Syntheses of Isomaltose, Isomaltotetraose, and Isomaltooctaose

Shinkiti Koto, Tetsuo Uchida, and Shonosuke Zen

School of Pharmaceutical Sciences, Kitasato University, Shirokane, Minato-ku, Tokyo 108

(Received September 27, 1972)

Ethyl 2,3,4-tri-*O*-benzyl-6-*O*-*p*-nitrobenzoyl-1-thio-α-D-glucopyranoside (I–1) was brominated and subsequently condensed with a deacylated derivative of I–1 in nitromethane in the presence of 2,6-lutidine to give ethyl 2,2′,3,3′,4,4′-hexa-*O*-benzyl-6′-*O*-*p*-nitrobenzoyl-1-thio-α-isomaltoside (I–2) in a 92% yield. These processes were repeated on I–2 to give the blocked tetrasaccharide (I–4) in a 49% yield. Finally, the octasaccharide (I–8) was synthesized in an 11% yield by one more repetition of this reaction cycle. The blocked glycosides, I–2, I–4, and I–8, gave isomaltose, isomaltotetraose, and isomaltooctaose respectively after the sequence of unblocking processes: bromolysis, hydrolysis, methanolysis, and hydrogenolysis.

In synthetic carbohydrate chemistry, it has long been one of the major tasks to synthesize oligosaccharides.[1,2] Since Helferich's gentiobiose synthesis,[3] very many oligosaccharides of various types have been synthesized by all sorts of methods.

At present, there are three general methods for oligosaccharide synthesis: the original and the modified Koenigs-Knorr method,[1,2,4] Kochetkov's ortho ester method,[5] and Lemieux's glycal method.[6] Until recently, however, the fundamental course for the synthesis of relatively higher members of oligosaccharide does not seem to have been laid out or checked, although there were some early investigations.[7] Quite recently, Fréchet and Schuerch started their investigations of the step-by-step oligosaccharide synthesis on polymer support[8] and recorded the synthesis of some higher homologs in excellent yields.[9]

This paper will deal with an approach to the synthesis of the higher oligosaccharide, as in our first program: a series of syntheses of isomaltose,[10] isomaltotetraose, and isomaltooctaose by a systematic repeating scheme in which the condensation between blocks of oligosaccharide was performed as is outlined in Chart 1.[11]

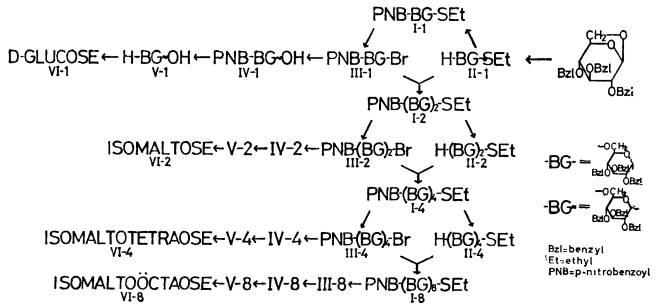

Chart 1.

At first, a suitable starting material was chosen so as to carry out the synthesis of the homolog of the 1–6 linked oligosaccharide. The blocking groups of the hydroxyl groups at the C–1 (head), C–2,3,4 (body), and C–6 (tail) positions of D-glucopyranose have to possess different properties. As the blocking group of the body, the non-anchimeric benzyl group was employed. The tail was blocked with the *p*-nitrobenzoyl group, which is stable under ordinary glycosidation and which is readily removable by mild methanolysis. As for the head, the hydroxyl group was replaced by the ethylthio group because of the necessity of generating the C–1 bromo compound.

The anhydro ring of the tri-*O*-benzyl derivative of levoglucosan[12] was cleaved with ethanethiol in the presence of zinc chloride to give the starting ethyl 2,3,4-tri-*O*-benzyl-1-thio-α-D-glucopyranoside (II–1) in a 40 % yield. This compound was then converted into the desired *p*-nitrobenzoate (I–1) and also 3,5-dinitrobenzoate (Ia–1) in the usual fashion. I–1 was then bromolyzed by Weygand's method[13] to generate 2,3,4-tri-*O*-benzyl-6-*O*-*p*-nitrobenzyl-α-D-glucopyranoside[14] (III–1) quantitatively.

Throughout this study, glycosidations were carried out by the so-called modified Koenigs-Knorr reaction.[1,2] The choice of the condensing reagent is important for the propagation of the glycoside chain up to a higher degree. 2,6-Lutidine[5] was selected since it is able to form a homogeneous reaction mixture without any activity toward the thio group and without any formation of water during the condensation. As for the solvent, nitromethane might be best for the glycosidation, because of its high polarity to ionize the glycosyl bromide as well as its excellent solvency to various organic materials.

The α-bromide (III–1) was condensed with II–1

1) R. W. Bailey and J. P. Pridham, *Adv. Carbohyd. Chem.*, **17**, 121 (1962); J. H. Pazur, "The Carbohydrates, Chemistry and Biochemistry," Vol. IIA, Academic Press, New York (1970), p. 69.
2) R. J. Ferrier, *Fortschr. Chem. Forsch.*, **14**, 389 (1970).
3) B. Helferich and W. Klein, *Ann. Chem.*, **450**, 219 (1926).
4) W. Koenigs and E. Knorr, *Ber.*, **34**, 957 (1901).
5) N. K. Kochetkov, A. J. Kholin, and A. F. Bochkov, *Tetrahedron*, **23**, 693 (1967).
6) R. U. Lemieux, R. Suemitsu, and S. W. Gunner, *Can. J. Chem.*, **46**, 1040 (1968); R. U. Lemieux, Y. Ito, K. James, and T. L. Nagabhushan, *Can. J. Chem.*, **51**, 7 (1973).
7) B. Helferich and R. Goots, *Ber.*, **64**, 109 (1931).
8) J. M. Frechét and C. Schuerch, *J. Amer. Chem. Soc.*, **93**, 492 (1971); *ibid.*, **94**, 604 (1972).
9) J. M. Frechét and Schuerch, *Carbohyd. Res.*, **22**, 399 (1972).
10) B. Lindberg, *Acta Chem. Scand.*, **3**, 1355 (1949); M. L. Wolfrom, A. O. Pittet, and I. C. Gillam, *Proc. Natl. Acad. Sci. U. S.*, **47**, 700 (1961).
11) S. Koto, T. Uchida, and S. Zen, *Chem. Lett.*, **1972**, 1049. Recently the synthesis of gentiohexaose by block condensation has been recorded by K. Takiura *et al.*, *Chem. Pharm. Bull.* (Tokyo), **20**, 438 (1972).
12) G. Zémplen, Z. Csüros, and S. Angyal, *Ber.*, **70**, 1848 (1937).
13) F. Weygand and H. Ziemann, *Ann. Chem.*, **657**, 179 (1962).
14) T. Ishikawa and H. G. Fletcher, Jr., *J. Org. Chem.*, **34**, 563 (1969).

in the above reaction system to give the blocked α-linked disaccharide (I–2) in a yield of over 90%. The oily I–2 was characterized as a deacylated crystalline compound, ethyl 2,2′,3,3′,4,4′-hexa-*O*-benzyl-1-thio-α-isomaltoside (II–2). When an analogous condensation was carried out by the use of Ia–1, the yield was about 50%.

I–2 was subjected to this sequence of unblocking processes: (1) bromolysis to split off the ethylthio group, giving a bromide (III–2); (2) hydrolysis to regenerate the hydroxyl group on the head; (3) methanolysis to remove the acyl group on the tail, and (4) catalytic hydrogenolysis to remove the benzyl groups of the body, giving isomaltose. II–2 was also bromolyzed, hydrolyzed, and hydrogenolyzed to give isomaltose.

The bromide (III–2) was condensed with II–2 to give the α-linked tetrasaccharide (I–4) in about 50% yield. The degree of polymerization was confirmed by PMR spectrum. I–4 was unblocked to give isomaltotetraose and was partially unblocked into the deacylated compound (II–4).

The connection of the tetrasaccharide blocks, II–4 and III–4, which had been derived from I–4 by bromolysis, gave the α-linked octasaccharide (I–8) in about 10% yield. The degree of polymerization was confirmed by PMR spectrum. The unblocking processes of I–8 gave isomaltooctaose (VI–8).

The synthesized blocked oligosaccharides, I–2, I–4, and I–8, had an α-linked structure, which was confirmed by their $[M]_D$ values within the range of experimental error as well as by PMR. In their PMR spectra, a doublet of an anomeric proton on the head appeared around 5.4 (J=5 Hz), whereas a signal of an internal anomeric proton(s) was observed around 5.0 (J=3 Hz). The very crucial problem of the propagation of the mixing of glycoside linkages by the formation of the undesired anomer at each condensation step, if any, may by overcome by the purification of the blocked glycosides, such as the I's of II's, before and/or after each condensation.

Turvey and Whelan[15] have investigated the partial hydrolyzate of a *Leuconostoc mesenteroides* dextran and found a linear correlation between the values of $[M]_D$ or R_M and the degree of polymerization (*D.P.*). It was found that all the values of the $[M]_D$'s and R_M's of the synthesized free oligosaccharides had a good linearity to the degree of polymerization (Figs. 1 and 2). In addition the $[M]_D$ values of the blocked oligosaccharides, I, II, IV, and V, also had good correlations to the degree of polymerization, although the ratios of the anomers of IV and V were not determined.

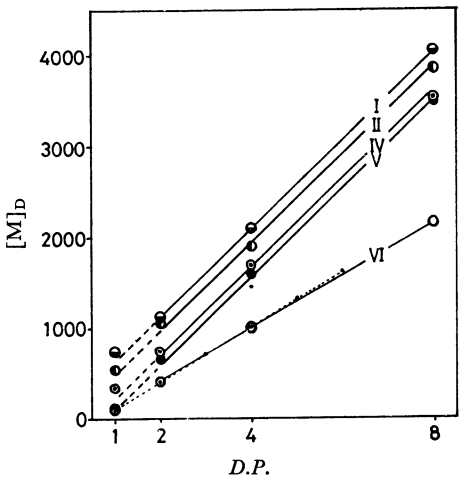

Fig. 1. Relations between *D.P.* and $[M]_D$.
····· Plotted by Turvey and Whelan

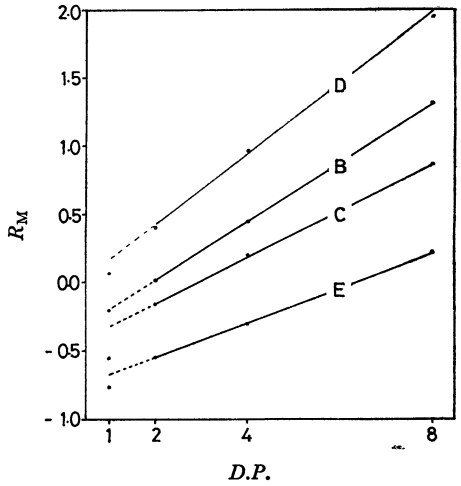

Fig. 2. Relations between *D.P.* and R_M.

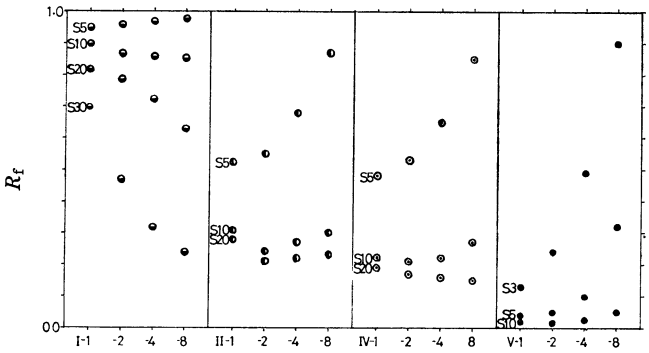

Fig. 3. Relations between *D.P.* and R_f of tlc.

The values of the R_f's in the tlc of the homolog had some correlation to the degree of polymerization (Fig. 3). The R_f's of II, IV, and V, all of which have hydroxy group(s), had roughly an inverse correlation to the degree of polymerization or the molecular size: the ratios of the occupancy of the polar group in the blocked oligosaccharide decrease as the molecular size increases.

Experimental

General Procedures. (1) The solvent systems used for the tlc (silica gel No. 7731, Merck; 10% sulfuric acid spray) and for the column chromatography over silica gel (Kanto Chemical Co.) were as follows: benzene : 2-butanone= (S40) 40 : 1, (S30) 30 : 1, (S20) 20 : 1 (S15) 15 : 1, (S10) 10 : 1, (S7) 7 : 1, (S5) 5 : 1, (S4) 4 : 1, and S(3) 3 : 1, by volume.

(2) The solvent systems used for the paper chromato-

15) J. R. Turvey and W. J. Whelan, *Biochem. J.*, **67**, 49 (1957).

graphy (Toyo filter paper No. 525, unless otherwise stated; ascending development as high as 35 cm at 25 °C; aniline hydrogenphthalate spray) and the column chromatography over cellulose powder (Toyo Roshi Co.,; 100—200 mesh) were as follows: (A) *n*-butanol : acetic acid : water=4 : 1 : 5, (B) *n*-propanol : ethyl acetate : water=6 : 1 : 3, (C) *n*-propanol : nitromethane : water=5 : 2 : 3, (D) *n*-butanol : pyridine : water=6 : 4 : 3, and (E) *n*-propanol : ethyl acetate : water=3 : 1 : 3, by volume.

(3) The melting point was determined by means of a Yanagimoto micromelting point apparatus; uncorrected values are given. The specific rotation was measured in a 1-dm tube by means of an Atago Polux apparatus. The IR spectrum was determined by means of a JASCO IRA-1 infrared spectrometer. The PMR spectrum was measured by means of a Varian S-60T spectrometer in $CDCl_3$, with TMS as the internal standard, unless otherwise mentioned. The elemental analysis was effected by means of a Perkin-Elmer Model 240 Elemental Analyzed apparatus.

(4) The bromolysis was performed by treatment with a bromine solution (Br : petroleum ether=1 : 25, by volume; prepared just before use) in ether (occasionally containing dioxane) at 25 °C for 5 min in a dark place. The bromolyzed mixture was evaporated below 20 °C and then dried at 40 °C. Three co-evaporation with toluene at 40 °C gave a hard yellowish syrup. These operations were performed as quickly as possible, using an oil pump (35 l/min).

Ethyl 2,3,4-Tri-O-benzyl-1-thio-α-glucopyranoside (II-1). A mixture of tri-*O*-benzyl-levoglucosan[12] (7.13 g), zinc chloride (2.08 g), and ethanethiol (35.7 ml) was stirred for 1.5 hr at 25 °C and then poured into iced aqueous sodium bicarbonate. The benzene extract was chromatographed with the S20 solvent to give II-1 after the elution of the unchanged material (1.35 g). Crystallization with *n*-hexane gave colorless needles (3.10 g; 47%); mp 61—63.5 °C, $[\alpha]_D^{25}$ +111° (*c* 1.8, $CHCl_3$). The PMR spectrum contained diagnostic signals of the thioethyl group at δ 1.27 (a triplet; CH_3-) and at δ 2.54 (a quartet; -CH_2-). A doublet of the anomeric proton appeared at δ 5.38, J=5.0 Hz.

Found: C, 70.07; H, 6.88%. Calcd for $C_{29}H_{34}O_5S$: C, 70.42; H, 6.93%.

Ethyl 2,3,4-Tri-O-benzyl-6-O-p-nitrobenzoyl-1-thio-α-D-glucopyranoside (I-1). II-1 (2.00 g) was treated with *p*-nitrobenzoyl chloride (1.23 g) in pyridine (10 ml) at 25 °C overnight. After the hydrolysis of the excess chloride, the benzene extract was chromatographed with the S40 solvent and crystallized with ethanol to give I-1 (2.32 g; 89%); mp 91—92 °C, $[\alpha]_D^{25}$ +116° (*c* 1.0, $CHCl_3$). The IR spectrum (film) showed characteristic bands of the ester group at 1729 cm^{-1} and of the nitro group at 1530 and 1350 cm^{-1}. The PMR spectrum showed the presence of three benzyl groups (δ 7.1~7.5) and one thioethyl group per *p*-nitrobenzoyl group (quasi-quartet, centered at δ 8.18) in I-1.

Found: C, 67.35; H, 5.83; N, 2.06%. Calcd for $C_{36}H_{37}NO_8S$: C, 67.17; H, 5.79; N, 2.18%.

Ethyl 2,3,4-Tri-O-benzyl-6-O-(3,5-dinitrobenzoyl)-1-thio-α-D-glucopyranoside (Ia-1). II-1 (200 mg) was treated with 3,5-dinitrobenzoyl chloride (140 mg) in pyridine (1.0 ml) overnight. After working-up as above, crystallization with diisopropyl ether gave Ia-1 (261 mg; 93%); mp 123—126 °C, $[\alpha]_D^{25}$ +110° (*c* 1.4, $CHCl_3$). The IR spectrum (KBr) had characteristic bands of an ester group at 1725 cm^{-1} and of the nitro group at 1545 and 1348 cm^{-1}. The PMR spectrum showed the presence of three benzyl groups per 3,5-dinitrobenzoyl group (δ 9.0—9.3) in Ia-1.

Found: C, 62.90; H, 5.35; N, 3.84%. Calcd for $C_{36}H_{36}N_2O_{11}S$: C, 62.78; H, 5.27; N, 4.07%.

Ethyl 2,2',3,3',4,4'-Hexa-O-benzyl-6'-O-p-nitrobenzoyl-1-thio-α-isomaltoside (I-2). From I-1. I-1 (0.87 g) was treated with a bromine solution (2.2 ml) in diethyl ether (14 ml) to give a bromide, III-1, $[\alpha]_D^{25}$ +127° (*c* 2.9, $CHCl_3$),[14] which was then condensed with II-1 (0.87 g) in nitromethane (6.0 ml) containing 2,6-lutidine (0.16 ml) for 20 hr. Subsequent chromatography with the S40 solvent gave a homogenous oil of I-2 (1.34 g; 92%), $[\alpha]_D^{25}$ +105° (*c* 2.0 $CHCl_3$). The PMR spectrum showed the presence of six benzyl groups and one thioethyl group per *p*-nitrobenzoyl group in I-2.

Found: C, 69.25; H, 7.22; N, 1.27%. Calcd for $C_{63}H_{65}NO_{13}S$: C, 70.30; H, 6.09; N, 1.30%.

Ethyl 2,2',3,3',4,4'-Hexa-O-benzyl-1-thio-α-isomaltoside (II-2). I-2 (1.32 g) was methanolyzed with dilute sodium methoxide (0.15 M; 20 ml) at 50 °C for 2 hr. Subsequent chromatography with the S20 solvent and crystallization with *n*-hexane gave colorless needles of II-2 (0.96 g; 84%); mp 98—100 °C, $[\alpha]_D^{25}$ +114° (*c* 1.1, $CHCl_3$). The IR(KBr) and PMR spectra indicated the absence of the *p*-nitrobenzoyl group. A couple of doublets of H-1 and H'-1 appeared at δ 5.41, J=4.5 Hz and at δ 5.08, J=4.0 Hz respectively.

Found: C, 72.65; H, 7.02%. Calcd for $C_{56}H_{62}O_{10}S$: C, 72.54; H, 6.74%.

From Ia-1. Ia-1 (100 mg) was bromolyzed in diethyl ether (1.6 ml) with a bromine solution (0.25 ml) to give a bromide, which was then condensed with II-2 (100 mg) in nitromethane (0.97 ml) in the presence of 2,6-lutidine (0.025 ml). Subsequent chromatography with the S40 solvent gave an amorphous disaccharide (76 mg; 47%), which was then methanolyzed with dilute sodium methoxide to give II-2 (52 mg; 39% from Ia-1). Admixture with the above II-2 showed no depression.

Isomaltose (VI-2). From I-2. I-2 (0.14 g) was bromolyzed with the bromine solution (0.22 ml) in diethyl ether (1.5 ml) and then stirred in moist dioxane (10%-H_2O; 10 ml) containing silver carbonate (0.2 g) at room temperature for 1 hr, followed by chromatography with the S15 solvent to give IV-2 (0.084 g; 63%), $[\alpha]_D^{25}$ +72° (*c* 0.5, $CHCl_3$). IV-2 (100 mg) was dissolved in dioxane (1 ml), treated with dilute sodium methoxide (0.15 M; 5 ml), and then chromatographed with the S5 solvent to give V-2 (77 mg; 90%), $[\alpha]_D^{25}$ +73° (*c* 0.5, $CHCl_3$). V-2 (67 mg) was hydrogenolyzed in moist methanol (initial, 2%-H_2O—final, 50%-H_2O), containing five drops of acetic acid over palladium black (20 mg×2)) to give a glass of VI-2 (18 mg; 69%), $[\alpha]_D^{25}$ +126° (*c* 0.8, H_2O)), which was identified with the authentic isomaltose by paper chromatography (Toyo filter paper No. 50; solvents, A, B, and D), as well as by a comparison with β-octa-*O*-*p*-nitro-benzoate[16] (mp 199—201 °C, $[\alpha]_D^{25}$ +97° (*c* 1.4, $CHCl_3$)), using the results of elemental analysis, a mixed-melting point determination, the specific rotation, IR(KBr), PMR, and the R_f's of tlc.

From II-2. II-2 (0.50 g) was bromolyzed in diethyl ether (5 ml) with the bromine solution (0.77 ml), hydrolyzed in moist dioxane (10%-H_2O; 10 ml) in the presence of silver carbonate (140 mg) for 2 hr, and then chromatographed with the S7 solvent to give V-2 (0.23 g; 48%). V-2 (117 mg) was hydrogenolyzed to give VI-2 (35 mg; 77%), which was identified with authentic isomaltose by paper chromatography (Toyo filter paper No. 50; solvents, A, B, and D), and also by comparison of β-octa-*O*-*p*-nitrobenzoate.[16]

16) *Cf.* E. M. Montgomery, F. B. Weakley, and G. E. Hilbert, *J. Amer. Chem. Soc.*, **71**, 1682 (1949). The octa-*O*-*p*-nitrobenzoate prepared from the authentic isomaltose according to their directions melted at 200—202 °C and had $[\alpha]_D^{25}$ +99° (*c* 1.0, $CHCl_3$) (Found: C, 52.90; H, 2.95; N, 7.17%).

Ethyl Dodeca-O-benzyl-terminal-6-O-p-nitrobenzoyl-terminal-1-thio-α-isomaltotetraoside (I-4). I-2 (0.95 g) in diethyl ether (14 ml) was treated with a bromine solution (1.41 ml) to give sirupy III-2, which was then condensed with II-2 (0.95 g) in nitromethane (8 ml) containing 2,6-lutidine (0.10 ml) for 2 days. A crude mixture was chromatographed with the S40 solvent to give a homogenous oil of I-4 (0.84 g; 59%), $[\alpha]_D^{25}$ +108° (*c* 1.0, $CHCl_3$) as the main product. The PMR spectrum showed the presence of 12 benzyl groups and one thioethyl group per *p*-nitrobenzoyl group in I-4.

Ethyl Dodeca-O-benzyl-terminal-1-thio-α-isomaltotetraoside (II-4). I-4 (85 mg) was dissolved in a mixture of methanol (20 ml) and dioxane (5 ml) and treated with dilute sodium methoxide (1.5 M; 0.25 ml) at 50 °C for 2 hr. The mixture was chromatographed with the S15 solvent to give II-4 (77 mg; 98%), $[\alpha]_D^{25}$ +106° (*c* 1.0, $CHCl_3$). The PMR spectrum had no signal of the *p*-nitrobenzoyl group.

Isomaltotetraose (VI-4). I-4 (60 mg) in diethyl ether (1.0 ml) containing dioxane (0.05 ml) was treated with a a bromine solution (0.057 ml) to give 66 mg of II-4, which was then stirred in moist dioxane (13%-H_2O; 4 ml) with silver carbonate (50 mg) for 1 hr. Subsequent chromatography with the S15 solvent gave IV-4 (38 mg; 65%), $[\alpha]_D^{25}$ +91° (*c* 1.0, $CHCl_3$), which was then methanolyzed in methanol (20 ml) containing dioxane (1 ml) with dilute sodium methoxide (1.5 M; 0.25 ml) and chromatographed with the S7 solvent to give V-4 (26 mg; 74%), $[\alpha]_D^{25}$ +91° (*c* 1.0, $CHCl_3$). V-4 (19 mg) was hydrogenolyzed in a mixed solvent of dioxane, methanol, and water (3+1+1 ml, initial→1+1+6 ml, final, containing 5 drops of acetic acid) to give a hygroscopic glass of VI-4 (4.5 mg; 63%), $[\alpha]_D^{25}$ +148° (*c* 0.5, H_2O); it was identified with the authentic isomaltotetraose by paper chromatography with the B, C, and D solvents, as well as by a comparison with β-tetradeca-*O*-benzoate[17] (mp 140.5—145.5 °C, $[\alpha]_D^{25}$ +169° (*c* 0.3, $CHCl_3$)), using the results of elemental analysis, a mixed-melting point determination, IR (KBr), and the R_f's of tlc.

Ethyl Tetracosa-O-benzyl-terminal-6-O-p-nitrobenzoyl-terminal-1-thio-α-isomaltooctaoside (I-8). I-4 (213 mg) was treated in diethyl ether (3.1 ml) containing dioxane (0.4 ml) with a bromine solution (0.16 ml) to give III-4, to which was then added a solution of II-4 (213 mg) in nitromethane (2.0 ml) containing 2,6-lutidine (0.013 ml). After one week, the mixture was chromatographed with the S30 solvent to give I-8 (45 mg; 11%), $[\alpha]_D^{25}$ +110° (*c* 1.1, $CHCl_3$) as the main product. The PMR spectrum showed the presence of 24 benzyl groups per *p*-nitro-benzoyl-group in I-8.

Ethyl Tetracosa-O-benzyl-terminal-1-thio-α-isomaltooctaoside (II-8). I-8 (10 mg) was treated in a mixed solvent of methanol (10 ml) and dioxane (1 ml) with dilute sodium methoxide (1.5 M; 0.15 ml) for 2 hr at 50 °C. Subsequent chromatography with the S20 solvent gave II-8 (8.3 mg; 86%), $[\alpha]_D^{25}$ +110° (*c* 0.8, $CHCl_3$). The IR spectrum (film) showed no absorption of the *p*-nitrobenzoyl group.

Isomaltooctaose (VI-8). I-8 (31.6 mg) was dissolved in a mixed solvent of diethyl ether (0.5 ml) and dioxane (0.1 ml) and then treated with a bromine solution (0.03 ml) to give III-8, which was then stirred in moist dioxane (14%-H_2O; 7 ml) with silver carbonate for 2 hr and chromatographed with the S20 solvent to give IV-8 (17.4 mg; 56%), $[\alpha]_D^{25}$ +99° (*c* 0.5, $CHCl_3$). IV-8 (11 mg) was treated in a mixed solvent of methanol (20 ml) and dioxane (5 ml) with dilute sodium methoxide (1.5 M; 0.25 ml) and chromatographed with the S15 solvent to give V-8 (9.8 mg; 93%), $[\alpha]_D^{25}$ +102° (*c* 0.3, $CHCl_3$).[18] V-8 (8.0 mg) was hydrogenolyzed in a mixed solvent of dioxane, methanol, and water (5+1+0.5 ml, initial→2+0.5+4 ml, final; containing five drops of acetic acid) over palladium balck (10 mg×5) to give a glass of VI-8 (1.8 mg; 59%), $[\alpha]_D^{25}$ +165° (*c* 0.2, H_2O), which had the same R_f values as the eighth spot of the partial hydrolyzate of Dextran T10 (Pharmacia Fine Chemicals Co.), developed with the B and C solvents.

17) *Cf.* Ref. 15; the preparation of the tetradeca-*O*-benzoate from the authentic isomaltotetraose according to their directions gave only an amorphous product (mp 142—146 °C, $[\alpha]_D^{25}$ +164° (*c* 0.9, $CHCl_3$) (Found: C, 68.60; H, 4.58%).

18) *Cf.* E. R. Ruckel and C. Schuerch, *J. Org. Chem.*, **31**, 2233 (1966). The reported value for the blocked polysaccharide with $\overline{DP_n}$=9 is $[\alpha]_D^{25}$ +109° ($CHCl_3$).