*Carbohydrate Research*, 38 (1974) 205–216


# SYNTHESIS OF THE 6-METHYL AND 3,6- AND 4,6-DIMETHYL ETHERS OF METHYL 2-ACETAMIDO-2-DEOXY-α-D-MANNOPYRANOSIDE*

NASIR-UD-DIN, DOROTHY A. JEANLOZ, AND ROGER W. JEANLOZ†

*Laboratory for Carbohydrate Research, Departments of Biological Chemistry and Medicine, Harvard Medical School and Massachusetts General Hospital, Boston, Massachusetts 02114 (U. S. A.)*

(Received August 10th, 1974; accepted September 6th, 1974)

ABSTRACT

The 6-mono- (**6**) and 4,6- (**16**) and 3,6-di-methyl (**25**) ethers of methyl 2-acetamido-2-deoxy-α-D-mannopyranoside have been synthesized from 6-*O*-trityl, 4,6-*O*-benzylidene, and 3-*O*-methyl derivatives, respectively, by way of *O*-benzoyl and of *O*-allyl derivatives. The yields were respectively 37 and 43% for **6**, 34 and 50% for **16**, and 14 and 25% for **25**. These ethers are used as standard compounds for the structure elucidation, by methylation, of polymers containing 2-amino-2-deoxy-D-mannose.

## INTRODUCTION

2-Amino-2-deoxy-D-mannose[1] and 2-amino-2-deoxy-D-mannuronic acid[2] are components of the bacterial cell-wall. For the structural investigation of the complex polymers containing these two sugars, methyl ethers of 2-amino-2-deoxy-D-mannose are needed, as the uronic acid derivatives can be reduced to the corresponding hexose derivatives. We have previously described[3] the synthesis of the 3- and 4-mono-, 3,4-di-, and 3,4,6-tri-methyl ethers of 2-amino-2-deoxy-D-mannose. We now describe the synthesis of the remaining methyl ethers susceptible to being formed by methylation of 2-amino-2-deoxy-D-mannopyranose, namely, the 6-mono- (**6**) and 4,6- (**16**) and 3,6-di-methyl (**25**) ethers. In addition to the benzoyl group conventionally used, the allyl group[4] was examined for the protection of the hydroxyl groups on C-3 and C-4; it was found to be very convenient, as etherification therewith readily proceeds to completion, the derivatives formed are isolated in good yield, and the removal of

*Amino Sugars XCII. This is publication No. 646 of the Robert W. Lovett Memorial Group for the Study of Diseases Causing Deformities, Harvard Medical School and the Massachusetts General Hospital, Boston, Massachusetts. This work was supported by research grants from the National Institute of Allergy and Infectious Diseases (AI-06692) and from the National Institute of Arthritis, Metabolism, and Digestive Diseases (AM-03564), National Institutes of Health, U. S. Public Health Service. A preliminary communication has been presented [*Abstr. Papers Amer. Chem. Soc. Meeting*, 168 (1974) CARB-16].

†To whom communications should be addressed.

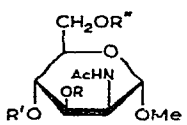
the protecting groups is conveniently performed with tris(triphenyl)phosphine-rhodium chloride and 1,4-diazabicyclo[2.2.2]octane (the Corey reagents[5]), followed by treatment with Dowex 50 ($H^+$) ion-exchange resin.

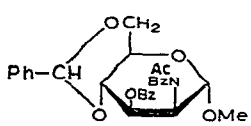
## RESULTS AND DISCUSSION

Benzoylation of methyl 2-acetamido-2-deoxy-6-*O*-trityl-α-D-mannopyranoside (**1**) with 2 molar equiv. of benzoyl chloride in the presence of pyridine at −60° (in order to avoid *N*-benzoylation[3,6]) gave the 3,4-dibenzoate (**2**) in 72% yield. A more polar compound, having properties corresponding to those of a monobenzoic ester, was isolated in 10% yield; it was probably[7] the 3-benzoate (**3**). Removal of the trityl group gave crystalline methyl 2-acetamido-3,4-di-*O*-benzoyl-2-deoxy-α-D-manno-pyranoside (**4**), which was methylated with methyl iodide and silver oxide (the Purdie reagents[8]) to give the crystalline 6-methyl ether (**5**), subsequently hydrolyzed under alkaline conditions to afford crystalline methyl 2-acetamido-2-deoxy-6-*O*-methyl-α-D-mannopyranoside (**6**) in an overall yield (from **1**) of ~37%.

Compound **6** was also synthesized from **1** through the crystalline 3,4-di-*O*-allyl-6-*O*-trityl derivative (**7**), obtained in 85% yield. In order to minimize *N*-substitution during the allylation of the hydroxyl groups[9], sodium hydride was replaced by sodium hydroxide, a milder reagent. Compound **7** was detritylated in 85% yield to give the syrupy 3,4-di-*O*-allyl derivative (**8**), which was methylated with the Purdie reagents. The resulting, crystalline 6-methyl ether (**9**), obtained in 82% yield, was treated with the Corey reagents[5], a procedure less cumbersome than the isomerization of allyl into 1-propenyl groups with dimethyl sulfoxide[4]; although, in this case, the isomerization was not complete, repetition of the procedure raised the yield to over 70%. The 1-propenyl derivative obtained was directly hydrolyzed, without purification, with a sulfonic resin to give **6** in an overall yield of 43% from **1**.

Comparison of the yields of **6** respectively obtained *via* the benzoate and the allyl derivatives shows that little migration of the benzoate groups occurred during methylation with the Purdie reagents. The acid- and alkali-stable allyl derivatives



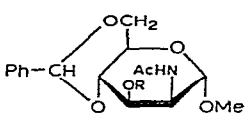



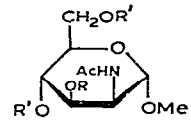

10

1 R = R' = H, R" = Tr
2 R = R' = Bz, R" = Tr
3 R = Bz, R' = H, R" = Tr
4 R = R' = Bz, R" = H
5 R = R' = Bz, R" = Me
6 R = R' = H, R" = Me
7 R = R' = $-CH_2-CH=CH_2$, R" = Tr
8 R = R' = $-CH_2-CH=CH_2$, R" = H
9 R = R' = $-CH_2-CH=CH_2$, R" = Me

were found to be convenient intermediates, and the removal of the allyl groups under mildly acidic conditions suggests their use in the synthesis of alkali-labile, oligosaccharide-containing compounds.

Re-investigation of the benzoylation of methyl 2-acetamido-4,6-*O*-benzylidene-2-deoxy-α-D-mannopyranoside (**11**) showed that treatment with only one molar equiv. of benzoyl chloride at −5° results in *N*-benzoylation, with formation of the *N*-benzoyl-3-*O*-benzoyl derivative (**10**) in 47% yield, and that a mixing temperature of −60°, followed by −20°, was needed in order to avoid *N*-benzoylation and obtain **12**, as previously reported[3]. Purdie methylation of the previously described[3] methyl 2-acetamido-3-*O*-benzoyl-2-deoxy-α-D-mannopyranoside (**14**), and saponification of the resulting crystalline 4,6-dimethyl ether (**15**) gave, in an overall yield of 34% from **11**, amorphous methyl 2-acetamido-2-deoxy-4,6-di-*O*-methyl-α-D-mannopyranoside (**16**). In a similar sequence of reactions, amorphous **16** was obtained, in an overall yield of 50%, by way of the crystalline 3-*O*-allyl-4,6-*O*-benzylidene (**13**), 3-*O*-allyl (**17**), and 3-*O*-allyl-4,6-di-*O*-methyl (**18**) derivatives.



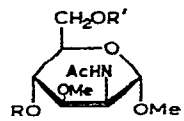

11 R = H
12 R = Bz
13 R = —$CH_2$—CH=$CH_2$

14 R = Bz, R' = H
15 R = Bz, R' = Me
16 R = H, R' = Me
17 R = —$CH_2$—CH=$CH_2$, R' = H
18 R = —$CH_2$—CH=$CH_2$, R' = Me

Treatment of methyl 2-acetamido-2-deoxy-3-*O*-methyl-α-D-mannopyranoside[3] (**19**) with chlorotriphenylmethane under vigorous conditions gave the 6-trityl ether (**20**) in only 61% yield. For an evaluation of the steric and electronic interactions of the hydroxyl groups on C-3 and C-4, it is of interest to compare the yield with that of the tritylation of methyl 2-acetamido-2-deoxy-α-D-glucopyranoside[10] (82%) and its 3-methyl ether[10] (70%), and of methyl 2-acetamido-2-deoxy-α-D-galactopyranoside[11] (88%) and its 3-methyl[12] (86%) and 3,4-dimethyl[13] (43%) ethers.

Benzoylation of **20** at very low temperature afforded the crystalline 4-*O*-benzoyl-3-*O*-methyl-6-*O*-trityl derivative (**21**), which was detritylated to give the crystalline 4-*O*-benzoyl-3-*O*-methyl derivative (**22**). Methylation with the Purdie reagents in tetrahydrofuran resulted in the formation of methyl 2-acetamido-4-*O*-benzoyl-2-deoxy-3,6-di-*O*-methyl-α-D-mannopyranoside (**23**), contaminated with some of the 6-*O*-benzoyl-3,4-di-*O*-methyl derivative (**24**). A similar migration of the benzoyl group from O-4 to O-6 under the mild alkaline conditions of the Purdie methylation, probably through the formation of an intermediate orthobenzoate, has previously been observed in the *gluco*[14] and *galacto*[12] series. Removal of the 4-*O*-benzoyl group gave amorphous methyl 2-acetamido-2-deoxy-3,6-di-*O*-methyl-α-D-mannopyranoside (**25**) in an overall yield of 14%.

In a similar sequence of reactions, **25** was obtained from **20** (in an overall yield of 25%) by way of the crystalline 4-*O*-allyl-3-*O*-methyl-6-*O*-trityl (**26**), amorphous 4-*O*-allyl-3-*O*-methyl (**27**), and crystalline 4-*O*-allyl-3,6-di-*O*-methyl (**28**) derivatives.




**19** R = R' = H
**20** R = H, R' = Tr
**21** R = Bz, R' = Tr
**22** R = Bz, R' = H
**23** R = Bz, R' = Me
**24** R = Me, R' = Bz
**25** R = H, R' = Me
**26** R = $-CH_2-CH=CH_2$, R' = Tr
**27** R = $-CH_2-CH=CH_2$, R' = H
**28** R = $-CH_2-CH=CH_2$, R' = Me

## EXPERIMENTAL

*General.* — Melting points were determined with a Mettler FP-2 apparatus and correspond to "corrected melting points". The i.r. spectra were recorded with a Perkin–Elmer Model 237 spectrophotometer. Optical rotations were measured in 1-dm semimicrotubes with a Perkin–Elmer Model 141 polarimeter. Gas–liquid chromatography (g.l.c.) of the per(trimethylsilyl)[15] ethers was performed with a Perkin–Elmer Model 900 gas chromatograph equipped with a flame-ionization detector and a stainless-steel column (1.5 m) of 3% of OV-225 on Gas Chrom Q (100–200 mesh) programmed for a rise of 10°.min$^{-1}$ from 80 to 250°, with nitrogen as the carrier gas. The chloroform used was analytical-reagent grade, and contained 0.75% of ethanol. Column chromatography was performed on Silica Gel Merck (70–325 mesh; E. Merck, Darmstadt, Germany), used without pretreatment. The proportion of weight of substance to weight of silica gel was 1:70 to 1:90. The volume of the fractions eluted was 3–4 ml per g of the substance to be chromatographed. Thin-layer chromatography (t.l.c.) was performed on plates of silica gel (without fluorescence indicator; layer thickness 0.25 mm; E. Merck). The compounds were detected by spraying the plates with (*A*) 1:1:18 (v/v) anisaldehyde–conc. sulfuric acid–ethanol, or (*B*) 1:10 (v/v) conc. sulfuric acid–ethanol. The $R_{TMG}$ values refer to the mobilities on plates of silica gel, relative to that of methyl 2-acetamido-2-deoxy-3,4,6-tri-*O*-methyl-α-D-glucopyranoside as unity. Evaporations were conducted *in vacuo*, with the bath temperature kept below 40°. G.l.c.–mass spectrometry (g.l.c.–m.s.) was performed with an analytical system consisting of an IBM 1800 computer fed raw data generated by a single-focussing, mass spectrometer (Perkin–Elmer–Hitachi RMU-6). In all cases, the analyses were conducted on the per(trimethylsilyl) ethers of the sugars. Microanalyses were performed by Dr. W. Manser, Zurich, Switzerland.

*Methyl 2-acetamido-3,4-di-O-benzoyl-2-deoxy-6-O-trityl-α-D-mannopyranoside* (**2**). — A solution of compound **1** (110 mg) in dry pyridine (10 ml) was treated with benzoyl chloride (39 μl) at −60°. The mixture was kept for 8 h at −20° and for 20 h at −10°, and then diluted with chloroform (10 ml) and successively washed with ice-cold, saturated solutions of sodium hydrogensulfate, sodium hydrogencarbonate, and ice-cold water, dried (sodium sulfate), and evaporated to give a syrup which was chromatographed on silica gel with 19:1 (v/v) dichloromethane–ethanol. The major, syrupy product was crystallized from ether to give small needles (111 mg, 72%), m.p. 129–131° (with softening at 113°), $[\alpha]_D^{20}$ −22° (*c* 0.65, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3290 (NH), 1770 (ester), 1655 (Amide I), and 1545 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) chloroform–ethanol: $R_F$ 0.63.

*Anal.* Calc. for $C_{41}H_{39}NO_8$: C, 73.09; H, 5.83; N, 2.08; O, 18.99. Found: C, 72.99; H, 5.78; N, 2.13; O, 18.86.

*Methyl 2-acetamido-3-O-benzoyl-2-deoxy-6-O-trityl-α-D-mannopyranoside* (**3**). — The later fractions eluted from the silica gel column, described in the preceding paragraph, gave a compound [$R_F$ 0.3 in 19:1 (v/v) chloroform–ethanol] which crystallized from benzene–hexane to give needles (15 mg), m.p. 223–225°, $[\alpha]_D^{20}$ +50° (*c* 0.59, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3355 (NH), 1740 and 1680 (ester), 1655 (Amide I), and 1560 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) chloroform–ethanol: $R_F$ 0.31.

*Anal.* Calc. for $C_{34}H_{35}NO_7$: C, 71.69; H, 6.19; N, 2.46; O, 19.66. Found: C, 72.04; H, 6.29; N, 2.40; O, 19.17.

A solution of **3** (3 mg) in dry methanol (1 ml) was treated with 0.1M methanolic sodium methoxide (0.01 ml) for 12 h at 4°, diluted with methanol (2 ml), de-ionized with Rexyn 300 ($H^+$, $OH^-$) ion-exchange resin (1 ml), and evaporated. T.l.c. of the residue in 9:1 (v/v) chloroform–methanol indicated the presence of **1**.

*Methyl 2-acetamido-3,4-di-O-benzoyl-2-deoxy-α-D-mannopyranoside* (**4**). — A suspension of **2** (90 mg) in acetic acid (5 ml, 60%) was heated on a water bath for 1 h at 80°, cooled to room temperature, diluted with water to 250 ml, and lyophilized. The syrupy residue was chromatographed on a column of silica gel with 14:1 (v/v) chloroform–ethanol. Triphenylmethanol was eluted first, and then **4**. Crystallization from ether–heptane gave 51 mg (86%) of prisms, m.p. 104–105°, $[\alpha]_D^{22}$ −26° (*c* 0.22, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3430 (broad, OH and NH), 1730 (ester), 1655 (Amide I), and 1545 $cm^{-1}$ (Amide II); t.l.c. in 14:1 (v/v) chloroform–ethanol: $R_F$ 0.35.

*Anal.* Calc. for $C_{23}H_{25}NO_8$: C, 62.30; H, 5.68; N, 3.16. Found: C, 62.41; H, 6.06; N, 2.81.

*Methyl 2-acetamido-3,4-di-O-benzoyl-2-deoxy-6-O-methyl-α-D-mannopyranoside* (**5**). — Compound **4** (40 mg) was dissolved in methyl iodide (4 ml), and silver oxide (100 mg) was added. The mixture was vigorously stirred for 12 h at room temperature, and then boiled under reflux for 1 h. The mixture was cooled, diluted with chloroform, and filtered, and the solids were washed with warm chloroform. The filtrate and washings were combined and evaporated, and the residue was crystallized from ether–hexane to give a chromatographically pure material (31 mg, 75%). Recrystallization from ether–hexane gave prismatic needles, m.p. 168–169°, $[\alpha]_D^{20}$ −9° (*c* 0.70,

chloroform); i.r. data: $\nu_{max}^{KBr}$ 3320 (NH), 1710 (ester), 1670 (Amide I), and 1520 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) dichloromethane–ethanol: $R_F$ 0.40.

*Anal.* Calc. for $C_{24}H_{27}NO_8$: C, 63.01; H, 5.95; N, 3.06; O, 27.98; OMe, 13.55. Found: C, 62.90; H, 5.98; N, 3.09; O, 28.04; OMe, 13.68.

*Methyl 2-acetamido-2-deoxy-6-O-methyl-α-D-mannopyranoside* (**6**). — *Method A. From* **5**. A solution of **5** (21 mg) in dry methanol (3 ml) was treated with 0.1M methanolic sodium methoxide (0.1 ml) for 12 h at 4°, and then for 8 h at room temperature. The solution was de-ionized with Rexyn 300 ($H^+$, $OH^-$) ion-exchange resin (2 ml), and evaporated. The residue was dissolved in methanol, the suspension filtered through charcoal–Celite, and the filtrate evaporated. The syrupy residue was crystallized from ethanol–ether to give needles (9 mg, 78%), m.p. 169–171°; $[\alpha]_D^{24}$ +38° (*c* 0.31, methanol); i.r. data: $\nu_{max}^{KBr}$ 3450–3250 (broad, OH and NH), 1655 (Amide I), and 1545 $cm^{-1}$ (Amide II); t.l.c. in 9:1 (v/v) chloroform–ethanol: $R_F$ 0.16; $R_{TMG}$ 0.31.

*Anal.* Calc. for $C_{10}H_{19}NO_6$: C, 48.19; H, 7.68; N, 5.62; OMe, 24.50. Found: C, 48.00; H, 7.64; N, 5.60; OMe, 25.07.

*Method B. From* **9**. To a solution of **9** (25 mg) in ethanol (3 ml, 90%) was added tris(triphenyl)phosphinerhodium chloride (12 mg, 13 μmoles) and 1,4-diazabicyclo-[2.2.2]octane (5 mg). The mixture was boiled under reflux for 3 h, and the solution cooled and evaporated, the residue dispersed in water, and the suspension extracted with ether. The extract was dried (magnesium sulfate) and evaporated. A solution of the residue in methanol (3 ml) was treated with Dowex-50 X-8 ($H^+$) (100–200 mesh, 2 ml) ion-exchange resin for 12 h at 37°, the suspension filtered, and the filtrate evaporated. The residue was chromatographed on silica gel with 9:1 (v/v) chloroform–ethanol, to give a compound that crystallized as platelets (9 mg), m.p. 170–172°; $[\alpha]_D^{24}$ +37° (*c* 0.42, methanol); t.l.c. in 4:1 (v/v) chloroform–ethanol: $R_F$ 0.16, $R_{TMG}$ 0.31. The first fractions eluted from the silica gel column gave unreacted **9**, which was treated as just described to give additional **6** (total yield: 15 mg, 73%).

*Methyl 2-acetamido-3,4-di-O-allyl-2-deoxy-6-O-trityl-α-D-mannopyranoside* (**7**). — A solution of **1** (150 mg) in dry acetone (1 ml) and benzene (10 ml) was mixed with allyl bromide (54 μl) and powdered sodium hydroxide (500 mg), and the mixture boiled under reflux for 4 h, and cooled; sodium hydroxide (150 mg) was added, and the mixture was stirred for 12 h at room temperature, and diluted with benzene (5 ml). The solid was filtered off and washed with 7:3 (v/v) benzene–acetone (5 ml), the filtrate and washings were combined and evaporated, and the residue was dissolved in chloroform. The solution was washed with water (3 × 5 ml), dried (sodium sulfate), and evaporated. The syrupy residue was chromatographed on silica gel with 19:1 (v/v) chloroform–ethanol. A syrupy material was obtained that crystallized from 2-iso-propoxypropane–heptane to give needles (145 mg, 85%), m.p. 110–112°, $[\alpha]_D^{24}$ +16° (*c* 0.82, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3295 (NH), 1665–1635 (–$CH_2$–CH=$CH_2$, Amide I), and 1560 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) chloroform–ethanol: $R_F$ 0.63.

*Anal.* Calc. for $C_{34}H_{39}NO_6$: C, 73.23; H, 7.05; N, 2.51; O, 17.21. Found: C, 73.20; H, 7.15; N, 2.37; O, 17.10.

*Methyl 2-acetamido-3,4-di-O-allyl-2-deoxy-α-D-mannopyranoside* (**8**). — A solution of **7** (125 mg) in glacial acetic acid (3 ml) was heated on a water bath (80°). The hot solution was diluted with water (1.5 ml), and heating was continued for 45 min. After dilution with cold water (150 ml), the solution was lyophilized, and the residue was chromatographed on silica gel with 14:1 (v/v) dichloromethane–ethanol to give 60 mg (85%) of a syrup, $[\alpha]_D^{24}$ +50° (*c* 0.60, methanol); i.r. data: $\nu_{max}^{KBr}$ 3440 (OH), 3250 (NH), 1660 (–$CH_2$–CH=$CH_2$), 1645 (Amide I), and 1545 $cm^{-1}$ (Amide II); t.l.c. in 14:1 (v/v) chloroform–ethanol: $R_F$ 0.31.

*Anal.* Calc. for $C_{15}H_{25}NO_6$: C, 57.13; H, 7.99; N, 4.44; O, 30.44. Found: C, 57.05; H, 8.01; N, 4.39; O, 30.44.

*Methyl 2-acetamido-3,4-di-O-allyl-2-deoxy-6-O-methyl-α-D-mannopyranoside* (**9**). — Compound **8** (50 mg) in dry tetrahydrofuran (2 ml) was treated with methyl iodide (3 ml) and silver oxide (250 mg) for 6 h under reflux, with vigorous stirring. A further 100 mg of silver oxide was added, and stirring was continued for 12 h. The solids were filtered off and washed with hot chloroform, and the combined filtrate and washings were evaporated. The residue was chromatographed on silica gel. Elution with 19:1 (v/v) chloroform–ethanol gave a material that crystallized from chloroform–heptane in plates (43 mg, 82%), m.p. 73–75°, $[\alpha]_D^{24}$ +44° (*c* 1.00, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3290 (NH), 1670 (–$CH_2$CH=$CH_2$), 1640 (Amide I), and 1550 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) chloroform–ethanol: $R_F$ 0.32.

*Anal.* Calc. for $C_{16}H_{27}NO_6$: C, 58.34; H, 8.26; N, 4.25; O, 29.14. Found: C, 58.27; H, 8.21; N, 4.33; O, 29.11.

*Methyl 2-(N-acetylbenzamido)-3-O-benzoyl-4,6-O-benzylidene-2-deoxy-α-D-mannopyranoside* (**10**). — Methyl 2-acetamido-4,6-*O*-benzylidene-2-deoxy-α-D-mannopyranoside[3] (**11**, 100 mg) in dry pyridine (2 ml) was treated with benzoyl chloride (54 ml) for 20 h at −5°. The mixture was diluted with chloroform (6 ml), and then processed as described for **2**. The chloroform extract was dried (sodium sulfate), filtered, and evaporated. The residue was chromatographed on silica gel with 19:1 (v/v) chloroform–ethanol to give a syrup (90 mg) that crystallized from methanol as needles (78 mg, 47%), m.p. 161–163°, $[\alpha]_D^{26}$ −70° (*c* 0.51, chloroform); i.r. data: $\nu_{max}^{KBr}$ 1715 (ester), 1680 (*N*-Ac and *N*-Bz), 1590 and 1580 (Ar), 1450 (CO), and 740 $cm^{-1}$ (Ar); no Amide II band was observed; t.l.c. in 19:1 (v/v) chloroform–methanol: $R_F$ 0.76.

*Anal.* Calc. for $C_{30}H_{29}NO_8$: C, 67.79; H, 5.50; N, 2.63; O, 24.08. Found: C, 68,24; H, 5.48; N, 2.73; O, 23.88.

*Methyl 2-acetamido-3-O-benzoyl-2-deoxy-4,6-di-O-methyl-α-D-mannopyranoside* (**15**). — Methyl 2-acetamido-3-*O*-benzoyl-2-deoxy-α-D-mannopyranoside[3] (**14**) (25 mg) in dry tetrahydrofuran (4 ml) was treated with methyl iodide (1.5 ml) and silver oxide (100 mg) for 16 h at room temperature, followed by boiling under reflux for 4 h. The solids were filtered off and washed with warm chloroform, the combined filtrate and washings were evaporated, and the residue was chromatographed on silica gel. Elution with 19:1 (v/v) chloroform–ethanol gave 18 mg (65%) of material that, on crystallization from chloroform–ether–pentane, gave platelets containing

0.5 molecule of water per molecule, m.p. 121–123°; $[\alpha]_D^{22}$ +35° (*c* 0.41, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3500 (OH), 3275 (NH), 1740 (ester), 1655 (Amide I), 1550 (Amide II), and 1455 $cm^{-1}$ (Ar); t.l.c. in 19:1 (v/v) chloroform–ethanol: $R_F$ 0.48.

*Anal.* for $C_{18}H_{25}NO_7 \cdot 0.5H_2O$: C, 57.43; H, 6.96; N, 3.72. Found: C, 57.59; H, 6.96; N, 4.12.

*Methyl 2-acetamido-2-deoxy-4,6-di-*O*-methyl-α-*D*-mannopyranoside* (**16**). — *Method A. From* **15**. Compound **15** (13 mg) in dry methanol (2 ml) was treated with 0.1M methanolic sodium methoxide (50 ml) for 18 h at 4°, and then the solution was diluted with methanol, de-ionized with Rexyn 300 ($H^+$, $OH^-$) ion-exchange resin (1 ml), and evaporated. The residue was chromatographed on silica gel with 9:1 (v/v) chloroform–ethanol to give 8 mg (86%) of material that crystallized from cold chloroform–ether–hexane as plates; at room temperature, the crystalline material turned partly amorphous; $[\alpha]_D^{20}$ +49° (*c* 0.36, ethanol); t.l.c. in 9:1 (v/v) chloroform–ethanol: $R_F$ 0.4, $R_{TMG}$ 0.67.

*Anal.* Calc. for $C_{11}H_{21}NO_6$: C, 50.18; H, 8.04; N, 5.32; OMe, 35.36. Found: C, 50.05; H, 8.13; N, 5.21; OMe, 35.05.

*Method B. From* **18**. A solution of **18** (30 mg) in 90% ethanol (4 ml) was treated with tris(triphenyl)phosphinerhodium chloride (8 mg) and 1,4-diazabicyclo[2.2.2]-octane (2 mg). The mixture was processed as described for the preparation of **6** from **9**, and the resulting product was chromatographed on silica gel with 9:1 (v/v) chloroform–ethanol to give an amorphous substance (14 mg) and unconverted **18**. A second treatment gave additional pure **16** (7 mg; total yield 81%), $[\alpha]_D^{20}$ +48° (*c* 0.42, ethanol); t.l.c. in 9:1 (v/v) chloroform–ethanol: $R_F$ 0.4, $R_{TMG}$ 0.67.

*Methyl 2-acetamido-3-*O*-allyl-4,6-*O*-benzylidene-2-deoxy-α-*D*-mannopyranoside* (**13**). — A solution of **11** (170 mg) in dry acetone (2 ml) and benzene (15 ml) was treated with allyl bromide (46 ml) and sodium hydroxide (400 mg). The mixture was boiled under reflux for 2 h, and then cooled, and sodium hydroxide (150 mg) was added. After being vigorously stirred for 12 h at room temperature, the mixture was filtered, and the insoluble material washed with warm, 1:1 (v/v) benzene–acetone (10 ml). The combined filtrate and washings were evaporated, the residue was dissolved in chloroform, and the solution was washed with water, dried (sodium sulfate), and evaporated. The residue was chromatographed on silica gel with 19:1 (v/v) dichloromethane–methanol to give a material that crystallized from 2-isopropoxypropane in prismatic needles (163 mg, 85%), m.p. 93–94°, $[\alpha]_D^{20}$ +9° (*c* 0.52, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3275 (NH), 1670 ($CH_2$–$CH$=$CH_2$), 1635 (Amide I), and 1550 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) dichloromethane–methanol: $R_F$ 0.4.

*Anal.* Calc. for $C_{19}H_{25}NO_6$: C, 62.80; H, 6.93; N, 3.85; O, 26.42. Found: C, 62.79; H, 6.89; N, 3.84; O, 26.49.

*Methyl 2-acetamido-3-*O*-allyl-2-deoxy-α-*D*-mannopyranoside* (**17**). — Compound **13** (140 mg) in 60% acetic acid (5 ml) was treated for 1 h on a water bath (80°). The solution was cooled and concentrated, and toluene was repeatedly added and distilled off. The residue was chromatographed on silica gel; elution with 7:3 (v/v) chloroform–ethanol gave 94 mg (89%) of **17** that tenaciously retained chloroform after being

dried *in vacuo*, as indicated by variable values for its chlorine content; $[\alpha]_D^{22}$ +33° (*c* 0.30, methanol); i.r. data: $\nu_{max}^{film}$ 3400–3250 (broad, OH, NH), 1660–1645 ($CH_2CH=CH_2$, Amide I), 1575 (Amide II), and 750 $cm^{-1}$ (C–Cl); t.l.c. in 4:1 (v/v) chloroform–ethanol: $R_F$ 0.60.

*Anal.* Calc. for $C_{12}H_{21}NO_6 \cdot 0.1CHCl_3$: C, 50.59; H, 7.40; N, 4.88. Found: C, 50.32; H, 7.49; N, 4.64.

*Methyl 2-acetamido-3-O-allyl-2-deoxy-4,6-di-O-methyl-α-D-mannopyranoside* (**18**). — A solution of **17** (65 mg) in dry tetrahydrofuran (2 ml) was mixed with methyl iodide (3 ml) and silver oxide (250 mg), and the mixture was boiled under reflux for 12 h. After a new addition of silver oxide (150 mg), the mixture was boiled under reflux for 8 h, and then cooled to room temperature and filtered. The residue was washed with warm chloroform, and the combined filtrate and washings were evaporated. The residue was chromatographed on silica gel with 19:1 (v/v) chloroform–ethanol to give a product that crystallized from chloroform–heptane as needles (59 mg, 82%), m.p. 86–88°, $[\alpha]_D^{24}$ +36° (*c* 0.50, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3275 (NH), 1670 ($CH_2$–CH=$CH_2$), 1635 (Amide I), and 1550 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) chloroform–ethanol: $R_F$ 0.25.

*Anal.* Calc. for $C_{14}H_{25}NO_6$: C, 55.43; H, 8.31; N, 4.62; O, 31.64. Found: C, 55.34; H, 8.29; N, 4.68; O, 31.58.

*Methyl 2-acetamido-2-deoxy-3-O-methyl-6-O-trityl-α-D-mannopyranoside* (**20**). — A solution of methyl 2-acetamido-2-deoxy-3-*O*-methyl-α-D-mannopyranoside[3] (**19**, 400 mg) in dry pyridine (40 ml) was treated with chlorotriphenylmethane (530 mg) for 6 h at 100°. The mixture was cooled and evaporated; repeated addition and distillation of toluene gave a residue that, on examination by t.l.c. in 14:1 (v/v) chloroform–ethanol, showed three components, corresponding to triphenylmethanol, unreacted **19**, and **20**. The mixture was chromatographed on silica gel with 14:1 (v/v) chloroform–ethanol to give a material that crystallized from ether as microcrystals (480 mg, 61%), m.p. 182–184°, $[\alpha]_D^{20}$ +3° (*c* 0.70, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3290 (NH), 1645 (Amide I), 1545 (Amide II), 1490, and 1450 $cm^{-1}$ (Ar); t.l.c. in 14:1 (v/v) chloroform–ethanol: $R_F$ 0.31.

*Anal.* Calc. for $C_{29}H_{33}NO_6$: C, 70.86; H, 6.77; N, 2.85; O, 19.53. Found: C, 70.74; H, 6.89; N, 2.75; O, 19.78.

From the later fractions from the silica gel column, **19** was recovered; it was converted into **20** to give an additional 180 mg of the latter.

*Methyl 2-acetamido-4-O-benzoyl-2-deoxy-3-O-methyl-6-O-trityl-α-D-mannopyranoside* (**21**). — A solution of **20** (270 mg) in dry pyridine (10 ml) was cooled to −60°, treated with benzoyl chloride (65 μl) for 6 h at −20° and 12 h at −10°, diluted with chloroform (10 ml), and processed as described for **2**. Chromatography of the resulting syrup on silica gel with 24:1 (v/v) chloroform–ethanol gave a material that crystallized from ether–pentane as prisms (240 mg, 76%), m.p. 198–199° (softening at 145°), $[\alpha]_D^{20}$ +12° (*c* 0.54, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3300 (NH), 1720 (ester), 1650 (Amide I), 1550 (Amide II), 1490, 1450 (Ar), and 700 $cm^{-1}$ (Ar); t.l.c. in 19:1 (v/v) dichloromethane–ethanol: $R_F$ 0.50.

*Anal.* Calc. for $C_{36}H_{37}NO_7$: C, 72.59; H, 6.26; N, 2.35; O, 18.80. Found: C, 72.54; H, 6.36; N, 2.39; O, 18.73.

*Methyl 2-acetamido-4-O-benzoyl-2-deoxy-3-O-methyl-α-D-mannopyranoside* (**22**). — Compound **21** (240 mg) was heated with 60% acetic acid (12 ml) for 1 h on a water bath (80°). The solution was cooled, diluted with water (150 ml), and lyophilized. The residue was chromatographed on silica gel with 14:1 (v/v) dichloromethane–ethanol to give a material that crystallized from chloroform–ether–pentane as plates (100 mg, 70%) containing 0.5 molecule of pentane per molecule, m.p. 76–77°, $[\alpha]_D^{20}$ −3° (*c* 1.32, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3440–3280 (broad, OH, NH), 1740 (ester), 1655 (Amide I), and 1540 $cm^{-1}$ (Amide II); t.l.c. in 14:1 (v/v) dichloromethane–ethanol: $R_F$ 0.22.

*Anal.* Calc. for $C_{17}H_{23}NO_7 \cdot 0.5C_5H_{10}$: C, 60.29; H, 7.26; N, 3.60. Found: C, 60.24; H, 6.83; H, 4.06.

*Methyl 2-acetamido-4-O-benzoyl-2-deoxy-3,6-di-O-methyl-* (**23**) *and methyl 2-acetamido-6-O-benzoyl-2-deoxy-3,4-di-O-methyl-α-D-mannopyranoside* (**24**). — Compound **22** (80 mg) in dry tetrahydrofuran (2 ml) was treated with methyl iodide (4 ml) and silver oxide (300 mg) for 24 h at room temperature, with vigorous stirring. A further 150 mg of silver oxide was added, and stirring was continued for 16 h. T.l.c. in 19:1 (v/v) chloroform–ethanol showed maximum conversion of **22** into two components moving very closely. After processing the mixture as described for **15**, the residue was chromatographed on silica gel with 29,1 (v/v) chloroform–ethanol. Early fractions gave an amorphous solid (50 mg, 60%) having 0.5 molecule of hexane per molecule, $[\alpha]_D^{20}$ +19° (*c* 0.24, chloroform); i.r. data: $\nu_{max}^{film}$ 3270 (NH), 1745 (ester), 1655 (Amide I), and 1550 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) chloroform–ethanol, $R_F$ 0.23.

*Anal.* Calc. for $C_{18}H_{25}NO_7 \cdot 0.5C_6H_{12}$: C, 61.60; H, 7.63. Found: C, 61.52; H, 8.07.

T.l.c. of the later fractions eluted from the column indicated the presence of two components (**23** and **24**); these were not separable by use of 29:1 (v/v) chloroform–ethanol, 29:1 (v/v) dichloromethane–ethanol, 19:1 or 14:1 (v/v) benzene–methanol, or 14:1 (v/v) ether–acetone. The mixture of **23** and **24** (22 mg) in methanol (2 ml) was treated for 16 h at 4° with 0.1M methanolic sodium methoxide (50 ml). The solution was de-ionized with Rexyn 300 ($H^+$, $OH^-$) ion-exchange resin, and examination of the product by t.l.c. in 19:1 (v/v) chloroform–ethanol showed the presence of two components. Examination of the mixture by g.l.c. after preparation of the per(trimethylsilyl) ethers indicated the presence of two components, one corresponding to methyl 2-acetamido-2-deoxy-3,4-di-*O*-methyl-6-*O*-(trimethylsilyl)-α-D-mannopyranoside. The mixture (16 mg) was chromatographed on silica gel in 19:1 (v/v) chloroform–ethanol. G.l.c.–m.s. of the isolated components as the per(trimethylsilyl) ethers showed the products to be methyl 2-acetamido-2-deoxy-3,6-di-*O*-methyl-α-D-mannopyranoside and methyl 2-acetamido-2-deoxy-3,4-di-*O*-methyl-α-D-mannopyranoside.

*Methyl 2-acetamido-2-deoxy-3,6-di-O-methyl-α-D-mannopyranoside* (**25**). — *Method A. From* **23**. A solution in dry methanol (4 ml) of the early fractions of the preparation of **23** (40 mg) was treated with 0.1M methanolic sodium methoxide

(100 ml) for 20 h at 4°, and the solution was processed as described for **16**. The residue was chromatographed on silica gel with 19:1 (v/v) chloroform–ethanol to give a material that crystallized from chloroform–hexane in the cold, and turned amorphous (21 mg, 76%) at room temperature, $[\alpha]_D^{24}$ +27° (*c* 1.13, methanol); i.r. data: $\nu_{max}^{film}$ 3450–3250 (OH, NH), 1655 (Amide I), and 1540 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) chloroform–ethanol: $R_F$ 0.23, $R_{TMG}$ 0.72.

*Anal.* Calc. for $C_{11}H_{21}NO_6$: C, 50.18; H, 8.04; N, 5.32. Found: C, 50.00; H, 7.98; N, 5.06.

*Method B. From* **28**. A solution of **28** (32 mg) in 90% ethanol (5 ml) was treated with tris(triphenyl)phosphinerhodium chloride (8 mg) and 1,4-diazabicyclo[2.2.2]-octane (2.5 mg). The mixture was processed as described for compound **6**, and the residue was chromatographed on silica gel with 14:1 (v/v) dichloromethane–ethanol to give an amorphous solid (14 mg). The earlier fractions eluted from the column contained **28**, which was again treated as just described, to give an additional amount of **25** (7 mg; total yield 76%), $[\alpha]_D^{24}$ +28° (*c* 0.42, methanol); t.l.c. in 19:1 (v/v) dichloromethane–ethanol: $R_F$ 0.23, $R_{TMG}$ 0.72; g.l.c.–m.s. of the per(trimethylsilyl) ether showed the product to be identical to that obtained from **23**.

*Methyl 2-acetamido-4-*O*-allyl-3-*O*-methyl-6-*O*-trityl-α-*D*-mannopyranoside* (**26**). — A solution of **20** (200 mg) in dry benzene (20 ml) was treated with allyl bromide (36 ml) and powdered sodium hydroxide (600 mg). The mixture was boiled under reflux for 3 h with vigorous stirring. A further 200 mg of sodium hydroxide was added, and stirring was continued for 12 h at room temperature. The mixture was processed as described for **7**, and chromatography of the resulting syrup with 19:1 (v/v) dichloromethane–ethanol gave a material that crystallized from chloroform–ether as needles (173 mg, 80%), m.p. 138–140°, $[\alpha]_D^{25}$ +18° (*c* 0.71, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3300 (NH), 1660 ($CH_2$–CH=$CH_2$), 1645 (Amide I), and 1540 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) dichloromethane–ethanol: $R_F$ 0.56.

*Anal.* Calc. for $C_{32}H_{37}NO_6$: C, 72.30; H, 7.01; N, 2.63; O, 18.06. Found: C, 72.21; H, 7.00; N, 2.58; O, 18.08.

*Methyl 2-acetamido-4-*O*-allyl-3-*O*-methyl-α-*D*-mannopyranoside* (**27**). — Compound **26** (150 mg) was treated with 60% acetic acid (6 ml) for 1 h on a water bath (80°). The solution was cooled and concentrated, and toluene was repeatedly added and distilled off. Chromatography of the residue on silica gel with 9:1 (v/v) chloroform–ethanol gave 70 mg (85%) of amorphous **27**, $[\alpha]_D^{24}$ −38° (*c* 0.67, chloroform); t.l.c. in 19:1 (v/v) chloroform–ethanol: $R_F$ 0.47.

*Anal.* Calc. for $C_{13}H_{23}NO_6$: C, 53.97; H, 8.01; N, 4.84; O, 33.18. Found: C, 53.89; H, 7.93; N, 4.84; O, 33.29.

*Methyl 2-acetamido-4-*O*-allyl-3,6-di-*O*-methyl-α-*D*-mannopyranoside* (**28**). — A solution of **27** (55 mg) in dry acetone (2 ml) mixed with methyl iodide (3 ml) and silver oxide (200 mg) was boiled for 12 h under reflux. A further 100 mg of silver oxide was added, and heating was continued for 8 h. After the mixture had been processed as described for **15**, the residue was chromatographed on silica gel. Elution with 19:1 (v/v) dichloromethane–ethanol gave a material that crystallized from chloroform–

ether as prisms (45 mg, 78%), m.p. 116–118°, $[\alpha]_D^{20}$ +48° (*c* 0.95, chloroform); i.r. data: $\nu_{max}^{KBr}$ 3300 (NH), 1660–1640 ($CH_2$–CH=$CH_2$, Amide I), and 1540 $cm^{-1}$ (Amide II); t.l.c. in 19:1 (v/v) dichloromethane–ethanol: $R_F$ 0.29.

*Anal.* Calc. for $C_{14}H_{25}NO_6$: C, 55.43; H, 8.31; N, 4.62; O, 31.64. Found: C, 55.35; H, 8.25; N, 4.65; O, 31.72.

## REFERENCES


1 J. D. Higginbotham and M. Heidelberger, *Carbohyd. Res.*, 23 (1972) 165–173.
2 H. R. Perkins, *Biochem. J.*, 86 (1963) 475–483; M. Torii, K. Sakakibara, and K. Kuroda, *Eur. J. Biochem.*, 37 (1973) 401–405.
3 Nasir-ud-Din and R. W. Jeanloz, *Carbohyd. Res.*, 28 (1973) 243–251.
4 J. Gigg and R. Gigg, *J. Chem. Soc., C*, (1966) 82–86; R. Gigg and C. D. Warren, *ibid.*, (1968) 1903–1911; P. A. Gent, R. Gigg, and R. Conant, *J. Chem. Soc. Perkin I*, (1972) 1535–1542.
5 E. J. Corey and J. W. Suggs, *J. Org. Chem.*, 38 (1973) 3224.
6 R. W. Jeanloz, *J. Amer. Chem. Soc.*, 76 (1954) 555–558.
7 K. Heyns, K. Propp, R. Harrison, and H. Paulsen, *Chem. Ber.*, 100 (1967) 2655–2663.
8 T. Purdie and J. C. Irvine, *J. Chem. Soc.*, 83 (1903) 1021–1037; H. Hibbert, R. S. Tipson, and F. Brauns, *Can. J. Res.*, 4 (1931) 221–239.
9 M. D. A. Halford and R. W. Jeanloz, unpublished results.
10 R. W. Jeanloz, *J. Amer. Chem. Soc.*, 74 (1952) 4597–4599.
11 P. J. Stoffyn and R. W. Jeanloz, *J. Amer. Chem. Soc.*, 80 (1958) 5690–5692.
12 D. K. Stearns, R. G. Naves, and R. W. Jeanloz, *J. Org. Chem.*, 26 (1961) 901–905.
13 R. W. Jeanloz, D. M. Schmid, and P. J. Stoffyn, *J. Amer. Chem. Soc.*, 79 (1957) 2586–2590.
14 R. W. Jeanloz, *J. Org. Chem.*, 26 (1961) 905–908.
15 V. N. Reinhold, *Methods Enzymol.*, 25 (1972) 244–249.