Hydrogel Formation by the 'Topological Conversion' of Cyclic PLA-PEO Block Copolymers

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Amphiphilic cyclic block copolymers consisting of poly(L- or D-lactide) and poly(ethylene oxide), i.e., PLLA–PEO or PDLA–PEO, were synthesized from their corresponding linear triblock precursors, PLLA–PEO–PLLA or PDLA–PEO–PDLA, respectively, with alkene end groups. A mixture of the respective micellar dispersions of linear PLLA–PEO–PLLA and linear PDLA–PEO–PDLA formed a gel upon heating, while a mixture of the cyclic counterparts did not show any sign of the phase transition. These results suggest that the gelation behaviour is directed by the topology of polymer components. Furthermore, cyclic photocleavable PLLA–PEO and PDLA–PEO block copolymers having *o*-nitrobenzyl units were synthesized. A mixture of the micellar dispersions of these block copolymers formed a gel upon UV irradiation via the 'topological conversion'.

INTRODUCTION

Polymers can be categorized as linear, branched, or cyclic, depending on the number of termini and branching points, and each exhibit different physical properties.[1, 2] For example, polyethylene, a typical general-use polymer, is used as high-density polyethylene, which has little branching in its structure, low-density polyethylene, which has a large amount of branching in its structure, linear low-density polyethylene having short-chain branching originating in the monomers. These polymers differ in physical properties such as density, crystallinity, and resistance to heat. Polymers are

used in applications based on their respective characteristics, and the ability to control the branching structures has greatly expanded applications.

Due to rapid technical advances in synthetic chemistry of polymers,[3] it is also now possible to control the complex branching structures of dendrimers and star polymers.[4, 5] There have thus been numerous studies involving the synthesis of polymers with many branching and terminal structures. However, by contrast, some polymers do not contain any branching or terminal structures, namely cyclic polymers. Cyclic polymers have been reported to differ from linear and branching polymers with regards to their physical properties such as viscosity, glass transition temperature, and hydrodynamic volume.[6, 7] In recent years, much success has been reported in further developing their functionality,[8] such as improved heat resistance in micelles,[9, 10] the control of micelle particle diameters,[11] and controlling domain intervals.[12] Increasing attention is being paid to such efforts.

In this study, we used polylactic acid (PLA) in an attempt to control the properties of the material. PLA can be synthesised using biomass such as corn as the raw material and is a carbon-neutral material[13] involving no increase or decrease in carbon dioxide concentration from the production to disposal. Polylactic acid has an additional benefit of being biodegradable; therefore, its use in research is very popular in these days of high demand for the development of environmentally friendly materials.[13] PLA occurs in two forms: poly(L-lactide) (PLLA) and poly(D-lactide) (PDLA), depending on its stereochemistry. It is known that when the two types interlock in a crystal, they form a strong stereocomplex; formation of a stereocomplex makes it possible to greatly improve the properties of PLA, which include weak heat-resistance and poor mechanical characteristics.[14, 15]

Due to the stereocomplex formation, PLLA-PEO-PLLA and PDLA-PEO-PDLA, which are block copolymers of poly(ethylene oxide) (PEO) and PLA, have the ability to gel.[16] These triblock copolymers form micelles in water; heat causes one end of the micelle-forming polymer to protrude out from the micelle and form a gel because of the formation of a three-dimensional structure which accompanies the stereocomplex formation (Scheme 1a). Arising from these characteristics and its biocompatibility, PLA-PEO-PLA shows promise for use in medical fields as a material in drug delivery systems.[17] Therefore, much research is being carried out with the objective of controlling the gelling properties of PLA-PEO-PLA.[17, 18] In some studies,

PLA-PEO-PLA with short hydrophilic segments was mixed together with PLA-PEO-PLA with long hydrophilic segments in order to finely control the gelation temperature.[17] Also, one example of controlling gel properties based on topological control is a PLA-PEO polymer with eight branches.[18] This research indicates that the temperature and density required for the gel formation of the eight-branched polymer is even lower than that required for the corresponding cyclic polymers.

Thus, it is clear that differences in topology and the ability to form a gel are related. On this basis, here we synthesised cyclic PLA-PEO and compared its ability to form a gel with the corresponding linear polymers. As noted above, the terminal structure plays an important role in the gel formation; therefore, we expected the cyclic polymers, which lack a terminal structure, to have less ability to gel (Scheme 1b). We also synthesised cyclic PLLA-PEO and cyclic PDLA-PEO, which have *o*-nitrobenzyl (NB) groups as photocleavable units, and devised a plan to use light to perform a 'topological conversion' to transform cyclic polymers,[19, 20] which are expected to have low ability to gel, to form linear polymers with the ability to gel (Scheme 1c).

MATERIALS AND METHODS

Materials

Unless otherwise noted, all commercial reagents were used as received. L-lactide (>99%, Musashino Chemical Laboratory, Ltd.) and D-lactide (>99%, Musashino Chemical Laboratory, Ltd.) were recrystallized from dry toluene twice prior to use. THF (>99.0%, Kanto Chemical Co., Inc.) was distilled over Na wire. Toluene (>99%, Godo Co., Inc.) was distilled over CaH_2. For column chromatography, Wakosil C-300 (Wako Pure Chemical Industries, Ltd.) was used. Synthesis was repeated several times for some polymers, resulting in different (although slight) number average molecular weight (M_n), peak molecular weight (M_p), and polydispersity index (PDI) values for such polymers.

Synthesis of HO-PLLA-PEO-PLLA-OH and HO-PDLA-PEO-PDLA-OH

L-lactide (1.5 g, 10.4 mmol) and PEG-4600 (2.0 g, 0.49 mmol) were vacuum dried, and tin(II) 2-ethylhexanoate ($Sn(Oct)_2$, 3.2 mg, 7.9 μmol) at a 32 mg/mL toluene solution was added. Toluene was removed by vacuum drying. The reaction mixture was heated at 130 °C for 12 h under nitrogen atmosphere. The reaction mixture was allowed to cool to ambient temperature and reprecipitated from CH_2Cl_2 into 1-propanol to yield **HO-PLLA-PEO-PLLA-OH** (2.3 g M_n(NMR) = 1100-4200-1100, M_p(SEC), = 8000,

PDI = 1.11) as white solid in 53% conversion. Likewise, **HO-PDLA-PEO-PDLA-OH** (2.1 g, M_n(NMR) = 700-4100-700, M_p(SEC) = 6800, PDI = 1.12) was synthesized using $_D$-lactide in 36% conversion.

^{1}H NMR: δ (ppm) 1.53–1.64 (m, –$CO_2CH(CH_3)$–), 3.61–3.67 (m, –CH_2CH_2O–), 4.21–4.40 (m, 6H, –$CH(CH_3)OH$, –$CO_2CH_2CH_2O$–), 5.09–5.27 (m, –$CO_2CH(CH_3)$–).

Synthesis of linear PLLA-PEO-PLLA and linear PDLA-PEO-PDLA

To a dry THF solution (200 mL) of a mixture of **HO-PLLA-PEO-PLLA-OH** (0.45 g, 70 μmol), 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (EDAC, 2.19 g, 11.4 mmol), and 4-dimethylaminopyridine (DMAP, 1.17 g, 9.58 mmol) was added 4-pentenoic acid (1.18 g, 11.7 mmol), and the resulting suspension was refluxed for 30 h under nitrogen atmosphere. The reaction mixture was evaporated to dryness under reduced pressure, and the residue was dissolved in CH_2Cl_2. The organic phase was washed with aqueous HCl (0.3 M) and saturated aqueous $NaHCO_3$, dried over anhydrous Na_2SO_4, and evaporated to dryness. The residue was reprecipitated from CH_2Cl_2 into 1-propanol to yield **linear PLLA-PEO-PLLA** (252 mg, M_n(NMR) = 1200-5300-1200, M_p(SEC) = 8800, PDI = 1.08) as brown solid in 46% yield. Likewise, **linear PDLA-PEO-PDLA** (380 mg, M_n(NMR) = 850-4300-850, M_p(SEC) = 8000, PDI = 1.09) was synthesized using **linear PDLA-PEO-PDLA** in 56% yield.

^{1}H NMR: δ (ppm) 1.55–1.62 (m, –$CO_2CH(CH_3)$–), 2.37–2.55 (8H, –$CH_2CH_2CO_2$–), 3.61–3.73 (m, –CH_2CH_2O–), 4.15–4.53 (m, 4H, –$CO_2CH_2CH_2O$–),4.96–5.06(d, 4H, CH_2=CH–), 5.07–5.31 (m, –$CO_2CH(CH_3)$–), 5.71–6.00(m, 2H, CH_2=CH–).

Synthesis of cyclic PLLA-PEO and cyclic PDLA-PEO

To a CH_2Cl_2 (600 mL) solution of **linear PLLA-PEO-PLLA** (120 mg, 15 μmol) was added the first-generation Grubbs catalyst (14.7 mg, 17.9 μmol) at 0, 24, and 48 h, and the resulting solution was refluxed for a total of 72 h. Ethyl vinyl ether (20 mL) was added, and the mixture was stirred at ambient temperature for 2.5 h. The reaction mixture was evaporated to dryness, and the residue was subjected to column chromatography on silica gel with CH_2Cl_2/MeOH (100/4 vol/vol) and subsequently with CH_2Cl_2/MeOH (10/1 vol/vol) to give crude **cyclic PLLA-PEO** (M_p(SEC) = 5100) as black solid containing the precursors. A portion of the crude was fractionated by preparative SEC to isolate **cyclic PLLA-PEO** (42 mg, M_n(NMR) = 1500-4200, M_p(SEC) = 5100, PDI = 1.09) in 46% yield. Likewise, **cyclic PDLA-PEO** (52 mg, M_n(NMR) = 1400-3900, M_p(SEC) = 5100, PDI = 1.09) was synthesized using **linear PDLA-PEO-PDLA** as brown solid in 33% yield.

^{1}H NMR: δ (ppm) 1.42–1.74 (m, –CO_2CH(CH_3)–), 2.28–2.58 (8H, =CHCH_2C$H_2$$CO_2$–), 3.53–3.84 (m, –C$H_2CH_2$O–), 4.16–4.43 (m, 4H, –$CO_2CH_2$$CH_2$O–), 5.10–5.24 (m, –$CO_2CH$($CH_3$)–), 5.37–5.53(m, 2H, –CH=CH–).

Synthesis of 4-((1-(4,5-dimethoxy-2-nitrophenyl)but-3-en-1-yl)oxy)-4-oxobutanoic acid

1-(4,5-Dimethoxy-2-nitro-phenyl)-but-3-ene-1-ol (4.31 g, 17.0 mmol) was dissolved in CH_2Cl_2 (100 mL). To this solution were added succinic acid anhydride (9.00 g, 89.9 mmol), triethylamine (9.0 mL, 64 mmol), and DMAP (215 mg, 1.76 mmol). The resulting solution was refluxed for 20 h. The reaction mixture was then washed with water and was concentrated under reduced pressure to afford 4-((1-(4,5-dimethoxy-2-nitrophenyl)but-3-en-1-yl)oxy)-4-oxobutanoic acid (3.66 g) in 61% yield.

^{1}H NMR: δ (ppm) 2.38–2.84(m, 6H, –CO_2CH_2C$H_2$$CO_2$H, CH_2=CH-CH_2–), 3.84–4.13(d, 6H, CH_3O-Ar), 5.00–5.24(t, 2H, CH_2=CH–), 5.76–5.96(m, 1H, CH_2=CH–), 6.45–6.53(q, 1H, Ar-CH($CO_2CH_2CH_2CO_2H$)–CH_2–CH=CH_2), 6.96–7.01(s, 1H, Ar–H meta to NO_2), 7.58–7.64(s, 1H, Ar–H ortho to NO_2)

Synthesis of photocleavable linear NB-PLLA-PEO-PLLA-NB and linear NB-PDLA-PEO-PDLA-NB

To a dry THF solution (375 mL) of a mixture of **HO-PLLA-PEO-PLLA-OH** (1.00 g, 151 μmol), EDAC (2.06 g, 10.7 mmol), and DMAP (1.31 g, 10.7 mmol) was added 4-((1-(4,5-dimethoxy-2-nitrophenyl)but-3-en-1-yl)oxy)-4-oxobutanoic acid (1.64 g, 4.65 mmol), and the resulting suspension was refluxed for 30 h under nitrogen atmosphere. The reaction mixture was evaporated to dryness under reduced pressure, and the residue was dissolved in CH_2Cl_2. The organic phase was washed with aqueous HCl (0.3 M) and saturated aqueous $NaHCO_3$, dried over anhydrous Na_2SO_4, and evaporated to dryness. The reaction mixture was fractionated by preparative SEC to isolation of **linear NB-PLLA-PEO-PLLA-NB** (651 mg, M_n(NMR) =900-3900-900, M_p(SEC) = 11000, PDI = 1.09) in 67% yield. Likewise, **linear NB-PDLA-PEO-PDLA-NB** (797 mg, M_n(NMR) = 1100-3800-1100, M_p(SEC) = 11000, PDI = 1.14) was synthesized using **HO-PDLA-PEO-PDLA-OH** as brown solid in 77% yield.

^{1}H NMR: δ (ppm) 1.55–1.65 (m, –CO_2CH(CH_3)–), 2.42–2.87 (12H, –CO_2CH_2C$H_2$$CO_2$–, CH_2=CH-CH_2-), 3.54–3.75 (m, –CH_2CH_2O–), 3.92–4.05(s, 12H, CH_3O-Ar) 4.18–4.42 (m, 4H, –CO_2C$H_2$$CH_2$O–), 5.10–5.25 (m, –$CO_2CH$($CH_3$)–,

C*H*$_2$=CH–), 5.73–5.97(m, 2H, CH$_2$=C*H*–), 6.42–6.57(m, 2H, Ar–C*H*(CHCH=CH$_2$)CO$_2$–) 6.97–7.06(d. 2H, Ar–*H* meta to NO$_2$) 7.56–7.69(d, 2H, Ar–*H* ortho to NO$_2$).

Synthesis of photocleavable cyclic NB-PLLA-PEO and cyclic NB-PDLA-PEO

To a toluene (1.0 L) solution of **linear NB-PLLA-PEO-PLLA-NB** (200 mg, 35 μmol) was added the second-generation Hoveyda-Grubbs catalyst (36.1 mg, 57.6 μmol) and refluxed for 50 h. Ethyl vinyl ether (20 mL) was added, and the mixture was stirred at ambient temperature for 15 h. The reaction mixture was evaporated to dryness, and the residue was subjected to column chromatography on silica gel with CH$_2$Cl$_2$/MeOH (100/2 vol/vol) and subsequently with CH$_2$Cl$_2$/MeOH (10/1 vol/vol) to give crude **cyclic NB-PLLA-PEO** (M_p(SEC) = 6100) as black solid containing the precursors. A portion of the crude was fractionated by preparative SEC to isolate **cyclic NB-PLLA-PEO** (120 mg, M_n(NMR) =1800-3700, M_p(SEC) = 7600) in 55% yield. Likewise, **cyclic NB-PDLA-PEO** (125 mg, M_n(NMR) = 2300-3600, M_p(SEC) = 8600, PDI = 1.11) was synthesized using **linear NB-PDLA-PEO-PDLA-NB** as brown solid in 56% yield.

^{1}H NMR: δ (ppm) 1.35–1.72 (m, –CO$_2$CH(C*H*$_3$)–), 2.37–2.94 (12H, –CO$_2$C*H*$_2$C*H*$_2$CO$_2$–, CH$_2$=CH-C*H*$_2$-), 3.49–3.83 (m, –C*H*$_2$C*H*$_2$O–), 3.90–4.04(s, 12H, C*H*$_3$O-Ar) 4.21–4.36 (m, 4H, –CO$_2$C*H*$_2$CH$_2$O–), 5.01–5.28 (m, –CO$_2$C*H*(CH$_3$)–), 5.53–5.70(m, 2H, –CH=C*H*–), 6.30–6.50(m, 2H, Ar–C*H*(CHCH=CH$_2$)CO$_2$–) 6.90–7.05(d. 2H, Ar–*H* meta to NO$_2$) 7.51–7.66(d, 2H, Ar–*H* ortho to NO$_2$).

Hydrogel Preparation

Micellar dispersions of **linear PLLA-PEO-PLLA**, **linear PDLA-PEO-PDLA**, **cyclic PLLA-PEO**, **cyclic PDLA-PEO**, **cyclic NB-PLLA-PEO**, and **cyclic NB-PDLA-PEO** were separately prepared. The copolymer was dissolved in THF. This solution was added dropwise into water in an ice bath. After sonication to the suspension, THF was evaporated from the suspension under reduced pressure in an ice bath to acquire an aqueous micellar dispersion. The polymer concentration was adjusted by the addition of water. Two of these dispersions were blended, and the mixture was heated or irradiated to UV light. The physical state of the mixture was reported at each temperature by turning the vial upside down. If the mixture flows, it was reported as a sol. If it did not flow for at least 10 s, it was reported as a gel.

RESULTS AND DISCUSSION

Synthesis of Cyclic PLLA-$(EO)_5$

We previously reported that the synthesis of cyclic PLA homopolymers,[21] and the synthetic methodology was here applied to prepare the cyclic amphiphilic block copolymers. Generally, mass spectrometry of block copolymers is difficult because there are multiple types of repeating units. In this study, we thus first used pentaethylene glycol as an initiator to synthesise cyclic PLLA-$(EO)_5$, and performed matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI TOF-MS) to verify the synthesis (Scheme 2).

Using pentaethylene glycol as an initiator in the presence of $Sn(Oct)_2$, L-lactide was heated to 130 °C with stirring and performed ring-opening polymerisation to obtain **HO-PLLA-$(EO)_5$-PLLA-OH**. The compound was analysed using ^{1}H NMR, SEC, and MALDI-TOF MS. In the ^{1}H NMR spectrum (Figure S1a), signal **a**, originating from methyl groups in the polylactide segments, was 1.54–1.66 ppm; signal **b**, originating from the ester-adjacent protons, was 5.10–5.23 ppm; signal **e,** originating from ester-adjacent methylene protons; and signal **f,** originating from methine groups of the terminal groups, was 4.20–4.41 ppm. We confirmed that a reaction proceeded, and determined M_n = 4700. In addition, a unimodal chromatogram was obtained by SEC (Figure S2a) and was calculated as M_p = 9000 and PDI = 1.17. We also confirmed by MALDI-TOF MS, that the molecular weight obtained was consistent with the calculated molecular weight. In particular, the calculated molecular weight for a 50-mer was 3864.45, while the measured value was 3863.90 (Figure S3a). A series of peak intervals was highly consistent with the molecular weight, 72, of $C_3H_4O_2$, a repeating unit in polylactic acid, and in particular the peak interval for 49-mers and 50-mers was 71.53.

Obtained **HO-PLLA-$(EO)_5$-PLLA-OH**, EDAC, DMAP, and 4-pentenoic acid were dissolved in THF, and refluxed and yielded **linear PLLA-$(EO)_5$-PLLA** (Scheme 2). The compound was analysed using ^{1}H NMR, SEC, and MALDI-TOF MS. In a ^{1}H NMR spectrum (Figure S1b), signal **f**, originating from the methylene adjacent to the terminal ester groups, and signal **g**, originating from methylene adjacent to the terminal alkene units, appeared at 2.35–2.54 ppm; signal **h**, originating from the terminal alkene units, appeared at 5.93 ppm; and signal **i**, originating from the terminal alkene units, appeared at 5.07–5.26 ppm, confirming that a condensation reaction proceeded. Furthermore, M_n was determined as 5200. Also, a unimodal chromatogram was obtained by SEC (Figure S2b), and M_p = 9700 and PDI = 1.07 were calculated for **linear PLLA-PEO-PLLA**. In

addition, using MALDI-TOF MS (Figure S3b), we confirmed that the peak molecular weight obtained was highly consistent with the calculated molecular weight. In particular, the calculated molecular weight for a 60-mer was 4749.28, while the measured molecular weight was 4749.93. A series of peak intervals was highly consistent with the molecular weight, 72, of $C_3H_4O_2$, a repeating unit in polylactic acid, and in particular the peak interval for 59-mers and 60-mers was 72.25.

The obtained **linear PLLA-(EO)$_5$-PLLA** and the first-generation Grubbs catalyst were dissolved in dichloromethane and refluxed in the diluted conditions, which caused an olefin metathesis reaction and yielded **cyclic PLLA-(EO)$_5$** (Scheme 2). The compound was analysed using ^{1}H NMR, SEC, and MALDI-TOF MS. In the ^{1}H NMR spectrum (Figure S1c), signal **h**, originating from the internal alkene units, shifted from 5.73–5.93 ppm to 5.36–5.56 ppm, and the terminal CH_2 units disappeared, confirming that an olefin metathesis reaction proceeded. Furthermore, M_n was calculated as 3900. Also, using SEC (Figure S2c), M_p was determined to be 6300 for **cyclic PLLA-(EO)$_5$**, which was then purified using preparative SEC (Figure S2d), and M_p = 6000 and PDI = 1.22 were determined for **cyclic PLLA-(EO)$_5$**. When the pre-fractionation molecular weight measured by SEC was compared to the pre-cyclisation molecular weight, a decrease from M_p = 9700 to M_p = 6300 was observed, confirming a reduction in the hydrodynamic volume brought about by cyclisation.

Moreover, using MALDI-TOF MS (Figure S3c), we confirmed that the peak molecular weight obtained was highly consistent with the calculated molecular weight. In particular, while the calculated molecular weight for 60-mers was 4721.23, the measured molecular weight was 4722.18. A series of peak intervals was highly consistent with the molecular weight, 72, of $C_3H_4O_2$, a repeating unit in polylactic acid, and in particular the peak interval for 59-mers and 60-mers was 71.98. Also, in the case of 60-mers, the molecular weight of 4277.18 was a reduction of 27.75 relative to that (4749.93) of the 60-mers prior to the cyclisation; therefore, it was also possible to confirm that a cyclisation reaction proceeded as this was highly consistent with the molecular weight of an ethylene molecule, 28.

The above results from ^{1}H NMR, SEC, and MALDI-TOF MS showed a significant difference between the states prior to and subsequent to the metathesis reaction, confirming the synthesis of **cyclic PLLA-(EO)$_5$**. On this basis, a method for synthesising **cyclic PLA-PEO** was also established.

Synthesis of Cyclic PLLA-PEO and Cyclic PDLA-PEO

Cyclic PLLA-PEO and **cyclic PDLA-PEO** were synthesised based on Scheme 3. Using PEO (M_n = 4600) as an initiator, $_L$-lactide was heated to 130 °C and stirred in the presence of $Sn(Oct)_2$, and **HO-PLLA-PEO-PLLA-OH** was obtained by ring-opening polymerisation (M_n(NMR) = 1100-4700-1100, M_p(SEC) = 8000, PDI = 1.11). The compound was analysed using ^{1}H NMR and SEC (Figures S4a and S6a). The same experimental procedures were applied to obtain **HO-PLLA-PEO-PLDA-OH** using $_D$-lactide (M_n = 700-4600-700, $M_{p\ SEC}$ = 6800, PDI = 1.12) (Figures S5a and S7a).

Obtained **HO-PLLA-PEO-PLLA-OH** or **HO-PDLA-PEO-PDLA-OH**, EDAC, DMAP, and 4-pentenoic acid were dissolved in THF. The hydroxy end groups were esterified by reflux, and **linear PLLA-PEO-PLLA** or **linear PDLA-PEO-PDLA**, respectively, was obtained. These polymers were analysed using ^{1}H NMR and SEC. In the ^{1}H NMR spectrum (Figure S4b), signal **f**, originating from the methylene groups adjacent to the terminal ester groups, and signal **g,** originating from methylene adjacent to the terminal alkene units, appeared at 2.37–2.55 ppm; signal **h**, originating from the terminal alkene units, appeared at 5.71–6.00 ppm; and signal **i**, originating from the alkene units, appeared at 4.96–5.06 ppm, confirming that the condensation reaction proceeded. Furthermore, M_n was calculated as 1200-5900-1200. **Linear PDLA-PEO-PDLA** (Figure S5b) was confirmed in the same manner, with a calculated M_n = 850-4800-850. In addition, unimodal chromatograms were obtained by SEC, and **linear PLLA-PEO-PLLA** was calculated as M_p = 8800, PDI = 1.08 (Figure S6b), while **linear PDLA-PEO-PDLA** was calculated as M_p = 8000, PDI = 1.09 (Figure S7b).

Obtained **linear PLLA-PEO-PLLA** or **linear PDLA-PEO-PDLA** and the first-generation Grubbs catalyst were dissolved in dichloromethane. In diluted conditions, they were refluxed, causing an intramolecular olefin metathesis reaction; the reaction yielded **cyclic PLLA-PEO** or **cyclic PDLA-PEO**, respectively. The compounds were analysed using ^{1}H NMR and SEC. In the ^{1}H NMR spectrum (Figure S4c), signal **h**, originating from the internal alkene unit, shifted from 5.71–6.00 ppm to 5.37–5.53 ppm, and the CH_2 group of the alkene units, which had been at 4.97–5.06 ppm, disappeared, confirming that an olefin metathesis reaction proceeded. Furthermore, M_n was determined as 4100-1500. **Cyclic PDLA-PEO** (Figure S5c) was confirmed in

the same manner, with M_n = 4400-1400. The reason for the moderate isolation yields (46% and 33%) was due to the formation of intermolecularly reacted products, which were removed by preparative GPC.

Furthermore, by SEC (Figures S6c and S7c), M_p = 5100 was determined for **cyclic PLLA-PEO** and M_p = 5100 was determined for **cyclic PDLA-PEO**; because we confirmed the formation of intermolecularly metathesized products, we performed purification with preparative SEC. As a result, a unimodal chromatogram was obtained by SEC (Figures S6d and S7d), and M_p = 5100, PDI = 1.09 for **cyclic PLLA-PEO** and M_p = 5100, PDI = 1.09 for **cyclic PDLA-PEO** were determined. Furthermore, as measured by SEC, a comparison of pre-fractionation molecular weight and pre-cyclisation molecular weight showed a decrease in **cyclic PLLA-PEO** from M_p = 8800 to M_p = 5100, and a decrease in **cyclic PDLA-PEO** from M_p = 8000 to M_p = 5100, which confirmed a reduction in the hydrodynamic volume brought about by cyclisation. The above results from ^{1}H NMR and SEC showed a significant difference between the states prior to and subsequent to the metathesis reaction, confirming the synthesis of **cyclic PLLA-PEO** and **cyclic PDLA-PEO**.

Gelation Behaviour of Non-photocleavable Cyclic PLLA-PEO and Cyclic PDLA-PEO

After fabricating **cyclic PLLA-PEO** and **cyclic PDLA-PEO**, we observed their gelation behaviour in the manner previously reported.[16] For comparison purposes, we performed the same gelation experiment for **linear PLLA-PEO-PLLA** and **linear PDLA-PEO-PDLA**. We also confirmed the formation of micelles using dynamic light scattering (DLS) measurements. Each of **linear PLLA-PEO-PLLA** (20 mg) and **linear PDLA-PEO-PDLA** (20 mg) was dissolved in THF, and 0.2 mL of water cooled by an ice bath was added. Whilst keeping the solution cool in an ice bath, after being exposed to ultrasound for a several tens of seconds, THF was evaporated under reduced pressure, giving a micellar dispersion of 10 wt%. We measured the micelle diameter by DLS. The diameters were 13.7 nm for **linear PLLA-PEO-PLLA** and 10.9 nm for **linear PDLA-PEO-PDLA** with monodispersity (Figures S13a and S13b, respectively). When each was mixed together and heated to 90 °C, gelation was observed as shown in Figure 1a. We also tested the gelation behaviour for **cyclic PLLA-PEO** and **cyclic PDLA-PEO**.[16] DLS indicated particle diameters of 13.5 nm for **cyclic PLLA-PEO** and 11.3 nm for **cyclic PDLA-PEO-PDLA** with monodispersity (Figures S13c and S13d,

respectively). However, as can be seen in Figure 1b, no gelation was observed. This suggests that a polymer network does not form for the micellar dispersion of the cyclic block copolymers.

Synthesis of Photocleavable Cyclic NB-PLLA-PEO and Cyclic NB-PDLA-PEO

As shown above, we experimentally ascertained that in comparison to **linear PLLA-PEO-PLLA** and **linear PDLA-PEO-PDLA**, it was difficult for **cyclic PLLA-PEO** and **cyclic PDLA-PEO** to form stereocomplexes in the micellar dispersion. Based on these experimental results, we fabricated cyclic PLLA-PEO and cyclic PDLA-PEO having photocleavable units. We investigated gelation based on the 'topological conversion' from cyclic polymers to linear polymers by photocleavage.

Based on the type of synthesis schemes illustrated below, we prepared **cyclic NB-PLLA-PEO** and **cyclic NB-PEO-PDLA**; here, we report on the synthesis methods. First, 1-(4,5-dimethoxy-2-nitro-phenyl)-but-3-ene-1-ol was synthesised on the basis of prior reports,[22] and 4-((1-(4,5-dimethoxy-2-nitrophenyl)but-3-en-1-yl)oxy)-4-oxobutanoic acid, a carboxylic acid with photocleavable unit, was synthesised in a condensation reaction with succinic anhydride. Through a condensation reaction of this carboxylic acid and **HO-PLLA-PEO-PLLA-OH** or **HO-PDLA-PEO-PDLA-OH**, alkene and photocleavable units were introduced at the termini, and **cyclic NB-PLLA-PEO** or **cyclic NB-PEO-PDLA**, respectively, was synthesised by metathesis under diluted conditions. 1-(4,5-dimethoxy-2-nitro-phenyl)-but-3-ene-1-ol, DMAP, and succinic anhydride were mixed with triethylamine and dichloromethane, and the mixture was refluxed to cause a condensation reaction between 1-(4,5-dimethoxy-2-nitro-phenyl)-but-3-ene-1-ol and succinic anhydride. The compound was analysed using ^{1}H NMR. In the ^{1}H NMR spectrum (Figure S8), all peaks were successfully attributed. In particular, signal **b**, originating from aromatic rings, had shifted after the reaction and appeared at 6.96–7.01 ppm, confirming the conversion of the groups into ester groups.

HO-PLLA-PEO-PLLA-OH or **HO-PDLA-PEO-PDLA-OH**, EDAC, 4-((1-(4,5-dimethoxy-2-nitrophenyl)but-3-en-1-yl)oxy)-4-oxobutanoic acid, and DMAP were dissolved in THF and refluxed, esterifying the hydroxy end groups to obtain **linear NB-PLLA-PEO-PLLA-NB** or **linear NB-PDLA-PEO-PDLA-NB**, respectively. The

compound was analysed using ^{1}H NMR and SEC. In the ^{1}H NMR spectrum (Figure S9a), signal **h**, originating from the methine groups adjacent to the photocleavable units, appeared at 6.42–6.57 ppm; signals **1** and **m**, originating from the methylene groups adjacent to the terminal alkene units and methylene groups adjacent to the terminal ester units, appeared at 3.45–3.75 ppm; signal **i**, originating from the terminal alkene units, appeared at 5.73–5.97 ppm; signals **f** and **g**, originating from aromatic rings, appeared respectively at 7.56–7.69 ppm and 6.97–7.06 ppm, respectively; and signal **k**, originating from methyl ether on aromatic rings, appeared at 3.92–4.05 ppm, confirming that a condensation reaction proceeded. Furthermore, M_n was determined as 900-4400-900. **Linear NB-PDLA-PEO-PDLA-NB** (Figure S10a) was confirmed in the same manner, with M_n = 1100-4300-1100. In addition, a unimodal chromatogram was obtained with the SEC measurement (Figures S11a and S12a), and **linear NB-PLLA-PEO-PLLA-NB** was calculated as M_p = 11000, PDI = 1.09, while **linear NB-PDLA-PEO-PDLA-NB** was calculated as M_p = 11000, PDI = 1.14.

The obtained **linear NB-PLLA-PEO-PLLA-NB** or **linear NB-PDLA-PEO-PDLA-NB** and the second-generation Hoveyda-Grubbs catalyst were dissolved in toluene and, under diluted conditions, stirred at 80 °C, causing an intramolecular olefin metathesis reaction yielding **cyclic NB-PLLA-PEO** and **cyclic NB-PDLA-PEO**. The compound was analysed using ^{1}H NMR and SEC. In the ^{1}H NMR spectrum (Figure S9b), signal **h**, originating from the olefins, shifted from 5.73–5.97 ppm to 5.53–5.70 ppm, confirming that the metathesis reaction proceeded. Furthermore, M_n was determined as 4200-1800. **Cyclic NB-PDLA-PEO** (Figure S10b) was confirmed in the same manner, with M_n = 4100-2300.

Furthermore, in SEC measurements (Figures S11b and S12b), M_p = 6100 was calculated for **cyclic NB-PLLA-PEO**, and M_p = 7800 was calculated for **cyclic NB-PDLA-PEO**. Because intermolecularly coupled products were formed, we performed purification with preparative SEC. As a result, a unimodal chromatogram was obtained by SEC (Figures S11c and S12c) and M_p = 7600, PDI = 1.20 for **cyclic NB-PLLA-PEO** and M_p = 8600, PDI = 1.11 for **cyclic NB-PDLA-PEO** were determined. When the pre-purification molecular weight measured by SEC was compared to the pre-cyclisation molecular weight, M_p was found to decrease from 11000 to 6100 for **cyclic NB-PLLA-PEO**, a reduction by 45%. Together with a decrease in **cyclic NB-PDLA-PEO**, from M_p = 11000 to M_p = 7800, a reduction by 30%, confirmed the synthesis. Analogous to **cyclic PLLA-PEO** and **cyclic PDLA-PEO**, the reason for the

moderate isolation yields (55% and 56%) was due to the formation of intermolecularly reacted products, which were removed by preparative GPC.

Gelation Behaviour of Photocleavable Cyclic NB-PLLA-PEO and Cyclic NB-PDLA-PEO

A mixture of the micellar dispersions of photocleavable **cyclic NB-PLLA-PEO** and **cyclic NB-PDLA-PEO** was tested for gelation. We examined gelation in the manner previously reported.[17] **Cyclic NB-PLLA-PEO** or **cyclic NB-PDLA-PEO** (10 mg) was dissolved in THF cooled by an ice bath, and 0.1 mL of water was added. After being exposed, in this cooled state, to ultrasound for several tens of seconds, the THF evaporated under reduced pressure in an ice bath, creating a micellar dispersion of 10 wt%. DLS measurement showed that the particle diameters were 13.4 nm for **cyclic NB-PLLA-PEO** and 13.0 nm for **cyclic NB-PDLA-PEO**, indicating that each formed micelles with a narrow size distribution (Figures S13e and S13f, respectively). After mixing these dispersions (Figure 2a), the mixture was UV radiated at 365 nm using an Asahi Spectra POT-365 light source (980 mW/cm^2 at φ = 6 mm) for 30 min while being cooled in an ice bath and gelled as shown in Figure 2b.

The polymer components of the gel were characterized using ^{1}H NMR, IR, and SEC after evaporating water under reduced pressure. The ^{1}H NMR spectrum (Figure S14a) confirmed that the peak (indicated by arrows in the figure) originating from the photocleavable units had decreased, and that UV irradiation caused photocleavage, as there were indications that topological conversion to linear polymers had occurred. The M_p value decreased, as determined by SEC (Figure S14b), to 6700 in comparison to pre-photocleaving measurements of **cyclic NB-PLLA-PEO** (M_p = 7600) and **cyclic NB-PDLA-PEO** (M_p = 8600). Essentially, the M_p value should have increased if the ideal topological conversion had occurred, but we considered that it may have decreased because of the removal of the two NB units. The IR measurement results are shown in Figure S14c. The peak originating in the carbonyl group of **cyclic NB-PLLA-PEO** was positioned at 1757.8 cm^{-1}, and the peak shifted to 1752.0 cm^{-1} upon the gel formation, suggesting that a stereocomplex formed.[23] From these points, we concluded that the polymers in this experiment, obtained by the topological conversion to a linear state, formed a stereocomplex and gelled.

In order to demonstrate that gelation stems from the stereocomplex formation, we

exposed a mixture of micellar dispersions based on **cyclic PLLA-PEO** and **cyclic PDLA-PEO** without photocleavable parts to UV radiation. There was no change in fluidity when the mixed micellar dispersion exposed to UV radiation under the same conditions (Figure 2c). Using NMR and SEC measurements, we ascertained that decomposition did not occur. Furthermore, a sodium carboxylate salt having photocleavable units was added to the mixed micellar dispersions of **cyclic PLLA-PEO** and **cyclic PDLA-PEO** without photocleavable units and exposed to UV light. However, no change in fluidity was observed (Figure 2d). Using NMR and SEC measurements, we confirmed that decomposition did not occur. This led us to conclude that the cleavage of the cyclic polymers had impacted gelation.

In addition, we attempted the same experiment using only **cyclic NB-PLLA-PEO**. In so doing, we were able to determine that the state of fluidity remained, and there was no gelation (Figure 2e). Because the peaks in the ^{1}H NMR spectrum of the photocleavable parts were decreasing (Figure S15a), we concluded that the photocleavage occurred due to UV radiation, suggesting the topological conversion towards linear polymers. A decrease in M_{p} (7300) was observed by SEC (Figure S15b). However, in the IR spectrum, as shown in Figure S15c, no peak shift was observed, and thus there was no stereocomplex formation. From the above, stereocomplexation stimulated the gel formation upon the topological conversion.

CONCLUSIONS

We conducted the synthesis of **cyclic PLLA-PEO** and **cyclic PDLA-PEO** and tested their gelation, and observed clear changes in the gelation behaviour, which indicated the decreased gelling ability of **cyclic PLA-PEO** in comparison to the linear polymers. By using **cyclic NB-PLLA-PEO** and **cyclic NB-PDLA-PEO** with photocleavable units, we also found the characteristic photoresponsive gelation behaviour based on topological conversion. Due to the potential for the use of PLA-PEO block copolymers as UV-responsive gelling agents, we anticipate their effective use in the medical field, particularly in drug delivery systems. Furthermore, we have been able to demonstrate the significance of the idea of ‘topological conversion’.

ACKNOWLEDGEMENTS

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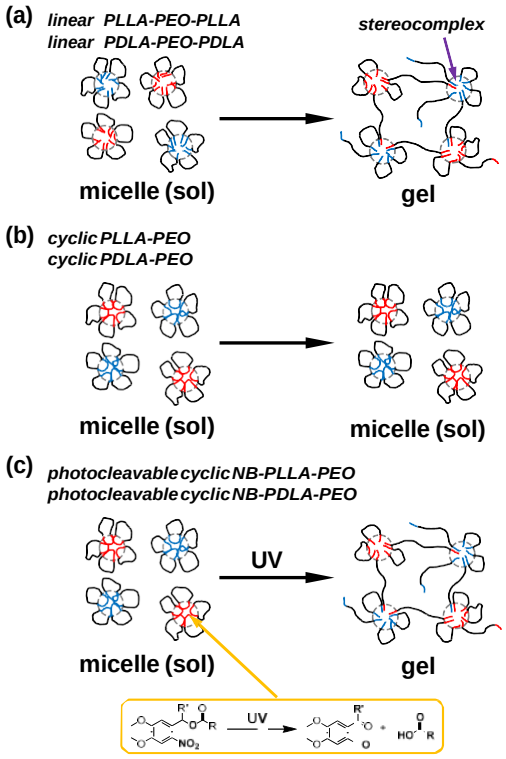
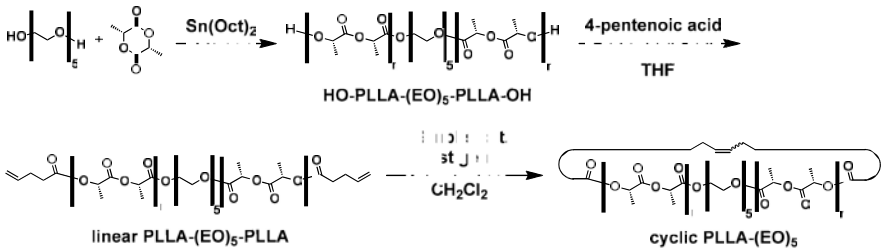
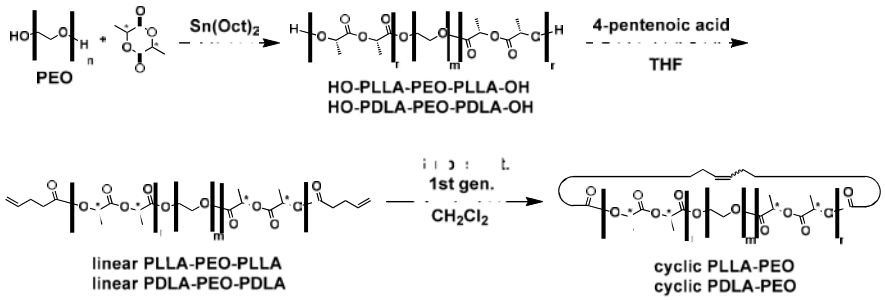


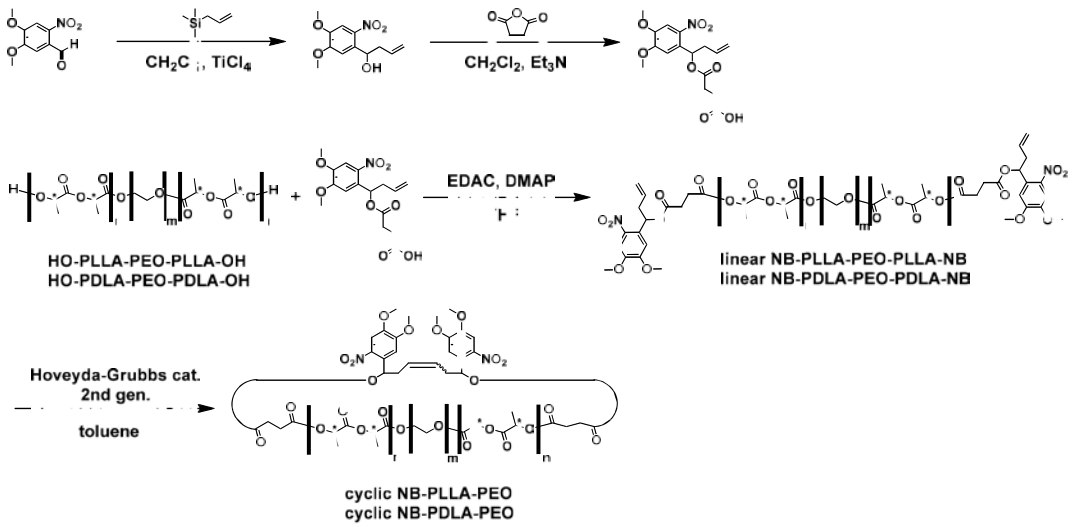
Scheme 1. Possible gelation of a mixture of micellar dispersions formed from (a) **linear PLLA–PEO–PLLA** and **linear PDLA–PEO–PDLA**, (b) **cyclic PLLA–PEO** and **cyclic PDLA–PEO**, and (c) photocleavable **cyclic NB-PLLA–PEO** and **cyclic NB-PDLA–PEO**.



Scheme 2. Synthesis of **cyclic PLLA-(EO)$_5$**.



Scheme 3. Synthesis of **cyclic PLLA-PEO** and **cyclic PDLA-PEO**.



Scheme 4. Synthesis of photocleavable **cyclic NB-PLLA-PEO** and **cyclic NB-PDLA-PEO**.

(a) (b)

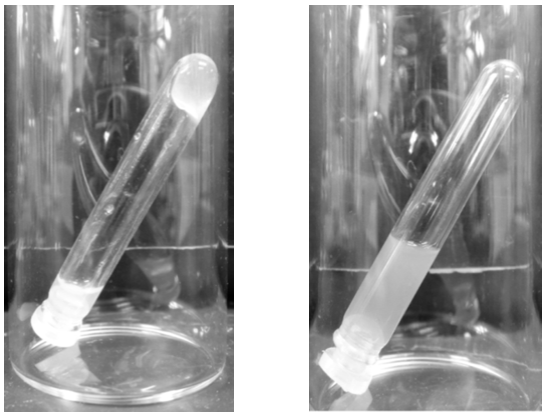


Figure 1. Photographs of mixtures of micellar dispersions of (a) **linear PLLA-PEO-PLLA** and **linear PDLA-PEO-PDLA** and (b) **cyclic PLLA-PEO** and **cyclic PDLA-PEO** after heating at 90 °C.

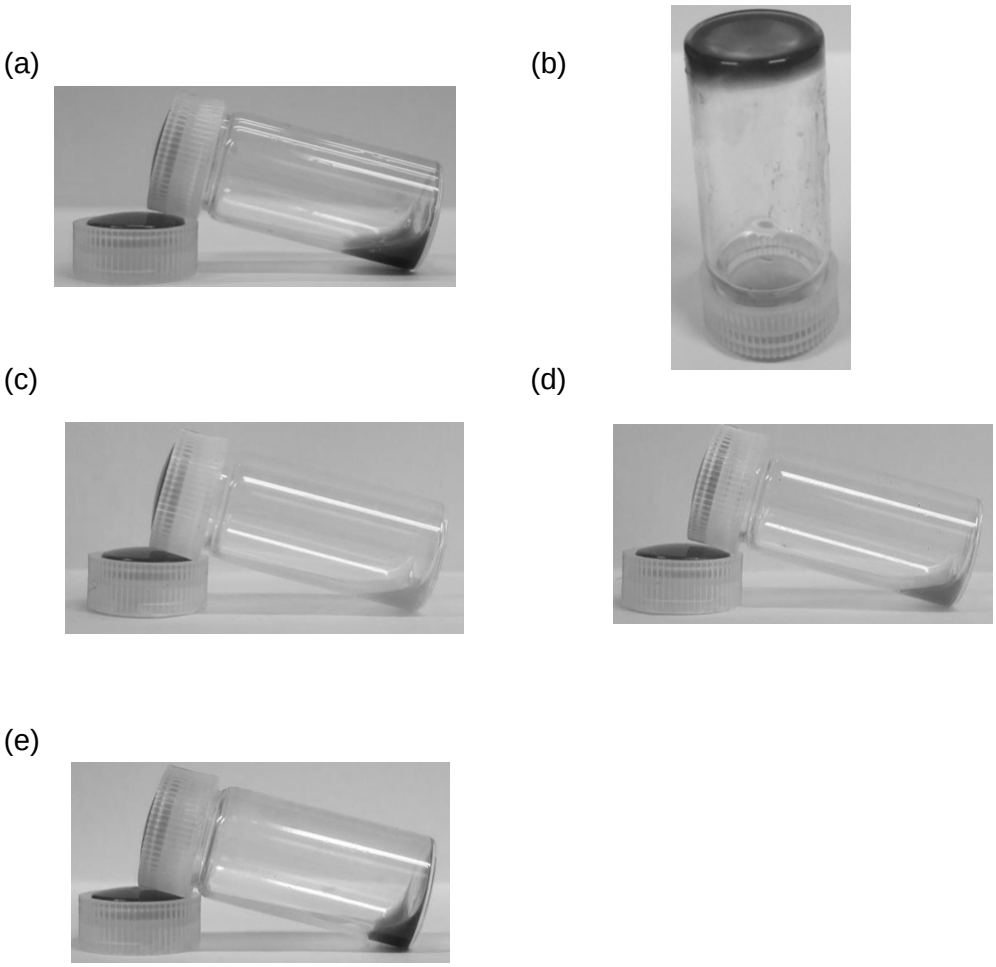


Figure 2. Photographs of mixtures of micellar dispersions of photocleavable **cyclic NB-PLLA-PEO** and **cyclic NB-PDLA-PEO** (a) before and (b) after UV irradiation, (c) non-photocleavable **cyclic PLLA-PEO** and **cyclic PDLA-PEO** after UV irradiation, (d) non-photocleavable **cyclic PLLA-PEO** and **cyclic PDLA-PEO** and a sodium carboxylate salt of a photocleavable unit after UV irradiation (e) photocleavable **cyclic NB-PLLA-PEO** after UV irradiation.