Tetrahedron, Vol. 37, Supplement No 1, pp 9 to 12, 1981
Printed in Great Britain.

0040-4020/81/Suppl. 1-0009-04 $02.00/0

STEREOSPECIFIC SYNTHESIS OF METHYL 2,6-DIACETAMIDO-2,3,6-TRIDEOXY-α-D-RIBO-HEXOFURANOSIDE AND HEXOPYRANOSIDE, DERIVATIVES OF TOBROSAMINE

JEAN-CLAUDE FLORENT and CLAUDE MONNERET*

Institut de Chimie des Substances Naturelles, 91190 Gif-sur-Yvette, France

(*Received in France* 9 *January* 1980)

Abstract—Title derivatives of 2,6-diamino-2,3,6-trideoxy-D-ribo-hexose (tobrosamine), the amino-sugar component of the antibiotic nebramycin factor 6, have been prepared in a stereospecific manner, based on the Tischenko rearrangement of methyl 3,6-dideoxy-α-arabino-hexopyranoside in the presence of benzaldehyde and Lewis acids.

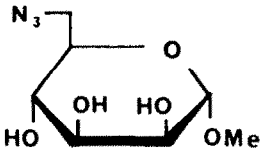
The highly successful clinical applications of aminoglycoside antibiotics has stimulated numerous investigations[1] in order to establish structure-activity relationships and to prepare more active compounds. At the same time an understanding of the mechanism of aminoglycosidic antibiotics resistance[2] has opened the way to the synthesis of new and improved compounds. For example, as one of the most frequently encountered means of inactivation of these antibiotics is phosphorylation at C-3',[3] gentamycin, sisomicin and tobramycin having a 3'-deoxy-sugar unit are among the most active derivatives of this class.[4] Following from these observations a large number of 3'-O-methyl and 3'-deoxy or 3',4'-dideoxy derivatives have been synthesized and in particular 3'-deoxy-kanamycin B or tobramycin.[5]

Derivatives of the sugar component of tobramycin, tobrosamine (2,6-diamino-2,3,6-trideoxy-D-*ribo-hexose*) **13** have already been described by Guthrie *et al.*[6] (methyl 2,6-diacetamido-4-*O*-mesyl tobrosaminide) and Gero *et al.*[7] (ethyl 2,6-diacetamido tobrosaminide). In a report of our laboratory[8] we have already described a new route to its methyl diacetamido derivative **12** by azidolysis at the allylic position of an enol ether sugar prepared from mannose.

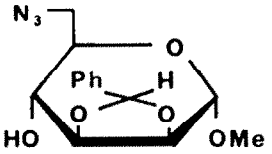
We now wish to report a new higher yielding and stereoselective synthesis of the hexoside **12** based upon a Tischenko type rearrangement[9] of the key intermediate **6**, methyl 6-acetamido-3,6-dideoxy-α-D-arabino-hexopyranoside.[10] As previously published,[8] the starting materials for this synthesis, methyl 6-azido-α-D-mannopyranoside **1** and its 2,3-*O*-benzylidene acetal **2** were prepared respectively in 90% and 72% overall yield from methyl α-D-mannoside. As observed for other 2,3-*O*-benzylidene acetals of hexose of manno configuration[8,11] and for the corresponding 4-*O*-methyl ether[8] treatment of **2** with NBS led regio- and stereospecifically to the bromo-sugar **3**. This latter was converted by lithium aluminium hydride reduction followed by N-acetylation into the crystalline intermediate **6**.

A more direct route from **1** to **6** was also explored. Barton *et al.*[12] have shown that treatment of 1,2-diols at room temperature with imidoyl chloride derived from dimethylbenzamide led to the regiospecific formation of the chlorhydrin benzoate in excellent yields. Under similar conditions but in presence of ten molar equivalents of imidoyl chloride, the azido-sugar **1** gave a mixture of the 3-chloro-sugars **4** and **5** (80% yield, ratio 4/5 = 0.75/1). The formation of **5** can be explained by esterification of the OH-4 due to the excess of reagent. If lower concentration of imidoyl chloride are used, the ratio 4/5 increases but unfortunately the total yield drops, a part of the reagent being probably consumed by the free OH on C-4. In fact the formation of **5** is not inconvenient since the next step is the $LiAlH_4$ reduction of the mixture followed by acetylation which gave exclusively the acetamido-sugar **6** with an overall yield of 40% from **1**.

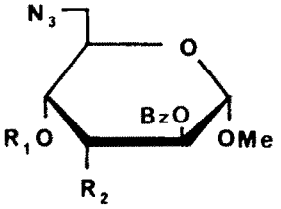
In contrast with the results obtained for methyl 3,6-dideoxy-α-D or L-arabino-hexopyranosides,[9] treatment of a benzaldehyde solution of **6** with different acids ($ZnCl_2$, $AlCl_3$ or $TiCl_4$) did not give any of the expected Tischenko type rearrangement product but led to a degradation of the starting material. Fortunately we found after many experiments that in the presence of an equal amount of $AlCl_3$ and $ZnCl_2$, **6** rearranged to **7** in rather good yield (65–70%). The structure of **7** was established from its spectroscopic data to be a furanoside derivative with a benzoic ester on C-2 and a benzyl ether on C-5. Catalytic transesterification of **7** gave **8** and subsequent treatment of **8** in a dichloromethane solution with triflic anhydride according to the procedure of Hall *et al.*[13] gave the sulfonate **9**. Azidolysis of **9** in HMPT at room temperature led to the azido-sugar **10** as the exclusive product (65% after purification). Hydrogenolysis of **10** in the presence of palladium-on-charcoal followed by acetylation in methanol of the crude product afforded the methyl furanoside of *N*,*N*'-acetyl tobrosamine **11**. Finally methyl 2,6-diacetamido-2,3,6-trideoxy-α-D-ribo-hexopyranoside (methyl 2,6-diacetamido-tobrosaminide) **12**[8] was



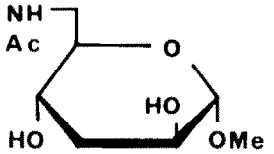
1



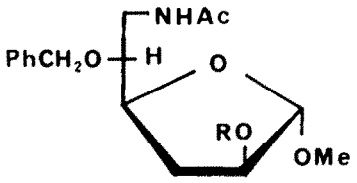
2



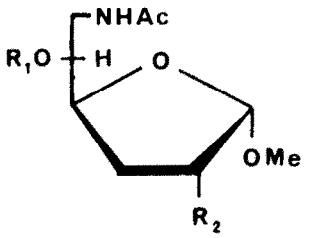
3 $R_1 = H$; $R_2 = Br$
4 $R_1 = H$; $R_2 = Cl$
5 $R_1 = Bz$; $R_2 = Cl$



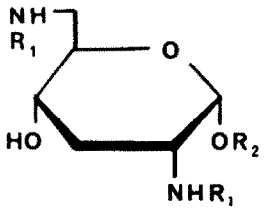
6



7 R = Bz
8 R = H
9 R = Tf



10 $R_1 = CH_2Ph$; $R_2 = N_3$
11 $R_1 = H$; $R_2 = NHAc$



12 $R_1 = Ac$ $R_2 = Me$
13 $R_1 = R_2 = H$ Tobrosamine

isolated after acid hydrolysis and glycosidation with methanolic solution of hydrochloric acid.

EXPERIMENTAL

General. All m.ps were determined on a Büchi apparatus and are uncorrected. IR spectra (film or Nujol suspension) were registered on a Perkin–Elmer Model 257. ^{1}H NMR spectra were recorded at 90 or 240 MHz with TMS as internal standard, the chemical shifts being expressed in ppm. Optical rotations were measured on a Perkin–Elmer Model 141 MC polarimeter and 1-dm tubes were used. TLC used Merck silica gel GF_{254} plates. Column chromatography was conducted using Merck silica gel type H. Microanalysis were performed by the Service Central de Microanalyse du CNRS. Extraction in usual manner means that the organic layer is washed with dil HCl or aq $NaHCO_3$, dried, and concentrated *in vacuo*.

Methyl 6-azido-2-O-benzoyl-3-bromo-3,6-dideoxy-α-D-altropyranoside **3**

To a solution of **2**[8] (10.9 g, 35 mmol) in carbon tetrachloride (500 ml) was added *N*-bromosuccinimide (8 g) and barium carbonate (10 g). The suspension was heated with stirring under reflux for 3 h. After cooling and filtration, the filtrate was extracted with dichloromethane in the usual manner. The solvent was removed *in vacuo* giving pure **3** (12.3 g, 90%) as a syrup, $[\alpha]_D^{20}$ + 18° ($CHCl_3$). IR (film): 2100 (N_3) 1725, 1265 and 1105 cm^{-1} (ester). NMR ($CDCl_3$, 60 MHz): δ = 5.52 (d, J = 2.5, H-2), 4.81 (s, H-1), 4.57 (dd, J=J'=2.5, H-3), 4.03 (m, H-4, H-5), 3.6 (m, H-6, H-6'), 3.47 (s, OMe). Calc. for $C_{14}H_{16}O_5N_3Br$ (386.21). C,H,N,Br.

Methyl 6-azido-2-O-benzoyl-3-chloro-3,6-dideoxy-α-D-altropyranoside **4** *and methyl 6-azido-2,4-di-O-benzoyl-3-chloro-3,6-dideoxy-α-D-altropyranoside* **5**

Reaction of the mannose derivative **1**[8] (236 mg, 1.08 mmol) with pyridine (0.5 ml) and *N,N*-dimethyl-α-chlorobenzylidene ammonium chloride (prepared as described[12] from N,N-dimethylbenzamide (2.5 g)) for 16h at reflux gave, on evaporation, a syrup. TLC (hexane–ethyl acetate–dichloro-methane, 5:1:1, v/v) showed two spots. Column chromatography on silica gel (same solvent as for TLC) gave **5** (197 mg) and in the later fractions **4** (146 mg): total yield 80%. **4**: syrup, $[\alpha]_D^{20}$ + 70° ($CHCl_3$). IR (film): 2100 (N_3), 1725, 1250 and 1105 cm^{-1} (ester). NMR ($CDCl_3$, 250 MHz): δ = 5.42 (dd, J = 0.7, J' = 3, H-2), 4.79 (s, H-1), 4.49 (dd, J = 3.5, J' = 3, H-3), 4.11 (m, H-4, H-5), 3.63 (m, H-6), 3.55 (m, H-6'), 3.47 (s, OMe). Calc. for $C_{14}H_{16}N_3O_5Cl$ (341.70). C,H,N. **5**: syrup, $[\alpha]_D^{20}$ + 34.5° ($CHCl_3$), NMR ($CDCl_3$, 250 MHz): δ = 5.48 (dd, J = 9, J' = 4, H-4), 5.46 (dd, J = 1, J' = 4, H-2), 4.87 (s, H-1), 4.71 (t, J = J' = 4, H-3), 4.56 (q, J = 9, J' = 5, J'' = 4, H-5), 3.55 (m, H-6 and H-6'), 3.51 (s, OMe). Calc. for $C_{21}H_{20}N_3O_6Cl$ (445.80). C,H,O.

Methyl 6-acetamido-3,6-dideoxy-α-D-arabino-hexopyranoside **6**

To a solution of **3** (13g, 35 mmol) in a mixture of tetrahydrofuran (150 ml) and ether (150 ml) was slowly added lithium aluminium hydride (5 g). After stirring under reflux for 4h, the excess of hydride reagent was destroyed by addition of a saturated solution of ammonium sulfate. The mixture was filtered and the solids washed with a mixture of chloroform and methanol (1:1, 2 × 100 ml). The combined filtrates were evaporated to dryness to give a syrup. This syrup was dissolved in anhydrous pyridine (75 ml) and acetic anhydride (25 ml) and stirred overnight. Water was then added (100 ml) and then aqueous bicarbonate and extraction was carried out with dichloromethane. The organic layer was washed with aqueous solution of hydrochloric acid 1 N, water and then with aqueous bicarbonate. After evaporation to dryness, the syrup (5g) was dissolved in a methanolic solution of sodium methanolate (0.2 N, 100 ml) and stirred for 2h. Neutralisation was effectued with Amberlite IR 50(H^+) and after filtration the solvent was removed under reduce pressure. The residue (3.5 g) was chromatographed on silica gel (solvent: dichloromethane-methanol, 9:1, v/v) to give **6** (3.1 g, 42%): m.p. 141–142 (methanol-ether), $[\alpha]_D^{20}$ + 93° (MeOH, c 2). IR (Nujol): 1650, 1570 cm^{-1} (amide). NMR (pyridine-d_5, 250 MHz): δ = 4.9 (s, H-1), 3.35 (s, OMe), 2.6–2.25 (m, H-3 and H-3'), 2.07 (s, OAc). Calc. for $C_9H_{17}O_5N$ (219.23). C,H,N.

Methyl 6-acetamido-2-O-benzoyl-5-O-benzyl-3,6-dideoxy-α-D-arabino-hexofuranoside **7**

Zinc chloride (1 g, 7 mmol) and aluminium chloride (0.9 g 7 mmol) were added to a solution of **6** (493 mg, 2.2 mmol) in benzaldehyde (15 ml). The suspension was stirred for 3 h and then the excess of reagent was destroyed by addition of methanol (2 ml) and prolonged stirring for 5 min. Extraction with dichloromethane in usual manner and purification by preparative chromatography (solvent: hexane-dichloromethane-ethylacetate, 1:1:1, v/v) gave **7** as a syrup: $[\alpha]_D^{20}$ + 16° (MeOH). IR: (film): 1725, 1275 and 1100 cm^{-1} (ester), 1610 and 1495 (acetamide). NMR ($CDCl_3$, 60 MHz): δ = 7.4 8 (m, Ph), 7.32 (s, $PhCH_2$), 5.30 (m, H-2), 5.05 (s, H-1), 4.68 (wide s, CH_2Ph), 3.40 (s, OMe), 1.87 (s, Ac). Calc. for $C_{23}H_{27}O_6N$ (413.45). C,H,N.

Methyl 6-acetamido-5-O-benzyl-3,6-dideoxy-α-D-arabino-hexofuranoside **8**

A solution of **7** (1.06 g, 2.6 mmol) in ethanolic KOH (4%, 50 ml) was stirred at room temperature for 3 h. Extraction with dichloromethane afforded pure **8** (TLC dichloromethane-methanol, 7:1, v/v) (595 mg, 75%) as a syrup, $[\alpha]_D^{20}$ + 58° (MeOH, c 1.75). IR (film): 1655 and 1540 cm^{-1} (amide). NMR ($CDCl_3$, 90 MHz): δ = 7.36 (m, Ph), 4.70–4.75 (m, 3 H, CH_2Ph and H-1) 3.25 (s, OMe), 1.87 (s, Ac). Calc. for $C_{16}H_{23}O_5N$ (309.35). C,H,N.

Methyl 6-acetamido-5-O-benzyl-3,6-dideoxy-2-O-trifluoromethanesulfonyl-α-D-arabino-hexofuranoside **9**

Pyridine (1.13 ml) and then a solution of triflic anhydride (1.13 g, 4 mmol) in dichloromethane (5 ml) were added dropwise to a solution of **8** (515 mg, 1.67 mmol) in dichloromethane (25 ml) chilled at −15°C. The mixture was kept at this temperature until TLC (solvent: dichloromethane-methanol, 7:1, v/v) showed complete disappearance of the starting material (3 h). The mixture was then poured into a chilled saturated solution of sodium bicarbonate and then extracted in the usual manner with dichloromethane. After concentration *in vacuo*, the residue (795 mg) was chromatographed on silica gel (dichloromethane-methanol, 33:1, v/v) to give **9** (569 mg, 90%) unstable, immediately converted into **10**.

Methyl 6-acetamido-2-azido-5-O-benzyl-2,3,6-trideoxy-α-D-ribo-hexofuranoside **10**

Sodium azide (100 mg) was added over 15 mn to a cooled (0°) solution of **9** (480 mg, 1.2 mmol) in HMPT (15 ml). Stirring was then continued for 2 h at room temperature. Extraction with benzene ether (1:1, v/v) gave a syrup which showed a single spot in TLC (dichloromethane-methanol 95:5, v/v). Purification by preparative TLC afforded **10** (234 mg, 60%) as a syrup: $[\alpha]_D^{20}$ + 115° ($CHCl_3$). IR: 2100 cm^{-1} (N_3). NMR ($CDCl_3$, 240 (MHz): δ = 4.92 (d, J = 4, H-1), 4.80 (m, CH_2Ph), 3.43 (s, OMe), 1.6–2.3 (m, H-3 and H-3'), 1.84 (s, OAc). Calc. for $C_{16}H_{22}N_4O_4$ (334.37). CHN.

Methyl 2,6-diacetamido-2,3,6-trideoxy-α-D-ribo-hexofuranoside **11**

A solution of **10** (141 mg, 0.42 mmol) in methanol (20 ml) and acetic anhydride (1 ml) was stirred for 2 h under hydrogen in the presence of palladium-on-charcoal (10%, 150 mg). The catalyst was then removed by filtration and the filtrate evaporated under reduced pressure, and evaporation repeated thrice after addition of toluene (10 ml).

Chromatography of the residue on silica gel (dichloromethane-methanol, 14:1, *v/v*) gave **11** (64 mg, 65%): mp 176–177° (acetone), $[\alpha]_D^{20}$ + 120° (c 2, methanol). IR (Nujol): 1635 and 1555 cm^{-1} (amide). NMR (240 MHz, pyridine-d_5): δ 5.07 (broad s, H-1), 3.30 (s, OMe), 2.09 (s, Ac). Calc. for $C_{11}H_{20}O_5$ (232.27). C,H,O.

Methyl 2,6-diacetamido-2,3,6-trideoxy-α-D-ribo-hexopyranoside **12**

A solution of **11** (50 mg) in a methanolic solution of hydrochloric acid (7 ml, 3%, v/v) was heated under reflux overnight. After cooling and neutralisation with Amberlite IR 45 (OH^-), the filtrate was evaporated *in vacuo*. TLC (dichloromethane-methanol, 12.5:1, v/v) showed two principal spots. Chromatography on silica gel gave starting material **11** (15 mg) and **12** (25 mg) whose constants are in agreement with those previously described in the literature.[8] mp, 207° (acetone-methanol); $[\alpha]_D^{20}$ + 90° ($CHCl_3$).

Acknowledgements—We thank Prof. D. H. R. Barton for his kind suggestion to use imidoyl chloride and Dr. P. Potier for his continuous interest and encouragement.

REFERENCES

[1]S. Umezawa, *Adv. Carbohydr. Chem. Biochem.* **30,** 111 (1974).
[2]J. E. Davies and R. Rownd, *Science* **176,** 758 (1972).
[3]H. Umezawa, M. Okanishi, S. Kondo, K. Hamana, R. Utahara, K. Maeda and S. Mitsuhashi, *Ibid.* **157,** 1559 (1967); H. Umezawa, *Adv. Carbohydr. Chem. Biochem.* **30,** 183 (1974); and refs therein; R. Benveniste and J. Davies, *Ann. Rev. Biochem.* **42,** 471 (1973).
[4]C. C. Crowe and E. Sanders, *Antimicrob. Agents Chemotherap.* **3,** 24 (1973); M. Kugelman, A. K. Mallams and H. F. Vernay, *J. Antibiotics* **26,** 394 (1973); K. F. Koch and J. A. Rhoades, *Antimicrob. Agents Chemotherap.* 309 (1970).
[5]Y. Takagi, T. Miyake, T. Tsuchiya and S. Umezawa, *J. Antibiotics* **26,** 403 (1973).
[6]C. L. Brewer and R. D. Guthrie, *J. Chem. Soc.* Perkin I 657 (1974).
[7]J. Leboul, J. Cleophax, S. D. Gero, A. Rolland and A. Forchioni, *Tetrahedron* **33,** 965 (1977).
[8]J. C. Florent, C. Monneret and Q. Khuong-Huu, *Tetrahedron* **34,** 909 (1978).
[9]J. C. Florent, C. Monneret and Q. Khuong-Huu, *Tetrahedron Letters* 243 (1977); J. C. Florent and C. Monneret, *Carbohydr. Res.* **81,** 225 (1980).
[10]C. Monneret, J. C. Florent and Y. Chapleur, *C.R. Acad. Sc. Paris* **285C,** 587 (1977).
[11]C. Monneret, J. C. Florent, N. Gladieux and Q. Khuong-Huu, *Carbohydr. Res.* **50,** 35 (1976); J. C. Florent, C. Monneret and Q. Khuong-Huu, *Ibid.* **56,** 301 (1977).
[12]T. G. Back, D. H. R. Barton and B. L. Rao, *J. Chem. Soc.* Perkin I 1715 (1977).
[13]L. D. Hall and D. C. Miller, *Carbohydr. Res.* **47,** 299 (1976).