Enzymatic Synthesis of Aromatic Polyesters by Lipase-Catalyzed Polymerization of Dicarboxylic Acid Divinyl Esters and Glycols

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ABSTRACT: Polyesters containing aromatic moiety in the main chain have been synthesized by enzymatic polymerization of dicarboxylic acid divinyl esters and glycols under mild reaction conditions. Divinyl esters of isophthalic acid, terephthalic acid, and *p*-phenylene diacetic acid, were used as new monomer for the enzymatic polymerization. Effects of the polymerization conditions were systematically investigated in the polymerization of divinyl isophthalate and 1,6-hexanediol. *Candida antarctica* lipase afforded the polymer of the highest molecular weight. Methylene chain length of the glycol affected the polymer yield and molecular weight. Divinyl terephthalate was enzymatically polymerized under the similar reaction conditions, yielding the polymer of lower molecular weight. Enzymatic polymerization of divinyl sebacate and *p*-xylylene glycol also afforded the aromatic polyester.

KEY WORDS Enzymatic Polymerization / Lipase / Aromatic Polyester / Divinyl Ester / Polycondensation /

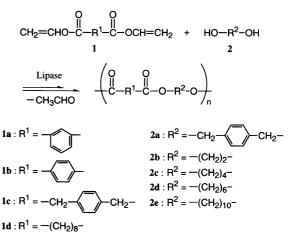
Aromatic polyesters, typically poly(ethylene terephthalate) (PET) and poly(butylene terephthalate) (PBT), are widely used in various fields since they have excellent properties, *e.g.*, high thermal and chemical stabilities.^{1,2} These polymers are industrially produced at high temperature under reduced pressure. Since PET and PBT are not environmentally benign due to their low biodegradability, aliphatic and aromatic ester copolymers have been developed as new biodegradable engineering plastics. However, their synthesis also involved severe reaction conditions (high temperature and/or reduced pressure).

Recently, hydrolase-catalyzed polymerizations have received much attention as environmentally friendly methodology of polyester syntheses owing to non-toxic enzyme catalyst and mild reaction conditions.³⁻⁷ In some cases, lipase showed unique catalytic behaviors for the polyester syntheses in comparison with those of conventional chemical catalysts. Lipase induced the ring-opening polymerization of small- and medium-size lactones as well as macrolides (12-, 13-, 16-, and 17-membered lactones).⁸⁻¹⁷ Using traditional chemical catalysts, the macrolides showed much lower reactivity and polymerizability than ε -caprolactone (7-membered lactone, ε -CL) due to their lower ring strain.^{18,19} However, they were enzymatically polymerized much faster than *ɛ*-CL, probably due to the stronger recognition of the macrolide by lipase.¹³⁻¹⁷

In lipase-catalyzed acylation for synthesis of functional organic products, vinyl esters have been often used as acylating agents because the irreversible tautomerization of the leaving vinyl alcohol group to acetaldehyde afforded the desired product in high yields.^{20,21} We reported that divinyl adipate was superior to the corresponding diacid or dialkyl ester for the enzymatic synthesis of polyester.²² Afterwards, several examples of the enzymatic polymerizations using divinyl esters were

reported.²³⁻²⁵ Very recently, we have found that the lipase-catalyzed polymerization of divinyl sebacate and glycerol proceeded regioselectively to give the soluble polyester mainly consisting of 1,3-diacylated structure of glycerol.²⁶ Vinyl ester derivatives also acted as an efficient terminator of enzymatic ring-opening polymerization of lactones; facile single-step production of polyester macromonomers and telechelics was achieved by lipase-catalyzed polymerization of 13-membered lactone in the presence of vinyl esters.²⁷⁻²⁹

We have performed the systematic investigation on the effects of the reaction parameters in the lipase-catalyzed polymerization of dicarboxylic acid divinyl esters and α, ω -glycols, yielding aliphatic polyesters with different methylene chain length.³⁰ This study deals with enzymatic synthesis of polyesters having an aromatic moiety in the main chain from divinyl esters and glycols (Scheme 1). Relevant to this study, there are two reports available on the enzymatic synthesis of aromatic polyesters. One is the protease-catalyzed polymerization of diethyl terephthalate and glycols to give the corresponding oligoesters with molecular weight from 400 to 1000.³¹



Scheme 1.

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Table I. Enzyme screen for polycondensation of 1a and 2d^a

Entry	Catalyst	Yield ^b		M_w/M_n^c
		%	M_n^{c}	
1	Lipase CA	74	5500	1.6
2	Lipase CC	0		
3	Lipase MM	40	2600	1.2
4	Lipase PC	13	2000	1.1
5	Lipase PF	36	2900	1.5
6	PPL	0		
7	d	0		

^a Polymerization of **1a** and **2d** (each 1.0 mmol) using lipase catalyst (100 mg) in heptane (5.0 mL) at 60°C for 48 h. ^b Methanol-insoluble part. ^c Determined by SEC. ^d Control experiment.

The other is synthesis of poly(1,6-hexanediyl isophthalate) and poly(1,6-hexanediyl terephthalate) from the corresponding dimethyl esters by using lipase catalyst, which involved a nitrogen bubbling procedure for the removal of methanol formed during the polymerization.³²

RESULTS AND DISCUSSION

In this study, four monomers containing an aromatic moiety, divinyl isophthalate (1a), divinyl terephthalate (1b), divinyl *p*-phenylene diacetate (1c), and *p*-xylylene glycol (2a), were used as new monomer of enzymatic polymerizations.

Enzymatic Polymerization of Divinyl Isophthalate and α, ω -Alkylene Glycols

The polymerization of divinyl isophthalate (1a) with 1,6-hexanediol (2d) was carried out by using six lipases of different origin as catalyst in heptane at 60°C for 48 h (Table I). The catalysts used were lipases derived from Candida antarctica (lipase CA), Candida cylindracea (lipase CC), Mucor miehei (lipase MM), Pseudomonas cepacia (lipase PC), Pseudomonas fluorescens (lipase PF), and porcine pancreas (PPL), which showed high catalytic activity for the lactone polymerizations.^{8,13-17} Lipases CA and MM are granular immobilized enzymes and the others are crude powdery ones. In most cases, the monomer was quantitatively consumed, however, the yield was not high due to the loss of the oligomers during the purification procedure. The molecular weight of the polymer was estimated by size exclusion chromatographic (SEC) analysis using tetrahydrofuran (THF) eluent.

Active were lipases CA, MM, PC, and PF for the present polymerization, which also efficiently catalyzed the polymerization of divinyl sebacate (1d) and 1,4-butanediol (2c).³⁰ On the other hand, no polymeric materials were obtained in using lipase CC and PPL. Lipase CA showed the highest catalytic activity; the yield and molecular weight of the polymer were the largest (entry 1). In the polymerization without enzyme (control experiment), the monomers were recovered unchanged (entry 7). These results indicate that the present polymerization proceeded through the enzyme catalysis and the polymerization behaviors depended on the lipase origin. In the subsequent experiments, therefore, lipase

Table II.	Enzymatic	polymerization	of	1a	and	2 ^a
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Entry	Glycol	Solvent	Temper- ature	Yield ^b	M_n^{c}	M_w/M_n^c
-	·		°C	%		
1	2b	Heptane	60	0		
2	2c	Heptane	60	60	1500	1.2
3	2d	Acetonitrile	60	0		
4	2d	Cyclohexane	60	72	5300	1.8
5	2d	Heptane	45	42	2500	1.5
6	2d	Heptane	60	74	5500	1.6
7	2d	Heptane	75	66	4200	1.8
8	2d	Isooctane	60	73	4400	1.7
9	2d	Tetrahydrofuran	60	0		
10	2d	Toluene	60	71	2000	1.4
11	2e	Heptane	60	54	7200	2.1

^a Polymerization of **1a** and **2** (each 1.0 mmol) using lipase CA catalyst (100 mg) in organic solvent (5.0 mL) for 48 h. ^b Methanol-insoluble part. ^c Determined by SEC.

CA is mainly used as catalyst.

The polymer structure was confirmed by ¹H and ¹³C NMR spectroscopies. In all cases, the desired product was obtained and peaks due to the units of the vinyl polymer from 1 were not detected. Besides the main peaks, there were small peaks due to α -methylene moiety of the terminal hydroxyl group: a triplet peak at δ 3.7 in ¹H NMR spectrum and a peak at δ 62 in that of ¹³C NMR. Peaks of the vinyl ester were not observed, suggesting that the hydrolysis of the terminal vinyl ester occurred during the polymerization.

Effects of the polymerization conditions were systematically investigated in the polymerization of divinyl isophthalate and 1,6-hexanediol using lipase CA catalyst (Table II). At first, the polymerization was carried out in various anhydrous organic solvents at 60°C. Hydrocarbons, cyclohexane, heptane, and isooctane, provided the polymer with molecular weight of ca. 5×10^3 in good yields (entries 4, 6, and 8), indicating hydrophobic solvents were suitable for the present polymerization. Similar results were obtained in the enzymatic synthesis of aliphatic polyesters from divinyl esters.³⁰ The polymerization in toluene afforded the polymer in a moderate yield, however, the molecular weight of the resulting polymer was relatively low (entry 10). It is to be noted that toluene afforded no polymeric materials from a combination of 1d and 2c.³⁰ No polymers were obtained in acetonitrile and THF (entries 3 and 9). This may be due to the inactivation of the enzyme in polar organic solvents.

Polymerization temperature also affected the polymerization results. In the temperature range from 45° C to 75° C (entries 5—7), yield and molecular weight of the polymer were the highest at 60° C (entry 6). These data are in contrast with those in the polymerization of 1d and 2c; the polymerization at 45° C afforded the best results.³⁰ This may be due to the difference of the enzymatic reactivity between divinyl esters of aromatic and aliphatic diacids.

Next, effects of the chain length in α, ω -alkylene glycols (**2b**—**2e**) were examined. The polymerization of ethylene glycol (**2b**) with **1a** did not produce the polymer (entry 1). From 1,4-buntanediol (**2c**), 1,6-hexanediol (**2d**), and

Table III. Lipase CA-catalyzed polycondensation of 1b and 2^a

Entry	Glycol	Yield ^b	M_n^{c}	M_w/M_n^c
1	2b	<5		
2	2c	<5 34 ^a	1100	1.1
3	2d	72	2300	1.3
4	2e	58	2000	1.3

^a Polymerization of **1b** and **2** (each 1.0 mmol) using lipase CA catalyst (100 mg) in diisopropyl ether (5.0 mL) at 60°C for 48 h. ^b Methanolinsoluble part. ^c Determined by SEC. ^d Diethyl ether-insoluble part.

Table IV. Lipase CA-catalyzed polycondensation of 1c and 2^a

Entry	Glucol	Yield ^b	M_{μ}^{c}	λ <i>Α</i> / Μ °
Entry	Glycol	%	M _n	M_w/M_n^c
1	2b	34	1800	1.2
2	2c	43	4400	1.4
3	2d	44	6000	1.4
4	2e	54	5800	1.8

^a Polymerization of **1c** and **2** (each 1.0 mmol) using lipase CA catalyst (100 mg) in heptane (5.0 mL) at 60° C for 48 h. ^b Methanol-insoluble part. ^c Determined by SEC.

1,10-decanediol (2e), the polymer formation was observed. The polymer yield was the highest in using 2d (entry 6) and 2e afforded the highest molecular weight (entry 11).

Enzymatic Synthesis of Polyesters from Other Aromatic Monomers

Terephthalic acid divinyl ester (1b) was also polymerized under the similar reaction conditions of 1a (Table III). From a combination of 1b and 2b, the polymer yield was very low (entry 1), whereas the polymer was obtained from other glycols in moderate yields. The polymer yield was the highest in using 2d as glycol monomer. A similar tendency was observed in the polymerization using 1a (Table II). The polymer molecular weight was lower than that from 1a. This is due to the lower solubility of the polymer having the parasubstituted diester structure than that with the metasubstituted structure. Polymer structure was confirmed by ¹H NMR spectroscopy. Besides main peaks of the polyester, there was a characteristic triplet peak at $\delta 3.7$ due to the α -methylene protons of the terminal hydroxy group. No peaks due to the vinyl ester group were detected.

The polymerization of divinyl *p*-phenylene diacetate (1c) with glycols proceeded in the presence of lipase CA (Table IV). The polymerization of 1c and ethylene glycol (2b) afforded the polymer with relatively low molecular weight (entry 1), on the other hand, the polymer from 2b was not obtained in using aromatic diacid divinyl esters (1a and 1b). The larger the methylene chain length of 2, the higher the polymer yield. In using 2d as glycol monomer, the highest molecular weight was achieved (entry 3).

Another approach to synthesize aromatic polyesters is use of aromatic diols as monomer. However, lipasecatalyzed polymerization of divinyl sebacate with dihy-

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Table V. Lipase CA-catalyzed polycondensation of 1d and $2a^a$

Entry	Catalyst	Yield ^b	14.6	M_w/M_n^{c}
		%	M_n^{c}	
1	Lipase CA	41	6200	1.7
2	Lipase MM	61	2600	1.3
3	Lipase PC	74	4000	1.5
4	Lipase PF	72	4200	1.5

^a Polymerization of **1c** and **2** (each 2.0 mmol) using lipase CA catalyst (100 mg) in heptane (5.0 mL) at 75°C for 48 h. ^b Methanol-insoluble part. ^c Determined by SEC.

droxybenzenes, catechol, resorcinol, and hydroquinone, did not take place. Thus, *p*-xylylene glycol (**2a**) was employed as new monomer in the lipase-catalyzed polymerization of **1d** (Table V). Lipases showing the high catalytic activity for the polymerization of **1a** and **2d** (Table I) also afforded the polymer from a combination of **1d** and **2a**. In using *Pseudomonas* family lipases (lipases PC and PF), the polymer yield was high (entries 3 and 4). The polymerization catalyzed by lipase CA produced the polymer of the highest molecular weight (entry 1).

CONCLUSION

Facile synthesis of polyesters having aromatic moiety in the main chain was achieved by the lipase-catalyzed polymerization of divinyl esters and glycols. The aromatic polyesters were synthesized by using new monomers of divinyl isophthalate, divinyl terephthalate, divinyl *p*-phenylene diacetate, and *p*-xylylene glycol under mild reaction conditions. In most cases, *Candida antarctica* lipase showed the high catalytic activity for the present polymerization to give the polymers with the molecular weight of several thousands.

Further investigations on enzymatic synthesis of polyesters from other vinyl ester monomers are under way in our laboratory.

EXPERIMENTAL

Materials

Divinyl sebacate (1d) was kindly donated by Shin-etsu Chemical Co. Lipases CA and MM were gifts from Novo Nordisk Bioindustry, Ltd. Lipase CC and PPL were purchased from Biocatalysts, Ltd. and Sigma Chemical Co., respectively. Lipases PC and PF were donated by Amano Pharmaceutical Co. These enzyme catalysts were used as received. Liquid monomers and polymerization solvents were stored over freshly activated type 4 molecular sieves. Other reagents were used without further purification.

Preparation of Divinyl Isophthalate³³

A mixture of isophthalic acid (15 g, 90 mmol), vinyl acetate (39 g, 450 mmol), palladium (II) acetate (1.0 g, 4.5 mmol), and potassium acetate (0.88 g, 9.0 mmol) was dissolved in 60 mL of N,N-dimethylformamide (DMF). The mixture was stirred at 45°C for 24 h under argon. The reaction mixture was filtrated through Celite, followed by concentration of the filtrate under reduced

pressure. The product was purified by sublimation at 80°C under 400 Pa, yielding 8.2 g of divinyl isophthalate (yield 42%). ¹H NMR (CDCl₃) δ 4.8 and 5.2 (d, CH₂=CHO-, 4H), 7.5 (q, CH₂=CHO-, 2H), 7.6, 8.3, 8.8 (Ar, 4H).

Terephthalic acid and *p*-phenylene diacetic acid divinyl esters were synthesized by the similar procedure.

Lipase-Catalyzed Polymerization of Dicarboxylic Acid Divinyl Esters and Glycols

A typical run was as follows (entry 1 in Table I). Divinyl isophthalate (0.22 g, 1.0 mmol) and 1,6-hexanediol (0.12 g, 1.0 mmol) in 5.0 mL of heptane were placed in a dried test tube. Lipase CA (100 mg) was added and the mixture was gently stirred at 60°C. After 48 h, 30 mL of chloroform was added to the reaction mixture. The lipase was removed by filtration and the filtrate was concentrated by evaporation under reduced pressure. A small amount of chloroform was added to the residue and the solution was poured into a large amount of methanol. The resulting precipitates were separated by filtration, following by drying *in vacuo* to give the polymer (0.18 g, 74% yield). ¹H NMR (CDCl₃) δ 1.5 (b, OCH₂CH₂CH₂, 4H), 1.8 (b, OCH₂CH₂, 4H), 4.3 (b, OCH₂, 4H), 7.5, 8.1, 8.6 (Ar, 4H).

¹H NMR of polymer from **1b** and **2c** (CDCl₃) δ 1.5 (b, OCH₂CH₂CH₂, 4H), 1.8 (b, OCH₂CH₂, 4H), 4.3 (b, OCH₂, 4H), 8.1 (b, Ar, 4H).

Measurements

SEC analysis was carried out using a Tosoh SC8010 apparatus with a refractive index (RI) detector under the following conditions: TSKgel G3000H_{HR} column and THF eluent at a flow rate of $1.0 \,\mathrm{mL\,min^{-1}}$. The calibration curves for SEC analysis were obtained using polystyrene standards. NMR spectra were recorded on a 270 MHz JEOL JNM-EX270J spectrometer.

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