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Preliminary communication

Glycosylation with Thioglycosides Activated by Dimethyl(methylthio)sulfonium Tetrafluoroborate: Synthesis of two Trisaccharide Glycosides Corresponding to the Blood Group A and B Determinants

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Two trisaccharide glycosides, *p*-trifluoroacetamidophenylethyl 3-*O*-(2-acetamido-2-deoxy-α-D-galactopyranosyl)-2-*O*-(α-L-fucopyranosyl)-β-D-galactopyranoside and *p*-trifluoroacetamidophenylethyl 2-*O*-(α-L-fucopyranosyl)-3-*O*-(α-D-galactopyranosyl)-β-D-galactopyranoside, corresponding to the human blood group A and B determinants, were synthesized. A key fucosylgalactosyl disaccharide derivative was glycosylated with galactosaminyl or galactosyl donors, respectively. Dimethyl (thiomethyl)sulfonium tetrafluoroborate was used for thioglycoside activation in coupling reactions.

The blood group A and B determinant trisaccharides have, in recent years, become increasingly demanded for biomedical purposes. These purposes include, e.g., production of monoclonal antibodies using trisaccharide-protein conjugates, or preparation of columns with immobilized carbohydrates to be used for specific adsorption of antibodies. Consequently, several chemical syntheses of the A or B trisaccharides (or derivatives of them) have been reported [1-13]. However, since some biomedical applications require substantial amounts of trisaccharide, improved synthetic procedures are still needed, especially such which use short synthetic paths, cheap reagents, and give rise to crystalline intermediates.

We have previously [14] reported a simple and efficient synthesis of the blood group B trisaccharide glycoside **10**, where the synthetic strategy was based on fucosylation of a digalactosyl derivative. This strategy gave satisfactory yields in all steps, and many intermediates were crystalline, which facilitated isolation. However, if feasible, the alternative synthetic strategy, encompassing glycosylation of a fucosylgalactosyl derivative, should represent a more rational approach if *both* the A and B trisaccharide derivatives are desired.

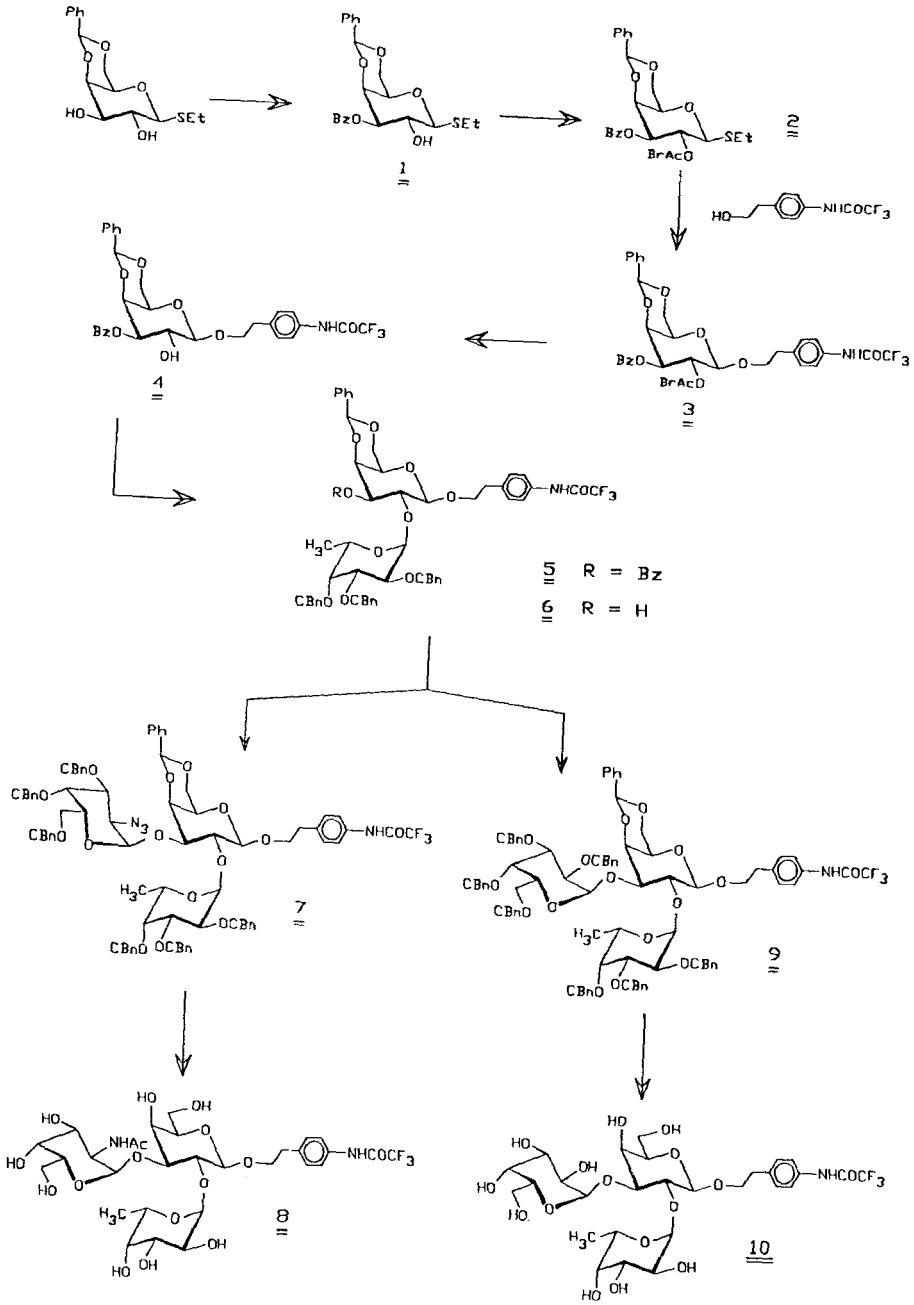

Table 1. NMR Data for compound **8** in deuterium oxide solution (acetone δ_H = 2.225, dioxane δ_C = 67.4)

^{1}H-NMR data:	H-1	H-2	H-3	H-4	H-5	H-6	H-6'
β-Gal	4.63	3.77	3.76	4.22	3.68	3.76	3.81
J(Hz)	$7.9_{1,2}$	$9.8_{2,3}$	$3.6_{3,4}$	ND[a]	ND	ND	ND
α-Fuc	5.23	3.64	3.33	3.14	3.78	0.97	-
J(Hz)	$4.1_{1,2}$	$10.4_{2,3}$	$3.3_{3,4}$	$<1.0_{4,5}$	-	$6.6_{5,6}$	-
α-GalNAc	5.16	4.23	3.85	3.97	4.19	3.75	3.75
J(Hz)	$3.8_{1,2}$	$10.0_{2,3}$	$3.0_{3,4}$	ND	ND	ND	ND

^{13}C-NMR data:	C-1	C-2	C-3	C-4	C-5	C-6
β-Gal	101.6	72.5	76.8	63.9	75.8	61.8
α-Fuc	99.0	68.5	70.4	72.4	67.5	15.9
α-GalNAc	92.2	50.3	68.6	69.3	71.9	62.2

$\mathbf{C}OCH_3$	175.6
$CO\mathbf{C}H_3$	22.8
O-$\mathbf{C}H_2$-CH_2	69.2
OCH_2-$\mathbf{C}H_2$-Ar	33.6
Ar	123.2, 129.9, 133.8, 138,5
$\mathbf{C}OCF_3$	157.8 (*J* 37 Hz)
$CO\mathbf{C}F_3$	116.8 (*J* 288 Hz)

[a] ND = not determined.

We now report on synthesis of the human blood group A and B trisaccharide derivatives **8** and **10**, respectively, using such an approach. Thioglycosides [15] and a recently described [16] glycosylation reagent, dimethyl(thiomethyl)sulfonium tetrafluoroborate (DMTSB) were used in most glycosylation reactions.

Results and Discussion

The starting material for the β-galactosyl unit was ethyl 4,6-*O*-benzylidene-1-thio-β-D-galactopyranoside [Nilsson SB, Lönn H, Norberg T, unpublished results] which on treatment with benzoyl chloride (1.1 equivalents) in pyridine gave the 3-*O*-benzoate **1** (83% yield, m.p. 123.5-125°C, $[\alpha]_D$ +55.4°). Treatment of **1** with bromoacetyl bromide and 2,4,6-trimethylpyridine/*N*,*N*-dimethylaminopyridine in dichloromethane gave **2** (90% yield, m.p.

126-127°C, $[\alpha]_D^{30}$ +79°). The thioethyl group in **2** was replaced by a *p*-trifluoroacetamidophenylethyl group by treatment with trifluoroacetamidophenylethanol [17] and DMTSB in dichloromethane-acetonitrile. The β-glycoside **3** was obtained (60% yield). The DMTSB reagent used here [16] is similar in performance to the widely used dimethyl(thiomethyl)sulfonium trifluoromethanesulfonate (DMTST) [15, 16, 18]. The advantage lies in its ease of preparation and handling. DMTSB is a relatively non-hygroscopic crystalline solid, easily prepared [19] from triethyloxonium tetrafluoroborate and dimethyl disulphide, whereas preparation of DMTST [16] require the use of the highly toxic methyl triflate.

The 2-bromoacetyl group in **3** was removed by treatment with pyridine-water to give **4** (70% yield, m.p. 224-225°C, $[\alpha]_D^{30}$ +79.2°). Fucosylation of **4** using ethyl 2,3,4-tri-*O*-*p*-chlorobenzyl-1-thio-β-L-fucopyranoside [20] and DMTSB in dichloromethane gave the disaccharide **5** (61% yield). In the NMR spectrum of **5**, signals from, *inter alia*, H-1 (δ 4.64, $J_{1,2}$ 7.8 Hz), H-2 (δ 4.40, $J_{2,3}$ 9.9 Hz), H-3 (δ 5.35, $J_{2,3}$ 9.9, $J_{3,4}$ 3.7 Hz) and H-1′ (δ 5.40, $J_{1,2}$ 3.7 Hz) were present, which verified the expected anomeric configuration and the 2-*O*-fucosyl-3-*O*-benzoyl substitution pattern.

Debenzoylation of **5** with sodium methoxide in methanol then gave the key fucosylgalactosyl disaccharide derivative **6** (77% yield, $[\alpha]_D^{30}$ -58°).

To obtain the A trisaccharide glycoside, glycosylation of compound **6** with methylphenyl 2-azido-3,4,6-tri-*O*-*p*-chlorobenzyl-2-deoxy-1-thio-β-D-galactopyranoside [21] or the corresponding glycosyl bromide was investigated using various promoters and reaction conditions. Use of DMTSB and the thioglycoside donor gave unsatisfactory yields. The use of silver triflate and the glycosyl bromide gave the best results. The α/β product ratio was increased in polar solvents. Thus, glycosylation of compound **6** with 2-azido-3,4,6-tri-*O*-*p*-chlorobenzyl-2-deoxy-1-thio-β-D-galactopyranosyl bromide in tetrahydrofuran:dioxane gave a 4:1 α/β mixture of trisaccharide derivatives (64% yield). The anomeric mixture was not resolved at this stage. Catalytic hydrogenation of the mixture (Pd/C) followed by *N*-acetylation with acetic anhydride then gave an α/β mixture of trisaccharide derivatives from which **8** could be isolated by silica gel chromatography (54% yield, $[\alpha]_D^{31}$ +28.5°, NMR data, see Table 1).

To obtain the B-trisaccharide glycoside, compound **6** was glycosylated with ethyl 2,3,4,6-tetra-*O*-*p*-chlorobenzyl-1-thio-β-D-galactopyranoside [Nilsson SB, Lönn H, Norberg T, unpublished results] in dichloromethane-tetrahydrofuran using DMTSB as promoter. Compound **9** was obtained (57% yield). Catalytic hydrogenation (Pd/C) of **9** then gave the target B-trisaccharide glycoside **10** (90% yield). Compounds **9** and **10** displayed physical data identical with those published [14].

In conclusion, the A and B trisaccharide glycosides **8** and **10** were synthesized from a common disaccharide intermediate **6** in good yields.

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