References

Broomhead, J. M. (1951). *Acta Cryst.* **4**, 92–100.

Bugg, C. E. (1970). *Proceedings, Fourth Jerusalem Symposium on the Purines*, pp. 178–204. New York: Academic Press.

Busing, W. R. & Levy, H. A. (1957). *Acta Cryst.* **10**, 180–182.

Cromer, D. T. (1965). *Acta Cryst.* **18**, 17–23.

Cromer, D. T. & Waber, J. T. (1965). *Acta Cryst.* **18**, 104–109.

Destro, R., Kistenmacher, T. J. & Marsh, R. E. (1974). *Acta Cryst.* B**30**, 79–85.

Hamilton, W. & Ibers, J. (1968). *Hydrogen Bonding in Solids*, pp. 181–182. New York: Benjamin.

Howells, E. R., Phillips, D. C. & Rogers, D. (1950). *Acta Cryst.* **3**, 210–214.

International Tables for X-ray Crystallography (1962). Vol. III, pp. 202–203. Birmingham: Kynoch Press.

Kim, S.-H. & Rich, A. (1967). *Science*, **158**, 1046–1048.

Larson, A. C. (1967). *Acta Cryst.* **23**, 664–665.

Lipson, H. & Woolfson, M. M. (1952). *Acta Cryst.* **5**, 680–682.

Marsh, R. E. (1968). *Structural Chemistry and Molecular Biology*, pp. 484–489. San Francisco: Freeman.

O'Brien, E. J. (1967). *Acta Cryst.* **23**, 92–106.

Pauling, L. (1960). *The Nature of the Chemical Bond*, p. 260. Ithaca: Cornell Univ. Press.

Pimentel, G. C. & McClellan, A. L. (1960). *The Hydrogen Bond*, pp. 288–289. San Francisco: Freeman.

Rogers, D. & Wilson, A. J. C. (1953). *Acta Cryst.* **6**, 439–449.

Stewart, R. F., Davidson, E. R. & Simpson, W. T. (1965). *J. Chem. Phys.* **42**, 3175–3187.

Thewalt, U., Bugg, C. E. & Marsh, R. E. (1970). *Acta Cryst.* B**26**, 1089–1101.

Voet, D. & Rich, A. (1970). *Progr. Nucl. Acid Res. Mol. Biol.* **10**, 183–265.

Acta Cryst. (1975). B**31**, 2867

A Refined Model for *N*-Acetyl-α-D-glucosamine

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A set of counter data has been collected for *N*-acetyl-α-D-glucosamine and the model refined by full-matrix least-squares calculations. For 1230 reflections 98·4% of all data within the Cu $K\alpha$ limit, $R(F)=0{\cdot}0237$, $R_w(F)=0{\cdot}0280$. Except for differences at the anomeric carbon the molecular conformation is similar to that of *N*-acetyl-α-D-glucosamine in the complex with triclinic lysozyme. The conformation found in a previous study of the compound [Johnson, *Acta Cryst.* (1966), **21**, 885–891] is generally confirmed. There are, however, systematic and in part significant differences between the two sets of bond lengths and valency angles. The present values for the glucopyranose structure are normal. The C–N bond of 1·346 (2) Å in the *N*-acetyl group is a long peptide-type C–N bond, all other structure parameters in this part of the molecule agree closely with averaged values for the peptide group. Most intermolecular hydrogen bonds are involved in helical chains running in the polar direction. These and a number of van der Waals interactions lead to tight packing and low thermal motion. The present study provides some evidence for a small amount of β sugar in the crystal as was suggested in the original analysis.

Introduction

N-acetylglucosamine (NAG) inhibits the enzymatic function of lysozyme (Wenzel, Lenk & Schütte, 1961; Rupley, 1964). Since it binds to lysozyme in both the tetragonal and triclinic forms, its structure in the crystalline state is of considerable interest. An analysis based on film data has already been reported (Johnson, 1966).

NAG is a derivative of D-glucose and it is a major constituent of a number of biological polymers such as chitin, hyaluronic acid, bacterial cell-wall polysaccharides, and some of the blood group polysaccharides. Thus, its structure is important in its own right, and a precise model would be useful for comparative purposes. We report such a model here based on counter data.

Experimental

A commercial sample of NAG was recrystallized from aqueous methanol containing a small amount of acetone. A prismatic crystal of dimensions 0·15 × 0·28 × 0·78 mm was mounted with its *b* axis (longest dimension) tilted *ca* 3° from the diffractometer φ axis and used for the X-ray measurements.

Crystallographic data are given in Table 1 together with those of Johnson (1966) (referred to hereinafter as J). Cell dimensions were determined from the setting angles of 20 reflections. The reciprocal vector $\mathbf{R}_{00l}$ chosen by J corresponds to $\mathbf{R}_{h0\bar{h}}$ in the present analysis. Transformation of our values according to the matrix

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Table 1. *Crystal data*

N-Acetylglucosamine, $C_8H_{15}O_6N$, F.W. 221·21

	Present work	Johnson (1966)	Recalculated
a	11·573 (2) Å	11·25 (3) Å	11·289 Å
b	4·849 (1)	4·82 (1)	
c	9·739 (2)	9·72 (2)	
β	116·72 (2)°	113·7 (1)°	113·70°
V	488·2 Å^3	483 Å^3	
D_x	1·505 g cm^{-3}	1·53 g cm^{-3}	(with $Z=2$)
D_m	–	1·55 (2)	

λ(Mo $K\bar{\alpha}$)=0·71069 Å
μ(Mo $K\bar{\alpha}$)=1·39 cm^{-1}
Space group: $P2_1$ (verified by the analysis)

$\mathbf{M}=(101/0\bar{1}0/00\bar{1})$ shows that the two sets of cell axes are within the 3σ limit given by J, but our dimensions are systematically larger.

The intensities of 2607 reflections of the equivalent classes $h\bar{k}\pm l, \bar{h}\bar{k}\pm l$ were measured with Nb-filtered Mo $K\alpha$ radiation on a computer-controlled diffractometer to a $\sin\theta/\lambda$ limit of 0·65. Operation was in the $\omega/2\theta$ scan mode at 2° min^{-1} in 2θ. The basic scan range before dispersion correction was 1·6°, and backgrounds were measured for 20 s at each end of the range. All 25 reflections below 2θ of 10° were remeasured manually to minimize errors caused by the Nb K absorption edge.

Three standard reflections, $50\bar{4}$, 305 and $0\bar{3}2$ were monitored at intervals of 100 reflections. The intensities of two of these decreased almost linearly to 98% of their initial values. The $50\bar{4}$ intensity, however, increased by about 1% during the first 24 h, and then gradually fell back to its initial value at the end of data collection. The data were scaled with a second degree polynomial to fit the normalized mean of the three standards.

All data were collected without attenuators, gross peaks ranging up to $3{\cdot}5\times10^6$ counts corresponding to a maximum rate of $2{\cdot}6\times10^5$ counts s^{-1}. 50 intense reflections were remeasured at reduced current, and a recovery constant for the system, $\tau=6{\cdot}5\times10^{-8}$ count^{-1}, determined graphically (*cf.* Sletten, Sletten & Jensen, 1969). A coincidence loss correction was applied to all reflections except the most intense one, $1\bar{1}\bar{1}$, which was scaled from its intensity at reduced current. No correction was made for absorption because the extreme variation in the transmission factor was estimated to be no more than ±1·5%.

Of the 61 unique $0kl$ reflections, one was measured only once and 54 were measured four times. Weighted averages of F^2 and $\sigma(F^2)$ were calculated for the two sets of pairs within the latter group and the averaging was repeated for the resulting total of 1249 pairs of equivalent reflections. Weighting was according to the expression $F^2_w=\sum(F^2_i/\sigma^2_i)/\sum(1/\sigma^2_i)$ and $\sigma_w=[1/\sum(1/\sigma^2_i)]^{1/2}$ where $\sigma_i=\sigma(F^2_i)=\sigma(I_i)\,(\mathrm{Lp})^{-1}$ (scale); $\sigma(I_i)=[(C'_i+T^2_iB'_i)+S^2(C_i-T_iB_i)^2]^{1/2}$, $i=1,2$. Symbols in the expression for $\sigma(I)$ are as defined by Mo & Adman (1975).

The differences $\Delta_i=|F^2_i-F^2_w|$ were calculated and the distribution of Δ_w was used to evaluate S. The weighted differences were assumed to follow a normal distribution, and S was adjusted from an initial value of 0·015 to 0·011 based on a frequency test of $z=\Delta_w/\sigma_w$. S=0·011 led to a value of 0·312 for the fraction of Δ's greater than σ and to 0·076 for the fraction greater than 2σ, the latter being considerably greater than expected for a normal distribution.

Among the 1249 pairs of intensities, there were 31 with $\delta_{12}=|F^2_1-F^2_2|>5\sigma_w$ and three with $\delta_{12}>7\sigma_w$. The discrepancy for all pairs of observations $D=\sum\delta_{12}/\sum F^2_w$ was 0·0124. 1230 reflections, 98·4% of the data, exceeded σ_w, and these were used in the least-squares refinement.

Structure determination and refinement

Although it was unnecessary to redetermine the structure, it was, in fact, solved by the multisolution tangent-refinement method. The conventional R for the non-hydrogen atoms from the correct E map was 0·179. Full-matrix least-squares refinement based on 1230 F's with $w=1/\sigma^2(F)$ converged quickly. Hydrogen atoms from a ΔF map were refined isotropically, nonhydrogen atoms anisotropically, and corrections for anomalous scattering were included. Data characterizing the refinement appear in Table 2.

The residual density in the final difference map ranges from −0·12 to 0·24 e Å^{-3} with the largest maxima in the C–C bonds. All atomic positions are within the density range −0·03 to 0·04 e Å^{-3}, but there are general negative regions above and below the main molecular plane at distances of *ca* 0·4–1·2 Å. Near some of the hydrogen atoms low maxima and minima occur, suggesting anisotropy not accounted for by the refinement. Positional and thermal parameters for all atoms are listed in Table 3.

Table 2. *Summary at the end of the least-squares refinement*

Total number of reflections	1250
Number of reflections in refinement, NO	1230
Number of variables, NV	196
$R(F)=\sum \lvert F_o\rvert-\lvert F_c\rvert /\sum\lvert F_o\rvert$	0·0237
$R_w(F)=[\sum w(\lvert F_o\rvert-\lvert F_c\rvert)^2/\sum wF^2_o]^{1/2}$	0·0280
$R(F^2)=[\sum w(F^2_o-F^2_c)^2/\sum wF^4_o]^{1/2}$	0·0337
$R(F)$ based on all 1250 F's	0·0245
$\mathrm{GOF}=[\sum w(\lvert F_o\rvert-\lvert F_c\rvert)^2/(\mathrm{NO}-\mathrm{NV})]^{1/2}$	4·7
Ave. shift/error	0·03
Max. shift/error	0·19

Atomic form factors for C, N and O atoms were from Doyle & Turner (1968); those for H (bonded) were from Stewart, Davidson & Simpson (1965).*

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* A list of structure factors has been deposited with the British Library Lending Division as Supplementary Publication No. SUP 31096 (7 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 13 White Friars, Chester CH1 1NZ, England.

Thermal motion

Three different models were considered in rigid-body analyses of thermal parameters according to the method of Schomaker & Trueblood (1968): (*A*) pyranose ring, (*B*) pyranose ring + O(1), N, O(3), O(4) and C(6), (*C*) all nonhydrogen atoms. With model *A* a good least-squares fit was obtained between thermal parameters calculated from the **T**, **ω** and **S** tensors, $U_{ij\mathrm{RBM}}$, and those from the structure refinement, $U_{ij\mathrm{exp}}$. The fit becomes poorer with increasing size of the model, but is still probably acceptable for model *B* (*cf.* Table 4). Results for the **T** and **ω** tensors in Table 5 have been calculated for model *B*. The largest libration of 4·0° is about an axis nearly normal to *b* and at an angle of approximately 15° to the long axis of

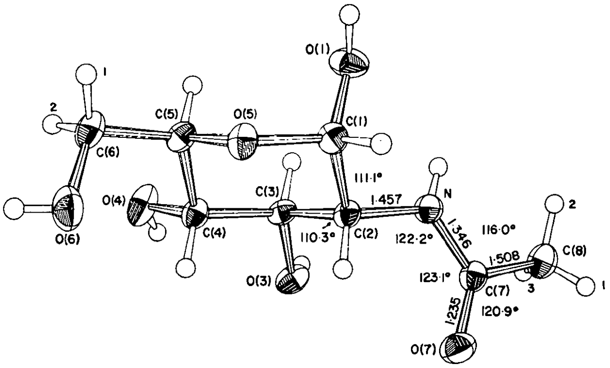

Fig. 1. Molecular conformation and atomic numbering of NAG with bond lengths and angles in the *N*-acetyl group. Sequential numbering of hydrogen atoms is indicated only where necessary. Thermal ellipsoids of the heavier atoms correspond to a 50% probability.

Table 4. *Results from least-squares fit to rigid-body motion for models A and B*

	Model *A*	Model *B*
R.m.s. $(U_{ij\,\mathrm{exp}} - U_{ij\,\mathrm{RBM}})$	0·0008 Å²	0·0016 Å²
Max. $(U_{ij\,\mathrm{exp}} - U_{ij\,\mathrm{RBM}})$	0·0018	0·0056
$\sigma(U_{ij\,\mathrm{RBM}})$	0·0012	0·0019
$\langle\sigma(U_{ij\,\mathrm{exp}})\rangle$	0·0006	0·0006

Table 5. *Rigid-body* **T** *and* **ω** *elements and r.m.s. values for the principal axes of the* **T** *and* **ω** *tensors referred to the axes* **a**, **b** *and* **c***

	135 (7)	5 (7)	−32 (6)	
T		188 (11)	−17 (7)	(Å² × 10⁴)
			228 (6)	
	9·9 (0·8)	−0·6 (0·6)	6·3 (0·5)	
ω		4·9 (0·6)	0·2 (0·7)	(deg²)
			9·8 (1·2)	

		Directions relative to axes		
Tensor	R.m.s. value	**a**	**b**	**c***
	0·16 Å	106·4°	108·2°	24·9°
T	0·14	95·6	18·2	72·7
	0·11	162·6	90·3	107·4
	4·0°	44·7	91·7	45·4
ω	2·3	76·9	159·4	105·7
	1·8	48·3	69·4	131·2

Table 3. *Final atomic parameters*

Thermal parameters, $U_{ij}(\times 10^4)$, as given here are defined by:

$$\exp\left[-2\pi^2(U_{11}a^{*2}h^2 + U_{22}b^{*2}k^2 + U_{33}c^{*2}l^2 + 2U_{12}a^*b^*hk + 2U_{13}a^*c^*hl + 2U_{23}b^*c^*kl)\right].$$

An average B_{iso} has been calculated for the heavier atoms where $B_{\mathrm{iso}} = \frac{1}{3}(B_1 + B_2 + B_3)$. Subscripts refer to the three principal axes of vibration and $B_{\mathrm{iso}} = 8\pi^2\langle U^2\rangle$. E.s.d.'s of the refined parameters appear in parentheses.

	x	*y*	*z*	U_{11}	U_{22}	U_{33}	U_{12}	U_{13}	U_{23}	B_{iso}
C(1)	0·3339 (1)	0·4195 (5)	0·4291 (2)	149 (6)	275 (8)	237 (7)	11 (7)	75 (5)	25 (7)	1·74 Å²
C(2)	0·2033 (1)	0·3149 (5)	0·4129 (2)	138 (6)	212 (7)	210 (6)	5 (6)	52 (5)	2 (6)	1·47
C(3)	0·1829 (1)	0·4096 (5)	0·5503 (2)	122 (6)	200 (7)	255 (6)	−12 (6)	75 (5)	−19 (6)	1·52
C(4)	0·2957 (1)	0·3222 (5)	0·7009 (2)	172 (6)	270 (8)	207 (6)	−3 (6)	81 (5)	−21 (6)	1·71
C(5)	0·4206 (1)	0·4390 (4)	0·7071 (2)	155 (6)	243 (8)	236 (7)	0 (6)	71 (5)	−26 (6)	1·67
C(6)	0·5425 (1)	0·3584 (5)	0·8484 (2)	163 (7)	379 (11)	241 (7)	0 (7)	36 (6)	−24 (7)	2·06
C(7)	−0·0044 (1)	0·2488 (4)	0·1843 (2)	184 (7)	278 (9)	217 (7)	−4 (6)	83 (5)	−11 (6)	1·79
C(8)	−0·1166 (2)	0·3898 (5)	0·0546 (2)	224 (7)	395 (11)	274 (8)	−15 (8)	23 (6)	10 (8)	2·35
O(1)	0·3272 (1)	0·7046 (4)	0·4124 (1)	208 (5)	299 (6)	442 (7)	−9 (5)	142 (5)	91 (6)	2·50
O(3)	0·0664 (1)	0·2913 (4)	0·5411 (1)	155 (5)	321 (7)	349 (6)	−54 (5)	132 (5)	−60 (5)	2·17
O(4)	0·2799 (1)	0·4317 (4)	0·8283 (1)	217 (5)	577 (9)	241 (5)	−32 (6)	112 (4)	−104 (6)	2·72
O(5)	0·4343 (1)	0·3366 (4)	0·5755 (1)	133 (4)	325 (7)	232 (5)	42 (5)	73 (4)	9 (5)	1·82
O(6)	0·5566 (1)	0·0680 (4)	0·8579 (2)	348 (7)	392 (8)	278 (6)	66 (6)	24 (6)	55 (6)	2·68
O(7)	−0·0082 (1)	0·0000	0·2092 (1)	283 (6)	274 (6)	367 (6)	−55 (5)	69 (5)	−14 (5)	2·43
N	0·0974 (1)	0·4103 (4)	0·2697 (1)	181 (5)	221 (7)	237 (6)	4 (6)	52 (5)	26 (6)	1·68

	x	*y*	*z*	*B* (Å²)		*x*	*y*	*z*	*B* (Å²)
H(Cl)	0·353 (1)	0·324 (3)	0·336 (1)	−0·05 (25)	H(C81)	−0·143 (2)	0·293 (6)	−0·028 (3)	5·2 (6)
H(C2)	0·205 (2)	0·114 (5)	0·412 (2)	1·7 (3)	H(C82)	−0·093 (2)	0·575 (7)	0·033 (3)	4·8 (6)
H(C3)	0·176 (2)	0·611 (5)	0·543 (2)	2·3 (4)	H(C83)	−0·177 (3)	0·432 (9)	0·090 (4)	8·1 (9)
H(C4)	0·299 (2)	0·118 (4)	0·704 (2)	1·5 (3)	H(O1)	0·399 (2)	0·766 (5)	0·406 (2)	3·7 (5)
H(C5)	0·417 (2)	0·638 (4)	0·701 (2)	1·7 (4)	H(O3)	0·022 (3)	0·400 (6)	0·535 (3)	4·2 (7)
H(C61)	0·616 (2)	0·443 (4)	0·839 (2)	2·5 (4)	H(O4)	0·205 (2)	0·416 (6)	0·816 (2)	3·7 (5)
H(C62)	0·536 (2)	0·435 (4)	0·938 (2)	2·4 (4)	H(O6)	0·604 (3)	0·043 (6)	0·945 (3)	4·6 (6)
					H(N)	0·088 (2)	0·575 (5)	0·256 (2)	2·4 (4)

Table 6. *Bond lengths and angles with standard deviations*

	Uncorr.	Corr.
C(1)–C(2)	1·534 (2) Å	1·537 Å
C(2)–C(3)	1·531 (3)	1·535
C(3)–C(4)	1·521 (2)	1·523
C(4)–C(5)	1·528 (3)	1·531
C(5)–O(5)	1·448 (2)	1·453
C(1)–O(5)	1·434 (2)	1·436
C(1)–O(1)	1·390 (3)	1·394
C(3)–O(3)	1·430 (2)	1·433
C(4)–O(4)	1·434 (2)	1·439
C(5)–C(6)	1·514 (2)	1·516
C(6)–O(6)	1·416 (3)	
C(2)–N	1·457 (2)	1·460

C(1)–H(C1)	1·12 (2)	O(5)C(1)C(2)	109·24 (15)°
C(2)–H(C2)	0·98 (2)	O(5)C(1)O(1)	111·90 (14)
C(3)–H(C3)	0·98 (2)	O(1)C(1)C(2)	108·42 (15)
C(4)–H(C4)	0·99 (2)	C(1)C(2)C(3)	110·11 (13)
C(5)–H(C5)	0·97 (2)	C(1)C(2)N	111·07 (15)
C(6)–H(C61)	0·99 (2)	NC(2)C(3)	110·31 (15)
C(6)–H(C62)	0·98 (2)	C(2)C(3)C(4)	110·88 (15)
C(8)–H(C81)	0·86 (3)	C(2)C(3)O(3)	109·68 (14)
C(8)–H(C82)	0·99 (3)	O(3)C(3)C(4)	108·97 (15)
C(8)–H(C83)	0·93 (4)	C(3)C(4)C(5)	108·77 (15)
O(1)–H(O1)	0·91 (3)	C(3)C(4)O(4)	110·06 (15)
O(3)–H(O3)	0·72 (3)	O(4)C(4)C(5)	108·34 (14)
O(4)–H(O4)	0·83 (3)	C(4)C(5)O(5)	108·41 (13)
O(6)–H(O6)	0·78 (3)	C(4)C(5)C(6)	114·77 (16)
N——H(N)	0·81 (3)	C(6)C(5)O(5)	106·72 (15)
		C(5)O(5)C(1)	114·97 (14)
		C(5)C(6)O(6)	110·44 (15)

Angles involving hydrogen

Type	Number	Range	Mean	$\sigma_{\rm ave}$
X*CH	22	105·9–111·9°	109·2°	1·1°
COH	4	103·2–112·0	108·5	2·1
CNH	2	117·2–117·7	117·5	1·2
HCH	4	101·6–117·4	109·4	2·6

* C, N, O.

the molecule. Libration corrections in bond lengths between nonhydrogen atoms change very little going from model *A* to *C*, the average difference being 0·0005 Å. Corrected bond lengths from the analysis of model *B* are given in Table 6.

Results and discussion

The molecular conformation

The present study of *N*-acetyl-α-*D*-glucosamine confirms the general features found by J but allows a more detailed analysis of the structure . Fig. 1 shows NAG in its *C*1 chair conformation. Some relevant torsion angles are given in Table 7.

Table 7. *Endo- and exocyclic torsion angles of NAG*

Only for exocyclic angles is the whole sequence of atoms *x–y–z–u* given. The sign convention for $\tau(xyzu)$ is that of Klyne & Prelog (1960).

Endocyclic	τ	Exocyclic	τ
C(1)C(2)	53·7°	O(1)C(1)C(2)N	54·0°
C(2)C(3)	−54·6	NC(2)C(3)O(3)	62·1
C(3)C(4)	56·8	O(3)C(3)C(4)O(4)	−63·8
C(4)C(5)	−58·4	O(4)C(4)C(5)C(6)	62·9
C(5)O(5)	62·5	C(5)O(5)C(1)O(1)	60·2
O(5)C(1)	−59·8	O(5)C(5)C(6)O(6)	−60·7
		C(4)C(5)C(6)O(6)	59·4

Endocyclic torsion angles τ vary from 53·7 to 62·5° which is a slightly narrower range than found, for example, in α-*D*-glucose, 51·3–62·2° (Brown & Levy, 1965), and the α-*D*-glucopyranose ring in α-lactose monohydrate, 51·2–62·9° (Fries, Rao & Sundaralingam, 1971). The difference results from a less flattened ring in NAG at the bonds C(2)–C(3) and C(3)–C(4). The primary hydroxyl group has a nearly ideal *gauche–gauche* or –synclinal orientation (Klyne & Prelog, 1960), one of the two favored arrangements of this group in glucopyranosides.

The atoms O(5), C(1), C(3) and C(4) in the ring are coplanar ($\chi^2=0·16$) and this plane, *r*, is nearly perpendicular (89·2°) to a plane *p* through atoms C(5)–C(6)–O(6). In the *N*-acetyl group only the atoms N–C(7)–O(7)–C(8) are coplanar ($\chi^2=2·42$). C(2) is displaced 0·214 Å from this plane which makes dihedral angles of 73·75° with plane *r* and 15·65° with plane *p*. The nonplanarity of the *N*-acetyl group is alternatively given as the torsion angle τ[C(2)NC(7)O(7)] = +9·9°. Similar nonplanar distortions of the peptide-type unit have been found for a number of amides and peptides; for example, a large torsion angle of 17·7° was reported for the peptide bond in *N*-acetyl-L-phenylalanyl-L-tyrosine (Stenkamp & Jensen, 1973). The hybridization at the nitrogen atom is not truly planar trigonal, H(N) being displaced 0·24 Å from a plane through atoms C(2)–N–C(7). CNDO/2 calculations by Ramachandran, Lakshminarayanan & Kolaskar (1973) indicate that the minimum energy conformations of simple amides involve pyramidal geometry at the nitrogen atom.

The sugar molecule in the triclinic lysozyme-NAG complex appears to be the β-anomer (Kurachi, Sieker & Jensen, 1974). Aside from the difference at the anomeric carbon the general conformation of complexed β-NAG as determined in the study at 2 Å resolution is similar to that found for α-NAG in the uncomplexed form. Both structures have the *C*1 chair conformation, the primary hydroxyl group is oriented *gauche–gauche* and O(6) and O(7) are located on the same side of the ring plane.

Bond lengths and angles

Bond lengths and valency angles with the e.s.d.'s are given in Table 6 and bond lengths and angles in the *N*-acetyl group are shown in Fig. 1. The discussion of these parameters will be based on values uncorrected for thermal motion.

There are systematic and in part significant differences between the present values of bond lengths and angles and those reported by J. 12 of the 15 bonds between nonhydrogen atoms are shorter in our analysis, the average and maximum deviations being 0·018 and 0·035 Å, respectively. The largest differences are in the bonds C(4)–O(4) and N–C(7). Of the 21 angles involving nonhydrogen atoms, 16 are larger in the present work, the average and maximum deviations being 1·95 and 3·9° respectively. Ten angles differ by more than $3\sigma_J$. The maximum deviation is in the angle O(5)–C(1)–O(1), the present value of 111·90(14)° appears to be normal for axial anomeric sugars.

All pyranose C–C bonds in NAG compare well with those of α-D-glucose (Brown & Levy, 1965) and the 1:1 α-D-glucose–urea complex (Snyder & Rosenstein, 1971). The results from the rigid-body analysis suggest that the shortening of C(5)–C(6) is a true structural feature, probably related to different substitution on the pyranosidic carbon atoms. Similar effects are observed, for example, in the condensed ring-systems of steroids and terpenes. In general, there is a larger spread among corresponding C–O bond lengths of the related carbohydrate structures, which appears to indicate that these bonds are more influenced than the C–C bonds by differences in polar and hydrogen bonding interactions in different crystal lattices. The axial anomeric C(1)–O(1)H bond of 1·390(3) Å is about 0·04 Å shorter than the other exocyclic C–O bonds [mean 1·432(2) Å] in agreement with previous observations (Berman, Chu & Jeffrey, 1967). The intracyclic C–O bonds differ in length by 7σ with C(1)–O(5) < C(5)–O(5).

None of the endocyclic C–C–X angles (X = C, O) deviates by more than 1·4° from the mean value of 109·5° ($\sigma_{\text{mean}} = 0{\cdot}14°$). The angle at the ring oxygen atom, 114·97(14)°, is within the normal range (Sundaralingam, 1968). Exocyclic valency angles at the ring atoms not involving hydrogen vary from 106·72 to 114·77° with a mean of 110·0° ($\sigma_{\text{mean}} = 0{\cdot}15°$). The pronounced angular asymmetry at C(5) is commonly observed in pyranose structures and seems to reflect the asymmetric intramolecular environment of this carbon.

With one exception the dimensions of the *N*-acetyl group agree with the average values for the peptide group calculated from ten X-ray structures by Marsh & Donohue (1967). The length of the peptide-type C–N bond varies considerably in different structures. Values ranging from 1·330 to 1·344 Å have been reported in a number of recent structure determinations, *cf.* Koetzle, Hamilton & Parthasarathy (1972), Kistenmacher, Hunt & Marsh (1972), Cotrait & Bideau (1974); however, in other cases this bond is in the range 1·310–1·320 Å (Cotrait & Barrans, 1974; Neuman, Gillier-Pandraud, Longchambon & Rabinovich, 1975).

The seven pyranosidic C–H bonds are 0·98 ± 0·01 Å except C(1)–H(C1) at 1·12 Å ($\sigma_{\text{mean}} = 0{\cdot}02$ Å). The O–H bonds vary over a wide range, 0·72–0·91 Å, as do the C–H bonds in the terminal methyl group, 0·86–0·99 Å. A survey of valency angles involving hydrogen atoms is given in Table 6.

Table 8. *Hydrogen-bond distances and angles*

Symmetry code

1	x	y	z	8	$-x$	$\frac{1}{2}+y$	$-z$
2	x	$1+y$	z	9	$1-x$	$-\frac{1}{2}+y$	$1-z$
3	x	$-1+y$	z	10	$1-x$	$\frac{1}{2}+y$	$1-z$
4	$-x$	$-\frac{1}{2}+y$	$1-z$	11	$1-x$	$-\frac{1}{2}+y$	$2-z$
5	$-x$	$\frac{1}{2}+y$	$1-z$	12	$1-x$	$\frac{1}{2}+y$	$2-z$
6	x	y	$-1+z$	13	x	y	$1+z$
7	$-x$	$-\frac{1}{2}+y$	$-z$				

Table 8 (*cont.*)

D–H···A	D···A	H···A	$(\text{H}\cdots A)^*_{\text{corr}}$	$(\angle D\text{–H}\cdots A)^*_{\text{corr}}$
O(1)–H(O1)···O(5)_{10}	2·784 (2) Å	1·88 (3) Å	1·82 Å	168°
O(3)–H(O3)···O(3)_5	2·789 (2)	2·13 (3)	1·90	150
O(6)–H(O6)···O(4)_{11}	2·856 (2)	2·08 (3)	1·88	172
O(4)–H(O4)···O(7)_5	3·016 (2)	2·21 (3)	2·07	163
N——H(N)····O(7)_2	3·061 (2)	2·29 (2)	2·10	158

* Distances and angles involving hydrogen have been recalculated (corr.) assuming lengths of 0·98 Å for the O–H and 1·015 Å for the N(sp^2)–H bonds.

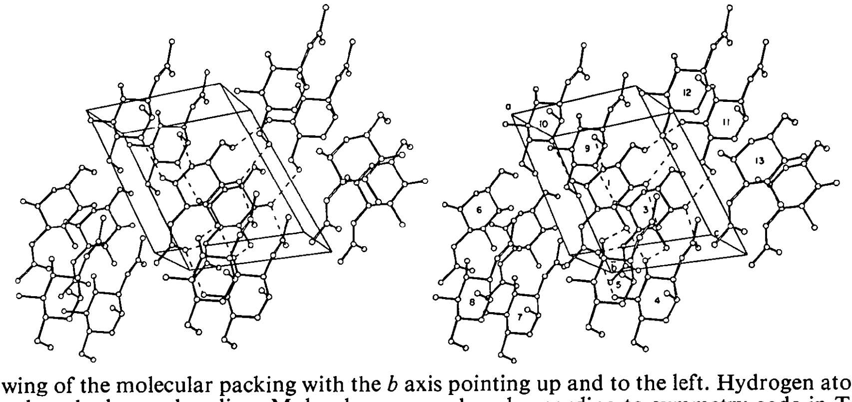

Fig. 2. Stereoscopic drawing of the molecular packing with the *b* axis pointing up and to the left. Hydrogen atoms are not drawn, broken lines show hydrogen bonding. Molecules are numbered according to symmetry code in Table 8.

The crystal structure

Fig. 2 is a stereo view of the molecular packing. Table 8 gives uncorrected hydrogen bond lengths and also corrected values for these bonds and angles assuming an O–H bond length of 0·98 Å (Brown & Levy, 1973) and a length of 1·015 Å for the N(sp^2)–H bond (Lehmann, Verbist, Hamilton & Koetzle, 1973).

The molecules form stacks along **b**. There are three normal van der Waals contacts (Kitaigorodskii,1973) between neighboring molecules within the same stack, $C(2)\cdots O(1)_3$:* 3·289(3), $C(8)\cdots O(7)_2$: 3·301(3) and $O(6)\cdots H(C5)_3$: 2·66(2) Å. Between molecules of adjacent stacks there are 11 different distances corresponding to van der Waals contacts. Atoms C(7), O(7) and the methyl group $C(8)H_3$ participate in about half of these contacts thereby accomplishing close packing and relatively low thermal motion in the *N*-acetyl group.

The hydrogen-bonding scheme is illustrated in Fig. 2. O(3) is both a donor and acceptor in hydrogen bonds between molecules related by screw axes at $0,y,\frac{1}{2}$. These bonds form left-handed helices; the sense donor to acceptor advancing along **b**. Another system of left-handed helical hydrogen bonds is formed around screw axes at $\frac{1}{2},y,\frac{1}{2}$ and involves O(1) and ring oxygen atoms O(5). A third set of helices, right-handed, include hydrogen bonds between O(6) and O(4) in adjacent molecules near screw axes at $\frac{1}{2},y,0$. The *N*-acetyl groups are packed around screw axes at $0,y,0$ and in these regions, therefore, van der Waals interactions predominate. A similar separation of intermolecular interactions in polar and nonpolar regions has been found in several other substituted carbohydrate structures (Berman & Kim, 1968).

In addition to the hydrogen-bond systems mentioned above there are pairs of much weaker hydrogen bonds in which O(4) and N are donors and carbonyl O(7) is acceptor. The hydrogen bond $O(4)\text{–}H(O4)\cdots O(7)_5$ which was not identified in the original analysis forms a second link between molecules related by screw axes at $0,y,\frac{1}{2}$. The hydrogen bond $N\text{–}H(N)\cdots O(7)_2$ is between adjacent molecules of the same stack.

Possible presence of the β-anomer

In the original analysis J found evidence pointing to the cocrystallization of both α and β anomers with 20–25% of the latter in the crystal [*cf.* also Strahs (1970), pp. 70–71]. A mixture of both sugar anomers in the same crystal is unusual, but has been reported, *e.g.*, for the structure of α-lactose monohydrate (Fries *et al.*, 1971). It was, therefore, a point of particular interest in the present analysis to study this possibility in more detail.

The position of H(C1) attached equatorially to C(1) was assigned to a peak in the ΔF map of density 0·64 e Å^{-3}, which is within the density range for hydrogen bonded to ring-carbon atoms. Refinement converged with the isotropic B for H(C1) at a physically unacceptable −0·05(25) Å^2 with the Stewart, Davidson & Simpson (1965) scattering-factor curve (SDS). Using the older f values of McWeeny (1951), J found the temperature factor of H(C1) refined to a large negative value as expected for a hydrogen atom with low thermal motion (Jensen & Sundaralingam, 1964). In the X-ray analysis of sucrose with the SDS f curve for H and data below $\sin\theta/\lambda = 0·64$ Å^{-1}, Hanson, Sieker & Jensen (1973) found B values for hydrogen atoms were consistently *ca* 2·0 Å^2 less than the values determined by Brown & Levy (1973) in the neutron diffraction study. There are other recent examples of B's attaining values near zero which emphasize the apparent underestimation of thermal parameters for hydrogen attached to carbon with the use of the SDS f curve, *cf.* Arora, Sundaralingam, Dancz, Stanford & Marsh (1973). Considering the value of B_{iso} of C(1) in NAG, the low B of H(C1) is not sufficient evidence of cocrystallized β anomer.

The relatively long C(1)–H(C1) bond of 1·12(2) Å, however, suggests the presence of some β anomer. The position of a β oxygen atom has been marked at a distance 1·39 Å from C(1) along the bond C(1)–H(C1) in Fig. 3 which shows the final residual densities near H(C1), H(O1) and H(O4), roughly in the plane of the covalent bonds. The highest density peak of 0·11 e Å^{-3} is on the 'inside' of the H(C1) position and it borders on negative troughs both above and below the section shown. The density features near other hydrogen atoms as *e.g.*, H(O1) and H(O4) are of comparable magnitude and appear to correspond to resid-

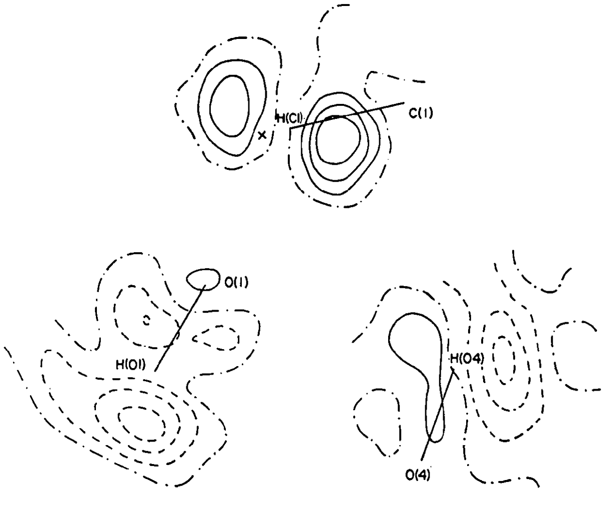

Fig. 3. Residual densities near some of the hydrogen atoms. The bonds are only approximately in the planes of the sections. Contour levels are at intervals of 0·03 e Å^{-3}; solid lines are positive, broken lines are negative and the zero level is chain dotted.

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* 3 denotes molecule at $x,-1+y,z$. For interpretation of symmetry code here and elsewhere, see Table 8.

ual anisotropy after the isotropic refinement of these atoms.

As was noted also by J, a β oxygen atom would make no short contacts with other atoms and could possibly form a hydrogen bond with O(6) in a neighboring molecule. We found an O(β) positioned 1·39 Å from C(1) to be at a distance of 2·63 Å from $O(6)_{10}$. The direction of approach suggests that a hydrogen bond could be formed with O(β) as a donor, not as an acceptor as suggested earlier (*cf* Fig. 2). This would bring about several changes in the hydrogen-bonding system, O(6) participating in two hydrogen bonds in the β anomer instead of one in the α form (see Table 8). O(5) would not be involved in hydrogen bonding in the β anomer and in addition there would be differences in the van der Waals interactions, notably a contact $O(\beta)\cdots C(5)_9$ of 3·19 Å instead of the $O(\alpha)\cdots C(2)_2$ contact at 3·289 Å. Such changes would probably affect the conformations of the α and β anomers differently, giving rise to apparent higher thermal motion for some of the ring atoms, but there is no indication of such effects. B_{iso} of the atoms in the pyranose ring range from 1·47 to 1·82 Å^2 and the movement of the ring fits closely the rigid-body model as shown in Table 4.

We cannot rule out the possibility that the β anomer might be present in small quantities, say a few per cent. In any case, it appears to be much less than in the earlier determination. This would not be surprising, since the relative amounts of α and β anomer may be sensitive to the conditions of crystallization.

Crystallographic programs from the X-RAY 70 System (Stewart, Kundell & Baldwin, 1970) were used for structure refinement and analyses of molecular geometry. Drawings of molecules were made by the *ORTEP* program (Johnson, 1965). Support by USPHS Grant GM-10828 from the National Institutes of Health is gratefully acknowledged.

References

Arora, S. K., Sundaralingam, M., Dancz, J. S., Stanford, R. H. & Marsh, R. E. (1973). *Acta Cryst.* B**29**, 1849–1855.

Berman, H. M., Chu, S. S. C. & Jeffrey, G. A. (1967). *Science*, **157**, 1576–1577.

Berman, H. M. & Kim, S. H. (1968). *Acta Cryst.* B**24**, 897–904.

Brown, G. M. & Levy, H. A. (1965). *Science*, **147**, 1038–1039.

Brown, G. M. & Levy, H. A. (1973). *Acta Cryst.* B**29**, 790–797.

Cotrait, M. & Barrans, Y. (1974). *Acta Cryst.* B**30**, 1018–1023.

Cotrait, M. & Bideau, J. P. (1974). *Acta Cryst.* B**30**, 1024–1028.

Doyle, P. A. & Turner, P. S. (1968). *Acta Cryst.* A**24**, 390–397.

Fries, D. C., Rao, S. T. & Sundaralingam, M. (1971). *Acta Cryst.* B**27**, 994–1005.

Hanson, J. C., Sieker, L. C. & Jensen, L. H. (1973). *Acta Cryst.* B**29**, 797–808.

Jensen, L. H. & Sundaralingam, M. (1964). *Science*, **145**, 1185–1187.

Johnson, C. K. (1965). *ORTEP*. Oak Ridge National Laboratory Report ORNL-3794.

Johnson, L. N. (1966). *Acta Cryst.* **21**, 885–891.

Kistenmacher, T. J., Hunt, D. J. & Marsh, R. E. (1972). *Acta Cryst.* B**28**, 3352–3361.

Kitaigorodskii, A. I. (1973). *Molecular Crystals and Molecules*. New York: Academic Press.

Klyne, W. & Prelog, V. (1960). *Experientia*, **16**, 521–523.

Koetzle, T. F., Hamilton, W. C. & Parthasarathy, R. (1972). *Acta Cryst.* B**28**, 2083–2090.

Kurachi, K., Sieker, L. C. & Jensen, L. H. (1974). Abstracts, ACA Meeting, Berkeley, California, March 23–28, Paper J9.

Lehman, M. S., Verbist, J. J., Hamilton, W. C. & Koetzle. T. F. (1973). *J. Chem. Soc. Perkin* II, pp. 133–137.

McWeeny, R. (1951). *Acta Cryst.* **4**, 513–519.

Marsh, R. E. & Donohue, J. (1967). *Advanc. Protein Chem.* **22**, 235–256.

Mo, F. & Adman, E. (1975). *Acta Cryst.* B**31**, 192–198.

Neuman, A., Gillier-Pandraud, H., Longchambon, F. & Rabinovich, D. (1975). *Acta Cryst.* B**31**, 474–477.

Ramachandran, G. N., Lakshminarayanan, A. V. & Kolaskar, A. S. (1973). *Biochim. Biophys. Acta*, **303**, 8–13.

Rupley, J. A. (1964). *Biochim. Biophys. Acta*, **83**, 245–255.

Schomaker, V. & Trueblood, K. N. (1968). *Acta Cryst.* B**24**, 63–76.

Sletten, E., Sletten, J. & Jensen, L. H. (1969). *Acta Cryst.* B**25**, 1330–1338.

Snyder, R. L. & Rosenstein, R. D. (1971). *Acta Cryst.* B**27**, 1969–1975.

Stenkamp, R. E. & Jensen, L. H. (1973). *Acta Cryst.* B**29**, 2872–2878.

Stewart, J. M., Kundell, F. A. & Baldwin, J. C. (1970). X-RAY 70 System of Crystallographic Programs, Computer Science Center, Univ. of Maryland.

Stewart, R. F., Davidson, E. R. & Simpson, W. T. (1965). *J. Chem. Phys.* **42**, 3175–3187.

Strahs, G. (1970). *Advanc. Carbohydr. Chem. Biochem.* **25**, 53–107.

Sundaralingam, M. (1968). *Biopolymers*, **6**, 189–213.

Wenzel, M., Lenk, H. & Schütte, E. (1961). *Z. Physiol. Chem.* **327**, 13–20.