

## Novel amphiphilic linear polymer/dendrimer block copolymer: Synthesis of poly(2-methyl-2-oxazoline)-*block*-poly(amido amine) dendrimer

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### SUMMARY:

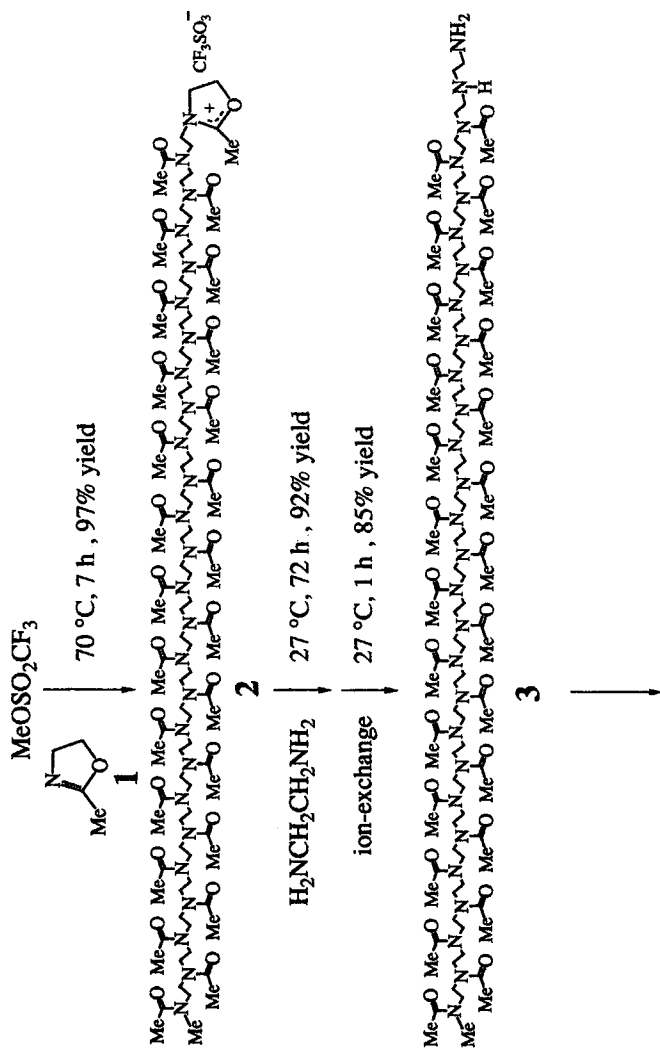
Novel linear polymer/dendrimer block copolymers, poly(2-methyl-2-oxazoline)-*block*-poly(amido amine) dendrimers (water-soluble full-generation type 4 ( $G = 4.0$  and  $5.0$ ) and amphiphilic half-generation type 5 ( $G = 3.5, 4.5,$  and  $5.5$ )), were synthesized by divergent-growth dendrimer construction with  $\omega$ -ethylenediamine-terminated poly(2-methyl-2-oxazoline), which was prepared by living ring-opening polymerization of 2-methyl-2-oxazoline. Assembly of the amphiphilic dendrimer-based block copolymer ( $G = 5.5$ ) was investigated by surface tension measurements (critical micelle concentration, 0.49 wt.-%) and by small-angle neutron scattering analysis (spherical particles; assembled number, ca.  $10^3$ ).

### Introduction

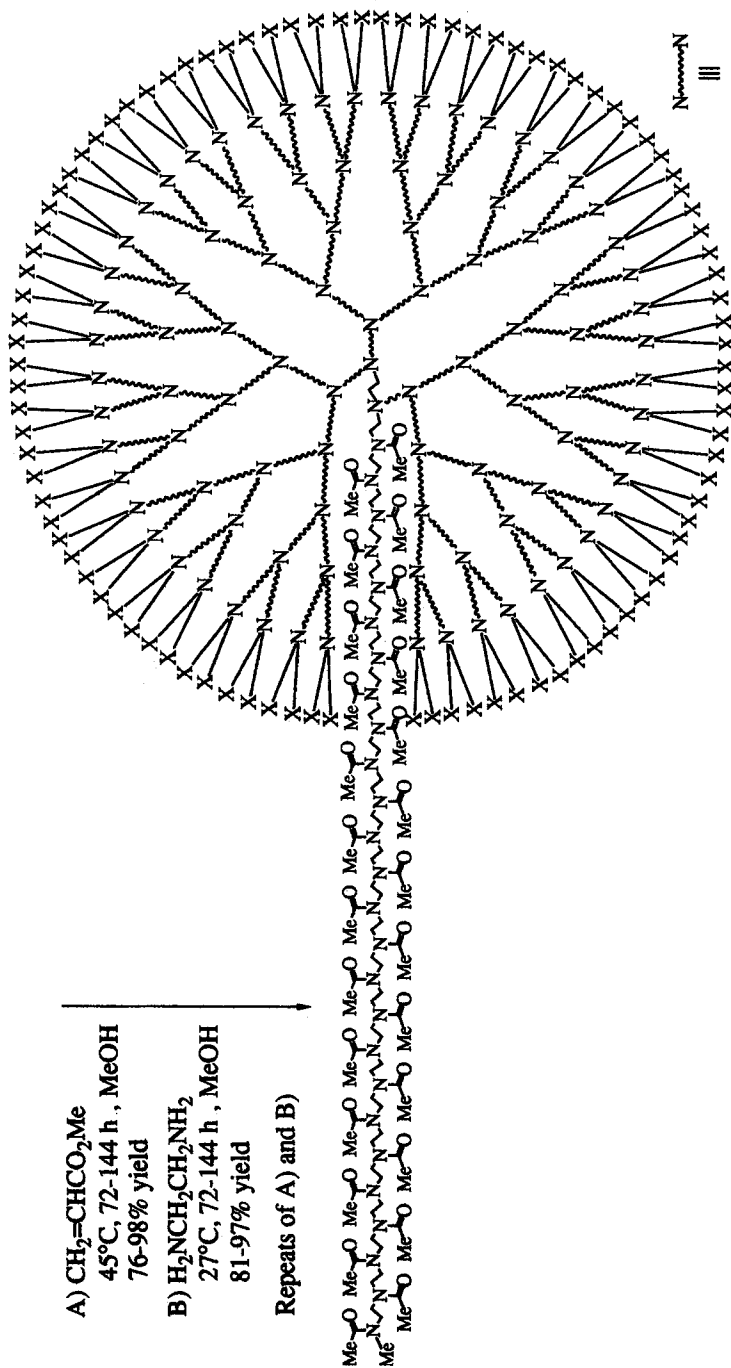
The combination of supramolecular chemistry<sup>1)</sup> and dendrimer chemistry<sup>2–5)</sup> has offered widespread possibilities. Self-assembly of dendrimers is of importance, since organization of native globular macromolecules such as globular proteins is a key to function in various life systems. However, little is known concerning dendrimers having molecular recognition ability and their association phenomena. Artificial assemblies of dendrimers were demonstrated by using a surface-block dendrimer<sup>5)</sup> and a dendrimer with a recognizing and assembling moiety<sup>6)</sup>. We have already reported the synthesis of globular carbohydrate macromolecules “Sugar Balls”, dendrimers covered with a surface sugar shell, as a typical example of dendritic functional materials with molecular information and molecular recognition ability<sup>7,8)</sup>. Aggregation phenomena between Sugar Balls and proteins such as lectins have been investigated. Meijer et al.<sup>9)</sup> have recently presented synthesis and properties of a block copolymer of polystyrene with dendritic poly(propylene imine), which is the hydrophilic part. “Hydraamphiphile”, a polymeric surfactant of poly(lysine) dendrimer/poly(ethylene oxide) block copolymer, has also been synthesized<sup>10)</sup>.

In this paper, we describe the synthesis of novel poly(2-methyl-2-oxazoline)/poly(amido amine) (PAMAM) dendrimer block copolymers. Supramolecular assemblies of a globular dendrimer, which is regarded as covalent-bonded unimolecular

Scheme 1:

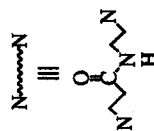


- A)  $\text{CH}_2=\text{CHCO}_2\text{Me}$   
45°C, 72-144 h, MeOH  
76-98% yield
- B)  $\text{H}_2\text{NCH}_2\text{CH}_2\text{NH}_2$   
27°C, 72-144 h, MeOH  
81-97% yield
- Repeats of A) and B)



**4** ( $G = 4.0$ ),  $X = \text{H}$ , Generation = 4.0

**5** ( $G = 4.5$ ),  $X = \text{CH}_2\text{CH}_2\text{CO}_2\text{Me}$ , Generation = 4.5



micelles<sup>2,11</sup>), are feasible by employing an amphiphilic “tadpole-shaped” structure of a globular dendrimer with a linear tail. Amphiphilic dendrimers will provide a new hierarchy of supramolecular nano-architecture consisting of covalent-linked organized dendritic macromolecules. The properties of globular dendrimers are considered to largely depend on the terminal substituents on the surface, instead of the inner branching skeleton. Although the amine-terminated PAMAM dendrimer is known to be water-soluble, the globular methyl ester-covered PAMAM dendrimer is taken to be rather hydrophobic compared with the strong hydrophilic poly(2-methyl-2-oxazoline) block, in the present work. This block copolymer has an advantage in the macromolecular design of dendrimer-based amphiphiles, because there is a potential to vary hydrophilicity and hydrophobicity by changing the substituents of the polyoxazolines and the terminal groups of the dendrimer.

## Results and discussion

### *Synthesis of poly(2-methyl-2-oxazoline)-block-poly(amido amine) (PAMAM) dendrimers*

The block copolymers poly(2-methyl-2-oxazoline)-*block*-poly(amido amine) (PAMAM) dendrimers (amine-terminated type **4** and methyl ester-terminated type **5**) were synthesized by divergent-growth dendrimer architecture with  $\omega$ -ethylenediamine-terminated poly(2-methyl-2-oxazoline) **3** according to *Scheme 1*. Polyoxazoline is known to possess useful characteristics such as high miscibility toward common polymers and good solubility in various solvents<sup>12</sup>. The solubility of polyoxazolines can be easily changed by the character of the 2-substituents of the oxazoline monomers. In order to make a hydrophilic chain, we employed poly(2-methyl-2-oxazoline), which exhibits high hydrophilicity<sup>13</sup>. Ring-opening polymerization of 2-methyl-2-oxazoline (**1**) was carried out to generate living polyoxazoline **2** with methyl trifluoromethanesulfonate as an initiator in acetonitrile under dry conditions<sup>12</sup>. The active end of **2** was successfully terminated by the reaction with 450-fold molar excess ethylenediamine at 27°C and subsequent ion-exchange<sup>14</sup>. A concerned by-product, *N,N'*-bis[poly(2-methyl-2-oxazoline)]-substituted ethylenediamine, was not detected by <sup>1</sup>H NMR spectroscopy and size exclusion chromatography (SEC). The resulting novel  $\omega$ -ethylenediamine-terminated poly(2-methyl-2-oxazoline) **3** has a relatively explicit chemical structure (DP = 26 by <sup>1</sup>H NMR,  $\bar{M}_w/\bar{M}_n = 1.1_2$  by SEC (in chloroform), functionality (terminal amino groups) = 0.96 by titration with 0.060 N HCl aq., 3400 ( $\nu_{N-H}$ ) cm<sup>-1</sup> by IR).

By using the  $\omega$ -amino group of **3** as an initiator site, PAMAM dendritic branches were extended by alternately repeating reaction sequences of Michael addition with methyl acrylate and amide formation with ethylenediamine according to the previously outlined procedure<sup>2</sup>. Especially, initial triple Michael addition of **3** with methyl acrylate was carefully confirmed by <sup>1</sup>H NMR spectra. Amine-terminated full-generational PAMAM dendrimer derivatives (generation 4.0 and 5.0) **4** (*G* = 4.0 and 5.0) and methyl ester-terminated half-generational derivatives **5** (*G* = 3.5, 4.5, and 5.5) were synthesized. The chemical structure of **4** and **5** was determined by <sup>1</sup>H

and  $^{13}\text{C}$  NMR and IR spectroscopies. The  $\bar{M}_w/\bar{M}_n$  value of **5** ( $G = 3.5$ ) estimated by SEC in dimethyl sulfoxide was 1.1<sub>8</sub>, which is close to the  $\bar{M}_w/\bar{M}_n$  value of polyoxazoline **3**.

The amphiphilic character of half-generational **5** was examined by a solubility test. **5** was readily soluble in water as well as **4**. However, for organic solvents, **5** was dissolved in *N,N*-dimethylformamide, acetone, chloroform, and 1,4-dioxane, whereas **4** was insoluble in these solvents. The results suggest amphiphilicity of **5** with the lipophilic dendritic globule.

#### *Assembly of poly(2-methyl-2-oxazoline)/methyl ester-ended PAMAM dendrimer block copolymer*

Self-assembly formation of **5** having a unique structure of a hydrophobic globular head with a hydrophilic linear tail was investigated in an aqueous solution by measuring the surface tension ( $\gamma$ ) values by means of the drop weight method<sup>15</sup>. Results are shown in Fig. 1. Critical micelle concentrations (CMCs) were clearly observed in both cases. CMC of **5** ( $G = 5.5$ ) was lower than that of **5** ( $G = 3.5$ ) (0.49 and 2.2 wt.-%, respectively). This means that **5** ( $G = 5.5$ ) with a relatively higher hydrophobic globule assembles effectively in comparison with **5** ( $G = 3.5$ ) having dendritic branches.

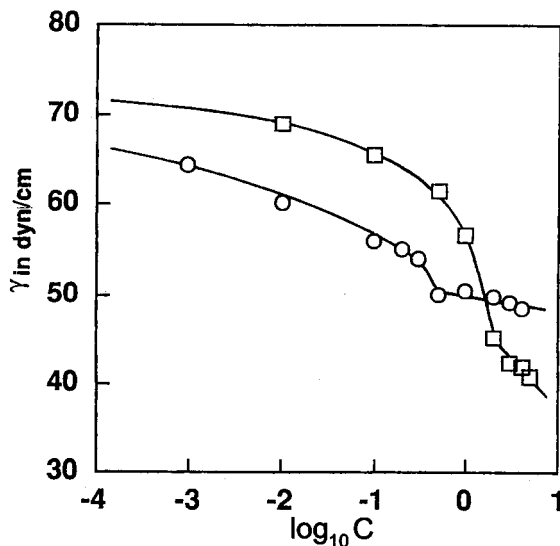


Fig. 1. Surface tension ( $\gamma$ ) of an aqueous solution of poly(2-methyl-2-oxazoline)-block-poly(amidoamine) dendrimer **5** (□;  $G = 3.5$ , ○;  $G = 5.5$ ) at 25 °C. The unit of concentration  $C$  is wt.-%. CMCs are indicated by the inflection points. The experimental error was within 0.5 dyn/cm

Moreover, small angle neutron scattering (SANS) investigations were carried out for assemblies of **5**. SANS was measured using a 3.0 wt.-%  $\text{D}_2\text{O}$  solution of **5** ( $G = 5.5$ ) at 25 °C. The resulting data reasonably fitted a Guinier equation of a spherical particle model, although rod-like models were not appropriate<sup>16</sup>. The evaluated average particle radius of assembled **5** ( $G = 5.5$ ) was 270 Å. An assembled number of **5** ( $G = 5.5$ ) was estimated to be approximately  $10^3$ .

In this study, we described the synthesis and assembly of a novel hydrophobic globular dendrimer-based amphiphile with a hydrophilic linear chain. Properties such as hydrophilic-lipophilic balance of the block copolymer will be easily regulated by derivatization of polyoxazoline<sup>12)</sup> and outer terminal groups of the dendrimer<sup>7)</sup>. The new class of assemblies affords a wide variety of interest from fundamental sciences to applications, e. g., detergents, cosmetics, drug delivery systems, emulsion polymerization, coating technology. Besides the amphiphilic character, water-soluble block copolymer **4** is taken as a new type of backbone for functional fine materials, especially for biochemical and biomedical fields. For example, cell-recognizable DNA carriers are designed by using polymerization of oxazoline with sugar-bearing initiators<sup>17,18)</sup>. Progress in synthesis and applications of dendrimer-based block copolymers is hopeful.

## Experimental part

### Materials

Purification of all compounds used for living polymerization of 2-methyl-2-oxazoline and dendrimer construction was carried out under dry nitrogen. Methyl trifluoromethanesulfonate, 2-methyl-2-oxazoline (Aldrich Co.), ethylenediamine, and methyl acrylate were purified by repeated distillations. The solvents were dried by conventional methods and purified by repeated distillations, and then stored over molecular sieves 3 Å.

### Instruments

<sup>1</sup>H and <sup>13</sup>C NMR spectra were recorded on a Bruker ARX-400 NMR spectrometer (400 MHz and 100 MHz, respectively). IR measurement was carried out with a Jasco FT/IR-5MP spectrophotometer. Size exclusion chromatography (SEC) was performed with a Jasco Model DIP-1 high performance liquid-chromatograph apparatus (column, Shodex KF803 → 804, 8 φ × 600 mm; solvent, dimethyl sulfoxide; temp., 27 °C) and by a Tosoh HLC-8020 system (column, Tosoh TSK-Gel G3000H<sub>XL</sub> and G2000H<sub>XL</sub>; solvent, chloroform; temp., 38 °C). Small angle neutron scattering (SANS) analysis was performed on a cold small angle neutron scattering instrument SANS-U at JRR-3M at the Japan Atomic Energy Research Institute, Tokai (the experimental conditions: wavelength of the spectrometer beam, 7 Å; scattering vector **Q** range, 0.004–0.08 Å<sup>-1</sup>; temp., 25 °C; the sample thickness, 4 mm). The SANS data were analyzed by a Guinier equation of a spherical particle model (Eqs. 1 and 2).

$$I(\mathbf{Q}) = I_0 \exp\left(-\frac{\langle R_G^2 \rangle}{3} \mathbf{Q}^2\right) \quad (1)$$

$$\langle R_G^2 \rangle = \frac{3}{5} \langle R^2 \rangle \quad (2)$$

**Q**: scattering vector, *I*(**Q**): intensity,

*I*<sub>0</sub>: constant, *R*<sub>G</sub>: radius of gyration

*R*: radius of spherical particle

*Termination of living poly(2-methyl-2-oxazoline) with ethylenediamine*

To 30 cm<sup>3</sup> (0.45 mol) of ethylenediamine in a flask equipped with a three-way stop-cock, an acetonitrile solution of 2.5 g (1.1 mmol) of living poly(2-methyl-2-oxazoline) (**2**) was added by a gastight syringe at 27 °C under nitrogen. After mixing for 72 h, **2** was purified by repeated reprecipitations (chloroform (solvent)/diethyl ether (non-solvent)). A part (2.4 g, 1.0 mmol) of the resulting product was dissolved in 100 cm<sup>3</sup> of dry chloroform, and then 11 g of ion-exchange resin Amberlist A-21 was added. The mixture was stirred at 27 °C for 1 h. Amberlist was removed by filtration.

*Poly(2-methyl-2-oxazoline)-block-poly(amido amine) dendrimer*

All operations in the stepwise dendrimer synthesis (Michael addition with methyl acrylate and amide formation with ethylenediamine) were carried out by using a gastight syringe under nitrogen, according to the procedure by Tomalia et al.<sup>2)</sup>

Poly(2-methyl-2-oxazoline)-*block*-PAMAM dendrimer (generation 4.5) **5** ( $G = 4.5$ ):

IR (KBr): 2950 ( $\nu_{C-H}$ ), 2850 ( $\nu_{C-H}$ ), 1740 ( $\nu_{C=O}$ , ester), 1640 ( $\nu_{C=O}$ , amide), 1550 ( $\delta_{N-H}$ , amide) cm<sup>-1</sup>.

<sup>1</sup>H NMR (CDCl<sub>3</sub>, TMS, 27 °C, 400 MHz):  $\delta = 7.17$  (br, NH), 3.67 (s, CO<sub>2</sub>CH<sub>3</sub>), 3.46 (m, CH<sub>2</sub> of polyoxazoline), 3.28 (m, NHCH<sub>2</sub> of PAMAM), 3.05 (s,  $\alpha$ -CH<sub>3</sub>N of polyoxazoline), 2.96 (m, NHCH<sub>2</sub>CH<sub>2</sub>N of PAMAM), 2.76 (t,  $J = 6.4$  Hz, NCH<sub>2</sub>CH<sub>2</sub>CO<sub>2</sub> of PAMAM), 2.63 (m, NCH<sub>2</sub>CH<sub>2</sub>CONH of PAMAM), 2.57–2.50 (m, CH<sub>2</sub>CONH of PAMAM), 2.44 (t,  $J = 6.4$  Hz, CH<sub>2</sub>CO<sub>2</sub> of PAMAM), 2.16, 2.12, and 2.09 (s  $\times 3$ , CH<sub>3</sub> of polyoxazoline).

<sup>13</sup>C NMR (CDCl<sub>3</sub>, TMS, 27 °C, 100 MHz):  $\delta = 173.1$  (C=O, ester), 171.3, 170.7 (C=O, amide), 52.9 (NHCH<sub>2</sub>CH<sub>2</sub>N of PAMAM), 51.7 (CO<sub>2</sub>CH<sub>3</sub>), 49.3 (NCH<sub>2</sub>CH<sub>2</sub>CONH of PAMAM), 47.9–43.4 (CH<sub>2</sub> of polyoxazoline), 37.2 (NHCH<sub>2</sub>CH<sub>2</sub>N of PAMAM), 32.7 (CH<sub>2</sub>CO<sub>2</sub> of PAMAM), 21.2 (CH<sub>3</sub> of polyoxazoline).

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