N-Acyl Derivatives of 2-Acylamino-2-deoxyhexoses. The Rearrangement of 2-(N-Acetylbenzamido)-2-deoxy-D-glucose

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On heating in chloroform solution, 2-(*N*-acetylbenzamido)-2-deoxy-D-glucose (II) undergoes rearrangement and, after acetylation, four substances were isolated. One of these, a minor product, is the tetraacetate of II and presumably arose from unchanged II. A second, minor product was shown to be 1,3,4,6-tetra-*O*-acetyl-2-benzamido-2-deoxy-α-D-glucopyranose (III). A third and major product proved to be 2-acetamido-3,4,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose (IV) which was independently obtained through the acetylation of the previously known 2-acetamido-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose (VII). The fourth product, formed in substantial quantity, had an nmr spectrum and mass spectrogram suggestive of a furanose structure. Ethyl 2-acetamido-3,5,6-tri-*O*-acetyl-2-deoxy-1-thio-α-D-glucofuranoside (XI) was *N*-benzoylated and the glycosidic link hydrolyzed to give a product with an nmr spectrum indistinguishable from the fourth product. Treatment of XI with mercuric benzoate in acetonitrile replaced the ethylthio group with a benzoyloxy group giving an anomeric mixture with an nmr spectrum which included the signals given by the fourth product. On this evidence, the latter is deemed to be 2-acetamido-3,5,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucofuranose (V). From the structures of the products isolated, the rearrangement of II is seen to consist of N → O acyl migrations. Either an acetyl or a benzoyl group shifts from the nitrogen atom to the oxygen atom at C-1 in a *cis* relationship, benzoyl migration predominating over acetyl migration. 2-Acetamido-3-*O*-benzoyl-2-deoxy-D-glucose was synthesized and its triacetate prepared for comparison purposes. No product from the rearrangement of II and subsequent acetylation matched this material in chromatographic properties and it is, therefore, concluded that a *trans* migration of an *N*-benzoyl group to the oxygen atom at C-3 is not a significant part of the rearrangement.

In the preceding paper[2] we have described the synthesis of the *N*-acylacylamino derivative, 2-(*N*-acetylbenzamido)-2-deoxy-D-glucose (II), through the hydrogenolysis of benzyl 2-(*N*-acetylbenzamido)-3,4,6-tri-*O*-benzyl-2-deoxy-β-D-glucopyranoside (I) (Scheme I). When heated in chloroform solution, this *N*-acylacylamide readily rearranges, the double peak at 1640 cm^{-1} characteristic of the *N*-acylacylamino derivative disappearing and bands, arising from ester carbonyl as well as NH groups, appearing. After acetylation of the rearranged material, thin layer chromatography showed the product to be a mixture; this mixture was separated by column chromatography into four components. The identification of these products will now be described.

The first substance isolated proved to be the previously described[2] 1,3,4,6-tetra-*O*-acetyl-2-(*N*-acetylbenzamido)-2-deoxy-β-D-glucopyranose. A minor component was then obtained in crystalline form [mp 178–179°, $[\alpha]^{20}D$ +111° ($CHCl_3$)] and found to have the composition and chromatographic properties of a 1,3,4,6-tetra-*O*-acetyl-2-benzamido-2-deoxy-D-glucopyranose. Its nuclear magnetic resonance spectrum included a doublet at τ 4.18 with a spacing of 3.5 cps, suggesting that the substance is an α anomer. Micheel, van de Kamp, and Petersen[3] acetylated 2-benzamido-2-deoxy-D-glucopyranose (VI) and found that the major product was 1,3,4,6-tetra-*O*-acetyl-2-benzamido-2-deoxy-α-D-glucopyranose (III) for which they gave mp 191° but no specific rotation. We have repeated their preparation and obtained a product of mp 185° and $[\alpha]^{20}D$ +107° ($CHCl_3$); while its nmr spectrum showed it to be contaminated with some of the β anomer VIII, the same spectrum clearly identified the minor product from the rearrangement as III.

A major component from the rearrangement proved to be a crystalline substance of mp 161–162° and $[\alpha]^{20}D$ +114° ($CHCl_3$); its nmr spectrum contained a doublet with a spacing of 3.5 cps centered at τ 3.55. Acetylation of the known 2-acetamido-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose[4] (VII) afforded 2-acetamido-3,4,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose (IV) which proved to be identical with the product from the rearrangement of II.

A second major product from the rearrangement was obtained as a syrup of $[\alpha]^{20}D$ +110° ($CHCl_3$); the general character of its nmr spectrum strongly suggested a furanose structure and a doublet at τ 3.32 with a spacing of 5 cps indicated that the substituents at C-1 and C-2 occupied a *cis* relationship.[5] The mass spectrum of the substance included a peak at *m/e* 306, corresponding to M − ($CH_2OAcCHOAc$–), which was not shown by IV. On the assumption that the substance was most probably 2-acetamido-3,5,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucofuranose (V) (Scheme II), the synthesis of authentic material of this structure was studied. The known ethyl 2-acetamido-3,5,6-tri-*O*-acetyl-2-deoxy-1-thio-α-D-glucofuranoside[6] (XI) was treated in acetonitrile solution with mercuric benzoate, the ethylthio group being replaced by a benzoyloxy group.[7] The crude syrupy product, $[\alpha]^{20}D$ +75° ($CHCl_3$), thus obtained gave a doublet of 5 cps centered at τ 3.32. However, a singlet at τ 3.6 was interpreted as probably arising from H_1 of the β anomer IX and, since all attempts to resolve the mixture by chromatography were unsuccessful, another approach to the synthesis of V was employed.

(1) Fellow in the Visiting Program of the National Institutes of Health, 1964–1965.

(2) T. D. Inch and H. G. Fletcher, Jr., *J. Org. Chem.*, **31**, 1815 (1966).

(3) F. Micheel, F.-P. van de Kamp, and H. Petersen, *Ber.*, **90**, 521 (1957).

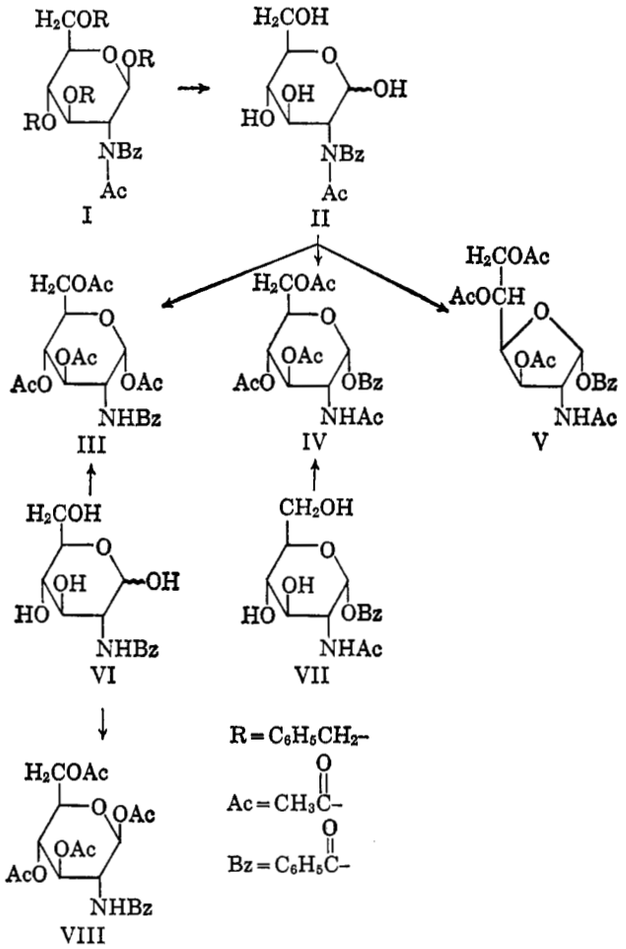
(4) R. Harrison and H. G. Fletcher, Jr., *J. Org. Chem.*, **31**, 2317 (1965).

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

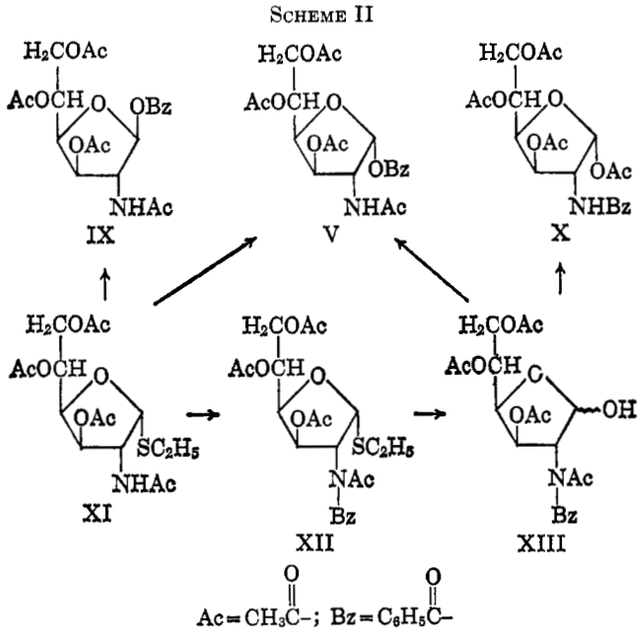
(6) M. L. Wolfrom, S. M. Olin, and W. J. Polglase, *ibid.*, **72**, 1724 (1950).

(7) In earlier studies of the behavior of 1-thioglycosides with mercuric and silver salts of carboxylic acids, acylated 1-thioglycosides (in contrast to the free 1-thioglycosides) were reported to be unreactive. *Cf.* H. B. Wood, Jr., B. Coxon, H. W. Diehl, and H. G. Fletcher, Jr., *J. Org. Chem.*, **29**, 461 (1964), footnote 7.

SCHEME I



SCHEME II



Benzoylation of XI with benzoyl chloride in pyridine afforded a syrup with the infrared absorption spectrum which would be expected for ethyl 3,5,6-tri-*O*-acetyl-2-(*N*-acetylbenzamido)-2-deoxy-1-thio-α-D-glucofuranoside (XII). Hydrolysis of this material in the presence of mercuric chloride and cadmium carbonate gave a mixture from which two substances, having the composition of a tetraacetylmonobenzoylhexosamine, were isolated through chromatography. One of these, obtained in 14% yield, showed $[\alpha]^{20}D$ +103° ($CHCl_3$) and gave an nmr spectrum identical with that of the second major product obtained from the rearrangement of II. The other product from XII was obtained in 34% yield and in crystalline form. It was dextrorotatory, $[\alpha]^{20}D$ +95° ($CHCl_3$), and gave an nmr spectrum with a doublet (τ 3.45, J = 5 cps) characteristic of an α derivative. This doublet being upfield of the corresponding signal from H_1 of V, indicates that the substance is the α 1-*O*-acetyl derivative X.

Whether derived from I, XII, or directly from XI, compound V was found in one respect to be unique among the substances tested. If, after thin layer chromatography on silica gel in any of a variety of solvents, the thin layer was exposed to iodine vapor and then sprayed with 10% sulfuric acid, a brilliant blue-green color developed. No explanation is offered for this interesting behavior.

In order to ascertain whether the rearrangement of II was solvent dependent, a sample was boiled in ethanol solution. After acetylation of the initial products, chromatography revealed the same spectrum of products found after the rearrangement of II in chloroform; in addition, a small amount of 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxy-D-glucopyranose was detected.[8]

Discussion

The structures of the substances isolated clearly show that *N*-acylacylaminohexose derivatives such as II and XIII readily undergo *N* → *O* acyl migration. Fodor and Ötvös[9] have shown that the *N*-acetyl group in methyl 2-acetamido-4,6-di-*O*-acetyl-2-deoxy-β-D-glucopyranoside easily migrates under the influence of acid to the oxygen atom at C-3. In the course of the present work, the triacetate of 2-acetamido-3-*O*-benzoyl-2-deoxy-D-glucose was synthesized for comparison purposes (see Experimental Section) but no component arising from the rearrangement of II (and subsequent acetylation) matched the chromatographic behavior of this substance. It may, therefore, be concluded that a *trans* migration of a benzoyl group from the nitrogen at C-2 to the oxygen at C-3 is not a significant part of the rearrangement of II. Furthermore, since III, IV, and V are all α anomers and since none of the corresponding β anomers were detected, it is apparent that *cis* migration to C-1 predominates.[10] Indeed, ring contraction to the furanose derivative V suggests that the juxtaposition of the groups at C-1 and C-2 in 2-(*N*-acetylbenzamido)-2-deoxy-α-D-glucofuranose is distinctly more favorable to N → O acyl migration than is the case with the α form of the pyranose structure II. This suggestion is confirmed by the fact that XIII rearranges at room temperature. The formation of 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxy-D-glucopyranose after the rearrangement of II in ethanol may be rationalized by either of two mechanisms; that the benzoyl group departed directly to the solvent in the absence of a catalyst is rendered unlikely by our previous work.[2] It is more likely that

(8) The anomeric forms of this substance are not distinguishable in the chromatographic system employed.

(9) G. Fodor and L. Ötvös, *Ber.*, **89**, 701 (1956).

(10) The evidence obtained renders migration of an acetyl from the nitrogen to C-3 highly unlikely since acetylation of 3-*O*-acetyl-2-benzamido-2-deoxy-D-glucopyranose would presumably give rise to a mixture of α and β anomers.

acyl migration initially gave 2-acetamido-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose (or -glucofuranose) and that solvolysis of the benzoyl group then took place prior to acetylation. In an earlier paper[11] we have shown that 2-acetamido-1-*O*-acyl-2-deoxyhexopyranoses are readily solvolyzed in the absence of a catalyst and one would expect the analogous furanose derivatives to cleave even more readily.

The behavior of an N-acyl group in an *N*-acylacylaminohexose resembles, in many respects, that of a normal *O*-acyl group rather than the behavior of the single acyl group in a mono-*N*-acylhexosamine. It undergoes solvolysis only in the presence of an acidic or basic catalyst but readily migrates intramolecularly, either at room temperature or under the influence of heat.

Where two dissimilar acyl groups are attached to the nitrogen (as in II and XIII) competition between them in migration is to be expected. It is interesting to note that with II benzoyl migration predominates over acetyl migration, IV and V being the major products isolated, while, on the other hand, XIII, presumably the immediate product from the hydrolysis of XII, largely undergoes acetyl migration to X. If acetyl migrates more readily than benzoyl in a furanose structure, one would expect X rather than V to have been formed from II. Such was not the case and it seems obvious that an understanding of the influences which govern the direction of these rearrangements must await further experimental study.

Experimental Section[12]

2-(*N*-Acetylbenzamido)-2-deoxy-D-glucopyranose (II).—This substance was prepared by the palladium-catalyzed reduction of benzyl 2-(*N*-acetylbenzamido)-3,4,6-tri-*O*-benzyl-2-deoxy-β-D-glucopyranoside (I) as described earlier.[2] The infrared absorption spectrum of the substance showed the two amide carbonyl bands, ν_{max}^{neat} (cm^{-1}) 1667 and 1616, characteristic of *N*-acylacylamino derivatives[2] and also a very small band at 1520 (NH), denoting partial acyl migration. When the hydrogenolysis was conducted rapidly on a small scale, rearrangement of the product was minimized.

Rearrangement of 2-(*N*-Acetylbenzamido)-2-deoxy-D-glucopyranose (II). A. In Chloroform.—Syrupy 2-(*N*-acetylbenzamido)-2-deoxy-D-glucopyranose (1.46 g) was dissolved in chloroform (U.S.P., 150 ml) and the solution boiled under reflux for 2 hr.[13] On removal of the chloroform, a solid residue was obtained. This was dissolved in pyridine, acetic anhydride (2 ml) was added, and the solution was stored at room temperature overnight. Toluene was then added and the solution was concentrated *in vacuo;* distillation of further quantities of toluene from the mixture was continued until the odor of pyridine could no longer be detected. The resulting crystalline solid (1.8 g) was examined by thin layer chromatography using ether, benzene–ether (1:1, v/v), and ether containing 2, 5, and 10% (v/v) of methanol, appropriate authentic substances being chromatographed simultaneously. The major component had a mobility indistinguishable from 2-acetamido-3,4,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose (IV). Minor components had the mobilities of 1,3,4,6-tetra-*O*-acetyl-2-benzamido-2-deoxy-α-D-glucopyranose (III) and 1,3,4,6-tetra-*O*-acetyl-2-(*N*-acetylbenzamido)-2-deoxy-β-D-glucopyranose; no 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxy-D-glucopyranose was detected.

The mixture was fractionated on a column (3 × 30 cm) of silica gel using ether as eluent and collecting 15-ml fractions. Fractions 10–26 contained **1,3,4,6-tetra-*O*-acetyl-2-(*N*-acetylbenzamido)-2-deoxy-β-D-glucopyranose** (0.26 g), mp 115–116° (from heptane–ether), mixture melting point with 1,3,4,6-tetra-*O*-acetyl-2-(*N*-acetylbenzamido)-2-deoxy-β-D-glucopyranose prepared from II as described earlier[2] 116–117°.

Fractions 27–41 contained **1,3,4,6-tetra-*O*-acetyl-2-benzamido-2-deoxy-α-D-glucopyranose** (III, 0.08 g), mp 178–179° (from methanol–ether), $[\alpha]^{20}D$ +111° (*c* 0.5, $CHCl_3$), mixture melting point with authentic material (mp 185–186°), prepared as described later in this paper by direct acetylation of 2-benzamido-2-deoxy-D-glucopyranose, 179–181°. The nmr spectrum of the substance gave a doublet at τ 3.67 ($J_{1,2}$ = 3.5 cps); this, together with the absence of a doublet at τ 4.18 suggests that the compound is probably the pure α anomer.

The eluent was changed to ether–methanol (1:1, v/v). Fractions 55–62 contained 0.95 g of material which solidified on trituration with cyclohexane. Crystallized from carbon tetrachloride–cyclohexane, the product had mp 78–82° and $[\alpha]^{20}D$ +102° (*c* 1.36, $CHCl_3$) and contained *ca.* 5% of carbon tetrachloride. After drying at 60° *in vacuo*, it was melted and dried further at 85° *in vacuo* and then it gave satisfactory elementary analyses for a tetraacetylmonobenzoylhexosamine. The nmr spectrum of the material suggested that it consisted of 2-acetamido-3,4,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose (IV) together with at least one other *O*-benzoyl derivative and thin layer chromatography in carbon tetrachloride–ethanol (13:1, v/v) revealed two components with nearly equal mobilities. The slower component was readily distinguished from the faster one by the intense blue-green color which it gave when exposed to iodine vapor and then sprayed immediately with 10% sulfuric acid.

The mixture (0.36 g) was dissolved in chloroform (1 ml) and the solution was poured on a column (3.3 × 27 cm) of silica gel; carbon tetrachloride–ethanol (13:1, v/v) was used as eluent and 15-ml fractions were collected. Fractions 18–26 contained a single component (158 mg) which was crystallized from carbon tetrachloride–cyclohexane: mp 161–162°, $[\alpha]^{20}D$ +114° (*c* 1.2, $CHCl_3$). The chromatographic behavior of the substance as well as its nmr spectrum was identical with that of **2-acetamido-3,4,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose** (IV); the preparation of an authentic specimen is described later in this paper. A mixture melting point was undepressed.

Fractions 27–29 contained a mixture of the two substances.

Fractions 30–46 contained the second component (180 mg) which was obtained as a syrup: $[\alpha]^{20}D$ +110° (*c* 0.3, $CHCl_3$); nmr data, τ 3.32 (H_1, $J_{1,2}$ = 5 cps), 7.88, 7.97, 8.00, and 8.03 (Ac). Its chromatographic behavior and nmr spectrum were indistinguishable from those of **2-acetamido-3,5,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucofuranose** (V) whose preparation is described later in this paper.

B. In Ethanol.—A solution of 2-(*N*-acetylbenzamido)-2-deoxy-D-glucopyranose (II, 0.91 g) in absolute ethanol (20 ml) was boiled under reflux for 1 hr and then concentrated *in vacuo*. The infrared spectrum of the residual syrup showed the rearrangement to be essentially complete. A solution of this syrup in a mixture of pyridine (10 ml) and acetic anhydride (4 ml) was stored at room temperature overnight and then was concentrated *in vacuo*, the last traces of acetic anhydride, acetic acid, and pyridine being removed by codistillation with toluene. The residue was examined by thin layer chromatography beside appropriate authentic samples. Using ether for chromatography, a trace of 1,3,4,6-tetra-*O*-acetyl-2-(*N*-acetylbenzamido)-2-deoxy-β-D-glucopyranose, together with a larger quantity of 1,3,4,6-tetra-*O*-acetyl-2-benzamido-2-deoxy-β-D-glucopyranose, were detected. Chromatography with benzene–ether–methanol (14:14:1, v/v) as solvent revealed the latter compound as well as some **2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxy-D-glucopyranose.** This appeared to be present in only a small amount; its presence had not been detected in A above. The major products of the rearrangement appeared to be 2-acetamido-3,4,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose (IV) and 2-acetamido-3,5,6-tri-*O*-acetyl 1-*O*-benzoyl-2-deoxy-α-D-glucofuranose (V).

(11) T. D. Inch and H. G. Fletcher, Jr., *J. Org. Chem.*, **31**, 1810 (1966).

(12) Melting points are corrected. Thin layer chromatography was conducted on silica gel G (E. Merck A.-G., Darmstadt) using the solvent systems specified; development was made with iodine vapor, followed in some cases with a spray of 10% (v/v) sulfuric acid. Column chromatography was carried out using silica gel (0.05–0.20 mm) of E. Merck A.-G. The nmr spectra were measured in $CDCl_3$ solution using a Varian A-60 spectrometer. Infrared spectra were obtained with a Perkin-Elmer Spectrocord spectrometer. Mass spectra were measured on an Associated Electrical Industry (UK) MS-9 double-focusing spectrometer at 70 ev. In all cases the sample was introduced directly into the electron beam.

(13) In a subsequent experiment, the progress of the reaction was monitored through the infrared absorption spectrum of the chloroform solution; the appearance of absorption bands of *ca.* equal intensity at 1700 (ester carbonyl), 1640 (amide carbonyl), and 1520 (NH) cm^{-1} indicating completion of the rearrangement.

Acetylation of 2-Benzamido-2-deoxy-D-glucopyranose (VI).—A solution of 2-benzamido-2-deoxy-D-glucopyranose[14] (VI, 2.2 g) in a mixture of acetic anhydride (9 ml) and pyridine (20 ml) was stored overnight at room temperature, poured into ice water, and worked up in conventional fashion, 2.1 g. Recrystallization from methanol gave a product of mp 185° (1 g, 29%), $[\alpha]^{20}D$ +107° (*c* 0.5, $CHCl_3$). Micheel, van de Kamp, and Petersen[3] reported mp 191° but no specific rotation for a substance to which they assigned structure VIII.

Anal. Calcd for $C_{21}H_{25}NO_{10}$ (451.44): C, 55.87; H, 5.58; N, 3.10. Found: C, 56.07; H, 5.62; N, 3.11.

A solution of the compound in $CDCl_3$ was shaken with D_2O for 24 hr; its nmr spectrum showed a strong doublet at τ 3.67 (J = 3.5 cps) and a very weak signal at τ 4.18 (J = 8 cps), corresponding to H_1 for the α and β anomers, respectively. Efforts to separate the two anomers by thin layer chromatography were unsuccessful.

2-Acetamido-3,4,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose (IV).—2-Acetamido-3,4,6-tri-*O*-benzyl-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose[4] (0.2 g) in ethanolic solution was hydrogenated using palladium as a catalyst. The solution was filtered and concentrated and, without isolation, the 2-acetamido-1-*O*-benzoyl-2-deoxy-α-D-glucopyranose[4] (VII) was acetylated with acetic anhydride (1 ml) and pyridine (5 ml). After storage overnight at room temperature, the solution was concentrated *in vacuo*, traces of pyridine, etc., being removed by codistillation with toluene. The product was crystallized from carbon tetrachloride–cyclohexane: 0.1 g (66%), mp 163–164°, $[\alpha]^{20}D$ +114° (*c* 1.2, $CHCl_3$). Owing to the presence of a trace of carbon tetrachloride, the product failed to give a fully satisfactory analysis. The material was, therefore, chromatographed on silica gel and the resulting pure syrup allowed to crystallize: melting point, either alone or in admixture with material crystallized from carbon tetrachloride–cyclohexane, 161°; nmr data, τ 3.55 ($J_{1,2}$ = 3.5 cps), 7.93 (OAc), 8.14 (NAc).

Anal. Calcd for $C_{21}H_{25}NO_{10}$ (451.44): C, 55.87; H, 5.58; N, 3.10. Found: C, 55.92; H, 5.62; N, 3.17.

Ethyl 3,5,6-Tri-*O*-acetyl-2-(*N*-acetylbenzamido)-2-deoxy-1-thio-α-D-glucofuranoside (XII).—A solution of ethyl 2-acetamido-3,5,6-tri-*O*-acetyl-2-deoxy-1-thio-α-D-glucofuranoside[15] (XI, 1.0 g) in a mixture of benzoyl chloride (0.34 ml) and pyridine (5 ml) was stored at room temperature overnight and then poured into ice-water. The product was extracted with dichloromethane and the combined extracts were washed successively with dilute hydrochloric acid, aqueous sodium bicarbonate, and water. Moisture was removed with magnesium sulfate and the solution was concentrated *in vacuo* to a syrup which was chromatographed on a column of silica gel using benzene–ether (5:1, v/v). A fraction was obtained which showed no absorption at 1727 cm^{-1} but gave bands at 1600 (aromatic), 1670, 1700 and 1750 cm^{-1} (CO). The product was subjected to a short-path distillation at 0.003 mm and 150° (bath temperature): 0.64 g (51%), $[\alpha]^{20}D$ +67° (*c* 1.6, $CHCl_3$). Repeated attempts to obtain satisfactory values for the elementary composition of the material were unsuccessful.

Hydrolysis of Ethyl 3,5,6-Tri-*O*-acetyl-2-(*N*-acetylbenzamido)-2-deoxy-1-thio-α-D-glucofuranoside (XII) and Spontaneous Rearrangement of Product XIII.—A solution of XII (0.8 g) in 50% (v/v) aqueous methanol (200 ml) containing mercuric chloride (10 g) and cadmium carbonate (10 g) was stirred at room temperature for 18 hr. It was then filtered through a layer of Celite and the solid residue was washed thoroughly with chloroform. The chloroform layer was separated from the filtrate and the aqueous layer was extracted with more chloroform. The original chloroform layer was combined with the extracts, washed with water, dried with magnesium sulfate, and concentrated *in vacuo*. Chromatographed on a column of silica gel using benzene–ether–methanol (14:14:1, v/v), the residue gave three fractions. The first fraction (50 mg) consisted of unchanged XII. The second fraction (250 mg, 34%) was twice crystallized from ether–ethanol: mp 128–129°, $[\alpha]^{20}D$ +95° (*c* 0.43, $CHCl_3$).

Anal. Calcd for $C_{21}H_{25}NO_{10}$ (451.44): C, 55.87; H, 5.58; N, 3.10. Found: C, 55.57; H, 5.58; N, 2.93.

The nmr spectrum showed τ 3.45 (H_1, $J_{1,2}$ = 5 cps), 7.87, 7.92, (Ac). The $H_{1,2}$ coupling constant together with the higher field position of the H_1 signal, compared with 2-acetamido-3,5,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucofuranose (V, see below) identified the substance as 1,3,5,6-tri-*O*-acetyl-2-benzamido-2-deoxy-α-D-glucofuranose (X).

The third fraction (100 mg, 14%) was obtained as a syrup, $[\alpha]^{20}D$ +103° (*c* 0.59, $CHCl_3$); its behavior on thin layer chromatography was indistinguishable from that of the product (V) of $[\alpha]^{20}D$ +110° obtained through the rearrangement of II. Like that product, it was unique among all those investigated in giving a blue-green color when chromatographed on thin layer and sprayed with 10% sulfuric acid immediately after exposure to iodine vapor. For analysis, a sample was subjected to a short-path distillation at 0.005 mm and 160° (bath temperature), whereupon it crystallized: mp 110–112°.

Anal. Calcd for $C_{21}H_{25}NO_{10}$ (451.44): C, 55.87; H, 5.58; N, 3.10. Found: C, 55.87; H, 5.73; N, 2.95.

The nmr data showed a doublet centered at τ 3.32 with a spacing of 5 cps (H_1), 7.88, 7.97, 8.00, 8.03 (Ac). The lower-field signal for H_1 (compared with that of its isomer X) distinguishes the two isomers.

Reaction of Ethyl 2-Acetamido-3,5,6-tri-*O*-acetyl-2-deoxy-1-thio-α-D-glucofuranoside (XI) with Mercuric Benzoate.—A mixture of ethyl 2-acetamido-3,5,6-tri-*O*-acetyl-2-deoxy-1-thio-α-D-glucofuranoside[6] (XI, 0.50 g) and mercuric benzoate (2.5 g) in acetonitrile (50 ml freshly distilled from P_2O_5) was stirred vigorously at room temperature for 20 hr. The suspension was filtered through a layer of Celite and concentrated. Chromatography on silica gel, using carbon tetrachloride–ethanol (13:1, v/v), gave a product contaminated with inorganic material. A second chromatography on silica gel, using benzene–ether–ethanol (14:14:1, v/v), gave a syrupy product free of inorganic material: 150 mg (26%), $[\alpha]^{20}D$ +75° (*c* 0.91, $CHCl_3$). The nmr spectrum of the product included acetyl proton signals at τ 7.87, 7.97, and 8.01. A doublet with a spacing of J = 5 cps, centered at τ 3.32, was interpreted as arising from H_1 of 2-acetamido-3,5,6-tri-*O*-acetyl-1-*O*-benzoyl-2-deoxy-α-D-glucofuranose (V) while a singlet of *ca.* equal area at τ 3.6 was viewed as arising from H_1 of the corresponding anomer IX. No chromatographic system capable of separating these anomers was found.

Benzyl 2-Acetamido-3-*O*-benzoyl-4,6-*O*-benzylidene-2-deoxy-D-glucopyranoside.—Crude benzyl 2-acetamido-2-deoxy-D-glucopyranoside was made by an adaptation of the procedure of Yoshimura, *et al.*[16] A solution of 2-acetamido-2-deoxy-D-glucose (5.0 g) in benzyl alcohol (35 ml) containing a solution of boron trifluoride in ether (0.7 ml, Eastman No. 4247) was heated at 95° for 2 hr, cooled, and poured into ether. Filtered off and washed with ether, the crude crystalline glycoside (7.0 g) was stirred at room temperature with a mixture of benzaldehyde (200 ml) and zinc chloride (30 g) for 5 hr. The solution was then poured into a vigorously stirred mixture of water and ligroin and the product was removed by filtration. Recrystallized from aqueous pyridine, the product melted at 245–250°. Kuhn, Baer, and Seeliger[17] reported mp 262° for benzyl 2-acetamido-4,6-*O*-benzylidene-2-deoxy-α-D-glucopyranoside.

A portion (1.0 g) of the benzylidene derivative was stored overnight at room temperature in a mixture of benzoyl chloride (1 molar equiv) and pyridine (10 ml). The solution was poured into ice–water, and the product was filtered off and crystallized from ethanol (0.8 g). Thin layer chromatography (benzene–methanol, 8:1, v/v) showed the product to be impure. It was chromatographed on a silica gel column using benzene–ether–ethanol (14:14:1); recrystallization from ethanol then gave a product (0.5 g, 40%) of mp 232°, $[\alpha]^{20}D$ +48.7° (*c* 1.1, $CHCl_3$), $[\alpha]^{20}D$ +31° (*c* 0.98, pyridine).

Anal. Calcd for $C_{29}H_{29}NO_7$ (503.56): C, 69.17; H, 5.80; N, 2.78. Found: C, 69.25; H, 5.77; N, 2.71.

Kuhn, Baer, and Seeliger[17] reported mp 218–220° and $[\alpha]D$ +44° (pyridine) for benzyl 2-acetamido-3-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranoside. It appears probable that the preparation described here is contaminated with the β anomer.

2-Acetamido-3-*O*-benzoyl-2-deoxy-D-glucose.—A solution of benzyl 2-acetamido-3-*O*-benzoyl-4,6-*O*-benzylidene-2-deoxy-D-glucopyranoside (0.34 g) in ethanol (50 ml) was shaken with hydrogen, using palladium black as catalyst until absorption of the

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gas had ceased. After filtration, the solution was concentrated *in vacuo* and the residue was crystallized from methanol: 0.2 g (91%), mp 198° dec, $[\alpha]^{20}D$ +35° (*c* 0.85 in 50% v/v aqueous dioxan, no mutarotation on standing overnight).

Anal. Calcd for $C_{25}H_{19}NO_7$ (325.33): C, 55.38; H, 5.87; N, 4.31. Found: C, 55.39; H, 5.90; N, 4.55.

The 3-*O*-benzoyl derivative was stored overnight with acetic anhydride in pyridine. Pyridine, etc., was removed by codistillation with toluene and the residue was shown to be essentially homogeneous by thin layer chromatography. The nmr spectrum of the product indicated that it was predominantly the α anomer: τ 3.7 (H_1, $J_{1,2}$ = 3.5 cps), 7.77, 7.88, 8.05, 8.15 (Ac).

Mass Spectra.—In an initial attempt to distinguish further between IV and V and to prove that V was a furanose[18] derivative, their mass spectra were obtained.

As expected, V showed a peak (although weak) at *m/e* 306 corresponding to M − ($CH_2OAcCHOAc$–). Such a peak was completely absent from IV and thus provided corroboration that V is indeed in the furanose form. Also, the mass spectra of V and of the anomeric mixture obtained by treatment of XI with mercuric benzoate were closely similar, thus substantiating the conclusion that this reaction had given an anomeric mixture of 1-*O*-benzoates and confirming that V was a 1-*O*-benzoate.

It has been previously observed[19] that the fragmentation pattern of acetylated aminosugars differs markedly from the fragmentation of acetylated sugars. In the compounds we have examined the presence of a benzoyl group further complicated the situation. With both IV and V, a peak at *m/e* 346 was obtained corresponding to M − C_6H_5CO. However with V a strong peak at *m/e* 304 was obtained corresponding to loss of C_6H_5CO (105) and CH_2=C=O (42) from the parent molecule. The corresponding peak in IV was negligibly small. Until more examples have been studied the reason for this difference must remain a matter of conjecture.

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N-Acyl Derivatives of 2-Acylamino-2-deoxyhexoses. Nuclear Magnetic Resonance Spectra and Conformations

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The nmr spectra of some 1,3,4,6-tetra-*O*-acetyl-2-(*N*-acylacylamino)-2-deoxyhexoses and of some 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxyhexoses of the D-glucose, D-galactose, and D-mannose series have been studied. In contrast to the spectra of the second class of compounds, those of the first class are readily analyzed. The coupling constants for representatives of the D-glucose and D-galactose series clearly show these compounds to exist in the normal chair conformation. In *N*-acylacylamino derivatives of the D-glucose series, the signal from the axial proton at C_1 occurs at lower field than the signal for the equatorial C_1 proton, a reversal of the normal situation which has not hitherto been observed at C_1. The conformational implications of the nmr spectra are discussed.

Since the pioneering experiments of Lemieux, *et al.*,[3] in 1958, nuclear magnetic resonance has become a widely used and nearly essential tool for the elucidation of structural and configurational problems in the carbohydrate field.[4] In only a few cases,[5,6] however, has it been possible to obtain the parameters thought necessary to describe unequivocally the precise conformation of a pyranose ring. Although nmr spectroscopy has provided an elegant means for the determination of the configuration of some of the more recently described aminosugars,[7] complete first-order analyses of the nmr spectra of the 2-amino-2-deoxyhexopyranoses (or, more usually, of 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxyhexopyranoses) have not been reported. It is immediately apparent from the spectrum of 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxy-β-D-glucopyranose (**5**, Figure 1) that the region of the spectrum from 4.95 to 5.44 ppm (H_3 and H_4) and the region from 3.7 to 4.5 ppm (H_2, H_5, H_6, and $H_{6'}$) cannot readily be analyzed although, of course, the coupling constant $J_{1,2}$ provides an indication of anomeric configuration.[8] Inch and Fletcher[8,9] have recently described 2-(*N*-acylacylamino)-2-deoxyhexopyranose derivatives which may readily be prepared through the *N*-acylation of substances such as **5** or its α anomer **6**. The nmr spectra of these derivatives proved to be much more informative. The present paper describes an analysis of the spectra of 2-(*N*-acylacylamino)-2-deoxyhexose derivatives of the D-glucose, D-galactose, and D-mannose series.

Experimental Section

Nmr Spectra.—Spectra were measured in deuteriochloroform using a Varian A-60 spectrometer at 60 Mc/sec, and the results quoted refer to these spectra unless otherwise stated. Chemical shifts are reported in parts per million from the internal tetramethylsilane standard. No true coupling constants have been calculated and the results, given in cycles per second (*e.g.*, $J_{1,2}$ = 4 cps), refer to measured line spacings. Spin–spin decoupling experiments[10] were performed using a Varian HA-100 internal proton stabilized spectrometer at 100 Mc/sec.

In order to identify (and eliminate) the NH signal from the 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxyhexopyranoses, the deuteriochloroform solutions of these substances were overlayered with D_2O and left at room temperature (with occasional shaking)

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