

Benzyl Glycosides from 2-Acetamido-2-deoxy-D-mannose. Benzyl 2-Acetamido-2-deoxy- α -D-mannofuranoside and Related Derivatives

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A recent paper from this laboratory³ has described the direct benzylation of 2-acetamido-2-deoxy-D-glucose and of 2-acetamido-2-deoxy-D-galactose in *N,N*-dimethylformamide solution using a mixture of barium oxide, barium hydroxide octahydrate, and benzyl bromide. In each case, the corresponding benzyl 2-acetamido-3,4,6-tri-*O*-benzyl-2-deoxy- β -D-hexopyranoside was obtained. We now wish to report, however, that the action of this combination of reagents on 2-acetamido-2-deoxy-D-mannose (1) gives benzyl 2-acetamido-3,4,6-tri-*O*-benzyl-2-deoxy- β -D-glucopyranoside (2) in 24% yield; it is, therefore, evident that at least considerable epimerization, prior to benzylation, makes this convenient combination of reagents unsuitable for the direct preparation of benzyl 2-acetamido-3,4,6-tri-*O*-benzyl-2-deoxy-D-mannopyranoside from 1. Attention was, therefore, turned to a possible two-step synthesis of such a substance, the first step being the preparation of a benzyl 2-acetamido-2-deoxy-D-mannopyranoside under nonalkaline conditions.

Yoshimura and his associates⁴ have described a study of the use of various Lewis acids for the conversion of *N*-acyl derivatives of 2-amino-2-deoxy-D-glucose to alkyl and aralkyl 2-acylamino-2-deoxy-D-glucosides. In particular, they reported the formation of a mixture of the anomeric benzyl 2-acetamido-2-deoxy-D-glucopyranosides through the action of benzyl alcohol on 2-acetamido-2-deoxy-D-glucose in the presence of boron trifluoride etherate. We therefore treated 2-acetamido-2-deoxy-D-mannose (1) in a similar manner. For convenience in chromatography, the initial reaction product was acetylated. However, extensive chromatography on columns of silica gel failed to resolve the resulting mixture completely although evidence (glpc and tlc) indicated that the main product was benzyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy- α -D-mannopyranoside (3), a substance for which a wholly different and somewhat more practicable synthetic pathway will be described shortly.⁵

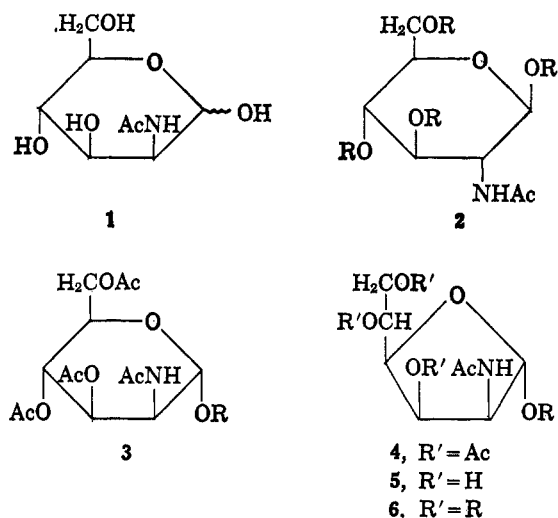
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(3) R. Harrison and H. G. Fletcher, Jr., *J. Org. Chem.*, **30**, 2317 (1965).

(4) T. Yoshimura, H. Ando, Y. Takahashi, H. Ono, and T. Sato, *Nippon Kagaku Zasshi*, **85**, 142 (1964); *Chem. Abstr.*, **62**, 623 (1965).

(5) N. Pravdić, T. D. Inch, and H. G. Fletcher, Jr., *J. Org. Chem.*, **32**, 1815 (1967).



A second component of the reaction mixture, perhaps the as yet unknown benzyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy- β -D-mannopyranoside, was detected by glpc but not identified. A third and minor product was obtained in crystalline form and shown to be benzyl 2-acetamido-3,5,6-tri-*O*-acetyl-2-deoxy- α -D-mannofuranoside (4); on de-*O*-acetylation of this substance benzyl 2-acetamido-2-deoxy- α -D-mannofuranoside (5) was obtained. The latter compound was found to reduce 1.06 molar equiv of periodate, liberating at the same time 0.97 molar equiv of formaldehyde; the furanoside structure of 4 and 5 was thus established. Benzylation of 5 in *N,N*-dimethylformamide solution with barium oxide, barium hydroxide octahydrate, and benzyl bromide afforded the tribenzyl ether 6. That the mannose configuration had been preserved was proved by successive hydrogenation and trimethylsilylation of 6; the product thus obtained was indistinguishable by glpc from that prepared through the trimethylsilylation of 1.

In view of the comparatively large dextrorotations of 4 and 5⁶ [$[\alpha]^{20}_D +128^\circ$ (CHCl₃) and $+132^\circ$ (CH₃OH), respectively] the α -D anomeric configuration is assigned to these substances. While not readily analyzable, the nmr spectrum of 4 included a one-proton singlet at τ 4.93, and the nmr spectrum of 6 contained a one-proton singlet at 5.02. Extensive experience with furanose derivatives has shown⁷ that H₁ in these substances normally appears as a singlet when H₁ is *trans* to H₂.

No attempt was made to maximize the yield of 4; the object of this Note is to draw attention to the fact

(6) The tri-*O*-benzyl derivative 6 had $[\alpha]^{20}_D +25.1^\circ$ (CHCl₃). However, since all of the fully benzylation 2-acylamino-2-deoxyhexopyranosides which have been made in this laboratory have specific rotations of approximately this numerical magnitude, it seems unwise to put much reliance on this value in assigning anomeric configurations. Cf. ref 3 and also T. D. Inch and H. G. Fletcher, Jr., *ibid.* **31**, 1810 (1966).

(7) C. P. J. Glaudemans and H. G. Fletcher, Jr., *J. Am. Chem. Soc.*, **87**, 4636 (1965), footnote 6.

that the process of Yoshimura, *et al.*,⁴ cannot with certainty be relied upon to give pyranosides exclusively.⁸

Experimental Section⁹

Benzylation of 2-Acetamido-2-deoxy-D-mannose (1) under Alkaline Conditions.—A solution of 1 (1.5 g) in a mixture of *N,N*-dimethylformamide (30 ml) and benzyl bromide (10 ml) was cooled to -10° and stirred while barium oxide (9.75 g of freshly ground lumps) and barium hydroxide octahydrate (3.75 g) were added. The reaction mixture was stirred at -10 to 0° for 5 hr and then at room temperature for 18 hr. The solid was removed by filtration on Celite and washed thoroughly with dichloromethane. The combined filtrate and washings were washed several times with water, dried with magnesium sulfate, and concentrated *in vacuo*, finally being held at *ca.* 1 mm and 100° (bath) to remove the excess of benzyl bromide. On treatment with ethanol and cooling, the residual material afforded crystalline benzyl 2-acetamido-3,4,6-tri-*O*-benzyl-2-deoxy- β -D-glucopyranoside (2, 0.95 g, 24%). After recrystallization from ethanol, the product was obtained as needles: mp 166 – 167° , $[\alpha]_{20}^D -13.4^{\circ}$ (*c* 0.95, CHCl_3). Harrison and Fletcher³ reported mp 160 – 165° and $[\alpha]_{20}^D -12.1^{\circ}$ (*c* 1.3, CHCl_3) for this substance, mmp 168 – 169° . Hydrogenation of a sample of the product in methanol solution and in the presence of palladium catalyst gave, after removal of the catalyst and solvent, a syrup which was trimethylsilylated; glpc failed to distinguish the product from the trimethylsilyl derivative of 2-acetamido-2-deoxy-D-glucose but readily showed it to be different from the trimethylsilyl derivative of 1.

Another sample of 2 was freed of its benzyl groups by catalytic hydrogenolysis and then acetylated with pyridine and acetic anhydride in conventional fashion to yield a crystalline product: mp 136 – 137° , $[\alpha]_{20}^D +88.5^{\circ}$ (*c* 0.26, CHCl_3). The infrared spectrum of this product was identical with that of an authentic specimen of 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxy- α -D-glucopyranose,¹⁰ mp 137 – 138° .

Reaction of 2-Acetamido-2-deoxy-D-mannose (1) with Benzyl Alcohol in the Presence of Boron Trifluoride.—A sample of 1 (11.6 g) which had been dried thoroughly *in vacuo* was stirred for 2 hr at 110 – 115° (bath) with a mixture of redistilled benzyl alcohol (70 ml) and boron trifluoride etherate (2 ml). The mixture was then cooled and poured into ether, and the brown syrup which precipitated was separated by decantation and acetylated with a mixture of acetic anhydride (40 ml) and pyridine (60 ml). The acetylation mixture was poured onto ice and the product was extracted with dichloromethane, the combined extracts being washed successively with dilute hydrochloric acid, aqueous sodium bicarbonate solution, and then with water. Moisture was removed with magnesium sulfate, then the solution was concentrated *in vacuo* to a brown syrup which was dissolved in ether, and this solution was poured on a column of silica gel (100 g). Elution with ether removed benzyl acetate from the column; ether containing 2% (v/v) of methanol was then used for elution to give, after evaporation of the eluate, a pale orange syrup (12.6 g). A portion (10.4 g) of this syrup was dissolved in ether and rechromatographed on a column of silica gel (300 g). The column was washed with ether (500 ml) and the product was then eluted with ether containing 2% (v/v) of methanol, 100-ml fractions being collected. The fractions were examined by tlc (1% methanol in ether) and pooled and evaporated to give three portions: (A) 5.8 g, 31%; (B) 2.7 g, 14%; (C) 0.9 g, 5%. Portion C proved to be benzyl 2-acetamido-3,5,6-tri-*O*-acetyl-2-

deoxy- α -D-mannofuranoside (4); recrystallized from ether, it was obtained as fluffy needles: mp 152° (after softening at 145°), $[\alpha]_{20}^D +128^{\circ}$ (*c* 0.94, CHCl_3), nmr signals at τ 4.93 and 5.00 (one proton singlets).

Anal. Calcd for $\text{C}_{21}\text{H}_{27}\text{NO}_9$ (437.45): C, 57.66; H, 6.22; N, 3.20. Found: C, 57.91; H, 5.95; N, 3.37.

Portion B was clearly a mixture (tlc); portion A was rechromatographed on a column of silica gel (3×54 cm) using ether containing 4% methanol as eluent and collecting 15-ml fractions. From fractions 46–54 was obtained a clear syrup which appeared to be homogeneous and identical with benzyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy- α -D-mannopyranoside (3)⁵ (tlc, ether-methanol, 19:1, v/v, and benzene-ether-methanol, 5:5:1, v/v). A sample (435 mg) of it was deacetylated with barium methoxide in conventional fashion to yield a foam (357 mg) which was trimethylsilylated and examined by glpc at 230° . Three peaks were observed. The first and major peak had a retention time identical with that of the trimethylsilyl derivative of benzyl 2-acetamido-2-deoxy- α -D-mannopyranoside.⁵ The second component migrated slightly faster than the first and was not identified; it may have been the as yet unknown benzyl 2-acetamido-2-deoxy- β -D-mannopyranoside. The third (and very minor) component had a retention time identical with that of the trimethylsilyl derivative of benzyl 2-acetamido-2-deoxy- α -D-mannofuranoside (5, see below).

Benzyl 2-Acetamido-2-deoxy- α -D-mannofuranoside (5).—A sample of 4 (230 mg) was dissolved in absolute methanol (5 ml) containing 0.1 *N* sodium methoxide (1 ml) and the solution was stored at room temperature. After 30 min, tlc showed the reaction to be complete. Amberlite IR-120 (H^+) was added and the solution was stirred, filtered, and then concentrated *in vacuo* to yield a crystalline residue (150 mg, 92%). The glycoside was recrystallized from methanol-hexane: mp 172 – 173° , $[\alpha]_{20}^D +132^{\circ}$ (*c* 0.3, CH_3OH).

Anal. Calcd for $\text{C}_{15}\text{H}_{21}\text{NO}_8$ (311.35): C, 57.87; H, 6.80; N, 4.50. Found: C, 57.75; H, 6.84; N, 4.37.

A sample of 5 (71.2 mg) was found to have consumed 1.06 molar equiv of sodium metaperiodate after 24 hr at room temperature. Analysis by the chromotropic acid method¹¹ showed the presence of 0.97 molar equiv of formaldehyde.

The trimethylsilyl derivative of 5 had a longer retention time than that of benzyl 2-acetamido-2-deoxy- α -D-mannopyranoside;⁵ in passing, it may be noted that the retention times of the trimethylsilyl derivatives of both of the methyl D-mannofuranosides have been found¹² to be greater than either of those of the methyl D-mannopyranosides.

Benzyl 2-Acetamido-3,5,6-tri-*O*-benzyl-2-deoxy- α -D-mannofuranoside (6).—One gram of 4 was dissolved in a mixture of *N,N*-dimethylformamide (20 ml) and benzyl bromide (6 ml), and the solution was cooled to 0° . To the stirred solution were added barium oxide (5 g of freshly powdered lumps) and barium hydroxide octahydrate (2 g). The suspension was stirred at 0° for 5 hr and then at room temperature for 15 hr; it was then diluted with dichloromethane and filtered through a layer of Celite, the residue being washed thoroughly with dichloromethane. The combined filtrate and washings were washed with water, dried with magnesium sulfate, and concentrated *in vacuo* to give a syrup which was held at *ca.* 1 mm and 120° (bath); on cooling, it crystallized. After recrystallization from methanol, the product was obtained as fluffy needles: 1.25 g (94%), mp 150° , $[\alpha]_{20}^D +25.1^{\circ}$ (*c* 0.23, CHCl_3), nmr signal at τ 5.02 (one-proton singlet).

Anal. Calcd for $\text{C}_{38}\text{H}_{39}\text{NO}_8$ (581.72): C, 74.33; H, 6.76; N, 2.41. Found: C, 74.35; H, 6.82; N, 2.29.

A sample of 6 (184 mg) in methanol solution was shaken with hydrogen in the presence of palladium catalyst. After removal of the catalyst and the solvent, the product was trimethylsilylated in conventional fashion; glpc showed the product to be indistinguishable from the trimethylsilyl derivative of 1.

Registry No.—2, 4171-69-1; 4, 10299-93-1; 5, 10299-94-2; 6, 10299-95-3.

(11) J. C. Speck, Jr., in "Methods in Carbohydrate Chemistry," Vol. 1, R. L. Whistler and M. L. Wolfrom, Eds., Academic Press Inc., New York, N. Y., 1962, p 441.

(12) V. Smirnyagin, C. T. Bishop, and F. P. Cooper, *Can. J. Chem.*, **43**, 3109 (1965).

(8) Yoshimura, *et al.*,⁴ assumed that only pyranosides had been formed; however, in only two cases (those of the anomeric ethyl 2-acetamido-2-deoxy-D-glucopyranosides) did they actually confirm the pyranoside ring structure of their products.

(9) Melting points are corrected. Thin layer chromatography was conducted on silica gel G (E. Merck A.G., Darmstadt) using the solvent systems specified, components being detected by spraying with 10% sulfuric acid and heating at 100° . Column chromatography was carried out with silica gel (0.05–0.20 mm) of E. Merck A.G. An F & M Model 500 instrument, equipped with a flame ionization detector, was used for glpc; the column employed (0.25 in. \times 6 ft) was filled with 3% SE 52 on Gaschrom A (Applied Science Laboratories, Inc., State College, Pa.). The "Tri-Sil" reagent of the Pierce Chemical Co., Rockford, Ill., was used for the preparation of trimethylsilyl derivatives. Nmr spectra were obtained in CDCl_3 solution using a Varian A-60 spectrometer and tetramethylsilane as an internal standard.

(10) O. Westphal and H. Holzmann, *Ber.*, **75**, 1274 (1942).

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Synthesis of Some 3'-O-Methyl Purine Ribonucleosides¹

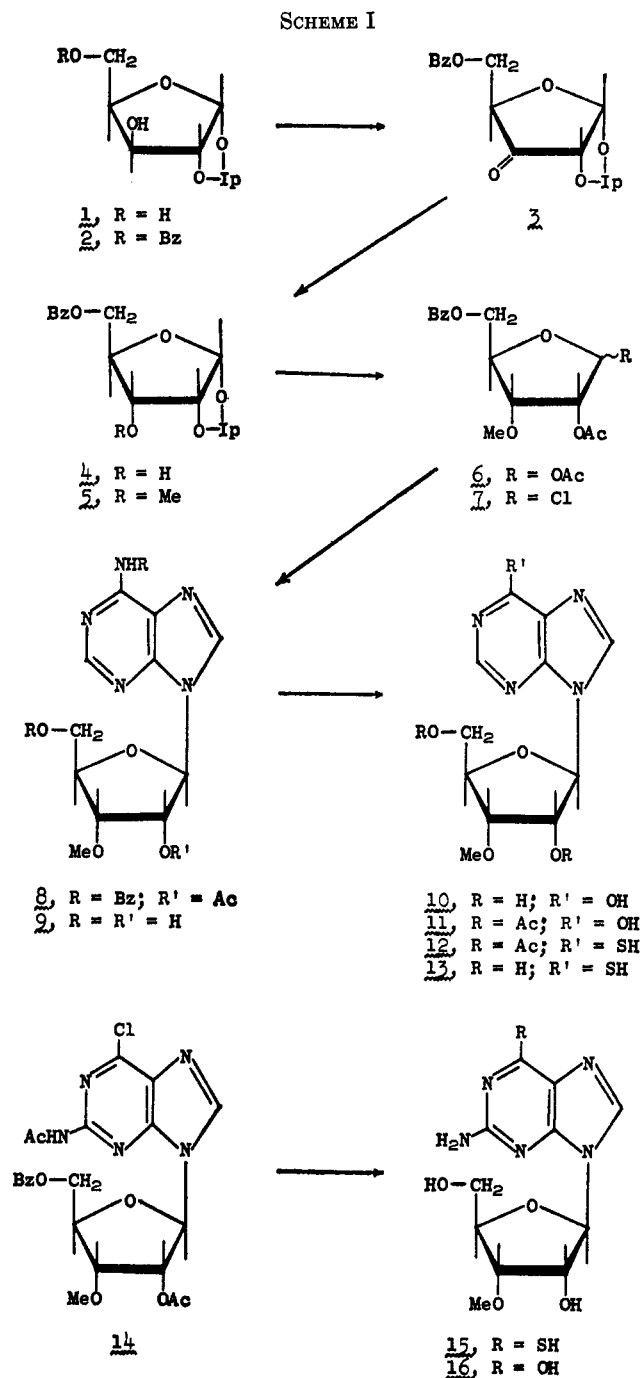
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A wide variety of methylated nucleosides have been isolated by degradation of ribonucleic acids.² Some of the methylated derivatives have been 2'-O-methyl ribonucleosides, and, recently, Robins and co-workers³ have described elegant methods for synthesizing these interesting compounds. We have been interested in the isomeric 3'-O-methyl ribonucleosides, both because one can imagine these as occurring in certain of the nucleic acids and as compounds where the alteration in the sugar moiety could result in unique biological properties. Certainly the change from a β -D-ribose to β -D-arabinoside and β -D-xyloside in adenine nucleosides has had profound biological effects.^{4,5} Other changes in the sugar moiety may be equally useful. The synthesis of 3'-O-methyluridine has recently been reported by Furukawa, *et al.*,⁶ using a method that seems restricted to the pyrimidine nucleosides. We report in this manuscript a preparation of 3'-O-methyl-D-ribose that permits a general synthesis of 3'-O-methylribofuranoside nucleosides and its application to the synthesis of some new purine 3'-O-methylribosides.

The key intermediate in the synthesis of the 3'-O-methyl nucleosides was 5-O-benzoyl-1,2-O-isopropylidene-3-O-methyl- α -D-ribofuranose (**5**) (see Scheme I). Although its precursor, **4**, is known,⁷ the literature preparation required considerable modification before **4** could be conveniently prepared. In the conversion of 1,2-O-isopropylidene- α -D-ribose **1** into the 5-O-benzoyl derivative **2**,⁸ it was not necessary to attempt the difficult isolation of crystalline **2**. The syrupy **2**, obtained in high yield, was equally suitable for oxidation to the crystalline ketone **3** where purification was easy. Oxidation of **2** with acetic anhydride and dimethyl sulfoxide⁹ was more convenient than the original chro-



mium trioxide procedure⁷ or the use of ruthenium tetroxide.^{10,11} Catalytic hydrogenation of **3** at 4 atm proceeded as satisfactorily to afford **4** as the hydrogenation at 60 atm.⁷

Reaction of **4** with methyl iodide and silver oxide in *N,N*-dimethylformamide¹² afforded **5**. Examination of the crude methylation product by nmr indicated the presence of a small amount of a second methyl ether that was removed during recrystallization. The high yield of crystalline **5** suggested that acyl migration was an insignificant factor in this methylation.¹³

- (10) V. M. Parikh and J. K. N. Jones, *Can. J. Chem.*, **43**, 3452 (1965).
 (11) R. F. Nutt, B. Arison, F. W. Holly, and E. Walton, *J. Am. Chem. Soc.*, **87**, 3273 (1965).
 (12) N. F. Taylor and G. M. Riggs, *J. Chem. Soc.*, 5600 (1963).
 (13) See W. A. Bonner, *J. Org. Chem.*, **24**, 1388 (1959), for a discussion of acyl migration during methylation of carbohydrates. See E. G. Gros and S. M. Flematti, *Chem. Ind.* (London), 1556 (1966), for the suggested use of diazomethane and boron trifluoride-etherate to avoid acyl migration during methylation.

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(2) See references given by R. H. Hall, *Biochemistry*, **3**, 876 (1964).

(3) (a) A. D. Broom and R. K. Robins, *J. Am. Chem. Soc.*, **87**, 1145 (1965);

(b) T. A. Khwaja and R. K. Robins, *ibid.*, **88**, 3640 (1966).

(4) (a) J. J. Brink and G. A. LePage, *Cancer Res.*, **24**, 312 (1964); (b) J. J. Brink and G. A. LePage, *Can. J. Biochem.*, **43**, 1 (1965).

(5) D. B. Ellis and G. A. LePage, *ibid.*, **43**, 617 (1965).

(6) Y. Furukawa, K. Kobayashi, Y. Kanai, and M. Honjo, *Chem. Pharm. Bull.* (Tokyo), **13**, 1273 (1965).

(7) K. Oka and H. Wada, *Yakugaku Zasshi*, **83**, 890 (1963); *Chem. Abstr.*, **60**, 1825 (1964).

(8) P. A. Levene and A. L. Raymond, *J. Biol. Chem.*, **102**, 317 (1933).

(9) (a) We are indebted to Kenneth J. Ryan of these laboratories for demonstrating the utility of the acetic anhydride-dimethyl sulfoxide oxidation^{9b} for converting **2** into **3**. (b) J. D. Albright and L. Goldman, *J. Am. Chem. Soc.*, **87**, 4214 (1965).