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SYNTHESIS OF METHYL *O*-(3-DEOXY-3-FLUORO-β-D-GALACTOPYRANOSYL)-(1→6)-*O*-β-D-GALACTOPYRANOSYL-(1→6)-3-DEOXY-3-FLUORO-β-D-GALACTOPYRANOSIDE AND RELATED N.M.R. STUDIES*

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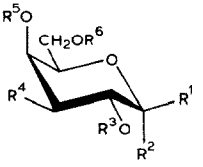
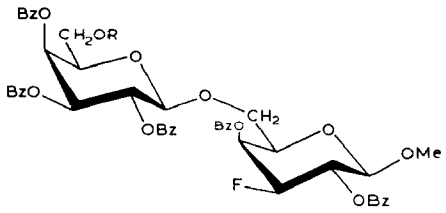
ABSTRACT

Sequential tritylation, benzoylation, and detritylation of methyl 3-deoxy-3-fluoro-β-D-galactopyranoside gave crystalline methyl 2,4-di-*O*-benzoyl-3-deoxy-3-fluoro-β-D-galactopyranoside (**9**), which was used as the initial nucleophile in the synthesis of the target oligosaccharide (**16**). Treatment of **9** with 2,3,4-tri-*O*-benzoyl-6-*O*-bromoacetyl-α-D-galactopyranosyl bromide gave the corresponding di-

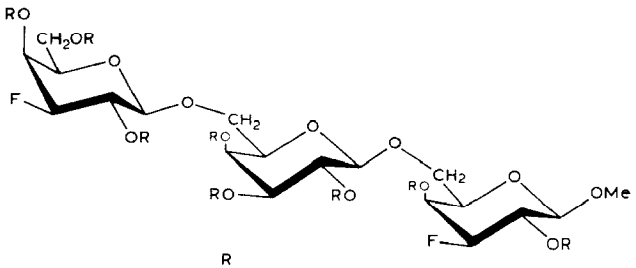


	R^1	R^2	R^3	R^4	R^5	R^6
1	H	OBz	Bz	OBz	Bz	Tr
2	OBz	H	Bz	OBz	Bz	Tr
3	OBz	H	Bz	OBz	Bz	H
4	OBz	H	Bz	OBz	Bz	$COCH_2Br$
5	H	Br	Bz	OBz	Bz	$COCH_2Br$
6	OMe	H	H	F	H	H
7	OMe	H	H	F	H	Tr
8	OMe	H	Bz	F	Bz	Tr
9	OMe	H	Bz	F	Bz	H
10	H	OBz	Bz	F	Bz	Bz
11	OBz	H	Bz	F	Bz	Bz
12	H	Br	Bz	F	Bz	Bz

*Synthesis of Specifically Fluorinated Methyl β-Glycosides of (1→6)-β-D-Galacto-oligosaccharides, Part V. For Part IV, see ref. 1.



	R
13	$COCH_2Br$
14	H



	R
15	Bz
16	H

saccharide derivative **13**, having a selectively removable blocking group at O-6′. Debromoacetylation of **13** afforded the disaccharide nucleophile **14** which, when treated with 2,4,6-tri-*O*-benzoyl-3-deoxy-3-fluoro-α-D-galactopyranosyl bromide, gave the fully protected trisaccharide **15**. Debenzoylation of **15** gave the title glycoside **16**. Condensation reactions were performed with silver trifluoromethanesulfonate as a promoter in the presence of *sym*-collidine under base-deficient conditions, and gave excellent yields of the desired *β(trans)*-products. Analyses of the ^1H- and ^{13}C-n.m.r. spectra, as well as determination of the J_{CF} and J_{HF} coupling constants, were made by using various one- and two-dimensional n.m.r. techniques.

INTRODUCTION

This laboratory has reported on the binding of (1→6)-β-D-galactan-specific monoclonal antibodies with a number of ligands. These ligands included (1→6)-β-D-galacto-oligosaccharides, some of which had HO-3 of the galactopyranosyl group replaced by fluorine[2]. That study permitted specific assignments of details of binding of the antigen at the antibody combining-site. In order to verify the proposed modes of binding, several additional, specifically fluorinated oligosaccharides have been prepared[1,3] and are currently being studied for their binding properties. Here we report the preparation of the methyl β-glycoside of (1→6)-β-D-galactotriose having a 3-deoxy-3-fluoro-D-galactopyranosyl group as well as a 3-deoxy-3-fluoro-D-galactoside residue.

RESULTS AND DISCUSSION

Synthesis. — We have previously described the syntheses of a series of homologous methyl β-glycosides of (1→6)-β-D-galacto-oligosaccharides having a 3-deoxy-3-fluoro-D-galactopyranosyl group[4] or a 3-deoxy-3-fluoro-D-galactopyranoside residue[1]. Extension of the oligosaccharide chain was achieved by using 2,3,4-tri-*O*-acetyl-6-*O*-chloroacetyl-α-D-galactopyranosyl bromide[5] or the corresponding 6-*O*-bromoacetyl analog[1] as the glycosyl donor. The latter reagent was an improvement over the 6-*O*-chloroacetyl analog in that *O*-debromoacetylation could be performed under milder conditions than for *O*-dechloroacetylation. Side reactions during this process[4] were minimal. However, formation of by-products during glycosylations[1,4] especially when silver triflate was used as a promotor[4], could not be overcome when acetylated glycosyl halides and nucleophiles were used. Garegg *et al.*[6] have rationalized the course of possible side-reactions accompanying triflate-promoted glycosylation reactions involving acetylated glycosyl halides. They found[7] that these may be minimized in the D-galactose series by the use of benzoylated, instead of acetylated, glycosyl donors. To meet these requirements, the nucleophile **9** and two new D-galactopyranosyl halides, **5** and **12**, have been now prepared and used in a stepwise synthesis of the title trisaccharide **16**.

The initial nucleophile **9** required by the present approach to the synthesis of **16** was prepared by sequential tritylation, benzoylation, and detritylation of **6** (ref. 10). In contrast to the corresponding acetyl derivative[1], acyl migration was not observed during detritylation of **8** with iodotrimethylsilane. The desired, crystalline compound **9** was obtained in excellent yield, and its structure was confirmed by ^{13}C and ^{1}H-n.m.r. spectroscopy (Tables I and II).

To obtain the glycosyl halide **5**, a compound of general usefulness in the synthesis of 6-*O*-substituted D-galactopyranosyl derivatives, we have followed a reaction sequence analogous to that leading to the corresponding *O*-acetyl derivative[8]. By conducting benzoylation of 6-*O*-trityl-D-galactose initially at −60°, the formation of variable proportions of furanose structures commonly encountered when acylation of D-galactose and some of its derivatives is performed at room or elevated temperatures (compare, for example, refs. 9 and 10) was largely minimized. Chromatographic resolution of the resulting, crude product yielded pure benzoates **1** (~30%) and **2** (~64%) in excellent combined yield. The formation of a large proportion of the β anomer **2** was in contrast to benzoylation of, for instance, D-galactose[11], where the proportion of β anomer was <10%, or of 3-deoxy-3-fluoro-D-galactose (see later). For preparative convenience, the amorphous β-benzoate **2** formed in higher yield was used in subsequent conversion to the glycosyl halide **5**, despite the fact that the α anomer **1** could be obtained crystalline. The reactions **2**→**5** were straightforward and high yielding, and the products in each step were fully characterized, and their structures substantiated by n.m.r.-spectral data (Tables I and II).

To avoid the formation of furanose structures during benzoylation of 3-

TABLE I

^{13}C-N M R CHEMICAL SHIFTS (δ) AND J_{CF} COUPLING CONSTANTS (Hz, IN PARENTHESES) FOR **1–12** IN $CDCl_3$[a]

Compound	*Carbon atom*								
	C-1	*C-2*	*C-3*	*C-4*	*C-5*	*C-6*	*CH_2Br*	*CPh_3*	*Me*
1[b]	90.73	67.97	68.78	68.50	70.56	60.92		87.21	
2[c]	93.08	67.74	71.69	68.91	73.44	60.43		86.99	
3[b]	93.24	69.02	71.81	68.71	75.21	60.53			
4[c]	92.99	68.72	71.59	68.03	72.18	63.21	25.39		
5[b]	88.00	68.32	68.71	67.83	71.41	62.80	25.08		
6[c]	103.17	69.51	93.20	66.93	73.90	60.71			57.38
	(11.0)	(19.5)	(183.1)	(17.1)	(7.3)	(3.7)			
7[c]	103.49	69.81	93.27	67.59	72.32	62.28		86.89	56.97
	(11.0)	(18.3)	(185.5)	(17.1)	(7.3)				
8[c]	101.64	70.81	89.59	67.76	71.52	60.72		86.94	
	(11.0)	(19.5)	(194.1)	(15.9)	(6.1)				
9[b]	101.80	70.72	89.04	68.68	72.88	60.05			57.13
	(10.5)	(19.2)	(195.2)	(16.1)	(4.5)	(1.8)			
10[b]	90.73	68.17	86.21	68.48	69.26	61.74			
	(9.2)	(17.4)	(194.1)	(18.9)	(4.9)				
11[b]	92.23	69.54	88.86	67.56	71.44	61.66			
	(10.1)	(20.3)	(195.6)	(16.5)	(5.5)	(1.3)			
12[b]	87.75	69.13	86.44	67.71	71.72	61.51			
	(8.7)	(19.7)	(193.7)	(17.1)	(5.1)				

[a]Spectrum of **6** taken in D_2O (internal standard MeOH). [b]Taken at 75.45 MHz; assignment confirmed by 2D heteronuclear proton–carbon correlation experiments. [c]Taken at 25.16 MHz.

deoxy-3-fluoro-D-galactose, a procedure similar to that used for the benzoylation of 6-*O*-trityl-D-galactose was followed. The crystalline products (**10** and **11**) showed n.m.r. data (Tables I and II) consistent with the suggested structures. When treated with hydrobromic acid in acetic acid, both **10** and **11** could readily be converted into the same glycosyl halide (**12**).

Treatment of the glycosyl halide **5** with the nucleophile **9**, with silver trifluoromethanesulfonate as the promotor in the presence of less than an equimolar proportion of *sym*-collidine, gave a straightforward reaction and, after purification of the crude product by chromatography, the crystalline disaccharide derivative **13** having a selectively removable blocking group at O-6′ was isolated in ~92% yield. Subsequent treatment of **13** with thiourea readily regenerated HO-6′ to afford the nucleophile **14**. Its reaction with the glycosyl halide **12** was as straightforward as the glycosylation leading to **13**, and the desired trisaccharide **15** was obtained in 93% yield, double that reported for the corresponding acetylated halides[10]. Debenzoylation of **15** under standard conditions produced the title trisaccharide glycoside **16**, isolated as a crystalline monohydrate.

N.m.r. spectroscopy. — Assignment of the ^{1}H and ^{13}C spectra for the monosaccharides **1–12** was first made routinely from the respective one-dimensional (1-D) spectra. First-order analysis of the coupling patterns in the ^{1}H-n.m.r.

TABLE II

^{1}H-N M.R DATA FOR **1–5** AND **9–12** IN $CDCl_3$

Compound	*Chemical shift (δ)*							*Coupling constant (Hz)*						
	H-1	*H-2*	*H-3*	*H-4*	*H-5*	*H-6a*	*H-6b*	$J_{1,2}$	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$	$J_{5,6a}$	$J_{5,6b}$	$J_{6a,6b}$
1[a]	6.868d	5.922dd	6.126dd	6.283d	4.418t	3.514dd	3.374t	3.7	10.8	3.2	[c]	5.5	9.0	9.0
2[b]	6.20b	5.98dd	5.80dd	6.22d	3.55dd	4.31dd	3.30t	8.0	10.0	3.5	[c]	5.5	9.0	9.0
3[a]	6.360dd	6.181dd	5.876dd	6.038d	4.391t	3.923m	3.756m	8.3	10.3	3.4	[c]	5.7	5.7	11.1
4[b]	6.31d	6.11dd	5.81dd	6.03d	4.59 ——		4.35m	8.0	10.5	3.5	[c]	[d]	[d]	[d]
5[a]	6.967d	5.674dd	6.031m	6.048m	4.827t	—— 4.408d ——		3.9	10.0	3.3	[c]	6.3	6.3	[e]
9[a]	4.593dd	5.730m	4.931ddd	5.783dd	3.833m	3.825m	3.630m	7.9	9.8	3.7	[c]	[d]	[d]	11.6
10[a]	6.921t	5.941dt	5.440ddd	6.213dd	4.691t	4.597ddd	4.422dd	3.8	10.2	3.8	[c]	6.5	6.5	12.7
11[a]	6.209d	6.073m	5.160ddd	6.139dd	4.464m	4.654m	4.482m	8.3	9.6	3.8	[c]	[d]	[d]	12.2
12[a]	6.944t	5.610dt	5.356ddd	6.150t	4.770t	4.602dd	4.481dd	4.2	10.0	3.6	[c]	6.7	5.8	11.6

[a]Spectrum taken at 300 MHz; proton signal signal-assignment effected by one-dimensional ^{1}H spectroscopy and 2-D ^{1}H–^{13}C shift-correlation experiment. [b]Spectrum taken 220 MHz; data based on first-order analysis of the spectrum. [c]<1 Hz. [d]Not determined because of overlapping of signals. [e]The H-6 protons are equivalent.

spectra yielded the J_{HH} values (Table II). The assignment was even more straightforward for the fluorinated derivatives because of the characteristic J_{CF} and J_{HF} values (Tables I and also Table V) shown by signals for carbon atoms and protons at various positions on the sugar ring. For most of these monosaccharides, a ^{13}C–^{1}H chemical-shift correlation map (CSCM)[12] was then generated for unambiguous confirmation of the assignment. For the disaccharide **13** and the trisaccharides **15** and **16**, assignment of the proton spectra was generally based on the ^{13}C–^{1}H CSCM, which in most instances gave unambiguous assignments. For **15** and **16**, containing two fluorinated rings having identical substituents, differentiation between the resonances of the corresponding carbon atoms and protons on the two rings cannot be done with absolute certainty because the corresponding proton chemical-shifts usually differ by <0.1 p.p.m. and $\delta(^{13}C)$ by <0.5 p.p.m. (Table III). Proton COSY[13] was, therefore, used in these instances to establish the sequencing and, consequently, to effect assignment of the corresponding resonances to the unprimed and double-primed rings (ring C being the galactoside residue and C″ the galactosyl group). As the $\delta(^{1}H)$ values of protons 6 and 6″ may be readily distinguished from the ^{13}C–^{1}H CSCM (the chemical shift of C-6 is ~68 p.p.m. and that of C-6″ 61 p.p.m.), assignment of the other resonances follows once the connectivity is established. A minor uncertainty in the proton-signal assignments constitutes protons H-2 and H-2″, where the difference in chemical shifts is the smallest. The assignment of signals at δ 4.442 and 4.736 to H-1 and H-1″ in the spectrum of **15** (Table IV) was based on comparison with the chemical shift for H-1 found in the spectrum of the closely related disaccharide derivative **13**. Likewise, assignment of the signals at δ 4.318 and 4.433 in the spectrum of **16** to H-1 and H-1″ was based on the chemical shift for H-1 (δ 4.319) found[4] in the spectrum of the methyl β-glycoside of a (1→6)-β-D-galactotriose containing a 3-deoxy-3-fluoro-D-galactopyranosyl residue. After the ^{1}H assignment for the trisaccharides had been established, most of the $J_{H,H}$ coupling constants for **15** were extracted from the 1-D spectrum.

By the ^{13}C–^{1}H chemical-shift correlation technique used in this study almost complete homonuclear decoupling in the ^{1}H dimension may be achieved[12]. It provides a unique and simple way to measure the magnitude and the relative sign of the ^{1}H–X (X is any nucleus other than ^{1}H and ^{13}C having spin 1/2) coupling constants, even in molecules having unresolvable ^{1}H spectra[12]. In this study, the couplings of ^{19}F with all protons on the same sugar ring (except H-6) have been determined by this method and are summarized in Table V. The magnitudes are generally in excellent agreement with previous work on simple fluorinated monosaccharides[14,15] and oligosaccharides[12]. The J_{HF} value between the equatorially oriented fluorine atom at C-3 and the ring protons are quite different from coupling constants observed when the fluorine is axially disposed[16]. The $^{2}J_{HF}$ couplings range from 46–49 Hz and have signs opposite to those of the corresponding $^{1}J_{CF}$. As the latter is known[12] to be negative, $^{2}J_{HF}$ is therefore absolutely positive. The α anomers usually show slightly larger $^{2}J_{HF}$ values than do the β anomers. The two $^{3}J_{HF}$ couplings, namely

TABLE III

^{13}C-N.M.R. CHEMICAL SHIFTS (δ) AND J_{CF} COUPLING CONSTANTS (Hz, IN PARENTHESES) FOR **13–15** (IN $CDCl_3$) AND **16** (IN D_2O)

Carbon atoms	*Compound*			
	13[a]	**14**[b]	**15**[a,c]	**16**[a,d]
C-1	101.32 (10.7)	101.39 (11.0)	101.50 (10.7)	103.17 (11.9)
C-2	70.54 (19.3)	70.63 (18.3)	70.9[e] (19.1)	69.58[e] (18.4)
C-3	88.97 (194.4)	89.02 (194.0)	89.12 (194.4)	92.98 (183.8)
C-4	68.12 (17.3)	68.14 (16.5)	68.34 (15.9)	66.97 (16.7)
C-5	71.97 (5.1)	71.85 (5.5)	71.86 (5.1)	72.57 (7.5)
C-6	67.86	67.69	67.82	68.88 (3.8)
C-1′	101.13	101.11	101.09	103.48
C-2′	69.47	69.92	69.81	70.80
C-3′	71.35	71.65	71.45	72.71
C-4′	67.97	68.69	68.31	68.76
C-5′	70.69	73.98	73.22	73.89
C-6′	63.21	60.29	66.64	69.45
C-1″			100.02 (10.8)	102.77 (12.3)
C-2″			70.75[e] (19.2)	69.45[e] (18.8)
C-3″			88.95 (193.4)	93.10 (183.7)
C-4″			67.,82 (16.4)	66.88 (16.6)
C-5″			70.37 (5.1)	73.93 (7.3)
C-6″			61.58	60.77 (3.6)
Me	56.62	56.65	56.72	57.57
CH_2Br	25.24			

[a]Spectra taken at 75.45 MHz; assignment confirmed by 2D heteronuclear ^{13}C–^1H correlation experiment. [b]Spectrum taken at 75.45 MHz; assignment made by comparison with spectral data of model compounds (**3, 4, 9,** and **13**). [c]Assignment of C-1 and C-1″ based on comparison with data for model compounds (**13** and **14**). [d]Assignment of C-1 and C-1″ based on comparison with two series of related oligosaccharides[1,4]. [e]Assignments for C-2 and C-2″ may be reversed.

$^3J_{F\text{-}3,H\text{-}2}$ and $^3J_{F\text{-}3,H\text{-}4}$, have rather different values and probably reflect different dihedral-angular relationships than those described[17] by the Karplus equation for $^3J_{HF}$. The correlation map showed[12] that both $^3J_{HF}$ values are of the same sign as the corresponding $^2J_{CF}$. As $^3J_{HF}$ values are known[18,19] to be mostly positive in saccharides, the $^2J_{CF}$ couplings are presumed also to be positive.

Longer-range J_{HF} were observed also in di- and tri-saccharides. The magnitude and signs of $^4J_{HF}$ were observed to be very stereospecific in the saccharides studied. Whereas the $^4J_{HF}$ values for Feq–Hax are rather small (usually <1 Hz), those for Feq–Heq ($^4J_{H\text{-}1,F\text{-}3}$ in 10 and 12) are significantly larger (~3 Hz). This observation is similar to that made previously[18]. Also worth noting is that, for the β anomers, $^4J_{H\text{-}1,F\text{-}3}$ values are of opposite sign as compared to the corresponding $^3J_{CF}$, whereas they are of the same sign in the α anomers. Therefore $^4J_{H\text{-}1,F\text{-}3}$ values for α and β anomers apparently do not have the same sign. The relative signs of $^4J_{HF}$ and the corresponding $^3J_{CF}$ value for other positions have also been established (Table V). As there was only scattered information[18] on the absolute sign of $^4J_{HF}$,

TABLE IV

^{1}H-N M R DATA FOR COMPOUNDS **13** AND **15** (IN $CDCl_3$), AND **16** (IN D_2O)

Proton	**13**		**15**		**16**	
	δ(p.p.m.)	$J_{H,H}$ *(Hz)*	*δ(p.p.m.)*	$J_{H,H}$ *(Hz)*	*δ(p.p.m.)*	$J_{H,H}$ *(Hz)*
H-1	4.452	$J_{1,2}$ 8.0	4.442	$J_{1,2}$ 8.0	4.318	8.0
H-2	5.632	$J_{2,3}$ 10.6	5.637[a]	$J_{2,3}$ 9.9	3.763[a]	[b]
H-3	4.869	$J_{3,4}$ 3.8	4.791	$J_{3,4}$ 3.8	4.533	[b]
H-4	5.921	$J_{4,5}$ [c]	5.861	$J_{4,5}$ [c]	4.191	[b]
H-5	4.003	$J_{5,6}$ 4.1, 7.2	3.969	$J_{5,6}$ [b]	3.848	[b]
H-6	4.195, 3.838	$J_{6a,6b}$ 10.3	4.150, 3.766	$J_{6a,6b}$ 10.5	4.006, 3.892	$J_{6a,6b}$ 10.7
OCH_3	3.208		3.201		3.531	
H-1′	4.893	$J_{1',2'}$ 7 9	4.758	$J_{1',2'}$ 7.8	4.412	[b]
H-2′	5.815	$J_{2',3'}$ 10.4	5.756	$J_{2',3'}$ 10.4	3.477	[b]
H-3′	5.611	$J_{3',4'}$ 3.4	5.528	$J_{3',4'}$ 3.4	3.591	[b]
H-4′	5.902	$J_{4',5'}$ [c]	5.907	$J_{4',5'}$ [c]	3.892	[b]
H-5′	4.232	$J_{5',5''}$ [b]	4.150	$J_{5',6'}$ [b], 6.6	3.850	[b]
H-6′	4.234	[d]	3.974, 3.778	$J_{6a,6b}$ 11.2	4.003, 3.874	$J_{6'a,6'b}$ 12.2
H-1″			4.736	$J_{1'',2''}$ 7.8	4.433	$J_{1'',2''}$ 8.0
H-2″			5.630[a]	$J_{2'',3''}$ 9.9	3.731[a]	[b]
H-3″			5.019	$J_{3'',4''}$ 3.9	4.519	[b]
H-4″			5.986	$J_{4'',5''}$ [c]	4.146	[b]
H-5″			4.111	$J_{5'',6''}$ 6.4, 6.5	3.635	[b]
H-6″			4.372, 4.218	$J_{6''a,6''b}$ 11.2	3.730	[d]

[a]Assignments for H-2 and H-2″ may be reversed. [b]Not determined because of overlapping of signals. [c]<1 Hz. [d]The two protons are equivalent.

TABLE V

THE $J_{^1H,^{19}F}$ COUPLING CONSTANTS (IN Hz) IN THE SPECTRA OF 3-DEOXY-3-FLUORO-D-GALACTOPYRANOSE DERIVATIVES **9–13**, AND **15** (IN $CDCl_3$), AND **16** (IN D_2O)

Compound	$^{19}F,^1H$-1	$^{19}F,^1H$-2	$^{19}F,^1H$-3	$^{19}F,^1H$-4	$^{19}F,^1H$-5
9	0.5[a]	12.3[b]	+47.4[c]	5.3[b]	1.6[b]
10[d]	2.7[b]	10.5[b]	+48.5[c]	5.1[b]	1.8[c]
11	0.9[c]	11.2[b]	+47.3[c]	5.8[b]	0.7[c]
12	3.1[b]	10.3[b]	+47.6[c]	5.2[b]	1.0[c]
13	1.0[c]	10.6[b]	+47.4[c]	5.5[b]	[e]
15	0.2[c], 0.9[c]	11.0[b], 11.9[b]	+48.6[c], +47.2[c]	3.5[b], 4.2[b]	1.0[c], 0.7[c]
16	0.5[c], 0.5[c]	13.0[b], 13.0[b]	+48.4[c], +47.7[c]	7.2[b], 6.6[b]	0.8[c], 0.9[c]

[a]Relative sign unknown. [b]Coupling is of the same sign as the corresponding ^{13}C–^{19}F coupling. [c]Coupling is of the opposite sign as the corresponding ^{13}C–^{19}F coupling; for absolute sign see text. [d]$^5J_{HF}$ = 0.8 Hz between fluorine and the H-6 proton resonating at 4.597 p.p.m. was observed in the 1-D ^1H spectrum; relative sign unknown. [e]Not observed.

and virtually no experimental evidence[20] on the sign of $^3J_{CF}$ (theoretical calculations predict that $^3J_{CF}$ is positive)[21], the present relative-sign determination by itself does not provide information on the absolute sign of these couplings. However, the information on the relative sign provides an important link between these parameters which will ultimately help determine their absolute signs.

EXPERIMENTAL

General methods. — Melting points were determined with a Büchi melting-point apparatus. Optical rotations were measured at 25° with a Perkin–Elmer automatic polarimeter, Model 241 MC. Thin-layer chromatography (t.l.c.) on precoated plates of silica gel (Analtech) was performed with *A*, 20:1 carbon tetrachloride–acetone; *B*, 10:1 carbon tetrachloride–acetone; *C*, 20:1 toluene–acetone; and *D*, 6:1 toluene–acetone. Detection was effected by charring with 5% (v/v) sulfuric acid in ethanol and, where applicable, by u.v. light. Preparative chromatography was performed by gradient elution from slurry-packed columns of Silica Gel 60 (Merck, Prod. No. 9385, 0.04–0.063 mm, or Prod. No. 15111, 0.015–0.04 mm). For purification of compound **14**, the silica gel was deactivated with 5% of water.

^{13}C-N.m.r. spectra were routinely recorded at room temperature for solutions in $CDCl_3$ (internal standard Me_4Si) or D_2O (internal standard MeOH, δ_{MeOH} *vs.* Me_4Si, 49.0 p.p.m.) with Varian FX-100 and Varian FX-300 spectrometers, operated at 25.16 and 75.45 MHz, respectively. ^{1}H-N.m.r. spectra were taken under the same conditions with a Varian HR-220 spectrometer.

All ^{13}C–^{1}H chemical-shift correlation and ^{1}H–^{1}H chemical-shift correlation (COSY)[13] experiments were performed with a Nicolet NT-300 spectrometer (^{1}H frequency 300.05 MHz; ^{13}C frequency 75.45 MHz) with 5-mm probes. For ^{13}C–^{1}H chemical-shift correlation, a new version that utilizes selective spin-flip pulses, resulting in homonuclear decoupling in the ^{1}H dimension[12], was used. The ^{13}C and ^{1}H pulse-widths were 11 and 30 μs, respectively. 2-D Data sets of 128 × 2K or 128 × 4K were usually generated. For ^{1}H–^{1}H chemical-shift correlation, a standard COSY[13] experiment was used where the ^{1}H pulse-width was 6 μs.

The $\delta(^1H)$ values determined were usually accurate to 0.005 p.p.m., $\delta(^{13}C)$ to 0.01 p.p.m. Coupling constants determined from 1-D spectra are accurate to 0.2 Hz, J_{FH} and $^2J_{HH}$ determined from 2-D spectra are accurate to 0.5 and 1.0 Hz, respectively.

The solution of hydrobromic acid in acetic acid (33%, almost colorless liquid) was a product of Fluka, A.G. Acetonitrile (u.v. Spectral Grade) was purchased from Burdick and Jackson Laboratories, Muskegon, MI 49442, and used as supplied. Silver trifluoromethanesulfonate was purchased from Aldrich Chemical Co. Reactions involving this reagent, as well others requiring anhydrous conditions, were performed under argon, and reagents and solvents were handled with Hamilton, Series 1000 gas-tight syringes. Nitromethane was distilled from urea (50 g/L, twice), and then from phosphorus pentaoxide. Toluene was dried with sodium

hydride and distilled. Solutions in organic solvents were dried with anhydrous sodium sulfate and, unless otherwise stated, concentrated at 40°/2 kPa.

*1,2,3,4-Tetra-*O*-benzoyl-6-*O*-trityl-α-* (**1**) *and β-*D*-galactopyranose* (**2**). — A solution of 6-*O*-trityl-D-galactose[5] (2.77 g, 6.55 mmol) in pyridine (50 mL) was cooled to −60° and benzoyl chloride (10.75 mL, 91 mmol) was added with stirring. When the addition was complete, cooling was removed and, with continued stirring, the semisolid mixture was allowed to warm to room temperature, kept for 3 h, and then held overnight at 60°. T.l.c. (solvent *A*) showed that the reaction was complete and that two major (R_F 0.25 and 0.35) and two, very minor, charring, trityl-positive products (R_F 0.3 and 0.4) were formed. Water (100 mL) was added slowly while the mixture was stirred and cooled in an ice bath. When the excess of benzoyl chloride had hydrolyzed, the mixture was poured with stirring into an excess of aqueous sodium hydrogencarbonate. The precipitate was collected, dissolved in dichloromethane, and the resulting solution washed with water, dried, and evaporated. The residue was chromatographed to give the two major products. The material having R_F 0.35 was the α anomer **1** (1.6 g, 29.2%), m.p. 128–130° (from acetonitrile), $[\alpha]_D$ +147° (*c* 1.2, chloroform).

Anal. Calc. for $C_{53}H_{42}O_{10}$: C, 75.88; H, 5.04. Found: C, 75.94; H, 5.42.

The material having R_F 0.25 was the β anomer **2** (amorphous solid, 3.5 g, 63.8%), $[\alpha]_D$ +60.7° (*c* 1, chloroform).

Anal. Calc. for $C_{53}H_{42}O_{10}$: C, 75.88; H, 5.04. Found: C, 75.71; H, 4.84.

*1,2,3,4-Tetra-*O*-benzoyl-β-*D*-galactopyranose* (**3**). — A solution of **2** (1.5 g, 1.78 mmol) and sodium iodide (0.8 g, 5.3 mmol) in acetonitrile (17 mL) was cooled to 0° and chlorotrimethylsilane (0.68 mL, 5.3 mmol) was added under anhydrous conditions. The mixture was stirred for 3 min at 0°, cold (0°) water (50 mL) was added, and the mixture was then shaken with aqueous sodium thiosulfate solution. The colorless solution was extracted with dichloromethane, and the extract dried. T.l.c. showed that the reaction was complete and that one charring product (R_F 0.2, solvent *B*) was formed. The crude product was chromatographed to give pure **3** (1 g, ~93%), $[\alpha]_D$ +157° (*c* 1.1, chloroform).

Anal. Calc. for $C_{34}H_{28}O_{10}$: C, 68.44; H, 4.73. Found: C, 68.24; H, 4.98.

*1,2,3,4-Tetra-*O*-benzoyl-6-*O*-bromoacetyl-β-*D*-galactopyranose* (**4**). — 2,6-Lutidine (0.4 mL, 3.45 mmol), followed by bromoacetyl bromide (0.26 mL, 3 mmol), were added at −30° to a solution of **3** (1.4 g, 2.3 mmol) in dry dichloromethane (10 mL). Cooling was removed and, after 10 min, while the solution was still very cold, t.l.c. (solvent *B*) showed that reaction was complete and that one faster-moving product had been formed. Water (20 mL) was added and the mixture was stirred for 1 h at room temperature. It was then partitioned between water and dichloromethane. The organic solution was dried, evaporated, and the residue eluted from a short column of silica gel to remove some non-carbohydrate, u.v.-positive, base-line material. Pure **4** was obtained as a solid, colorless foam (1.55 g, 92%) having $[\alpha]_D$ +121.4° (*c* 0.86, chloroform).

Anal. Calc. for $C_{36}H_{29}BrO_4$: C, 60.25; H, 4.07. Found: C, 60.34; H, 4.20.

2,3,4-Tri-O-benzoyl-6-O-bromoacetyl-α-D-galactopyranosyl bromide (**5**). — A solution of hydrobromic acid in acetic acid (5 mL) was added to a solution of **4** (1.3 g) in dry dichloromethane (5 mL). After 40 min at room temperature, t.l.c. (solvent *C*) showed that all of the starting material had been converted into a faster-moving product. The solution was concentrated with concurrent evaporation of toluene, to remove the excess of reagent, and the solution of the residue in toluene was filtered through a layer (30 g) of silica gel, to remove some u.v.-positive, non-carbohydrate material. Pure **5** (1.2 g, 98%) was obtained as a colorless, glassy solid, $[\alpha]_D$ +297.5° (*c* 1.25, chloroform). When kept under dry conditions at −20°, the compound is stable (t.l.c.) for a period of at least 3 months.

Anal. Calc. for $C_{29}H_{24}Br_2O_9$: C, 51.49; H, 3.57; Br, 23.63. Found: C, 51.50; H, 3.39; Br, 23.93.

Methyl 2,4-di-O-benzoyl-3-deoxy-3-fluoro-β-D-galactopyranoside (**9**). — Chlorotriphenylmethane (1.56 g, 5.6 mmol) was added to **6** (1 g, 5.1 mmol) in dry pyridine (5 mL) and the mixture was stirred overnight at room temperature. T.l.c. (solvent *B*) showed that **6** was almost completely converted into the tritylated product **7**. Benzoyl chloride (3 mL, 25.5 mmol) was added dropwise. After 6 h at room temperature, t.l.c. (solvent *A*) showed the reaction to be practically complete. After conventional processing, the major trityl-positive product (R_F 0.6) was isolated by elution of the crude product from a column of silica gel to give pure, amorphous **8** (3.0 g, 93.7%), $[\alpha]_D$ +14.7° (*c* 1.3, chloroform).

Anal. Calc. for $C_{40}H_{35}FO_7$: C, 74.28; H, 5.45; F, 2.93. Found: C, 74.08; H, 5.75; F, 2.66.

A solution of the fully protected compound **8** (2.9 g, 4.64 mmol) and sodium iodide (2.09 g, 13.82 mmol) in acetonitrile (45 mL) was cooled to 0° and chlorotrimethylsilane (1.8 mL, 14.2 mmol) was added with stirring. After 3 min, the reaction was quenched, and the mixture processed as for the preparation of **3**. Pure **9** (1.6 g, 86.2%) was isolated by chromatography. Crystallization from ether–isopropyl ether (twice) gave material melting at 138–139°, $[\alpha]_D$ +108° (*c* 1, chloroform).

Anal. Calc. for $C_{21}H_{21}FO_7$: C, 62.36; H, 5.23; F, 4.69. Found: C, 62.47; H, 5.51; F, 4.93.

1,2,4,6-Tetra-O-benzoyl-3-deoxy-3-fluoro-α- (**10**) *and β-D-galactopyranose* (**11**). — Benzoyl chloride (12.9 mL, 110 mmol) was added dropwise to a solution of 3-deoxy-3-fluoro-D-galactose (2 g, 11 mmol) in pyridine (22 mL) while the mixture was kept below −50°. Cooling was removed and, with occasional shaking, the mixture was allowed to warm to room temperature and then left overnight. T.l.c. (solvent *A*) showed the reaction to be complete and that two major products (R_F 0.4 and 0.3) were formed. The mixture was processed conventionally and the crude product was chromatographed to give first the α-benzoate **10** (4.85 g, 73.9%), m.p. 158–158.5° (from dichloromethane–methanol), $[\alpha]_D$ +122° (*c* 0.7, chloroform).

Anal. Calc. for $C_{34}H_{27}FO_9$: C, 68.22; H, 4.54; F, 3.17. Found: C, 67.97; H, 4.51; F, 3.38.

Eluted next was the β-benzoate **11** (1.6 g, 24.3%), m.p. 211–212° (from dichloromethane–methanol), $[\alpha]_D$ +27° (*c* 2.2, chloroform).

Anal. Calc. for $C_{34}H_{27}FO_9$: C, 68.22; H, 4.54; F, 3.17. Found: C, 68.03; H, 4.61; F, 3.21.

Methyl O-(2,3,4-tri-O-benzoyl-6-O-bromoacetyl-β-D-galactopyranosyl)-(1→6)-2,4-di-O-benzoyl-3-deoxy-3-fluoro-β-D-galactopyranoside (**13**). — A solution of the nucleophile **9** (0.65 g, 1.6 mmol) and the glycosyl halide **5** (1.195 g, 1.76 mmol) in toluene–nitromethane (1:1, 6 mL) was concentrated under diminished pressure and anhydrous conditions to approximately one-half of the original volume. The solution was cooled to −25° and dropwise, with stirring, a solution (5 mL) of silver triflate (2 mmol) and *sym*-collidine (0.18 mL, 1.36 mmol) was added. The mixture was stirred for at −25° while silver bromide precipitated. After 15 min, t.l.c. (solvent *B*) showed the reaction to be complete and that the mixture, which was acidic to litmus, contained essentially one component only (R_F 0.4, compare 0.2 and 0.8 for the starting materials **9** and **5**, respectively). *sym*-Collidine was added to neutralize triflic acid and, after dilution with dichloromethane, the mixture was filtered and the filtrate washed with aqueous sodium thiosulfate. The organic phase was dried and evaporated, and the residue was chromatographed to give pure **13** (1.47 g, 91.9%) which readily crystallized from carbon tetrachloride, m.p. 126–129°. A portion was dissolved in dichloromethane, carbon tetrachloride was added, and the disaccharide **13** crystallized after dichloromethane had been evaporated off; m.p. 125–127°, $[\alpha]_D$ +123.5° (*c* 0.8, chloroform).

Anal. Calc. for $C_{50}H_{44}BrFO_{16}$: C, 60.06; H, 4.43; Br, 7.99. Found: C, 59.86; H, 4.41; Br, 8.19.

Methyl O-(2,3,4-tri-O-benzoyl-β-D-galactopyranosyl)-(1→6)-2,4-di-O-benzoyl-3-deoxy-3-fluoro-β-D-galactopyranoside (**14**). — A solution of thiourea (456 mg, 6 mmol) in methanol (40 mL) was added to a solution of **13** (2 g, 2 mmol) in dichloromethane (20 mL). The mixture was stirred at room temperature for 20 min, whereupon t.l.c. showed only traces of unchanged starting-material (R_F 0.7). After evaporation, the solid residue was extracted with dichloromethane, and the organic solution was washed with water, dried, and evaporated. Elution of the residue from a column of silica gel afforded pure **14** (1.75 g, 84%), as a colorless foam, $[\alpha]_D$ +159° (*c* 1.1, chloroform).

Anal. Calc. for $C_{48}H_{43}FO_{15}$: C, 65.59; H, 4.93; F, 2.16. Found: C, 65.78; H, 5.12; F, 2.14.

Methyl O-(2,4,6-tri-O-benzoyl-3-deoxy-3-fluoro-β-D-galactopyranosyl)-(1→6)-O-(2,3,4-tri-O-benzoyl-β-D-galactopyranosyl)-(1→6)-2,4-di-O-benzoyl-3-deoxy-3-fluoro-β-D-galactopyranoside (**15**). — Compound **10** or **11** (1.5 g, 2.5 mmol) was treated with hydrobromic acid in acetic acid as described for the preparation of the non-fluorinated analog **5**. The crude product was purified by chromatography on 20 g of silica gel to give pure **12** (1.3 g, 93.2%) as a colorless foam that had $[\alpha]_D$ +154° (*c* 1.25, chloroform).

The disaccharide nucleophile **14** (1.5 g, 1.7 mmol) and the glycosyl halide **12**

(1.09 g, 2.12 mmol) were treated with silver triflate (0.55 g, 2.12 mmol) and *sym*-collidine (0.19 mL, 1.45 mmol) as described for the preparation of **13**. After processing as already described and purification of the crude product by chromatography, the trisaccharide **15** was obtained as a colorless foam (2.4 g, 93%), $[\alpha]_D$ +104.6° (*c* 1.2, chloroform).

Anal. Calc. for $C_{75}H_{64}BrFO_{22}$: C, 66.46; H, 4.76. Found: C, 66.40; H, 4.75.

Methyl O-(3-deoxy-3-fluoro-β-D-galactopyranosyl)-(1→6)-O-β-D-galactopyranosyl-(1→6)-3-deoxy-3-fluoro-β-D-galactopyranoside (**16**). — A solution of sodium methoxide (M) in methanol was added to a suspension of **15** (1.7 g) in methanol (80 mL) until the supernatant solution was strongly alkaline to litmus. The mixture was stirred overnight at room temperature, the clear solution was made neutral with Dowex 50 W (H^+-form) resin, filtered, and the resin washed with methanol. The combined filtrate was concentrated until a crystalline mass formed. After cooling, the crystals were filtered off and washed several times with methanol and ether to give a white, odorless product **16** (517 mg), m.p. 233–235°. The mother liquor was concentrated at 60°/133 Pa, to remove methyl benzoate, leaving a solid residue which, when crystallized from aqueous ethanol, gave a second crop of **16** (70 mg, total yield 89.7%). Recrystallization of a portion from aqueous ethanol gave material melting at 233–234°, $[\alpha]_D$ +14.2° (*c* 0.63, water).

Anal. Calc. for $C_{19}H_{32}F_2O_{14} \cdot H_2O$: C, 42.22; H, 6.34; F, 7.03. Found: C, 42.23; H, 6.55; F, 7.00.

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