

## Article

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# Synthesis and Micellar Characterization of an Amphiphilic Diblock Copolyphosphazene

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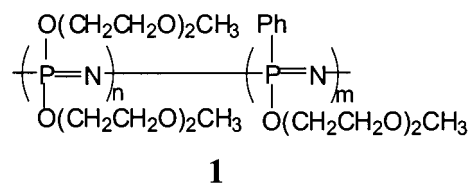
**ABSTRACT:** An amphiphilic polyphosphazene diblock copolymer (MEEP–Ph/MEEP) based on  $[N=P(OCH_2CH_2OCH_2CH_2OCH_3)_2]_n$  (MEEP) as a hydrophilic block and  $[N=PPh(OCH_2CH_2OCH_2CH_2OCH_3)]_m$  (Ph/MEEP) as a hydrophobic block was synthesized via the controlled cationic block copolymerization of phosphoranimine. The molecular weight of the block copolymer was estimated by GPC to be 3300. The molar composition ratio of repeating unit in the MEEP block to those in the Ph/MEEP block was 0.5. The micellar characterization of MEEP–Ph/MEEP in an aqueous phase was carried out by using fluorescence techniques, dynamic light scattering, and TEM. The block copolymer formed micelles with a critical micelle concentration (cmc) of 80 mg/L. The hydrodynamic radius ( $R_h$ ) of micelles, measured by dynamic light scattering, was 120 nm at 25 °C with a narrow distribution. The TEM image showed micelles with a spherical shape. Intermicellar aggregation was induced in the micellar solution of the block copolymer above 57 °C. The partition equilibrium constant,  $K_v$ , of pyrene in the micellar solution of the block copolymer was  $7.0 \times 10^3$ . The steady-state fluorescence anisotropy value ( $r$ ) of 1,6-diphenyl-1,3,5-hexatriene (DPH) was 0.237. The fluorescence lifetime value for DPH in the block copolymer solution showed that the core region of the micelles consisted of two regions with different hydrophobicities.

## Introduction

Polyphosphazenes, with the general formula  $(N=PR_2)_n$ , have a highly flexible phosphorus–nitrogen inorganic backbone, and their properties can be tailored by the incorporation of the suitable substituent R.<sup>1,2</sup> In particular, amphiphilic or water-soluble polyphosphazenes have attracted much attention due to their potential application in biomaterials and drug delivery area.<sup>3,4</sup> These polyphosphazenes can be categorized as follows. The first one includes hydrogels formed from  $[N=P(OCH_2CH_2OCH_2CH_2OCH_3)_2]_n$ , MEEP, and its relatives. The hydrogels can be utilized in membranes and have been used for the immobilization of enzymes.<sup>5</sup> Second, polyphosphazenes with lower critical solution temperatures (LCST) have been reported for the potential applications in thermosensitive drug release devices.<sup>3</sup> Recently, Allcock et al. reported on the ambient temperature route for the synthesis of polyphosphazenes via the controlled cationic polymerization of phosphoranimines, e.g.,  $Cl_3P=NSiMe_3$ .<sup>6–11</sup> This  $PCl_5$ -induced polymerization method is useful for the production of a variety of polymeric phosphazene systems with controlled architectures, such as block copolyphosphazenes. This synthetic method provides an opportunity to design well-defined amphiphilic diblock copolyphosphazenes which consist of hydrophilic and hydrophobic polyphosphazene blocks. These amphiphilic phosphazene block copolymers are expected to form supramolecular architectures such as micelles by self-assembly in an aqueous phase and consequently serve as a nanosize vehicle for delivery application.<sup>12–15</sup> In this work, therefore, we report the synthesis of an amphiphilic diblock copolyphosphazene **1**, constructed with a hydrophilic block of

phobic block of  $[N=PPh(OCH_2CH_2OCH_2CH_2OCH_3)]_m$  (Ph/MEEP).

In addition, the micellar characteristics of the amphiphilic diblock copolymer **1** was investigated by using fluorescence techniques, dynamic light scattering, and transmission electron microscopy.



## Experimental Section

**Materials and Equipment.** The phosphoranimines  $Cl_3P=NSiMe_3$  and  $PhCl_2P=NSiMe_3$  have been synthesized according to referenced methods.<sup>3,7,9</sup> Dichloromethane (Aldrich) was dried and distilled from  $CaH_2$  and then from  $P_2O_5$  into the reaction flask. All glassware was flame-dried under vacuum before use. All reactions were performed under an atmosphere of dry argon or nitrogen.  $^{31}P$ ,  $^{13}C$ , and  $^1H$  NMR spectra were recorded with a Bruker WM-360 NMR spectrometer operated at 146, 90.27, and 360 MHz, respectively.  $^1H$  and  $^{13}C$  NMR spectra are referenced to internal  $CDCl_3$  or  $CD_3C(O)CD_3$ .  $^{31}P$  NMR chemical shifts are relative to 85% phosphoric acid as an external reference, with positive shift values downfield from the reference. Molecular weights were estimated using a Hewlett-Packard HP 1090 gel permeation chromatograph equipped with an HP-1047A refractive index detector, American Polymer Standards AM gel 10  $\mu m$  and Am gel 10  $\mu m$  10<sup>4</sup> Å columns, and calibrated versus polystyrene standards (Polysciences). The samples were eluted with 0.1 wt % solution of tetra-*n*-butylammonium nitrate (Aldrich) in THF (OmniSolv). Thermal analysis was carried out using a Perkin-Elmer DSC

7 instrument. UV-vis spectra were obtained using a Hewlett-Packard 8452A spectrophotometer.

**Synthesis of Block Copolymer** {[N=P(OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>)<sub>2</sub>]<sub>n</sub>-[N=PPh(OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>)]<sub>m</sub>} (MEEP-Ph/MEEP). The room-temperature polymerizations were conducted in the following manner: To a stirred solution of PCl<sub>5</sub> (ca. 0.19 g, 0.91 mmol) in 20 mL of CH<sub>2</sub>Cl<sub>2</sub> was added Cl<sub>3</sub>P=NSiMe<sub>3</sub> (2.00 g, 8.95 mmol), and the resultant reaction mixture was stirred at 25 °C. After complete conversion of Cl<sub>3</sub>P=NSiMe<sub>3</sub> to living poly(dichlorophosphazene), as determined by <sup>31</sup>P NMR spectroscopy (ca. 3 h for [N=PCl<sub>2</sub>]<sub>n</sub>, <sup>31</sup>P NMR (CH<sub>2</sub>Cl<sub>2</sub>): δ = -17.4 ppm), 1.18 g (4.4 mmol) of PhCl<sub>2</sub>P=NSiMe<sub>3</sub> was added to the reaction solution, and then the mixture was stirred for 1 week at 35 °C with occasional monitoring by <sup>31</sup>P NMR spectroscopy. After complete conversion of PhCl<sub>2</sub>P=NSiMe<sub>3</sub> to polymer, all volatiles were removed at reduced pressure. Treatment of the resultant {[N=PCl<sub>2</sub>]<sub>n</sub>[N=PPh(Cl)]<sub>m</sub>} with NaOCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub> in dioxane produced {[N=P(OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>)<sub>2</sub>]<sub>n</sub>-[N=PPh(OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>)]<sub>m</sub>} (MEEP-Ph/MEEP). The polymer was purified by dialysis against H<sub>2</sub>O and MeOH (1 week each).

<sup>1</sup>H NMR (CDCl<sub>3</sub>): δ 3.26 (s, -OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>), 3.34-3.65 (m, -OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>), 3.98 (br, -P-OCH<sub>2</sub>CH<sub>2</sub>O-), 7.24 (br, -P-Ph (o)), 7.77 (br, -P-Ph (m, p)). <sup>13</sup>C NMR (CDCl<sub>3</sub>): δ 58.6, 63.8, 64.7, 70.0, 71.7, 127.4, 130.1, 131.5. <sup>31</sup>P NMR (CH<sub>2</sub>Cl<sub>2</sub>): δ -0.5 (br s, [N=PPh(OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>)]<sub>m</sub>), -6.3 (s, [N=P(OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub>)<sub>2</sub>]<sub>n</sub>).

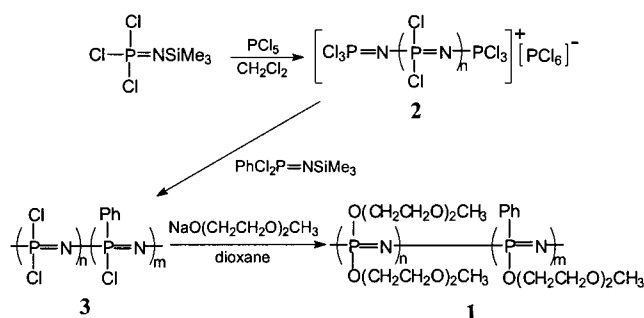
**Sample Preparation for Fluorescence and Light Scattering Measurements.** To prepare micellar solutions, MEEP-Ph/MEEP was dispersed in a doubly distilled water with vigorous stirring at 90 °C for 6 h. The micellar solution in doubly distilled water was diluted to obtain a concentration range from 5 × 10<sup>-5</sup> to 2.5 g/L. For the measurement of fluorescence spectra of pyrene in micellar solutions, samples were prepared following a literature procedure.<sup>16</sup> The final concentration of pyrene in the sample solutions was 6.0 × 10<sup>-7</sup> M. For the measurements of steady-state fluorescence anisotropy and fluorescence lifetime of DPH in micellar solutions, samples were prepared following a literature procedure.<sup>16</sup> The sample solution was filtered through a Millipore 0.45 μm filter and was purged with dry nitrogen for 30 min before measurement.

**Fluorescence Measurements.** The fluorescence spectra of pyrene were obtained using a Shimadzu RF-5301 PC spectrofluorometer. For the measurement of pyrene excitation spectra, emission and excitation slit widths were set at 3 and 1.5 nm, respectively. For the excitation spectra, the emission wavelength was 390 nm. The steady-state fluorescence anisotropy value and fluorescence lifetimes of DPH were obtained using an ISS K2 spectrofluorometer with a thermostat cell unit. The steady-state fluorescence anisotropy value of DPH was determined in the L-format geometry of detection. The excitation wavelength was 360 nm, and the emission was measured at 430 nm. The anisotropy value (*r*) was calculated from the following relationship:

$$r = \frac{(I_{VV} - I_{VV}^S) - G(I_{VH} - I_{VH}^S)}{(I_{VV} - I_{VV}^S) + 2G(I_{VH} - I_{VH}^S)} \quad (1)$$

where *I*<sup>S</sup> is the contribution of scattered light from a sample solution without DPH; *G* (= *I*<sub>HV</sub>/*I*<sub>HH</sub>) is the instrumental correction factor; and *I*<sub>VV</sub>, *I*<sub>VH</sub>, *I*<sub>HV</sub>, and *I*<sub>HH</sub> refer to the resultant emission intensities polarized in the vertical or the horizontal detection planes (second subindex) when excited with vertically or horizontally polarized light (first subindex).<sup>17</sup> Fluorescence lifetimes of DPH were measured using an ISS K2 fluorometer equipped with a frequency synthesizer (Marconi Instruments) and an ISS-ADC interface for data collection and analysis.<sup>16</sup> The phase-shift and demodulation ratios, using 1,4-bis(5-phenyloxazol-2-yl)benzene (POPOP) in ethanol as the reference (τ = 1.35 ns),<sup>18</sup> were recorded at 10 different modulation frequencies, logarithmically spaced (5.0, 7.0, 9.7, 13.6, 18.9, 26.4, 36.8, 51.4, 71.7, and 100.0 MHz). The estimated errors

Scheme 1



used in the decay law fittings were 0.008 in the demodulation and 0.4° in the phase shift. The excitation was operated at 360 nm. The emission was collected through a 408 nm cut-on filter. Fluorescence lifetimes were determined using a nonlinear least-squares program (ISS187) which minimized the reduced  $\chi^2$  for an exponentially good fit.

**Light Scattering Measurements.** Dynamic light scattering measurements were performed using a Brookhaven BI-200SM goniometer, BI-9000AT autocorrelator, and He-Ne laser (632.8 nm) (Research Electro-Optics, 35 mW). The sample solution (0.5 g/L) was purified by passing through a Millipore 0.45 μm filter. All the measurements were carried out at 25 °C. The scattered light of a vertically polarized He-Ne laser was measured at an angle of 90° and was collected on an autocorrelator. The hydrodynamic radius (*R*<sub>h</sub>) of the micelles was calculated by using the Stokes-Einstein equation  $R_h = k_B T / 6\pi\eta D$ , where *k*<sub>B</sub> is the Boltzmann constant, *T* is the absolute temperature, *η* is the solvent viscosity, and *D* is the diffusion coefficient. The polydispersity factor of micelles, represented as  $\mu_2/\Gamma^2$ , where  $\mu_2$  is the second cumulant of the decay function and  $\Gamma$  is the average characteristic line width, was calculated by the cumulant method.<sup>19,20</sup> CONTIN algorithms were used in the Laplace inversion of the autocorrelation function to obtain micelle size distribution.<sup>21</sup>

**Transmission Electron Microscopy.** Transmission electron microscopy (TEM) was performed on a Philips CM 200, operating at an acceleration voltage of 80 kV. A drop of aqueous polymer solution (0.5 g/L) was applied onto a 200 mesh copper grid coated with carbon, followed by air-drying. The sample was stained by applying a droplet of a 5 wt % uranyl acetate solution.

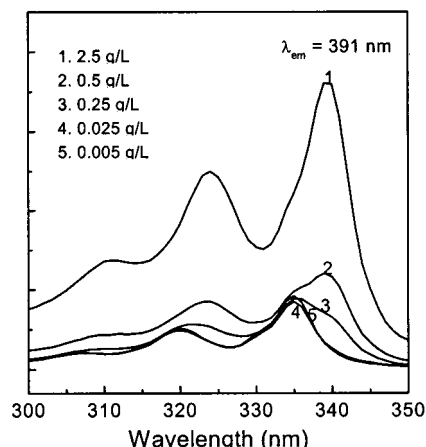
## Results and Discussion

**Synthesis and Characterization.** The synthesis of MEEP-Ph/MEEP block copolymer **1** was performed using the method reported by Allcock et al. as illustrated in Scheme 1.<sup>9</sup> The reaction of the phosphoranimine, Cl<sub>3</sub>P=NSiMe<sub>3</sub>, with PCl<sub>5</sub> yielded living poly(dichlorophosphazene) **2**. The addition of a second phosphoranimine, PhCl<sub>2</sub>P=NSiMe<sub>3</sub>, to this living species resulted in the formation of the block copolymer **3**. Subsequent treatment of this reaction mixture with NaOCH<sub>2</sub>CH<sub>2</sub>OCH<sub>2</sub>CH<sub>2</sub>OCH<sub>3</sub> replaced the chlorine atoms to yield the amphiphilic MEEP-Ph/MEEP diblock copolymer **1**. Diblock copolyphosphazene **1** was soluble in THF, chloroform, methylene chloride, and DMF. The molecular weight and molecular weight distribution of the block copolymer, estimated by GPC, were 3300 and 1.98, respectively. The block composition was determined by the analysis of the <sup>1</sup>H NMR spectrum. The molar ratios of the repeating units in the MEEP and MEEP-Ph blocks were determined by the peak integration ratios of the methyl protons (3.26 ppm) of methoxyethoxy units in the MEEP block and protons (7.24 ppm) of phenyl units in Ph/MEEP block. The molar composition ratio of repeating units in the MEEP block to that of the Ph/MEEP block was found to be 0.5 (Table 1).

**Table 1. Properties of MEEP–Ph/MEEP Block Copolymer**

block copolymer	$M_n^a$	composition ratio <sup>b</sup> [MEEP]/[Ph/MEEP]	wt % of hydrophobic block <sup>c</sup>	$M_w/M_n^d$
MEEP– Ph/MEEP	3300	0.5	67	1.98

<sup>a</sup> Estimated by GPC. <sup>b</sup> Molar composition ratio of the repeating units of MEEP to that of Ph/MEEP by <sup>1</sup>H NMR. <sup>c</sup> Weight percentage of the hydrophobic Ph/MEEP block in the block copolymer. <sup>d</sup> Estimated by GPC.

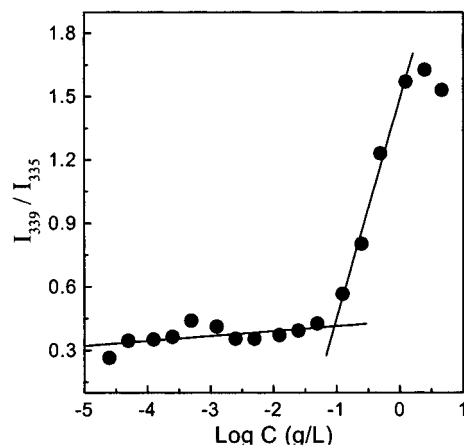
**Figure 1.** Excitation spectra of pyrene as a function of MEEP–Ph/MEEP concentration in water.

The block copolymer was amorphous and had a glass transition at  $-74$  °C without crystallinity.

**Micelles of MEEP–Ph/MEEP Block Copolymer.** The phosphazene block copolymer consists of MEEP and Ph/MEEP blocks. The MEEP homopolymer is totally soluble in water. However, Ph/MEEP homopolymer in which one phosphorus atom possesses both phenyl and 2-(2-methoxyethoxy)ethoxy substituents is not soluble but precipitates. The amphiphilic nature of the MEEP–Ph/MEEP diblock copolymer, consisting of hydrophilic MEEP and hydrophobic Ph/MEEP blocks, provides an opportunity of self-aggregation to form micelles in the aqueous phase. The micellar characteristics of the amphiphilic MEEP–Ph/MEEP diblock copolymer in an aqueous phase were investigated by using fluorescence techniques, dynamic light scattering, and TEM. The micelle formation in an aqueous phase was confirmed by a fluorescence technique with pyrene as a fluorescence probe.<sup>21–25</sup> When the micelles are formed in an aqueous phase, pyrene molecules preferably locate inside or close to the hydrophobic microdomain of micelles, and consequently their photophysical characteristics are changed, compared to pyrene molecules in an aqueous phase. Figure 1 shows the excitation spectra of pyrene at various concentrations of MEEP–Ph/MEEP.

The characteristic feature of the pyrene excitation spectra, (0,0) band shift from 335 to 339 nm upon pyrene partition into micellar hydrophobic microdomain, was utilized to determine the cmc value of MEEP–Ph/MEEP. Figure 2 shows the intensity ratio ( $I_{339}/I_{335}$ ) of pyrene excitation spectra versus logarithm of MEEP–Ph/MEEP block copolymer concentration.

The cmc value was determined from the threshold concentration, where the intensity ratio  $I_{339}/I_{335}$  begins to increase markedly. Therefore, the interception of two straight lines in the low concentration range is determined as cmc. The cmc value of the MEEP–Ph/MEEP

**Figure 2.** Plot of  $I_{339}/I_{335}$  (from pyrene excitation spectra) vs  $\log C$  for MEEP–Ph/MEEP.

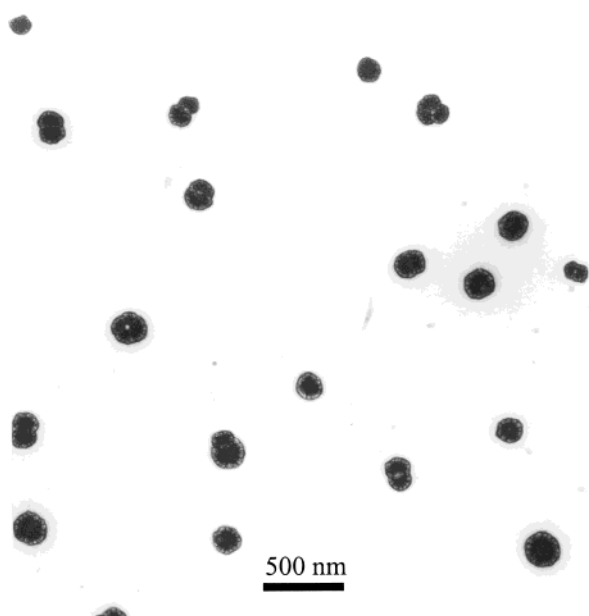
block copolymer is 80 mg/L, which is much higher than those of other polymeric amphiphiles.<sup>16,21–24</sup> This might be due to the low hydrophobicity of the Ph/MEEP block. The mean hydrodynamic radius ( $R_h$ ) of block copolymer micelles, measured by dynamic light scattering, was 120 nm at 25 °C. The  $R_h$  of micelles was not significantly affected by the change of the polymer concentration in the range of 0.5–5 g/L. Considering the block length of MEEP–Ph/MEEP, it does not seem to generate micelles with  $R_h$  as large as 120 nm in an aqueous phase, provided that they are core–shell structure. Therefore, it can be suggested that the micelles of MEEP–Ph/MEEP would be multicore structure formed by association of individual micelles rather than simple core–shell structure. In previous reports, micelles with a PEO outer shell are frequently found to form large aggregates in an aqueous phase.<sup>26,27</sup> In particular, Eisenberg's group recently reported that the formation of large aggregates of individual micelles was more pronounced in the case of micelles with a shell from a relatively short PEO block ( $M_n$  = ca. 2000), which could not outweigh the hydrophobic interaction and van der Waals interaction between exposed hydrophobic cores of micelles.<sup>27</sup> This explanation might also be applied to the case of MEEP–Ph/MEEP block copolymer systems which generate the micelles with the multicore structure. From the cumulant method, the polydispersity factor ( $\mu_2/\Gamma^2$ ) of micelles at 25 °C was obtained fairly low (0.12), suggesting narrow size distribution.

The micelles of MEEP–Ph/MEEP observed by TEM were generally spherical as shown in Figure 3.

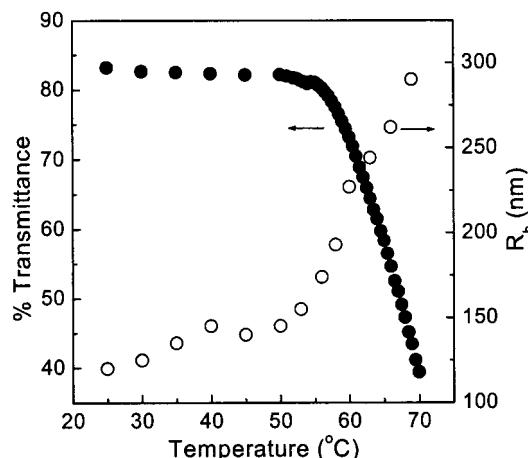
MEEP, comprising the hydrophilic outer shell of the micelles, is known to show a lower critical solution temperature (LCST) in an aqueous phase.<sup>3</sup> Therefore, the micellar solutions of MEEP–Ph/MEEP are expected to exhibit temperature-dependent phase transition behavior, which might be associated with the LCST of MEEP homopolymer in water. The temperature-dependent phase transition of MEEP–Ph/MEEP aqueous solution was monitored by observing the change of transmittance at 500 nm as shown in Figure 4.

MEEP–Ph/MEEP solutions showed a remarkable increase in turbidity above 57 °C with increasing temperature. Figure 4 also illustrates the temperature dependence of the hydrodynamic radius ( $R_h$ ) of the micelles. In the temperature range 25–50 °C, the  $R_h$  of the micelles was slightly increased from 120 to 140 nm. However, as the temperature was increased, a dramatic increase of  $R_h$  from 140 to 290 nm was also detected in





**Figure 3.** TEM micrograph of MEEP-Ph/MEEP micelles.

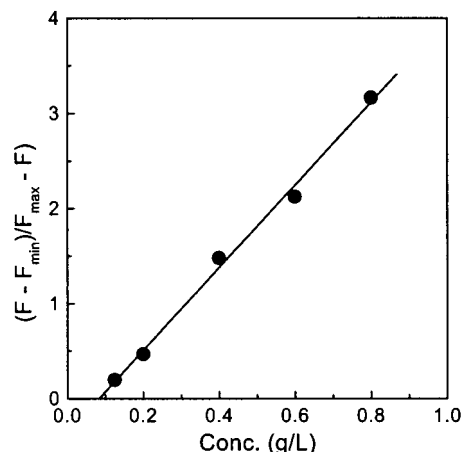


**Figure 4.** (●) Percent transmittance change of a 0.5 g/L MEEP-Ph/MEEP solution as a function of temperature. (○) Temperature dependence of hydrodynamic radius ( $R_h$ ) of MEEP-Ph/MEEP micelles in a 0.5 g/L solution.

the temperature range 53–60 °C. This suggests that the temperature increase makes the hydrophilic MEEP block somewhat hydrophobic and consequently induces the intermicellar aggregation. This type of temperature-dependent aggregation of micelles is observed from the block copolymer, poly(*N*-isopropylacrylamide)–poly(DL-lactide), which has a hydrophilic poly(*N*-isopropylacrylamide) block with LCST.<sup>28</sup> It is also of interest to note that the aggregates of micelles at the high-temperature level were found to retain the narrow size distribution, as judged by low  $\mu_2/\Gamma^2$  (0.05–0.17).

#### Partitioning of Pyrene in a Micellar Solution.

The hydrophobicity of the hydrophobic microdomain of the micelles was estimated by measuring the partition equilibrium constant  $K_v$  of pyrene, a hydrophobic probe, in the micellar solutions of the block copolymer MEEP-Ph/MEEP. In this work, the equilibrium constant  $K_v$  was calculated by the method of Wilhelm et al.<sup>21</sup> In this approach, a simple equilibrium between a micellar (Ph/MEEP) phase and a water phase was assumed for pyrene binding to the micelles. In this assumption, the ratio of pyrene concentration in the micellar phase to



**Figure 5.** Plots of  $(F - F_{\min})/(F_{\max} - F)$  vs concentration of MEEP-Ph/MEEP in water.

the water phase ( $[Py]_m/[Py]_w$ ) can be correlated to the ratio of volume of each phase as expressed in eq 2.

$$[Py]_m/[Py]_w = K_v V_m/V_w \quad (2)$$

Equation 2 can be rewritten as

$$[Py]_m/[Py]_w = K_v x(c - cmc)/1000\rho \quad (3)$$

where  $x$  is the weight fraction of Ph/MEEP,  $c$  is the concentration of the block copolymer, and  $\rho$  is the density of the MEEP-Ph/MEEP core of micelles, which is assumed as the bulk density of Ph/MEEP (1.08 g/mL). In the intermediate range of polymer concentration with substantial increases of intensity ratios ( $I_{339}/I_{335}$ ),  $[Py]_m/[Py]_w$  can be written as

$$[Py]_m/[Py]_w = (F - F_{\min})/(F_{\max} - F) \quad (4)$$

where  $F_{\max}$  and  $F_{\min}$  correspond to the average magnitude of  $I_{339}/I_{335}$  in the flat region of high- and low-concentration ranges in Figure 2, and  $F$  is the intensity ratio ( $I_{339}/I_{335}$ ) in the intermediate concentration range of the conjugates. Combining eqs 3 and 4,  $K_v$  values of pyrene are determined by using a plot  $(F - F_{\min})/(F_{\max} - F)$  versus MEEP-Ph/MEEP concentration as shown in Figure 5.

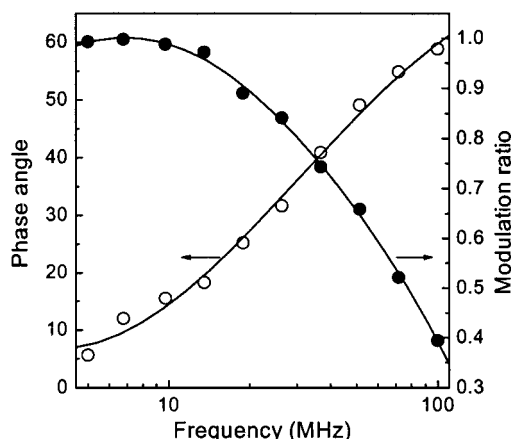
The  $K_v$  value of pyrene for MEEP-Ph/MEEP micelles was  $7.0 \times 10^3$ . It is interesting to note that the  $K_v$  value of pyrene for MEEP-Ph/MEEP is about 2 orders of magnitude lower than those of other polymeric micelles, indicating a low hydrophobicity of the microdomain constructed from Ph/MEEP block in which one phosphorus atom possesses both phenyl and 2-(2-methoxyethoxy)ethoxy substituents.<sup>16,21</sup> For PEO–polystyrene block copolymer micelle, the  $K_v$  value was reported as  $3.0 \times 10^5$ .<sup>21</sup> In addition, poly(2-ethyl-2-oxazoline)–poly( $\epsilon$ -caprolactone) and poly(2-ethyl-2-oxazoline)–poly(L-lactide) had  $K_v$  values on the order of  $\sim 10^5$ .<sup>16</sup>

**Microviscosity of the Micellar Core.** The microviscosity of the micellar hydrophobic microdomain was estimated by the measurement of the steady-state fluorescence anisotropy originated from the depolarization of DPH fluorescence due to the rotational diffusion of DPH in the micellar core. The anisotropy value increases with increasing the microviscosity of the micellar core because the rotational diffusion of DPH is increasingly hindered. The anisotropy value,  $r$ , measured for MEEP-Ph/MEEP block copolymer micelles is

**Table 2. Properties of MEEP–Ph/MEEP Micelles**

block copolymer	cmc <sup>a</sup> (mg/L)	R <sub>h</sub> <sup>b</sup> (nm)	μ <sub>2</sub> /Γ <sup>2 c</sup>	K <sub>v</sub> (×10 <sup>-3</sup> )	r <sup>d</sup>
MEEP–Ph/MEEP	80	120	0.12	7.0	0.237

<sup>a</sup> Measured at 25 °C. <sup>b</sup> Hydrodynamic radius determined by dynamic light scattering at 25 °C. <sup>c</sup> Polydispersity factor. <sup>d</sup> Steady-state fluorescence anisotropy of DPH.

**Figure 6.** Frequency-dependent phase (○) and modulation (●) data for DPH in MEEP–Ph/MEEP solution.**Table 3. Lifetime Data for DPH in Block Copolymer Solution**

block copolymer	τ <sub>1</sub> (ns)	f <sub>1</sub> <sup>a</sup>	τ <sub>2</sub> (ns)	f <sub>2</sub> <sup>a</sup>	χ <sup>2</sup>	⟨τ⟩ <sup>b</sup> (ns)
MEEP–Ph/MEEP	4.1	0.91	0.6	0.09	11.07	3.84

<sup>a</sup> Fractional intensity,  $f_i = \alpha_i \tau_i / \sum \alpha_j \tau_j$ , where  $\alpha_i$  is preexponential factor representing the fractional contribution to the time-resolved decay of the component with a lifetime  $\tau_i$ . <sup>b</sup> Average lifetime,  $\langle \tau \rangle = \tau_1 f_1 + \tau_2 f_2$ .

0.237 (Table 2). It is worth comparing  $r$  values with those of poly(ethylene-*co*-maleic acid) (0.187), poly(1-decene-*co*-maleic acid) (0.225), and poly(1-octadecene-*co*-maleic acid) (0.273).<sup>29</sup> Considering the high flexibility of phosphazene backbone, the relatively high anisotropy value (0.237) might be probably due to rigid phenyl units of the hydrophobic Ph/MEEP block. The  $r$  value of DPH for the diblock copolymer based on PEO and poly[bis(trifluoroethoxy)phosphazene] was about 0.10.<sup>30</sup>

**Fluorescence Lifetime.** The site-specific polarity for the hydrophobic microdomain of micelles was estimated by the measurement of the fluorescence lifetime of DPH using a phase and modulation method.<sup>16,31</sup> The lifetime value of DPH decreases with increasing the polarity of the hydrophobic core in micelles.<sup>31</sup> The phase and modulation data at logarithmically spaced frequencies for DPH in the presence of MEEP–Ph/MEEP are plotted in Figure 6. When using discrete biexponential functions, the goodness of fit ( $\chi^2$ ) was obtained more satisfactorily than with any other exponential function, as judged by the  $\chi^2$  values.

The fluorescence lifetimes of DPH were determined to be 4.1 ns for a major component (91%) and 0.6 ns for a minor component (9%), respectively (Table 3). These lifetime values obtained in a discrete biexponential function indicated that DPH predominantly associated with the interior of the hydrophobic microdomain. The fast component may arise from the DPH, located in the relatively polar domain such as the interface between the hydrated MEEP shell and hydrophobic microdomain.

For SDS micelles, the DPH lifetime of the main fraction was reported as 5.52 ns.<sup>29</sup> For block copolymers such as poly(1-decene-*co*-maleic acid) and poly(1-octadecene-*co*-maleic acid), the DPH lifetime values of the main domain were reported as 10.62 and 11.58 ns, respectively.<sup>29</sup> The lifetime data of DPH were consistent with the  $K_v$  value of pyrene in the MEEP–Ph/MEEP solution in terms of indicating low hydrophobicity of the micellar domain formed by Ph/MEEP block.

## Conclusions

The amphiphilic diblock copolymer (MEEP–Ph/MEEP) with  $[N=P(OCH_2CH_2OCH_2CH_2OCH_3)_2]_n$  (MEEP) as a hydrophilic block and  $[N=PPh(OCH_2CH_2OCH_2CH_2OCH_3)]_m$  (Ph/MEEP) as a hydrophobic block was synthesized via a controlled cationic polymerization of phosphoranimine. MEEP–Ph/MEEP self-associated to form micelles with a critical micelle concentration of 80 mg/L in an aqueous solution. The hydrodynamic radius of the micelles was 120 nm at 25 °C with a narrow distribution. The micellar solutions of MEEP–Ph/MEEP exhibited a temperature-dependent phase transition behavior due to the intermicellar aggregation, which might be associated with the LCST of hydrophilic MEEP block. The hydrophobicity of the micellar core was estimated by the measurement of partition equilibrium constant,  $K_v$ , of pyrene in the micellar solution of MEEP–Ph/MEEP. The  $K_v$  value was  $7.0 \times 10^3$ , which was much lower than those of other reported polymeric micelles. The steady-state fluorescence anisotropy value ( $r$ ) of DPH in a block copolymer solution was 0.237. The analysis of the fluorescence lifetime of DPH in the MEEP–Ph/MEEP solution indicated that the micellar core region was fairly polar and consisted of two hydrophobic domains with different polarity.

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## References and Notes

- (1) (a) Mark, J. E.; Allcock, H. R.; West, R. *Inorganic Polymers*; Prentice Hall: Englewood Cliffs, NJ, 1992. (b) Allcock, H. R. *Science* **1992**, *255*, 1106.
- (2) Allcock, H. R. *Adv. Mater.* **1994**, *6*, 106.
- (3) (a) Allcock, H. R.; Pucher, S. R.; Turner, M.; Fitzpatrick, R. *Macromolecules* **1992**, *25*, 5573. (b) Allcock, H. R.; Dudley, G. K. *Macromolecules* **1996**, *29*, 1313.
- (4) Allcock, H. R. In *Biodegradable Polymers as Drug Delivery Systems*; Langer, R., Chasin, M., Eds.; Marcel Dekker: New York, 1990.
- (5) (a) Allcock, H. R. *Chem. Mater.* **1994**, *6*, 1476. (b) Allcock, H. R.; Kwon, S.; Riding, G.; Fitzpatrick, R.; Bennet, J. *Biomaterials* **1988**, *9*, 509. (c) Allcock, H. R.; Pucher, S. R.; Visscher, K. *Biomaterials* **1994**, *15*, 502.
- (6) Honeyman, C. H.; Manners, I.; Morrissey, C. T.; Allcock, H. R. *J. Am. Chem. Soc.* **1995**, *117*, 7035.
- (7) Allcock, H. R.; Nelson, J. M.; Reeves, S. D.; Honeyman, C. H.; Manners, I. *Macromolecules* **1997**, *30*, 50.
- (8) Nelson, J. M.; Allcock, H. R. *Macromolecules* **1997**, *30*, 1854.
- (9) Allcock, H. R.; Reeves, S. D.; Nelson, J. M.; Crane, C. A. *Macromolecules* **1997**, *30*, 2213.
- (10) Nelson, J. M.; Allcock, H. R.; Manners, I. *Macromolecules* **1997**, *30*, 3191.
- (11) Nelson, J. M.; Primrose, A. P.; Hartle, T. J.; Allcock, H. R. *Macromolecules* **1998**, *31*, 947.
- (12) Gref, R.; Minamitake, Y.; Peracchia, M. T.; Trubetskoy, V.; Torchilin, V.; Langer, R. *Science* **1994**, *263*, 1600.

- (13) Kataoka, K.; Kwon, G. S.; Yokoyama, M.; Okano, T.; Sakurai, Y. *J. Controlled Release* **1993**, *24*, 119.
- (14) Yokoyama, M.; Kwon, G. S.; Okano, T.; Sakurai, Y.; Seto, T.; Kataoka, K. *Bioconjugate Chem.* **1992**, *3*, 295.
- (15) Kwon, G. S.; Suwa, S.; Yokoyama, M.; Okano, T.; Sakurai, Y.; Kataoka, K. *J. Controlled Release* **1994**, *29*, 17.
- (16) Lee, S. C.; Chang, Y.; Yoon, J.-S.; Kim, C.; Kwon, I. C.; Kim, Y.-H.; Jeong, S. Y. *Macromolecules* **1999**, *32*, 1847.
- (17) Ringsdorf, H.; Venzmer, J.; Winnik, F. M. *Macromolecules* **1991**, *24*, 1678.
- (18) Lakowicz, J. R. *Principles of Fluorescence Spectroscopy*; Plenum Press: New York, 1983.
- (19) Harada, A.; Kataoka, K. *Macromolecules* **1995**, *28*, 5294.
- (20) Harada, A.; Kataoka, K. *Macromolecules* **1998**, *31*, 288.
- (21) Wilhelm, M.; Zhao, C.; Wang, Y.; Xu, R.; Winnik, M. A.; Mura, J.; Riess, G.; Croucher, M. D. *Macromolecules* **1991**, *24*, 1033.
- (22) Nagasaki, Y.; Okada, T.; Scholz, C.; Iijima, M.; Kato, M.; Kataoka, K. *Macromolecules* **1998**, *31*, 1473.
- (23) Kwon, G. S.; Naito, M.; Yokoyama, M.; Okano, T.; Sakurai, Y.; Kataoka, K. *Langmuir* **1993**, *9*, 945.
- (24) Kabanov, A. V.; Nazarova, I. R.; Astafieva, I. V.; Batrakova, E. V.; Alakhov, V. Y.; Yaroslavov, A. A.; Kabanov, V. A. *Macromolecules* **1995**, *28*, 2303.
- (25) Astafieva, I.; Zhong, X. F.; Eisenberg, A. *Macromolecules* **1993**, *26*, 7339.
- (26) La, S. B.; Okano, T.; Kataoka, K. *J. Pharm. Sci.* **1996**, *85*, 85.
- (27) Allen, C.; Yu, Y.; Maysinger, D.; Eisenberg, A. *Bioconjugate Chem.* **1998**, *9*, 564.
- (28) Kohori, F.; Sakai, K.; Aoyagi, T.; Yokoyama, M.; Sakurai, Y.; Okano, T. *J. Controlled Release* **1998**, *55*, 87.
- (29) McGlade, M. J.; Randall, F. J.; Tcheurekdjian, N. *Macromolecules* **1987**, *20*, 1782.
- (30) Prange, R.; Allcock, H. R.; Chang, Y.; Lee, S. C.; Kim, C., unpublished results.
- (31) Lee, K. Y.; Jo, W. H.; Kwon, I. C.; Kim, Y.-H.; Jeong, S. Y. *Macromolecules* **1998**, *31*, 378.

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