831. *The Structure of Some* N-*Aryl*-D-*glucosylamines* *

By Brian Capon and Brian E. Connett

Evidence is presented that some dextrorotatory *N*-aryl-D-glucosyamines have the α-pyranose structure and some lævorotatory ones the β-pyranose structure.

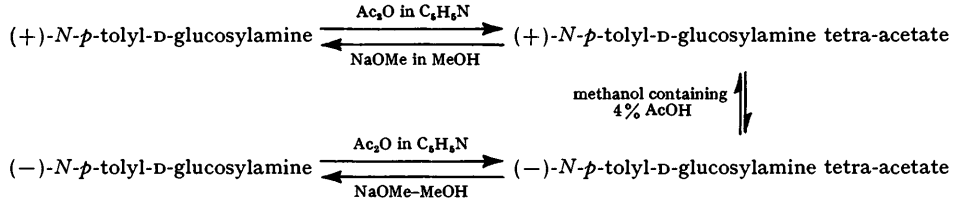
When the subject of the glucosylamines was last reviewed [1] only one, *N*-*p*-tolyl-D-glucosylamine, was known in more than one isomeric form despite two pyranose, two furanose, and two acylic Schiff's-base structures being possible. Since then Bognár and Nánási [2] have succeeded in isolating several isomeric pairs of *N*-aryl-D-glucosylamines and D-galactosylamines. The structures having generally been assumed to be pyranose, the positively

* For a preliminary account of this work see B. Capon and B. E. Connett, *Tetrahedron Letters*, 1964, 1391.

[1] G. P. Ellis and J. Honeyman, *Adv. Carbohydrate Chem.*, 1955, **10**, 95.
[2] R. Bognár and P. Nánási, *J.*, 1961, 323; *Magyar Kém. Folyóirat*, 1962, **68**, 32.

rotating isomers having the α-configuration and the negatively rotating isomers, the β-configuration. Evidence for these assumptions has, however, been lacking since the two classical methods for assigning ring-size to carbohydrate derivatives—methylation and periodate oxidation—both fail to give useful information. Methylation of glycosylamines gives only poor yields (<25%) of permethylated derivatives, and periodate oxidation results in complete oxidation.[1] Also, in view of the several reported failures of Hudson's rules,[3] the assignment of configuration to the anomeric carbon from the sign of rotation in the D-line must now be considered to be of dubious validity, especially with compounds containing a glycosyl–nitrogen bond. It was therefore decided that before embarking on the detailed mechanistic investigation of the hydrolysis of the *N*-aryl-D-glucosylamines more definite evidence for the structure of the compounds to be used should be obtained.

Acetylation Experiments.—The method chosen for determining ring-size was acetylation, but because this process might itself change the ring size it was necessary to design the experiments so that any such effect would be easily detected. This was done by carrying out the acetylation–deacetylation cycle shown for the *p*-tolyl compound in Scheme 1. The starting



Scheme 1

point is (—)-*N*-*p*-tolyl-D-glucosylamine tetra-acetate which has the β-pyranose structure, since it is formed from the reaction between tetra-*O*-acetyl-α-D-glucopyranosyl bromide and *p*-toluidine, which proceeds with inversion of configuration; [4] this structure is also supported by the n.m.r. spectrum (see below). On deacetylation, under conditions in which the product is precipitated the moment it is formed, the (—)-*N*-*p*-tolyl-D-glucosylamine was obtained in quantitative yield. This may be reconverted into the (—)-tetra-acetate in 98% yield by careful acetylation with 50% excess of acetic anhydride in dry pyridine (it was shown that the glucosylamine was stable in pyridine containing acetic acid equivalent to that formed in the acetylation). Thus if a change in ring-size occurred on deacetylation it would have occurred quantitatively and stereospecifically and have been quantitatively reversed in the acetylation step. This is considered to be extremely unlikely and hence the negatively rotating *N*-*p*-tolyl-D-glucosylamine almost certainly has a pyranose structure. The β-tetra-acetate undergoes a smooth isomerisation in methanol in the presence of acetic acid to yield an equilibrium mixture containing 31·6% of a positively rotating tetra-acetate, separable by fractional crystallisation. Kinetically, this behaves as a first-order reversible reaction and is certainly an anomerisation. If change in ring-size occurred, there would have to be extensive acetyl migration, and the simple kinetic behaviour would not be observed. The α-pyranose structure for the (+)-tetra-acetate is also supported by the n.m.r. spectrum (see below). This could also be made to undergo an acetylation–deacetylation cycle with the (+)-glucosylamine similar to that described with the negatively rotating isomers, thus indicating that this has a pyranose structure.

A similar series of reactions was carried out with both the phenyl-D-glucosylamines.

Nuclear Magnetic Resonance Spectra.—The spectra of both isomers of *N*-*p*-tolyl-D-glucosylamine in pyridine are reproduced in Figures 1 and 2. Integrals, present in the original spectra, have been omitted. In both cases the hydroxylic and nitrogen-bound

[3] J. J. Fox, N. C. Yung, and M. Hoffer, *J. Amer. Chem. Soc.*, 1961, **83**, 4066; R. U. Lemieux and M. Hoffer, *Canad. J. Chem.*, 1961, **39**, 110.

[4] Cf. L. J. Haynes and F. H. Newth, *Adv. Carbohydrate Chem.*, 1955, **10**, 243.

protons were exchanged twice with deuterium oxide–dioxan. The three *para*-methyl protons appeared as a very sharp singlet at 2·20 p.p.m. The six protons on C-2 ⟶ C-6 for the dextrorotatory isomer gave rise to a doublet at 4·37 p.p.m. The signal occurring at lowest field was a doublet, corresponding to one proton. This signal was absent from the spectrum of the glucosylamine prepared from D-[$^2H_1$]-glucopyranose and was therefore that of the proton at C_1. The splitting of 4 c./sec. is that to be expected from the α-D-glucopyranose configuration.[5] The lævorotatory isomer showed a rather poorly resolved group of peaks at 3·8 ⟶ 4·3 p.p.m. corresponding to the protons on C-2 ⟶ C-6.

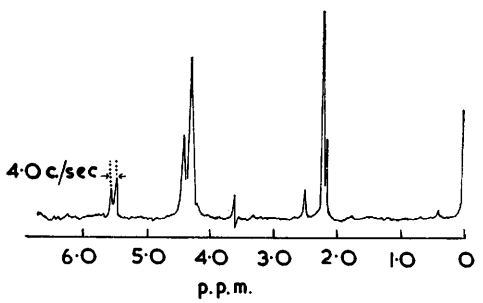


FIGURE 1. Nuclear magnetic resonance spectrum of *N*-*p*-tolyl-α-D-glucosylamine

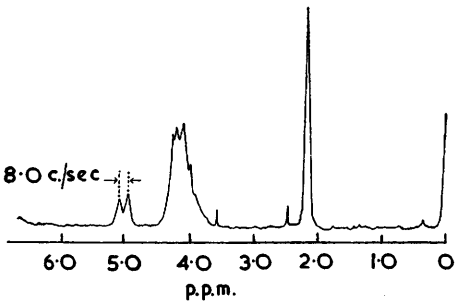


FIGURE 2. Nuclear magnetic resonance spectrum of *N*-*p*-tolyl-β-D-glucosylamine

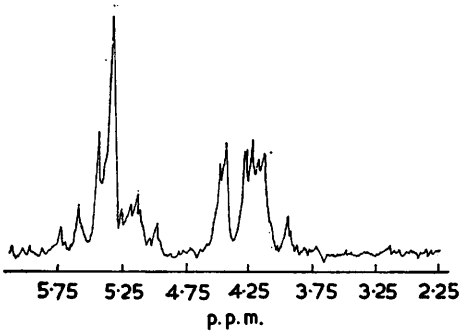


FIGURE 3. Nuclear magnetic resonance spectrum of 2,3,4,6-tetra-*O*-acetyl-*N*-*p*-tolyl-α-D-glucosylamine

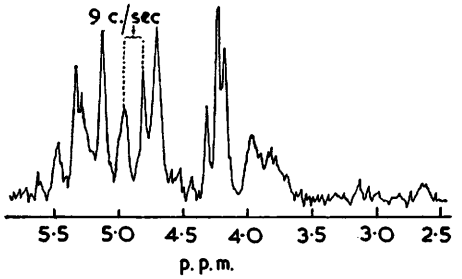


FIGURE 4. Nuclear magnetic resonance spectrum of 2,3,4,6-tetra-*O*-acetyl-*N*-*p*-tolyl-β-D-glucosylamine

The anomeric proton signal, which disappeared as before on substitution with deuterium, was at 5·05 p.p.m. with a splitting of 8·0 c./sec., thus confirming the β-D-glucopyranose configuration.[5] Additional peaks at 2·5 and 3·6 p.p.m. were observed. The former is due to a small amount of methyl-substituted pyridines in the pyridine and the latter to a small amount of residual nitrogen-bound hydrogen.

The chemical shifts and splittings for the anomeric protons of all the glucosylamines studied are given in Table 1, confirming the structures given. With one compound, the *N*-*o*-carboxyphenyl-D-glucosylamine, two well-resolved doublets were obtained, and it is clear that this is a mixture of isomers. All attempts to separate them failed.

Analysis of the n.m.r. spectra of the acetylated glucosylamines is more difficult as the signals from the anomeric protons do not appear at lower fields than the signals from the other ring protons (cf. Figures 3 and 4). The chemical shifts for all the ring protons are therefore very similar, and analysis of the spectra by a simple first-order treatment is not justified. The spectrum of tetra-*O*-acetyl-*N*-*p*-tolyl-β-D-[$^2H_1$]-glucosylamine lacked two signals centred at 4·87 p.p.m. and 9·0 c./sec. apart and that of its α-anomer lacked a signal

[5] Cf. R. U. Lemieux and D. R. Lineback, *Ann. Rev. Biochem.*, 1963, **32**, 155.

TABLE 1

Glucosylamine	Chemical shift (p.p.m.*)	Splitting (c./sec.)
N-*p*-Tolyl-α-D-	5·54	J_{1e2a} : 4·0
N-*p*-Tolyl-β-D-	5·05	J_{1a2e} : 8·0
N-Phenyl-α-D-	5·48	J_{1e2a} : 4·0
N-Phenyl-β-D-	5·15	J_{1a1a} : 7·5
N-*p*-Hydroxyphenyl-β-D-	5·13	J_{1a2a} : 8·5
N-*p*-Trifluoromethylphenyl-β-D-	5·05	J_{1a2a} : 8·0
N-*p*-Carboxyphenyl-β-D-	5·20	J_{1a2a} : 7·0
N-*o*-Carboxyphenyl-D-	5·15	J_{1a2a} : 7·5
	5·62	J_{1e2a} : 5·0
N-*p*-Nitrophenyl-α-D-	5·62	J_{1a2a} : 4·0
N-*p*-Nitrophenyl-β-D-	5·23	J_{1a2a} : 7·0

* From tetramethylsilane.

at 5·30 p.p.m., present in the protonated compounds. Although a detailed theoretical analysis of these spectra is not possible the results support the configurational assignments given, the signal from the β-compound with the axial anomeric proton appearing at higher field than that from the α-compound with an equatorial proton.

EXPERIMENTAL

The properties of the glucosylamines and their acetylated derivatives used in this work are given in Tables 2 and 3.

TABLE 2

N-Aryl-D-glucosylamines

Aryl	Confign.	M. p. (lit. value)	$[\alpha]_D$ in MeOH (lit. value)	Footnote
N-*p*-Tolyl	α	140—141° (135—136°)	+239° (+209)	A
	β	112—113 (117—118)	−102 (−101)	A
N-*p*-Nitrophenyl	α	193—194 (174)	+342 (+337)	B, D
	β	160—161 (184)	−161 (−165)	B
N-*p*-Trifluoromethylphenyl	β	155—156	−102	C
N-Phenyl	α	149—150 (139—140)	+237 (+221)	B
	β	135—136 (123—124)	−105 (−105)	B
N-*p*-Hydroxyphenyl	β	149—150 (148—149)	−102 (−89·4 in H_2O)	A
N-*p*-Carboxyphenyl	β	128—129 (127)	−111 (−112)	A
N-*o*-Carboxyphenyl	α + β	131—132 (131—132)	+87·6 (+65 in 50% EtOH)	A

A, See G. P. Ellis and J. Honeyman, *Adv. Carbohydrate Chem.*, 1955, **10**, 136. B, R. Bognár and P. Nánási, *Magyar Kém. Folyóirat*, 1962, **67**, 32. C, See Experimental section. D, Found: C, 47·2; H, 5·3; N, 9·0. Calc. for $C_{12}H_{16}N_2O_7$: C, 48·0; H, 5·3; N, 9·3%.

TABLE 3

Tetra-*O*-acetyl *N*-aryl-D-glucosylamines

Aryl	Confign.	M. p. (lit. value)	$[\alpha]_D$ in $CHCl_3$ (lit. value)	Footnote
N-*p*-Tolyl	α	143—144° (143°)	+177° (+223 in MeOH)	A
	β	150—151 (145—146)	−50·2 (−34·2)	B
N-*p*-Nitrophenyl	α	168—169 (153)	+300 (+310)	A
	β	180—181 (180)	−119 (−120)	B
N-*p*-Trifluoromethylphenyl	α	178—179	+179	C
	β	176—177 (184—185)	−67 (−61·1 in EtOH)	D
N-Phenyl	α	159—150 (149—150)	+185 (+180)	B
	β	93—94 (95—96)	−57·3 (−59·9)	B
N-*p*-Acetoxyphenyl	α	160—161	+165	C
	β	134—135 (133)	−52·0	B

A, R. Bognár and P. Nánási, *Magyar Kém. Folyóirat*, 1962, **67**, 32. B, See G. P. Ellis and J. Honeyman, *Adv. Carbohydrate Chem.*, 1955, **10**, 136. C, See Experimental section. D, See J. F. Sibley, Ph.D. Thesis, University of London, 1960.

N-p-*Trifluoromethylphenyl*-β-D-*glucosylamine*.—α-D-Glucose (3·6 g., 0·02 mole) and *p*-aminobenzotrifluoride (3·6 g., 0·01 mole) were refluxed for 7 hr. in dry methanol (60 ml.). The solution was concentrated to half bulk *in vacuo* and diethyl ether–petroleum (b. p. 40—60°)(1 : 1 v/v) was added to give a slight turbidity. It was left at 0° for 2 days. Crystals of the

glucosylamine were deposited slowly and were recrystallised from dry methanol–petroleum (b. p. 40—60°) (Found: C, 47·0; H, 4·8; F, 16·9; N, 4·2. $C_{13}H_{16}F_3NO_5$ requires C, 48·3; H, 5·0; F, 17·6; N, 4·7%).

N-*Phenyl*-α- *and* -β-D-*glucosylamines*.—α-D-Glucose (36 g., 0·2 mole) and redistilled aniline (18·6 g., 0·2 mole) were refluxed for 2 hr. in dry methanol (350 ml.). The solution was then concentrated to half bulk *in vacuo* (temp. <45°) and ether added to give slight turbidity. The white wax-like solid, $[\alpha]_D$ +54·5° (MeOH), which separated overnight at 0°, was filtered off dried, dissolved in pyridine, and acetylated at 0° with a 50% excess of acetic anhydride. The resulting anomeric tetra-acetate, $[\alpha]_D$ +72·0° ($CHCl_3$), was recrystallised twice from alcohol to give needles of 2,3,4,6-tetra-*O*-acetyl-*N*-phenyl-α-D-glucosylamine. They were made into a slurry with dry methanol and N-sodium methoxide (1·0 ml.) was added. A clear solution was obtained in a few seconds and almost immediately the deacetylated material was precipitated.[2] It was crystallised twice from dry methanol to give needle-shaped crystals of *N*-phenyl-α-D-glucosylamine. More of the mixture of the anomeric tetra-acetates was dissolved in the minimum quantity of boiling carbon tetrachloride. On cooling, the needle-shaped crystals that separated were filtered off and crystallised from diethyl ether–petroleum (b. p. 40—60°) to give 2,3,4,6-tetra-*O*-acetyl-*N*-phenyl-β-D-glucosylamine. This was deacetylated as described for the α-isomer to give *N*-phenyl-β-D-glucosylamine, which was recrystallised from dry methanol.

2,3,4,6-*Tetra*-O-*acetyl*-N-p-*Trifluoromethylphenyl*-α-D-*glucosylamine and* N-p-*Acetoxyphenyl*-2,3,4,6-*tetra*-O-*acetyl*-α-D-*glucosylamine*.—The following chromatographic method is similar to the method used by Wickberg for the separation of acetylated sugars.[6] A 24 × 36 mm. sheet of Whatmann No. 3 chromatography paper was impregnated twice with a 20% solution of dimethyl sulphoxide in toluene. The toluene was removed by drying in an oven at 60°. An anomeric mixture of the tetra-acetate (250 mg.) was applied, in chloroform solution, as a strip along the top of the paper. Development by the descending method, with isopropyl ether containing 5% chloroform, was stopped when the solvent front reached the base of the paper. The mixture separated into two distinct bands which were positioned by spraying two strips (cut from each vertical edge of the paper) with aniline hydrogen phthalate and drying them at 120°. The strip of paper containing the α-isomer (the upper band in both cases) was removed and eluted with chloroform. This was evaporated to dryness and the residue (the tetra-acetate dissolved in dimethyl sulphoxide) was warmed *in vacuo* in the presence of a large quantity of water. This removed the last traces of dimethyl sulphoxide by steam distillation the *tetra-acetate* then being precipitated; it was filtered off and recrystallised from aqueous methanol (Found: (i) C, 51·7; H, 4·9; F, 11·6; N, 2·85. $C_{21}H_{24}F_3NO_9$ requires C, 51·3; H, 4·9; F, 11·6; N, 2·85%. (ii) C, 54·7; H, 6·1; N, 2·9. $C_{22}H_{27}NO_{11}$ requires C, 54·7; H, 5·9; N, 2·9%).

Acetylation–Deacetylation Cycles.—These were performed with both isomers of the *N*-phenyl and *N*-*p*-tolyl compounds. The tetra-acetates were deacetylated by making into a slurry in a little dry methanol and adding a few drops of M-sodium methoxide solution. After about a minute a clear solution resulted and after a short time the free glucosylamine was deposited in quantitative yield. The glucosylamines were reacetylated in dry pyridine at 0° by slowly adding acetic anhydride (50% excess). The solution was left overnight at 0° and then poured on to crushed ice. The resulting tetra-acetates were crystallised from aqueous methanol. Yields were better than 95%.

TABLE 4

Anomerisation of the glucosylamine tetra-acetates

(Solvent, 4% v/v acetic acid in methanol; temp., 25°)

Glucosylamine acetate	$k \times 10^4$ (sec.$^{-1}$)	Glucosylamine acetate	$k \times 10^4$ (sec.$^{-1}$)
N-*p*-Trifluoromethylphenyl-β-D- ...	0·16	*N*-*p*-Tolyl-β-D-	17·7
N-*p*-Hydroxyphenyl-β-D-	3·56	*N*-*p*-Tolyl-α-D-	18·2
N-Phenyl-α-D-	5·95		

Kinetic Measurements.—Kinetics of the anomerisation of the glucosylamines and their tetra-acetates were followed with a Bendix–Ericsson NPL/ETL automatic polarimeter fitted with a thermostatically controlled cell-holder. The results are given in Table 4.

[6] B. Wickberg, *Acta Chem. Scand.*, 1958, **12**, 615.

Nuclear Magnetic Resonance Spectra.—Nuclear magnetic resonance spectra were recorded for the free bases in dry pyridine and for the tetra-acetate in deuterochloroform.

We thank Professor W. B. Whalley for placing a Varian A-60 n.m.r. spectrometer at our disposal and Miss J. Lovenack for measuring the spectra. We thank also Professor W. G. Overend for his interest and encouragement.

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