

## Supplementary Material

### Phase Selective Solubility of Poly(*N*-alkylacrylamide)s

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## Experimental Section

***N*-Octadecylacrylamide.** Octadecylamine (26.7 g) was suspended in 500 mL of dichloromethane. Then 16.9 mL of triethylamine was added, and the mixture was cooled to 0 °C using an ice-water bath. At this point, 50 mL of a CH<sub>2</sub>Cl<sub>2</sub> solution containing 8.9 mL of acryloyl chloride was added dropwise to the amine suspension. The reaction mixture was warmed to room temperature and stirred for 1h. The solvent was removed under reduced pressure and the residue was taken up in 250 mL of chloroform, washed with water and brine, dried over sodium sulfate and filtered. The filtrate was concentrated under reduced pressure and the residue was recrystallized from acetone. The product was isolated by filtration and had mp 70-71 °C (lit<sup>1</sup> 74.5-75.1 °C); <sup>1</sup>H NMR: δ (CDCl<sub>3</sub>) 6.3 (d, 1H), 6.1 (dd, 1H), 5.6 (overlapping d and br s, 2H), 3.3 (q, 2H), 1.5 (t, 2H), 1.3 (bs, 30H), 0.9 (t, 3H); <sup>13</sup>C NMR δ (CDCl<sub>3</sub>) 165.78, 131.18, 126.35, 39.84, 32.13, 29.90, 29.87, 29.86, 29.85, 29.79, 29.78, 29.75, 29.57, 29.51, 27.15, 22.90, 14.33; IR (thin film) 1670 cm<sup>-1</sup>.

**Poly(*N*-isopropylacrylamide-co-NASI)(PNIPAM-NASI).** This copolymer was prepared using a published procedure.<sup>2</sup>

**Poly(*N*-octadecylacrylamide-co-NASI)(PNODAM-NASI).** Benzene (CAUTION: benzene is a known carcinogen and should be handled appropriately) (50 mL) was degassed with bubbling N<sub>2</sub> for 30 min. Then *N*-octadecylacrylamide monomer (4.5 g, 13.9 mmol) and 240 mg of *N*-acryloxysuccinimide (1.39 mmol) were added. The reaction mixture was warmed to 80°C and a solution of AIBN (12.6 mg, 0.077 mmol) in 2 mL of benzene was added. The resulting polymerization reaction mixture was stirred at 80°C under nitrogen for 36 h. At that point, the solvent was removed at reduced pressure using a rotary evaporator and the residue was taken up in 30 mL of CHCl<sub>3</sub> and precipitated into 150 mL of cold methanol to give 4.1 g (87%) of a white powder. <sup>1</sup>H NMR δ (CDCl<sub>3</sub>) 3.1 (br s, 20H), 2.8 (br s, 4H), 1.5 (br s, 27H), 1.2 (br s, 395H), 0.8 (t, 30H); IR (KBr) 1812, 17383, 1737, 1654 and 1542 cm<sup>-1</sup>.

**Methyl Red Labeled Poly(*N*-alkylacrylamide).** The necessary amine-terminated methyl red azo dye was made according to a literature procedure.<sup>3</sup> It was coupled to a sample of a copolymer of NIPAM or NODAM and NASI prepared as described above. To effect this coupling, the NASI containing copolymer was dissolved in THF. To this solution was added 0.1-0.2 equivalent (relative to NASI) of amine-terminated methyl red. This solution was stirred for 4 hours. Then excess amine (isopropyl or octadecylamine for PNIPAM or PNODAM, respectively) was added to consume any unreacted NASI groups and stirring was continued at room temperature overnight. Any precipitate that formed was removed by centrifugation. The product polymer was isolated by solvent precipitation with the exact procedure varying for the polar and nonpolar polymer products. In the case of the methyl red-labeled PNIPAM, the product polymer solution was added dropwise via pipette to an excess of hexanes. This formed an orange powder

that was isolated by filtration. This product was characterized spectroscopically; the  $^1\text{H}$  NMR and IR spectrum was essentially the same as PNIPAM ( $^1\text{H}$  NMR  $\delta$  ( $\text{CDCl}_3$ ) 4.0 (br, s, 1H), 2.4-1.4 (br, m, 3H), 1.1 (br, 6H); IR (thin film)  $1648\text{ cm}^{-1}$ ). However, the presence of azo dye label was evident in UV-visible spectroscopy (EtOH); 513 nm (the protonated form of the azo dye), 437 nm (the neutral form of the azo dye). The product PNIPAM polymer's molecular weight was characterized by viscosimetry,  $M_v = 600,000$  (THF,  $30\text{ }^\circ\text{C}$ ).<sup>4</sup> In the case of the methyl red-labeled PNODAM, the solvent from the product polymer solution was first removed under reduced pressure. The resulting red polymer solid was then dissolved in  $\text{CHCl}_3$  and added dropwise to an excess of methanol to form an orange powder that was isolated by filtration. As was true for PNIPAM, the  $^1\text{H}$  NMR and IR spectra were the same as PNODAM ( $^1\text{H}$  NMR  $\delta$  ( $\text{CDCl}_3$ ) 3.3(q, 2H), 1.8-1.3 (br, 5H), 1.2 (br, 30H), 0.8 (t, 3H); IR (thin film)  $1651\text{ cm}^{-1}$ ) but the presence of the azo dye was seen in UV-visible spectroscopy (heptane); 515 nm (the protonated form of the azo dye), 419 nm (the neutral form of the azo dye).

**Dansyl Labeled Poly(*N*-alkylacrylamide).** The necessary amine-terminated dansyl probe was made according to a literature procedure.<sup>5</sup> It was then coupled to a sample of a copolymer of NIPAM or NODAM and NASI prepared as described above. To effect this coupling, the NASI containing copolymer was dissolved in THF and 1.5 equivalent of  $\text{Et}_3\text{N}$  (relative to NASI) was added. To this solution was added 0.01 equivalent of amine-terminated dansyl derivative in THF. The solution was stirred at room temperature for an hour, then 2 equiv of isopropylamine or octadecylamine were added and the mixture was stirred overnight. Any precipitate that formed during this reaction was removed by centrifugation. In the case of the PNIPAM support, the PNIPAM derivative

was isolated after precipitation using excess hexanes. In the case of the PNODAM support, the PNODAM derivative was isolated after precipitation using excess methanol. The characterization results were similar to those of the corresponding dye-labeled polymers except for the polymers' fluorescence properties (vide infra).

**Poly(*N*-acryloxysuccinimide) (PNASI).** *N*-Acryloxysuccinimide (NASI) was made according to the procedure described by Whitesides.<sup>6</sup> This monomer was then used in a polymerization reaction as follows. Benzene (300 mL) was first placed in a 500-mL flask that contained a reflux condenser. Bubbling of N<sub>2</sub> for 30 min was used to degas this solution. Then NASI (5 g, 29.6 mmol) was added to the flask. After it dissolved, the flask was placed in a 60 °C oil bath and allowed to heat for 30 min under nitrogen. Then a solution of azobisisobutyronitrile (AIBN) (19 mg, 0.118 mmol) in 2 mL of benzene was added and the reaction was allowed to stir at 60 °C overnight. The initially homogeneous reaction contained a significant amount of white precipitate the next day. The solvent was removed from this benzene suspension of product at reduced pressure using a rotary evaporator and the residue was dissolved in ~70 mL of dry DMF with heating. This dissolution takes some time. After the polymer had dissolved, the solution was added dropwise quickly from a pipette into 500 mL of a mixed solution containing a 40:60 (vol:vol) mixture of hexane and THF. A white powder formed which was isolated by filtration (3.7 g). The product was characterized by <sup>1</sup>H NMR spectroscopy but the <sup>1</sup>H NMR was of poor quality with broad peaks: <sup>1</sup>H NMR δ (DMSO-*d*<sub>6</sub>) 2.0 (s, br 2 H), 2.8 (s, br 4 H), 3.1 (s, br 1 H); <sup>13</sup>C NMR δ (DMSO-*d*<sub>6</sub>) (broad peaks) 171-169, 39-31, 27-24; IR (Nujol mull) 1810, 1781, 1740 cm<sup>-1</sup>.

**Synthesis of a Library of Methyl Red-Labeled Poly(*N*-alkylacrylamide)s from a Common Poly(*N*-acryloxysuccinimide) Precursor.** Poly(*N*-acryloxysuccinimide) was first prepared according to a literature procedure.<sup>7</sup> Portions of this polymer were then placed in separate reactors and treated as follows. Each sample of PNASI (0.59 mequiv of active ester, 100 mg) was first dissolved in DMF. Then a mixture of the primary alkylamine and a primary amine derivative of methyl red were added. Each mixture of methyl red amine and alkylamine used 0.003 g (0.00887 mmol) of the methyl red derivative and 0.887 mmol of the alkylamine. Each reaction used the same amount (ca. 0.9 mmol) of the alkylamine. Each reaction used a total volume of 20 mL of DMF. Each amidation reaction was carried out under identical conditions. The mixture of active ester polymer and amines was heated to 50 °C for 24 hours. At this point, each of the reddish solutions contained the desired methyl red-labeled poly(*N*-alkylacrylamide) copolymer, excess unreacted primary amine-terminated methyl red and excess alkylamine. While the product polymers at this point could have been isolated by a solvent precipitation as described above, such a precipitation process could fractionate the polymer, separating the polymer into fractions with differing degrees of polymerization and differing polydispersities. To avoid this, a slightly different work-up was used. In this workup, the excess amine was removed from these solutions using Amberlyst 15. This treatment involved shaking each solution with 0.35 g of Amberlyst 15 for 24 h at room temperature using a wrist action shaker. The red suspensions were then filtered to remove the Amberlyst resin. The eight different polymers were then isolated from each experiment by removing the solvent under reduced pressure. Drying in vacuo led to a mixture of polymer samples that were then tested for phase selective solubility as described below.

The molecular weight of the PNIPAM member of this library was determined by both viscosity and GPC ( $M_n = 1.8 \times 10^5$ ,  $M_w = 3.4 \times 10^5$ ,  $M_v = 3.3 \times 10^5$ ).

**Tests of Non Reactivity of Polymer Dyes and Sulfonated Polystyrene Resin.** To test whether the Amberlyst-15 had removed all of the low molecular weight amines in each of the above experiments, one of the samples (the isopropylamine-containing derivative) was analyzed by UV-visible spectroscopy. The absorbance was noted. Then this sample was treated with an additional 300 mg of Amberlyst-15 with an additional 1 h of shaking at room temperature. At this point, the suspension was allowed to settle and the supernatant was again analyzed by UV-visible spectroscopy. At this point, the absorbance of the supernatant was unchanged. Since there was no change in intensity of the absorbance peak for the methyl red dye, this indicated that all the excess methyl red had been removed by the Amberlyst-15. This same experiment showed that Amberlyst-15 does not absorb the methyl red-labeled poly(*N*-alkylacrylamide)s since the polymer most likely to have been absorbed by the sulfonic acid-containing resin was the smallest most polar polymer, the methyl red derivative of poly(*N*-isopropylacrylamide).

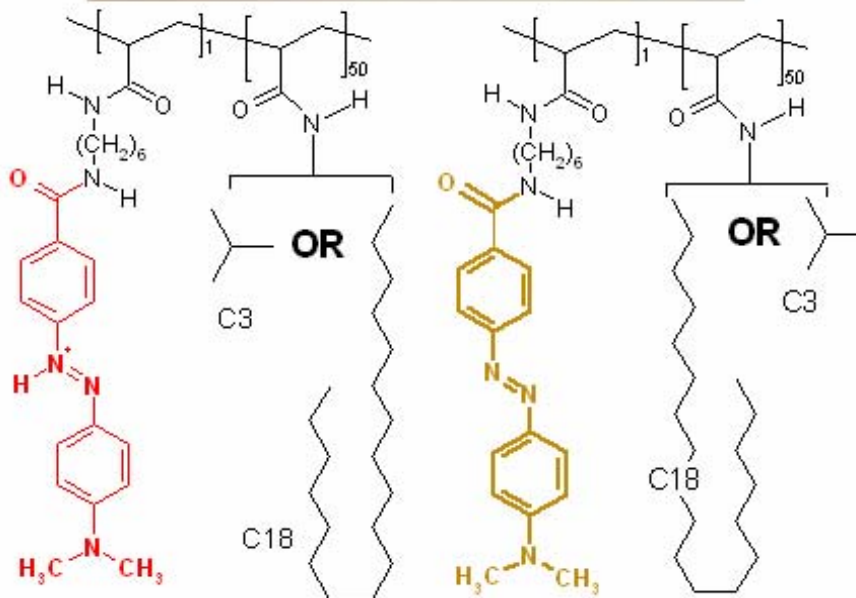
**UV-visible Analysis using the Methyl Red-Labeled Polymers 3 and 4.** The procedure for analysis of phase selective solubility of the neutral form of **3** is typical of the analysis used for either **3** or **4** with the difference being the initial solvent (90% EtOH-H<sub>2</sub>O for **3** and heptane for **4**). The dye labeled polymer **3** was first dissolved in 90% aqueous ethanol so that the methyl red concentration was around  $10^{-3}$  M. Then an equal volume of heptane was added and the biphasic mixture was heated to 70 °C. At this point a homogeneous solution formed. This solution was then cooled down to room temperature and this cooling led to phase separation. This process was completely

reversible. To analyze the dye content of the respective phases, the nonpolar phase and the polar phase were separated from one another. Each phase was analyzed by UV-visible spectroscopy. The  $\lambda_{\text{max}}$  of these two polymers in different solvents is given in the text and in the experimental section above. The extent of phase separation was estimated by assuming the polymer bound dye would have similar extinction coefficients in either phase.

**Fluorescence Studies using the Dansyl-Labeled Polymers 5 and 6.** The dansyl-labeled polymer **5** was first dissolved in DMF or 90% aqueous ethanol so that the dansyl label concentration was  $5 \times 10^{-5}$  M. Then an equal volume of heptane was added and the biphasic mixture was heated to 70 °C to form a homogenous solution. The solution was then cooled down to room temperature to induce phase separation. The nonpolar phase and the polar phase were separated from one another, and each phase was analyzed by fluorescence spectroscopy. Experiments using **6** were identical except that the polymer was initially dissolved in heptane and then mixed with the polar solvent. All the analyses used a  $\lambda_{\text{EX}}$  of 343 nm. The  $\lambda_{\text{EM}}$  values depended on the individual experiment. Experiments where **5** ended up in the EtOH-rich phase had a  $\lambda_{\text{EM}} = 513$  nm. Experiments where **5** ended up in the DMF-rich phase had a  $\lambda_{\text{EM}} = 507$  nm. Experiments where **6** ended up in the heptane-rich phase from an EtOH-H<sub>2</sub>O-heptane mixture had a  $\lambda_{\text{EM}} = 496$  nm. Experiments where **6** ended up in the heptane-rich phase from a DMF-heptane mixture had a  $\lambda_{\text{EM}} = 492$  nm. Experiments where **5** ended up in the Et<sub>3</sub>N phase from a miscible Et<sub>3</sub>N-H<sub>2</sub>O mixture had a  $\lambda_{\text{EM}} = 511$  nm. The extent of phase separation was estimated by assuming the polymer bound fluorophore would have equivalent intensity if it were present in either phase. In the examples studied, the fluorophore was exclusively

in one phase. Estimates of the amount of fluorophore that could have been detected in the other phase were based on a series of serial dilutions of the phase that had the fluorophore. A lower limit for the extent of fluorophore was estimated based on the amount of fluorophore that was detectable in the most diluted of these samples.

**Photographs.** Photographs of phase separated solutions of the protonated (red) and unprotonated (yellow) forms of the EtOH-H<sub>2</sub>O-soluble PNIPAM-bound methyl red and the heptane-soluble PNODAM-bound methyl red dyes **3** and **4**.



## Experimental Section References

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