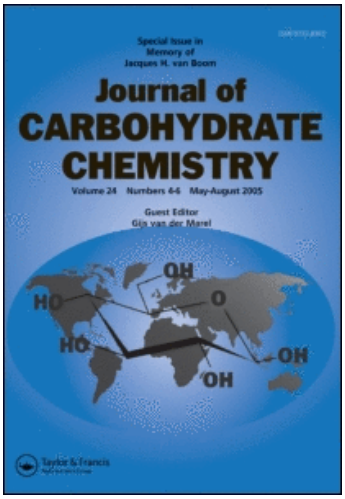

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CYTOTOXIC CYCLITOLS. SYNTHESIS OF ETHANOLAMINO AND HALOETHYLAMINO DERIVATIVES OF METHYLSULFONYLATED CYCLITOLS

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ABSTRACT

Starting from a mesylated inosamine several ethanol-, β-chloroethyl-, β-mesyloxy ethyl-, and β-iodoethylamino compounds were synthesized. Different chlorinated derivatives were obtained through variations in the chlorinating reagent, solvent, and reaction time.

INTRODUCTION

The importance of nitrogenated cyclitols in the biological activity of many complex substances is well known.[1] A small group of these cyclitols are nitrogen mustards and nitrosourea derivatives,[2] which have shown antitumor activity similar to that of analogous sugar compounds.[3] The efficacy of the active, nitrogenated groups is related to the general structure of the carrier molecule,[4] a fact which explains the broad-spectrum antitumor activity generally shown by synthetic nitrogen mustards.

On the basis of these facts, we considered inserting the active grouping $(R\text{-}CH_2CH_2)_2N$- (R= OH, halogen, mesyl) in the structure of some mesylated inositols which had already been synthesized.[5] To this effect we started from 3-amino-3-deoxy-

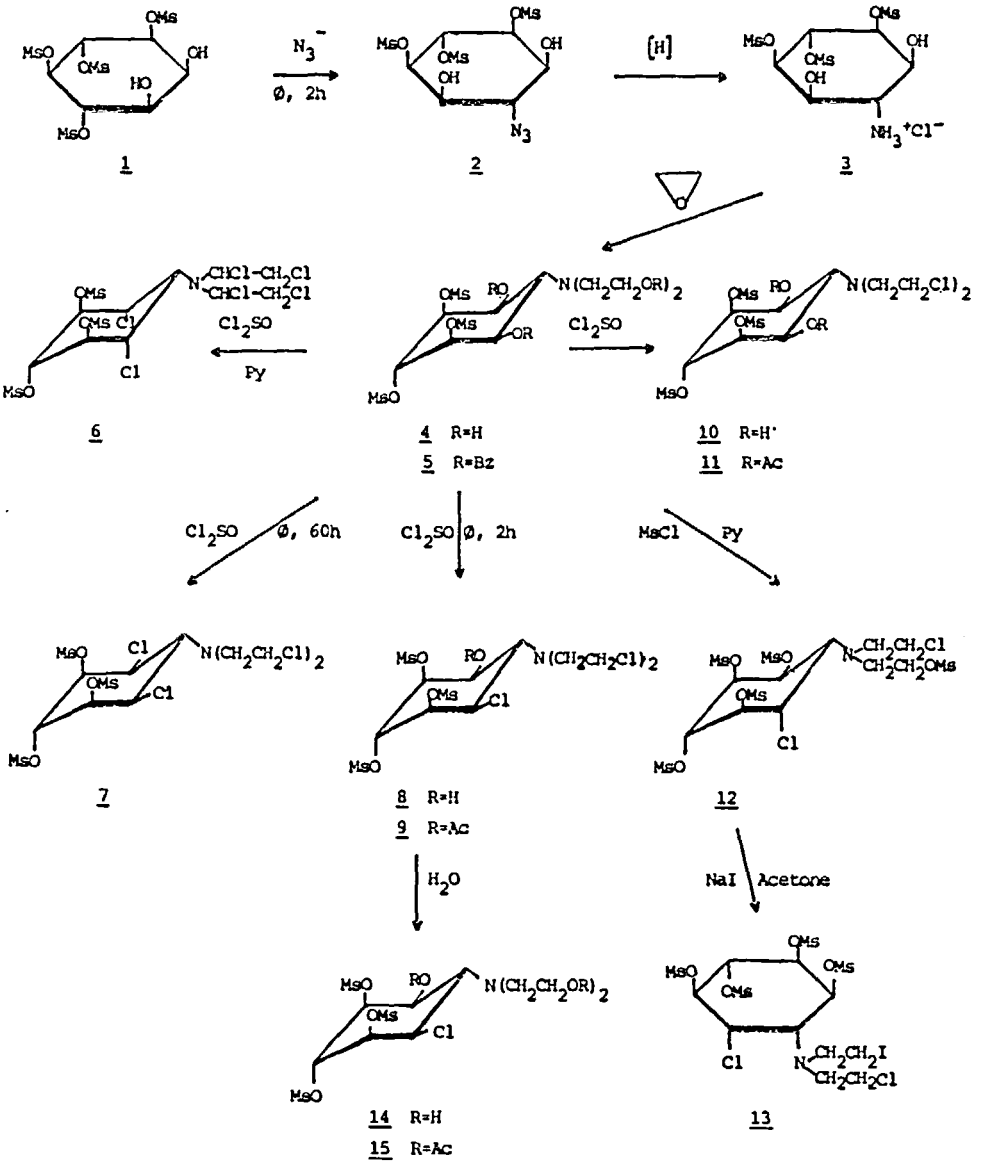
1,5,6-tri-O-(methanesulfonyl)-muco-inositol (3), previously obtained either by anchimerically assisted displacement[5] of a mesyl group in 1, or by nucleophilic opening of an oxirane ring by azide ion[6] to give the azido compound 2, whose reduction afforded 3 (see SCHEME 1). The synthesis of 2 and 3 was improved in order to work on a preparative scale.

RESULTS AND DISCUSSION

Compound 3 (as the hydrochloride) reacted in aqueous medium with ethylene oxide to give the diethanolamino derivative 4. This compound was shown to be a complex mixture by TLC analysis, although its ^{13}C and ^{1}H NMR spectra and analytical data indicated that it was pure. Benzoylation of 4 gave only one crystalline product (5), which on debenzoylation by mild alkali regenerated the apparently complex starting material.

In the ^{1}H NMR spectrum of 4 in D_2O, the protons on the hydroxylated carbon atoms (H-2 and H-4) appear, owing to the molecular symmetry, as a wide pair of doublets (δ 4.54) whose spacings (11 and 3 Hz) reflect, respectively, the axial-axial and the gauche relationships of these protons with the vicinal protons as shown in the conformation depicted for 4 (SCHEME 1). The resonances of H-1 and H-5, which appeared superimposed as a narrow triplet (δ 5.25), confirm this conformation. The ring proton of the nitrogenated carbon atom (H-3) appeared noticeably shielded and partially superimposed with the methylene groups at δ 4.00. Benzoylation of 4 did not markedly change these conformational features but deshielded strongly the two ring protons (H-2 and H-4 in 5, δ 5.62) related to the benzoylated positions. The symmetrical structure of 4 was also evident through its ^{13}C NMR spectrum which showed C-1 and C-5 as a singlet (78.8 ppm) and C-2 and C-4 (65.7 ppm) also a singlet, both of double intensity compared with that of C-6 (74.4) and C-3 (61.0 ppm).

Chlorination of 4 with thionyl chloride gave divergent results, depending on the presence or absence of pyridine in the medium and on the duration of the reaction. For example, when pyridine and thionyl chloride were used in equimolar amounts and the reaction was carried out at reflux, strong darkening occurred and low yields of the hexachloro derivative



SCHEME 1*

*All of the compounds, except for the meso ones, are racemic, and the formulas depict one enantiomer of the corresponding racemate.

6 was obtained. On the other hand, boiling of 4 with thionyl chloride alone for 60 h afforded the tetrachloro derivative 7 in good yield.

The ^{1}H NMR spectrum of the tetrachloro derivative 7, when compared with that of 4, showed that no configurational change had occurred in the ring, i.e., the chlorination took place without inversion of configuration at the chiral centers involved. This result is attributed to the experimental conditions employed, i.e., thionyl chloride in absence of pyridine. It is known that chlorination occurs with inversion of configuration at a chiral center when the thionyl chloride-pyridine reagent is employed.[7] The protons of the mesylated carbon atoms in 7 (H-2, H-3, and H-4) appeared as narrow triplets identical to that of the corresponding protons in 4, and the protons on the chlorinated, ring carbon atoms (H-1, H-5) appeared as an identical pair of doublets with spacings of 3 and 10 Hz, showing the gauche and trans-diaxial relationship of both protons with their neighbours, as that shown by the corresponding protons (H-2 and H-4) in 4. In the ^{13}C NMR spectrum the resonances for C-2, C-3, and C-4 are identical with the similar, mesylated carbon atoms in 4, the only differences being noticed for those carbons which have been chlorinated (i.e., C-1 and C-5). These resonances appeared superimposed and displaced to higher field (-6.8 ppm with respect to C-2 and C-4 in 4). The terminal chloromethylene groups in the sidechain appeared strongly displaced to higher field (-15.7 ppm) in comparison with the corresponding hydroxymethylene groups in 4. Similar drastic displacements on chlorination were observed with other sugars.[8]

The mass spectrum of 7 showed the parent peak (m/z 573, 8.6%) and a common tendency of some inositols to give cyclic, three-carbon fragments on electron impact[9] attributable to dichlorocyclopropyl ion and nitrogenated dichlorocyclopropane. The nitrogen mustard sidechain afforded an important fragment, m/z 92 (64.2%), attributable to $CH_2=\overset{+}{N}H-CH_2CH_2Cl$.

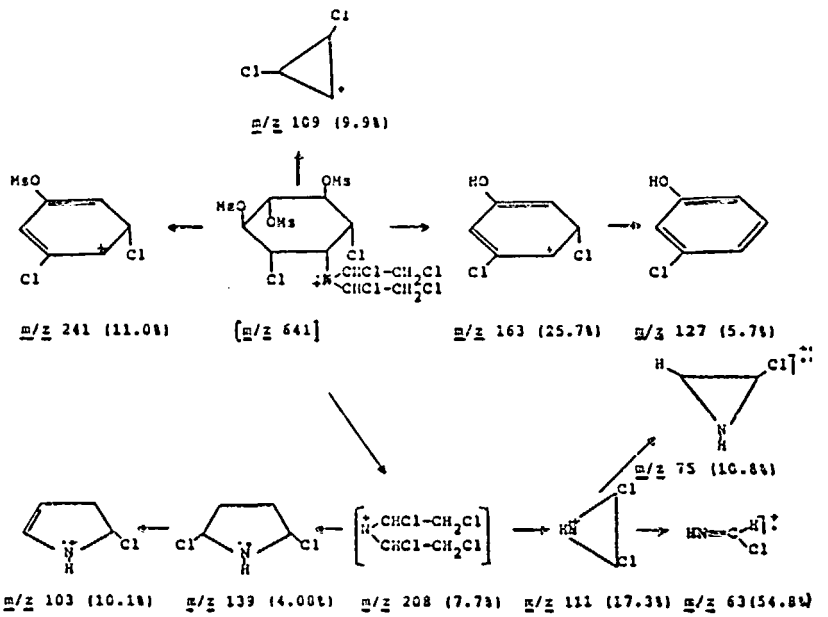
Chlorination of 4 by refluxing with thionyl chloride-pyridine occurred with inversion of configuration at the hydroxylated carbon atoms of the ring and with substitution at the secondary methylene groups of the side-chain, to give the hexachloro derivative 6. The ^{1}H NMR spectrum showed the six ring protons (δ 6.10-5.76) as narrow multiplets, sug-

gesting a gauche relationship among protons around the ring. The proton on the nitrogenated carbon atom (H-5), which usually resonates at high field owing to shielding by the electron pair of the nitrogen atom, in compound 6 appeared strongly deshielded and in the range of the other ring protons, indicating the electron-atracting influence of the chlorine atoms on the sidechain upon the vicinal nitrogen atom. The nitrogenated sidechain appeared as two multiplets comprising four (δ 4.35) and two (δ 4.05) protons, respectively. This compound, unfortunately, proved to be practically insoluble in solvents at concentrations required for a ^{13}C NMR determination in solution.

The mass spectrum of 6 showed a peak at m/z 208 and others that can be related to it (see SCHEME 2) that are useful in supporting this particular structure.

When compound 4 was heated for only two hours with thionyl chloride alone, a mixture of chloro derivatives was obtained, in which two main products different from either 6 or 7 could be distinguished. These new compounds differred noticeably from these on TLC and were separated and purified by successive selective solvent extractions, recrystallizations, and preparative TLC. A trichloro derivative (8, R_f 0.49) was isolated (21.1% yield) and its structure was established on a crystalline basis, whereas the other compound (10, R_f 0.25) was a syrup (23% approximate yield), which was identified as a dichloro derivative through the data afforded by its crystalline diacetate 11.

The structure of 8 was ascertained from the ^1H NMR spectrum of its monoacetate (9), which showed a narrow multiplet for three protons at δ 5.80-6.00 attributable to the equatorial protons on the mesylated carbon atoms (H-2, H-3, and H-4). The proton corresponding to the acetylated and chlorinated ring carbon atoms appeared well differentiated as two pairs of doublets at δ 5.67 (H-1) and 5.15 (H-5), respectively, with $J_{1,6}$=10 Hz and $J_{1,2}$=3 Hz for H-1 and $J_{5,6}$=10 Hz and $J_{4,5}$=3 for H-5, supporting the conformation depicted for 9. The proton on the nitrogenated carbon atom (H-6) appeared at higher field (δ 3.81) with $J_{5,6}$=$J_{1,6}$=10 Hz. These data agree for a stereochemistry of monochlorination on the ring with retention of configuration, as observed for compound 7, when

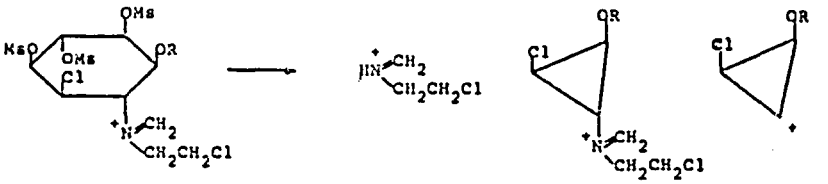


SCHEME 2

the reaction is conducted by heating with thionyl chloride in absence of pyridine. The ^{13}C NMR spectrum of 8 compared with that of 7 indicated an identical chlorinated sidechain for both compounds, supporting a structure with the free hydroxyl group on the ring. The resonances for the carbon atoms of the ring showed predictable differences for C-2, C-3, and C-4, indicating an asymmetric pattern of substitution, whereas the mesylated carbon atoms (C-1, C-5, and C-6) showed almost identical resonances to that of 7.

The structurally significant features of the mass spectrum of 8 were the peak at m/z 506 and others produced by its fragmentation, as depicted in SCHEME 3 for 8 and also for related peaks from 9.

The dichloro derivative 10 gave a diacetate 11, which in its ^{1}H NMR spectrum showed two acetyl groups superimposed as a singlet, indicating that both groups were either in the ring or in the side chain. The former possibility seemed supported by the appearance of the ring protons as a multiplet at rela-



9 R=H	m/z 506 (86.1%)	m/z 92 (19.7%)	m/z 182 (8%)	-
9 R=Ac	m/z 548 (91.6%)	m/z 92 (44.1%)	-	m/z 134 (16.0%)

SCHEME 3

tively low field, whereas ring chlorination would imply some relative displacement to higher field of these two ring protons. The resonance of the acetyl singlet at δ 2.22 suggest the equatorial orientation[10] of both acetyl groups.

The mass spectrum of 11 (at 20 eV) supports the postulated structure mainly on the basis of the peak at m/z 573 (100%) which originated by the loss of a CH_2Cl fragment from the bis-chlorinated sidechain.

Compound 4, when treated with mesyl chloride in pyridine, afforded the chloropermesylated compound 12. Analytical and spectroscopic data agree for five mesyl groups and two chlorine atoms, and its structure appeared unambiguous since its reaction with sodium iodide in acetone gave a monoiodo derivative 13. This reaction of chloromesylation was assumed to proceed with inversion of configuration at the chlorinated ring carbon atom to give a chiro derivative, on the basis of previous knowledge about chlorinations in pyridine.[7] This result was supported by the signal of the proton at the nitrogenated carbon atom (H-6, δ 4.17) which appeared as a pair of doublets with $J_{5,6}$=5 Hz and $J_{1,6}$=8 Hz at variance with the usual, broad triplet that appears when the trans-diaxial stereochemistry of that proton with its neighbours is maintained; (cf. compounds 9 and 15). Mass spectral data did not show the molecular ion, but showed two important ions at m/z 644 (100%) and 584 (30%) attributable to the loss

of CH_2Cl and CH_2OMs groups, respectively. The remaining five protons of the ring appeared as a multiplet (δ 5.80-6.10) that did not allow further configurational inferences.

The ^{13}C NMR spectrum of 12 was also consistent with the proposed structure, showing a signal at 75.4 ppm attributable to C-2 and C-3 which, being in a symmetric environment, appeared as a double-intensity singlet. Then, two single intensity, close resonances (74.2 and 73.5 ppm) are attributed to the other mesylated carbon atoms (C-1, C-4). At higher field appeared the two remaining resonances corresponding to the cyclic carbons, i.e., C-6 (61.2 ppm) and the chlorinated ring carbon atom (C-5, 53.8 ppm). The chloro- and mesyloxymethylene groups of the sidechain were observed to resonate at 42.3 and 38.6 ppm, respectively, whereas $(CH_2)_2N$ and the five mesyl groups appeared as higher intensity peaks at 39.4 and 38.8 ppm, respectively.

The iodo compound 13 showed tendency to lose iodine on recrystallization from hydroxylated solvents. From non-hydroxylated solvents, it precipitated in amorphous form, which did not give acceptable elemental analysis data, although they were reasonably close to the calculated values for the proposed structure. The ^1H NMR spectrum of 13 showed four singlets for the mesyl groups. The ^{13}C NMR spectrum showed a pattern for the carbon atoms of the cyclic moiety analogous to that of compound 12. The halogenated carbon atoms of the side chain appeared well differentiated (CH_2Cl at 42.3 ppm and CH_2I at 38.7 ppm). The mass spectrum showed structurally significant peaks as that of m/z 92 (51%) and m/z 184 (6.7%) attributable to $CH_2\overset{+}{N}H\text{-}CH_2CH_2Cl$ and $CH_2\overset{+}{N}H\text{-}CH_2CH_2I$, respectively.

When the trichloro derivative 8 was refluxed with water for 5 h, compound 14 was obtained as the principal product. This monochloro derivative showed, in its ^1H NMR spectrum, a narrow triplet for H-3 (δ 5.99, $J_{2,3}=J_{3,4}=3$ Hz), indicating its equatorial orientation, and a broad triplet for H-6 (δ 3.70, $J_{5,6}=J_{1,6}=10$ Hz) in an axial orientation as depicted for 14 (see SCHEME 1). The ring protons H-1 and H-5 appeared superimposed as a doublet of doublets (δ 5.08), but when the compound was acetylated to give 15, the annular acetoxy group was shown to deshield to corresponding proton to δ 5.66 (H-1), whereas the proton on the chlorinated carbon atom remained

practically unchanged from that observed in 14(δ 5.02, H-5). The triacetate 15 showed the two sidechain acetoxyl groups as a singlet and the third, located on the ring, at δ 2.22, indicating its equatorial orientation.[10] The four methylene groups on the sidechain appeared as two triplets comprising four protons each, at 3.14 ppm ($(CH_2)_2N$) and 4.16 ppm (2 CH_2OAc).

EXPERIMENTAL

General procedures. Melting points (Kofler hot stage) are uncorrected. TLC was conducted on Silica Gel G (Merck) plates (0.25 mm layer thickness) with the following solvents: A) water-saturated butanone, B) 19:1 (v/v) benzene-abs. ethanol, C) 9:1 (v/v) benzene-abs. ethanol. The spots were detected with 1) iodine vapor and 2) alkaline hydroxylamine-ferric nitrate (for esters).[11] IR spectra were recorded, for Nujol mulls with a Perkin-Elmer 710B spectrophotometer; ^1H and ^{13}C NMR spectra were recorded at 20-25 °C with a Varian XL-100 spectrometer at 100 (^1H) and 25.2 (^{13}C) MHz, respectively, with tetramethylsilane as the internal standard. Mass spectra were recorded, unless otherwise stated, with a Varian MAT 7 spectrometer commanded by a Varian-MAT data system 166 computer at an ionizing potential of 70 eV; the temperature of the direct-insertion probe was 200-220 °C.

3-Azido-3-deoxy-1,5,6-tri-O-(methanesulfonyl)-muco-inositol (2). The tetramesylate 1[5] (500 mg, 1.02 mmol) and sodium azide (250 mg, 3.85 mmol) were dissolved in 2-methoxyethanol (20 mL) and refluxed for 2 h. The solution was evaporated to dryness, and the residue was extracted with hot acetone (3 x 30 mL). The acetone extract was evaporated to dryness, and the residue, triturated in ethanol, afforded 2 (367 mg, 82% yield), mp and mixed mp[5] 227 °C.

3-Amino-3-deoxy-1,5,6-tri-O-(methanesulfonyl)-muco-inositol hydrochloride (3). Compound 2 (2 g, 5.56 mmol) was dissolved in 2-methoxyethanol (100 mL), and 10% Pd on charcoal (0.3 g) was added. This suspension was hydrogenated at room temperature for 20 h at 50 lb. in^{-2}, then was filtered and the catalyst was extracted with hot ethanol (3 x 30 mL). The 2-methoxyethanol solution and the ethanolic extracts were

combined and evaporated to dryness. The residue was suspended in boiling 2-propanol (200 mL), and concentrated hydrochloric acid (50 mL) was added to achieve dissolution of the product. Upon cooling, compound 3 was obtained (1.73 g, 80.8% yield), mp and mixed mp[5] 186-187 °C.

3-Deoxy-3-N-bis(2-hydroxymethyl)amino-1,5,6-tri-O-(methanesulfonyl)-muco-inositol (4). Compound 3 (7.82 g, 0.017 mol) was dissolved in water (100 mL) and ethylene oxide (60 mL) was added. The solution was kept 10 days at room temperature, at the end of which time it was evaporated to dryness. The residue crystallized as the free base from 2-propanol (7.87 g, 92% yield), mp 138-140 °C. Recrystallization successively from 2-propanol and from methanol saturated with gaseous hydrochloric acid, gave the hydrochloride, mp 108-110 °C. ^{1}H NMR data (D_2O): δ 3.41 (s, 6H, CH_3SO_2), 3.43 (3H, CH_3SO_2), 3.70 ($N(CH_2)_2$), 4.00 (5H, H-3, $(CH_2OH)_2$), 4.54 (H-2, H-4), 5.25 (H-1, H-5), 5.38 (t, H-6, $J_{1,6}=J_{5,6}=3$ Hz). ^{13}C NMR (C_5D_5N): δ 78.8 (C-1, C-5), 74.4 (C-6), 65.7 (C-2, C-4), 61.0 (C-3), 58.8 (CH_2OH), 37.8 (CH_3SO_2, $(CH_2)_2N$).

Anal. Calcd for $C_{13}H_{27}NO_{13}S_3 \cdot HCl$: C, 29.02; H, 5.21; N, 2.60; S, 17.86. Found: C, 28.74; H, 5.40; N, 2.91; S, 17.60.

From the hydrochloride, the free base was reobtained by treatment with methanolic ammonia; TLC R_f 0.45 (solvent A, reagent 1).

Anal. Calcd for $C_{13}H_{27}NO_{13}S_3 \cdot H_2O$: C, 30.06; H, 5.59; N, 2.70; S, 18.50. Found: C, 30.00; H, 5.49; N, 2.89; S, 18.13.

The pure substances, however, showed a complex mixture by TLC analysis. Acetylation afforded a mixture of syrupy acetates, but benzoylation was straightforward, and the benzoylated derivative 5 (see below) confirmed the purity of this substance.

1,5-Di-O-benzoyl-6-N-bis(2-benzoyloxyethyl)amino-6-deoxy-2,3,4-tri-O-(methanesulfonyl)-muco-inositol (5). Compound 4 (146 mg, 0.29 mmol) was dissolved in pyridine (2 mL), and benzoyl chloride (2 mL) was added to the previously cooled (0 °C) solution. After 24 h at room temperature, the solution was heated for 30 min in a boiling water bath and then was poured into ice-water. The solid obtained was washed repeatedly with water, dried and recrystallized from ethanol (155 mg). Recrystallization from 2-propanol-acetone (5:1) gave mp

160-163 °C; TLC R_f 0.60 (solvent B, reagent 1), double development); ^{1}H NMR data (Cl_3CD): δ 2.94 (s, 6H, CH_3SO_2), 3.22 (s, 3H, CH_3SO_2), 3.14 (t, 4H, $N(CH_2)_2$), 3.96 (H-6), 4.22 (t, 4H, 2 CH_2), 5.26 (3H, H-2, H-3, H-4), 5.62 (2H, H-1, H-5), 7.32-7.88 (m, 20H, benzoyl groups).

Anal. Calcd for $C_{41}H_{43}O_{17}NS_3$: C, 53.65; H, 4.69; N, 1.53; S, 10.47. Found: C, 53.54; H, 5.04; N, 1.76; S, 10.33.

Debenzoylation. Benzoate 5 was treated with 0.01 N sodium methoxide in methanol, and the solution was kept 3 days at room temperature. After neutralization with Dowex-50 $|H^+|$ resin, the solution was evaporated to dryness, and the residue was recrystallized from 2-propanol, to give a product with mp 140 °C. A TLC analysis revealed a complex mixture similar to that observed with a pure sample of 4 (q.v.).

1,3-Dichloro-2-N-bis(1,2-dichloroethyl)amino-1,2,3-trideoxy-4,5,6-tri-O-(methanesulfonyl)-myo-inositol (6). Compound 4 as the free base (1.17 g, 2.3 mmol) was dissolved in pyridine (5 mL), and thionyl chloride (5 mL) was added. The solution was refluxed 5 h, then kept 24 h at room temperature, and evaporated to dryness. The residue, triturated in 2-propanol, gave 1.12 g of dark powder which was extracted twice with acetone (2 x 20 mL), and the acetone extract was evaporated to dryness. The residue recrystallized from methanol to afford needles of 6 (307 mg, 20.6% yield), mp 213-216 °C; TLC (solvent B, reagent 1) R_f 0.47. The mother liquors showed a complex mixture of chlorinated derivatives, which was not further resolved; ^{1}H NMR (C_5D_5N): δ 3.53 (3H, CH_3SO_2), 3.55 (7H, H-6, 2 CH_3SO_2), 4.05 (m, 2H, -CHCl-), 4.35 (m, 4H, CH_2Cl), 5.76-6.10 (6H, ring protons). Mass spectrum: see SCHEME 2.

Anal. Calcd for $C_{13}H_{21}Cl_6NO_9S_3$: C, 24.22; H, 3.26; Cl, 33.07; N, 2.17; S, 14.91. Found: C, 24.53; H, 3.52; Cl, 32.73; N, 2.27; S, 14.74.

1,5-Dichloro-6-N-bis(2-chloroethyl)amino-1,5,6-trideoxy-2,3,4-tri-O-(methanesulfonyl)-muco-inositol (7). The hydrochloride of compound 4 (1.11 g, 2.1 mmol) was suspended in thionyl chloride (15 mL) and refluxed for 60 h. The light yellow solution was evaporated to dryness, and the residue was extracted with hot chloroform (3 x 30 mL) which on evaporation gave a residue that was recrystallized from methanol to afford

7 (787 mg, 61.1% yield), mp 209-211 °C; TLC R_f 0.39 (solvent B, reagent 1), ^1H NMR (C_5D_5N): δ 3.10-3.90 (m, 9H, H-6 and methylene protons), 3.38 (s, 3H, CH_3SO_2), 3.51 (s, 6H, CH_3SO_2), 4.98 (H-1, H-5), 5.79 (H-2, H-4), 5.93 (t, H-3, $J_{2,3}=J_{3,4}=3$ Hz). ^{13}C NMR (C_5D_5N): δ 78.4 (C-2, C-4), 74.5 (C-3), 63.4 (C-6), 58.8 (C-1, C-5), 43.1 (CH_2Cl), 38.4 (CH_3SO_2), 37.7 $(CH_2)_2N$).

Anal. Calcd for $C_{13}H_{23}Cl_4NO_9S_3$: C, 27.13; H, 4.00; Cl, 24.69; N, 2.43; S, 16.70. Found: C, 27.44; H, 4.29; Cl, 24.09; N, 2.50; S, 16.80.

D,L-4-Chloro-3-N-bis(2-chloroethyl)amino-3,4-dideoxy-1,5,6-tri-O-(methanesulfonyl)-muco-inositol (8). The hydrochloride of 4 (2.27 g, 4.2 mmol) was suspended in thionyl chloride (50 mL) and refluxed for 2 h. The solution was filtered to eliminate some unreacted material (0.169 g), the solution was evaporated to dryness, and the residue was triturated in cold water to obtain a powder that was filtered (1.84 g) and extracted by refluxing successively with benzene, chloroform, and dichloromethane (3 x 30 mL, each). The extracts were combined, evaporated to dryness, and recrystallized from methanol to give pure 8 (460 mg, 21.2% yield) as plates mp 168-171 °C; TLC R_f 0.49 (solvent B, reagent 1); ^1H NMR (DMSO-d_6): δ 3.12 (m, 4H, $(CH_2)_2N$), 3.28 (3H, CH_3SO_2), 3.38 (3H, CH_3SO_2), 3.40 (3H, CH_3SO_2), 3.58 (5H, H-3, $2CH_2Cl$), 4.63 (dd, H-4), 4.96 (m, H-2), 5.04-5.24 (m, H-1, H-5, H-6). ^{13}C NMR (C_5D_5N): δ 79.0 and 78.3 (C-1, C-5), 74.5 (C-6), 66.6 (C-2), 63.4 and 62.9 (C-3, C-4), 43.5 (CH_2Cl), 38.4 (CH_3SO_2), 37.6 (CH_2N).

Anal. Calcd for $C_{13}H_{24}Cl_3NO_{10}S_3.H_2O$: C, 27.15; H, 4.53; Cl, 18.54; N, 2.44; S, 16.71. Found: C, 27.40; H, 4.65; Cl, 18.90; N, 2.44; S, 16.60.

D,L-1-O-Acetyl-5-chloro-6-N-bis(2-chloroethyl)amino-5,6-dideoxy-2,3,4-tri-O-(methanesulfonyl)-muco-inositol (9). Compound 8 (140 mg, 0.25 mmol) was dissolved in pyridine-acetic anhydride (1:1, 3 mL), the solution was kept 24 h at room temperature, and then 0.5 h in a boiling water bath. After evaporation to dryness, the residue was triturated in water, filtered (117 mg), and recrystallized from ethanol-acetone (1:1, 12 mL). Compound 9 was obtained (89 mg), mp 188-191 °C;

TLC R_f 0.50 (solvent B, reagent 1). ^1H NMR (C_5D_5N): δ 2.23 (3H, CH_3CO), 3.33 (4H, $(CH_2)_2N$), 3.39, 3.55, 3.58 (9H, CH_3SO_2), 3.61 (4H, $2CH_2Cl$), 3.81 (t, H-6, $J_{1,6}=J_{5,6}=10$ Hz), 5.15 (dd, H-5, $J_{4,5}=3$ Hz, $J_{5,6}=10$ Hz), 5.67 (H-1, $J_{1,2}=3$, $J_{1,6}=10$ Hz), 5.80-6.00 (m, H-2, H-3, H-4).

Anal. Calcd for $C_{15}H_{26}Cl_3NO_{11}S_3$: C, 30.07; H, 4.34; Cl, 17.79; N, 2.33; S, 16.04. Found: C, 30.31; H, 4.57; Cl, 17.48; N, 2.50; S, 15.84.

1,5-Di-O-acetyl-6-N-bis(2-chloroethyl)amino-6-deoxy-2,3,4,-tri-O-(methanesulfonyl)-muco-inositol (11). The residue (504 mg) from the successive extractions to obtain 8, on TLC showed one principal spot, R_f 0.25 (solvent B, reagent 1). An aliquot (99 mg) of this residue, purified by preparative TLC, gave the dichloro derivative 10 as an amorphous powder which was acetylated with acetic anhydride-pyridine (1:1) as for compound 8. A crystalline acetate was obtained which was recrystallized from ethanol-acetone (1:1) to give 11 (21 mg), mp 203-205 °C. ^1H NMR (C_5D_5N): δ 2.22 (6H, CH_3CO), 3.20 (4H, $N(CH_2)_2$), 3.30 (3H, CH_3SO_2), 3.53 (6H, CH_3SO_2), 3.58 (4H, 2 CH_2Cl), 3.88 (t, H-6), 5.74-6.00 (5H, ring protons).

Anal. Calcd for $C_{17}H_{29}Cl_2O_{13}NS_3$: C, 32.79; H, 4.66; Cl, 11.41; N, 2.25; S, 15.43. Found: C, 33.12; H, 4.80; Cl, 11.30; N, 1.97; S, 15.21.

D,L-5-Chloro-6-[N-(2-chloroethyl)-N-(2-O-(methanesulfonyl)ethyl]amino-5,6-dideoxy-1,2,3,4-tetra-O-(methanesulfonyl)-chiro-inositol (12). Compound 4 as the free base (1.0 g, 1.4 mmol) was dissolved in boiling pyridine (15 mL) and, after cooling at room temperature, mesyl chloride (0.5 mL) was added. The mixture was kept 2 h at 0 °C, and a new portion (0.5 mL) of mesyl chloride was added. After four days at room temperature, the solution was poured onto crushed ice, and the powder obtained was repeatedly washed with water by decantation. The product (622 mg) was recrystallized from methanol to give 12 as plates mp 202-204 °C; TLC R_f 0.58 (solvent C, reagent 1). Yield 46.7%, ^1H NMR (C_5D_5N): δ 3.32 (s, 3H, CH_3SO_2), 3.50 (m, 4H, $(CH_2)_2N$), 3.60, 3.66 (12H, CH_3SO_2), 3.80 (m, CH_2Cl, CH_2OMs), 4.17 (t, H-6, $J_{5,6}=6$ Hz, $J_{1,6}=8$ Hz), 5.82-6.10 (m, 5H, ring protons); ^{13}C NMR (C_5D_5N): δ 75.4 (C-2, C-3), 74.2, 73.5 (C-1, C-4), 61.2 (C-6), 53.8 (C-5), 42.3 (CH_2Cl), 39.4 ($(CH_2)_2N$), 38.8 (CH_3SO_2), 38.6 (CH_2OMs).

Anal. Calcd for $C_{15}H_{29}Cl_2NO_{15}S_5$: C, 25.94; H, 4.18; Cl, 10.23; N, 2.02; S, 23.05. Found: C, 26.27; H, 4.27; Cl, 10.32; N, 2.17; S, 22.79.

D,L-5-Chloro-6-[N-(2-chloroethyl)-N-(2-iodoethyl)]amino-5,6-dideoxy-1,2,3,4-tetra-O-(methanesulfonyl)-chiro-inositol (13). Compound 12 (348 mg, 0.5 mmol) was dissolved in acetone (50 mL) containing sodium iodide (245 mg, 1.6 mmol). The mixture was refluxed 20 h, filtered and evaporated to dryness. The dried residue was triturated in cold water and the precipitate obtained (379 mg) was filtered and disolved in ethyl acetate. This solution was washed with a saturated solution of sodium thiosulfate, then water, and dried (Na_2SO_4). Concentration of the solution and precipitation of the product with light petroleum gave an amorphous solid (245 mg) and subsequently a second crop (77 mg, total yield of 13 88.5%). The solid was again precipitated with light petroleum from its ethyl acetate solution and gave mp 199-201 °C; TLC R_f 0.63 (solvent C, reagent 1). ^1H NMR (C_5D_5N): δ 3.35 (CH_2I), 3.51 ($N(CH_2)_2$), 3.54 (6H, CH_3SO_2), 3.61 (6H, CH_3SO_2), 3.80 (CH_2Cl), 4.20 (H-6, $J_{1,6}$=8 Hz, $J_{5,6}$=5 Hz), 5.80-6.06 (m, 5H, ring protons). ^{13}C NMR (C_5D_5N): δ 75.6, 75.4 (C-2, C-3), 74.2, 73.5 (C-1, C-4), 61.1 (C-6), 53.8 (C-5), 42.3 (CH_2Cl), 39.9 ($(CH_2)_2N$), 39.4 (CH_3SO_2), 38.7 (CH_2I).

Anal. Calcd for $C_{14}H_{26}Cl_2INO_{12}S_4$: C, 23.14; H, 3.58; N, 1.93. Found: C, 23.98; H, 3.50; N, 1.74.

D,L-1-Chloro-1,6-dideoxy-6-N-bis(2-hydroxyethyl)amino-2,3,4-tri-O-(methanesulfonyl)-muco-inositol (14). Compound 7 (275 mg, 0.48 mmol) was suspended in water (10 mL) and refluxed for 5 h. The solution was evaporated to dryness, and the dried residue was triturated in methanol to afford 14 as a powder (129 mg, 47% yield), mp 178-181 °C; TLC R_f 0.66 (solvent A, reagent 1). Recrystallization from 5% hydrochloric acid in methanol gave the hydrochloride mp 137-141 °C. ^1H NMR (C_5D_5N): δ 3.17 (4H, $N(CH_2)_2$), 3.51 (3H, CH_3SO_2), 3.55 (6H, CH_3SO_2), 3.70 (H-6, $J_{5,6}=J_{1,6}$=10 Hz), 3.88 (4H, $2CH_2Cl$), 5.10 (dd, H-1, H-5), 5.84 (H-2, H-4), 5.99 (t, H-3, $J_{3,4}=J_{2,3}$=3 Hz).

Anal. Calcd for $C_{13}H_{26}ClNO_{12}S_3 \cdot HCl$: C, 28.06; H, 4.86; Cl, 12.77; N, 2.52; S, 17.27. Found: C, 28.05; H, 4.41; Cl, 13.14; N, 2.44; S, 17.47.

D,L-6-N-Bis(2-acetoxyethyl)amino-1-O-acetyl-5-chloro-5,6-dideoxy-2,3,4-tri-O-(methanesulfonyl)-muco-inositol (15). Compound 14 (150 mg, 0.29 mmol) was dissolved by warming in a 1:1 mixture of acetic anhydride-pyridine. The solution was kept 24 h at room temperature, then 0.5 h in a boiling water-bath, and filtered (146 mg). Recrystallization from methanol-acetone gave 15 (113 mg), mp 122-126 °C; TLC R_f 0.82 (solvent B, reagent 1). ^{1}H NMR (C_5D_5N): δ 2.06 (6H, CH_3CO), 2.22 (3H, CH_3CO), 3.14 (4H, $N(CH_2)_2$, 3.42 (3H, CH_3SO_2), 3.52 (3H, CH_3SO_2), 3.54 (3H, CH_3SO_2), 3.86 (H-6, $J_{5,6}=J_{1,6}=11$), 4.16 (4H, $2CH_2OAc$), 5.02 (dd, H-5, $J_{5,6}=11$ Hz, $J_{4,5}=4$ Hz), 5.66 (dd, H-1, $J_{1,6}=11$ Hz, $J_{1,2}=4$ Hz), 5.90 (m, H-2, H-3, H-4).

Anal. Calcd for $C_{19}H_{32}ClNO_{15}S_3$: C, 35.32; H, 4.96; Cl, 5.50; N, 2.17; S, 14.87. Found: C, 35.05; H, 5.07; Cl, 5.45; N, 2.58; S, 14.53.

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