958. *The Preparation and Properties of Acetochloroglucosamine and its Use in the Synthesis of 2-Acetamido-2-deoxy-β-D-glucosides* (N-*Acetyl-β-D-glucosaminides*).

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A new preparation of acetochloroglucosamine is described. This compound has been used in the synthesis of alkyl and aryl 2-acetamido-2-deoxy-β-D-glucosides. In wet solvents it is transformed into 1 : 3 : 4 : 6-tetra-*O*-acetyl-2-amino-2-deoxy-α-D-glucose hydrochloride, properties and reactions of which are described.

Methods were required for the synthesis of alkyl and aryl 2-acetamido-2-deoxy-β-D-glucosides (*N*-acetyl-β-D-glucosaminides) for use as substrates in a study of the enzyme "*N*-acetyl-β-glucosaminidase."[1] Such alkyl derivatives have been prepared by the condensation[2] of alcohols with 2-acetamido-2-deoxy-D-glucose, and the phenyl compound by condensation of phenol with 2-amino-2-deoxyglucose (glucosamine) penta-acetate.[3] Both these reactions gave mixtures of α- and β-glycosides from which the β-isomers were separated by column chromatography[2] or fractional crystallisation.[4]

Moggridge and Neuberger[5] claimed to have prepared acetobromoglucosamine, but

[1] Borooah, Leaback, and Walker, *Biochem. J.*, 1957, **65**, 15P.
[2] Zilliken, Rose, Braun, and Gyorgy, *Arch. Biochem. Biophys.*, 1951, **54**, 392.
[3] Helferich and Iloff, *Z. physiol Chem.*, 1933, **221**, 252.
[4] Roseman and Dorfman, *J. Biol. Chem.*, 1951, **191**, 607.
[5] Moggridge and Neuberger, *J.*, 1938, 745.

were unable to use their product in the synthesis of 2-acetamido-2-deoxy-β-D-glucosides. Kuhn and Kirschenlohr,[6] however, described the preparation, in good yield, of such alkyl glycosides by condensation of alcohols with acetobromoglucosamine which was said to have been prepared by Moggridge and Neuberger's method. In our hands, Kuhn and Kirschenlohr's method was successful provided that acetobromoglucosamine was used as an uncrystallised syrup. Attempts to crystallise acetobromoglucosamine from the syrup invariably resulted in the isolation of 1 : 3 : 4 : 6-tetra-*O*-acetyl-2-amino-2-deoxy-α-D-glucose hydrobromide, thus confirming the work of Micheel, Van de Kamp, and Wulff.[7] Unsuccessful attempts were made to employ acetobromoglucosamine in the synthesis of aryl 2-acetamido-2-deoxy-β-D-glucosides.[8] Other reports of difficulty in the use of acetobromoglucosamine have appeared.[7,9,10]

Baker *et al.*[9] described the preparation of crystalline acetochloroglucosamine (2-acetamido-3 : 4 : 6-tri-*O*-acetyl-1-chloro-2-deoxy-α-D-glucose) (II), which was employed successfully in the synthesis of an *N*-glycosylpurine and of 2-acetamido-2-deoxy-α-D-glucose 1-phosphate.[10] The present paper describes further work on the preparation and properties of acetochloroglucosamine and its use in the synthesis of alkyl and aryl 2-acetamido-2-deoxy-β-D-glucosides.[11]

Much of the present work was carried out with acetochloroglucosamine prepared according to the method of Baker *et al.*, by the action of ether, acetic anhydride, and hydrogen chloride on the β-penta-acetate (I). Difficulties were experienced with this preparation: frequently the penta-acetate did not dissolve in the reagent or was precipitated therefrom. This was even more marked in the preparation of acetochloroglucosamine from the α-penta-acetate (VII), and has been attributed to the basicity of the *N*-acetyl group.[12] The difficulties have been overcome by using dry hydrogen chloride and acetic anhydride as the reagent: reaction is faster with the β- than with the α-penta-acetate.

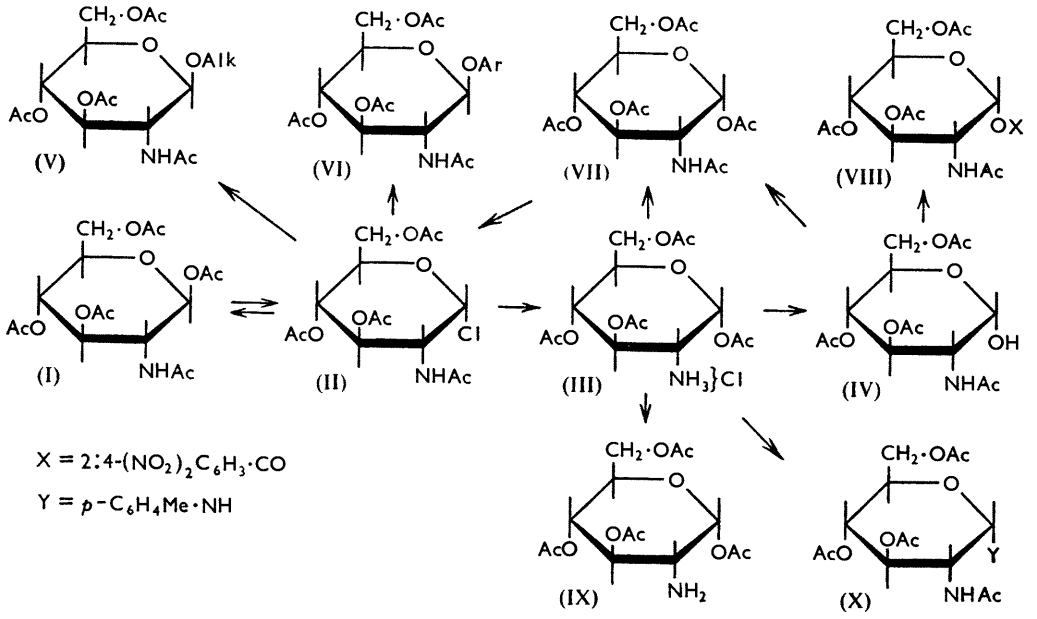
Acetochloroglucosamine and methyl or ethyl alcohol, with silver carbonate as the condensing agent, gave good yields of methyl and ethyl 2-acetamido-3 : 4 : 6-tri-*O*-acetyl-2-deoxy-β-D-glucoside (V). Removal of the *O*-acetyl groups from the ethyl compound with ammoniacal methanol brought about some anomerisation.[13] While the present work was in progress, Morel[14] reported briefly that the alkyl β-D-glycosides can be prepared from acetochloroglucosamine with mercuric cyanide as the condensing agent.

Aryl tetra-acetyl-2-amino-2-deoxy-β-D-glucosides (VI) were prepared by condensing acetochloroglucosamine with the appropriate phenol under conditions similar to those used by Glaser and Wulwek;[15] no single method was found convenient for removal of *O*-acetyl groups from all the tetra-acetates prepared (Table 2). Glaser and Wulwek reported that *o*-nitrophenyl tetra-*O*-acetyl-β-D-glucoside gave an anomalous positive optical rotation; the 2-amino-compound is similar in this respect. In conditions known to give aryl α-glycosides with acetobromo-sugars,[16] acetochloroglucosamine gave phenyl tetra-acetyl-2-amino-2-deoxy-β-D-glucoside.

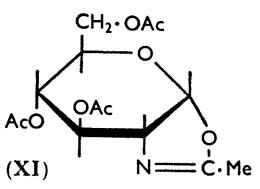
Acetochloroglucosamine (II) is stable in dry solvents but in the presence of water is converted into 1 : 3 : 4 : 6-tetra-*O*-acetyl-2-amino-2-deoxy-α-D-glucose hydrochloride (III). Inouye, Kitaoka, and Ochiai[17] independently came to a similar conclusion. The infrared spectra of compounds (II) and (III) agree with the proposed structures (Table 1); further, compound (III) was converted into the free base (IX) which had constants similar to those

[6] Kuhn and Kirschenlohr, *Chem. Ber.*, 1953, **86**, 1331.
[7] Micheel, Van de Kamp, and Wulff, *Chem. Ber.*, 1955, **88**, 2011.
[8] Leaback and Walker, unpublished results.
[9] Baker, Joseph, Schaub, and Williams, *J. Org. Chem.*, 1954, **19**, 1786.
[10] Leloir and Cardini, *Biochim. Biophys. Acta*, 1956, **20**, 33.
[11] Cf. Leaback and Walker, *Chem. and Ind.*, 1956, 1017.
[12] Baker, Joseph, and Schaub, *J. Amer. Chem. Soc.*, 1955, **77**, 5905.
[13] Cf. Hough and Taha, *J.*, 1956, 2042.
[14] Morel, *Experentia*, 1956, **12**, 419.
[15] Glaser and Wulwek, *Biochem. Z.*, 1924, **145**, 514.
[16] Helferich and Jung, *Annalen*, 1954, **589**, 77.
[17] Inouye, Kitaoka, and Ochiai, *Bull. Agric. Chem. Soc. Japan*, 1956, **20**, 157.

reported by Micheel *et al.*[7] for this compound. The transformation (II) ⟶ (III) is particularly rapid in nitromethane and acetonitrile, and is accelerated by free acid. A similar reaction probably occurred in our unsuccessful attempts to crystallise acetobromoglucosamine.



Compound (III) had physical properties similar to those of the corresponding hydrobromide described by Micheel *et al.*,[7] and to those of the β-anomer;[18] however, it differs from the latter in that, in aqueous solution, its optical rotation falls slowly. The process is faster in presence of pyridine or sodium acetate and is similar to that described by Micheel *et al.*[7] for the corresponding hydrobromide. Micheel *et al.* ascribed this fall in rotation to the conversion of 1 : 3 : 5 : 6-tetra-*O*-acetyl-2-amino-2-deoxy-α-D-glucose hydrobromide into the oxazoline (XI). White[19] started with what was probably 1 : 3 : 4 : 6-tetra-*O*-acetyl-2-amino-2-deoxy-α-D-glucose hydrobromide (then thought to be acetobromoglucosamine) and claimed to have prepared this oxazoline (XI). By White's method, Micheel *et al.*[7] converted the 1 : 3 : 4 : 6-tetra-acetate hydrobromide into what was said to be the oxazoline (XI); under anhydrous conditions the product was then converted into 2-amino-2-deoxy-α-D-glucose penta-acetate and the 1-(3 : 5-dinitrobenzoate) 3 : 4 : 6 : *N*-tetra-acetate (VIII).



Starting from the hydrochloride (III) and using conditions similar to White's, we have isolated a product with constants similar to those reported for the oxazoline; however, we consider this product to be the tetra-acetate (IV) since (i) it has an infrared band at 3300 cm.$^{-1}$ corresponding to the groups –NH– and/or –OH (see Table 1), (ii) it was converted under anhydrous conditions into compounds (VII) and (VIII), and (iii) its optical rotation (in water) fell (mutarotation?) to a value identical with the equilibrium value obtained when the salt (III) was treated at room temperature with a solution of sodium acetate.

[18] Bergman and Zervas, *Ber.*, 1931, **64**, 978.
[19] White, *J.*, 1940, 428.

TABLE 1. *Infrared spectra* (*max. in cm.*$^{-1}$).

Compound	–OH and –NH– Region	$NH_3^+ X^-$	*O*-Acyl	Amide I & II	2*a* Band
(II)	3250	None	1735	1640, 1540	755
(III)	None	3000—2500	1750	None	None
Hydrobromide analogous to (III)	None	3000—2500	1750	None	None
(IV)	3300	None	1735	1650, 1540	None

TABLE 2. *Removal of* O-*acetyl groups from aryl* 2-*acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-2-*deoxy*-β-D-*glucosides*.

Aryl	Products [a] Yield (%)	M. p. (decomp.)	$[\alpha]_D$ [e]	Formula	Found (%) C	H	N	Required (%) C	H	N
Ph	71 [b]	249°	− 8·4	$C_{14}H_{19}O_6N$	56·3	6·6	4·9	56·6	6·4	4·7
p-C_6H_4Ac	55 [c]	224—225	− 5·5	$C_{16}H_{21}O_7N$	58·8	6·2	4·5	59·6	6·2	4·4
o-$NO_2{\cdot}C_6H_4$	69 [d]	192—194	−33·1	$C_{14}H_{18}O_8N_2$	49·2	5·3	8·2	49·1	5·3	8·2
p-$NO_2{\cdot}C_6H_4$	83 [d]	204	−18·6	$C_{14}H_{18}O_8N_2$	48·8	5·4	8·2	49·1	5·3	8·2
1-$C_{10}H_7$	92 [d]	244—246	−64·5	$C_{18}H_{21}O_6N$	60·9	5·8	4·0	62·2	6·0	4·0

[a] Recryst. from H_2O. [b] Prep. by $Ba(OMe)_2$–MeOH (Isbell, *Bur. Stand. J. Res.*, 1930, **5**, 1185). [c] As (*b*) but in 1 : 1 MeOH–$CHCl_3$. [d] Prep. by Westphal and Schmidt's method (*Annalen*, 1952, **575**, 84). [e] In H_2O (0·2—0·5% solution) at *ca.* 20°.

Bertho and Koziollek [20] prepared 2-acetamido-2 : 3 : 4 : 6-tetra-*O*-acetyl-1 : 2-dideoxy-1-*p*-toluidino-D-glucopyranose by treating *p*-toluidine with "acetobromoglucosamine" prepared by Moggridge and Neuberger's method.[5] We treated the salt (III) with *p*-toluidine under Bertho and Koziollek's conditions [20] and isolated the same compound (X). On removal of *O*-acetyl groups, the product had constants similar to those described for 2-acetamido-1 : 2-dideoxy-1-*p*-toluidino-D-glucopyranose and behaved chromatographically as did this compound prepared by the condensation of 2-acetamido-2-deoxy-D-glucose and *p*-toluidine.[20] The nature of the reaction between the salt (III) and *p*-toluidine is under investigation but it is thought that the amide (IV) may be involved.

Table 1 summarises the infrared data presented in this paper.

EXPERIMENTAL

Paper chromatography was carried out on Whatman No. 1 filter paper by the descending method with (*a*) butan-1-ol–ethanol–water (4 : 1 : 5 v/v) or (*b*) butan-1-ol–pyridine–water (6 : 4 : 3 v/v), and the separated substances were detected by Rydon and Smith's method.[21]

All the compounds except acetochloroglucosamine were dried overnight at 100° (over P_2O_5) before analysis. Infrared spectra were determined on potassium chloride pellets. Solutions were evaporated under reduced pressure. The light petroleum had b. p. 80—100°.

Acetochloroglucosamine (II).—(i) Dry crystalline 2-acetamido-tetra-*O*-acetyl-2-deoxy-β-D-glucose [22] was shaken with acetic anhydride (30 ml.) saturated at 0° with dry hydrogen chloride. After dissolution was complete the mixture was set aside for 16 hr. at room temperature. It was added to chloroform (125 ml.), cooled, and extracted twice with cold saturated sodium hydrogen carbonate solution, and twice with cold water. The chloroform layer was dried ($MgSO_4$) and evaporated to a syrup which crystallised. The residue was triturated with dry ether and the solid filtered off, to give acetochloroglucosamine, m. p. 126—127° (6·5 g., 69%).

The product recrystallised when dissolved in warm ethyl acetate, treated with light petroleum to turbidity, and kept cold overnight. It then decomposed at 133—134° and had $[\alpha]_D^{18}$ +118° (*c* 1 in $CHCl_3$) (Found: C, 46·1; H, 5·3; N, 3·7; Cl, 9·7. Calc. for $C_{14}H_{20}O_8NCl$: C, 46·0; H, 5·5; N, 3·8: Cl, 9·7%).

(ii) The crystalline α-penta-acetyl compound [prepared by acetylation of the salt (III)] (5 g.) was kept in acetic anhydride (15 ml.) saturated at 0° with dry hydrogen chloride for 16 hr. at room temperature. The solution was cooled to 0°, re-saturated with dry hydrogen chloride,

[20] Bertho and Koziollek, *Chem. Ber.*, 1954, **87**, 934.
[21] Rydon and Smith, *Nature*, 1952, **169**, 922.
[22] Levene, "Hexosamines and Mucoproteins," Longmans, Green & Co., London, 1925.

set aside for 3 days at room temperature, and worked up as above, to give 70% of acetochloroglucosamine identical with that obtained as in (i). Procedure (ii) is necessary for the preparation of the compound (II) from the mixed penta-acetates.[23]

Acetochloroglucosamine is soluble in chloroform, acetone, acetic acid, or nitromethane but insoluble in ether or cold water. The infrared spectrum (Table 1) includes bands at 3250 (banded absorption of secondary amides in the solid state), 1640 (carbonyl absorption of mono-substituted amides), 1540 (CO·NHR), and 755 cm.$^{-1}$ (sharp) (C–Hal stretching).

2-*Acetamido*-1 : 3 : 4 : 6-*tetra*-O-*acetyl*-2-*deoxy*-β-D-*glucopyranose* (I).—Acetochloroglucosamine (0·5 g.) was shaken with dry acetic acid (10 ml.) for 16 hr. in the absence of light with silver carbonate (0·5 g.). The silver salts were filtered off and the filtrate was evaporated under reduced pressure. Recrystallisation of the residue from ethanol gave the penta-acetyl derivative (0·31 g., 58%), m. p. 187°, $[\alpha]_D^{20}$ +1·0° (*c* 1 in $CHCl_3$), which did not depress the m. p. of an authentic sample.[18]

Ethyl 2-*Acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-2-*deoxy*-β-D-*glucoside*.—Anhydrous ethanol (125 ml.) was shaken with silver carbonate (5 g.) and anhydrous calcium sulphate (5 g.). After 30 min., crystalline acetochloroglucosamine (5 g.) was added and the mixture shaken for 16 hr. with the exclusion of light, then filtered. The filtrate was evaporated to dryness, the residue dissolved in chloroform (100 ml.) and shaken twice with cold 2% aqueous ammonia and twice with water, and the solution dried, and evaporated to dryness. The residue recrystallised from ethanol, to give *ethyl* 2-*acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-2-*deoxy*-β-D-*glucoside*, m. p. 167° (2·6 g., 51%), $[\alpha]_D^{16.5}$ −24·1° (*c* 1 in MeOH) (Found: C, 51·1; H, 6·7; N, 3·7. $C_{16}H_{25}O_9N$ requires C, 51·2; H, 6·7; N, 3·7%).

Treatment with methanolic ammonia [6] gave a crude product which on chromatography in solvent (*a*) gave spots of R_F 0·57 and 0·45 which correspond respectively to those of the ethyl 2-acetamido-2-deoxy-α- and -β-D-glucoside. Recrystallisation of the crude product from ethanol gave the β-*anomer*, m. p. 178°, $[\alpha]_D^{20}$ −48·9° (*c* 1 in H_2O) (Found: C, 48·9; H, 7·8; N, 5·6. $C_{10}H_{19}O_6N$ requires C, 48·2; H, 7·7; N, 5·5%).

Methyl 2-*acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-2-*deoxy*-β-D-*glucoside*, prepared similarly (42% yield), had m. p. 163°, $[\alpha]_D^{16.5}$ −22·2° (*c* 1 in MeOH) (Found: C, 49·9; H, 6·1; N, 3·8. $C_{15}H_{23}O_9N$ requires C, 49·9; H, 6·3; N, 3·9%), and gave *methyl* 2-*acetamido*-2-*deoxy*-β-D-*glucoside*, m. p. 204° (from ethanol), $[\alpha]_D^{19}$ −47·1° (*c* 1·5 in H_2O) (Found: C, 45·8; H, 7·5; N, 6·1. $C_9H_{17}O_6N$ requires C, 46·0; H, 7·3; N, 6·0%).

Phenyl 2-*Acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-2-*deoxy*-β-D-*glucoside*.—(i) Phenol (5 g.) and acetochloroglucosamine (5 g.) in cold acetone (105 ml.) were treated with 3·3% aqueous sodium hydroxide (45 ml.) and kept for 6 hr. at room temperature and overnight at 5°. The acetone was removed at room temperature and the products were shaken with chloroform (100 ml.). The chloroform layer was extracted with cold dilute alkali and with water, dried, and evaporated *in vacuo*. The residue was recrystallised from propan-2-ol, to give the *phenyl glycoside* (1·8 g., 32%), m. p. 204°, $[\alpha]_D^{18}$ −14·5° (*c* 1 in acetone) (Found: C, 57·5; H, 6·1; N, 3·3. $C_{20}H_{25}O_9N$ requires C, 56·7; H, 5·9; N, 3·3%).

(ii) Phenol (0·66 g.) was melted with mercuric cyanide (0·48 g.), acetochloroglucosamine (1 g.) was added, and the melt heated at 100° for 1 hr. with the exclusion of moisture. The product was dissolved in chloroform (20 ml.), extracted with water, dilute alkali, and water. After drying, the chloroform layer was evaporated and the residue recrystallised from propan-2-ol, to give the phenyl glycoside (150 mg., 13%), m. p. and mixed m. p. 204°, $[\alpha]_D^{20}$ −15·5° (*c* 1 in acetone).

p-*Nitrophenyl* 2-*Acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-2-*deoxy*-β-D-*glucoside*.—*p*-Nitrophenol (5 g.) and acetochloroglucosamine (5 g.) were kept in acetone (105 ml.) and 3·3% aqueous sodium hydroxide (45 ml.) for 6 hr. at room temperature and overnight at 5°. The crystals were filtered off. The acetone was evaporated at room temperature and the solid removed. The combined solids were washed with water and then ether, to give the *glycoside* (2·5 g., 39%). Recrystallisation from 1 : 1 methanol–chloroform gave a product, m. p. 240°, $[\alpha]_D^{18}$ −46·2° (*c* 0·5 in acetone) (Found: C, 51·3; H, 5·1; N, 6·0. $C_{20}H_{24}O_{11}N_2$ requires C, 51·2; H, 5·2; N, 6·0%).

The o-*nitrophenyl analogue*, prepared similarly in 42% yield, had m. p. 196—197° (from ethanol), $[\alpha]_D^{23}$ +3·4° (*c* 1 in acetone) (Found: C, 50·9; H, 5·0; N, 5·7%).

The p-*acetylphenyl analogue* (55% yield) had m. p. 219—220° (from water), $[\alpha]_D^{22}$ −14·6°

[23] Lobry de Bruyn and Van Eckenstein, *Rec. Trav. chim.*, 1899, **18**, 83.

(*c* 0·5 in acetone) (Found; C, 56·8; H, 5·8; N, 2·9. $C_{22}H_{27}O_{10}N$ requires C, 56·8; H, 5·8; N, 3·0%).

The 1-*naphthyl analogue* was obtained similarly. After evaporation of the reaction mixture the products were treated with ether (50 ml.). The solid was filtered off and washed with water and then ether to give a 23% yield. Recrystallisation from propan-2-ol gave a product, m. p. 210—211°, $[\alpha]_D^{21}$ −80·0° (*c* 0·5 in acetone) (Found: C, 60·6; H, 5·8; N, 2·7. $C_{24}H_{27}O_9N$ requires C, 60·9; H, 5·7; N, 2·9%).

Table 2 records removal of the *O*-acetyl groups from the aryl compounds.

1 : 3 : 4 : 6-*Tetra*-O-*acetyl*-2-*amino*-2-*deoxy*-α-D-*glucose Hydrochloride* (III).—Acetochloroglucosamine (3·5 g.) was kept in nitromethane (25 ml.) containing 2N-hydrochloric acid (0·025 ml.) overnight; the white needles were then filtered off, water (0·025 ml.) was added, and the procedure repeated until no further product crystallised out. The *hydrochloride* (3·1 g., 85%), recrystallised from acetic acid, decomposed at 160—186° and had $[\alpha]_D^{22}$ +141° (*c* 1 in H_2O) (Found: C, 44·3; H, 5·7; N, 3·7; Cl, 9·1. $C_{14}H_{22}O_9NCl$ requires C, 43·9; H, 5·5; N, 3·7; Cl, 9·2%). It is soluble in water and acetic acid but insoluble in most other organic solvents including chloroform and nitromethane.

The infrared spectrum (Table 1) shows a complicated set of partly unresolved bands between 3000 and 2500 cm.$^{-1}$, corresponding to reports that amino-acid hydrochlorides show an almost continuous series of bands between 3300 and 2500 cm.$^{-1}$. A small band at 2015 cm.$^{-1}$ is in the position where many hydrochlorides absorb, and the double peaks at 1595 and 1580 cm.$^{-1}$ are found in amino-acid hydrochlorides containing NH_3^+.

2-*Acetamido*-1 : 3 : 4 : 6-*tetra*-O-*acetyl*-2-*deoxy*-α-D-*glucose* (VII).—The salt (III) (5 g.) was kept in pyridine (50 ml.) and acetic anhydride (12·5 ml.) overnight. The mixture was evaporated under reduced pressure and the residue evaporated to dryness several times with absolute methanol. The crystalline residue was dissolved in water (100 ml.) and extracted with chloroform (5 × 15 ml.). The combined chloroform extracts were dried and evaporated to dryness. The residue, recrystallised from ethyl acetate–light petroleum, gave compound (VII), m. p. and mixed m. p. 134—135°, $[\alpha]_D^{21}$ +94° (*c* 1 in $CHCl_3$) (4·2 g., 86%).

1 : 3 : 4 : 6-*Tetra*-O-*acetyl*-2-*amino*-2-*deoxy*-α-D-*glucose* (IX).—The salt (III) (2·1 g.) was shaken in the absence of light for 6 hr. with dry chloroform (50 ml.), anhydrous calcium sulphate (2 g.), and silver carbonate (3 g.), then filtered, and the filtrate was evaporated to dryness at 37°. The crystalline residue was dissolved in warm ethyl acetate (5 ml.) and centrifuged and the supernatant liquid was treated with light petroleum to turbidity and left at 5° overnight, to give the 2-*amino-compound* (1·5 g., 79%). A further recrystallisation gave a product having m. p. 119°, $[\alpha]_D^{20}$ +145·2° (*c* 1 in $CHCl_3$) (Found: C, 48·3; H, 6·1; N, 3·9. $C_{14}H_{31}O_9N$ requires C, 48·4; H, 6·1; N, 4·0%).

2-*Acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-1 : 2-*dideoxy*-1-p-*toluidino*-D-*glucose* (X).—The salt (III) (1·1 g.) was shaken for 24 hr. with *p*-toluidine (0·96 g.) in chloroform (120 ml.), then set aside for several days. Toluidine hydrochloride separated and was filtered off; the brownish filtrate was evaporated to dryness and the semicrystalline residue washed with ether to give the *product* (0·68 g., 54%) which, recrystallised from ethanol, had m. p. 182—183° (decomp.), $[\alpha]_D^{20}$ −38·8° (*c* 1 in $CHCl_3$) (Found: C, 56·9; H, 6·8; N, 6·2. $C_{21}H_{28}O_8N_2$ requires C, 57·7; H, 6·5; N, 6·4%). This product with sodium methoxide [20] gave 2-acetamido-1 : 2-dideoxy-1-*p*-toluidino-D-glucose, $[\alpha]_D^{20}$ −9·5° ⟶ + 2·0° in 6 hr. (*c* 1 in H_2O), decomp. 193°. On chromatography in solvents (*a*) and (*b*) this compound gave spots of R_F 0·74 and 0·63 respectively, identical with those of the compound synthesised from 2-acetamido-2-deoxyglucose and *p*-toluidine.[20]

2-*Acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-2-*deoxy*-α-D-*glucose* (IV).—(i) The salt (III) (2·68 g.) and anhydrous sodium acetate (1 g.) were heated in water (50 ml.) at 65° for 6 hr. and, after cooling, extracted with chloroform (6 × 25 ml.). The combined chloroform extracts were dried (Drierite) and evaporated at 30°. The resulting syrup was dissolved, with slight warming, in anhydrous ether (75 ml.) and kept at −20° overnight. The ether was decanted from the white crystals (which are very deliquescent at this stage), and the flask and contents were dried *in vacuo* over sulphuric acid. The *product* (1·1 g., 45%) softened and melted at 65—75° and had $[\alpha]_D^{20}$ +49·4° (*c* 2·1 in $CHCl_3$), +50·4° ⟶ +26·9° in 4 hr. (*c* 0·5 in H_2O) (Found: C, 47·3, 48·0; H, 6·3, 6·1; N, 4·1, 4·0. $C_{14}H_{21}O_9N$ requires C, 48·4; H, 6·1; N, 4·0%).

(ii) The salt (III) (2·68 g.) and anhydrous sodium acetate (1 g.) were left in water (50 ml.) at room temperature. α (in a 1 dm. tube) of this solution fell in 4 days to a constant value of

+1·41° which corresponds to $[\alpha]_D^{20}$ +26·7° [assuming the transformation (III) ⟶ (IV)]. The solution was extracted as before and the above product isolated. This product was a white, slightly hygroscopic powder which was very soluble in water or chloroform, moderately soluble in ether, and gave a weak colour with Ehrlich's reagent: 10 mg. of the product in water (0·5 ml.) was mixed with acetic acid (4 ml.) and a 2% solution (0·5 ml.) of *p*-dimethylaminobenzaldehyde in acetic acid containing 2% v/v of 10N-hydrochloric acid; a colour developed which deepened during 48 hr.; then the solution had absorption max. at 540 (ε 206) and 580 mμ (ε 209).

The infrared spectrum (Table 1) shows bands at 3300 (intense) (NH and/or OH), and 1660 and 1540 cm.$^{-1}$ (secondary amide).

2-*Acetamido*-1 : 3 : 4 : 6-*tetra*-O-*acetyl*-2-*deoxy*-α-D-*glucose* (VII).—The compound (IV) (0·2 g.), with pyridine and acetic anhydride,[7] gave a penta-acetyl derivative (60% yield) identical in m. p. and [α] with an authentic sample.

2-*Acetamido*-3 : 4 : 6-*tri*-O-*acetyl*-2-*deoxy*-1-O-(3 : 5-*dinitrobenzoyl*)-α-D-*glucose* (VIII).—The compound (IV) (1 g.) with pyridine and 3 : 5-dinitrobenzoyl chloride [4] gave the product (VIII) (15%), m. p. 172—173°, $[\alpha]_D^{20}$ +94·4° (*c* 1 in $CHCl_3$).

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