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Communications

Amylose Selectively Includes One from a Mixture of Two Resemblant Polyethers in Vine-Twining Polymerization

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In this paper, we describe that amylose almost selectively includes poly(tetrahydrofuran) (PTHF) from a mixture of poly(oxetane) (POXT) and PTHF having resemblant chemical structures and molecular weights in vine-twining polymerization. This was performed by the phosphorylase-catalyzed enzymatic polymerization of α-D-glucose 1-phosphate from maltoheptaose as a primer in the presence of a mixture of POXT and PTHF to produce an amylose−PTHF inclusion complex.

Introduction

Amylose, a linear polysaccharide with helical structure linked through α(1→4) glycosidic linkages, is a well-known host compound that forms inclusion complexes with relatively lower molecular weight compounds such as fatty acids by noncovalent interaction between guest molecules and the cavity of amylose.[1] In addition, the relationship between the including abilities and alkyl chain lengths of fatty acids has been investigated.[2] However, little has been reported regarding the formation of inclusion complexes between amylose and polymeric compounds.[3] The main difficulty in incorporating polymeric compounds into the cavity of amylose is that the driving force for the binding is only caused by hydrophobic interactions. Amylose, therefore, does not have sufficient ability to include the long chains of guest polymers into its cavity.

Enzymatic polymerization is a useful tool for the preparation of precisely regio- and stereocontrolled polysaccharides.[4] For example, phosphorylase-catalyzed enzymatic polymerization using α-D-glucose 1-phosphate (G-1-P) proceeds with the regio- and stereoselective construction of an α-glycosidic bond under mild conditions, leading to the direct formation of amylose in aqueous media. This polymerization is initiated from a maltooligosaccharide primer such as maltoheptaose (Glc_7). Then, the propagation proceeds through the following reversible reaction to produce a (1→4)-α-glucan chain, that is, amylose:

$$[(\alpha,1\rightarrow4)\text{-G}]_n + \text{G-1-P} \Leftrightarrow [(\alpha,1\rightarrow4)\text{-G}]_{n+1} + \text{P}$$

In the reaction, a glucose unit is transferred from G-1-P to the nonreducing 4-OH terminus of a (1→4)-α-glucan chain, resulting in inorganic phosphate (P).[5]

So far, we have found a new methodology for the formation of inclusion complexes composed of amylose and synthetic polymers,[6] which was achieved by means of the above-mentioned enzymatic polymerization forming amylose in the presence of guest polymers. As the guest polymers for this polymerization system, polyethers,[6a,c] polyesters,[6b,d] and a poly(ester−ether)[6d] have been employed to form the corresponding inclusion complexes with amylose. The image of the system is similar to the way vines of plants grow twining around a rod. Accordingly, we have proposed that this polymerization method be named "vine-twining polymerization."

The hydrophobicity of the guest polymers on the vine-twining polymerization can be considered to be a very important factor in whether they can be included by amylose or not. For example, when this polymerization was carried out using polyethers with different alkyl chain lengths, that is, poly(ethylene glycol) (PEG, $-(CH_2CH_2O)_n-$), poly(oxetane) (POXT, $-(CH_2CH_2CH_2O)_n-$), or poly(tetrahydrofuran) (PTHF, $-(CH_2CH_2CH_2CH_2O)_n-$), as the guest polymers, POXT and PTHF

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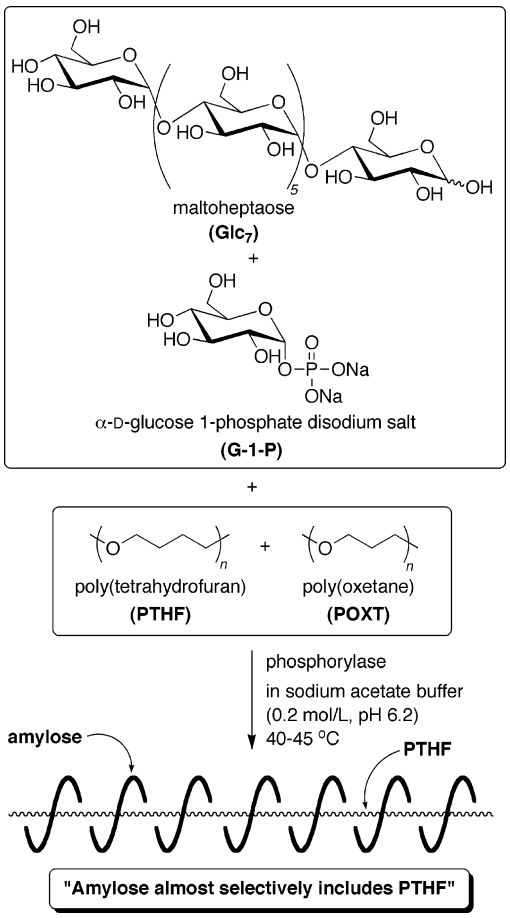
Table 1. Including Selectivity of Amylose on the Vine-Twining Polymerization[a]

	unit ratio in feed[b]	feed amount (mmol [mg])			unit ratio in the product[b]
run	PTHF/POXT	Glc_7	G-1-P	yield (mg)	PTHF/POXT/amylose
1	1.00:0.90	4.0×10^{-4} [0.5]	0.1 [30.4]	7.0	1.00:0.02:4.00
2	1.00:0.94	2.0×10^{-3} [2.3]	0.5 [152.1]	14.2	1.00:0.07:3.37
3	1.00:0.92	6.0×10^{-3} [6.9]	1.5 [456.2]	49.0	1.00:0.17:4.26
4	1.00:1.02	1.2×10^{-2} [13.8]	3.0 [912.3]	137.1	1.00:0.21:4.12

[a] Polymerization was carried out in the presence of a mixture of PTHF (0.5 mmol unit = 36.1 mg) and POXT (0.5 mmol unit = 29.0 mg) catalyzed by phosphorylase (16 unit) in sodium acetate buffer (0.2 mol/L, pH = 6.2, 5 mL) for 6 h at 40−45 °C. [b] Estimated by ^{1}H NMR measurements.

Scheme 1. Selective Inclusion of Amylose in Vine-Twining Polymerization



of the hydrophobic polyethers were included in the cavity of amylose to form the inclusion complexes, whereas no inclusion complex was obtained from the hydrophilic PEG.[6c,] Since the difference of included abilities between the two hydrophobic polyethers with resemblant chemical structures was not investigated, we have continued to study vine-twining polymerization using guest polymers.

As a recent finding in the course of this work, in this communication we would like to report that amylose almost selectively includes a type of the polymer, that is, PTHF from a mixture of POXT and PTHF (Scheme 1). The selective inclusion is probably attributed to the difference in the including ability of amylose toward these two polyethers in the vine-twining polymerization. The difference in the chemical structures between POXT and PTHF is only one methylene length, and those having similar molecular weights were used in this study. Therefore, this study is considered a tool for recognition of the polymers by means of the cavity of amylose, and thus has the potential to be extended to a separation system of the polymers in future work.

Experimental Section

Materials. Phosphorylase was supplied from Ezaki Glico Co., Ltd., Osaka, Japan.[7] POXT and PTHF were prepared by ring-opening polymerization of the corresponding cyclic ethers according to the experimental method reported in previous literature.[8] The degree of polymerization values of the PTHF and POXT were calculated from the integrated ratio of the peaks due to the $-CH_2OH$ of the polymer end to the peak due to CH_2O of the polymer units in the ^{1}H NMR spectra. Glc_7 was prepared according to the literature.[9] Other reagents and solvents were used without further purification.

Typical Procedures for the Preparation of the Inclusion Complex (Run 1 in Table 1). A mixture of POXT (M_n = ∼1800; 29.0 mg = 0.5 mmol unit) and PTHF (M_n = ∼1600; 36.1 mg = 0.5 mmol unit) was suspended in sodium acetate buffer (4.5 mL, 0.2 mol/L, pH = 6.2) using an ultrasonic wave. After the addition of Glc_7 primer (0.5 mg = 0.4 μmol) and α-D-glucose 1-phosphate disodium salt-hydrate (G-1-P; 30.4 mg = 0.1 mmol), adjustment of the pH value to 6.2 using acetic acid aqueous solution (0.2 mol/L) was carried out. Phosphorylase (16 units) was added to this solution, and then the solution was stirred vigorously for 6 h at 40−45 °C. The precipitated product was collected by filtration, washed with acetone and water, and dried under reduced pressure at room temperature to yield ca. 7.0 mg of the inclusion complex. ^{1}H NMR (600 MHz, DMSO-d_6): δ 1.40−1.60 (br, β), δ 1.65−1.72 (br, β'), δ 3.10−3.92 (m, $\mathbf{H_2}$−$\mathbf{H_6}$, α, α′ overlapping with HOD), δ 4.61, 5.44, 5.55 (OH), δ 5.10 (br, $\mathbf{H_1}$),

Measurements. The ^{1}H NMR spectra were recorded using a JEOL ECA600 spectrometer (JEOL Ltd.). The X-ray diffraction (XRD) measurements were performed at a scanning speed of 2θ = 2°/min using a Geigerflex RAD-IIB diffractometer (Rigaku Co.) with Ni-filtered Cu Kα radiation (= 0.15418 nm).

Results and Discussion

Vine-twining polymerization was performed by the phosphorylase-catalyzed enzymatic polymerization of G-1-P from Glc_7 as a primer in the presence of a mixture of POXT (M_n = ∼1800) and PTHF (M_n = ∼1600) in sodium acetate buffer (0.2 mol/L, pH = 6.2) for 6 h at 40−45 °C in this study (Scheme 1). The precipitated product was collected and characterized by the following ^{1}H NMR and XRD measurements.

Figure 1 shows the ^{1}H NMR spectra of the employed mixture of the guest polyethers ($CDCl_3$, (a)) and the product obtained by the above vine-twining polymerization (DMSO-d_6, (b)). The unit ratio of PTHF to POXT in feed was assessed to be 1.00:0.90 on the basis of the integrated ratio of the β signal due to PTHF to the β' signal due to POXT (Figure 1a). In the ^{1}H NMR spectrum of the product, the signals due to PTHF and amylose are prominently observed, whereas the β' signal due to POXT only slightly appeared (Figure 1b). These data indicate that PTHF was almost selectively included in the cavity of amylose (PTHF/POXT = 1.00:0.02) (Figure 1b and run 1 in Table 1). When PTHF or POXT was independently used as the guest for

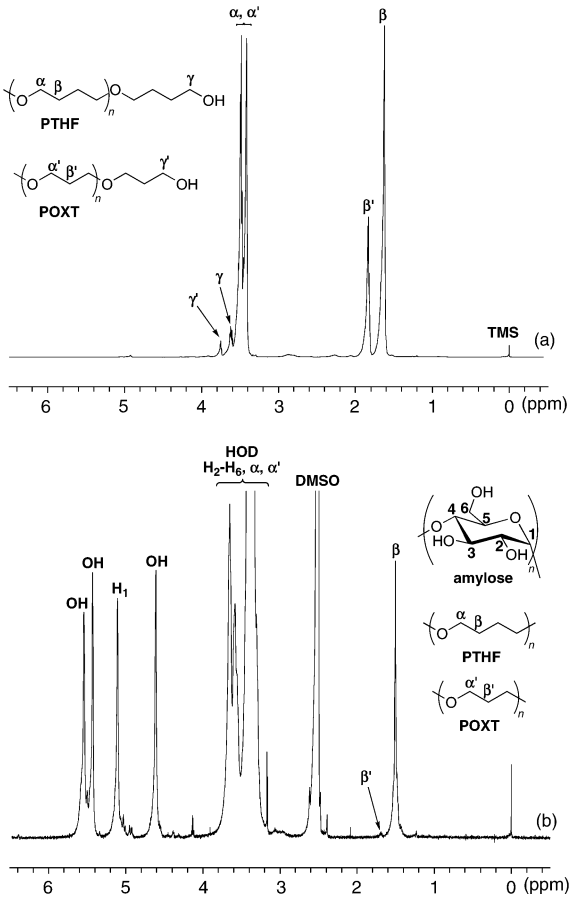


Figure 1. ^{1}H NMR spectra of (a) the mixture of guest polyethers in $CDCl_3$ and (b) the product obtained by vine-twining polymerization in DMSO-d_6.

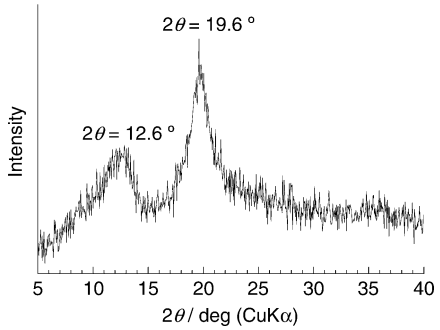


Figure 2. XRD pattern of the product (run 1 in Table 1).

the vine-twining polymerization, the inclusion complex was formed well.[6c] These results indicate that amylose selectively includes PTHF from a mixture of the two polyethers in vine-twining polymerization. The selectivity was apparent by the difference in the including ability of amylose toward the polyethers, probably attributed to the slight difference in their hydrophobicities.

The XRD pattern of the product shows two diffraction peaks at $2\theta = 12.6$ and 19.6°, corresponding to $d = 0.70$ and 0.45 nm, respectively (Figure 2), which was similar to that of the inclusion complexes of amylose with monomeric compounds[10] and with polymers, as shown in our previous studies.[3] This indicates that the product is an inclusion complex composed of amylose and PTHF.

When the enzymatic polymerization was carried out in higher concentrations of Glc_7 and G-1-P than those described above (run 1), the ratios of POXT to PTHF in the products were increased upon increasing the yields of the products (runs 2−4). Because the concentration of PTHF decreases with the progress of the polymerization due to inclusion, POXT probably started to be included at a later stage of the polymerization.

Conclusions

In the present study, we found that amylose selectively included PTHF from a mixture of two polyethers in vine-twining polymerization. This indicates that amylose discriminated PTHF from the mixture, probably because of the slight difference in the hydrophobicities. The present study provides a new method for recognition of polymers by means of the cavity of amylose. Studies on selective inclusion using other guest polymers (e.g., polyesters) by this vine-twining polymerization method are now in progress.

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