Carbohydrate Research, 218 (1991) 63–73
Elsevier Science Publishers B.V., Amsterdam

Determination of the absolute configuration of monosaccharides using (+) or (−) 2-*tert*-butyl-2-methyl-1,3-benzodioxole-4- carboxylic acid and high-resolution ^{1}H-n.m.r. spectroscopy*

Yoshihiro Nishida, Mariko Kumagai, Akiko Kamiyama, Hiroshi Ohrui, and Hiroshi Meguro

Department of Food Chemistry, Faculty of Agriculture, Tohoku University, Tsutsumidohri-Amamiyamachi 1-1, Aobaku, Sendai 981 (Japan)

(Received August 13th, 1990; accepted for publication in revised form March 9th, 1991)

ABSTRACT

Reaction of (−) 2-*tert*-butyl-2-methyl-1,3-benzodioxole-4-carboxylic acid with per-*O*-acetyl-D- and L-glucopyranosyl bromides yielded diastereomeric per-*O*-acetylated glycopyranosyl 2-*tert*-butyl-2-methyl-1,3-benzodiazole-4-carboxylates. Their ^{1}H-n.m.r. signals, especially the strong singlet peaks of *tert*-Bu and Me groups in the 1-ester portion were diagnostic for the determination of the D,L-configuration and optical purity of monosaccharides.

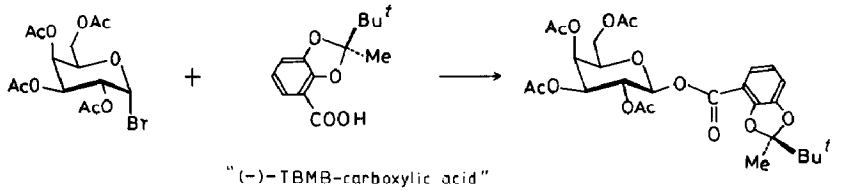
INTRODUCTION

Determination of the enantiomeric configuration of sugars is a problem when either enantiomer may be present in natural products, as with galactose, fucose, and arabinose[1]. Polarimetric or enzymic methods have been most commonly used to differentiate these enantiomers[2]. More-recent methods have employed chiral alcohols, such as (+)- or (−)-2-butanol to give diastereomeric glycosides that are separated by gas–liquid chromatography (g.l.c.)[3], chiral-phase g.l.c.[4], or high-performance liquid chromatography (h.p.l.c.)[5].

High-resolution n.m.r. spectroscopy provides another approach for determining the absolute configuration and optical purity of chiral alcohols or amines by using chiral shift reagents[6] or chiral derivatizing agents[7]. Application with carbohydrate molecules, has, however, been limited because of their many OH groups reactive to these reagents. Recently[8], we have developed a new type of chiral derivatizing agent, (*S*)-(+)- or (*R*)-(−)-2-*tert*-butyl-2-methyl-1,3-benzodioxole-4-carboxylic acid [(+)- or (−)-TBMB-carboxylic acid]. This reagent has two isolated methyl groups at C-2 (Me and *tert*-Bu) which give sharp singlet peaks diagnostic for the determination of the absolute configuration of chiral alcohols or amines[8]. In this paper, we apply this reagent for the analysis of D,L monosaccharides. Our approach involves the derivatization of the D or L-sugar

* Presented at the 15th International Carbohydrate Symposium, Yokohama, Japan, August 12–17, 1990.

with both (−)-TBMB-carboxylic acid and racemic TBMB-carboxylic acid and comparison of the ^{1}H-n.m.r. signals, especially of the *tert*-Bu and Me groups, between the two diastereomers thus derived.



RESULTS AND DISCUSSION

A. Derivatization of D or L-*sugars with TBMB-carboxylic acid.* — Sugars have several sites available for derivatization with TBMB-carboxylic acid, through esterification of one of the OH groups. Selective esterification, however, requires prior protection of the other OH groups. We have adopted another method as shown in the accompanying formulas because it can be applied to any monosaccharide.

The monosaccharides D-glucose, D-galactose, D-mannose, D-fucose, L-rhamnose, D-xylose, L-arabinose, and D-ribose, were derivatized with both of (−)- and (+)-TBMB carboxylic acid in this way. All sugars studied here gave the corresponding per-*O*-acetylglycopyranosyl 1-TBMB-carboxylates (**1–11**, 30–70% yield, based on the sugars used) which were identified by e.i.-m.s. and n.m.r. spectroscopy (Tables I–V). The e.i.-m.s. (70 eV) gave a strong ion peak of $m/z = (M-57)^+$ and a smaller molecular-ion peak (Tables I and II) for all derivatives. This pattern is characteristic of the 1-TBMB-carboxylates. The anomeric configurations, ring structures, and ring conformations were determined mainly from n.m.r. data of the anomeric protons (chemical shifts and coupling constants), as summarized in Tables I and II.

Most of the sugars gave a single pyranoside anomer, except for mannose, rhamnose, and ribose. Mannose and rhamnose produced a ~3:1 mixture of *α-axial* (**3** and **6**) and *β-equatorial* pyranosides (**4** and **7**) in ~30% total yields. These anomers were readily separated by preparative t.l.c. Production of the α-pyranoside from mannose and rhamnose indicates significant neighboring-group participation by the 2-acetoxyl group[10]. For ribose, the *β*-D-pyranoside (**10**) and a furanoside(**11**) were obtained in ~2:1 ratio, probably because the two ring forms had already formed during the acetylation step. The pyranoside and furanoside were inseparable by t.l.c. on silica gel or silica gel column chromatography.

All of the hexopyranosides (**1–7**) adopted the $^4C_1$ (D) ring conformation, whereas the pentopyranosides (**8–10**) occured as mixtures of the $^4C_1$ and $^1C_4$ conformations. The conformation of the ribofuranoside (**11**) was not determined as its spectrum was too complex

B. ^{1}H-N.m.r. differentiation between D- *and* L-*sugars.* — The ^{1}H-n.m.r. signals of the Me and *tert*-Bu groups of per-*O*-acetyl-D- or L-glycosyl 1-[(+)-TBMB-carboxy-

TABLE I

^{1}H-N.m.r. and e.i.–m.s. data for per-*O*-acetyl-D- and L-glycopyranosyl 1-[(−)-2-*tert*-butyl-2-methyl-1,3-benzodioxole -4-carboxylates][a]

Compound		*Chemical shifs (δ p.p.m.)*			*Ring conformations*	*E.i.–m.s. (70 eV)*	
		tert-*Bu*	*Me*	*H-1*($J_{H1,H2}$)[b]	(D *form*)[c]	M^+	$[M-57]^+$
1	β-D-Glc*p*	1.095	1.582	5.898(8.1)	$^4C_1$	566 (6)[d]	509 (90)
	L-Glc*p*	1.077	1.618	5.937(8.1)			
	Δ	+0.018	−0.036	−0.039			
2	β-D-Gal*p*	1.100	1.598	5.878(8.4)	$^4C_1$	566 (6)	509 (64)
	L-Gal*p*	1.083	1.623	5.902(8.4)			
	Δ	+0.017	−0.025	−0.024			
3	α-D-Man*p*	1.111	1.621	6.390(1.8)	$^4C_1$	566 (10)	509 (162)
	L-Man*p*	1.094	1.701	6.405(1.8)			
	Δ	+0.017	−0.080	−0.015			
4	β-D-Man*p*	1.079	1.597	6.073(1.1)	$^4C_1$	566 (14)	509 (182)
	L-Man*p*	1.058	1.607	6.045(1.1)			
	Δ	+0.021	−0.010	+0.028			
5	β-D-Fuc*p*	1.099	1.591	5.841(8.4)	$^4C_1$	508 (11)	451 (68)
	L-Fuc*p*	1.078	1.622	5.875(8.4)			
	Δ	+0.021	−0.031	−0.034			
6	α-D-Rha*p*	1.112	1.628	6.312(1.8)	$^4C_1$	508 (7)	451 (101)
	L-Rha*p*	1.117	1.670	6.322(1.8)			
	Δ	−0.005	−0.042	−0.034			
7	β-D-Rha*p*	1.078	1.604	6.030(1.1)	$^4C_1$	508 (5)	451 (42)
	L-Rha*p*	1.057	1.628	6.017(1.1)			
	Δ	+0.021	−0.024	+0.013			

[a] Measured at 400 MHz in $CDCl_3$ solution at 27–28° with internal Me_4Si standard. ^{1}H-N.m.r. and e.i.-m.s. data were obtained from the spectra of per-*O*-acetyl-D-or L-glycopyranosyl 1-[(−)-TBMB-carboxylates] and the corresponding 1-(+)-TBMB-carboxylates]. Assignment of D-or L was based on the assumption that (−)-TBMB-carboxylic acid was used for the derivatization (see text). [b] Coupling constant, J ±0.2 Hz. [c] Predicted from the $J_{H1,H2}$ values or the other vicinal H–H couplings. [d] Relative intensity of the M^+ and $[M-57]^+$ ion peaks.

lates] are depicted in Figs. 1 and 2, respectively. The assignment of D or L is based on the assumption that (−)-TBMB-carboxylic acid is used for the derivatization. Thus, the resonances attributable to per-*O*-acetyl-D-glycosyl 1-[(+)-TBMB-carboxylate] are labeled L and those for the L-glycosyl 1-[(+)-TBMB-carboxylates are labeled D as they are equivalent to those of their enantiomeric per-*O*-acetyl-L and D-glycosyl 1-[(−)-TBMB-carboxylates], respectively.

The significants results in Figs. 1 and 2 may be summarized as follows: 1. The signals of these protons are well separated between the diastereomers for determining the D or L-configuration. 2. Kinetic resolution in the coupling reaction was small (within

TABLE II

^{1}H-N.m.r. and e.i.-m.s. data for tri-*O*-acetyl-D- and L-pentosyl 1-[(−)-2-*tert*-butyl-2-methyl-1,3-benzodioxole-4-carboxylates][a]

Compounds		*Chemical shifts (δ p.p.m.)*			*Ring conformations*	*e.i.–m.s. (70 eV)*	
		tert-*Bu*	*Me*	*H-1* ($J_{H1,H2}$)	$^4C_1$:$^1C_4$[b]	M^+	$[M-57]^+$
8	β-D-Xyl*p*	1.088	1.584	5.912(6.6)	73:27	494 (23)	437 (193)
	L-Xyl*p*	1.078	1.613	5.956(6.6)			
	Δ	+0.010	−0.029	−0.044			
9	α-D-Ara*p*	1.081	1.624	5.878(6.8)	24:76	494 (11)	437 (64)
	L-Ara*p*	1.097	1.595	5.849(7.0)			
	Δ	+0.016	+0.029	+0.029			
10	β-D-Rib*p*[c]	1.083	1.651	6.368(3.7)	29:71	494 (8)	437 (66)
	L-Rib*p*	1.072	1.624	6.339(3.7)			
	Δ	+0.011	+0.027	+0.029			
11	β-D-Rib*f*[c]	1.098	1.617	6.393(<0.3)	—	494 (8)	437 (66)
	L-Rib*f*	1.092	1.639	6.420(<0.3)			
	Δ	+0.006	−0.022	−0.022			

[a] See footnote [a] of Table I.
[b] Calculated from the equation: $^4C_1(\%) = (100 \times J_{H1,H2} - 180)/6.6$; where anti-diaxial and anti-diequatorial couplings were assumed to be 8.4 Hz and 1.8 Hz, respectively. [c] As a mixture of pyranoside and furanoside.

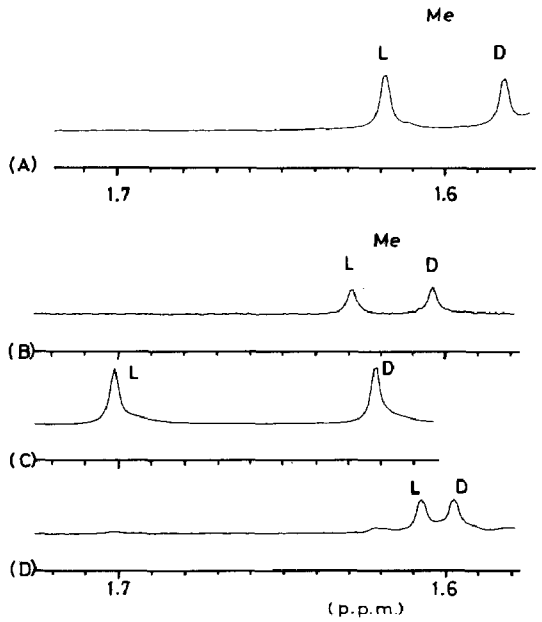


Fig. 1. ^{1}H-N.m.r. signals of the Me group in the TBMB-carboxylate moiety of per-*O*-acetylglycopyranosyl-D-1-[(±)-TBMB-carboxylates: A = Glc*p* (1), B = Gal*p* (2), C = α Man*p* (3), and D = β Man*p* (4). The resonances for the D-sugar 1-[(−)-TBMB-carboxylates] are labelled D, and those for D-sugar 1-[(+)-TBMB-carboxylates are labelled L, as they are equivalent to those of the enantiomeric L-sugar 1-[(−)-TBMB-carboxylates.

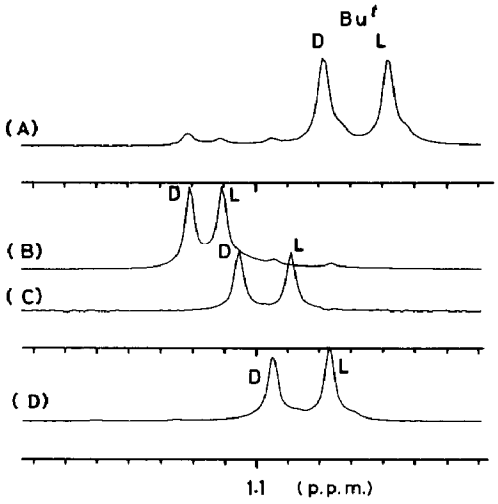


Fig. 2. ^{1}H-N.m.r. signals of the *tert*-butyl group in the TBMB-carboxylate moiety of per-*O*-acetyl-D-glycopyranosyl 1-[(±)-TBMB carboxylates: A = β Man*p* (**4**), B = α Man*p* (**3**), C = Gal*p* (**2**), and D = Glc*p* (**1**). The D,D assignments are as in Fig. 1.

3% *e.e.*), and therefore, the relative ^{1}H-n.m.r. peak-areas of each of the diastereomers may be used to estimate the D/L ratio if the sugar exists as a D,L mixture. 3. The analysis may be performed even with a mixture of sugars, as the *tert*-Bu or Me peaks have different chemical shifts specific for each sugar. However, care should be exercised in that yields from the derivatization may vary extensively among monosaccharides, and some of the sugars (mannose, rhamnose, and ribose) yield mixtures of isomers.

The results for all monosaccharides studied here are compiled in Tables I and II, together with the chemical-shift differences of the *tert*-Bu, Me, and H-1 signals between the diastereomers. In Tables III–V, n.m.r. data for ring protons are summarized. The magnitude of the chemical-shift differences [$\Delta(\delta_D - \delta_L)$] between diastereomers is in the approximate order of H-1 ≧ Me > *tert*-Bu >. In some cases, separation was also observed for the other ring signals, such as H-2 and H-3. The large separation of H-2 and H-3 in compounds **3** and **6** is particularly noteworthy. This result may be rationalized by the fact that the C-1–O-CO-TBMB group in **3** and **6** has the 1,2-*syn* and 1,3-*syn*-diaxial relationship with H-2 and H-3, respectively.

The foregoing results show that comparison of the n.m.r. spectrum with those of authentic samples of known configuration gives a simple method for determining the D,L configurations of monosaccharides. It would be convenient if the signs (+ or −) of the chemical-shift differences of the *tert*-Bu, Me, or H-1 signals were correlated with the absolute configurations of sugars. However, the signs of [$\Delta(\delta_D - \delta_L)$] do not seem to be correlated directly with the D,L configurations of sugars. This is because the position of derivatization (C-1) is remote from the C-5 or C-4 position which defines the enantiomeric identities of hexoses or pentoses, respectively. Instead, the signs of the chemical-shift differences of the *tert*-Bu, Me and H-1 signals correlate well with the absolute stereochemistries around the anomeric position (Figs. 3 and Table VI).

TABLE III

^{1}H-N.m.r. data (sugar portions) of 1-*O*-[(−)-TBMB carbonyl]-per-*O*-acetyl-D- and L-hexopyranosyl 1-[(−)-2-*tert*-butyl-2-methyl-1,3-benzodioxole-4-carboxylates][a]

	Compound	*Chemical shifts/coupling constants (p.p.m. Hz)*										
		H-1	*H-2*	*H-3*	*H-4*	*H-5*	*H-6*proR	*H-6*proS[b]	*Ac(1)*	*Ac(2)*	*Ac(3)*	*Ac(4)*
1	β-D-Clc*p*[b]	5.898 m	~ 5.31 m	~ 5.31 m	~ 5.2 m	3.914 m	4.337 (4.7)	4.143 (2.1,12.6)	1.996	2.031	2.048	2.083
	L-Glc*p*	5.937 m	~ 5.31 m	~ 5.31 m	~ 5.2 m	3.914 m	4.326 (4.6)	4.143 (2.1,12.6)	1.988	2.031	2.048	2.080
	Δ	−0.39	0.00	0.00	0.00	0.000	0.011	0.000	0.008	0.000	0.000	0.003
2	β-D-Gal*p*	5.878 (8.4)	5.488 (10.6)	5.133 (3.3)	5.459	~ 4.18 m	~ 4.1 m	~ 4.1 m	2.176	2.044	2.006	1.994
	L-Gal*p*	5.902 (8.4)	5.488 (10.6)	5.137 (3.3)	5.459	~ 4.18 m	~ 4.1 m	~ 4.1 m	2.184	2.044	2.006	1.985
	Δ	0.024	0.00	0.00	0.00	0.0	0.0	0.0	−0.008	0.000	0.000	0.009
3	α-D-Man*p*	6.390 (1.8)	5.383 (3.7)	5.513 (10.3)	~ 5.4 (10.3)	~ 4.26	~ 4.29	~ 4.13	2.209	2.077	2.032	1.991
	L-Man*p*	6.405 (1.8)	5.342 (3.7)	5.611 (10.3)	~ 5.4 (10.3)	~ 4.25	~ 4.31	~ 4.11	2.206	2.080	2.019	1.999
	Δ	−0.015	0.041	−0.098	0.0	0.01	−0.02	0.02	0.003	−0.003	0.013	−0.008
4	β-D-Man*p*	6.073 (1.1)	5.608 (3.9)	5.181 (9.9)	5.338 (9.8)	3.85	4.324 (5.3,12.3)	4.179 (2.2)	2.244	2.092	2.066	2.019
	L-Man*p*	6.045 (1.1)	5.600 (3.9)	5.181 (9.9)	5.338 (9.8)	3.85	4.350 (5.3,12.0)	4.184 (2.2)	2.236	2.092	2.066	2.019
	Δ	0.028	0.008	0.000	0.000	0.00	−0.026	−0.005	−0.008	0.000	0.000	0.000

[a] Measured at 400 MHz in $CDCl_3$ solution with Me_4Si as standard. [b,c] Assignments of the H-6*proR* and H-6*proS* signals were based on the ^{1}H-n.m.r. studies on per-*O*-acetyl D-glucopyransides stereospecifically deuterated at the C-6 position[12].

TABLE IV

^{1}H-N.m.r. data (sugar portions) of per-*O*-acetyl-6-deoxy-D- and L-hexopyranosyl 1-[(−)-2-*tert*-butyl-2-methyl-1,3-benzodioxole-4-carboxylates][a]

Compound		*Chemical shifts/coupling constants (p.p.m./Hz)*								
		H-1	*H-2*	*H-3*	*H-4*	*H-5*	*6-Me*	*Ac(1)*	*Ac(2)*	*Ac(3)*
5	β-D-Fuc*p*	5.841	5.480	5.130	5.306	4.020	1.240	2.208	2.173	2.099
		(8.4)	(10.3)	(3.5)	(0.7)	m	(6.2)			
	L-Fuc*p*	5.875	5.480	5.122	5.306	4.027	1.248	2.216	2.173	2.046
		(8.4)	(10.3)	(3.5)	(0.7)	m	(6.2)			
	Δ	−0.034	0.000	0.008	0.000	−0.007	−0.008	−0.008	0.000	0.053
6	α-D-Rha*p*	6.312	5.379	5.468	5.162	4.141	1.251	2.198	2.044	1.990
		(1.8)	(3.5)	(10.4)	(9.9)	m	(6.2)			
	L-Rha*p*	6.322	5.344	5.568	5.171	4.141	1.251	2.198	2.033	1.997
		(1.8)	(3.5)	(10.4)	(9.9)	m	(6.2)			
	Δ	−0.010	0.035	−0.100	−0.009	0.000	0.000	0.000	0.007	−0.007
7	β-D-Rha*p*	6.030	5.599	~5.13	~5.13	3.72	1.310	2.245	2.077	2.015
		(1.1)	(b s)			m	(6.2)			
	L-Rha*p*	6.017	5.599	~5.13	~5.13	3.72	1.323	2.237	2.077	2.012
		(1.1)	(b s)			m	(6.2)			
	Δ	0.013	0.000	0.0	0.0	0.0	−0.013	0.008	0.000	0.003

[a] Measured at 400 MHz in $CDCl_3$ solution; Me_4Si.

TABLE V

^{1}H-n.m.r. data for tri-*O*-acetyl-D- and L-pentosyl 1-[(−)-*tert*-butyl-2-methyl-1,3-benzodioxole-4-carboxylates][a]

Compound		*Chemical shifts/coupling constants (p.p.m./Hz)*								
		H-1	*H-2*	*H-3*	*H-4*	*H-5*eq	*H-5*ax	*Ac(1)*	*Ac(2)*	*Ac(3)*
8	β-D-Xyl*p*	5.912 (6.6)	5.196 (8.2)	5.267 (8.2)	5.05 m	4.225 (5.2,12.1)	3.598 (8.4)	2.077	2.062	2.031
	L-Xyl*p*	5.956 (6.6)	5.196 (8.2)	5.267 (8.2)	5.045 m	4.240 (5.2,12.1)	3.612 (8.4)	2.077	2.062	2.027
	Δ	−0.044	0.000	0.000	0.000	−0.015	−0.014	0.000	0.000	0.004
9	α-D-Ara*p*	5.878 (6.8)	5.432 (8.8)	5.194 (3.7,8.8)	5.340 m	4.121 (4.0,13.2)	3.846 (2.0)	2.158	2.064	2.042
	L-Ara*p*	5.849 (7.0)	5.438 (9.2)	5.181 (3.7,8.8)	5.340 m	4.110 (3.9,13.2)	3.838 (2.0)	2.158	2.064	2.046
	Δ	0.029	−0.006	0.013	0.00	0.011	0.008	0.000	0.000	−0.004
10	β-D-Rib*p*	6.368 (3.7)	~5.21 (3.7)	5.578 (3.3)	~5.21	4.237 (2.6,12.8)	3.985 (3.8)	2.057	2.142	2.140
	L-Rib*p*	6.339 (3.7)	~5.21 (3.7)	5.534 (3.3)	~5.23	4.181 (2.6,12.8)	3.968 (3.8)	2.061	2.157	2.154
	Δ	0.029	0.0	0.044	−0.02	0.056	0.017	−0.004	−0.015	−0.014
							H-5a	H-5b		
11	β-D-Rib*f*	6.393 (<0.3)	~5.48	~5.48	4.434 m	4.35 (4.2,12.3)	4.146 (6.0)	1.941	2.082	2.103
	L-Rib*f*	6.420 (<0.3)	~5.48	~5.48	4.434 m	4.35 (4.1,12.3)	---[b]	1.952	2.082	2.103
	Δ	−0.027	0.0	0.0	0.0	0.0	---[b]	−0.011	0.000	0.000

[a] Measured at 400 MHz in $CDCl_3$ solution. Me_4Si standard. [b] Not estimated because of the overlapping of signals.

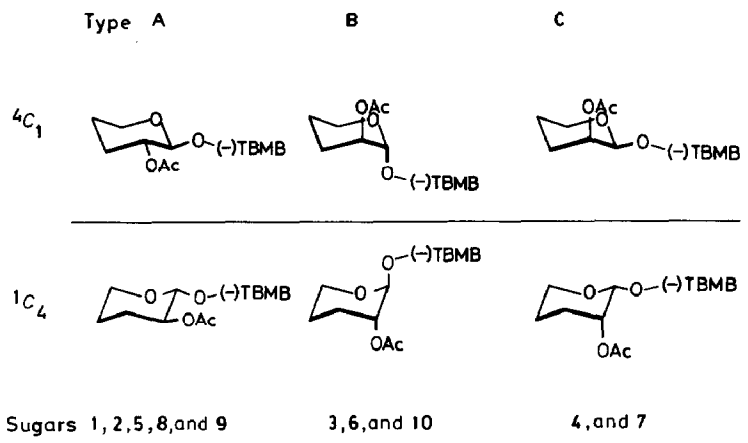


Fig. 3. Three types of sugar **A**, **B**, and **C** and classification of sugar derivatives **1–10**.

TABLE VI

The signs of the chemical-shift differences $\Delta[\delta(^4C_1 - {}^1C_4)]$ of the *tert*-butyl group, methyl group, and anomeric proton between the D-and L-sugar 1-[(−)-tert-butyl-2-methyl-1,3-benzodioxole-4-carboxylates] of the three types **A**, **B**, and **C**

	Signs of $\delta(^4C_1 - {}^1C_4)$		
	A	**B**	**C**
tert-Bu	+	+,−	+
Me	−	−	−
H-1	−	−	+

The glycosyl 1-TBMB-carboxylate derivatives may be classified into three types **A**, **B**, and **C** from their stereochemistries at C-1 and C-2, as depicted in Fig. 3. Each type involves a pair of diastereomers in rings having the $^4C_1$ and $^1C_4$ conformations. Sugars (**1**, **2**, **5**, **8**, and **9**) in type **A** have *diequatorial* substituents at C-1 and C-2, sugars (**3**, **6**,**10**) in type **B** have *diaxial* substituents, and sugars (**4** and **7**) in type **C** have an *axial* substituent at C-2 and an *equatorial* one at C-1. Sugars in the same type group showed similar chemical-shift relations between the diastereomers ($^4C_1$ and $^1C_4$) for any of the *tert*-Bu, Me, and anomeric-proton signals. The typical patterns are summarized in Table VI, where it may be noted that the Me signal has a negative value for the difference, $\Delta[\delta(^4C_1) - \delta(^1C_4)]$, for each type of sugar derivative. The observed relations in Table VI will be useful for predict the D,L configurations of sugars without having recourse to authentic samples.

EXPERIMENTAL

Per-*O*-acetylated glycosyl bromides were obtained from the reducing sugars by standard methods[9]. A mixture of the reducing D-or L-sugar (~0.1 mmol) and catalytic amount of $HClO_4$ in Ac_2O (1 mL) was stirred for 15 min at room temperature. The solution was made neutral with $KHCO_3$, the mixture, filtered, and the filtrate concentrated repeatedly from EtoH-PhMe. The residue was treated with 25% HBr in HOAc (1 mL) for 30 min. The mixture was concentrated *in vacuo* repeatedly with benzene to give the per-*O*-acetylated glycosyl bromide as a crude syrup (~75–85% by t.l.c.) which was used in the next reaction with (−)- or (+)-TBMB-carboxylic acid without further purification.

The crude bromide (~0.1 mmol) was dissolved in dry *N,N*-dimethylformamide (2 mL) containing $KHCO_3$ powder (10 mg) and (−)- or (+)-TBMB-carboxylic acid (0.15 mmol). The mixture was kept for 1 h at 60°, diluted with EtOAc (2 mL), washed with aq. $NaHCO_3$ and dried over Na_2SO_4 T.l.c. on silica gel [Kieselgel 60 F_{254}. (Merck), 3:1 benzene EtOAc] showed a fluorescent spot at R_F ~0.6–0.7 under a u.v. lamp and a minor, non-fluorescent spot at R_F ~0.2–0.3. The fluorescent material was collected by preparative t.l.c. and used for the n.m.r. analyses.

ACKNOWLEDGEMENT

This research was supported by a Grant-in-Aid for Scientific Research from the Ministry of Education, Science and Culture of Japan (02856029) and also by the Research Development Corporation of Japan (JRDC).

REFERENCES

1 M. S. G. Pavano, R. M. Albano, and P. A. S. Mourao, *Carbohydr. Res.*, 189 (1989) 374–379; D. J. Bell and D. E. Baldwin, *J. Chem. Soc.*, (1941) 125–131; J. B. C. Correa, A. Dmytraczenko, J. H. Dwarte, *Carbohydr. Res.*, 3 (1967) 445–452; H. Bretting, N. F. Whittaker, E. A. Kabat, K. Konigsmann-Lange, J. Thiem, *Carbohydr. Res.*, 98 (1981) 213–236; E. E. Percival and R. A. J. Foyle, *Carbohydr. Res.*, 72 (1979) 165–176; D. G. Medcalf, J. R. Scott, J. H. Brannon, G. A. Hemerick, R. L. Cunningham, J. H. Chessen, and J. Shah, *Carbohydr. Res.*, 44 (1975) 87–96.

2 R. J. Sturgeon, *Methods Carbohydr. Res.*, 8 (1980) 131–133; J. Melrose and R. J. Sturgeon, *Carbohydr. Res.*, 118 (1983) 247–253; R. J. Sturgeon, *Carbohydr. Res.*, 200 (1990) 499–501.

3 G. J. Gerwig, J. P. Kamerling, and J. F. G. Vliegenthart, *Carbohydr. Res.*, 62 (1978) 349–357; G. J. Gerwig, J. P. Kamerling, and J. F. G. Vliegenthart, *Carbohydr. Res.*, 77 (1979) 1–7; K. Leontein, B. Lindberg, and J. Lonngren, *Carbohydr. Res.*, 62 (1978) 359–362.

4 W. A. Konig, P. M. Lubbecke, B. Brassat, and S. Litz, *Carbohydr. Res.*, 183 (1988) 11–17.

5 S. Tsukamoto, K. Kaneko, and K. Hayashi, *Chem. Pharm. Bull.*, 37 (1989) 637–641.

6 W. H. Pirkle and D. J. Hoover, *Top. Stereochem.*, 13 (1982) 263–331.

7 C. R. Johnson, R. C. Elliott, and T. D. Penning, *J. Am. Chem. Soc.*, 106 (1984) 5019–5020; R. E. Doolittle and R. R. Heath, *J. Org. Chem.*, 49 (1984) 5041–5050; D. Terunuma, M. Kato, M. Kamei, H. Uchida, and H. Nohira, *Chem. Lett.*, (1985), 13–14; B. L. Feringer, B. Strijveen, and R. M. Kollog, *J. Org. Chem.*, 51 (1986) 5484–5486; A. Nabeya and T. Endo, *J. Org. Chem.*, 53 (1988) 3358–3361.

8 Y. Nishida, H. Ohrui, and H. Meguro, *Tetrahedron Lett.*, 30 (1989) 5277–5280; Y. Nishida, H. Ohrui, C. Kabuto, and H. Meguro, *Anal. Sci.*, in press.

9 F. L. Humoller, *Methods Carbohydr. Chem.*, 1 (1962) 83–84; F. Weygand, *ibid.*, 1 (1962) 183–184; R. W. Jeanloz and P. J. Stoffyn, *ibid.*, 2 (1962) 224–225; R. U. Lemieux, *ibid.*, 2 (1963) 221–222.
10 H. Paulsen, *Angew. Chem. Int. Ed*, 21 (1982) 155–172; R. U. Lemieux, in K. J. Laidler (Ed.), *IUPAC frontiers of chemistry*, Pergamon Press, New York, 1982 pp. 3–24.
11 Y. Nishida, H. Hori, H. Ohrui, and H. Meguro, *J. Carbohydr. Chem.*, 7 (1988) 239–250, and related references therein.