

## Transglycosylation of Permethylated Methyl D-Glycopyranosides in the Presence of Trimethylsilyl Trifluoromethanesulfonate

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Transglycosylation reactions among methyl 2,3,4,6-tetra-*O*-methyl-D-glycopyranosides and isomeric butyl alcohols or cyclohexanol took place in the presence of trimethylsilyl trifluoromethanesulfonate (TMSOTf) in dichloromethane. The extent of the reaction after 1 h and 24 h from mixing was determined by gas chromatography (GC). Anomerization of the substrate took place during the course of transglycosylation, which favors  $\alpha$  anomer regardless of the anomeric configurations of the starting glycosides. Transglycosylation also favors the  $\alpha$  anomer regardless of the steric bulkiness of the alcohol. *tert*-Butyl alcohol did not give any transglycosylation, suggesting the steric hindrance of approaching the bulky alcohol to the oxonium intermediate. A mechanism for the transglycosylation have been proposed.

### Introduction

The phenomena of anomerization and transglycosylation of glycosidic linkage have been widely investigated in literature.<sup>1</sup> Substrates commonly employed were methyl D-glycopyranosides and their tetraacetates. On the other hand, there is few example in which permethylated glycopyranosides are used. Considering the fact that most saccharides in nature contain free OH groups, it is not surprising that no attention has been made to the chemistry of the permethylated glycosides.

Recently we reported the phenomena of anomerization and transglycosylation of methyl 2,3,4,6-tetra-*O*-methyl-D-glycopyranosides under the conditions of reductive cleavage reaction of the glycosidic linkage.<sup>2</sup> The information is essential to deduce the total structure of a polysaccharide by reductive-cleavage method. For example, the information of monosaccharide sequence in a polysaccharide can be deduced from the structural information of disaccharides isolated from the partial cleavage of the permethylated polysaccharides, provided that no anomerization and transglycosylation take place during the course of reaction.<sup>3</sup> We reported such complication occurring in the partial reductive-cleavage of permethylated pullulan.<sup>4</sup>

Pullulan (Glc  $\alpha_4$ , Glc  $\alpha_4$ , Glc  $\alpha_6$ ) is a polysaccharide which has a repeating unit containing two 1,4- $\alpha$  and one 1,6- $\alpha$  linkages. Therefore, a partial reductive cleavage should give only three disaccharide derivatives; 4-OH-Glc  $\alpha_4$ -Glc, 4-OH-Glc  $\alpha_6$ -Glc, and 6-OH-Glc  $\alpha_4$ -Glc. However, we observed the formation of 6-OH-Glc  $\alpha_6$ -Glc, which should be formed only by transglycosylation of the monomeric 6-OH-Glc to the glycosidic linkage of the parent polysaccharide.

These findings prompted us to examine the transglycosylation with isomeric butyl alcohols and cyclohexanol. The latter may be considered as a 2° alcohol which has structural similarity to the 2,3,6-tri-*O*-methyl-1,5-anhydroalditol. We report the results of the transglycosylation in order to explore the scope of the reaction and to provide a mechanism, which should be useful in structural determination of polysaccharides.

### Experimental

**Starting Materials.** Methyl D-glycopyranosides (**1a $\alpha$**  and **1a $\beta$** ), methyl  $\alpha$ -D-mannopyranosides (**2a $\alpha$** ), and methyl D-galactopyranosides (**3a $\alpha$**  and **3a $\beta$** ), methyl iodide, trimethylsilyl trifluoromethanesulfonate, cyclohexanol, *n*-butyl alcohol, ( $\pm$ )-*sec*-butyl alcohol, *tert*-butyl alcohol, and dimethyl sulfoxide (DMSO) were all commercial products.

**Instruments.** <sup>1</sup>H NMR spectra were recorded in CDCl<sub>3</sub> as the solvent and were referenced to tetramethylsilane as internal reference on a Bruker DPX-400 MHz FT NMR spectrometer in the Central Lab of Kangwon National University. Infrared (IR) spectra were recorded on a Perkin-Elmer Model 1410R spectrophotometer. GC-MS spectra were obtained at the probe temperature of 200 °C and at 70 eV using a JEOL JMS-AX 505 WA mass spectrometer at Research Center for New Bio-Materials in Agriculture in Seoul National University. GC analyses were performed using a Hewlett-Packard 6890 gas-liquid chromatograph equipped with a split-splitless injector connected to a T-shaped splitter which, in turn, was connected to a HP-5 capillary column (30 m, film thickness 1.0  $\mu$ m, ID 0.53 mm) and a HP-50+ capillary column (30 m, film thickness 1.0  $\mu$ m, ID 0.53 mm), a flame-ionization detector for each column, and a Hewlett-Packard workstation. For retention indices experiments a Hewlett-Packard 5890A gas-liquid chromatograph equipped with a split-splitless injector connected to a T-shaped splitter which, in turn, was connected to a HP-1 capillary column (30 m, film thickness 0.25  $\mu$ m, ID 0.25 mm) and a Rtx-200 capillary column (30 m, film thickness 0.25  $\mu$ m, ID 0.25 mm), a flame-ionization detector for each column, and a Hewlett-Packard workstation. N<sub>2</sub> was used as the carrier gas. The column temperature conditions are as follows: initial temperature, 110 °C; initial hold, 2 min; temperature increase, 6 °C/min; final temperature, 200 °C; final hold, 25 min; injector temperature, 220 °C; detector temperature, 250 °C.

**Calculation of Retention Indices (RI).**<sup>5,6</sup> The reaction mixture after the transglycosylation and quenching was injected to the GC with a mixture of *n*-C<sub>11</sub>H<sub>24</sub>~*n*-C<sub>26</sub>H<sub>54</sub>. The retention index value for each glycoside component X was

calculated as follow:

$$RI = 100n + 100 \frac{t_x - t_n}{t_{(n+1)} - t_n}$$

$n$ : number of the carbon atoms of the alkane whose peak is immediately before that of the component X.

$t_x$ : recorded retention time of the component X on the chromatogram.

$t_n$ : recorded retention time of the  $n$ th alkane on the chromatogram.

$t_{n+1}$ : recorded retention time of the  $(n+1)$ th alkane on the chromatogram.

For example, the peak corresponding to **1a $\alpha$**  appears at 12.573 min when it was injected with  $n$ -C<sub>11</sub>H<sub>22</sub>~ $n$ -C<sub>26</sub>H<sub>54</sub> to the HP-5 capillary column. At this time the peaks corresponding to  $n$ -C<sub>14</sub>H<sub>30</sub> and  $n$ -C<sub>15</sub>H<sub>32</sub> appear at 10.953 min and 13.005 min, respectively. The RI value should be calculated as:

$$RI = 100 \times 14 + 100 \times \frac{12.573 - 10.953}{13.005 - 10.953} = 1478.95$$

The values of the retention indices are listed in Table 1.

**General Procedure for Methylation of Methyl D-glycosides.**<sup>7</sup> Methyl D-glycopyranosides (1.94 g, 0.01 mol) was dissolved in DMSO (30 mL) and NaOH (4.8 g, 0.12 mol) was added. The mixture was stirred for 15 h and methyl iodide (7.5 mL, 0.12 mol) was added. The solution was capped and stirred for 10 h and then poured into H<sub>2</sub>O (50 mL). The aqueous solution was extracted with chloroform (15 mL  $\times$  2) and the organic extract was washed with water (15 mL  $\times$  2). The solution was dried over Na<sub>2</sub>SO<sub>4</sub> and evaporated to dryness. The residual liquid was distilled under vacuum to give a colorless liquid. In most cases the bp and the yields of the literature<sup>7</sup> were reproduced.

**General Procedure for Preparation of Alkyl 2,3,4,6-tetra-O-methyl  $\alpha$ (and  $\beta$ )-D-glycopyranosides: an Illustrative Procedure with D-glucose.** A mixture of D-glucose (4.0 g, 0.022 mol),  $n$ -butyl alcohol (20 mL), and IRA-120 (H<sup>+</sup>) resin (1 g) was heated with stirring in an oil-bath at 75-80 °C for 6 days. The resin was removed by filtration and the filtrate was distilled under vacuum to remove  $n$ -butyl alcohol. The residual yellow liquid did not solidify by trituration with various solvents. So, it was dried under vacuum to remove solvent completely (yield 50%). The residue was dissolved in DMSO (30 mL) and NaOH (6.24 g, 0.156 mol) was added. The mixture was stirred for 11 h and then cooled in an ice bath. Methyl iodide (9.7 mL, 0.156 mol) was added and stirred for 10 h. Following the similar workup described for the preparation of permethylated methyl D-glycopyranosides, 96% of both  $\alpha$  and  $\beta$  forms of **1b** were obtained. The ratio of  $\alpha$ : $\beta$  was 2.3:1 by <sup>1</sup>H NMR. For other substrates the following product mixtures were obtained: **2b**, 60%, only  $\alpha$  form; **3b**, 75%,  $\alpha$ : $\beta$  = 3:1; **1c**, total 45%,  $\alpha$ : $\beta$  = 3:1 for one diastereomer and 4:1 for the other diastereomer; **2c**; total 20%, only  $\alpha$  for both diastereomers; **3c**; total 35%,  $\alpha$ : $\beta$  = 2.5:1 for one diastereomer and 3:1 for the other diastereomer; **1d**, total 25%,  $\alpha$ : $\beta$  = 9:1; **2d**, total 30%, only  $\alpha$  form; **3d**, total 42%,  $\alpha$ : $\beta$  = 4.5:1.

**Transglycosylation of Methyl 2,3,4,6-tetra-O-methyl  $\alpha$ (and  $\beta$ )-D-glycopyranosides: An Illustrated Procedure.** In a 3-mL Wheaton V-vial methyl 2,3,4,6-tetra-O-methyl  $\alpha$ (and  $\beta$ )-D-glycopyranosides (**1a $\alpha$** ,  **$\beta$** ) was dissolved

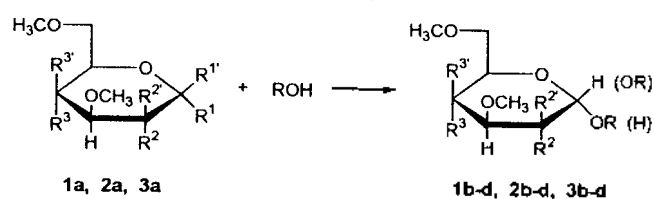
in dried dichloromethane to make 4 mM solution.  $n$ -Butyl alcohol (10 equiv) and TMSOTf (10 equiv) were added and the vial was capped. The solution was stirred for 1 h at 25 °C and quenched by adding sat'd NaHCO<sub>3</sub> solution. The organic layer was carefully taken out and was examined by GC. The results with various alcohols are listed in Table 2.

## Results and Discussion

We chose a series of permethylated methyl D-glycopyranosides such as glucose (**1a $\alpha$** , **1a $\beta$** ), mannose (**2a $\alpha$** ) and galactose (**3a $\alpha$** , **3a $\beta$** ) as substrates for the transglycosylation of glycosidic linkages. These compounds were prepared by exhaustive methylation of each methyl D-glycopyranosides which are available commercially. But  $\beta$  anomer of the mannopyranoside **2a $\beta$**  could not be prepared because of no starting material available. We attempted to synthesize methyl  $\beta$ -D-mannopyranoside but the purity was not satisfactory for the purpose of our investigation. Permethylation was carried out by following a method in literature.<sup>7</sup> However, the final products were purified by distillation under vacuum.

We also prepared permethylated D-glycopyranosides having different aglycons such as  $n$ -butyl (**1-3b**), ( $\pm$ )-*sec*-butyl (**1-3c**), and cyclohexyl (**1-3d**) group in order to use as standards for GC analyses. However, an attempts to prepare permethylated *tert*-butyl D-glycopyranosides were all failed. Direct glycosidation of D-glucose with any *tert*-butyl alcohol in the presence of various acid catalysts did not produce *tert*-butyl glycopyranoside. An alternative approach using transglycosylation of methyl D-glycopyranosides with *tert*-butyl alcohol was also unsuccessful. The failure of preparing the *tert*-butyl glycopyranosides is consistent with our observation of no transglycosylation of permethylated methyl D-glycopyranosides with the same alcohol.

Transglycosylation of permethylated methyl D-glycopyranosides with the alcohols at 25 °C produced mixtures of the  $\alpha$  and  $\beta$  anomers of the substrates (**1-3a**) as well as transglycosylated products (**1-3b-d**).



	R <sup>1</sup>	R <sup>1'</sup>	R <sup>2</sup>	R <sup>2'</sup>	R <sup>3</sup>	R <sup>3'</sup>
<b>1a-d<math>\alpha</math></b> ( $\alpha$ -Glc)	OR	H	OCH <sub>3</sub>	H	OCH <sub>3</sub>	H
<b>1a-d<math>\beta</math></b> ( $\beta$ -Glc)	H	OR	OCH <sub>3</sub>	H	OCH <sub>3</sub>	H
<b>2a-d<math>\alpha</math></b> ( $\alpha$ -Man)	OR	H	H	OCH <sub>3</sub>	OCH <sub>3</sub>	H
<b>2a-d<math>\beta</math></b> ( $\beta$ -Man)	H	OR	H	OCH <sub>3</sub>	OCH <sub>3</sub>	H
<b>3a-d<math>\alpha</math></b> ( $\alpha$ -Gal)	OR	H	OCH <sub>3</sub>	H	H	OCH <sub>3</sub>
<b>3a-d<math>\beta</math></b> ( $\beta$ -Gal)	H	OR	OCH <sub>3</sub>	H	H	OCH <sub>3</sub>

a R = CH<sub>3</sub>

b R =  $n$ -C<sub>4</sub>H<sub>9</sub>

c R = ( $\pm$ )-*sec*-C<sub>4</sub>H<sub>9</sub>

d R = cyclo-C<sub>6</sub>H<sub>11</sub>

Therefore, it is essential to identify all the components in the reaction mixture. This was done by the GC analyses of the reaction mixture. The NMR spectra of the reaction

mixture gives rough estimation of the ratios of the  $\alpha$  and  $\beta$  anomers, but the integration of the NMR peaks has error of 5-10% and it cannot be reliable, especially when one of the components is present in less than 5%. The GC analysis also has a problem of coelution of components depending on the conditions of run. We solved the problem by introducing a splitter which splits the injected sample into two columns of different polarities. The columns for the purpose are HP-1 (crossed-linked polydimethylsiloxane), HP-5 (5%-diphenyl and 95%-dimethylsiloxane), HP-50+ (50%-diphenyl and 50%-dimethylsiloxane) and Rtx-200 (methyl trifluoropropylpolysiloxane). HP-1 and H-5 are nonpolar ones and Rtx-200 has medium polarity. HP-50+ is the most polar column among the four.

Since the retention time is variable by many different factors it is not useful to list the retention time corresponding to each component. Instead we have calculated the retention indices by running the GC of each sample together with a mixture of alkanes ranging from undecane ( $C_{11}H_{24}$ ) to hexacosane ( $C_{26}H_{54}$ ). The procedure has been reported in literature for various 1,5-anhydroalditol acetates.<sup>5,6</sup> Our results are listed in Table 1.

More polar compound is expected to show longer retention time. As shown in Table 1, the  $\alpha$  anomers of the permethylated methyl D-gluco- and -galactopyranosides have larger values of RI than the  $\beta$  anomers regardless of the columns being used for analysis with one exception of **3a $\alpha$**  moving faster than **3a $\beta$**  in the Rtx-200 column. On the other hand, the opposites were the case with the methyl D-mannopyranosides, **2a $\alpha$**  showing the smaller RI values than **2a $\beta$** . Although we have no definite explanation, it may be due to changes in polarity of the permethylated methyl D-glycopyranosides as the anomeric configuration changes from  $\alpha$  to  $\beta$ . In the cases of **1a $\alpha$**  and **3a $\alpha$**  the methoxy

groups at C-1 and C-2 are *cis*. Consequently, they are more polar than their  $\beta$  anomers in which C-1—OCH<sub>3</sub> and C-2—OCH<sub>3</sub> are *trans* to each other. All methoxy groups in **1a $\beta$**  are equatorial, making the chair form least polar, and it has the smallest RI values for each column. On the other hand, **2a $\beta$**  may be the most polar because its 1,2-dimethoxy groups are *cis* to each other and 2-methoxy group is axial. The net dipole moment of **3a** should also be affected by the presence of the axial C-4—OCH<sub>3</sub> group, but the two methoxy groups at C-1 and C-4 are fairly separated even in **3a $\beta$**  although they are *cis* to each other. The retention times of these compounds are well consistent with the prediction based on the polarities of the substrates although a few exceptions are present.

As the aglycon is changed from methyl to *n*-butyl group the interaction between C-4—OCH<sub>3</sub> and C-1—OC<sub>4</sub>H<sub>9</sub> groups in **3b $\beta$**  becomes significant, causing the drastic change in polarity. As a result **3b $\beta$**  elutes after **3b $\alpha$**  in all the four columns. In case of the *n*-butyl mannopyranosides (**2b**) the  $\beta$  anomer elutes after the  $\alpha$  anomer, which is similar to the methyl mannopyranosides (**2a**). But the  $\alpha$  anomer of the *n*-butyl D-glucopyranoside (**1b $\alpha$** ) show different elution feature depending on the column: that is, no separation of the  $\alpha$  and  $\beta$  anomers with HP-5, and the  $\alpha$  anomer moving faster with HP-1 but slower with HP-50+ and Rtx-200 column.

When ( $\pm$ )-*sec*-butyl alcohol is employed for the transglycosylation a mixture of the diastereomers are formed. The diastereomers of **1c $\beta$**  and **2c $\alpha$**  show distinct RI values for each column, but those of **1c $\alpha$** , **3c $\alpha$** , and **3c $\beta$**  are not separable with the HP columns. Only Rtx-200 column provides good separation. The elution order of the *sec*-butyl glycopyranosides are pretty similar to that of the *n*-butyl glycopyranosides, as shown in Table 1.

The cyclohexyl glycopyranosides show the largest RI

**Table 1.** Retention indices (RI) of the permethylated alkyl D-glycosides

			<b>1<math>\alpha</math></b>	<b>1<math>\beta</math></b>	<b>2<math>\alpha</math></b>	<b>2<math>\beta</math></b>	<b>3<math>\alpha</math></b>	<b>3<math>\beta</math></b>
			$\alpha$ -Glc	$\beta$ -Glc	$\alpha$ -Man	$\beta$ -Man	$\alpha$ -Gal	$\beta$ -Gal
<b>a</b>	Me	A	1454.99	1416.06	1446.71	1459.14	1468.12	1457.85
		B	1479.13	1436.59	1473.08	1491.48	1497.09	1484.94
		C	1642.93	1585.24	1633.84	1682.65	1661.56	1669.54
		D	1732.86	1655.05	1743.47	1791.86	1778.03	1752.71
<b>b</b>	<i>n</i> -Bu	A	1644.44	1703.71	1657.15	1674.11	1660.46	1681.92
		B	1667.96	1667.96	1683.04	1706.27	1683.91	1707.70
		C	1823.68	1800.00	1837.32	1874.92	1823.31	1866.38
		D	1931.76	1870.22	1939.96	1988.49	1947.96	1966.26
<b>c</b>	<i>sec</i> -Bu	A	1600.00	1576.35	1585.76	1557.84	1608.32	1625.00
			1602.89	1592.49	1594.95	1566.95	1608.32	1625.00
		B	1619.73	1595.63	1623.82	1640.99	1632.43	1650.36
			1619.73	1610.27	1631.80	a	1632.43	1650.36
		C	1748.73	1713.59	1776.70	1747.92	1759.32	1763.25
			1751.67	1733.94	1792.01	a	1763.25	1772.04
		D	1845.77	1778.42	1872.03	1889.99	1884.07	1909.31
			1845.77	1800.00	1876.78	1900.00	1884.07	1909.31
<b>d</b>	Cyclohexyl	A	1883.58	1763.33	1896.67	1907.67	1893.64	1918.22
		B	1910.51	1910.51	1924.18	1940.45	1921.81	1949.32
		C	2054.46	2026.99	2098.32	2128.03	2068.96	2121.42
		D	2206.74	2169.28	2237.24	2274.38	2240.41	2270.07

A, HP-1; B, HP-5; C, Rtx-200; D, HP-50+

<sup>a</sup>Not observed. It is not certain if the diastereomers are not separable, or if it is not detected because they are very small amount.

**Table 2.** <sup>1</sup>H NMR and mass spectral data of the permethylated alkyl D-glycopyranosides

	$\delta\text{H-1(J, Hz)}^a$	$\delta\text{OCH}_3^b$	O-R	EI-mass, m/z (%)	CI-mass, m/z (%)
<b>1b<math>\alpha</math></b>	4.90 (3.59)	3.39, 3.47 3.52, 3.61	0.88 (t, 3H) 1.38 (m, 2H) 1.60 (m, 2H) 3.21 (t, 2H)	219(6), 187(5), 117(35), 101(42), 88(100)	310(40), 236(100), 187(36), 117(58), 88(80)
<b>1b<math>\beta</math></b>	4.19 (7.65)	3.38, 3.51 3.56, 3.60	0.88 (t, 3H) 1.34 (m, 2H) 1.57 (m, 2H) 3.19 (t, 2H)	219(10), 117(32), 101(38), 98(100)	310(68), 236(94), 187(44), 117(72), 88(100)
<b>1c<math>\alpha</math></b>	5.06 (3.86)	3.41, 3.54 3.57, 3.63	0.91 (t, 3H) 1.19 (d, 2H) 1.55 (m, 2H)	219(3), 187(6), 117(40), 101(48), 88(100)	310(80), 236(100), 187(30), 117(36), 101(21), 88(51)
	5.04 (3.70)	3.41, 3.47 3.52, 3.62	0.90 (t, 3H) 1.15 (d, 2H) 1.52 (m, 2H)	219(3), 187(9), 117(42), 101(55), 88(100)	310(77), 236(100), 187(30), 117(36), 101(24), 88(47)
<b>1c<math>\beta</math></b>	4.28 (7.79)	3.54, 3.61 3.66, 3.67	0.95 (t, 3H) 1.20 (d, 2H) 1.55 (m, 2H)	219(3), 187(8), 117(45), 101(52), 88(100)	310(40), 236(100), 187(23), 117(26), 88(35)
	4.14 (7.66)	3.48, 3.61 3.70, 3.73	0.95 (t, 3H) 1.21 (d, 2H) 1.55 (m, 2H)	219(3), 187(8), 117(40), 101(50), 88(100)	310(25), 236(80), 187(28), 117(45), 88(100)
<b>1d<math>\alpha</math></b>	5.09 (3.69)	3.41, 3.47 3.54, 3.63	1.0~2.0 (m)	219(13), 143(43), 111(11), 102(12), 101(80), 88(100)	336(41), 236(100), 187(32), 143(40), 101(30), 88(49)
<b>1d<math>\beta</math></b>	4.33 (7.80)	3.50, 3.56 3.58, 3.61	1.0~2.0 (m)	219(13), 143(39), 102(11), 101(69), 88(100)	336(85), 236(100), 187(42), 143(51), 101(43), 88(63)
<b>2b<math>\alpha</math></b>	4.87 (0.30)	3.39, 3.48 3.46, 3.51	0.90 (t, 3H) 1.35 (m, 2H) 1.50 (m, 2H) 3.64 (t, 2H)	219(10), 117(25), 101(50), 88(100)	310(72), 236(100), 187(33), 88(66)
<b>2c<math>\alpha</math></b>	5.03 (0.30)	3.50, 3.51 3.51, 3.52	0.91 (t, 3H) 1.18 (d, 2H) 1.52 (dq, 2H)	219(20), 117(22), 101(55), 88(100)	310(46), 236(100), 187(32), 88(50)
	4.98 (0.30)	3.40, 3.41 3.48, 3.49	0.89 (t, 3H) 1.12 (d, 2H) 1.46 (dq, 2H)	219(22), 117(21), 101(55), 88(100)	310(45), 236(100), 187(23), 88(65)
<b>2d<math>\alpha</math></b>	5.05 (1.57)	3.40, 3.47 3.50, 3.52	1.2~1.8 (m)	219(15), 143(55), 101(40), 88(100)	336(41), 236(100), 143(50), 88(60)
<b>3b<math>\alpha</math></b>	4.97 (3.24)	3.39, 3.48 3.51, 3.56	0.90 (t, 3H) 1.38 (m, 2H) 1.60 (m, 2H) 3.87 (t, 2H)	219(3), 117(45), 101(35), 88(100)	310(45), 236(100), 187(22), 88(90)
<b>3b<math>\beta</math></b>	4.20 (7.52)	3.55, 3.58 3.60, 3.64	0.89 (t, 3H) 1.35 (m, 2H) 1.60 (m, 2H) 3.69 (t, 2H)	219(4), 117(30), 101(42), 88(100)	310(75), 236(100), 187(38), 88(80)
<b>3c<math>\alpha</math></b>	5.09 (3.72)	3.40, 3.49 3.52, 3.57	0.92 (t, 3H) 1.15 (d, 2H) 1.48 (dq, 2H)	219(4), 117(20), 101(65), 88(100)	310(70), 236(100), 187(33), 88(80)
	5.07 (3.52)	3.39, 3.48 3.51, 3.57	0.93 (t, 3H) 1.21 (d, 2H) 1.63 (dq, 2H)	219(5), 117(25), 101(66), 88(100)	310(70), 236(100), 187(23), 88(68)
<b>3c<math>\beta</math></b>	4.27 (2.61)	3.41, 3.42 3.51, 3.57	0.90 (t, 3H) 1.16 (d, 2H) 1.50 (dq, 2H)	219(5), 117(22), 101(65), 88(100)	310(65), 236(100), 187(41), 88(90)
	4.17 (7.60)	3.40, 3.41 3.48, 3.58	0.90 (t, 3H) 1.18 (d, 2H) 1.63 (dq, 2H)	219(5), 117(22), 101(55), 88(100)	310(65), 236(100), 187(33), 88(80)
<b>3d<math>\alpha</math></b>	5.13 (3.17)	3.40, 3.47 3.52, 3.57	1.1~2.0 (m)	219(3), 143(55), 101(45), 88(100)	336(86), 236(100), 187(48), 88(65)
<b>3d<math>\beta</math></b>	4.32 (7.45)	3.49, 3.56 3.59, 3.60	1.1~2.0 (m)	219(3), 143(52), 101(50), 88(100)	336(72), 236(100), 187(32), 88(90)

<sup>a</sup> Doublet, 1H. <sup>b</sup> Singlet, 3H.

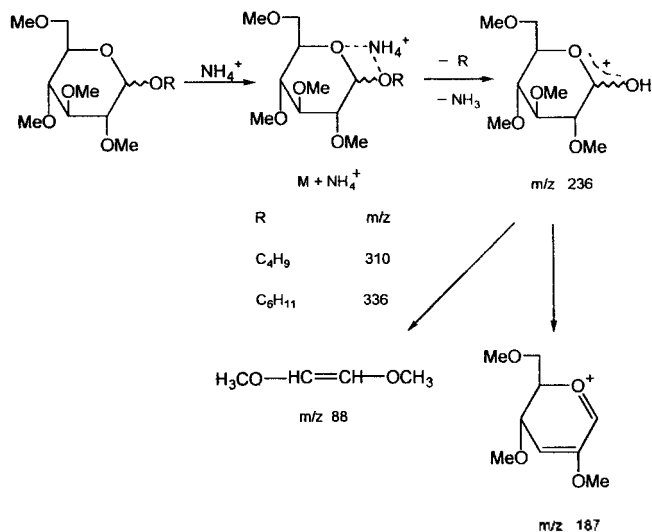
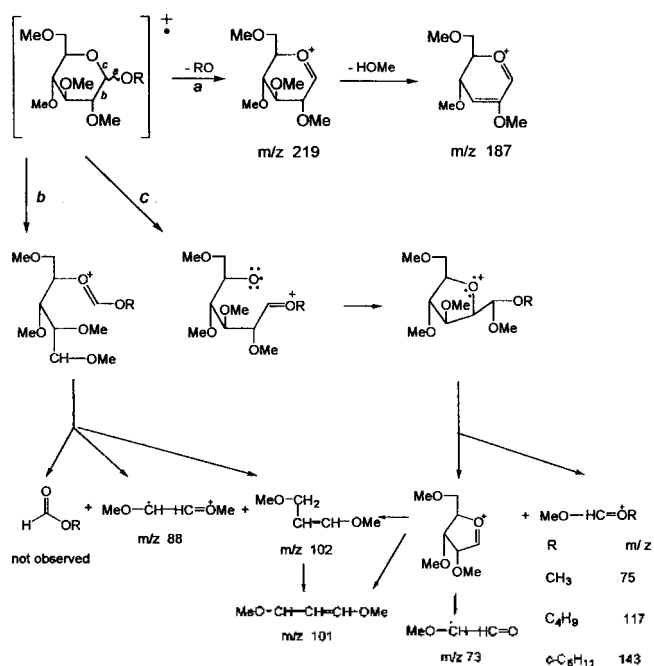
values as expected for the larger size of aglycon<sup>8</sup> and the elution order is similar to that of the *n*-butyl glycopyranosides. It is apparent that the anomeric configuration of the glycopyranoside, the size of the aglycon, and the presence of the axial methoxy group in **2** and **3** are determining factors which govern the relative order of elution.

The reaction mixtures of the transglycosylation were analyzed by GC-EI-mass and GC-CI-mass spectrometry (Table 2). The EI-mass spectra show an interesting pattern of fragmentation.<sup>9</sup> There is not a single case in which  $M^+$  ion is detected. The molecular ion, once it is formed, seems to undergo the cleavage of the three bonds, designated as *a*, *b*, and *c* from C-1. The resulting ionic species may undergo further fragmentation as shown in Scheme 1.

The base peak,  $m/z$  88, from the all permethylated glycosides should be corresponding to  $\text{CH}_3\text{OCH}=\text{CH}=\text{O}^+\text{CH}_3$  which can be formed not only from the pathway *b* but from other intermediates because of the presence of the vicinal dimethoxy groups. Other notable ions is  $m/z$  101 and its formation should be common to all the glycosides. The identity of the aglycon could be readily confirmed by the fragments corresponding to  $\text{MeOCH}=\text{O}^+\text{R}$  ( $m/z$  75 for methyl,  $m/z$  117 for butyl, and  $m/z$  143 for cyclohexyl). Apparently, the acyclic oxonium ion (from *c*) undergoes ring closure to form a furanoside skeleton and the methoxy group at C-2 undergoes rearrangement to C-1 simultaneously.

The CI-mass spectra show peaks corresponding to  $M+\text{NH}_4^+$  for all the components. Although the fragmentation patterns on the CI-mass are similar in all the glycosides regardless of aglycon, they readily confirm the presence of the transglycosylation products. The base peak in the CI-mass is  $m/z$  236 which should be driven by the loss of the aglycon. The fragmentation pattern of the CI-mass spectra is illustrated in Scheme 2.

The <sup>1</sup>H NMR spectrum of the reaction mixture also provides a definite evidence of the transglycosylation. The  $\alpha$  and  $\beta$  anomers of the methyl glycosides show the anomeric pro-



ton signals usually at  $\delta$  4.80 and 4.15, respectively. Those of the transglycosylated products show signals shifted to down-field significantly. The shift is most profound when the aglycon is cyclohexyl.

The coupling constant of the anomeric proton signal becomes larger as the aglycon becomes bulkier, but the change is not much significant except the mannosides. The NMR spectra of the transglycosylation mixture show only a trace of the  $\beta$  anomer present in the mixture. Therefore, the detection of the  $\beta$  mannosides has to depend on the GC-mass. The coupling constants of the permethylated methyl  $\alpha$ -D-mannopyranoside (**2a $\alpha$** ) is about 3Hz, but those of **2b $\alpha$** , **2c $\alpha$** , and **2d $\alpha$**  are 0.30, 0.30, and 1.57 Hz, respectively.

The axial arrangements of C-2—OCH<sub>3</sub> and C-1—OR may cause the dihedral angle of H—C-1—C-2—H close to 90° and the coupling constant becomes smaller as the aglycon becomes larger.

The extents of transglycosylations of **1-3** with various alcohols in the presence of TMSOTf after 1 h and 24 h are listed in Table 3. In general, the  $\beta$  anomers underwent faster reaction than their  $\alpha$  counterparts. There are several noteworthy characteristics in Table 3.

First of all, the extent of transglycosylation is roughly similar (70-90%) for all the substrates within 1 h regardless of the nature of the alcohols. Secondly, the  $\alpha$  anomers are the major products in all the cases. Thirdly, the ratios of  $\alpha$  to  $\beta$  of the products vary depending on the alcohol in case of the glucopyranosides, but the mannosides favor  $\alpha$  almost exclusively. Finally, the extent of anomerization is the smallest when the transglycosylation is taking place with *n*-butyl alcohol. The anomeric effect is most prominent in the case of the mannoside (**2a $\alpha$** ) in which the C-1 and C-2 substituents are *trans* to each other and only less than 1% of the  $\beta$ -anomer forms by the anomerization reaction.

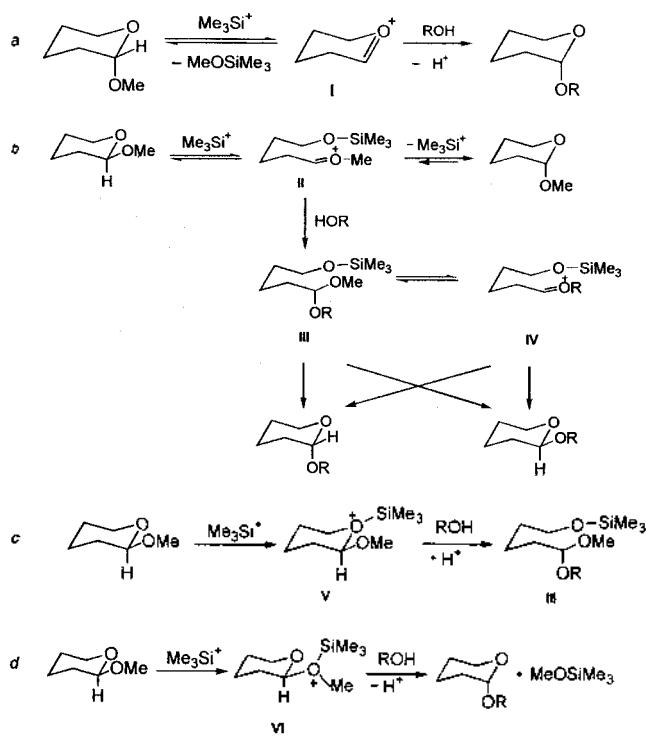
The transglycosylation of the  $\alpha$ -glycopyranosides seems to take place via a cyclic oxonium ion intermediate (**I**) as shown in the pathway *a* of Scheme 3. One of the lone pair orbitals in the  $\alpha$ -glycopyranoside is in the *anti* arrangement to C-1—OCH<sub>3</sub> bond, and therefore, the formation of the cyclic oxonium ion is greatly assisted by that orbital. Once the oxonium ion is formed it can be attacked by ROH and

**Table 3.** Molar compositions of the alkyl D-glycopyranosides present in the reaction mixture of the transglycosylation at 25°C after 1 h and 24 h (parenthesis)

ROH		$\alpha$ -Glc	$\beta$ -Glc	$\alpha$ -Man	$\alpha$ -Gal	$\beta$ -Gal
<i>n</i> -Bu	<b>a<math>\alpha</math></b>	16.0 ( 6.5)	6.9 ( 1.5)	5.9 ( 4.4)	4.9 ( 3.2)	3.6 ( 3.9)
	<b>a<math>\beta</math></b>	1.4 ( 0.8)	1.7 ( 0.4)	0.3 ( 0.2)	0.8 ( 0.7)	0.4 ( 0.3)
	<b>b<math>\alpha</math></b>	63.9 (72.2)	74.9 (80.6)	89.1 (91.3)	78.1 (79.4)	76.4 (79.1)
	<b>b<math>\beta</math></b>	18.7 (20.5)	16.4 (17.5)	4.7 ( 4.1)	16.2 (16.8)	19.6 (16.7)
<i>sec</i> -Bu	<b>a<math>\alpha</math></b>	20.7 (13.5)	13.6 ( 2.8)	20.1 (14.6)	23.8 (11.9)	15.0 (12.5)
	<b>a<math>\beta</math></b>	4.1 ( 2.5)	3.0 ( 0.8)	0.0 ( 0.7)	5.0 ( 2.2)	3.5 ( 2.5)
	<b>c<math>\alpha</math></b>	33.0 (39.4)	35.0 (46.3)	40.1 (42.3)	30.0 (31.7)	30.5 (37.1)
	<b>c<math>\beta</math></b>	30.0 (32.2)	34.8 (37.0)	39.8 (41.8)	32.9 (38.6)	37.0 (38.0)
		6.1 ( 6.7)	7.1 ( 7.1)	0.0 ( 0.4)	4.3 ( 9.2)	8.9 ( 5.0)
Cyclohexyl	<b>a<math>\alpha</math></b>	20.6 (19.0)	16.6 ( 3.4)	14.1 (9.0)	12.4 (13.1)	14.3 ( 9.7)
	<b>a<math>\beta</math></b>	4.1 ( 1.8)	3.9 ( 1.0)	0.6 ( 0.5)	2.8 ( 2.6)	3.1 ( 2.3)
	<b>d<math>\alpha</math></b>	68.5 (70.3)	66.1 (79.9)	81.4 (86.0)	67.1 (67.0)	65.5 (70.2)
	<b>d<math>\beta</math></b>	6.8 ( 8.9)	13.4 (15.7)	3.8 ( 4.5)	17.7 (17.3)	17.1 (17.8)

the so-called anomeric effect should favor the  $\alpha$  product. However, the methoxy group at C-1 should depart not as methoxide ion which is one of the poorest leaving groups but as methoxytrimethylsilane.<sup>10</sup> This step should be the rate-determining because the cyclic oxonium ion is energetically less favorable than the pyranoside ring.

Once the cyclic oxonium ion is formed the attack of ROH may favor  $\alpha$  direction, but the preference seems to be significantly different depending on the nature of the alcohol. The ratios of  $\alpha$ : $\beta$  are approximately 3:1, 5:1, and 9:1 for *n*-butyl, *sec*-butyl, and cyclohexyl, respectively, for the glucopyranoside. They are 5:1, 7:1, and 4:1 for the galactopyranoside. The ratio must have been determined by complex factors such as product stability, relative rate of the attack of ROH to the cyclic oxonium ion, etc, and any rationale is out of the scope of the present study.

**Scheme 3**

There are three different pathways leading to transglycosylation in the  $\beta$  anomer as shown in Scheme 3. The pathway *b* involves an acyclic oxonium intermediate (II) which may anomerize to the  $\alpha$  form or react with ROH to give an acetal III. An intramolecular  $S_N2$  type of reaction will lead to the transglycosylated product. It is not certain whether the acetal III gives the transglycosylation products or it is further transformed to another acyclic oxonium ion IV and then recyclize to give the final products.

The pathway *c* also gives the acetal intermediate III but it is formed by the nucleophilic attack of ROH to V which is the result of the silylation of the ring oxygen atom. On the other hand, a similar silylation of the oxygen atom at C-1 like VI and subsequent intermolecular  $S_N2$  type of reaction should directly give the  $\alpha$  product (pathway *d*). But the pathways *c* and *d* may be ruled out because the extents of the transglycosylation of **1a $\beta$**  with *n*-butyl alcohol and *sec*-butyl alcohol are not much different within 1 h. If a direct  $S_N2$  type of reaction is involved, a 1° alcohol should be a better nucleophile than a 2° alcohol and a faster rate is expected with the 1° alcohol.

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