Synthesis of Novel Comb-Type Polylactide and Its Biodegradability

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Received January 16, 2001; Revised Manuscript Received May 10, 2001

ABSTRACT: The control of biodegradation and the improvement of moldability of poly-L-lactide, PLA, have been difficult because of the high crystallinity and the lability of melt viscosity. The synthesis of biodegradable PLA having the branched structure, comb-type PLA, was carried out by using only metabolizable materials to obtain a new material showing the controlled degradation and good molding properties, and to provide information on correlation of branched structure with biodegradability. A depsipeptide−lactide random copolymer having pendant hydroxy groups, poly[(Glc-Ser)-LA], was obtained by ring-opening copolymerization of L-lactide, LA, with a protected cyclodepsipeptide, cyclo[Glc-Ser(OBzl)], and consequent deprotection. The obtained copolymer was used as a macroinitiator for graft-polymerization of LA to give comb-type PLA. The obtained comb-type PLA showed a lower glass transition point, melting point, and apparent crystallinity than linear PLA. Furthermore, it was suggested that the degradation rate of comb-type PLA could be adjusted by controlling the molecular architecture such as molecular weight of main chain and/or side chain and number of graft chain.

Introduction

Biodegradable polymers have become of interest from the standpoints of pharmaceutical and environmental applications as well as biomedical applications. Various properties, such as degradation rate and its pattern, mechanical property, biocompatibility, and safety are desired for biodegradable medical materials. The plasticity and the reactivity are also required for the extension of their use. Among them, poly-L-lactide, PLA, is one of the most widely utilized class of biodegradable and bioabsorbable polymers in the field of biomedical materials and has been used clinically in wound closure,[1,2] tissue repair and regeneration,[3] and/or drug delivery.[4] PLA has good biocompatibility and biodegradability, high mechanical strength, and excellent shaping and molding properties. However, the application scope of PLA is limited, because of the difficulty of controlled degradation and the poor compatibility with soft tissue based on its high crystallinity and because of the induction of material defects based on lability of melt viscosity. To control the crystallinity and degradation rate, many approaches, for example chirality control, copolymerization with other lactones, and copolymerization with polyethers, have been tried.[5−17] In practice, to modify PLA with a view toward a decrease of the crystallinity, we also reported previously the synthesis of random copolymers of L-lactide (LA) with depsipeptides consisting of glycolic acid (Glc) and α-amino acids [lysine (Lys) and aspartic acid (Asp)] having pendant amino or carboxyl groups, poly[(Glc-Lys)-LA] and poly-[(Glc-Asp)-LA], through ring-opening copolymerization of cyclo[Glc-Lys(Z)] and cyclo[Glc-Asp(OBzl)] with LA and subsequent deprotection of benzyloxycarbonyl (Z) and benzyl (Bzl) groups, respectively.[18,19] Furthermore, we reported the synthesis of graft copolymers composed of PLA and polysaccharides such as pullulan or amylose through the trimethylsilyl protection method.[20,21] As a result, the biodegradability of polymers obtained could be controlled by introduction of hydrophilic units in PLA.

One of the promising approaches to overcome these problems is to introduce the branched structure into PLA. Recently, a number of ways have been reported to obtain the synthesis of branched polymers such as hyperbranched, star-shaped, and dendritic polymers because of interesting rheological and mechanical properties.[22−29] In general, it is well-known that branched compounds have different physicochemical properties compared with their linear counterparts owing to their molecular architecture. Long-chain branches predominantly affect the viscoelasticity of fluidity range, decrease the viscosity, and increase the elasticity. On the other hand, short-chain branches predominantly affect the crystallinity. By introducing the branched structure into linear PLA, that is, physical properties such as crystallinity, glass transition point (T_g), and melting point (T_m) will be influenced. The characteristics of the biodegradation by hydrolysis of these biodegradable polymers will in turn also be influenced. Thus, to get more suitable rheological and mechanical properties by varying the molecular architecture as well as to control the degradation rate by varying the crystallinity, we tried to provide PLA with a branched structure. It is expected that PLAs having desirable biodegradation and/or elasticity could be synthesized by controlling the molecular architecture, as the need arises.

In this paper, we report on the synthesis of a novel biodegradable comb-type PLA having a branched structure and its biodegradability. Biodegradable comb-type PLA was synthesized using only metabolizable materials by graft polymerization of LA onto depsipeptide−lactide copolymer containing serine residues as a macroinitiator. The obtained comb-type PLA can control the degradation rate by varying the molecular architecture and can be of interest for biodegradable and bioabsorbable biomedical materials.

Experimental Section

Materials. LA supplied by Wako Pure Chemical Co. (Tokyo) was recrystallized twice from ethyl acetate before using. DL-Poly(lactide) ($M_w = 10.6 \times 10^4$) was purchased from Sigma Chemical Co. (St. Louis, MO) and used without further

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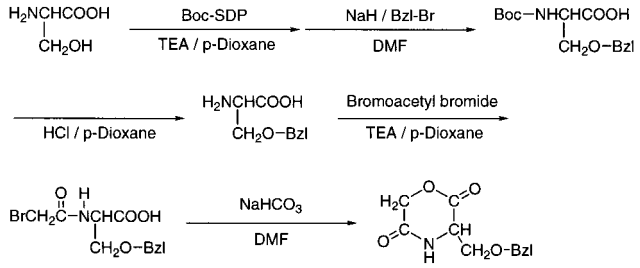
10.1021/ma010067m CCC: $20.00
Published on Web 06/28/2001

Scheme 1. Synthetic Route of Cyclo[Glc-Ser(OBzl)]



purification. L-Serine (Ser), tin 2-ethylhexanoate, and other chemicals were purchased from Wako Pure Chemical Co. Organic solvents were purified by usual distillation methods. Other reagents were used without purification.

Instrumentation. IR spectra were recorded with a Perkin-Elmer 1600 series FTIR. ^{1}H NMR and ^{13}C NMR spectra were recorded on a JEOL GSX-400 using tetramethylsilane (TMS) or 2,2-dimethyl-2-silapentane-5-sulfonate (DSS) as internal reference. Gel-permeation chromatography (GPC) analysis was carried out using a Tosoh GPC-8020 series system with a refractive index (RI) detector under the following conditions: TSK Gel G4000H_{XL} and G1000H_{XL} columns, and tetrahydrofuran (THF) eluent at the flow rate of 1 mL/min. The calibration curve for GPC analysis was obtained using polystyrene standards. The molecular weight of comb-type PLA was measured by size-exclusion chromatography (SEC) using a dual detector system, set in the direction of flow, consisting of a multiangle laser light scattering (MALLS) device and a differential refractometer in sequence. SEC-MALLS measurements were carried out at 40 °C in THF using a Shodex GPC KF-806L × 5 columns, at the polymer concentration of 3 mg/mL and the flow rate of 1 mL/min. The MALLS device was a DAWN model F (Wyatt Technology Co.) where the laser beam of wavelength of 632.8 nm was focused on the 67 μL flow cell. The progress of reaction was monitored by thin-layer chromatography (TLC) with a Merck Kieselgel 60 F_{254} silica gel plate. T_g and T_m of polymers were measured by a differential scanning calorimeter (DSC) (Rigaku TAS-200) in the range −50 to +200 °C at a heating rate of 10 °C/min. Samples were quenched with liquid nitrogen to −50 °C; then first heating and second heating were conducted. The aspect of films prepared from the polymers was observed by using a JEOL JSM-35 scanning electron microscope (SEM).

Synthesis of Cyclo[Glc-Ser(OBzl)]. The synthesis of a protected cyclodepsipeptide consisting of glycolic acid and *O*-benzyl-L-serine, cyclo[Glc-Ser(OBzl)], was performed by the reaction steps shown in Scheme 1. Ser(OBzl) was prepared by means of the methods of Nagasawa et al.[30] and Sugano et al.[31] First, 5.7 mL of triethylamine was added to a cooled suspension of Ser(OBzl) (5.3 g, 27.2 mmol) in 80 mL of *p*-dioxane. The resulting solution was vigorously stirred, and 10 mL of *p*-dioxane solution containing bromoacethyl bromide (3.5 mL, 40.8 mmol) was gradually added, while maintaining the reaction temperature at 5−10 °C by cooling with an ice bath. The mixture was extracted twice with 75 mL portions of diethyl ether. The combined organic layers were washed with water, dried with $MgSO_4$, and concentrated in vacuo. The resulting brown oily *N*-bromoacetyl-Ser(OBzl) was used without further purification in the next reaction step. Yield: 11.2 g, R_f = 0.79 (*n*-butanol:acetic acid:water = 4:1:1 v/v).

A solution of 11.2 g of *N*-bromoacetyl-Ser(OBzl) in 50 mL of *N*,*N*-dimethylformamide (DMF) was added dropwise to a mixture of 9.3 g of $NaHCO_3$ in 450 mL of DMF with vigorous stirring at 60 °C over a period of 5 h. The reaction mixture was stirred at 60 °C for 36 h after dropping of *N*-bromoacetyl-Ser(OBzl) to the mixture. After filtration and removal of the solvent in vacuo, the residue was partitioned between chloroform and water. The organic layer was separated and subsequently extracted with water, dried with $MgSO_4$, and concentrated in vacuo to yield a brown oil. The purification of cyclo[Glc-Ser(OBzl)] was carried out by column chromatography on silica gel using chloroform or chloroform/methanol as the eluent and recrystallization from chloroform. Total yield: 1.19 g (18.6%). R_f = 0.49 (ethyl acetate). Mp: 168−169 °C. IR (cm^{-1}, KBr disk): 3269(CONH), 1747, 1646(CO), 1064(−O−). ^{1}H NMR ($CDCl_3$): δ (ppm) = 3.85 (dd, 2H, CHCH_2OBzl), 4.32 (t, 1H, CH), 4.56 (dd, 2H, COCH_2O), 4.76 (dd, 2H, OCH_2Ph), 6.87 (s, 1H, NH), 7.26−7.38 (m, 5H, Ph). ^{13}C NMR ($CDCl_3$): δ (ppm) = 54.2 (CH*C*H$_2$OBzl), 67.7 (CHNH), 71.1 (CO*C*H$_2$O), 73.8 (O*C*H$_2$Ph), 127.8, 128.3, 128.7, 136.7(Ph), 163.8 (*C*OCH), 165.4 (NH*C*OCH$_2$). MS: m/z = 235.9 (M^+).

Synthesis of Poly{[Glc-Ser(OBzl)]-LA}. The synthesis of poly{[Glc-Ser(OBzl)]-LA} was performed according to the similar method of Morita et al.[6,32] reported recently. Cyclo-[Glc-Ser(OBzl)], LA and a freshly prepared 1.11 × 10^{-2} M solution of tin 2-ethylhexanoate in anhydrous THF (molar ratio of cyclo[Glc-Ser(OBzl)]/tin 2-ethylhexanoate = 1000) were added to a glass tube in a glovebox purged with dry argon. After the THF was removed under vacuum and the tube purged with dry argon, it was sealed in vacuo. The sealed tube was placed in an oil bath at 170 °C for 2 min and subsequently at 135 °C for 48 h. The reaction mixture was dissolved in a small amount of chloroform mixed with acetic anhydride and poured into a large amount of diethyl ether to precipitate poly-{[Glc-Ser(OBzl)]-LA} as a light yellow solid. IR (cm^{-1}, KBr disk): 3348 (CONH), 1758 (CO). ^{1}H NMR ($CDCl_3$): δ (ppm) = 1.58 (t, 3H, CH_3), 5.16 (q, 1H, C*H*CH$_3$), 3.65−4.10 (m, 2H, CHCH_2O), 4.40−4.60 (m, 1H, CH), 4.65 (s, 2H, OCH_2CO), 4.80−4.90 (m, 2H, OCH_2Ph), 6.88 (s, 1H, NH), 7.27−7.31 (m, 5H, Ph). ^{13}C NMR ($CDCl_3$): δ (ppm) = 16.6 (CH_3), 69.0 (*C*HCH$_3$), 169.6 (*C*OCHCH$_3$O), 52.1 (CHNH), 63.1 (CO*C*H$_2$O), 65.8 (CH*C*H$_2$O), 73.4 (O*C*H$_2$Ph), 127.7, 127.9, 128.4, 128.5 (Ph), 168.9 (*C*OCH), 169.4 (NH*C*OCH$_2$).

Synthesis of Poly[(Glc-Ser)-LA]. To achieve the deprotection perfectly, poly{[Glc-Ser(OBzl)]-LA} was treated with 1.0 M of TFMSA−thioanisole/TFA at 0 °C for 30 min. The reaction mixture was poured into a large amount of diehyl ether to precipitate poly[(Glc-Ser)-LA] as a white solid. IR (cm^{-1}, KBr disk): 3512 (OH), 3435 (CONH), 1758 (CO). ^{1}H NMR ($CDCl_3$): δ (ppm) = 1.58 (t, 3H, CH_3), 5.16 (q, 1H, C*H*CH$_3$), 3.80−4.30 (m, 2H, CHCH_2O), 4.60−4.90 (m, 1H, CH), 4.69 (s, 2H, OCH_2CO), 6.93 (s, 1H, NH). ^{13}C NMR ($CDCl_3$): δ (ppm) = 16.7 (CH_3), 69.0 (*C*HCH$_3$), 169.6 (*C*OCHCH$_3$O). Other small peaks assigned to the depsipeptide unit could not be detected.

Synthesis of Comb-Type PLA. Comb-type PLA was obtained by the following method. LA was graft-polymerized in bulk using tin 2-ethylhexanoate catalyst in the presence of poly[(Glc-Ser)-LA] having pendant hydroxy groups as a macroinitiator. The sealed tube prepared by the procedure described above was placed in an oil bath at the melting temperature of the macroinitiator for 2 min, and then the reaction was allowed to proceed for 4 h at 130 °C. Purification was carried out similar to that described above. IR (cm^{-1}, KBr disk): 3504 (OH), 3445 (CONH), 1758 (CO). ^{1}H NMR ($CDCl_3$): δ (ppm) = 1.59 (t, 3H, CH_3), 5.16 (q, 1H, C*H*CH$_3$), 4.30−4.50 (m, 2H, CHCH_2O), 4.60−4.70 (m, 1H, CH), 4.95−5.00 (m, 2H, OCH_2CO), 6.93 (s, 1H, NH). ^{13}C NMR ($CDCl_3$): δ (ppm) = 16.7 (CH_3), 69.0 (*C*HCH$_3$), 169.6 (*C*OCHCH$_3$O). Other small peaks assigned to the depsipeptide unit could not be detected.

Biodegradation Test. The polymers were casted from chloroform solution (4 wt %) and dried overnight to give colorless films having ca. 7 mm diameter × 0.1 mm thickness. Linear L-PLA (M_n = 5.3 × 10^4) obtained by a procedure similar to that described above without cyclo[Glc-Ser(OBzl)] was used as a reference polymer. After dried in vacuo, the films were incubated in $^1/_{15}$ M KH_2PO_4/$NaHPO_4$ buffer (pH = 7.0) at 37 °C for 28 days. After 1, 2, 3, 5, 7, 14, and 28 days, the films were washed with distilled water and dried in vacuo. The aspect of the films was observed by SEM and optical microscopy. The molecular weights of the polymers were measured by GPC. The degradation rates were estimated by the molecular weight reduction (%) calculated with the equation shown in the caption of Figure 8.

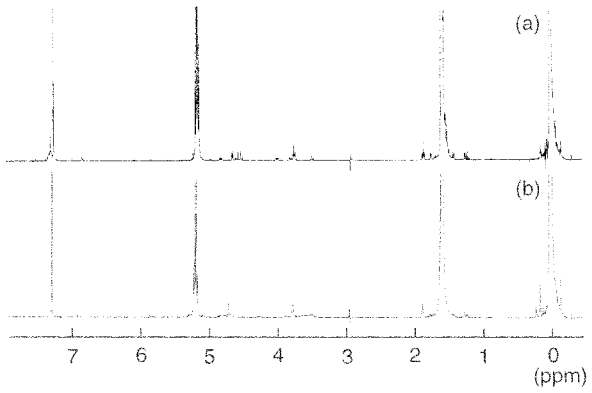


Figure 1. ^{1}H NMR spectra of (a) poly{[Glc-Ser(OBzl)]-LA} and (b) poly[(Glc-Ser)-LA].

Table 1. Results of Copolymerization of LA with Cyclo[Glc-Ser(OBzl)][a]

Y^b (mol %)	y^c (mol %)	yield (wt %)	$M_n{}^d$ ($\times 10^4$)	$M_w/M_n{}^d$
5.0	4.2	90	1.88	2.01
10.0	9.5	91	2.49	1.85
15.0	13.5	76	2.07	1.60

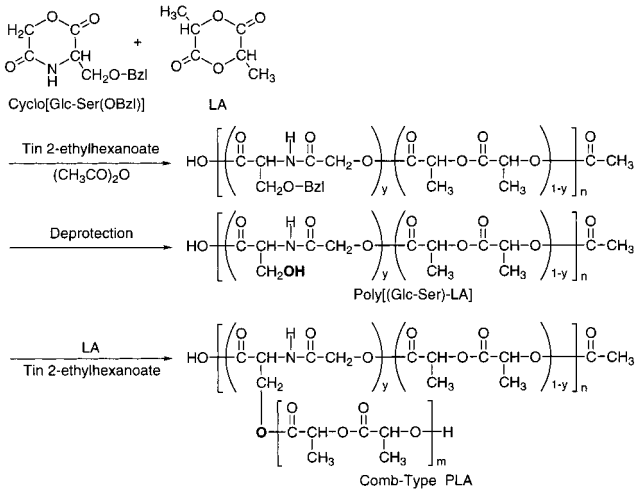
[a] Polymerization was carried out in bulk at 135 °C for 2 days. Polymerization flask was placed in an oil bath at 170 °C for 2 min; subsequently, the temperature was lowered to 135 °C. Ratio of monomer to initiator (M/I) = 1000. I = Tin 2-ethylhexanoate. [b] *Y*: mole fraction of cyclo[Glc-Ser(OBzl)] in feed. [c] *y*: mole fraction of [Glc-Ser(OBzl)] unit in copolymer. [d] Estimated by GPC.

Results and Discussion

The synthesis of protected cyclodepsipeptide, which consisted of Glc and Ser(OBzl), was performed through the reaction steps shown in Scheme 1. To avoid side reactions, the hydroxy group of Ser unit had to be protected by benzyl group. The benzyl group was stable under the condition used in the synthesis of protected cyclodepsipeptide and in the ring-opening copolymerization.

The ring-opening copolymerization of LA with cyclo[(Glc-Ser(OBzl)] using tin 2-ethylhexanoate was carried out under low cyclo[(Glc-Ser(OBzl)] feed conditions at 135 °C to give poly{[Glc-Ser(OBzl)]-LA}. The content of the [Glc-Ser(OBzl)] unit in the copolymer was calculated from the ratio of the integration value of the 1.58 ppm ($-CH_3$) peak of LA to that of the 3.65−4.90 ppm (CH, CH_2) peaks of Glc-Ser(OBzl) in ^{1}H NMR (Figure 1a). The results of composition and molecular weight of the obtained poly{[Glc-Ser(OBzl)]-LA} are summarized in Table 1. By variation of the mole fraction of cyclo[(Glc-Ser(OBzl)] in feed, the mole fraction of [Glc-Ser(OBzl)] unit in copolymer could be controlled. To obtain poly[(Glc-Ser)-LA], the deprotection of benzyl group in poly{[Glc-Ser(OBzl)]-LA} was carried out by the TFMSA-thioanisole/TFA method, as shown in Scheme 2. The results of IR and NMR spectra indicated the achievement of complete elimination of the benzyl groups from the obtained poly{[Glc-Ser(OBzl)]-LA}. The GPC data of the obtained poly[(Glc-Ser)-LA] suggested that main-chain cleave in this copolymer occurred slightly under our deprotection reaction conditions. However, benzyl groups could be completely deprotected as shown in Figure 1b. Thus, hydroxy groups were found to be easily introduced into PLA by using the technique of ring-opening copolymerization of LA with a small amount of cyclo[(Glc-Ser(OBzl)].

Scheme 2. Synthetic Route of Comb-Type PLA



The bulk graft polymerization was carried out in the presence of poly[(Glc-Ser)-LA] as a macroinitiator using tin 2-ethylhexanoate catalyst at 130 °C to give comb-type PLA as shown in Scheme 2. In this research, macroinitiator-1, -2, and -3 (macroinitiator-1: $M_n = 1.11 \times 10^4$, $y = 4.2$ mol %; macroinitiator-2: $M_n = 1.20 \times 10^4$, $y = 6.4$ mol %; macroinitiator-3: $M_n = 1.98 \times 10^4$, $y = 9.2$ mol %) were used. Comb-type PLAs obtained from macroinitiator-1, -2, and -3 are abbreviated comb-PLA-1, comb-PLA-2, and comb-PLA-3, respectively. The results of graft polymerization are shown in Table 2. The graft polymerization was performed under rigorously anhydrous conditions. Poly[(Glc-Ser)-LA] used as a macroinitiator was carefully dried to avoid an initiation by water. The GPC profiles of graft polymer, comb-type PLA, were symmetrical and unimodal as shown in Figure 2. Nonreacted macroinitiator was not found in the profile for comb-type PLA. This means grafting was successful. However, GPC analysis is not the appropriate method for the determination of molecular weight, since it always underestimates the molecular weight of the branched polymer due to smaller hydrodynamic volume in solution, compared with that of linear polystyrene as a reference material. Therefore, comb-type PLA was analyzed by a combination of SEC and MALLS to characterize its effective molecular weight and hydrodynamic volume in THF solution. Plots of real molecular weight vs elution

Table 2. Results of Graft Polymerization of LA Using Poly[Glc-Ser-LA] as Macroinitiator[a]

macroinitiator[b]			comb-type PLA					
y^c (mol %)	$M_n{}^{GPC}$ ($\times 10^4$)	$M_w/M_n{}^{GPC}$	yield (wt %)	y^c (mol %)	$M_n{}^{GPC}$ ($\times 10^4$)	$M_w/M_n{}^{GPC}$	$M_n{}^{MALLS}$ ($\times 10^4$)	$M_w/M_n{}^{MALLS}$
4.2	1.11	2.66	86	1.6	4.30	2.80	5.06	2.83
6.4	1.20	2.07	78	2.6	2.49	1.93	3.91	1.97
9.2	1.98	1.77	78	1.4	4.58	3.05	5.95	3.10

[a] Polymerization was carried out in bulk at the melting temperature of the macroinitiator for 2 min and then at 130 °C for 4 h. Molar ratio of monomer to initiator (M/I) = 100 or 30. I: Tin 2-ethylhexanoate. [b] Poly[(Glc-Ser)-LA] was used as a macroinitiator. [c] *y*: mole fraction of (Glc-Ser) unit in copolymer. GPC: estimated by GPC. MALLS: determined by SEC-MALLS.

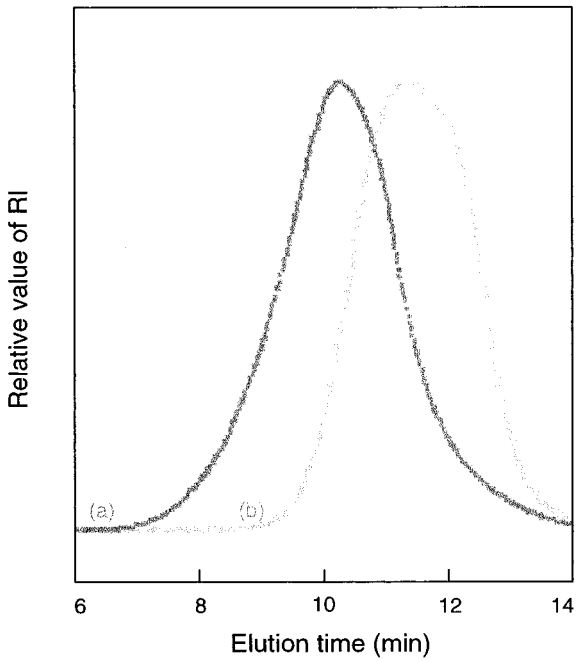


Figure 2. GPC elution profiles of (a) comb-PLA-3 and (b) macroinitiator-3.

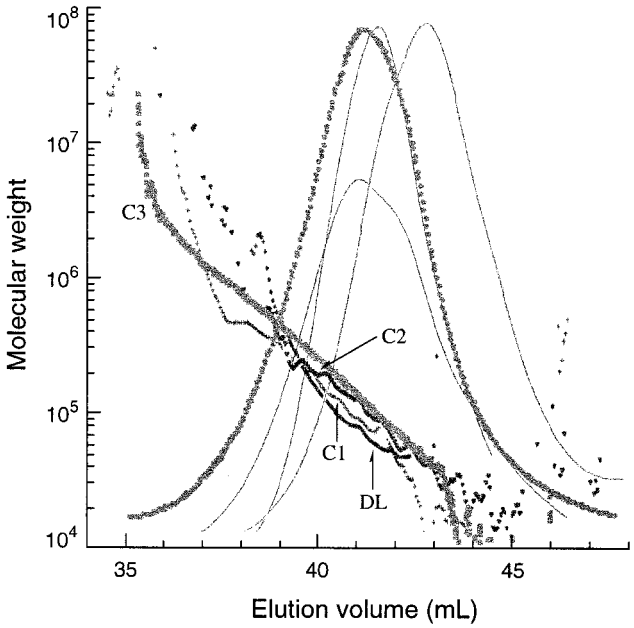


Figure 3. Comparison of comb-type PLA and linear DL-PLA by RI monitored SEC curves and correlation of molecular weight vs elution volume: (C1) comb-PLA-1; (C2) comb-PLA-2; (C3) comb-PLA-3; (DL) linear DL-PLA.

volume obtained from SEC-MALLS analysis suggested the comb-type PLA, in order of comb-PLA-1, -2, and -3, showed delayed elution time compared with linear DL-PLA having the same real molecular weight (Figure 3) or used macroinitiator (data not shown). That is, the hydrodynamic volume of comb-type PLA, especially comb-PLA-3, in THF solution was obviously small compared with that of linear DL-PLA due to introduction of the branched structure. In practice, the molecular weight obtained by SEC-MALLS was higher than that obtained by GPC in the case of comb-type PLA. On the other hand, no difference of the molecular weight between SEC-MALLS and GPC was confirmed in the case of linear DL-PLA or macroinitiator. Since linear L-PLA was not dissolved in THF owing to its high crystallinity, that of comb-type PLA could not be directly compared with linear L-PLA having the same real molecular weight. However, it is not clear that it is appropriate to claim that the solubility differences between linear PLA and the branched copolymer are based upon crystallinity and not the actual makeup of the PLA and the copolymer or the differences one might expect for the solubility of a linear vs graft copolymer. Although the obtained comb-type PLA has from ca. three to ca. 12 branch points, plots of molecular weight vs hydrodynamic volume for comb-type PLA were observed to be obviously different compared with those for linear DL-PLA. Figure 4 shows schematically the average molecular architecture of the obtained comb-type PLA based on the results of SEC-MALLS, GPC, and ^{1}H NMR measurements. It is assumed that comb-PLA-1 is composed of 11 100 Da of main chain and about three graft chains having 13 000 Da, comb-PLA-2 is composed of 12 000 Da of main chain and about five graft chains having 5500 Da, and comb-PLA-3 is composed of 19 800 Da of main chain and about 12 graft chains having 3300 Da. If the molecular weight is same, shrinkage factor (g_s) is defined by the expression

$$g_s = \langle S^2 \rangle_{comb} / \langle S^2 \rangle_{linear}$$

where $\langle S^2 \rangle_{comb}$ and $\langle S^2 \rangle_{linear}$ are the square of radius of gyration of comb-type polymer and linear polymer, respectively. Here, assuming that both main chain and graft chains have no polydispersity about comb-type polymer, g_s is given by the following equation:[33]

$$g_s = \alpha + 2\alpha(1-\alpha)/p + (3p-2)(1-\alpha)^3/p^2$$

p and α is the number of graft chain and the weight fraction of main chain in comb-type polymer, respectively. The g_s results are shown in Table 3. These results correlate with the results of elution order obtained by SEC-MALLS, and suggested that comb-type PLA having the molecular architecture as shown in Figure 4 could be synthesized.

Table 3. Shrinkage Factor of Comb-Type PLA

	p^a	α^b	$g_s{}^c$
comb-PLA-1	3	0.22	0.70
comb-PLA-2	5	0.31	0.57
comb-PLA-3	12	0.33	0.44

[a] p: number of graft-chains. [b] α: weight fraction of main-chain in comb-type PLA. [c] g_s: shrinkage factor.

Table 4 summarizes the results of DSC analysis of comb-type PLA, poly[(Glc-Ser)-LA] of low content of (Glc-Ser) unit, and linear PLAs. T_g and T_m values of poly[(Glc-Ser)-LA], which is a random copolymer, decreased with increasing content of the hydrophilic (Glc-Ser) unit. The presence of the serine residues at relatively low percentage disrupted the crystalline region slightly, causing the decrease of T_m and the appearance of two melting endotherms as shown in Table 4. This quite small additional peak appeared at ca. 10 °C lower temperature than the dominant peak. When the content of the (Glc-Ser) unit in random copolymer exceeded 15.6 mol %, the random copolymer became amorphous. On the other hand, those of comb-type PLA decreased with introducing the branched structure despite increasing the average molecular weight. The apparent crystallinity (X_c) of the polymers

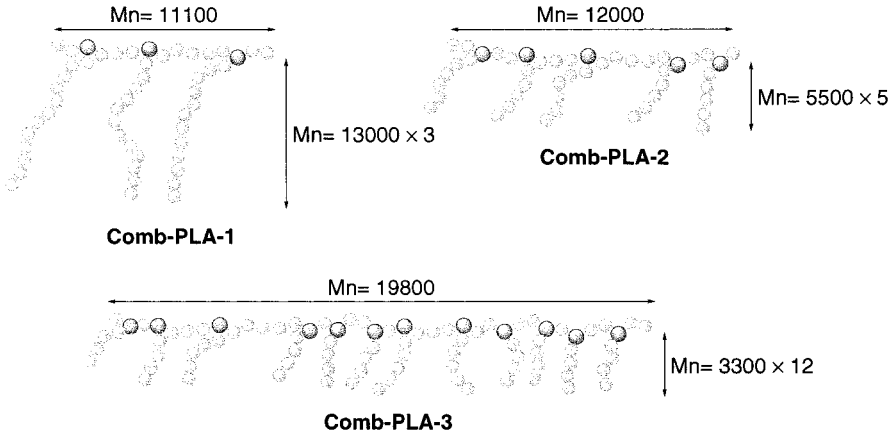


Figure 4. Schematic molecular architecture of comb-type PLA.

Table 4. Thermal Properties of Comb-Type PLA, Macroinitiator, and Linear PLAs

(co)polymer	M_n[a] ($\times 10^4$)	M_w/M_n[a]	y[b] (mol %)	T_g[c] (°C)	T_m[c] (°C)	ΔH[c] (J/g)	X_c[d] (%)
comb-PLA-1	5.06[f]	2.83[f]	1.6	40.4	135.1, 146.0	−20.5	21.9
comb-PLA-2	3.91[f]	1.97[f]	2.6	44.0	135.8, 144.3	−18.8	20.1
comb-PLA-3	5.95[f]	3.10[f]	1.4	42.9	140.5, 150.9	−13.7	14.6
macroinitiator-1[e]	1.11	2.66	4.2	59.7	141.6, 151.6	−47.3	50.5
macroinitiator-2[e]	1.20	2.07	6.4	58.9	138.9, 150.7	−24.8	26.5
macroinitiator-3[e]	1.98	1.77	9.2	42.5	126.8, 137.9	−21.4	22.8
linear L-PLA	1.80	1.81	0	62.1	166.6	−51.5	55.0
	5.30	2.37	0	63.8	173.5	−51.9	55.4
linear DL-PLA	4.76	2.11	0	51.4	–	0	0

[a] Estimated by GPC. [b] y: mole fraction of (Glc-Ser) unit in copolymer. [c] Determined by DSC. [d] X_c: crystallinity estimated from ΔH values. [e] Indicates poly[(Glc-Ser)-LA]. [f] Determined by SEC-MALLS.

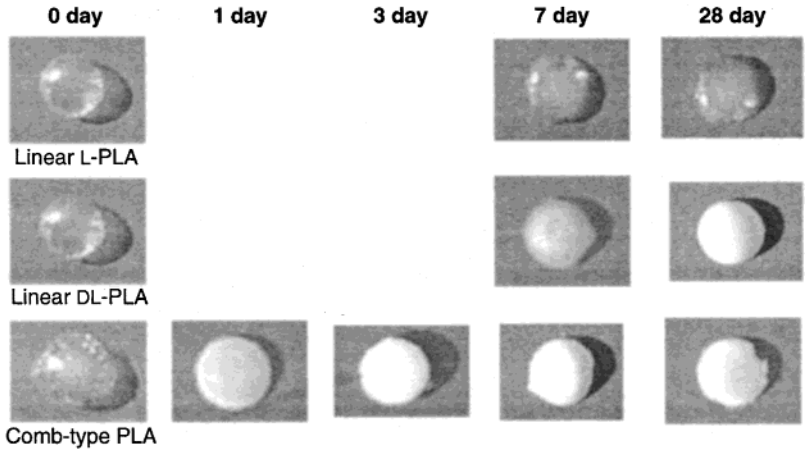


Figure 5. Optical microscope photographs of films during biodegradation test.

was calculated with the aid of the enthalpy of fusion of −93.7 J/g[34] for the perfectly crystalline PLA by the equation X_c (%) = $\Delta H/\Delta H_{theo} \times 100$. The X_c results shown in Table 4 suggested that the crystallinity of PLA could be easily controlled by the introduction of very low content of depsipeptide units with the OH group and/or branched structure.

To evaluate in vitro biodegradability of the comb-type PLA, its biodegradation behavior was investigated in $^1/_{15}$ M $KH_2PO_4/NaHPO_4$ buffer (pH = 7.0) at 37 °C and compared with that of linear PLAs. The optical microscope photographs of films obtained from the polymers before and after the biodegradation test are shown in Figure 5. All of films were transparent immediately after preparation. The films of comb-type PLA and linear DL-PLA became white on the first and seventh days with proceeding degradation, respectively. However, the film of linear L-PLA kept its transparency for 28 days. Next, to examine the surface state of films, SEM observation was carried out. Figure 6 shows the SEM photographs of films prepared from comb-type PLA before and after the biodegradation test. Before exposure to phosphate buffer solution, the film surface is smooth. In contrast, the transparent film became opaque and was eroded after exposure to phosphate buffer solution, which would be due to the degradation caused by hydrolysis. The change in the GPC elution profiles of the polymers as a function of time is given in Figure 7. The GPC elution profiles of the polymers remained unimodal over time. These results suggested that no entrapment of degradation products such as oligomers occurred, and the degradation homogeneously proceeded because films were relatively small size devices.[35] All comb-type PLA gave results of the same tendency. Figure 8 shows the time course of molecular weight change of the polymers during the biodegradation test. In vitro degradation rate of comb-type PLA was significantly faster than that of linear L-PLA and

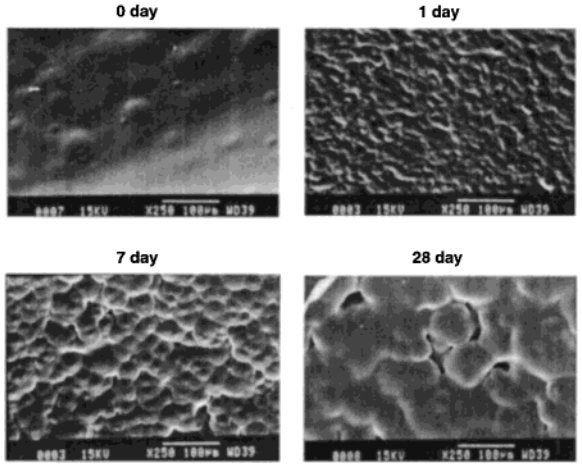


Figure 6. SEM photographs of the surface of comb-type PLA films during biodegradation test.

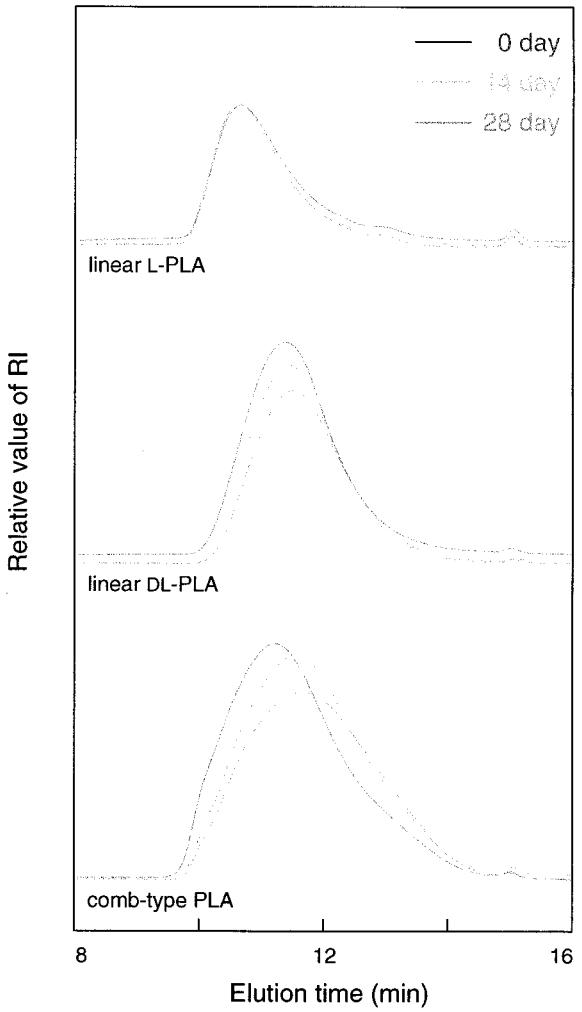


Figure 7. GPC elution profiles of original and degraded polymers.

amorphous linear DL-PLA. In the comparison among three comb-type PLAs, although their apparent crystallinities were almost the same, the degradation rate of comb-PLA-3 was faster than that of comb-PLA-1 and -2. This is probably because comb-PLA-3 has a larger number of end groups per molecule. In general, it has been reported that the degradation of PLA takes place predominantly in the amorphous region at the initial stage. Therefore, it is expected that the degradation rate

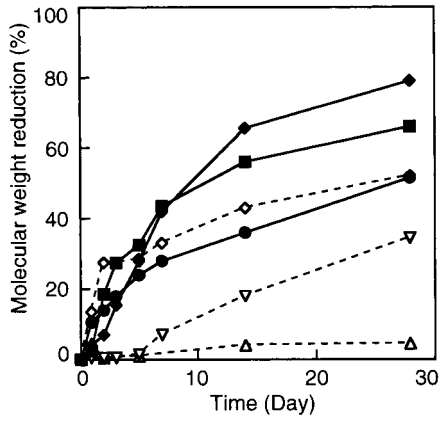


Figure 8. Degradation behavior of comb-type PLA and linear PLAs in $^1/_{15}$ M KH_2PO_4−Na_2HPO_4 (pH = 7.0) at 37 °C in vitro: (●) comb-PLA-1; (■) comb-PLA-2; (◆) comb-PLA-3; (◇) macroinitiator-3; (▽) linear DL-PLA; (△) linear L-PLA. Molecular weight reduction (%) = $100\times(M_0 - M_t)/M_0$, where M_0 = initial molecular weight and M_t = molecular weight after degradation.

of the polymer having lower crystallinity be faster than that of the polymer having higher crystallinity. In practice, the degradation rate of comb-type PLA was faster than that of the used macroinitiator and linear L-PLA, which have higher crystallinity than comb-type PLA. If the degradation rate is determined by only the crystallinity, the degradation rate of amorphous linear DL-PLA should be the fastest in all used PLA samples, and those of three comb-type PLAs should be almost same. However, the obtained results were not so. This difference of degradation rate is attributable to increase of the number of end groups based on the introduction of branched structure. That is, the increase of the number of end groups per molecule causes the increase of degradation rate. Thus, these results suggest that the degradation rate of PLA can be controlled by not only the crystallinity but also the molecular architecture, such as the branched structure.

Conclusions

Novel biodegradable comb-type PLA was successfully synthesized by graft-polymerization of LA using a depsipeptide−lactide random copolymer having pendant hydroxy groups as a macroinitiator. The obtained comb-type PLA showed a decrease of crystallinity and an increase of biodegradability compared with linear PLA. The degradation rate of PLA could be controlled by varying the molecular architecture.

By varying the molecular weight of main chain and/or graft chain and the number of graft chains, we can obtain various comb-type PLAs having different molecular architecture. We expected to control the solution, thermal, and morphology properties, by controlling the molecular architecture. So, such a comb-type PLA can be expected to be applied as novel biomedical materials. We plan to report on the correlation of molecular architecture with physical properties such as mechanical properties in a subsequent paper.

Acknowledgment. The authors are very grateful to Professor A. Matsumoto of Kansai University for his help with the SEC-MALLS measurements. We would like to thank Wako Pure Chemical Co. for an adequate supply of LA.

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MA010067M