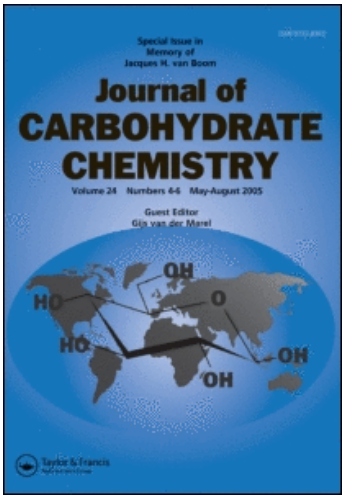


Journal of Carbohydrate Chemistry
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713617200

Synthetic Studies of the Cororubicin Oligosaccharide: Glycosylation of Branched Amino and Nitro Sugars

Lincoln Noecker[a]; Franco Duarte[a]; Robert M. Giuliano[a]
[a] Department of Chemistry, Villanova University, Villanova, PA

To cite this Article Noecker, Lincoln , Duarte, Franco and Giuliano, Robert M.(1998) 'Synthetic Studies of the Cororubicin Oligosaccharide: Glycosylation of Branched Amino and Nitro Sugars', Journal of Carbohydrate Chemistry, 17: 1, 39 — 48
To link to this Article: DOI: 10.1080/07328309808005767
URL: http://dx.doi.org/10.1080/07328309808005767

J. CARBOHYDRATE CHEMISTRY, 17(1), 39-48 (1998)

SYNTHETIC STUDIES OF THE CORORUBICIN OLIGOSACCHARIDE: GLYCOSYLATION OF BRANCHED AMINO AND NITRO SUGARS

Lincoln Noecker, Franco Duarte, and Robert M. Giuliano*

Department of Chemistry, Villanova University, Villanova, PA 19085

Received April 28, 1997 - Final Form October 16, 1997

ABSTRACT

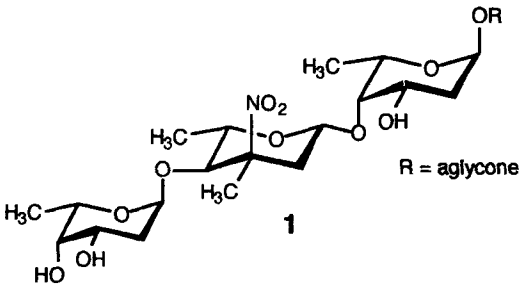
Derivatives of the branched amino sugar methyl 3-amino-2,3,6-trideoxy-3-*C*-methyl-α-L-*ribo*-hexopyranoside and its 3-nitro analog were coupled with glycosyl donors of 3,4-di-*O*-acetyl-2,6-dideoxy-L-fucopyranose. Successful glycosylations were developed using the fucosyl bromide, activated with silver triflate, and the fucosyl oxysilane, activated with trimethylsilyl triflate. High stereoselectivities for the desired α-1,4 linkage were observed in both cases. The *N*-trifluoroacetamido disaccharide was deacylated and the amino group oxidized to nitro with dimethyldioxirane. An alternate route based on coupling of the fucosyl bromide with the nitro sugar methyl α-L-decilonitroside also gave the α-linked disaccharide, which is related to oligosaccharides found in the antibiotics cororubicin and arugomycin.

INTRODUCTION

Cororubicin, arugomycin, and decilorubicin are complex antibiotics belonging to the anthracyclinone family.[1-3] All three possess an aglycone similar to that found in nogalamycin[4] and each occurs as a glycoside of an oligosaccharide that contains the branched nitro sugar decilonitrose[5] (2,3,6-trideoxy-3-*C*-methyl-3-nitro-L-*ribo*-hexopyranose). Decilonitrose is attached to two digitoxose (2,3,6-trideoxy-L-*ribo*-hexopyranose) residues in cororubicin by β-1,4 and α-1,4 linkages, resulting in trisaccharide **1**. While there has been considerable success in the synthesis of branched

nitro sugars,[6] the synthesis of oligosaccharides that contain them has remained a difficult problem owing to the lengthy syntheses of the precursor monosaccharides, the presence of 2-deoxy residues, and the sensitivity of the nitro group. In the first example of the synthesis of disaccharides containing nitro sugars, Baer and coworkers reported the coupling of a nitro sugar to a glycosyl bromide via the Koenigs-Knorr method.[7] A glycosyl donor for evernitrose, a branched nitro sugar found in the everninomicin antibiotics, has been recently developed by Scharf and Jütten, resulting in the first synthesis of a disaccharide containing one of the four naturally occurring nitro sugars.[8] Glycosylation of the branched-chain amino sugars constitutes an alternate route to the synthesis of oligosaccharides that contain nitro sugars, provided that efficient methods are available for the deprotection of the amino group and oxidation to nitro. The synthesis of oligosaccharides that contain the branched nitro sugar decilonitrose, or the related amino sugar, have not been described.

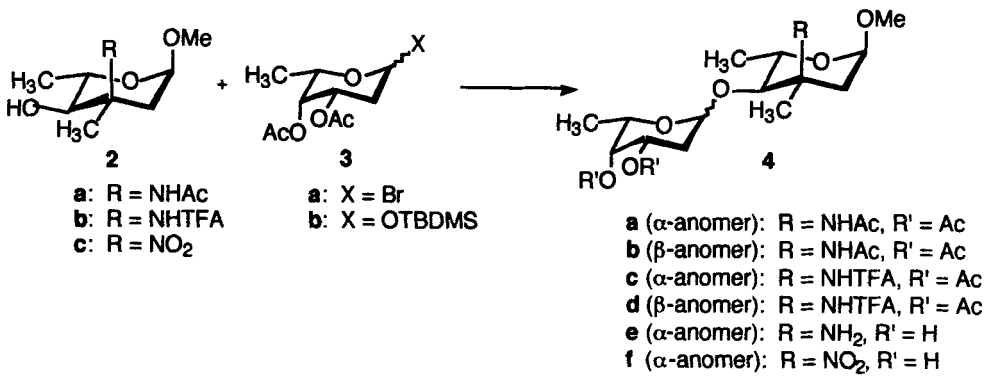
During the course of our studies of the synthesis of oligosaccharides that contain nitro sugars, we have developed two methods for the stereoselective glycosylation of the protected branched amino sugar methyl 2-trifluoroacetamido-2,3,6-trideoxy-3-*C*-methyl-α-L-*ribo*-hexopyranoside **2b** with 2-deoxyfucosyl donors. Both fucosyl bromides and glycosyl silanes were used successfully in this study, each giving the desired α-1,4-linked disaccharides in good yields and high stereoselectivity. Deblocking and oxidation of the amino group to nitro were carried out on the disaccharide, by hydrolysis with lithium hydroxide and oxidation with dimethyldioxirane. We have also been able to couple the corresponding branched nitro sugar **2c** directly to the 2-deoxyfucosyl bromide.



1

RESULTS AND DISCUSSION

Several syntheses of the nitro sugar L-decilonitrose have been reported.[6] For the purpose of this study, *N*-protected derivatives **2a**[9] and **2b**[10] of methyl 3-amino-2,3,6-



Scheme 1. Synthesis of Disaccharides of Branched Amino and Nitro Sugars

trideoxy-3-*C*-methyl-α-L-*ribo*-hexopyranoside[11] were prepared by selective protection of the amino group with acetic anhydride in methanol[12] or trifluoroacetic anhydride followed by treatment with guanidine,[13] in yields of 90% (**2a**) and 89% (**2b**). The f2-deoxyfucosyl bromide **3a** was prepared from di-*O*-acetyl-L-fucal[14] by treatment with HBr in benzene.[15] Di-*O*-acetyl-L-fucal was hydrated by treatment with Dowex resin and lithium bromide in acetonitrile-water,[16] and the resulting 2-deoxyfucopyranose was subsequently converted to 2-deoxyfucosyl silane **3b** by reaction with *t*-butyldimethyl-chlorosilane and imidazole.[17] The glycosyl bromide **3a** was prepared as needed and used immediately, while the glycosyl silane **3b** could be stored for prolonged periods without decomposition.

Coupling reactions of the branched sugar derivatives and glycosyl donors are shown in Scheme 1, and conditions, anomer ratios, and yields are given in Table 1. The first coupling reactions attempted were those of *N*-acetyl derivative **2a** and 2-deoxyfucosyl bromide **3a**. Using silver triflate[18] as the promoter and benzene as the solvent, a 2:3 ratio of α-linked to β-linked disaccharides was obtained in a combined yield of 69% after purification by column chromatography on Florisil. The mixture was not separable by this method, but most ^1H and ^{13}C NMR resonances were well resolved, and the structures were assigned without difficulty. The stereochemistry of the disaccharide linkage was assigned on the basis of the value observed for $J_{1,2'ax}$ on the 2-deoxyfucose residue (3.1 Hz in the α-anomer **4a**; 9.5 Hz in the β-anomer **4b**). In view of the resistance of the *N*-acetyl group toward removal in branched amino sugars of this type,[19] subsequent coupling reactions were carried out on *N*-trifluoroacetyl derivative **2b**. Coupling of **2b** with 2-deoxyfucosyl bromide **3a** gave the desired α-linked disaccharide in 83% isolated yield when carried out at low temperature. Unlike their *N*-

Table 1. Conditions for Synthesis of Disaccharides

acceptor	donor	conditions	product	α/β	%yield
2a	**3a**	AgOTf, 3A MS collidine, O °C - rt 18 h, benzene	**4a + 4b**	2:3	69
2b	**3a**	AgOTf, 3A MS collidine, O °C - rt 3 h, benzene	**4c + 4d**	30:1	70
2b	**3a**	AgOTf, 3A MS collidine, 18 h -78 - -10 °C dichloromethane	**4c**	α only	83
2b	**3b**	TMSOTf, 3A MS -50 - -20°C, 33 h dichloromethane	**4c + 4d**	40:1	56
2c	**3a**	AgOTf, 4A MS collidine, O °C - rt benzene, 18 h	**4f**	α only	20

acetyl counterparts, disaccharides **4c** and **4d** were separable by column chromatography on Florisil and only trace amounts of β-anomer **4d** were obtained. In considering a non-halogen donor for the fucal residue, we decided to attempt the coupling reaction using a glycosyl silane,[17] based on the successful application of this donor in the synthesis of anthracycline oligosaccharides, in which the acceptor is a derivative of an amino sugar (daunosamine), and high selectivity for α-glycosides was observed. Silylation of 2-deoxy-3,4-di-*O*-acetylfucopyranose in dichloromethane gave the β-glycosyl silane as reported. Coupling of trifluoroacetyl derivative **2b** with the 2-deoxyfucosyl silane (**3b**) in dichloromethane at -50 °C gave a 40:1 mixture of α- and β-linked disaccharides **4c** and **4d** in 56% yield along with recovered starting materials.

Deblocking of **4c** was not observed with sodium borohydride in ethanol, an established procedure[20] for cleaving trifluoroacetamido groups. However, treatment of **4c** with lithium hydroxide gave the completely deacetylated disaccharide **4e** in yields of 80-90%. Oxidation of the amino group to nitro in the disaccharide was carried out with dimethyldioxirane (DMDO)[21] in quantitative yield to give **4f**. We have found that DMDO oxidation is the most efficient method for this oxidation in branched amino sugars.[22] The production of excess benzoic acid that occurs when MCPBA is used is avoided, and chromatographic purification is unnecessary.

Coupling of nitro sugar[11] **2c** with 2-deoxyfucosyl donors was attempted under numerous conditions.[23] The best results were obtained with bromide **3a** and silver triflate as the promoter. Coupling reactions with **2c** were slower than those of the corresponding protected amino sugars, and isolation of the product proved more difficult because the unreacted nitro sugar (**2c**) co-eluted with the disaccharide. By careful experimentation, it was observed that treatment of the acetylated disaccharide with lithium hydroxide under mild conditions effected both deacetylation and epimerization of **2c** to a diastereomeric nitro sugarwith a different R_f value. Deacetylated disaccharide isolated from this mixture was identical with **4f** in all respects; however, the use of trifluoroacetamido derivative **2b** as an acceptor is technically less complicated as well as more efficient in terms of overall yield. These results are the first reported studies of the synthesis of oligosaccharides related to those found in the cororubicin and arugomycin. Further studies leading to the trisaccharide are in progress.

EXPERIMENTAL

General Methods. ^1H and ^{13}C NMR spectra were recorded on a Varian XL200 spectrometer at 200.06 and 50.3 MHz, respectively, in deuteriochloroform solution. Proton chemical shifts are relative to tetramethylsilane (0.00 ppm), and carbon chemical shifts are relative to deuteriochloroform (76.91 ppm). High resolution mass spectra were measured at the University of Pennsylvania under CI conditions using ammonia as the reagent gas. Flash chromatography [24] was performed on Merck silica gel 60 using mixtures of ethyl acetate and hexane as indicated. Visualization of TLC plates was carried out with ceric sulfate-ammonium molybdate in 2M sulfuric acid. Anhydrous DMF and THF were purchased as such from Aldrich Chemical Co. Dry benzene was purchased from Aldrich. Methanol was dried by distillation from magnesium. Dry dichloromethane was purchased from Aldrich and dried further by passing through a column of basic alumina (Woelm activity 1). Molecular sieves were dried overnight under vacuum at 130 °C.

Methyl 3-Acetamido-2,3,6-trideoxy-3-*C*-methyl-α-L-*ribo*-hexopyranoside (2a). Methyl 3-amino-2,3,6-trideoxy-3-*C*-methyl-α-L-*ribo*-hexopyranoside[11] (70 mg, 0.4 mmol) was dissolved in acetic anhydride (2.0 mL) and methanol (0.5 mL) and the reaction was stirred overnight at room temperature. Thin layer chromatographic analysis in ethyl acetate-hexane (2:3 v/v) showed the presence of a major component with R_f 0.18 (*N*-acetyl derivative) and a trace component with R_f 0.63 (presumably *N*,*O*-diacetyl derivative). Acetic anhydride and acetic acid were removed by azeotropic distillation with toluene under reduced pressure and the product was further dried under vacuum to give 0.078 g of *N*-acetyl

derivative **2a** which was used without further purification. The ^{1}H NMR spectrum of **2a** matched that reported for material prepared by another route.[9]

Methyl 2,3,6-Trideoxy-3-*C*-methyl-3-trifluoroacetamido-α-L-*ribo*-hexopyranoside (2b). Trifluoroacetic anhydride (0.29 mL, 0.436 g, 2.08 mmol) was added to a solution of the amino sugar (0.1654 g, 0.94 mmol) in dry dichloromethane (3 mL) at 0 °C with stirring under nitrogen. After 1 h, additional TFAA (0.1 mL) was added and the starting material was consumed after a total of 2.5 h, as evidenced by TLC with ethyl acetate-hexane (1:3 v/v). Dry ethanol (2 mL) was added and the reaction was stirred for 15 min, after which a solution of guanidine in 1:1 ethanol-dichloromethane (3.0 mL, 0.48M) was added. The cooling bath was removed and additional guanidine solution (0.5 mL) was added and stirring was continued for 1 h. The reaction mixture was transferred to a separatory funnel containing dichloromethane (20 mL) and water (10 mL) and the mixture was swirled. The layers were separated and the aqueous layer was extracted with dichloromethane (10 mL) and the combined organic extracts were dried (Na_2SO_4) and concentrated to give 0.231 g (91%) of **2b** that matched that reported.[10]

Methyl 4-*O*-(3,4-Di-*O*-acetyl-2,6-dideoxy-α,β-L-*lyxo*-hexopyranosyl) 3-acetamido-2,3,6-trideoxy-3-*C*-methyl-α-L-*ribo*-hexopyranoside (4a, 4b). A solution of *N*-acetyl derivative **2a** (50 mg, 0.23 mmol), freshly prepared glycosyl bromide **3a** (82 mg, 0.28 mmol) and *s*-collidine (0.34 g, 0.28 mmol) in dry benzene (3 mL) was added to a mixture of silver triflate (0.086 g, 0.34 mmol) and crushed 3Å molecular sieves (0.15 g) in dry benzene (2 mL) with stirring under nitrogen at 0 °C. The addition was carried out over a period of 10 min, and stirring was continued at 0 °C for 40 min, after which the ice bath was removed, and the reaction was allowed to warm to room temperature and stirred for 18 h. The reaction mixture was filtered and the cake was rinsed with dichloromethane (5 mL). The filtrate was concentrated under reduced pressure to give a crude syrup which was determined to contain a 2:3 mixture of **4a** and **4b** by ^{1}H NMR spectroscopy. Purification by flash chromatography in ethyl acetate-hexane (1:1 v/v) gave a mixture of disaccharides with R_f = 0.10 (1:1 ethyl acetate-hexane), R_f = 0.43 (ethyl acetate); yield, 68 mg (69%): ^{1}H NMR (**4a**, 300 MHz) δ 6.37 (bs, 1H, NH), 5.28 (d, 1H, $J_{1',2'ax}$ = 3.1 Hz, H-1'), 5.22 (ddd, 1H, $J_{2',3'}$ = 11.9 Hz, $J_{2e',3'}$ = 5.6 Hz, $J_{3',4'}$ = 3.1 Hz, H-3'), 5.20 (m, 1H, $J_{2'eq,4'}$ = 0.7 Hz, H-4'), 4.64 (dd, 1H, $J_{1,2ax}$ = 3.5 Hz, $J_{1,2eq}$ = 1.1 Hz, H-1), 4.16 (m, 1H, $J_{5'6'}$ = 6.6 Hz, H-5'), 3.82 (dq, 1H, $J_{5,6}$ = 6.3 Hz, $J_{4,5}$ = 9.5 Hz, H-5), 3.34 (s, 3H, OCH_3), 3.21 (d, 1H, $J_{4,5}$ = 9.5 Hz, H-4), 2.54 (dd, 1H, $J_{2ax,2eq}$ = 14.7 Hz, H-2eq), 2.16 (s, 3H, $OCOCH_3$), 2.14-2.05 (m, 2H, H-2ax', H-2eq'), 1.99 (s, 3H, $OCOCH_3$), 1.93 (s, 3H, $OCOCH_3$), 1.59 (dd, 1H, $J_{2ax,2eq}$ = 14.6 Hz, H-2ax), 1.58 (s, 3H, 3-CH_3), 1.29 (d, 3H, H-6), 1.13 (d, 3H, H-6'); ^{13}C NMR δ 170.3, 169.6 ($O\underline{C}OCH_3$), 99.7 (C-1'), 97.6 (C-1), 84.8 (C-4), 69.3 (C-4'), 66.3 (C-3'), 65.4 (C-5'),

63.0 (C-5), 55.3 (C-3), 54.9 (OCH_3), 39.0 (C-2), 29.1 (C-2'), 25.1 (3-CH_3), 20.7, 20.4 ($OCO\underline{C}H_3$), 18.2 (C-6), 16.1 (C-6'); ^1H NMR (**4b**, 300 MHz) δ 5.99 (bs, 1H, NH), 5.09 (dt, 1H, $J_{3',4'}$ = 3.1 Hz, $J_{4',5'}$ = $J_{2'eq,4'}$ = 0.9 Hz, H-4'), 4.98 (ddd, $J_{3',4'}$ = 3.1 Hz, $J_{2'ax,3'}$ = 11.9 Hz, $J_{2'eq,3'}$ = 5.2 Hz, H-3'), 4.61 (dd, 1H, $J_{1,2ax}$ = 4.5 Hz, $J_{1,2eq}$ = 2.0 Hz, H-1), 4.57 (dd, 1H, $J_{1',2'ax}$ = 9.5 Hz, $J_{1',2'eq}$ = 2.4 Hz, H-1'), 3.91 (dq, 1H, $J_{5,6}$ = 6.3 Hz, $J_{1,5}$ = 0.6 Hz, H-5), 3.66 (m, 1H, $J_{5'6'}$ = 6.4Hz, H-5'), 3.29 (s, 3H, OCH_3), 3.18 (d, 1H, $J_{4,5}$ = 9.5 Hz, H-4), 3.13 (dd, 1H, $J_{2ax,2eq}$ = 14.7 Hz, H-2eq), 2.17 (s, 3H, $OCOCH_3$), 2.01 (s, 3H, $OCOCH_3$), 1.93 (s, 3H, $OCOCH_3$), 2.00 (dddd, 1H, $J_{2ax',2eq'}$ = 12.3 Hz, $J_{2'eq,3'}$ = 5.2 Hz, $J_{1',2'eq}$ = 2.4 Hz, $J_{2'eq,4'}$ = 0.9 Hz, H-2eq), 1.89 (m, 1H, $J_{2'ax,3'}$ = 12.4 Hz, $J_{1',2'ax}$ = 9.5 Hz, H-2'ax), 1.73 (dd, 1H, $J_{2ax,2ee}$ = 14.5 Hz, H-2ax), 1.48 (s, 3H, 3-CH_3), 1.26 (d, 3H, H-6), 1.18 (d, 3H, H-6'); ^{13}C NMR δ 170.3, 169.6 ($O\underline{C}OCH_3$), 100.3 (C-1'), 98.1 (C-1), 85.0 (C-4), 69.2 (C-5'), 68.6 (C-3'), 68.0 (C-4'), 63.6 (C-5), 53.7 (C-3), 54.9 (OCH_3), 35.6 (C-2), 31.5 (C-2'), 24.7 (3-CH_3), 20.7, 20.4 ($OCO\underline{C}H_3$), 18.2 (C-6), 16.1 (C-6'). HRMS Calcd for $C_{20}H_{34}NO_9$ $(M+H)^+$: 432.2233. Found: 432.2314.

Methyl 4-*O*-(3,4-Di-*O*-acetyl-2,6-dideoxy-α-L-*lyxo*-hexopyranosyl)-2,3,6-trideoxy-3-*C*-methyl-3-trifluoroacetamido-α-L-*ribo*-hexopyranoside (4c). Method A: A solution of *N*-trifluoroacetyl derivative **2b** (94 mg, 0.35 mmol) and *s*-collidine (54 mg, 0.45 mmol) in dry dichloromethane (3 mL) was added to a mixture of silver triflate (0.122 g, 0.47 mmol) and powdered 3Å molecular sieves (0.2 g) in dry dichloromethane (3 mL) with stirring under nitrogen at -78 °C (dry ice-acetone). The addition was carried out over a period of 2 min, after which a solution of crude glycosyl bromide **3a** (freshly prepared from 97 mg (0.45 mmol) of 3,4-di-*O*-acetyl-L-fucal) in 3 mL dry benzene; the volume of benzene was reduced by 2/3 by evaporation after bromination) was diluted with 2 mL dichloromethane and added over a period of 10 min while maintaining the bath temperature at or below -75 °C. Stirring was continued for 5 h, after which the reaction flask was stoppered securely and placed in a freezer at -15 °C overnight. for 18 h. The reaction mixture warmed to room temperature and filtered through a pad of Celite. The pad was rinsed with dichloromethane (5 mL) followed by ethyl acetate (10 mL), and the filtrate concentrated under reduced pressure to give syrupy product which was purified by flash chromatography with ethyl acetate-hexane (20:80 to 40:60 v/v) to give 141 mg (83%) of disaccharide **4c**: R_f = 0.4 (40:60 ethyl acetate-hexane); $[\alpha]_D^{25}$ -134° (*c*, 0.7, chloroform); ^1H NMR δ 7.94 (bs, 1H, NH), 5.29 (ddd, ^1H, $J_{1',2'ax}$ = 4.0 Hz, $J_{1',2'eq}$ = 1.2 Hz, $J_{1',5'}$ = 0.6 Hz, H-1'), 5.20 (m, 1H, H-4'), 5.16 (m, $J_{3',4'}$ = 2.9 Hz, H-3'), 4.71 (ddd, 1H, $J_{1,2ax}$ = 2.9 Hz, $J_{1,2eq}$ = 1.2 Hz, $J_{1,5}$ = 0.6 Hz, H-1), 4.14 (m, 1H, $J_{5'6'}$ = 6.7 Hz, $J_{1'5'}$ = 0.6 Hz, H-5'), 3.76 (m, 1H, $J_{5,6}$ = 6.2 Hz, $J_{1,5}$ = 0.6 Hz, H-5), 3.38 (s, 3H, OCH_3), 3.27 (d, 1H,

$J_{4,5}$ = 9.5 Hz, H-4), 2.24 (m, 1H, $J_{2ax,2eq}$ = 14.7 Hz, H-2eq), 2.17 (ddt, 1H, $J_{2'3'}$ = 6.5 Hz, $J_{3'4'}$ = 2.9 Hz, $J_{2'eq,4'}$ = 1.2 Hz, $J_{2'ax,2'eq}$ = 12.6 Hz, H-2'eq), 2.15 (s, 3H, $OCOCH_3$), 2.05 (td, 1H, $J_{2'ax,3'}$ = 11.9 Hz, H-2'ax), 1.99 (s, 3H, $OCOCH_3$), 1.72 (dd, 1H, $J_{2ax,2eq}$ = 14.6 Hz, H-2ax), 1.69(s, 3H, 3-CH_3), 1.32 (d, 3H, H-6), 1.13 (d, 3H, H-6'); ^{13}C NMR δ 170.3, 170.0 ($O\underline{C}OCH_3$), 99.9 (C-1'), 97.2 (C-1), 84.0 (C-4), 69.4 (C-4'), 66.2 (C-3'), 65.7 (C-5'), 63.4 (C-5), 57.0 (C-3), 54.9 (OCH_3), 40.3 (C-2), 28.9 (C-2'), 24.3 (3-CH_3), 20.8, 20.6 ($OCO\underline{C}H_3$), 18.3 (C-6), 16.3 (C-6').

Anal. Calcd for $C_{20}H_{30}F_3NO_9$: C, 49.48; H, 6.23; H, 2.88. Found: C, 49.59; H, 6.32; N, 2.85.

When the reaction of **2b** and **3a** was conducted in benzene at 0 °C, the ratio of **4c** to **4d** obtained was 30:1; β-anomer (**4d**): ^1H NMR δ 7.15 (bs, 1H, NH), 5.08 (d, 1H, $J_{3'4'}$ = 3.2 Hz, H-4'), 4.97 (ddd, 1H, $J_{2'ax,3'}$ = 12.2 Hz, $J_{3',4'}$ = 3.2 Hz, H-3'), 4.65 (dd, 1H, $J_{1,2ax}$ = 4.2 Hz, $J_{1,2eq}$ = 2.3 Hz, H-1), 4.58 (dd, 1H, $J_{1,2'ax}$ = 9.0 Hz, $J_{1,2'eq}$ = 2.9 Hz, H-1'), 3.84 (dd, 1H, $J_{5,6}$ = 6.4 Hz, $J_{1,5}$ = 0.6 Hz, H-5), 3.65 (dd, 1H, $J_{4',5'}$ = 1.2 Hz, H-5'), 3.30 (s, 3H, OCH_3), 3.28 (d, 1H, $J_{4,5}$ = 9.6 Hz, H-4), 2.82 (dd, 1H, H-2eq), 2.20-1.74 (m, 2H, H-2'ax, H-2'eq), 2.17 (s, 3H, $OCOCH_3$), 2.01 (s, 3H, $OCOCH_3$), 1.84 (dd, 1H, $J_{2ax,2eq}$ = 14.8 Hz), 1.58 (s, 3H, 3-CH_3), 1.28 (d, 3H, H-6), 1.17 (d, 3H, $J_{5',6'}$ = 6.4 Hz, H-6'). Insufficient **4d** was available for determinations of elemental analysis or optical rotation.

Method B: A mixture of **2b** (102 mg, 0.375 mmol), 2-deoxyfucosyl oxysilane **3b** (130 mg, 0.375 mmol) and powdered 4Å molecular sieves (0.2 g) in dry dichloromethane (6 mL) was stirred under nitrogen and cooled to -50 °C. Trimethylsilyl triflate (22 μL, 0.025 g, 0.3 eq) was added via syringe and the reaction was carefully monitored by TLC using ethyl acetate-hexane (1:1). Additional TMSOTf (36 μL) was added in two portions over the next 10 h during which the reaction temperature was maintained below -40 °C. After 12 h, the reaction was sealed and stored at -12 °C for an additional 13 h. TMSOTf (88 μL) was added at -25 °C in three portions over the next 8 h bringing the total to 2.0 equivalents. Triethylamine (100 μL) was added and the reaction was diluted with dichloromethane (5 mL), filtered through a Celite pad which was washed with citrate buffer (pH 5, 10 mL) then H_2O (10 mL), and dried (Na_2SO_4). Concentration under reduced pressure gave syrupy product which was purified by column chromatography on Florisil with ethyl acetate-hexane (12:88 then 25:75) to give **4c** (66 mg, 56%). Trace amounts of β-anomer **4d** eluted in a more polar chromatographic fraction. The ratio of **4c** to **4d** is estimated to be 40:1 by NMR and found to be consistent with isolated yields of **4d** (29 mg **4c** to 1 mg **4d** in an earlier preparation.) Starting materials **2b** and **3b** were recovered in yields of 14% and 25%.

Methyl 4-*O*-(2,6-Dideoxy-α-L-*lyxo*-hexopyranosyl)-2,3,6-trideoxy-3-*C*-methyl-3-nitro-α-L-*ribo*-hexopyranoside (4f). Method A: A mixture of

disaccharide **4c** (140 mg, 0.29 mmol) and lithium hydroxide hydrate (113 mg, 2.69 mmol) in water (6 mL) was stirred under reflux for 4 h, cooled, and extracted wtih chloroform (12x15 mL). The combined organic extracts were dried (Na_2SO_4) and concentrated to give crude amino sugar **4e** which was added as a solution in chloroform (5 mL) to a solution of dimethyldioxirane (20 mL, prepared by the method of Murray and Singh,[21] 0.55M as determined by iodometric titration[25]). The reaction was stirred at 0 °C for 1.5 h after which additional DMDO solution (2 mL) was added in two 1 mL portions over the next 2 h. The solvent was removed under reduced pressure and the product was dissolved in ethyl acetate and the solution loaded onto a column of Florisil in a Pasteur pipet. Elution with ethyl acetate and concentration under reduced pressure gave nitro disaccharide **4f**; yield, 77 mg (79% overall): $[\alpha]_D^{25}$ -139° (*c*, 0.7, chloroform); ^1H NMR δ 4.86 (d, 1H, $J_{1',2'ax}$ = 3.0 Hz, H-1'), 4.73 (dd, 1H, $J_{1,2ax}$ = 6.4 Hz, $J_{1,2eq}$ = 6.4 Hz, H-1), 4.02-3.84 (m, 3H, H-4', H-5', H-5), 3.64 (m, 1H, H-3'), 3.60 (dd, 1H, $J_{4,5}$ = 6.3 Hz, H-4), 3.37 (s, 3H, OCH_3), 2.48 (dd, 1H, H-2eq), 2.33 (ddd, 1H, $J_{2ax,2eq}$ = 14.2 Hz, $J_{2ax,4}$ = 1.5 Hz, H-2ax), 2.26 (bs, OH), 1.78 (dd, 1H, $J_{2'eq,3'}$ = 3.7 Hz, H-2'eq), 1.73 (s, 3H, 3-CH_3), 1.67 (ddd, 1H, $J_{2'ax,2'eq}$ = 13.2 Hz, $J_{2'ax,3}$ = 12.5 Hz, H-2'ax), 1.37 (d, 3H, $J_{5,6}$ = 6.4 Hz, H-6), 1.25 (d, 3H, $J_{5',6'}$ = 6.6 Hz, H-6'); ^{13}C NMR δ 101.0 (C-1'), 97.3 (C-1), 87.9 (C-3), 85.1 (C-4), 70.8 (C-3'), 68.7, 66.6, 65.3 (C-5, C-4', C-5'), 55.0 (OCH_3), 33.8 (C-2), 32.8 (C-2'), 24.8 (3-CH_3), 19.3 (C-6), 16.5 (C-6').

Anal. Calcd for $C_{14}H_{25}NO_8$: C, 50.14; H, 7.51; H, 4.18. Found: C, 49.94; H, 7.83; N, 3.71.

Method B: A solution of nitro alcohol **2c** (73 mg, 0.36 mmol), freshly prepared glycosyl bromide **3a** (from 94 mg (0.44 mmol) of 3,4-di-*O*-acetyl-L-fucal in 3 mL dry benzene, 1.2 eq) and *s*-collidine (0.53 g, 1.2 eq) in dry benzene (3 mL) was added to a mixture of silver triflate (115 mg, 0.44 mmol) and 4Å molecular sieves (50 mg) in dry benzene (1 mL) with stirring under nitrogen in an ice bath. The reaction temperature was maintained between 5 °C and 15 °C during the addition. The ice bath was removed, and the reaction was allowed to warm to room temperature and stirred for 18 h. The reaction mixture was filtered through a pad of Celite and the cake was rinsed with ethyl acetate, then methanol. The filtrate was concentrated under reduced pressure to give a crude syrup which was loaded on a 1x10 cm column of silica gel and eluted with ethyl acetate-hexane (20:80 v/v). The crude product containing a mixture of α-linked disaccharide and unreacted nitro sugar was dissolved in dichloromethane-methanol (3 mL, 1:2 v/v) and treated with lithium hydroxide hydrate (5 mg) at room temperature for 2 h. Solvent was removed under reduced pressure and the product was purified by chromatography on a 1x12 cm column of silica gel with ethyl acetate-hexane (15:85 v/v) to give 23 mg (20 % from **2c**) of disaccharide **4f**.

ACKNOWLEDGMENTS

The authors thank the Petroleum Research Fund, administered by the American Chemical Society, and Villanova University for financial support of this work.

REFERENCES AND NOTES

1. K. Ishigami, Y. Hayakawa and H. Seto, *J. Antibiot.*, **47**, 1219 (1994).
2. H. Kawai, Y. Hayakama, M. Jakagawa, K. Furihata, H. Seto and N. Otake, *Tetrahedron Lett.*, **25**, 1941 (1984).
3. K. Ishii, Y. Nishimura, H. Nakagawa, S. Kondo and H. Umezawa, *J. Antibiot.*, **37**, 344 (1984).
4. P. F. Wiley, D. W. Elrod, D. J. Houser, J. L. Johnson, L.M. Pschigoda, W. C. Krueger and A. Moscowitz, *J. Org. Chem.*, **44**, 4030 (1979).
5. K. Ishii, S. Kondo, S. Nishimura, M. Hamada and H. Umezawa, *J. Antibiot.*, **36**, 454 (1983).
6. P. A. Wade and R. M. Giuliano in *Nitro Compounds: Recent Advances in Synthesis and Chemistry*, H. Feuer and A. T. Nielsen, Eds.; VCH: New York, 1993, p 137.
7. H. H. Baer, J. M. J. Frechet and U. Williams, *Can. J. Chem.*, **52**, 3337 (1974).
8. P. Jutten, H. D. Scharf and G. Raabe, *J. Org. Chem.*, **56**, 7144 (1991).
9. J. S. Brimacombe, R. Hanna, and L. C. N. Tucker, *J. Chem. Soc., Perkin Trans. 1*, 2273 (1983).
10. J. S. Brimacombe and K. M. M. Rahman, *J. Chem. Soc., Perkin Trans. 1*, 1073 (1985).
11. R. M. Giuliano and T. W. Deisenroth, *Carbohydr. Res.*, **158**, 249 (1986).
12. C. L. Stevens, R. P. Glinski, K. G. Taylor, B. Blumbergs, and S. K. Gupta, *J. Am. Chem. Soc.*, **92**, 3160 (1970).
13. N. Kunesch, C. Meit and J. Poisson, *Tetrahedron. Lett.*, **28**, 3569 (1987).
14. K. P. R. Kartha and H. Jennings, *J. Carbohydr. Chem.*, **9**, 777 (1990).
15. H. S. El Khadem, D. L. Swartz, J. K. Nelson and L. A. Berry, *Carbohydr. Res.*, **58**, 230 (1977).
16. S. Sabesan and S. Neira, *J. Org. Chem.*, **56**, 5468 (1991).
17. C. Kolar and G. Kneissel, *Angew. Chem. Int. Ed. Engl.*, **29**, 809 (1990).
18. C. Schuerch and F.J. Kronzer, *Carbohydr. Res.*, **27**, 379 (1973); S. Hanessian and J. Banoub, in *Synthetic Methods for Carbohydrates, ACS Symposium Series 39*, H.S. El Khadem, Ed.; American Chemical Society, Washington, D.C., 1977, p 36; S. Hanessian and J. Banoub, *Carbohydr. Res.*, **53**, C13 (1977).
19. J. S. Brimacombe and K. M. M. Rahman, *Carbohydr. Res.*, **113**, C6 (1983).
20. F. Weygand and E. Frauendorfer, *Chem. Ber.*, **103**, 2437 (1970).
21. R. Murray and M. Singh, *Org. Synth.*, **74**, 91 (1996).
22. A study of the oxidation of amino sugars lacking the α-methyl substituent with DMDO has been described: M. D. Wittman, R. L. Halcomb, and S. J. Danishefsky, *J. Org. Chem.*, **55**, 1981 (1990).
23. The phenylthioglycoside derived from the 3-*O*-methyl-4-*O*-benzyl analog of **3**, as well as its sulfoxide, did not couple with **2c** in satisfactory yield.
24. W. C. Still, M. Kahn, and A. Mitra, *J.Org.Chem.*, **43**, 2923 (1978).
25. A 1 mL-aliquot of freshly prepared DMDO solution in acetone (stored on dry ice over Na_2SO_4) was added to cold aqueous KI (5.0 mL, 0.20M) and starch solution (2 drops of 1%). After warming to room temperature, liberated I_2 was titrated to a colorless endpoint with aqueous sodium thiosulfate (0.005M).