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Functionalized cyclodextrins: Synthesis and structural characterization of 6-deoxy-6-{4-[N-tert-butoxycarbonyl-2-aminoethyl]-imidazolyl}-cyclomaltoheptaose

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Functionalized cyclodextrins: synthesis and structural characterization of 6-deoxy-6-{4-[N-tert-butoxycarbonyl-2-aminoethyl]-imidazolyl}-cyclomaltoheptaose

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The synthesis and the structural characterization by X-ray diffraction analysis of a monofunctionalized β-cyclodextrin (β-CD), the 6-deoxy-6-{4-[N-tert-butoxycarbonyl-2-aminoethyl]-imidazolyl}-cyclomaltoheptaose, are reported. It crystallizes in the orthorhombic space group $P2_12_12_1$, with a = 17.250(9)Å, b = 19.45(1)Å, c = 23.24(1)Å, d_{calc} = 1.304 g/cm^3 and Z = 4. The structure was refined to final indices R1 = 0.083 and wR2 = 0.24 (based on F_o^2) for the 4843 observed reflection with I ≥ 2σ(I). At the end of the refinement the presence of 12 water molecules per β-CD molecule distributed over 16 sites was detected. In the solid state the monofunctionalized β-CD molecule shows a 'sleeping swan'-like shape with the covalently bonded Boc-amino-ethyl-imidazolyl moiety forming a folded structure with its terminal part inserted inside the hydrophobic cavity of the β-CD ring. The β-CD macrocycle presents only small differences with respect to the conformation observed in hydrated uncomplexed or methylated β-CDs. The macrocycle structure maintains an approximate seven-fold symmetry. The substituted β-CD molecules pack in layers parallel to the bc plane. The layers are stacked in an head-to-tail arrangement of the monomeric units, with formation of columns of molecules along the a axis. The layers are connected to each other by H-bonds through water molecules. Channels generated in the crystal by the packing of the macrocycles are filled with water molecules.

Keywords: β-Cyclodextrins, monofunctionalized cyclodextrins, synthesis, solid state structure, conformation

INTRODUCTION

Cyclodextrins (CDs), also known as cyclomaltoheptaoses or cycloamyloses, are a family of macrocyclic oligosaccharides containing six, seven, eight or nine D-glucopyranosyl units (α-CD, β-CD, γ-CD, and δ-CD, respectively) in the $^4C_1$ chair conformation, linked by α-1,4 glycosidic bonds.[1–3] They have the shape of a truncated cone with the wide and narrow rims occupied by secondary and primary hydroxyl groups, respectively, and an hydrophobic cavity, the surface of which is dominated by H atoms and glycosidic O atoms. Because of the intramolecular cavity, CDs show remarkable ability to act as hosts for a wide variety of guest molecules or ions having appropriate dimensions to fit into the cavity.[4–5] For this reason, they have been used successfully to build up models and to study non-covalent intermolecular interactions and enzymes mechanisms. The inclusion and catalytic abilities of chemically modified CDs have been extensively studied both in solution and solid state.[6–7] Many structural data on inclusion complexes of CDs and various guest molecules have been reported, but only a few examples of X-ray structures of functionalized CDs have been inves-

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tigated. In particular, among functionalized compounds, differently methylated CDs have been prepared[8–9] in order to increase the hydrophobicity of their internal cavity. However, for the investigation of the factors contributing to the molecular inclusion mechanism and stabilization of host-guest complexes more efficient models can be obtained with CDs covalently bonded to larger functional groups. Recently, we have reported an example of structural characterization by X-ray diffraction analysis of 6-deoxy-6-cyclo(L-histidyl-L-leucyl)-β-CD (cHL-βCD),[10] a β-CD monosubstituted with a cyclic dipeptide. In this molecule, the cyclodipeptide moiety assumes a folded structure, with the terminal part, represented by the hydrophobic leucine side chain, directed inside the CD cavity. This study has proved the existence in the solid state of a monomeric inclusion compound formed by an intramolecular host (β-CD) and guest (peptide) interaction. In order to elucidate the molecular interactions of such inclusion compounds, structural characterization of modified CDs with functional groups of different sizes and polarities are being performed in our laboratories, both in solid state and in solution. In the present paper, we report the synthesis and the structural characterization by X-ray diffraction analysis of 6-deoxy-6-{4-[N-tert-butoxycarbonyl-2-aminoethyl]-imidazolyl}-β-CD (CDmhboc).

RESULTS AND DISCUSSION

The 6-deoxy-6-{4-[N-**tert**-butoxycarbonyl-2-aminoethyl] -imidazolyl}-β-CD (CDmhboc) is used as an intermediate product in the synthesis of the 6-deoxy-6-[4-(2-aminoethyl)imidazolyl]-cyclomaltoheptaose (CDmh). The copper(II) complexes of CDmh have been used as chiral eluent to resolve racemate mixtures of aromatic amino acids. The ligand CDmh has been previously isolated as by-product in low yield.[11] Here a new synthetic route to prepare CDmh in good yield is described.

The ^{1}H NMR spectra of CDmhboc (Figure 1) was assigned on the basis of COSY spectra. In addition to the groups of peaks which are typical of CD derivatives, other peaks are seen. The signals at 4.61 and at 4.17 ppm were assigned to the 6A diastereotopic protons which are shifted downfield because of the substitution of an OH group with the imidazole derivative, as reported in the case of 6-deoxy-6-(imidazolyl)-cyclomaltoheptaose (CDIm) and CDmh.[11] The signal at 3.51 ppm was assigned to the 4A proton shifted as a consequence of the functionalization as typically observed in other 6-derivatives.[12] The signals at 3.32 and at 2.96 ppm were assigned to the 6 protons of an adjacent ring that shows an upfield shift due to the ring current effect of imidazole ring placed almost perpendicularly to the cavity of the macrocycle, as found for other derivatives.[11,12] The signal at 3.25 ppm was assigned to one of the methylenic protons in α to the amino groups. The signal of other methylenic protons is in the 2, 4-H region (at about 3.6 ppm). These protons are diastereotopic. The signals at 2.90 ppm were assigned to the methylene chain in β to the NH group.

Conformation of the β-CD molecule. A perspective view of the molecular structure is represented in Figure 2. Bond distances, bond angles and torsion angles observed for the β-CD molecule and the covalently bonded functional group are unexceptional. A selection of torsion angles which define the linkage bonds between the glucose units and the orientation of the primary hydroxyl groups is given in Table 1a. The Cremer and Pople puckering parameters of glucose units, which with some degree of flexibility are found in the usual $^4C_1$ chair conformation, are listed in Table 2. The total puckering amplitude Q for all residues (0.54–0.56 Å) is slightly lower than the corresponding value found for an ideal cyclohexane chair conformation (0.63 Å), whereas the parameter which measures the magnitude of ring distortion is in the range 1.5°–4.7°. With the exception of the glucose units 2 and 6, all primary hydroxyl groups assume *gauche*$^+$- *gauche*$^-$ orientation (mean torsion angles C(4)-C(5)-C(6)-O(6) and O(5)-C(5)-C(6)-O(6), 53° and −69°, respectively). They point outside the macrocycle cavity. As for the primary hydroxyl groups of the glucose units 2 and 6, the C(6)-O(6) bond is trans to the C(4)-C(5) bond and *gauche*$^+$ to the O(5)-C(5) bond (mean torsion angles 173° and 69°, respectively); consequently, these two hydroxyl groups point inward the cavity.

The β-CD macrocycle presents only slight differences with respect to hydrated uncomplexed or methylated β-CDs.[13–15] The macrocycle structure maintains an approximate seven-fold symmetry. Geometrical data of

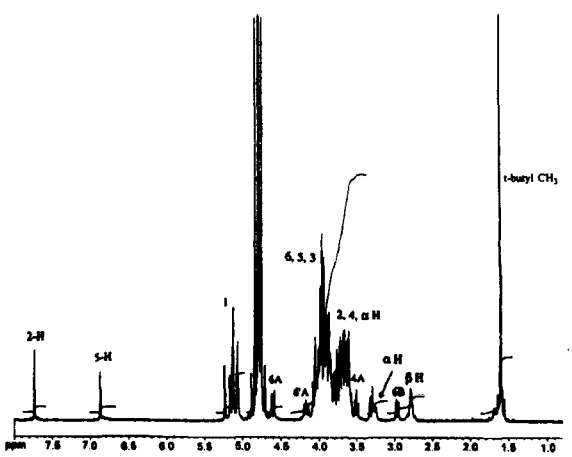


Figure 1 ^{1}H-NMR spectrum of CDmhboc at 400 MHz.

Table 1 Selected torsion angles(°) describing (a) the linkage bonds between the Glucose residues and the orientations of the primary hydroxyl; (b) the conformation of the Bocamino-ethyl-imidazolyl moiety.

a)	G1	G2	G3	G4	G5	G6	G7
C(3)n-C(4)n-O(4)n-C(1)n+1	124.2	111.6	134.8	130.4	130.1	114.8	135.9
C(5)n-C(4)n-O(4)n-C(1)n+1	−114.6	−127.9	−103.2	−106.0	−107.6	−124.9	−105.4
O(5)n-C(1)n-O(4)n-1-C(4)n−1	118.7	113.1	103.7	128.4	107.3	102.9	101.8
C(2)n-C(1)n-O(4)n-1-C(4)n−1	−120.6	−126.6	−135.7	−111.5	−128.5	−135.1	−136.5
C(4)n-C(5)n-C(6)n-O(6)n	—	−174.5	56.7	46.4	50.7	−170.6	58.9
O(5)n-C(5)n-C(6)n-O(6)n	—	67.1	−65.5	−76.6	−69.0	70.3	−63.7

b)	Angle°		Angle°
O(5)1-C(5)1-C(6)1-N3I	69.7	C5I-C3I-C2I-C1I	−5.4
C(4)1-C(5)1-C(6)1-N3I	−172.4	N2I-C3I-C2I-C1I	175.4
C(5)1-C(6)1-N3I-C4I	−92.2	C3I-C2I-C1I-N1I	−69.7
C(5)1-C(6)1-N3I-C5I	78.9	C1I-N1I-C'-OB	179.0
C(6)1-N3I-C5I-C3I	−173.1	N1I-C'-OB-C4B	168.7
C(6)1-N3I-C4I-N2I	173.1	C'-OB-C4B-C1B	−175.5
N3I-C5I-C3I-C2I	−179.0	C'-OB-C4B-C2B	65.0
N3I-C4I-N2I-C3I	−0.4	C'-OB-C4B-C3B	−59.5
C4I-N2I-C3I-C2I	179.4		

β-CD are listed in Table 3. The glucosydic O(4) atoms form a heptagon with radius and side length in the range 4.66–5.20 Å and 4.27–4.45 Å, respectively. The O(4) atoms are nearly coplanar: a maximum deviation from the least-squares plane of 0.20 Å is observed for the O(4) atoms of units 1 and 2. The dihedral angles between the O(4) plane and the least-squares plane through O(4)n+1, C(1)n, C(4)n, O(4)n lie in the range 0.7°–18.6°. The round shape of β-CD ring is stabilized by intramolecular H-bonds between the secondary hydroxyl groups of neighboring glucose residues: O(2)n-O(3)n−1 H-bonds lie in the usual range (2.76–2.90 Å) of hydrogen bond distances found in β-CDs, with the exception of the O(2)5-O(3)6 distance of 3.10 Å which is indicative of a slightly weaker hydrogen bond.

The host-guest interaction. The molecular model of the structure reveals a 'sleeping swan'-like shape with the covalently bonded Boc-amino-ethyl-imidazolyl moiety forming a folded structure with its terminal part inserted inside the hydrophobic cavity of the β-CD ring. This structure is quite similar in dimensions and conformation to the one we previously reported[10] for the β-CD monosubstituted with the cyclic dipeptide c(L-histidyl-L-leucyl), cHL-βCD (Figure 3). In spite of different sizes, polarities and flexibilities, the two functional groups assume the same type of folded conformation (with respect to β-CD macrocycle) indicating the existence of similar host-guest binding forces between these groups and the hydrophobic cavity. The conformation of the Boc-amino-ethyl-imidazolyl moiety is best described by the torsion angles given in Table 1b. The relative orientations of the imidazole ring and of the -CO-NH- group with respect to the plane of the seven glucosydic O(4) atoms (42° and 84°, respectively), indicate the presence of a bent conformation involving the C1-C2 and N1-C1 bonds; in fact, the observed values for the N1-C1-C2-C3 and C'-N1-C1-C2 torsion angles (−70°, 121°, respectively) are close to the ideal *gauche*−

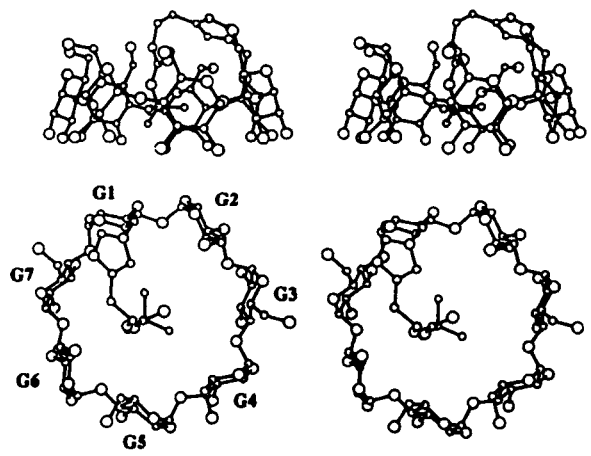


Figure 2 Stereo view of the of CDmhboc molecular model.

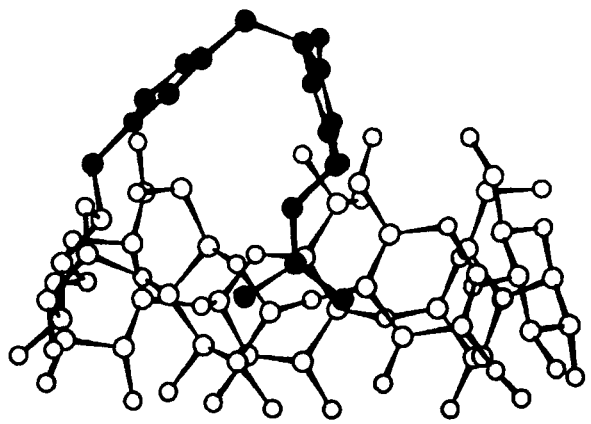


Figure 3 Molecular model of β-CD monosubstituted with the cyclic dipeptide c(L-histidyl-L-leucyl)[10].

Table 2 Puckering parameters of glucose units

	G1	G2	G3	G4	G5	G6	G7
Q(Å)	0.55	0.56	0.54	0.54	0.56	0.56	0.56
θ(°)	4.68	1.96	1.52	2.67	4.07	2.50	4.07

(−60°) and *skew+* (120°) conformations. Thus, the terminal Boc group is almost buried inside the β-CD macrocycle and a favorable intramolecular host-guest interaction occurs with the hydrophobic cavity. The baricentre of tert-butyl group is positioned just below the O(4) plane at a distance of 0.18 Å from it, whereas the C1B, C2B and C3B methyl groups lie in the wider part of the cavity at distances of 1.14, 0.40 and 0.24 Å, respectively. In addition to van der Waals interactions between the hydrophobic **tert**-butyl group and the β-CD cavity, the folded conformation of Boc-amino-ethyl-imidazolyl moiety is further stabilized by intramolecular H-bonds involving the amide N-H group and the primary hydroxylic group of the glucose unit 6. More stabilization of the molecular structure is obtained in the crystal by the presence of the Ow2 water molecule, which is H-bonded to the C = O carbonyl group of the urethane moiety and to the primary hydroxyl group of the glucose unit 2.

Molecular packing and water molecules. In Figure 4, the mode of packing of the functionalized β-CD molecules as viewed along the **a** direction is shown. The substituted β-CD molecules form layers which are parallel to the **bc** plane. The layers are stacked in an head-to-tail arrangement of the monomeric units, (the "head" and the "tail" being the O(6) and the O(2)-O(3) sides, respectively), forming columns of molecules along the **a** axis. The layers are connected to each other by H-bonds through water molecules. Because the seven-fold molecular axis of the β-CD macrocycle ring lies about 1.0 Å away from the crystallographic twofold screw axis in the **a** direction, slightly deformed channels similar to those found in dimeric β-CD channel structures, are formed[16–17]. In contrast with the herringbone packing mode occurring in monomeric β-CD complexes[18–20], the presence of layers in this structure could be related to the substitution of the monofunctionalized

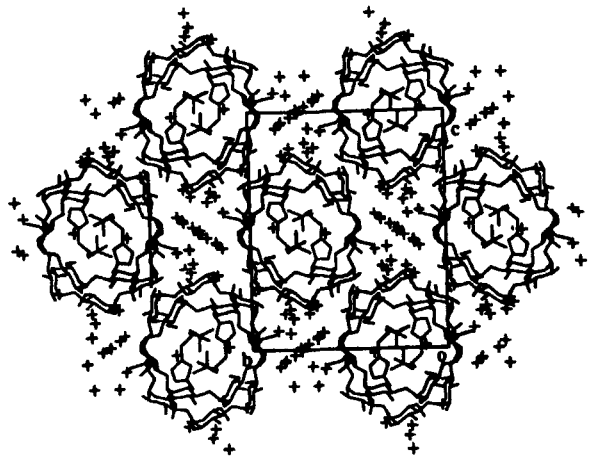


Figure 4 Crystal packing of the β-CDmhboc as viewed down the crystallographic *a* axis.

β-CD molecules, whose steric hindrance and orientation with respect to the macrocyclic cavity seems to play an important and decisive role. Within the layers the β-CD macrocycles are held together by a complicated intermolecular hydrogen bond network in which numerous water molecules and hydroxyl groups are involved (Table 4). No direct intermolecular H-bond involving hydroxyl groups exists in the crystal, except for the intermolecular hydrogen bond between the secondary hydroxyl O(3) of glucose unit 2 and the primary hydroxyl group O(6) of glucose unit 4 of a symmetry related β-CD molecule. The arrangement of β-CD molecules produces continuous hydrophilic channels filled with water molecules. Of the 12 water molecules, distributed over 16 molecular sites present in the crystal, the Ow2, located inside the cavity on the O(6) side of the β-CD macrocycle, links together two-fold symmetry related molecules in the **a** direction, whereas the remaining 15 water molecules are distributed along the hydrophilic channel. The Ow1, Ow3, Ow4, Ow5 and Ow10 water molecules are H-bonded to neighboring primary and secondary hydroxyl groups of symmetry related β-CD molecules. Other water molecules are involved in H-bonds with hydroxyl groups of the macrocycles within the layers or they interact among themselves.

Table 3 Geometrical Data

Residue	Radius(Å)[a]	Distance(Å)[b]	Angle (°)[c]	Tilt angle[d]	Planarity[e]
G1	4.89	4.29	126.0	16.6	−0.20
G2	5.04	4.32	128.3	16.5	0.19
G3	5.14	4.34	129.5	9.4	0.02
G4	5.19	4.45	129.3	13.5	−0.10
G5	5.20	4.27	126.4	18.6	−0.04
G6	4.94	4.39	127.7	18.4	0.16
G7	4.66	4.36	131.7	0.7	−0.02

[a]The radius is measured from the centre of gravity of the seven O(4) atoms to each O(4) atom [b]The distance is defined as the $O(4)_n$-$O(4)_{n+1}$ distance. [c]The angle is defined as the $O(4)_{n-1}$-$O(4)_n$-$O(4)_{n+1}$ angle. [d]The title angle is defined as the angle made by O(4) atoms plane and the plane formed by $O(4)_{n+1}$, $C(1)_n$, $C(4)_n$, $O(4)_n$ of each glucose residue. [e]Planarity is the defined as the O(4)n distance from the O(4) atoms plane.

Table 4 Inter- and Intramolecular H-bonds

A. O(2)n-O(3)n-1 distances

		Distance(Å)
O(2)1	O(3)7	2.81
O(2)2	O(3)1	2.76
O(2)3	O(3)2	2.78
O(2)4	O(3)3	2.79
O(2)5	O(3)4	2.90
O(2)6	O(3)5	3.09
O(2)7	O(3)6	2.83

B. Intramolecular and Intermolecular β-CD interactions

		Distance(Å)	Symmetry
N1I	O(6)6	2.97	x,y,z
N2I	O(3)3	2.65	x,y,z
O(6)4	O(3)2	2.82	−x+1,1/2+y,−z−1/2
O(3)4	O(3)6	2.82	−x+1,1/2+y,−z−1/2
O(3)4	O(2)6	2.95	−x+1,1/2+y,−z−1/2

C. β-CD and water molecules interactions

		Distance(Å)	Symmetry
O(6)2	Ow2	2.79	x,y,z
O(6)2	Ow4	2.67	x+1/2,1/2−y,−z
O(6)3	Ow10	2.64	x,y,z
O(6)3	Ow11	2.85	−x+1,1/2+y,1/2−z
O(6)4	Ow5	2.73	x,y,z
O(6)5	Ow1	2.72	x,y,z
O(6)5	Ow8	2.71	x,y,z
O(6)6	Ow1	2.79	x,y,z
O(6)7	Ow3	2.83	x,y,z
O(2)1	Ow1	2.72	−x+3/2,−y,1/2+z
O(3)1	Ow5	2.74	x−1/2,1/2−y,−z
O(2)2	Ow10	2.67	3/2−x,1−y,1/2+z
O(3)2	Ow1	2.80	x−1/2,1/2−y,−z
O(2)3	Ow6	2.72	x−1/2,1/2−y,−z
O(3)3	Ow3	2.79	x−1/2,1/2−y,−z
O(2)4	Ow13	2.87	−x+1,1/2+y,−z−1/2
O(2)5	Ow7	2.77	−x+1,1/2+y,−z−1/2
O(2)5	Ow9	2.89	x,y,z
O(3)5	Ow3	2.74	3/2−x,−y,z−1/2
O(3)5	Ow4	2.85	x,y,z
O(3)6	Ow9	3.02	−x+1,y−1/2,−z−1/2
O(2)7	Ow7	2.74	x,y,z
O(3)7	Ow2	2.77	x−1/2,1/2−y,−z
O	Ow2	2.79	x,y,z

D. Water-Water interactions

		Distance(Å)	Symmetry
Ow5	Ow6	2.84	2−x,1/2+y,−1/2−z
Ow5	Ow8	2.82	x,y,z
Ow6	Ow15	2.69	1−x,y−1/2,1/2−z
Ow7	Ow11	2.84	1/2−x,−y,−1/2+z
Ow7	Ow14	2.81	1/2+x,1/2−y,−z
Ow8	Ow10	2.83	2−x,1/2+y,−1/2+z
Ow8	Ow16	2.71	3/2−x,1−y,−1/2+z
Ow9	Ow13	2.73	x,y,z
Ow10	Ow12	3.11	x,y,z
Ow11	Ow15	2.80	x,y,z
Ow11	Ow16	2.47	−x+1,y−1/2,1/2−z
Ow12	Ow15	2.63	−x+1,y+1/2,−z+1/2

CONCLUSION

We have carried out the synthesis and the structural characterization by X-ray diffraction analysis of the monofunctionalized β-CD, 6-deoxy-6-{4-[N-**tert**-butoxycarbonyl-2-aminoethyl]-imidazolyl}-cyclomaltoheptaose, CDmhboc. In the solid state the macrocycle crystallizes together with 12 water molecules per unit cell distributed over 16 sites. The monofunctionalized β-CD molecule shows a 'sleeping swan'-like shape with the covalently bonded Boc-amino-ethyl-imidazolyl moiety forming a folded structure with its terminal part inserted inside the hydrophobic cavity of the β-CD ring. The encapsulation of the Boc group inside the CD cavity is achieved by the folded conformation assumed by the amino-ethyl-imidazolyl bridge. Van der Waals contacts between atoms of the Boc group and atoms of the cavity contribute to the stabilization of the self-assembled host-guest complex. This conformation is probably the result of the tendency to exclude the hydrophobic Boc group from the highly hydrophilic surrounding of a β-CD molecule. The β-CD macrocycle presents only slight differences with respect to uncomplexed hydrated or methylated β-CDs. In fact, the macrocyclic structure, although substituted, approximately maintains the characteristic seven-fold symmetry shown by β-CDs. The substituted β-CD molecules form layers which are parallel to the **bc** plane. The layers are stacked in an head-to-tail arrangement of the monomeric units, forming columns of molecules along the **a** axis. The layers are connected to each other by H-bonds through water molecules. Channels generated in the crystal by the packing of the macrocycles are filled with water molecules.

MATERIALS AND METHODS

β-CD was purchased from Sigma and anhydrous dimethylformamide (DMF) was purchased from Aldrich. T.l.c. was carried out on silica gel plates (Merck 60-F254). β-CD derivatives were detected with u.v. light and with anisaldehyde as a reagent. Merck Lichroprep RP-8 (40–63 μm) was used for reverse-phase column chromatography.

4-[N-**tert**-butoxycarbonyl-2-aminoethyl]-imidazole (hmboc) was synthesized from histamine with tert-butyl-pyrocarbonate in methanol as described in the literature[21]. ^{1}H NMR spectra were recorded in D_2O on a Bruker AC-400 spectrometer at 400.13 MHz, while ^{13}C NMR spectra were recorded on a Bruker AC-200 spectrometer at 50.33 MHz.

Synthesis of 6-deoxy-6-{4-[N-tert-butoxycarbonyl-2-aminoethyl]-imidazolyl}-cyclo maltoheptaose (CDmhboc).

To a solution of β-CD[22] (1.0 g) in DMF (30 mL) hmboc (1.0 g) was added. The solution was stirred at 85°C for 24h until all β-CD had reacted; the reaction was monitored by TLC (eluent PrOH/H_2O/AcOEt/NH_3 5:3:2:1,

Table 5 Crystal data and structure refinement parameters for 6-deoxy-6-{4-[N-tert-butoxycarbonyl-2-aminoethyl]-imidazolyl}-β-cyclodextrin

Crystal Data	
Empirical formula	$C_{52}H_{86}N_2O_{36}12H_2O$
Crystal size	0.3 × 0.5 × 0.5 mm
Crystal system	Orthorhombic
Space group	$P2_12_12_1$
Unit cell dimensions	a = 17.250(9) Å
	b = 19.45(1) Å
	c = 23.24(1) Å
Volume	7797(9) $Å^3$
Z	4
Molecular weight	1531.5 u.m.a
Density (calculated)	1.304 g/cm^3
Absorption coefficient	0.967 mm^{-1}
Data Collection Parameters	
Wavelength/Temperature	1.54178/298(2) K
Scan type	ω-2θ
Scan speed	variable depending upon intensity
Index ranges	$0 \le h \le 21$; $0 \le k \le 23$;
	$0 \le l \le 28$
Reflections collected	8103
Independent reflections	8066
Observed reflections	4843
[$I \ge 2\sigma(I)$]	
Refinement Parameters	
Refinement method	Full-matrix least-squares on F^2
Data/restraints/parameters	4843/0/960
Goodness of fit on F^2	1.052
Final R indices [$I \ge 2\sigma(I)$]	R1 = 0.085, wR2 = 0.240
R indices (all data)	R1 = 0.109, wR2 = 0.267
Largest diff. peak and hole	0.496 and −0.309 $eÅ^{-3}$

β-CD Rf = 0.34, CDmhboc Rf = 0.28). The solvent was evaporated under vacuum at 40°C and the remaining solid was passed through a reverse-phase Rp-8 column (35 × 500 mm) with a linear gradient of ethanol-water (0 → 30%). The appropriate fractions were combined and the final product CDmhboc was crystallized from water. Yield 30% (based on β-CD) ^{1}H NMR: δ 7.74 (s, 1H, 2-H of imidazole), 6.87 (s, 1H, 5-H of imidazole), 5.01–5.33 (m, 7H, H-1 of CD), 4.61 (d, 1H, H-6A, $J_{6A,6'A}$ 14 Hz), 4.17 (dd, 1H, H-6'A, $J_{5A,6'A}$ 9.6 Hz), 3.55–4.10 (m, 38H, 5-,6-,3-,2-,3-H of Cdand one of the H in α to the amino group), 3.51 (t, 1H, H-4A, $J_{4A,5A}$ 9.5 Hz), 3.32 (d, 1H, H-6B, $J_{6B,6'B}$ 12Hz), 3.25 (m, 1H, the other H in α to the amino group), 2.96 (d, 1H, H-6'B), 2.80 (m, 2H, H in β to the amino group), 1.60 (s, 9H, t-butyl CH_3's) ^{13}C NMR: δ 105–103 (C-1), 85.8 (C-4A), 85–82 (C-4), 76.0–74.0 (C-5,2,3), 63.5–60.0 (C-6), 40.1 (C in α to the amino group), 32.9 (C in β to the amino group), 31.3 (t-butyl CH_3's).

X-Ray Diffraction Analysis

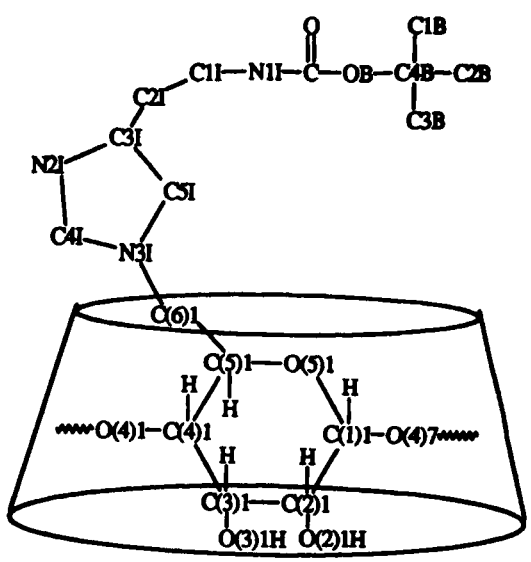
Colorless crystals of 6-deoxy-6-{4-[N-tert-butoxycarbonyl-2-aminoethyl]-imidazolyl}-cyclomaltoheptaose (Cdmhboc) in the form of regular parallelepiped suitable for X-ray diffraction, were obtained at room temperature by slow evaporation of an aqueous solution. A crystal of dimensions 0.3 × 0.5 × 0.5 mm was sealed in a Lindmann glass capillary with a drop of mother liquor to avoid water loss, which quickly leads to opaque crystals. Crystal system and space group were determined by preliminary oscillation and Weissenberg photographs. Cell dimensions were determined by a least-squares fit of the experimentally determined positions of 25 high angle reflections (2θ range 45–50°). Data were collected on a turbo-CAD4 Enraf-Nonius diffractometer with graphite-monochromatized CuKα radiation (λ = 1.54178Å) with an ω-2θ scan mode up to 140 in 2θ. The intensities of three reflections, used as standards, were remeasured every 60 min of X-ray exposure time and they showed an average random fluctuation of ±3%. Detailed crystallographic data are presented in Table 5. A total of 8103 independent reflections were measured and corrected for Lorentz and polarization factors. Of these only 4843 reflections were considered as "observed", having net intensity $I \ge 2.0\sigma(I)$. No absorption correction was applied. The space group symmetry and unit cell dimensions suggested the presence in the cell of one β-CD independent molecule and a number of water molecules. Many attempts to solve the structure by straightforward application of direct methods failed. The structure solution was obtained by the Patterson search program PATSEE[23] by using the atomic coordinates of two glycosidic units of one of the two molecules of the cHL-βCD dimer, recently determined[10], as input fragment for the rotation search. The best solution of this procedure was used to assign the phases to 500 E values and to partially expand the set by SIR-92[24]. The E-map revealed the position of all atoms of the molecule and some water molecules. Subsequent difference electron density maps, revealed the positions of the remaining water molecules. The structure was refined using the full-matrix least-squares program SHELXL-93[25]. The refinement carried out on F_0^2 values, as opposed to conventional refinement on F_0 values, allows the use of all data rather than just data with F_0 greater than a specified threshold. As a result the experimental information is more fully exploited, which for weakly scattering crystals can appreciably improve the precision of the structure determination. However, the R-factor, wR2, is for statistical reasons about twice as large as the conventional R-factor, R1, which is based on F_0 values (Table 5). Also, any R-factors based on all data will inevitably be larger than those based only on data greater then a given threshold. It should also be noted that wR2 is not related to wR or Rw since the weighting schemes for F_0^2 and F_0 refinements are quite different. The refinement was carried out on 960 parameters including atomic coordinates and anisotropic thermal factors for N, C and O atoms. All hydrogen atoms were included in their stereochemically expected positions with thermal

Table 6 Fractional atomic coordinates and equivalent isotropic thermal parameters for non hydrogen atoms

$(Å^2)$ Ueq = $(1/3)\Sigma_i\Sigma_j U_{ij} a_i^* a_j^* a_i a_j$

Atom	x	y	z	Ueq
C1B	0.6108(5)	0.1840(6)	−0.0891(5)	0.088(3)
C2B	0.6648(6)	0.3029(6)	−0.0900(5)	0.085(3)
C3B	0.6680(6)	0.2325(6)	0.0001(5)	0.088(3)
C4B	0.6715(4)	0.2328(5)	−0.0648(4)	0.061(2)
OB	0.7448(3)	0.1999(3)	−0.0821(3)	0.071(2)
C'	0.8141(5)	0.2240(5)	−0.0659(4)	0.069(2)
O	0.8241(4)	0.2810(4)	−0.0437(4)	0.086(2)
N1I	0.8707(4)	0.1802(4)	−0.0775(4)	0.071(2)
C1I	0.9511(5)	0.1935(5)	−0.0654(4)	0.072(2)
C2I	0.9846(4)	0.1379(5)	−0.0248(4)	0.068(2)
C3I	0.9538(4)	0.1411(4)	0.0358(4)	0.057(2)
N2I	0.9755(3)	0.0914(4)	0.0736(3)	0.065(2)
C4I	0.9397(5)	0.1087(4)	0.1230(4)	0.066(2)
N3I	0.8977(3)	0.1664(3)	0.1172(3)	0.056(2)
C5I	0.9065(4)	0.1874(4)	0.0605(4)	0.060(2)
C(1)1	0.6919(4)	0.0667(4)	0.1591(4)	0.058(2)
C(2)1	0.6282(5)	0.1008(4)	0.1945(4)	0.062(2)
O(2)1	0.5558(3)	0.0685(3)	0.1844(3)	0.071(2)
C(3)1	0.6237(4)	0.1766(4)	0.1833(4)	0.060(2)
O(3)1	0.5710(3)	0.2077(3)	0.2202(3)	0.083(2)
C(4)1	0.7040(4)	0.2084(4)	0.1885(3)	0.051(2)
O(4)1	0.6973(3)	0.2783(3)	0.1696(2)	0.054(1)
C(5)1	0.7636(4)	0.1716(4)	0.1507(3)	0.049(2)
O(5)1	0.7632(3)	0.0997(3)	0.1672(2)	0.056(1)
C(6)1	0.8456(4)	0.1962(5)	0.1590(4)	0.062(2)
C(1)2	0.7211(5)	0.3310(4)	0.2072(3)	0.059(2)
C(2)2	0.6546(5)	0.3811(4)	0.2132(4)	0.063(2)
O(2)2	0.5873(3)	0.3479(3)	0.2351(3)	0.073(2)
C(3)2	0.6376(4)	0.4130(4)	0.1557(4)	0.059(2)
O(3)2	0.5799(3)	0.4661(3)	0.1639(3)	0.078(2)
C(4)2	0.7106(4)	0.4449(4)	0.1319(3)	0.057(2)
O(4)2	0.6913(3)	0.4659(3)	0.0730(3)	0.061(1)
C(5)2	0.7758(4)	0.3952(4)	0.1282(3)	0.054(2)
O(5)2	0.7866(3)	0.3643(3)	0.1847(2)	0.059(1)
C(6)2	0.8541(5)	0.4301(5)	0.1130(4)	0.073(2)
O(6)2	0.9126(4)	0.3792(4)	0.1048(3)	0.097(2)
C(1)3	0.6878(5)	0.5377(4)	0.0609(4)	0.063(2)
C(2)3	0.6143(5)	0.5482(5)	0.0250(4)	0.068(2)
O(2)3	0.5486(3)	0.5254(4)	0.0576(3)	0.080(2)
C(3)3	0.6215(4)	0.5134(4)	−0.0307(4)	0.060(2)
O(3)3	0.5550(3)	0.5247(3)	−0.0674(3)	0.072(2)
C(4)3	0.6937(4)	0.5334(4)	−0.0615(4)	0.058(2)
O(4)3	0.7039(3)	0.4925(3)	−0.1115(3)	0.070(2)
C(5)3	0.7656(4)	0.5238(5)	−0.0230(4)	0.067(2)
O(5)3	0.7536(3)	0.5590(3)	0.0312(3)	0.071(2)
C(6)3	0.8398(5)	0.5499(7)	−0.0484(6)	0.094(4)
O(6)3	0.8350(5)	0.6199(5)	−0.0632(5)	0.121(3)
C(1)4	0.7254(5)	0.5261(5)	−0.1629(5)	0.082(3)
C(2)4	0.6584(6)	0.5217(6)	−0.2030(5)	0.085(3)
O(2)4	0.5936(4)	0.5569(4)	−0.1809(3)	0.092(2)
C(3)4	0.6410(6)	0.4473(6)	−0.2172(5)	0.082(3)
O(3)4	0.5840(5)	0.4418(5)	−0.2618(4)	0.114(3)
C(4)4	0.7145(5)	0.4093(5)	−0.2387(4)	0.074(3)
O(4)4	0.6973(4)	0.3392(4)	−0.2469(3)	0.076(2)
C(5)4	0.7800(5)	0.4196(5)	−0.1985(5)	0.075(3)
O(5)4	0.7920(4)	0.4920(4)	−0.1863(3)	0.085(2)
C(6)4	0.8580(6)	0.3929(6)	−0.2201(5)	0.093(3)
O(6)4	0.8730(5)	0.4127(4)	−0.2780(4)	0.105(3)
C(1)5	0.7179(6)	0.3061(6)	−0.2991(4)	0.082(3)
C(2)5	0.6513(8)	0.2719(8)	−0.3214(5)	0.105(4)
O(2)5	0.5865(6)	0.3166(5)	−0.3295(4)	0.125(3)
C(3)5	0.6245(6)	0.2081(6)	−0.2819(5)	0.085(3)
O(3)5	0.5631(5)	0.1726(6)	−0.3067(5)	0.139(4)
C(4)5	0.6969(5)	0.1653(6)	−0.2726(4)	0.077(3)
O(4)5	0.6737(3)	0.1123(3)	−0.2328(3)	0.071(2)
C(5)5	0.7637(5)	0.2065(5)	−0.2490(5)	0.076(2)
O(5)5	0.7808(4)	0.2615(4)	−0.2897(3)	0.086(2)
C(6)5	0.8371(6)	0.1690(6)	−0.2409(6)	0.093(3)
O(6)5	0.8597(4)	0.1320(5)	−0.2911(4)	0.109(3)
C(1)6	0.6913(6)	0.0434(5)	−0.2461(4)	0.075(3)
C(2)6	0.6184(6)	−0.0010(6)	−0.2339(5)	0.083(3)
O(2)6	0.5566(5)	0.0203(5)	−0.2679(4)	0.117(3)
C(3)6	0.6016(5)	0.0011(6)	−0.1711(4)	0.080(3)
O(3)6	0.5382(4)	−0.0447(5)	−0.1581(4)	0.111(3)
C(4)6	0.6710(5)	−0.0189(5)	−0.1374(4)	0.076(3)
O(4)6	0.6516(4)	−0.0041(3)	−0.0775(3)	0.071(2)
C(5)6	0.7419(4)	0.0238(5)	−0.1526(4)	0.064(2)
O(5)6	0.7536(3)	0.0203(3)	−0.2148(3)	0.074(2)
C(6)6	0.8156(5)	−0.0014(5)	−0.1270(5)	0.081(3)
O(6)6	0.8788(3)	0.0429(4)	−0.1332(3)	0.081(2)
C(1)7	0.6461(5)	−0.0579(5)	−0.0397(5)	0.074(2)
C(2)7	0.5749(5)	−0.0480(6)	−0.0020(5)	0.082(3)
O(2)7	0.5064(4)	−0.0406(4)	−0.0387(3)	0.098(2)
C(3)7	0.5845(40	0.0161(5)	0.0361(4)	0.069(2)
O(3)7	0.5206(3)	0.0218(4)	0.0733(3)	0.084(2)
C(4)7	0.6601(4)	0.0080(4)	0.0699(4)	0.058(2)
O(4)7	0.6688(3)	0.0713(3)	0.0996(2)	0.058(1)
C(5)7	0.7279(4)	−0.0032(5)	0.0290(4)	0.065(2)
O(5)7	0.7143(3)	−0.0627(3)	−0.0051(3)	0.074(2)
C(6)7	0.8047(5)	−0.0112(6)	0.0585(5)	0.085(3)
O(6)7	0.8040(5)	−0.0658(6)	0.0959(4)	0.119(3)
OW1	0.9535(4)	0.0377(4)	−0.2398(3)	0.089(2)
OW2	0.9312(5)	0.3825(5)	−0.0145(4)	0.121(3)
OW3	0.9490(6)	−0.1317(6)	0.0808(5)	0.142(4)
OW4	0.4629(6)	0.1517(9)	−0.2105(5)	0.171(5)
OW5	0.9917(8)	0.3319(7)	−0.3167(5)	0.155(4)
OW6	0.9567(7)	−0.1383(7)	−0.0697(6)	0.154(4)
OW7	0.4334(9)	−0.1646(8)	−0.0530(6)	0.193(6)
OW8	0.9305(9)	0.2132(10)	−0.3698(7)	0.212(7)
OW9	0.4247(14)	0.3142(10)	−0.2965(8)	0.220(10)	[0.85]
OW10	0.9136(19)	0.6691(20)	−0.1514(6)	0.280(24)	[0.50]
OW11	0.2184(32)	0.2145(11)	0.4768(18)	0.311(29)	[0.50]
OW12	0.8040(43)	0.7937(24)	−0.1519(15)	0.341(33)	[0.50]
OW13	0.3523(17)	0.1976(14)	−0.03307(18)	0.255(15)	[0.65]
OW14	0.0447(29)	0.7535(15)	0.0070(23)	0.276(22)	[0.50]
OW15	0.1906(12)	0.3320(8)	0.5443(11)	0.104(7)	[0.39]
OW16*	0.7050(33)	0.8056(29)	0.0698(23)	0.190(19)	[0.28]

Starred atom was isotropically refined.
The occupancy factors are reported in square brackets.



Scheme 1

factors equal to the equivalent U of the carrier atom (C-H distance 0.96 Å), except those of the water molecules. The refinement of the occupancy factor of water molecules indicated the presence of 12 water molecules per β-CD molecule distributed over 16 sites, 8 of which were refined with full occupancy and 8 with a site occupancy factor varying from 0.28 to 0.85 because of their high disorder. Scattering factors were taken from International Tables for X-ray Crystallography[26]. All measurements and calculations were carried out at the Biocrystallography Research Centre of the CNR at the University of Naples. The structure was refined to final indices R1 = 0.083 and wR2 = 0.24. These somehow high values reflect the poor quality of the crystal and, in part, are due to the thermal disorder of the cocrystallized water molecules in the unit cell. The final positional parameters and equivalent isotropic thermal factors for the N, C and O atoms are given in Table 6. Bond lengths, bond angles and anisotropic thermal factors have been deposited with the Cambridge Crystallographic Data Bank as Supplementary Material. Selected torsion angles describing the conformation of the β-CD macrocycle and that of Boc-amino-ethyl-imidazolyl moiety, the Cremer and Pople puckering parameters of the glucose residues[27], and the geometrical data relative to the cyclodextrin ring are given in Tables 1–3 respectively. Hydrogen bond parameters are listed in Table 4. The atoms of the glucose residues are indicated as C(m)n or O(m)n where m denotes the m^{th} atom within the n^{th} glycosidic residue Gn. In Scheme 1 a cartoon rapresentation of the molecule is reported with the atom labelling of the first glycosidic residue and of the Boc-amino-ethyl-imidazolyl moiety.

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REFERENCES

1 Saenger, W. *Angew. Chem., Int. Ed. Engl.* **1980**, **19**, 344.
2 Szejtli, J. *Cyclodextrins and their Inclusion Complexes*; Akademiai Kiado: Budapest, 1982
3 Szejtli, J. *Cyclodextrins Technology*, Kluwer Academic Publ., Dordrecht, The Netherlands, 1988.
4 Jones, S.P.; Grant, J.W.; Hadgraft, J.; Parr, G.D. *Acta Pharm. Technol.* **1984**, **30**, 213.
5 Jones, S.P.; Grant, J.W.; Hadgraft, J.; Parr, G.D. *Acta Pharm. Technol.* **1984**, **30**, 263.
6 Tabushi, I. *Tetrahedron* **1984**, **40**, 269.
7 Breslow, R. *Science* **1982**, **218**, 532.
8 Harata, K.; Uekama, K.; Otagiri, M.; Hirayama, F. *Bull. Chem. Soc. Jpn.* **1983**, **56**, 1732.
9 Harata, K. *Chem. Lett.* **1984**, 1961.
10 Di Blasio, B.; Pavone, V.; Nastri, F.; Isernia, C.; Saviano, M.;Pedone, C.; Cucinotta, V.; Impellizzeri, G.; Rizzarelli, E. *Proc. Natl. Acad. Sci. USA* **1992**, **89**, 7218.
11 Cucinotta, V.; D'Alessandro, F.; Impellizzeri, G.; Vecchio, G. *J. Chem. Soc., Chem. Commun.* **1992**, 1743.
12 Cucinotta, V.; D'Alessandro, F.; Impellizzeri, G.; Rizzarelli, E.; Vecchio, G. *J. Chem. Soc., Chem. Commun.* **1991**, 294.
13 Betzel, C.; Saenger, W.; Hingerty, B.E.; Brown, G.M. *J. Am. Chem. Soc.* **1984**, **106**, 7545.
14 Lindner, K.; Saenger, W. *Carbohydr. Res.* **1982**, **99**, 103.
15 Zabel, V.; Saenger, W.; Mason, S.A. *J. Am. Chem. Soc.* **1986**, **108**, 3663.
16 Le Bas, G. PhD Thesis, Univ. de Pierre et Marie Curie, Paris, France.
17 Mavridis, I.; Hadjoudis, E.; Tsoucaris, G. *Mol. Cryst. Liq. Cryst.* **1990**, **186**, 185.
18 Lindner, K.; Saenger, W. *Carbohydr. Res.* **1982**, **107**, 7.
19 Harata, K.; Kawano, K.; Fukunaga, K.; Ohtani, Y. *Chem. Pharm. Bull. Jpn.* **1983**, **31**, 1428.
20 Harata, K.; Uekama, K.; Otagiri, M.; Hirayama, F.; Ohtani, Y. *Bull. Chem. Soc. Jpn.* **1985**, **58**, 1234.
21 Tardel, D.S.; Yamoto, Y.; Pope B.M. *Proc. Nat. Acad. Sci. USA* **1972**, **69**, 730.
22 Bonomo, R.P.; Cucinotta, V.; D'Alessandro, F.; Impellizzeri, G.; Maccarrone, G.; Vecchio, G.; Rizzarelli, E. *Inorg. Chem.* **1991**, **30**, 2708.
23 Egert, E.; Sheldrick, G.M. *Acta Cryst.* **1985**, **A41**, 262.
24 Altomare, A.; Burla, M.C.; Camalli, M.; Cascarano, G.; Giacovazzo, C.; Gugliardi, A.; Polidori, G. *J. Applied Cryst.* **1994**, **27**, 635.
25 Sheldrick, G.M. *SHELXL93. Program for Crystal Structure Refinement.* Univ. of Göttingen, Germany, 1993
26 Cromer, D.T.; Waber, J.T. in *International Tables for X-Ray Crystallography*, vol. 4, Kynoch Press, Birmingham, United Kingdom, Table 2.2.B, 1974.
27 Cremer, D.; Pople, J.A. *J. Am. Chem. Soc.* **1975**, **97**, 1354.