) dissolved in the aqueous IL ($\lambda_{\text{max}} = 462 \text{ nm}$),^{6b} indicating the presence of the excitonic interactions among parallel-oriented azobenzene chromophores.^{6b} Such a parallel molecular orientation of **1** is typical of bilayer membranes whose molecular orientation would be stabilized by screening the electrostatic repulsion between anionic sulfonate groups by the electrostatic adsorption of $[\text{C}_2\text{mim}]^+$ at the aggregate surface.^{6b} This spectral blue shift was observed for dispersions with the IL content of 2-30 vol% (Figure S5A, ESI), indicating that the ordered bilayer assemblies of **1** are formed in aqueous ILs with a fairly wide range of IL content.

In contrast to the as-prepared dispersions, the heat-cooled dispersions (1 heating cycle) of **1** in water- $[\text{C}_2\text{mim}][\text{BF}_4]$ mixtures (10 vol%, Fig. 1B) showed broadened spectra with a maximum at 500 nm. The diazo ($-\text{N}=\text{N}-$) moiety of amphiphile **1** possesses a higher pK_a (~3.4)^{9a} as compared to those of aromatic sulfonates ($\text{pK}_a \sim -2.5$).^{9b} Therefore, the diazo moiety would undergo protonation at the low pH caused by the heat treatment.⁹ The λ_{max} at 500 nm along with a shoulder around 560 nm is ascribed to the protonated azobenzene chromophores (Figure S5B, †ESI),^{9a} whereas the presence of a shoulder peak around at 420 nm suggests the coexistence of unprotonated species in the self-assembly.

Interestingly, when the heating-cooling procedure was repeated for 3 cycles, large spherical aggregates of **1** with diameters of 400-1500 nm were observed in optical microscopy (Fig. 2).

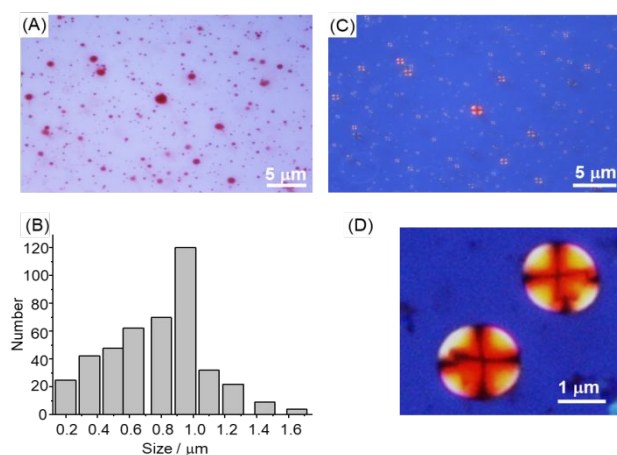


Fig. 2. Optical microscopic images of LCMs prepared from amphiphile **1** (500 μM, above cac) in water–[C₂mim][BF₄] (IL; 10 vol%) binary mixtures after three heating-cooling cycles. (A) under bright field and (C) under crossed polarizers. (B) The size histogram of LCMs performed for around 400 aggregates at different locations of the image. (D) Magnified image of LCMs under crossed polarizers.

These mesoscopic-size aggregates showed liquid crystalline characteristics (Fig. 2C, 2D) when viewed using polarized optical microscopy. The observed liquid crystalline texture shows Maltese cross patterns, which are characteristic of a highly ordered LC phase with a director pointing away from the centre of the droplet.^{11,12}

The formation of LCMs was widely observed for other water–[C₂mim][BF₄] mixtures having IL content up to 30 vol% (Fig. S6, †ESI), and was accompanied by decreases in the peak absorption intensity (Fig. S7, †ESI) and increase in D_h (Fig. S8, †ESI). These observations are ascribed to lower dispersibility of the developed LCMs. As sulfonate head groups of **1** are complexed with [C₂mim]⁺ ions abundantly present in the binary aqueous IL media, it is natural to assume that the protonated azobenzene units are stabilized by up-taking BF₄⁻ counter-ions to compensate the positive charges, rather than forming zwitterionic forms. We thus consider that the LCMs are formed from the ternary complex [1-H⁺][C₂mim][BF₄]. This is supported by FTIR measurements (Fig. S9, †ESI).

To further gain information on the morphological changes occurring in the course of heat-treatment, transmission electron microscopy (TEM) was conducted. Fig. 3 shows TEM images of **1** dispersed in water–[C₂mim][BF₄] (IL, 10 vol%) mixture at different stages. The as-prepared and the heat-cooled (one cycle) samples give spherical nanoparticles with average diameters of ca. 7 nm (Fig. 3A) and 16 nm (Fig. 3B), respectively, which are consistent with the observed D_h (Fig. S2, ESI). After repeating the heating-cooling procedure for 3 times, larger LCMs along with small aggregates were observed (Fig. 3C). The observed morphological changes (Fig. 3A→3C) indicate the growth of LCMs occurred by coalescence of nano-aggregates (Fig. 3B→3C). The surface structure and morphology of LCMs was also observed by scanning electron microscopy (SEM, Fig. S10, †ESI).

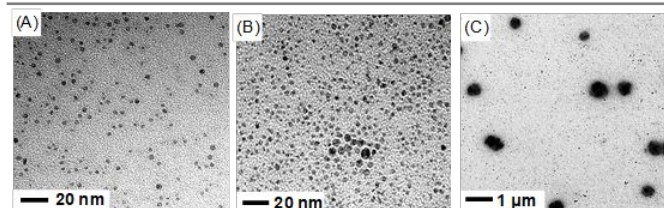


Fig. 3. TEM images of **1** dispersed in water–[C₂mim][BF₄] (IL; 10 vol%) binary mixtures. (A) nano-aggregates of **1** in as-prepared dispersion; (B) nano-aggregates of **1** after one heating-cooling procedure; (C) and LCMs after three heating-cooling procedures. The samples were post-stained with uranyl acetate.

To generalize the proton-triggered formation of LCMs in binary water–IL media, a series of hydrolysis-prone ILs with varied cationic species and BF₄⁻ (Scheme 1) were investigated. The other binary IL–water mixtures similarly showed a decrease in pH upon the heating-cooling treatment (Table S1, †ESI) and gave rise to the formation of LCMs (Fig. S11, †ESI) as in the case for [C₂mim][BF₄]. The growth of LCMs in the presence of different ILs (Fig. S12, †ESI) follows the order: [C₂mim][BF₄] > [C₂Py][BF₄] ≥ [C₁C₂Pyrr][BF₄] > [C₁C₃Pip][BF₄]. This suggests that the growth of LCMs is considerably promoted by ILs containing aromatic cations (i.e., [C₂mim]⁺ and [C₂Py]⁺). As discussed, the amphiphile **1** in the protonated form would be electrostatically complexed with both of the cations and BF₄⁻ anions of ILs, respectively. In this model, the promoted increase of D_h by aromatic cations indicates their stronger tendency to bind sulfonate groups of the azobenzene amphiphiles, which promoted the coalescence of smaller nano-aggregates into LCMs.¹³

Further to elucidate the role of hydrolysis-prone ILs in the proton-triggered transformation process, self-assembly of **1** was investigated in dilute aqueous hydrochloric acid solutions ([HCl]_{aq} = 0.06M) in the absence of ILs. Although **1** is protonated in aqueous HCl (Fig. S13A, †ESI), the formation of LCMs was not observed. Likewise, no LCM formation was confirmed when **1** was dispersed in aqueous NaBF₄ (Fig. S13B, †ESI). LCMs were also not formed when **1** was dispersed in water–[C₂mim][C₂OSO₃] (IL, 10 vol%) mixture because [C₂mim][C₂OSO₃] is stable against hydrolysis under the condition employed (Fig. S14(A,B), †ESI).^{6b} On the other hand, when the mixture of dilute aqueous HCl (0.02 M) and **1** dispersed in the water–[C₂mim][C₂OSO₃] (IL, 10 vol%) was subjected to the heat-treatment, LCMs were formed (Fig. S14(D-F), †ESI). These observations confirm that the both of the protonation of pre-organized **1** and the use of binary water–IL mixtures as dispersion media are indispensable to produce LCMs.

These observations are summarized in Scheme 1. In the as-prepared binary water–[C₂mim][BF₄] dispersions (pH~4), azobenzene amphiphiles **1** form bilayer membrane-based aggregates with parallel chromophore orientation. The generation of protons occurs *in situ* by the simple heat treatment of aqueous [C₂mim][BF₄], which transformed proton-responsive azobenzene bilayer membranes into the ordered LCMs. As the cationic charge of the protonated diazo group is well separated from the anionic sulfonate group, their

charges are most likely to be compensated by taking BF_4^- and $[\text{C}_2\text{mim}]^+$ ions, forming the ternary complex $[\text{1-H}^+][\text{C}_2\text{mim}][\text{BF}_4]$ (Scheme 1).

In summary, we demonstrated that the binary water-hydrolytic ILs media serve not only as a versatile dispersion media for the self-assembly of azobenzene amphiphiles but also as thermally triggered proton-releasing media. This phenomenon is widely observed for binary aqueous mixtures of hydrolytic ILs. This work demonstrates the first beneficial use of hydrolysis prone ILs for the supramolecular transformation process. It would be widely applicable to the design of pH-responsive phenomena in the binary aqueous ILs media. Together with the reported biphasic water-IL systems,⁴ the active use of hydrolysis susceptible ILs in aqueous systems offers a new perspective in the chemistry of ILs.

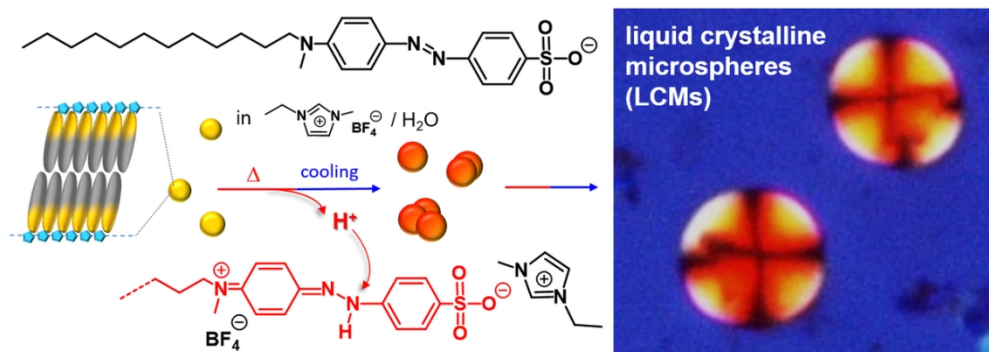
Conflicts of interest

There are no conflicts to declare.

Notes and references

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