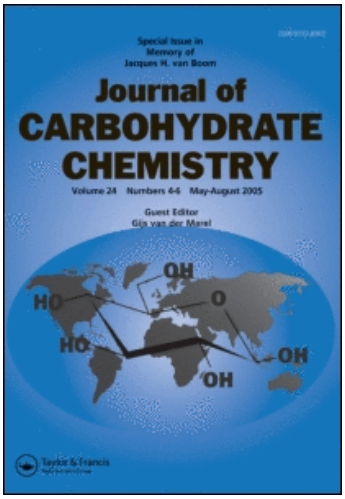


Journal of Carbohydrate Chemistry

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

Chemical Modification of Kanamycin A. II. Nucleophilic Displacement Reactions of Kanamycin-A-4''-sulfonates

R. Albert[a]; K. Dax[a]; A. E. Stutz[a]; H. Weidmann[a]
[a] Institute of Organic Chemistry Technical University Graz, Graz, Austria

To cite this Article Albert, R. , Dax, K. , Stutz, A. E. and Weidmann, H.(1982) 'Chemical Modification of Kanamycin A. II. Nucleophilic Displacement Reactions of Kanamycin-A-4''-sulfonates', Journal of Carbohydrate Chemistry, 1: 3, 289 — 299

To link to this Article: DOI: 10.1080/07328308208085101
URL: http://dx.doi.org/10.1080/07328308208085101

J. CARBOHYDRATE CHEMISTRY, 1(3), 289-299 (1982-83)

CHEMICAL MODIFICATION OF KANAMYCIN A. II.[1]

NUCLEOPHILIC DISPLACEMENT REACTIONS OF KANAMYCIN-A-4"-SULFONATES

R. Albert[2], K. Dax, A. E. Stütz[2], and H. Weidmann[+]

Institute of Organic Chemistry
Technical University Graz
A-8010 Graz, Austria

Received November 1, 1982

ABSTRACT

Reactions of 2',3',4',2",6"-penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-kanamycin-A-4"-brosylate (4b) or -4"-triflate (4c) with acetate, thiolacetate, azide, and fluoride, respectively, result in the formation of the corresponding derivatives of 4"-epi-kanamycin A (5a-d). While 4b invariably forms an elimination by-product (9), the only side-reaction of 4c consists in a neighboring group attack with formation of a 3",epi-4"-cyclic urethane (7). Removal of the protecting groups yields 4"-epi- (6a), 4"-thio-4"-epi-(6b), 4"-deoxy-4"-fluoro-4"-epi- (6d), 4"-azido-4"-deoxy-4"-epi- (6c), and after hydrogenation of the latter, 4"-amino-4"-deoxy-4"-epi-kanamycin A (6f).

Methyl 2,6-di-O-acetyl-3-amino-3-N-tert-butyloxycarbonyl-3-deoxy-4-O-triflyl-β-D-glucopyranoside (1b) served as a model to anticipate preparation, handling, and reactivity of 4c.

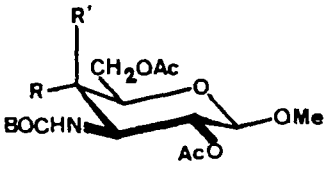
INTRODUCTION

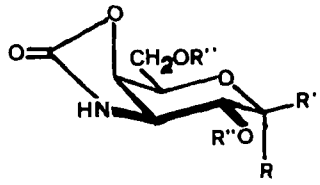
O-4→6 Acetyl migration was recently found[3] to be a generally applicable, valuable preparative tool for the regiospecific liberation of hydroxyl groups at C-4 in hexopyranosides and aminodeoxy-hexopyranosides, respectively. This method proved to be particularly useful for the regio- and stereospecific synthesis of 4"-halogeno-4"-deoxy-4"-epi-kanamycins A by way of various phosphorous containing reagents.[1]

In our continuing efforts toward the chemical modification of the kanosamine moiety of kanamycin A, we turned our attention to the nucleophilic displacement reactions of kanamycin-A-4"-sulfoates. In addition to the products of such reactions, general information on the reactivity of 3-amino-3-deoxy-3-N-tert-butyloxycarbonyl-4-O-sulfonyl-hexopyranosides, still lacking from the literature, was of interest.

RESULTS AND DISCUSSION

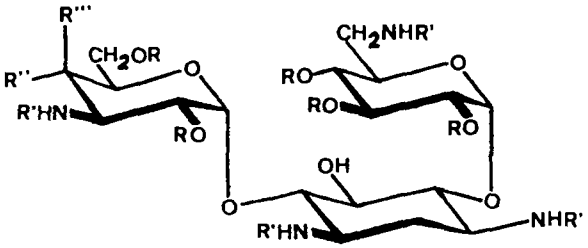
Inasmuch as the brosyloxy group was reported[4] to be the most favorable leaving group, even towards weak nucleophiles, in D-gluco→D-galacto inversion reactions, we prepared 2',3',4',2",6"-penta-O-acetyl-4"-O-brosyl-tetra-N-tert-butyloxycarbonyl-kanamycin A (4b) by esterification of 2',3',4',2",6"-penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-kanamycin A (4a) with 4-bromobenzene sulfonyl chloride in pyridine/dichloromethane. Treatment of 4b in dimethyl formamide at 100-110°C for 6-10 h with sodium acetate, thiolacetate, and azide, respectively, yielded the corresponding 4"-epi-acetate (5a), 4"-epi-thiolacetate (5b), and 4"-epi-azide (5c), respectively, in all cases together with minor amounts of 2',3',4',2",6"-penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-4"-deoxy-4"-eno-kanamycin A (9), an elimination product reported previously.[1] However, attempts to convert 4b into the corresponding 4"-epi-fluoride employing tetrabutyl ammonium fluoride in refluxing acetonitrile not only proceeded slowly but also resulted in intractable product mixtures.





1a R=OH, R'=H
1b R=OTf, R'=H
2 R=H, R'=F

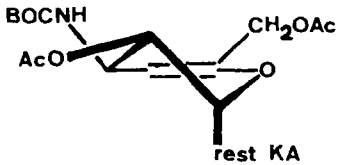
3 R=H, R'=OMe, R''=Ac
7 R=rest KA, R'=H, R''=Ac
8 R=rest KA, R'=R''=H



4a R=Ac, R'=BOC, R''=OH, R'''=H
4b R=Ac, R'=BOC, R''=OBros, R'''=H
4c R=Ac, R'=BOC, R''=OTf, R'''=H

5a R=Ac, R'=BOC, R''=H, R'''=OAc
5b R=Ac, R'=BOC, R''=H, R'''=SAc
5c R=Ac, R'=BOC, R''=H, R'''=N_3
5d R=Ac, R'=BOC, R''=H, R'''=F
5e R=Ac, R'=BOC, R''=H, R'''=NHAc

6a R=R'=R''=H, R'''=OH
6b R=R'=R''=H, R'''=SH
6c R=R'=R''=H, R'''=N_3
6d R=R'=R''=H, R'''=F
6e R=R'=R''=H, R'''=NHAc
6f R=R'=R''=H, R'''=NH_2



9

In order to enhance the reactivity of the substrate towards nucleophilic substitution, 2',3',4',2",6"-penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-4"-O-triflyl-kanamycin A (4c) as well as methyl 2,6-di-O-acetyl-3-amino-3-N-tert-butyloxycarbonyl-3-deoxy-4-O-triflyl-ß-D-glucopyranoside (1b), a monosaccharide model, were prepared. When 1b was treated with tetrabutyl ammonium fluoride in acetonitrile at ambient temperature, a 70 % yield of methyl 2,6-di-O-acetyl-3-amino-3-N-tert-butyloxycarbonyl-4-deoxy-4-fluoro-ß-D-galactopyranoside (2) was obtained within 2 h. Reaction of 4c under identical conditions afforded 2',3',4',2",6"-penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-4"-deoxy-4"-fluoro-4"-epi-kanamycin A (5d) in a yield of 73 %. Each of these reactions proceeds with formation of a by-product, a cyclic urethane, resulting from intramolecular attack of the respective neighboring tert-butyloxycarbonyl group. Thus compound 2 is accompanied by methyl 2,6-di-O-acetyl-3-amino-3-N,4-O-carbonyl-3-deoxy-ß-D-galactopyranoside (3, 20 %) and 5d by 2',3',4',2",6"-penta-O-acetyl-1,3,6'-tri-N-tert-butyloxycarbonyl-3"-N,4"-O-carbonyl-4"-epi-kanamycin A (7, 12 %). It is noteworthy that the mere substitution of acetonitrile for benzene in this reaction almost reverses the ratio of 2 to 3 to For comparison, 4c was treated at ambient temperature with the same nucleophiles as 4b.

The results of all nucleophilic substitutions are collected in the Table and can be summarized as follows:

In keeping with the good leaving group character of the triflyloxy group,[5] all nucleophilic displacement reactions of 4c proceed considerably faster and lead to higher yields than those of 4b. Aside from this distinctive quantitative effect, there is a noteworthy qualitative difference between 4-brosylates and 4-triflates of N-alkyloxycarbonyl-3-amino-3-deoxy-D-glucopyranosides. While the side-reaction of the former invariably results in a regiospecific elimination, that of the latter leads to a 3,epi-4-cyclic urethane, exclusively. Obviously, the comparatively severe conditions required to obtain reasonable rates of substitution in the brosylate favour elimination, while reactions at ambient temperature

TABLE

Nucleophilic Displacement Reactions

Starting Material	Reagent	Conditions	Time	Products (% Yield)
4b	NaOAc	DMF/100°C	8 h	5a (69.3), 9 (10.2)
	NaSAc	DMF/100°C	6 h	5b (62.8), 9 (4.0)
	NaN_3	DMF/100°C	10 h	5c (73.4), 9 (6.1)
4c	NaOAc	DMF/20°C	3 h	5a (81.9), 7 (10.7)
	NaSAc	DMF/20°C	2 h	5b (83.1), 7 (6.7)
	NaN_3	DMF/20°C	2 h	5c (80.7), 7 (4.0)
	Bu_4NF	MeCN/20°C	4 h	5d (73.3), 7 (12.1)

of the triflates only allow substitution. The ratio of S_N 2 to S_N i products is dependant on the nucleophility of the respective reagent as apparent from the reaction of 1b with tetrabutyl ammonium fluoride in an aprotic dipolar and a nonpolar solvent, respectively.

Hydrogenation of the azide 5c in the presence of acetic anhydride and Raney nickel gave 4"-acetamido-2',3',4',2",6"-penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-4"-deoxy-4"-epi-kanamycin A (5e).

Removal of O-acetyl groups by sodium methoxide and of N-tert-butyloxycarbonyl functions by trifluoroacetic acid afforded 4"-epi- (6a), 4"-thio-4"-epi- (6b), 4"-deoxy-4"-fluoro-4"-epi- (6d), 4"-acetamido-4"-deoxy-4"-epi- (6e), 3"-N,4"-O-carbonyl-4"-epi- (8), 4"-azido-4"-deoxy-4"-epi- (6c), and after hydrogenation of an aqueous solution of the latter in the presence of Raney nickel, 4"-amino-4"-deoxy-4"-epi-kanamycin A (6f).

In this context it is interesting that no oxidative coupling, a reaction quite common to thiosugars, could be observed on deprotection of 5b.

From these compounds, only 6a, 6c, and 6d show antibiotic activity comparable to that of kanamycin A.

EXPERIMENTAL

Melting points were obtained with a Tottoli apparatus and are uncorrected. Optical rotations were measured with a Perkin-Elmer 141 polarimeter. TLC was performed on silica gel 60 F_{254} precoated plates (Merck 5554) and column chromatography [6] on silica gel 60, 230-400 mesh (Merck 9385). ^{13}C NMR spectra, in full agreement with the structures assigned, were recorded with a Bruker WH-90 DS instrument.

Methyl 2,6-di-O-acetyl-3-amino-3-N-tert-butyloxycarbonyl-3-deoxy-4-O-triflyl-β-D-glucopyranoside (1b). A solution of 1a [2] (4.0 g, 11 mmol) in dichloromethane/pyridine (19:1, 80 mL) and a solution of trifluoromethane sulfonic anhydride (4.1 g, 14.5 mmol) in CH_2Cl_2 (40 mL) were combined at 0°C and kept at that temperature for 1 h. After rapid extraction with ice cold 1 N HCl (100 mL) followed by extraction with a saturated solution of $NaHCO_3$ (100 mL), the organic layer was dried and rapidly concentrated under reduced pressure to form a 20 % solution, ready for further reactions. Because of its marked inclination to form the cyclic urethane 3, any lengthy manipulation is to be avoided.

2',3',4',2",6"-Penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-4"-O-triflyl-kanamycin A (4c). From 4a [3] (4.0 g, 3.65 mmol) and trifluoromethane sulfonic anhydride (1.38 g, 4.87 mmol) as described for the preparation of 1b.

2',3',4',2",6"-Penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-4"-brosyl-kanamycin A (4b). To a solution of 4a (11.0 g, 10.04 mmol) in pyridine (50 mL) and CH_2Cl_2 (50 mL) a solution of 4-bromobenzene sulfonyl chloride (4.0 g, 15.7 mmol) was added. After 24 h at ambient temperature, the mixture was kept at 50°C for 2 h, methanol (10 mL) added, evaporated and the solution of the residue in $CHCl_3$ (100 mL) extracted with 1 N HCl followed by saturated $NaHCO_3$ solution. After drying and evaporation, chromatography (toluene/ethyl acetate 1:1) yielded 10.3 g (78.1 %) of 4b;

m.p. 150-155°C (dec.), $[\alpha]_D^{20}$ + 58.1° (c 1.3, $CHCl_3$), R_f 0.71 (toluene/ethyl acetate 1:2).

Methyl 2,6-di-O-acetyl-3-amino-3-N-tert-butyloxycarbonyl-3,4-dideoxy-4-fluoro-β-D-galactopyranoside (2) and Methyl 2,6-di-O-acetyl-3-N,4-O-carbonyl-β-D-galactopyranoside (3). To a 20 % solution of 1b (5 mL) in CH_2Cl_2 a solution of tetrabutyl ammonium fluoride (3.0 g, 11.5 mmol) in (a) acetonitrile or (b) benzene (20 mL) was added. After stirring for (a) 2 h or (b) 2 days at room temperature, the reaction mixture was evaporated to dryness and products separated by column chromatography (toluene/ethyl acetate 1:1); 2: (a) 0.70 g (69.5 %), (b) 0.14 g (13.9 %); m.p. 116-118°C, $[\alpha]_D^{20}$ - 19.8° (c 1.35, $CHCl_3$), R_f 0.78 (toluene/ethyl acetate 1:2); ^{13}C NMR ($CDCl_3$): 170.6 (Ac), 155.6 (BOC), 102.1 (C-1), 88.3 (d, $J_{4,F}$ 182.4 Hz; C-4), 80.4 (BOC), 71.8 (d, $J_{5,F}$ 19.1 Hz; C-5), 69.9 (C-2), 61.7 (d, $J_{6,F}$ 5.9 Hz; C-6), 56.8 (OMe), 53.3 (d, $J_{3,F}$ 17.6 Hz; C-3), 28.3 (BOC), 20.8 (Ac);
3: (a) 0.16 g (20.1 %), (b) 0.55 g (69.1 %); m.p. 158-158.5°C, $[\alpha]_D^{20}$ - 51.6° (c 0.78, $CHCl_3$), R_f 0.20 (toluene/ethyl acetate 1:2); ^{13}C NMR ($CDCl_3$): 170.8 (Ac), 158.3 (urethane), 100.2 (C-1), 74.0 and 73.1 (C-2 and -5), 70.5 (C-4), 62.9 (C-6), 56.5 (OMe), 55.2 (C-3), 20.9 (Ac).

2',3',4',2",6"-Penta-O-acetyl-tetra-N-tert-butyloxycarbonyl-4"-deoxy-4"-fluoro-4"-epi-kanamycin A (5d) and 2',3',4',2",6"-Penta-O-acetyl-1,3,6'-tri-N-tert-butyloxycarbonyl-3"-N,4"-O-carbonyl-4"-epi-kanamycin A (7). A solution of 4c (20 ml) was treated with a solution of tetrabutyl ammonium fluoride (5.0 g, 19.1 mmol) in acetonitrile (20 mL) at ambient temperature until no starting material could be detected by TLC. After evaporation of the solvent the products were separated by chromatography (toluene/ethyl acetate 2:3); 5d: 2.9 g (72.3 %), m.p. 140-145°C (dec.), $[\alpha]_D^{20}$ + 83.0° (c 0.9, $CHCl_3$), R_f 0.70 (toluene/ethyl acetate 1:2);
7: 0.45 g (12.1 %), m.p. 172-175°C (dec.), $[\alpha]_D^{20}$ + 78.0° (c 2.8, $CHCl_3$), R_f 0.68 (ethyl acetate).

General Procedure for Nucleophilic Substitution Reactions in Dimethyl Formamide.

With Brosylate 4b (Method A): A 10 % solution of the brosylate 4b in dimethyl formamide was stirred at 100-110°C in the presence of a 4-8 fold molar excess of reagent until the starting material had disappeared (t.l.c.). After filtration and evaporation , the residue was dissolved in $CHCl_3$ and, after filtration, chromatographed (toluene/ethyl acetate 3:2).

With Triflate 4c (Method B): A 20 % solution of the triflate 4c (20 mL) in CH_2Cl_2 was diluted with absol dimethyl formamide (100 mL), a 5-10 fold molar excess of reagent added and the mixture stirred at ambient temperature until no starting material could be detected by TLC. The mixture was worked up as described above.

2',3',4',2",4",6"-Hexa-O-acetyl-tetra-N-tert-butyloxycarbonyl-4"-epi-kanamycin A (5a). Method A: 4b (6.0 g, 4.57 mmol) in the presence of sodium acetate (3.0 g, 36.6 mmol) after 8 h gave 5a [3.6 g (69.3 %), m.p. 123-125°C, $[\alpha]_D^{20}$ + 91.0° (c 2, $CHCl_3$), R_f 0.56 (toluene/ethyl acetate 1:2)] together with 9 [1] (0.5 g, 10.2 %). Method B: From 4c by treatment with sodium acetate (3.0 g, 36.6 mmol) for 3 h, 5a (3.4 g, 81.9 %) and 7 (0.4 g, 10.7 %) were obtained.

2',3',4',2",6"-Penta-O-acetyl-4"-S-acetyl-tetra-N-tert-butyloxycarbonyl-4"-thio-4"-epi-kanamycin A (5b). Method A: 4b (4.0 g, 3.04 mmol) with sodium thiolacetate (1.1 g, 11.2 mmol) after 6 h yielded 5b [2.2 g (62.8 %), m.p. 145-150°C (dec.), $[\alpha]_D^{20}$ + 82.3° (c 1, $CHCl_3$), R_f 0.56 (toluene/ethyl acetate 1:2)] and 9 (0.13 g, 4 %). Method B: 4c with sodium thiolacetate (2.0 g, 20.4 mmol) after 2 h gave 5b (3.5 g, 83.1 %) together with 7 (0.25 g, 6.7 %).

2',3',4',2",6"-Penta-O-acetyl-4"-azido-tetra-N-tert-butyloxycarbonyl-4"-deoxy-4"-epi-kanamycin A (5c). Method A: Treatment of 4b (4.0 g, 3.04 mmol) with sodium azide (2.0 g, 30.8 mmol) for 10 h gave 5c [2.5 g (73.4 %), m.p. 140-145°C (dec.), $[\alpha]_D^{20}$ + 81.0° (c 1.75, $CHCl_3$), R_f 0.70 (toluene/ethyl acetate 1:2)] and 9 (0.2 g, 6.1 %). Method B: Reaction of 4c with sodium azide (2.0 g, 30.8 mmol) for 2 h led to 5c (3.3 g, 80.7 %) and 7 (0.15 g, 4.0 %).

2',3',4',2",6"-Penta-O-acetyl-4"-acetamido-tetra-N-tert-butyl-oxycarbonyl-4"-deoxy-4"-epi-kanamycin A (5e). A 10 % solution of 5c (3.0 g, 2.68 mmol) in acetic anhydride was hydrogenated at 3 bar (Parr apparatus) in the presence of Raney nickel (Fluka 83440, 3.0 g) for 8 h. After filtration and evaporation the residue was chromatographed (toluene/ethyl acetate 1:1) to give pure 5e [2.8 g (92.1 %); m.p. 175-180°C (dec.), $[\alpha]_D^{20}$ + 86.4° (c 0.95, $CHCl_3$), R_f 0.60 (ethyl acetate)].

General Procedure for the Removal of the Protecting Groups

A 5 % solution of the respective fully protected kanamycin A derivative in 0.01 N sodium methoxide in methanol was kept at ambient temperature for 30 minutes. After stirring with Amberlite IR 120 $[H^+]$ ion exchange resin (3 g) for 5 minutes, filtration and evaporation to dryness, the residue was treated with 98 % trifluoroacetic acid (5 mL/g) until gas evolution has ceased (5 min). After absol ethyl ether (50 mL/g) was added and the precipitate collected by filtration, a 10 % aqueous solution of this solid was treated with excess of Dowex 1X1 $[OH^-]$ followed by ion exchange chromatography on Amberliete CG 50 $[NH_4^+]$ (0.1-0.3 N ammonia).

Applying this procedure to 5a-e the following kanamycin A derivatives were obtained (R_f from $CHCl_3$/25 % NH_4OH/CH_3OH 1:2:2):

4"-epi-Kanamycin A (6a, 88.0 %), $[\alpha]_D^{20}$ + 142.2° (c 1.22, H_2O), R_f 0.51; ^{13}C-NMR[7]: C-3" 54.1 (54.3), C-4" 71.9 (68.4).

4"-Thio-4"-epi-kanamycin A (6b, 76.4 %), $[\alpha]_D^{20}$ + 119.2° (c 1.25, H_2O), R_f 0.52.

4"-Azido-4"-deoxy-4"-epi-kanamycin A (6c, 86.2 %), $[\alpha]_D^{20}$ + 139.5° (c 1.1, H_2O), R_f 0.70; ^{13}C-NMR[7]: C-3" 54.2 (53.8), C-4" 66.6 (62.2).

4"-Deoxy-4"-fluoro-4"-epi-kanamycin A (6d, 82.3 %), $[\alpha]_D^{20}$ + 137.9° (c 1.3, H_2O), R_f 0.70; ^{13}C NMR[7]: C-3" 53.1 (d, $J_{3",F}$ 17.7 Hz) [54.2 (17.7 Hz)], C-4" 93.6 (d, $J_{4",F}$ 176.5 Hz) [90.5 (178 Hz)], C-6" 62.6 (d, $J_{6",F}$ 5.9 Hz) [63.0 (5.9 Hz)].

4"-Acetamido-4"-deoxy-4"-epi-kanamycin A (6e, 85.5 %), $[\alpha]_D^{20}$ + 138.0° (c 1.25, H_2O), R_f 0.58; ^{13}C NMR[7]: C-3" 53.3 (54.9), C-4" 53.3 (51.0), 178.3 and 25.0 (Acetyl).

3"-N,4"-O-Carbonyl-4"-epi-kanamycin A (8, 78.9 %), $[\alpha]_D^{20}$ + 90.2° (c 1.25, H_2O), R_f 0.61; ^{13}C NMR[7]: C-3" 56.4, C-4" 71.0, C=O 164.1.

4"-Amino-4"-deoxy-4"-epi-kanamycin A (6f). A 5 % aqueous solution of 6c was hydrogenated at 3 bar in the presence of Raney nickel overnight. After filtration and chromatographic purification as described above, 6f was obtained as as amorphous powder (74.2 %), $[\alpha]_D^{20}$ + 132.8° (c 0.85, H_2O), R_f 0.48; ^{13}C NMR[7]: C-3" 54.2 (53.7), C-4" 54.2 (52.7), C-5" (67.2).

ACKNOWLEDGEMENT

Financial support by the Fonds zur Förderung der wissenschaftlichen Forschung, projects 3471 and 4528, as well as generous gift of kanamycin A and biological screening by Sandoz-Forschungsinstitut, Vienna, is gratefully acknowledged.

FOOTNOTES AND REFERENCES

1. R. Albert, K. Dax, A. E. Stütz, and H. Weidmann, Tetrahedron Lett., 23, 2645 (1982).

2. Part of Ph.D. Thesis, Technical University Graz, 1982.

3. R. Albert, K. Dax, A. E. Stütz, and H. Weidmann, J. Carbohydrate Chemistry, submitted for publication.

4. A. Maradufu and A. S. Perlin, Carbohydr. Res., 32, 261 (1974).

5. R. W. Binkley and D. G. Hehemann, J. Org. Chem., 43, 3244 (1978);

P. J. Stang, M. Hanack, and L. R. Subramanian, Synthesis, 1982, 85.

Y. Ishido and N. Sakairi, Carbohydr. Res., 97, 151 (1981).

6. H. Loibner and G. Seidl, Chromatographia, 12, 600 (1979).

7. D_2O, (ppm), shifts upon acidification (pD 2) with DCl in parenthesis; kanamycin A: C-3" 57.2 (58.0), C-4" 72.3 (68.7).