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The crystal and molecular structure of a diglycosylamine: the *N*-analogue of peracetylated α,β-trehalose

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Abstract

Crystals of the disaccharide, (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6–tetra-*O*-acetyl-β-D-glucopyranosyl)amine belong to the space group $P2_1$ with $a=14.848(2)$, $b=21.838(3)$, $c=11.534(2)$ Å, and $\beta=113.72(4)°$ and $Z=4$. The crystal structure was solved by direct methods and refined by the full-matrix least squares procedure to an R value of 0.049 ($Rw=0.46$) for 4262 observed reflections. This work provides the first crystalline features in the diglycosylamine series and this molecule can be considered as a *N*-analogue of peracetylated α, β-trehalose. The conformational features of the two independent diglycosylamine molecules do not display any major significant differences. The D–glucopyranose residues have the usual $^4C_1$ chair conformations, whereas all the primary and secondary acetate groups display usual orientations. In the crystal structure the hydrogen atom covalently linked to the nitrogen at the glycosidic linkage could be located; its configuration is (*R*) providing the priority is given to the (*R*) carbon neighbor (C-1β). The torsional angles at the glycosidic linkages Φα (O-5–C-1α–N-1–C-1β), Φβ (C-1α–N-1–C-1β–O-5) of the two molecules have values of (57.6°,−78.8°) and (59.8°,−85.9°), respectively, slightly different from the conformation found in α, β-trehalose monohydrate. These conformations have been rationalized through the use of molecular mechanics calculations. The packing mode is highly anisotropic, with the existence of chains of molecules along the *c* axis, stabilized by numerous van der Waals contacts and by a strong hydrogen bond, involving the NH group of the glycosidic linkage and the carbonyl group of one acetate.

Keywords: Crystal structure; *N*-trehalose; Diglycosylamine

1. Introduction

Aminosugars are potent reversible inhibitors of glycosidases [1–3] and can be used for many therapeutic applications. Among these compounds, the glycosylamines are of interest since they are reversible inhibitors that bind to the active site. Some well known compounds used are the *pseudo* glycosylamines such as acarbose, amylostatin or adiposine that are powerful inhibitors of α-glycosidases.

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Diglycosylamines have been synthesized with the aim of obtaining a more stable compound than monoglycosylamine [4,5]. In diglycosylamines, the basicity of the glycosylamine nitrogen is expected to be decreased by the combined electron withdrawing effect of the two neighboring ring oxygen atoms. The β-D-glucopyranosylamine and the α, β anomer were both reported to be strong competitive inhibitors of β-D-glucosidase from *Aspergillus phoenicis* and *Aspergillus niger* [4].

The present study reports on the crystalline and molecular feature of the peracetylated α, β-glucopyranosylamine: (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*–acetyl-β-D-glucopyranosyl)amine. This molecule can be considered as a *N*-analogue of per acetylated α, β-trehalose. Since this is the first crystal structure of a NH linkage between two carbohydrates, there is a special interest for investigating the structural features of the linkage.

2. Materials and methods

Data collection.—The title compound was prepared following a route that has been reported previously [4]. Single crystals suitable for X-ray work were grown by slow evaporation of an ethanolic solution. A whitish ribbon-like crystal with dimensions of 0.005×0.03×0.95 mm was used for the X-ray measurement. Determination of the unit cell dimensions and collection of the diffraction intensities were performed on a four circle–diffractometer Philips PW1100 with monochromatised CuK_α radiation ($\lambda = 1.5418$ Å) performed at room temperature (292 K). The lattice constants were obtained by a least–squares procedure using 25 reflections with 2θ in the range 40–50°. The 'flying stepscan' scan mode was selected in the 2θ–ω mode throughout the data collection. The intensity of three reference reflections (measured every 2 h) did not show any significant change during the duration of the data collection. The observed intensities were corrected from Lorentz-polarization factor, but due to the small dimensions of the crystal, no absorption correction was made ($\mu = 0.91$ mm^{-1}). A total of independent 5606 reflections were measured up to $\theta = 122°$ of which 4262 with $I/\sigma(I) > 3$ were accepted and used for the following calculations. The scattering factors were taken from International Tables for X-ray Crystallography [6]. The density of the crystal was measured, as 1.32 (2), by the flotation method using a mixture of carbon tetrachloride and toluene; it compares well with the calculated density of 1.314, thereby confirming the occurrence of two independent molecules in the asymmetric unit. Crystal data are given in Table 1.

Structure elucidation.—The initial atomic coordinates of about half of the non-hydrogen atoms in the asymmetric unit were obtained by direct method using the SHELX program [7]. The remaining atoms were obtained by a series of Fourier differences. The quantity minimized in the refinement was $\Sigma w(Fo-Fc)^2$, with $w = 1/\sigma(Fo)$ where $\sigma(Fo)$ was the standard deviation of Fo estimated from counting statistics. The final discrepancy factor was R value of 0.049 and the $R_w = 0.46$ for all the non-hydrogen atoms with anisotropic temperature factors and the hydrogen atoms with isotropic temperature factor of the corresponding parent atom, for the 4262 observed reflections. Geometrical calculations and ORTEP illustration were obtained with PLATON [8].

Conformational analysis.—The potential energy surfaces of both *N*-α, β-trehalose (with the hydrogen of NH adopting the same chirality has in the crystal) and α,β-trehalose were calculated using the Tripos force-field [9]. The systematic search option was used. The hydrogen atoms of the

Table 1
Crystal data of (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)amine

Formula	$C_{28}H_{39}NO_{18}$
Molecular weight	677.61
Crystal system	Monoclinic
Space group	$P2_1$
a(Å)	14.848(2)
b(Å)	21.838(3)
c(Å)	11.534(2)
β(°)	113.72(4)
V(Å^3)	3424.0
D_{calc} (g cm^{-3})	1.314
D_{obs} (g cm^{-3})	1.32
Z	4
F (000)	1432.0
μ (cm^{-1})	0.91
Crystal size (mm)	0.005×0.03×0.95
T (K)	292
Θ range (°)	2.0–122.05
Wavelength (Cu K_α) (Å)	1.5418
Data set	−16:16, 0:24, 0:12
No. unique reflections	5606
No. observed [$F_o^2 > -3\sigma(F_o^2)$] reflections	4262
No. refined parameters	849
Final R	0.0495
Final wR	0.0458

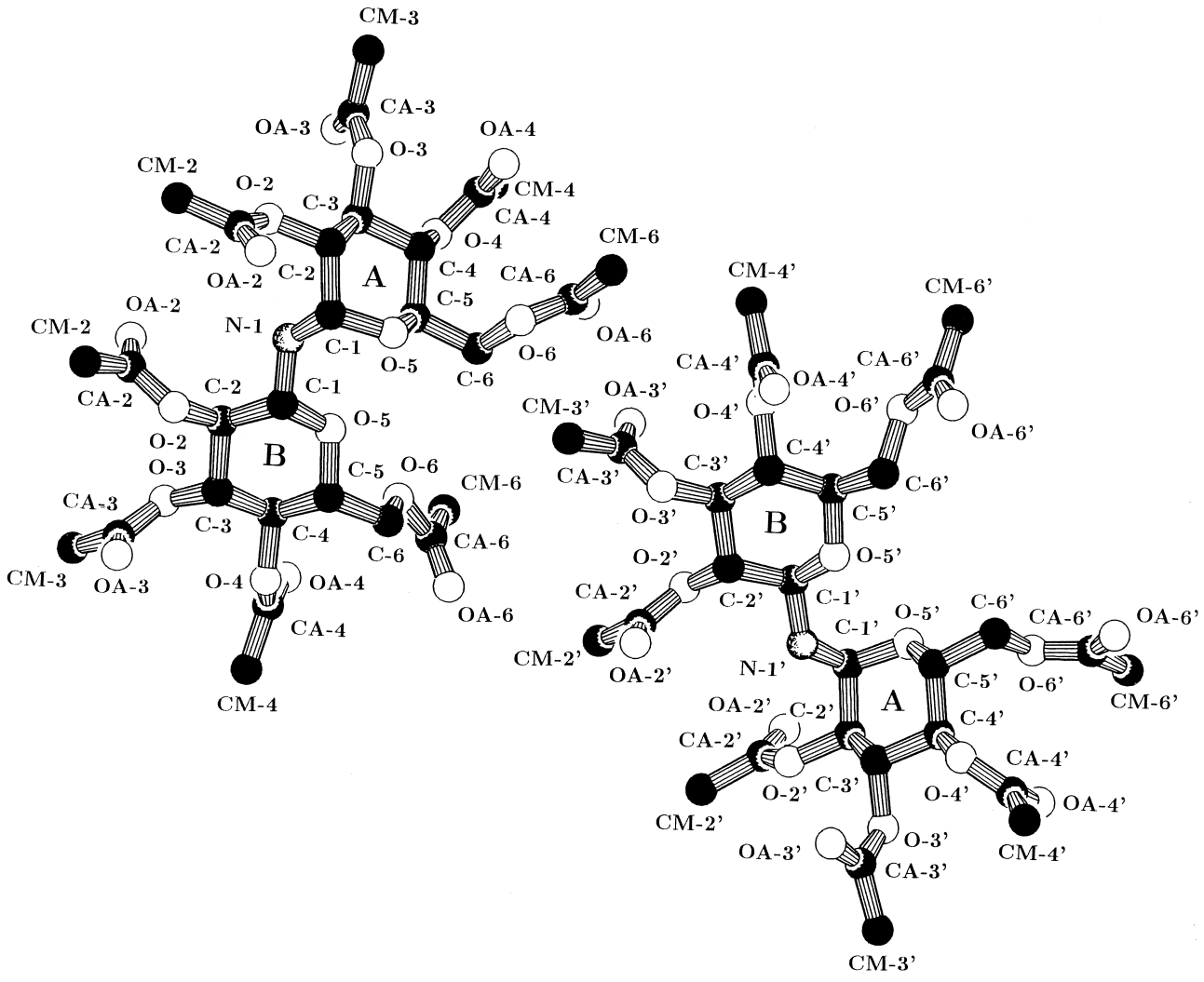

Fig. 1. Graphical representation of (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6–tetra-*O*-acetyl-β-D-glucopyranosyl)amine along with the labeling of the atoms. With respect to the coordinates listed in Table 1, the second molecule has been applied the symmetry operation $(-x, 1/2+y, -z)$.

hydroxyl groups were omitted and electrostatic interaction were not considered. The aim of the calculation was to estimate the steric energy surface of both compounds; therefore no special parametrization was used.

3. Results

The two independent molecules are represented in Fig. 1 together with the labeling of the atoms. The positional and isotropic thermal parameters for the non-hydrogen atoms are given in Table 2. An ORTEP representation of the two molecules constituting the asymmetric unit along with the thermal ellipsoids is shown in Fig. 2. The atomic coordinates of the hydrogen atoms and the anisotropic temperature factors have been deposited.[2] The bond lengths, bond angles, and torsion angles of interest are given in Tables 3–5, respectively. The estimated standard deviations of the bond lengths range from 0.007 to 0.013 Å, from 0.4 to 0.9° for the bond angles, and from 0.4 to 1.3° for torsion angles involving non–hydrogen atoms.

Conformation of the glucose rings.—The C–C bond lengths of the glucose rings have a mean value of 1.52 Å which conforms with 1.527 Å that is the mean value for carbohydrate ring [10]. The

[2] Data have been deposited with the Cambridge Crystallographic Data Center. These data may be obtained, on request, from the Director, Cambridge Crystallographic Data Center, 12 Union Road, Cambridge CB2 1EZ, UK. Tel: +44 223 336408; Fax: +44 223 336033.

Table 2
Final fractional coordinates and equivalent isotropic thermal parameters for the non hydrogen atoms of (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)amine

Atom	*x*/*a*	*y*/*b*	*z*/*c*	U(iso) (Å^2)
Molecule 1				
C-1A	0.2110(4)	0.3849(3)	0.6736(5)	0.042(3)
C-2A	0.0994(4)	0.3867(3)	0.6285(5)	0.043(3)
C-3A	0.0497(4)	0.4184(3)	0.5022(5)	0.033(3)
C-4A	0.0987(4)	0.4802(3)	0.5016(6)	0.043(3)
C-5A	0.2091(4)	0.4729(3)	0.5513(6)	0.049(4)
C-6A	0.2615(5)	0.5343(3)	0.5638(8)	0.058(4)
O-2A	0.0558(3)	0.3259(2)	0.6095(3)	0.048(2)
O-3A	−0.0505(3)	0.4302(2)	0.4874(4)	0.039(2)
O-4A	0.0662(3)	0.5007(2)	0.3727(4)	0.056(3)
O-5A	0.2438(3)	0.4468(2)	0.6749(4)	0.046(2)
O-6A	0.2274(4)	0.5772(2)	0.6300(4)	0.082(3)
CA-2A	0.0754(4)	0.2930(3)	0.7149(6)	0.041(4)
CA-3A	−0.1233(5)	0.4203(4)	0.3749(7)	0.056(5)
CA-4A	−0.0002(5)	0.5462(3)	0.3339(7)	0.059(4)
CA-6A	0.1924(6)	0.6311(3)	0.5735(8)	0.068(5)
CM-2A	0.0260(6)	0.2329(4)	0.6851(8)	0.099(6)
CM-3A	−0.2202(5)	0.4370(4)	0.3738(9)	0.038(4)
CM-4A	−0.0241(6)	0.5633(4)	0.1975(7)	0.083(5)
CM-6A	0.1572(7)	0.6694(4)	0.6511(8)	0.105(7)
OA-2A	0.1246(3)	0.3112(2)	0.8186(4)	0.075(3)
OA-3A	−0.1092(4)	0.4048(4)	0.2848(6)	0.068(4)
OA-4A	−0.0358(4)	0.5700(3)	0.3981(5)	0.108(4)
OA-6A	0.1907(5)	0.6454(2)	0.4730(6)	0.137(5)
C-1B	0.3403(4)	0.3344(3)	0.6276(5)	0.038(3)
C-2B	0.3588(4)	0.2741(3)	0.5744(6)	0.051(4)
C-3B	0.4635(4)	0.2679(3)	0.5855(6)	0.051(4)
C-4B	0.4966(4)	0.3243(3)	0.5388(5)	0.033(3)
C-5B	0.4812(4)	0.3798(3)	0.6090(6)	0.035(3)
C-6B	0.5187(4)	0.4396(3)	0.5827(6)	0.045(4)
N-1	0.2401(3)	0.3428(2)	0.6006(4)	0.037(3)
O-2B	0.3413(3)	0.2254(2)	0.6479(4)	0.054(3)
O-3B	0.4690(3)	0.2166(2)	0.5101(4)	0.060(3)
O-4B	0.6003(3)	0.3151(2)	0.5693(4)	0.048(2)
O-5B	0.3773(3)	0.3838(2)	0.5754(4)	0.036(2)
O-6B	0.4735(3)	0.4542(2)	0.4498(4)	0.045(2)
CA-2B	0.2720(6)	0.1838(3)	0.5906(8)	0.069(5)
CA-3B	0.5254(6)	0.1682(4)	0.5704(9)	0.078(5)
CA-4B	0.6303(4)	0.3191(3)	0.4730(6)	0.051(4)
CA-6B	0.5295(5)	0.4876(3)	0.4056(7)	0.052(4)
CM-2B	0.2701(7)	0.1362(4)	0.679(1)	0.102(7)
CM-3B	0.5316(8)	0.1243(4)	0.475(1)	0.150(1)
CM-4B	0.7309(5)	0.2947(4)	0.5102(8)	0.066(5)
CM-6B	0.4766(6)	0.5046(4)	0.2720(7)	0.109(7)
OA-2B	0.2179(5)	0.1878(3)	0.4812(6)	0.145(6)
OA-3B	0.5644(5)	0.1635(3)	0.6819(7)	0.158(6)
OA-4B	0.5789(4)	0.3399(3)	0.3709(4)	0.075(3)
OA-6B	0.6117(4)	0.5023(3)	0.4705(6)	0.065(4)
Molecule 2				
C-1A′	0.2100(4)	0.3921(3)	0.1466(5)	0.049(4)
C-2A′	0.0992(4)	0.3915(3)	0.1095(5)	0.042(3)
C-3A′	0.0448(4)	0.4144(3)	−0.0234(5)	0.043(3)
C-4A′	0.0812(4)	0.4781(3)	−0.0353(5)	0.033(3)
C-5A′	0.1907(4)	0.4756(3)	0.0015(6)	0.058(4)
C-6A′	0.2351(5)	0.5378(3)	−0.0029(7)	0.068(5)
O-2A′	0.0611(3)	0.3300(2)	0.1097(4)	0.048(2)
O-3A′	−0.0592(3)	0.4193(2)	−0.0504(4)	0.039(2)
O-4A′	0.0322(3)	0.4966(2)	−0.1654(4)	0.056(3)
O-5A′	0.2365(3)	0.4531(2)	0.1272(4)	0.046(2)
O-6A′	0.2154(4)	0.5776(2)	0.0855(5)	0.082(3)

(continued)

Table 2—*contd*

Atom	*x*/*a*	*y*/*b*	*z*/*c*	U(iso) (Å^2)
CA-2A′	0.0821(4)	0.3055(3)	0.2245(6)	0.041(4)
CA-3A′	−0.1221(5)	0.3928(4)	−0.1572(7)	0.056(5)
CA-4A′	−0.0259(5)	0.5463(3)	−0.1906(7)	0.038(3)
CA-6A′	0.2088(7)	0.6364(4)	0.0615(9)	0.105(7)
CM-2A′	0.0339(6)	0.2440(4)	0.2132(8)	0.099(6)
CM-3A′	−0.2256(6)	0.4082(5)	−0.1842(9)	0.038(4)
CM-4A′	−0.0812(7)	0.5549(4)	−0.3310(7)	0.083(5)
CM-6A′	0.1862(7)	0.6745(4)	0.1560(9)	0.059(4)
OA-2A′	0.1298(3)	0.3314(2)	0.3210(4)	0.075(3)
OA-3A′	−0.0962(4)	0.3615(4)	−0.2238(7)	0.068(4)
OA-4A′	−0.0353(5)	0.5757(3)	−0.1096(6)	0.108(4)
OA-6A′	0.2230(8)	0.6575(3)	−0.0234(8)	0.137(5)
C-1B′	0.3383(4)	0.3376(3)	0.1058(5)	0.051(4)
C-2B′	0.3596(4)	0.2726(3)	0.0734(6)	0.035(3)
C-3B′	0.4673(4)	0.2660(3)	0.0999(6)	0.033(3)
C-4B′	0.5011(4)	0.3150(3)	0.0332(5)	0.051(4)
C-5B′	0.4704(4)	0.3788(3)	0.0586(6)	0.045(4)
C-6B′	0.4845(4)	0.4290(3)	−0.0227(6)	0.052(4)
N-1′	0.2380(3)	0.3452(2)	0.0807(5)	0.037(3)
O-2B′	0.3389(3)	0.2302(2)	0.1544(4)	0.054(3)
O-3B′	0.4833(3)	0.2076(2)	0.0519(5)	0.060(3)
O-4B′	0.6072(3)	0.3106(2)	0.0840(4)	0.048(2)
O-5B′	0.3669(3)	0.3783(2)	0.0277(4)	0.036(2)
O-6B′	0.5841(3)	0.4513(2)	0.0410(4)	0.045(2)
CA-2B′	0.2786(6)	0.1832(4)	0.1015(8)	0.102(7)
CA-3B′	0.5522(6)	0.1696(4)	0.1335(9)	0.150(1)
CA-4B′	0.6531(5)	0.3115(3)	0.0032(6)	0.066(5)
CA-6B′	0.6189(6)	0.4871(4)	−0.0268(8)	0.109(7)
CM-2B′	0.2655(7)	0.1435(4)	0.199(1)	0.043(3)
CM-3B′	0.5625(7)	0.1147(4)	0.066(1)	0.069(5)
CM-4B′	0.7587(5)	0.3027(3)	0.0708(7)	0.078(5)
CM-6B′	0.7180(6)	0.5105(4)	0.0497(9)	0.051(4)
OA-2B′	0.2401(5)	0.1750(3)	−0.0108(6)	0.145(6)
OA-3B′	0.5976(5)	0.1816(3)	0.2419(6)	0.158(6)
OA-4B′	0.6084(3)	0.3188(3)	−0.1075(5)	0.075(3)
OA-6B′	0.5720(4)	0.4981(3)	−0.1345(5)	0.065(4)

mean C-5–O-5 bond lengths are 1.42 and 1.43 Å for the α- and β-Glc rings which agrees with tabulated values. However, the mean O-5–C-1 bond lengths, 1.45 Å for the α-Glc and 1.43 Å for the β-Glc are slightly longer that average. The mean C–C–C ring bond angle value is 110.7° which agrees with the carbohydrate mean values whereas the C–C–O ring bond angle value of 107.6° is significantly shorter than the average value of 110.3°. The Cremer–Pople puckering parameters [11] are ($Q = 0.575$, $\Theta = 8.71$, $\phi = 354.3$) and ($Q' = 0.583$, $\Theta' = 1.90$, $\phi' = 245.2$) for the two α-Glc rings and ($Q = 0.588$, $\Theta = 11.2$, $\phi = 308.5$) and ($Q' = 0.583$, $\Theta' = 7.5$, $\phi' = 36.4$) for the two β-Glc rings. These values are in complete accordance with an $^4C_1$ conformation of the rings.

Conformation of the acetate groups.—The average bond lengths and angles of the acetate groups are in good agreement with the values observed for other acetylated carbohydrate derivatives [12]. As with other carbohydrate acetates, the secondary acetate groups are arranged in such a way that the carbonyl nearly eclipses the axial hydrogen at the corresponding ring carbon atom. The only exception is the secondary acetate at C-2 of the α–glucose. In both independent molecules, the H-2–C-2–O-2–CA-2 torsion angles adopts a value close to 50°. This deviation from the *cis* orientation could be due to steric hindrance with the acetate group at C-2 if the β-ring. The conformation of the primary acetate groups can be described by two contiguous torsion angles: O-5–C-5–C-6–OA-6, and C-5–C-6–OA-6–CA-6. For the α-residue of each disaccharide, the torsion angles orientations are found to be (−69.6°, −125.1°) and (−60.2°, −152.2°). The first value of each couple corresponds to a

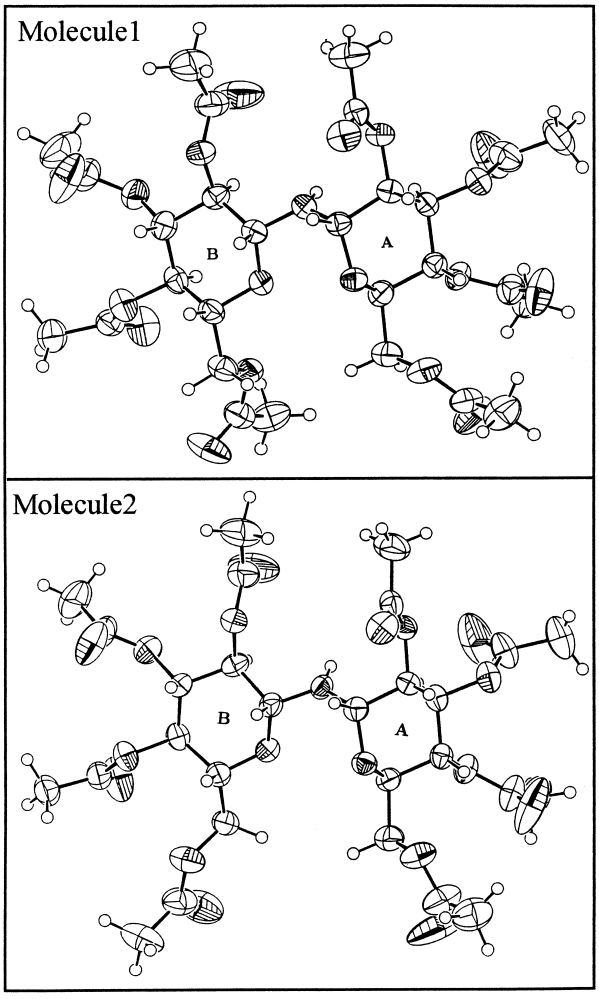

Fig. 2. ORTEP views of the two independent molecules of (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)amine. Thermal ellipsoids are drawn at the 50% probability level.

gauche–gauche arrangement of the primary acetate. As for the β-residue, the orientations are (−62.0°, 122.6°) and (154.0°, 166.6°) corresponding to a *gauche–gauche* and *trans–gauche* arrangement, respectively. Whereas the first three orientations fall within the range of usually observed conformations the last one is somehow unusual [13] as it involves a 1,3 diaxial interactions between the acetate groups at position C-4' and C-6'. Nevertheless, such departure from the lowest energy conformations have already been observed in other per acetylated carbohydrate structures [12]. The different orientations of these primary acetate groups are the largest conformational differences between the two independent molecules in the asymmetric unit.

Geometry and molecular conformation at the glycosidic linkage.—The two Glc residues are linked 1–1 which corresponds to an α,β-D-anomeric configuration as indicated by the values of the torsion angles C-5A–O-5A–C-1A–N-1 and C-5B–O-5B–C-1B–N-1 of (59.4°, 175.4°) and (64.1°, 172.7°), respectively. The hydrogen atom of the NH group is well located and no indication of a "umbrella" effect is observed. To describe the position of the hydrogen atom, we could assess that the nitrogen has the *R* chirality if the β-Glc is given priority (due to the *R* configuration of C-1B). In the two independent molecules the anomeric hydrogen atoms are transoid with respect to the hydrogen covalently linked to N-1 at the glycosidic linkage; H-1A–C-1A–N-1–H-N1 and H-1B–C-1B–N-1–HN-1 are respectively (−156.6°, −159.3°) and (143.0°, −173.8°) in the two independent molecules. This fact, together with the occurrence of a N-H...O=C hydrogen bond could explain that the hydrogen atom carried by the nitrogen group is well located instead of being disordered by the so-called "umbrella effect".

The mean value of the C-1A–N-1 bond is 1.430 Å that is significantly larger that the mean value of the C-1B–N-1 bond (1.406 Å). The mean value of the valence angle C-1A–N-1–C-1B is 119.5°. Both Φ torsion angles at the glycosidic linkage adopt a gauche conformation (57.6° and 59.8°) for the axial bond and (−78.8° and −85.9°) for the equatorial bond. The comparison between NH-linkage and O-linkage will be discussed below, together with the influence of the *exo*-anomeric effect.

Intermolecular interaction and molecular packing.—Two hydrogen bonds are observed in the crystal structure, each one involving the NH of one of the two independent molecules and the O=C of the acetate group at C-2 of the α-ring of the other molecule. The N...O distances are 2.98 and 2.90 Å, respectively. These distances are longer than the optimum value for O...O hydrogen bond (2.75 Å) but it has been reported that N–H are not as good donor as O–H groups [14]. The H...O distances are 1.92 and 1.89 Å (after correcting the N–H distance to 1.07 Å). This is in agreement with the mean value of 1.95 Å observed in nucleotide and nucleoside [14]. The N–H...O valence angles are 168.7 and 155.4°, respectively. As indicated below, these two hydrogen bonds have a strong influence on the packing arrangement.

The mode of molecular packing has been analyzed by applying the atom-pair procedure of

Table 3
Bond lengths for (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)amine

Bond	Molecule 1	Molecule 2	Bond	Molecule 1	Molecule 2
C-1A–C-2A	1.525(9)	1.524(9)	C-1B–C-2B	1.524(9)	1.533(9)
C-2A–C-3A	1.510(8)	1.502(8)	C-2B–C-3B	1.514(9)	1.509(9)
C-3A–C-4A	1.535(9)	1.518(9)	C-3B–C-4B	1.504(9)	1.516(9)
C-4A–C-5A	1.512(9)	1.507(9)	C-4B–C-5B	1.525(9)	1.530(9)
C-5A–C-6A	1.528(10)	1.520(10)	C-5B–C-6B	1.498(9)	1.511(9)
N-1–C-1A	1.426(8)	1.433(8)	N-1–C-1B	1.404(8)	1.408(8)
O-2A–C-2A	1.455(8)	1.458(8)	O-2B–C-2B	1.447(8)	1.434(8)
O-2A–CA-2A	1.339(7)	1.343(8)	O-2B–CA-2B	1.332(9)	1.337(10)
O-3A–C-3A	1.451(8)	1.451(8)	O-3B–C-3B	1.441(8)	1.447(8)
O-3A–CA-3A	1.331(9)	1.341(9)	O-3B–CA-3B	1.354(10)	1.359(11)
O-4A–C-4A	1.438(8)	1.437(7)	O-4B–C-4B	1.449(8)	1.446(8)
O-4A–CA-4A	1.343(9)	1.343(8)	O-4B–CA-4B	1.356(8)	1.358(9)
O-5A–C-1A	1.435(8)	1.432(8)	O-5B–C-1B	1.447(8)	1.446(8)
O-5A–C-5A	1.425(8)	1.419(8)	O-5B– C-5B	1.434(8)	1.431(8)
O-6A–C-6A	1.424(9)	1.456(9)	O-6B–C-6B	1.440(8)	1.446(8)
O-6A–CA-6A	1.345(8)	1.309(10)	O-6B–CA-6B	1.351(9)	1.347(10)
CA-2A–CM-2A	1.475(11)	1.503(11)	CA-2B–CM-2B	1.464(10)	1.493(11)
CA-3A–CM-3A	1.479(12)	1.479(13)	CA-3B–CM-3B	1.490(12)	1.471(12)
CA-4A–CM-4A	1.514(11)	1.504(10)	CA-4B–CM-4B	1.478(11)	1.456(11)
CA-6A–CM-6A	1.466(13)	1.512(14)	CA-6B–CM-6B	1.468(11)	1.470(13)
OA-2A–CA-2A	1.191(8)	1.194(8)	OA-2B–CA-2B	1.196(11)	1.200(11)
OA-3A–CA-3A	1.189(10)	1.201(11)	OA-3B–CA-3B	1.183(12)	1.187(11)
OA-4A–CA-4A	1.189(10)	1.187(10)	OA-4B–CA-4B	1.205(8)	1.189(8)
OA-6A–CA-6A	1.191(11)	1.175(14)	OA-6B–CA-6B	1.191(10)	1.179(10)

Kitaigorodsky [15]. For a reference molecule, all the contacts with surrounding molecules are explored. In addition to counting intermolecular contacts within 1.5 times the sum of atomic van der Waals radii, the energy of interaction was also calculated using a 6-exp potential with the coefficients proposed by Fillipini and Gavezzoti [16]. For each of the two independent molecules, the ten neighbors with strongest interactions are listed in Table 6. The strongest interaction occurs between the two independent molecule. Since the molecule2 is almost equivalent to molecule1 with a translation of $c/2$, this interaction is almost reproduced along c. This gives rise to a chain of molecules, involving also the occurrence of the two hydrogen bonds previously described. Two different views of the packing arrangement are displayed in Fig. 3.

4. Discussion

Characteristics of the N-*glycosyl linkage*.—The present work provides the first experimental description of the geometry of *N*-glycosyl bond. It can readily be compared to the *O*-glycosyl bond in disaccharide exhibiting similar type of linkage in a α,β-D-anomeric configuration, such as in α,β-trehalose [17]. The geometrical data of interest are summarized in Table 7; they illustrate the significant perturbation that occurs at the glycosidic center upon substitution of O by N–H. The glycosidic C-1A–N-1–C-1B bond angles are 119.9° and 118.9°, i.e., 6° larger than that of α,β-trehalose. This opening of the valence angle could be due in part to steric effect from the acetate pendent group since, from ab initio calculations, the theoretical value for a C–N–C angle at C-1 is 115.3° [18]. A lengthening of the O-5–C-1 bond is also observed. This phenomenon was predicted from ab initio calculations and was attributed to a stabilizing delocalization of the nitrogen lone pair into an antibonding C–O orbital ($n_N \rightarrow \sigma_{C-O}^*$) [18]. For the β-linkage in the observed orientation, the nitrogen lone pair is *ap* to the C-1–O-5 bond, allowing particularly efficient delocalization which is reflected in the elongation of the C–O bond and the shortening of the C–N bond.

Anomeric and exo-*anomeric effect*.—The anomeric effect was introduced after the empirical observation that a polar exocyclic group would prefer the axial over the equatorial position at the anomeric carbon of a pyranose ring [19–20]. The reverse anomeric effect describes the equatorial preference for quaternary nitrogen at the same

Table 4
Bond angles for (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)amine

Bond angle	Molecule 1	Molecule 2	Bond angle	Molecule 1	Molecule 2
C-2A–O-2A–CA-2A	115.6(4)	115.6(5)	C-2B–O-2B–CA-2B	119.7(5)	118.6(5)
C-3A–O-3A–CA-3A	118.6(5)	116.9(5)	C-3B–O-3B–CA-3B	118.1(6)	117.8(6)
C-4A–O-4A–CA-4A	118.1(5)	117.7(5)	C-4B–O-4B–CA-4B	117.4(5)	119.1(5)
C-1A–O-5A–C-5A	112.4(4)	115.7(5)	C-1B–O-5B–C-5B	112.9(5)	113.9(5)
C-6A–O-6A–CA-6A	117.9(6)	117.2(6)	C-6B–O-6B–CA-6B	115.3(5)	117.0(5)
O-5A–C-1A–N-1	115.6(5)	115.2(5)	O-5B–C-1B–N-1	110.9(5)	110.5(5)
O-5A–C-1A–C-2A	107.3(5)	107.3(5)	O-5B–C-1B–C-2B	108.5(5)	106.0(5)
C-2A–C-1A–N-1	111.2(5)	110.8(5)	N-2B–C-1B–N-1	111.7(5)	111.3(5)
O-2A–C-2A–C-1A	112.6(5)	112.5(5)	O-2B–C-2B–C-1B	107.2(5)	108.7(5)
O-2A–C-2A–C-3A	104.9(4)	105.0(5)	O-2B–C-2B–C-3B	107.8(5)	107.1(5)
C-3A–C-2A–C-1A	112.5(5)	111.1(5)	C-1B–C-2B–C-3B	112.9(5)	110.2(5)
O-3A–C-3A–C-2A	106.0(5)	109.4(5)	O-3B–C-3B–C-2B	108.9(5)	109.3(5)
O-3A–C-3A–C-4A	108.0(5)	107.4(5)	O-3B–C-3B–C-4B	108.3(5)	106.8(5)
C-2A–C-3A–C-4A	111.0(5)	109.4(5)	C-2B–C-3B–C-4B	111.6(5)	111.7(5)
O-4A–C-4A–C-3A	108.4(5)	107.6(5)	O-4B–C-4B–C-3B	105.5(5)	106.0(5)
O-4A–C-4A–C-5A	106.7(5)	109.9(5)	O-4B–C-4B–C-5B	110.7(5)	110.0(5)
C-3A–C-4A–C-5A	110.4(5)	108.9(5)	C-3B–C-4B–C-5B	108.7(5)	111.1(5)
O-5A–C-5A–C-4A	108.8(5)	108.6(5)	O-5B–C-5B–C-4B	106.2(5)	108.5(5)
O-5A–C-5A–C-6A	106.9(5)	107.9(5)	O-5B–C-5B–C-6B	109.8(5)	104.7(5)
C-4A–C-5A–C-6A	112.1(6)	113.1(6)	C-4B–C-5B–C-6B	116.0(5)	114.5(5)
O-6A–C-6A–C-5A	110.5(6)	107.3(6)	O-6B–C-6B–C-5B	110.9(5)	107.3(5)
O-2A–CA-2A–OA-2A	123.6(6)	123.1(6)	O-2B–CA-2B–OA-2B	122.1(7)	123.2(8)
O-2A–CA-2A–CM-2A	111.1(6)	110.9(6)	O-2B–CA-2B–CM-2B	111.5(7)	111.6(6)
OA-2A–CA-2A–CM-2A	125.3(6)	126.0(6)	OA-2B–CA-2B–CM-2B	126.4(8)	125.2(8)
O-3A–CA-3A–O-A3A	122.6(7)	123.3(8)	O-3B–CA-3B–OA-3B	123.8(8)	123.0(8)
O-3A–CA-3A–CM-3A	112.0(7)	112.1(7)	O-3B–CA-3B–CM-3B	109.4(7)	109.9(7)
OA-3A–CA-3A–CM-3A	125.1(8)	124.6(8)	OA-3B–CA-3B–CM-3B	126.7(8)	127.0(8)
O-4A–CA-4A–OA-4A	124.5(7)	122.4(7)	O-4B–CA-4B–OA-4B	122.5(6)	121.4(7)
O-4A–CA-4A–CM-4A	111.3(6)	111.0(6)	O-4B–CA-4B–CM-4B	111.5(6)	111.1(6)
OA-4A–CA-4A–CM-4A	124.2(7)	126.4(7)	OA-4B–CA-4B–CM-4B	126.0(7)	127.5(7)
O-6A–CA-6A–OA-6A	123.6(7)	122.5(9)	O-6B–CA-6B–OA-6B	122.6(7)	121.9(8)
O-6A–CA-6A–CM-6A	111.5(7)	114.0(8)	O-6B–CA-6B–CM-6B	112.5(7)	112.2(7)
OA-6A–CA-6A–CM-6A	124.8(7)	123.4(8)	OA-6B–CA-6B–CM-6B	124.9(8)	125.9(9)
C-1B–N-1–C-1A	119.9(5)	118.9(5)			

position [22]. Recent ab-initio calculations performed on different 2-substituted derivatives of tetrahydropyran [18] established that the magnitude of the anomeric effect is slightly negative (EA = −0.4 kcal/mol) for the $NHCH_3$ substituent. This value has been compared to the anomeric effect of the OCH_3 substituent (EA = 2.4 kcal/mol) or reverse anomeric effect of the CH_2CH_3 one (EA = −1.5 kcal/mol). Therefore the NH bond is predicted to have a slight preference for the equatorial form.

The *exo*-anomeric effect has been defined by analoguey with the anomeric effect to describe the orientational preference of the aglycon around the glycosidic C–O bond [23]. Theoretical calculations demonstrated a clear preference for the *sc* orientation for both axial and equatorial linkage for the 2-methylamino substituted tetrahydropyran [18]. The *exo*-anomeric effect in this compound was evaluated to be only slightly lower than that of methoxy derivative. These conclusions are in agreement with the present crystal structure where both the axial and equatorial glycosidic linkages adopt Φ angle consistent with the *exo*-anomeric effect.

Conformational analysis of the N-*glycosidic linkage*.—The α,β-trehalose energy map, computed as a function of Φα (O-5A–C-1A–O-1–C-1B) and Φβ (O-5B–C-1B–O-1–C-1A) with the Tripos forcefield is represented in Fig. 4. Even though a simple energy function has been used, together with a rigid residue approach, this map exhibits the same characteristics as the ones published previously with the relaxed residue approach, using the MM3 program [24]. They are also very similar to the energy map of a model compound, α, β–2-(tetrahydropyran-2-yloxy)tetrahydropyran, as calculated using the semi-empirical PCILO method with the inclusion of solvent effect [25]. The main low energy

Table 5
Selected torsion angles for (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)amine

Torsion angle	Molecule 1	Molecule 2	Torsion angle	Molecule 1	Molecule 2
CA-2B–O-2B–C-2B–C-1B	120.0(6)	127.2(7)	CA-2A–O-2A–C-2A–C-1A	68.9(6)	73.6(6)
C-2B–O-2B–CA-2B–OA-2B	−5.9(12)	−0.7(12)	C-2A–O-2A–CA-2A–OA-2A	0.4(9)	−0.9(9)
CA-3B–O-3B–C-3B–C-2B	−115.7(7)	−125.6(7)	CA-3A–O-3A–C-3A–C-2A	−137.7(6)	−129.3(6)
C-3B–O-3B–CA-3B–OA-3B	6.0(13)	1.8(13)	C-3A–O-3A–CA-3A–OA-3A	−3.4(12)	6.6(11)
CA-4B–O-4B–C-4B–C-3B	124.6(5)	134.0(5)	CA-4A–O-4A–C-4A–C-3A	105.1(6)	118.1(6)
C-4B–O-4B–CA-4B–OA-4B	13.1(9)	3.7(9)	C-4A–O-4A–CA-4A–OA-4A	−1.7(10)	2.2(10)
C-1B–O-5B–C-5B–C-4B	69.0(6)	63.7(6)	C-1A–O-5A–C-5A–C-4A	67.2(6)	62.5(7)
C-1B–O-5B–C-5B–C-6B	−164.9(5)	−173.6(5)	C-1A–O-5A–C-5A–C-6A	−171.5(5)	−174.5(5)
C-5B–O-5B–C-1B–C-2B	−61.6(6)	−66.5(6)	C-5A–O-5A–C-1A–C-2A	−65.2(6)	−59.8(6)
C-5B–O-5B–C-1B–N-1	175.4(5)	172.7(5)	C-5A–O-5A–C-1A–N-1	59.4(7)	64.1(7)
CA-6B–O-6B–C-6B–C-5B	−149.8(6)	166.6(6)	CA-6A–O-6A–C-6A–C-5A	−125.1(7)	−152.2(7)
C-6B–O-6B–CA-6B–OA-6B	2.4(10)	−2.7(11)	C-6A–O-6A–CA-6A–OA-6A	−2.0(13)	−3.9(15)
O-5B–C-1B–C-2B–C-3B	49.3(6)	59.1(6)	O-5A–C-1A–C-2A–C-3A	55.2(6)	55.3(6)
O-5B–C-1B–C-2B–O-2B	167.8(4)	176.1(5)	O-5A–C-1A–C-2A–O-2A	173.6(4)	172.7(4)
N-1–C-1B–C-2B–C-3B	171.8(5)	179.4(5)	N-1–C-1A–C-2A–C-3A	−72.0(7)	−71.3(7)
N-1–C-1B–C-2B–O-2B	−69.7(6)	−63.6(6)	N-1–C-1A–C-2A–O-2A	46.4(6)	46.1(6)
O-2B–C-2B–C-3B–C-4B	−166.5(5)	−171.8(5)	O-2A–C-2A–C-3A–C-4A	−171.8(5)	−178.4(5)
C-1B–C-2B–C-3B–C-4B	−48.4(7	−53.9(7)	C-1A–C-2A–C-3A–C-4A	−49.0(7)	−56.5(7)
C-1B–C-2B–C-3B–O-3B	−167.9(5)	−171.8(5)	C-1A–C-2A–C-3A–O-3A	−166.0(5)	−174.0(5)
C-2B–C-3B–C-4B–C-5B	54.4(7)	50.2(7)	C-2A–C-3A–C-4A–C-5A	49.0(7)	57.2(6)
C-2B–C-3B–C-4B–O-4B	173.1(5)	169.8(5)	C-2A–C-3A–C-4A–O-4A	165.5(5)	176.2(5)
C-3B–C-4B–C-5B–C-6B	174.7(5)	−169.4(5)	C-3A–C-4A–C-5A–C-6A	−174.7(5)	−178.0(5)
C-3B–C-4B–C-5B–O-5B	−62.9(6)	−52.8(6)	C-3A–C-4A–C-5A–O-5A	−56.7(6)	−58.3(6)
O-5B–C-5B–C-6B–O-6B	−62.0(7)	154.0(5)	O-5A–C-5A–C-6A–O-6A	−69.6(7)	−60.2(7)
C-4B–C-5B–C-6B–O-6B	58.4(7)	−87.2(6)	C-4A–C-5A–C-6A–O-6A	49.5(8)	60.0(7)
C-1B–N-1–C-1A–O-5A	57.6(7)	59.8(7)	C-1A–N-1–C-1B–O-5B	−78.8(6)	−85.9(6)
C-1B–N-1–C-1A–C-2A	−179.9(6)	−178.2(5)	C-1A–N-1–C-1B–C-2B	160.1(5)	156.5(5)

Table 6
Intermolecular packing interactions of (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl) amine and α, β–trehalose

Packing environment of molecule 1				Packing environment of molecule 2			
Molecule and symmetry	Contacts	E (kcal mol^{-1})	Hydrogen bonds	Molecule and symmetry	Contacts	E (kcal mol^{-1})	Hydrogen bonds
Mol 2 (I)	316	−21.9	N-1′–H-1′...OA-2A	Mol 1 (I)	316	−21.9	N-1′–H-1′...OA-2A
Mol 2 (I+*c*)	295	−23.2	N-1–H-1...OA-2A′	Mol 1 (I-*c*)	295	−23.2	N-1–H-1...OA-2A′
Mol 1 (II+*a*+*c*)	81	−6.9		Mol 2 (II+*a*)	88	−6.9	
Mol 1 (II+*a*−*b*+*c*)	81	−6.9		Mol 2 (II+*a*−*b*)	88	−6.9	
Mol 2 (I−*a*)	66	−4.7		Mol 1 (I+*a*)	66	−4.7	
Mol 1 (II+*c*)	58	−5.0		Mol 2 (II)	50	−4.6	
Mol 1 (II−*b*+*c*)	58	−5.0		Mol 2 (II−*b*)	50	−4.6	
Mol 1 (I+*a*)	53	−3.7		Mol 2 (I+*a*)	62	−4.3	
Mol 1 (I−*a*)	53	−3.7		Mol 2 (I−*a*)	62	−4.3	
Mol 2 (I+*a*+*c*)	49	−3.0		Mol 1 (I−*a*−*c*)	49	−3.0	

I = *x*,*y*,*z*; II = −*x*,1/2+*y*,−*z*.

region is centered around ($\Phi\alpha = 60°$, $\Phi\beta = -60°$) as dictated by the double *exo*-anomeric effect. The potential energy surface is more extended about the $\Phi\beta$ axis and secondary minima can exist for $\Phi\beta = 60°$ and $\Phi\beta = -150°$. In both MM3 and Tripos map, the conformation observed in the crystal structure of α,β-trehalose does not correspond closely to the calculated global minimum. This could be explained by the contribution of a water molecule, that stabilizes by hydrogen bonding the peculiar conformation observed in the crystal [17]. The potential energy surface computed for the *N*-analogue exhibits the same aspect than the one calculated for α,β-trehalose, except that the accessible area is larger (Fig. 4). This may result from the opening of glycosidic angle which results in

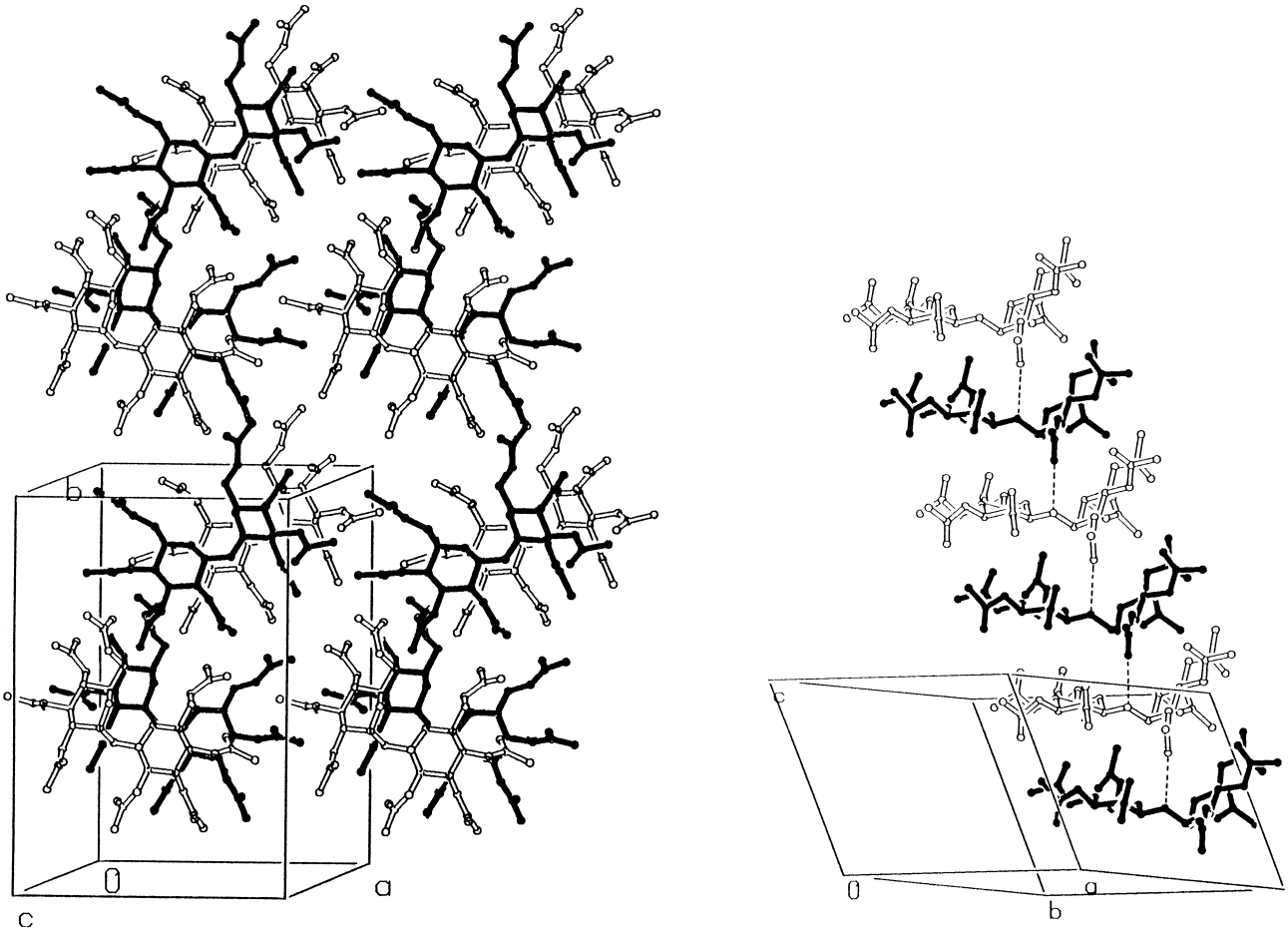

Fig. 3. Two different views of the packing arrangement of (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)amine. The two independent molecules have been given different colors. Hydrogen atoms have been omitted for clarity. Hydrogen bonds are represented by dotted lines.

Table 7
Comparison of the glycosidic linkage stereochemical features between (2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl) (2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)amine, α,β-trehalose and 2–methylamino-tetrahydropyran. Bond lengths and angles are indicated in Å and °, respectively (lengths in Å and angles in °)

	Molecule 1	Molecule 2	α, β-trehalose[a]	2-Methylamino-tetrahydropyran[b]
O-5–C-1α	1.435	1.432	1.410	1.414[c]
C-1α–N/O	1.426	1.433	1.414	1.434[c]
N/O–C-1β	1.404	1.408	1.401	1.425[d]
C-1β–O-5	1.447	1.446	1.419	1.413[d]
O-5–C-1α–N/O	115.55	115.18	112.63	114.7[c]
C-1α–N/O–C1-β	119.94	118.93	113.72	115.3[c,d]
N/O–C-1β–O-5	110.86	110.55	107.34	110.3[d]
C-5–O-5–C-1α–N/O	59.4	64.1	60.9	70.3[c]
O-5–C-1α–N/O–C-1β	57.6	59.8	68.8	54.8[c]
C-1α–N/O–C-1β–O-5	−78.8	−85.9	−93.4	−62.3[d]
N/O–C-1β–O-5–C-5	175.4	172.7	175.3	176.4[d]

[a] From the crystal structure [17].
[b] From ab initio (6-31G*) study [18].
[c] Axial (*S*-$NHCH_3$).
[d] Equatorial (*R*-$NHCH_3$).

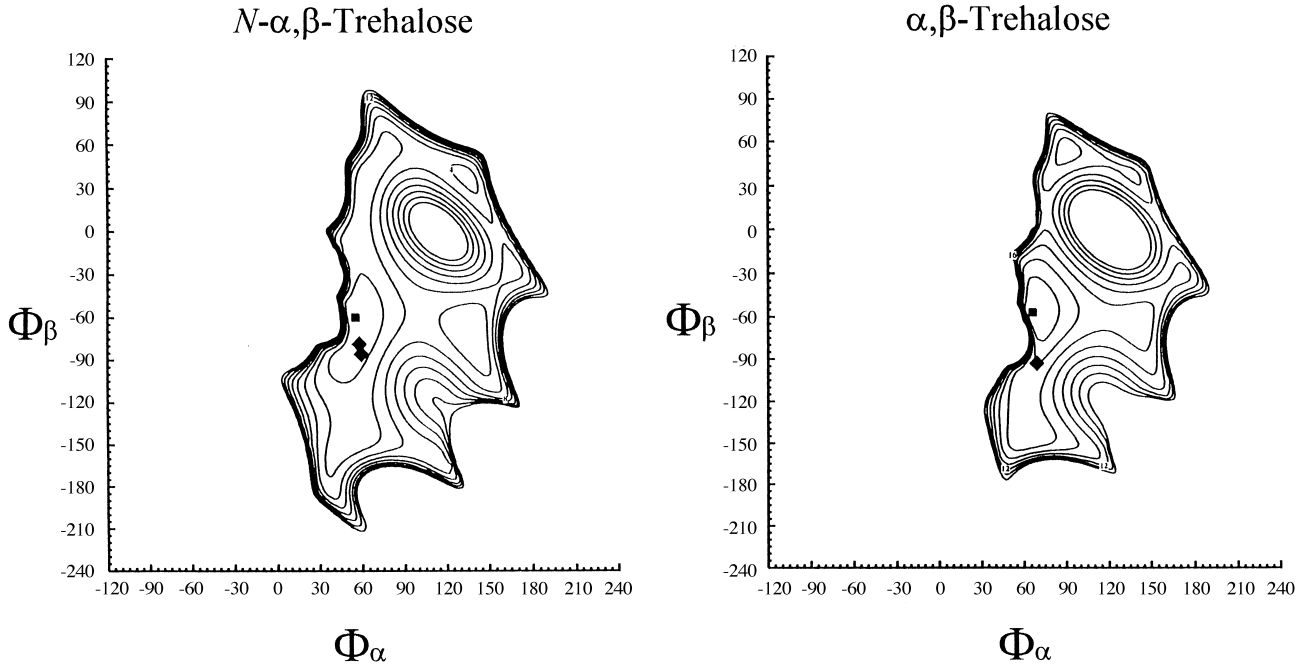

Fig. 4. Potential energy surface of *N*-α,β-trehalose and α,β-trehalose as a function of the Φα and Φβ torsion angles. Iso-energy contours are drawn with interpolation of 2 kcal mol^{-1}, up to the external contour at 16 kcal mol^{-1}. The crystallographic conformations are indicated by diamonds and the global minima by squares.

less steric conflict. The conformations observed in the crystal structure correspond closely to the global minimum.

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