Host–guest complexation of aromatic carboxylic acids and their conjugate bases by 6^A-(ω-aminoalkylamino)-6^A-deoxy-β-cyclodextrins† in aqueous solution

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A potentiometric titration study of the complexation of the guests benzoic acid, 4-methylbenzoic acid and (*RS*)-2-phenylpropanoic acid and their conjugate bases by host 6^A-(ω-aminoalkylamino)-6^A-deoxy-β-cyclodextrins, where the ω-aminoalkylamino groups are -$NH(CH_2)_nNH_2$ (n = 2, 3, 4 and 6), is reported. Of the 64 host–guest complexes whose formation is statistically possible over the pH range 2.0–12.0 studied, 35 were detected. Their stability constants range from 140 ± 35 $dm^3\ mol^{-1}$ for the [βCDNH$(CH_2)_2NH_3$·4-methylbenzoate] complex to 1760 ± 150 $dm^3\ mol^{-1}$ for the [βCDNH$(CH_2)_6NH_2$·(*S*)-2-phenylpropanoate]$^-$ complex at 298.2 K and I = 0.10 mol dm^{-3} ($NaClO_4$). The charge and hydrophobicity of both host and guest appear to be significant factors in the variation of host–guest complex stability. Qualitative structural information on the host–guest complexes obtained from 600 MHz ^{1}H NMR ROESY spectroscopy is generally consistent with the structures generated by molecular modelling.

Introduction

The range of cyclodextrins (CDs), their modified forms and the host–guest complexes formed by them is extensive.[1–16] In most cases, entry of a hydrophobic moiety of the guest into the hydrophobic region of the host CD annulus occurs during host–guest complexation, and the CD may discriminate between guests on the basis of their size, hydrophobicity, charge and chirality. We are particularly interested in the effects of variation of charge and hydrophobicity of both the host CD and the guest on complex stability. Our reported syntheses of 6^A-(ω-aminoalkylamino)-6^A-deoxy-βCDs (βCDNH$(CH_2)_n$-NH_2) (Fig. 1) and the determination of the pK_as of their protonated forms[17] facilitate a systematic study of the effects of the simultaneous variation of these factors in both host and guest on complex stability. We have studied the relative importance of these factors in βCDNH$(CH_2)_nNH_2$ (n = 2, 3, 4 and 6) where the hydrophobicity of the -$NH(CH_2)_nNH_2$ substituent increases with n, and a variation of charge occurs through amine protonation.[14] The guests selected for this study, benzoic acid, 4-methylbenzoic acid and (*R*)- or (*S*)-2-phenylpropanoic acid and their conjugate bases, also exhibit differences in hydrophobicity and charge depending on protonation, and in the case of the (*R*)- or (*S*)-2-phenylpropanoic acids and their conjugate bases, differences in chirality. We have previously reported[2] the complexation of these guests by βCD and 6^A-amino-6^A-deoxy-βCD which provides a comparison with our new studies.

Experimental

The modified βCDs were prepared as previously described,[17] and were dried to constant weight and stored over P_2O_5 prior to use. The carboxylic acids were high quality commercial grade materials. Deionized water was purified with a MilliQ-Reagent system to produce water with a specific resistance of >15 MΩ cm and, after boiling to remove CO_2, was used in the preparation of all solutions. A Metrohm Dosimat E665 titrimator, an Orion SA 720 potentiometer and an Orion 8172 Ross Sureflow combination pH electrode filled with 0.10 mol dm^{-3} $NaClO_4$ were used in all titrations. During each titration a fine stream of nitrogen bubbles (previously passed through aqueous 0.10 mol dm^{-3} NaOH to remove any CO_2 traces, and then through aqueous 0.10 mol dm^{-3} $NaClO_4$) was passed through the titration solution which was magnetically stirred and thermostatted at 298.2 ± 0.1 K in a water-jacketed 20 cm^3 titration vessel which was closed to the atmosphere with the exception of a small vent for the nitrogen stream. In each titration, 10 cm^3 of a solution 1.0×10^{-3} mol dm^{-3} in [βCDNH$(CH_2)_nNH_2$] and [carboxylic acid] and 2.0×10^{-3} mol dm^{-3} in [$HClO_4$] and I = 0.10 mol dm^{-3} ($NaClO_4$) were measured into the titration vessel and allowed 30 min to reach thermal equilibrium. Sodium hydroxide solution (0.10 mol dm^{-3}) was the titrant. All titrations were carried out in duplicate at least and a typical titration curve is shown in Fig. 2. Complex stability constants were derived from the titration data using the program SUPERQUAD.[18]

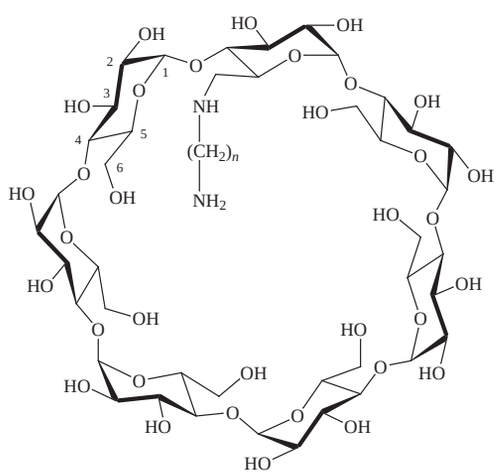

Fig. 1 6^A-(ω-Aminoalkylamino)-6^A-deoxy-β-cyclodextrins where n = 2, 3, 4 or 6.

† β-Cyclodextrin = cycloheptamaltose.

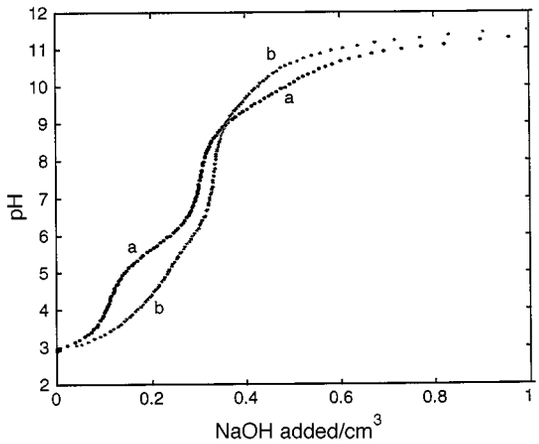

Fig. 2 The titration curves for a) a 0.010 mol dm^{-3} solution of βCDNH$(CH_2)_6NH_2$, 0.020 mol dm^{-3} in [$HClO_4$], and b) a 0.010 mol dm^{-3} solution of both βCDNH$(CH_2)_6NH_2$ and 4-methylbenzoic acid, 0.020 mol dm^{-3} in [$HClO_4$] against 0.100 mol dm^{-3} NaOH, at 298.2 K where I = 0.10 mol dm^{-3} ($NaClO_4$).

The ^{1}H NMR 1D and 2D ROESY (mixing time of 0.35 s)[19] experiments were run on a Varian Inova 600 spectrometer. Either βCDNH$(CH_2)_6NH_2$ alone or equimolar amounts of the guest species and βCDNH$(CH_2)_6NH_2$ were dissolved in D_2O to give total concentrations of 0.06 mol dm^{-3} of each species, and the pH was adjusted to ≥11.5. The resultant solutions were filtered and degassed by freeze–thawing before the spectra were recorded. The spectral assignments below are according to the glucopyranose numbering system H1–H6 (where a superscript A specifies the glucopyranose unit bearing the 1,6-diaminohexyl substituent) in Fig. 1. The hexyl protons are labelled Ha–Hf as distance increases from the secondary amine group, and aromatic guest protons are labelled as H*o* and H*m* where the former is adjacent to the carboxylate group.

The spectral assignments for βCDNH$(CH_2)_6NH_2$ alone at pH ≥ 11.5 are: 1D ^{1}H spectrum: δ_H 4.80 (s, 7H + solvent, H1); 3.5–3.8 (m, 26H, H3, H5, H6); 3.2–3.4 (m, 13H, H2, H4); 3.11 (t, J = 9.3 Hz, 1H, H4^A); 2.93 (d, J = 12.4 Hz, 1H, H6^A); 2.70 (m, 1H, H6$^{A'}$); 2.65 (m, 2H, Hf); 2.46 (m, 2H, Ha); 1.40 (br s, 4H, Hb, He); 1.26 (br s, 4H, Hc, Hd). The 2D ROESY spectrum shows the following cross-peaks: δ_H 1.26 (Hc,d) shows cross-peaks with 1.40 (Hb,e), 2.46 (Ha), 2.65 (Hf), 3.7 (H5), 3.8 (H3); 1.40 (Hb,e) shows cross-peaks with 1.26 (Hc,d), 2.46 (Ha), 2.65 (Hf), 3.7 (H5), 3.8 (H3); 2.46 (Ha) shows cross-peaks with 1.26 (Hc,d), 1.4 (Hb,e), 3.9 (H5^A); 2.65 (Hf) shows cross-peaks with 1.26 (Hc,d), 1.4 (Ha,e), 3.7 (H5), 3.8 (H3). These cross-peaks are absent from the 2D-ROESY spectrum obtained after acidification of the sample solution to pH 1 with hydrochloric acid.

The stability constants discussed below indicate that the [βCDNH$(CH_2)_6NH_2$·4-methylbenzoate]$^-$ complex constitutes 85% of the total [βCDNH$(CH_2)_6NH_2$] and [4-methylbenzoate] at the pH ≥ 11.5 of the NMR study. The spectral assignments are: 1D ^{1}H spectrum: δ_H 7.80 (d, J = 7.8 Hz, 2H, H*o*); 7.29 (d, J = 7.8 Hz, 2H, H*m*); 5.00 (m, 7H + solvent, H1); 3.6–3.9 (m, 26H, H3, H5, H6); 3.4–3.6 (m, 13H, H2, H4); 3.26 (t, J = 9.6 Hz, 1H, H4^A); 2.99 (d, J = 13.2 Hz, 1H, H6^A); 2.71 (m, 1H, H6$^{A'}$); 2.64 (t, J = 7.2 Hz, 2H, Hf); 2.45 (t, J = 7.2 Hz, 2H, Ha); 2.41 (s, 3H, Me-); 1.4–1.5 (m, 4H, Hb, He); 1.2–1.3 (m, 4H, Hc, Hd). The 2D ROESY spectrum shows the following cross-peaks: δ_H 1.2–1.3 (Hc,d) shows cross-peaks with 1.4–1.5 (Hb,e), 2.45 (Ha), 2.64 (Hf), 3.8 (H5), 3.9 (H3); 1.4–1.5 (Hb,e) shows cross-peaks with 1.2–1.3 (Hc,d), 2.45 (Ha), 2.64 (Hf), 3.8 (H5), 3.9 (H3); 2.41 (Me) shows a cross-peak with 7.29 (H*m*); 2.45 (Ha) shows cross-peaks with 1.2–1.3 (Hc,d), 1.4–1.5 (Hb,e), 3.8 (H5), 3.9 (H3); 2.64 (Hf) shows cross-peaks with 1.2–1.3 (Hc,d), 1.4–1.5 (Hb,e), 3.8 (H5), 3.9 (H3); 2.71 (H6$^{A'}$) shows a cross-peak with 3.26 (H4^A); 3.26 (H4^A) shows a cross-peak with 2.71 (H6$^{A'}$); 3.8 (H5) shows cross-peaks with 1.2–1.3 (Hc,d), 1.4–1.5 (Hb,e), 7.29 (H*o*), 7.80 (H*m*); 3.9 (H3) shows cross-peaks with 1.2–1.3 (Hc,d), 1.4–1.5 (Hb,e), 2.45 (Ha), 2.64 (Hf), 7.29 (H*o*), 7.80 (H*m*); 7.29 (H*o*) shows cross-peaks with 2.41 (Me-), 3.8 (H5), 3.9 (H3), 7.80 (H*m*); 7.80 (H*m*) shows cross-peaks with 3.8 (H5), 3.9 (H3), 7.29 (H*o*).

The stability constants discussed below indicate that the [βCDNH$(CH_2)_6NH_2$·(*S*)-2-phenylpropanoate]$^-$ complex constitutes 90% of the total [βCDNH$(CH_2)_6NH_2$] and [(*S*)-2-phenylpropanoate] at the pH ≥ 11.5 of the NMR study. The spectral assignments are: 1D ^{1}H spectrum: δ_H 7.30 (m, 4H, H*m*, H*o*); 7.22 (m, 1H, H*p*); 4.92 (br s, 7H + solvent, H1); 3.6–4.0 (m, 26H, H3, H5, H6); 3.58 (q, J = 7.2 Hz, αCH); 3.2–3.5 (m, 13H, H2, H4); 3.19 (t, J = 9.6 Hz, 1H, H4^A); 2.93 (d, J = 12.6 Hz, 1H, H6^A); 2.67 (dd, J = 12.6, 9.5 Hz, 1H, H6$^{A'}$); 2.62 (t, J = 7.2 Hz, 2H, Hf); 2.45 (t, J = 7.8 Hz, 2H, Ha); 1.39 (m, 4H, Hb,e); 1.36 (d, J = 7.2 Hz, 3H, Me); 1.25 (m, 4H, Hc,d). 2D ROESY cross-peaks: δ_H 1.25 (Hc,d) shows cross-peaks with 1.39 (Hb,e), 2.45 (Ha), 2.62 (Hf), 3.7 (H5), 3.75 (H3); 1.36 (Me) shows a cross-peak with 3.58 (αCH); 1.39 (Hb,e) shows cross-peaks with 1.25 (Hc,d), 2.45 (Ha), 2.62 (Hf), 3.7 (H5), 3.75 (H3); 2.45 (Ha) shows cross-peaks with 1.25 (Hc,d), 1.39 (Hb,e), 3.7 (H5), 3.8 (H5^A); 2.62 (Hf) shows cross-peaks with 1.25 (Hc,d), 1.39 (Hb,e), 3.7 (H5), 3.75 (H3); 2.67 (H6$^{A'}$) shows cross-peaks with 2.93 (H6^A), 3.19 (H4^A); 2.93 (H6^A) shows cross-peaks with 2.67 (H6$^{A'}$), 3.7 (H5), 3.8 (H5^A); 3.19 (H4^A) shows cross-peaks with 2.67 (H6$^{A'}$), 3.75 (H3), 3.8 (H5^A); 3.58 (αCH) shows cross-peaks with 1.36 (Me), 7.30 (H*o*, H*m*); 3.7 (H5) shows cross-peaks with 1.25 (Hc,d), 1.39 (Hb,e), 2.62 (Hf), 7.30 (H*o*, H*m*); 3.75 (H3) shows cross-peaks with 1.25 (Hc,d), 1.39 (Hb,e), 2.62 (Hf), 7.30 (H*o*, H*m*); 3.8 (H5^A) shows cross-peaks with 2.45 (Ha), 2.93 (H6^A), 3.19 (H4^A); 7.30 (H*o*, H*m*) shows cross-peaks with 3.58 (αCH), 3.75 (H3). Very similar spectra were recorded for the βCDNH$(CH_2)_6NH_2$·(*R*)-2-phenylpropanoate complex.

Molecular modelling[20] was carried out using a Silicon Graphics Iris Indigo X2 400 Unix workstation. Computational results were obtained using the force-field programme CVFF with the 6-12 ε function with geometric averages for the heteronuclear interactions. Energy minimisations were performed with the Discover programme, using a steepest descents algorithm until the root mean square of the residuals (RMS) derived <10, whereafter a conjugate gradients algorithm was used until RMS <1 and the global minimisation was obtained using a quasi Newton–Raphson algorithm. Several local energy minima were found before the global minimum was reached. Graphical displays were printed through the Insight II molecular modelling programme.

Results and discussion

Complex stabilities

The various weak secondary bonding interactions between the CD host and the guest can sum to produce quite high stabilities in CD complexes. It has been concluded from a wide range of studies dominated by natural CDs that the complexation process involves conformational change in the CD host and the guest accompanied by dehydration of both to an extent depending on the nature of both entities.[1,3,11,13,15,16] These aspects of CD complexation have been extensively discussed, and as a consequence only those interactions which appear to dominate the variations in stability observed in this study are discussed in detail here. On a statistical basis, 64 different host–guest complexes could be formed between the four host βCDNH_2-$(CH_2)_nNH_3^{2+}$ (n = 2, 3, 4 and 6) and the four guest carboxylic acids and their respective conjugate bases, but only 35 of these complexes were detected under the conditions of this study. The complex stability range is encompassed by stability constants K = 140 and 1760 dm^3 mol^{-1} for equilibria (7) and (16) when n = 2 and 6, respectively, as shown in Table 1. While each pos-

Table 1 Equilibria, K and pK_a[a] values determined in aqueous solution at $I = 0.10$ mol dm^{-3} ($NaClO_4$) and 298.2 K

Equilibrium	K/dm^3 mol^{-1}			
	$n = 2$	$n = 3$	$n = 4$	$n = 6$
(1) $\beta CDNH_2(CH_2)_nNH_3^{2+}$ + benzoic acid ⇌ $[\beta CDNH_2(CH_2)_nNH_3 \cdot$benzoic acid$]^{2+}$	820 ± 170	350 ± 80	740 ± 100	545 ± 140
(2) $\beta CDNH_2(CH_2)_nNH_3^{2+}$ + benzoate$^-$ ⇌ $[\beta CDNH_2(CH_2)_nNH_3 \cdot$benzoate$]^+$	870 ± 70			
(3) $\beta CDNH(CH_2)_nNH_3^+$ + benzoate$^-$ ⇌ $[\beta CDNH(CH_2)_nNH_3 \cdot$benzoate]	180 ± 15		305 ± 35	275 ± 30
(4) $\beta CDNH(CH_2)_nNH_2$ + benzoate$^-$ ⇌ $[\beta CDNH(CH_2)_nNH_3 \cdot$benzoate$]^-$		915 ± 60	950 ± 120	1000 ± 120
(5) $\beta CDNH_2(CH_2)_nNH_3^{2+}$ + 4-methylbenzoic acid ⇌ $[\beta CDNH_2(CH_2)_nNH_3 \cdot$4-methylbenzoic acid$]^{2+}$		420 ± 60	415 ± 65	
(6) $\beta CDNH_2(CH_2)_nNH_3^{2+}$ + 4-methylbenzoate$^-$ ⇌ $[\beta CDNH_2(CH_2)_nNH_3 \cdot$4-methylbenzoate$]^+$	1000 ± 170			
(7) $\beta CDNH(CH_2)_nNH_3^+$ + 4-methylbenzoate$^-$ ⇌ $[\beta CDNH(CH_2)_nNH_3 \cdot$4-methylbenzoate]	140 ± 35			180 ± 20
(8) $\beta CDNH(CH_2)_nNH_2$ + 4-methylbenzoate$^-$ ⇌ $[\beta CDNH(CH_2)_nNH_2 \cdot$4-methylbenzoate$]^-$		885 ± 65	535 ± 50	750 ± 20
(9) $\beta CDNH_2(CH_2)_nNH_3^{2+}$ + (*R*)-2-phenylpropanoic acid ⇌ $[\beta CDNH_2(CH_2)_nNH_3 \cdot$(*R*)-2-phenylpropanoic acid$]^{2+}$	850 ± 170	395 ± 50	420 ± 65	
(10) $\beta CDNH_2(CH_2)_nNH_3^{2+}$ + (*R*)-2-phenylpropanoate$^-$ ⇌ $[\beta CDNH_2(CH_2)_nNH_3 \cdot$(*R*)-2-phenylpropanoate$]^+$	790 ± 80			
(11) $\beta CDNH(CH_2)_nNH_3^+$ + (*R*)-2-phenylpropanoate$^-$ ⇌ $[\beta CDNH(CH_2)_nNH_3 \cdot$(*R*)-2-phenylpropanoate]				250 ± 35
(12) $\beta CDNH(CH_2)_nNH_2$ + (*R*)-2-phenylpropanoate$^-$ ⇌ $[\beta CDNH(CH_2)_nNH_2 \cdot$(*R*)-2-phenylpropanoate$]^-$		760 ± 75	630 ± 30	1150 ± 295
(13) $\beta CDNH_2(CH_2)_nNH_3^{2+}$ + (*S*)-2-phenylpropanoic acid ⇌ $[\beta CDNH_2(CH_2)_nNH_3 \cdot$(*S*)-2-phenylpropanoic acid$]^{2+}$	345 ± 40	690 ± 70	570 ± 80	
(14) $\beta CDNH_2(CH_2)_nNH_3^{2+}$ + (*S*)-2-phenylpropanoate$^-$ ⇌ $[\beta CDNH_2(CH_2)_nNH_3 \cdot$(*S*)-2-phenylpropanoate$]^+$	630 ± 30			
(15) $\beta CDNH(CH_2)_nNH_3^+$ + (*S*)-2-phenylpropanoate$^-$ ⇌ $[\beta CDNH(CH_2)_nNH_3 \cdot$(*S*)-2-phenylpropanoate]				285 ± 80
(16) $\beta CDNH(CH_2)_nNH_2$ + (*S*)-2-phenylpropanoate$^-$ ⇌ $[\beta CDNH(CH_2)_nNH_2 \cdot$(*S*)-2-phenylpropanoate$]^-$		465 ± 60	630 ± 105	1760 ± 150
(17) βCD + benzoic acid ⇌ [βCD·benzoic acid]	590[b]			
(18) βCD + benzoate$^-$ ⇌ $[\beta CD \cdot$benzoate$]^-$	60[b]			
(19) $\beta CDNH_3^+$ + benzoic acid ⇌ $[\beta CDNH_3 \cdot$benzoic acid$]^+$	340[b]			
(20) $\beta CDNH_3^+$ + benzoate$^-$ ⇌ $[\beta CDNH_3 \cdot$benzoate]	120[b]			
(21) $\beta CDNH_2$ + benzoate$^-$ ⇌ $[\beta CDNH_2 \cdot$benzoate$]^-$	50[b]			
(22) βCD + 4-methylbenzoic acid ⇌ [βCD·4-methylbenzoic acid]	1680[b]			
(23) βCD + 4-methylbenzoate$^-$ ⇌ $[\beta CD \cdot$4-methylbenzoate$]^-$	110[b]			
(24) $\beta CDNH_3^+$ + 4-methylbenzoic acid ⇌ $[\beta CDNH_3 \cdot$4-methylbenzoic acid$]^+$	910[b]			
(25) $\beta CDNH_3^+$ + 4-methylbenzoate$^-$ ⇌ $[\beta CDNH_3 \cdot$4-methylbenzoate]	330[b]			
(26) $\beta CDNH_2$ + 4-methylbenzoate$^-$ ⇌ $[\beta CDNH_2 \cdot$4-methylbenzoate$]^-$	100[b]			

[a] The pK_as of $\beta CDNH_2(CH_2)_nNH_3^{2+}$ are 9.42 and 5.70 when $n = 2$, 9.90 and 7.39 when $n = 3$, 10.26 and 8.06 when $n = 4$ and 10.26 and 8.72 when $n = 6$ under the conditions of this study.[17] The pK_a of $\beta CDNH_3^+ = 8.49$.[2] The pK_as of benzoic, 4-methylbenzoic and (*R*)- or (*S*)-2-phenylpropanoic acid are 4.06, 4.20 and 4.23, respectively.[17] [b] From reference 2.

sible type of complex was detected for each guest species, each type of complex was not detected for every value of n for the $\beta CDNH(CH_2)_nNH_2$ host. There are systematic absences in the detected species about the diagonal of data sets for $n = 2$, 3, 4 and 6 in Table 1. The negatively charged complex was not detected when $n = 2$, the neutral and unipositively charged complexes were not detected when $n = 3$ and 4, except when the guest was benzoate in the last case. When $n = 6$, the dipositively charged complex was only detected when benzoic acid was the guest. (It should be noted that complexes present at concentrations at ≤ 5% of total $[\beta CDNH(CH_2)_nNH_2]$ are not reliably detected by the potentiometric method used in this study and are listed as not detected.)

A broad pattern emerges in the variation of complex stability in Table 1. This may be represented through Fig. 3 where the rectangle represents the set of stability constants for a carboxylic acid and its carboxylate being complexed by $\beta CDNH(CH_2)_nNH_2$ and its protonated analogues. Generally, the most stable complexes occur in the top left and bottom right corners of the rectangle (with the exception of $[\beta CDNH_2(CH_2)_2NH_3 \cdot$4-methylbenzoic acid$]^{2+}$ which was not detected), while there is an absence of detected complexes about the diagonal running from the bottom left to the top right of the rectangle. This

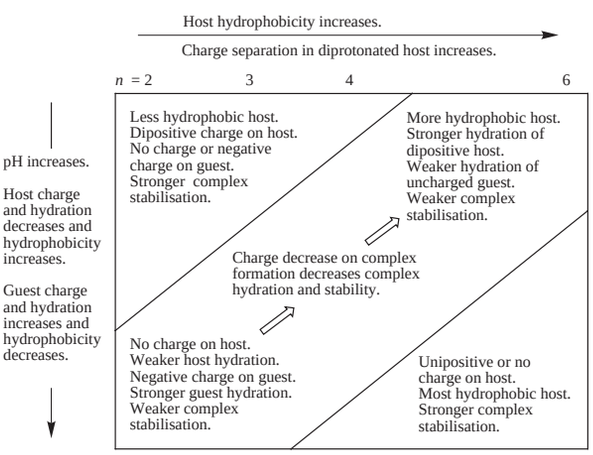

Fig. 3 A schematic representation of the variation of complex stability and the factors contributing to it.

pattern of variation in stability may be attributed to the effects of changes in i) the charge, ii) the hydrophobicity, iii) the stereochemistry of both host and guest, and iv) changes in CD host,

guest and complex hydration superimposing on the stabilising effect of entry of the hydrophobic phenyl group of the guest into the hydrophobic centre of the βCD annulus.

The βCD annulus and the two amine groups represent constant structural features in the four $\beta CDNH(CH_2)_nNH_2$ hosts. Hence, variations in effects i)–iv) generated by the CD hosts arise predominantly from the changes in the length of the $-NH(CH_2)_nNH_2$ substituent indicated by *n*. The phenyl ring and the carboxylic acid group are invariant in the four guest carboxylic acids, and therefore differences in their complexation characteristics arise from differences in hydrophobicity and stereochemistry engendered by the CH_3- and $CH_3CH<$ moieties in 4-methylbenzoic acid and (*R*)- or (*S*)-2-phenylpropanoic acid, respectively, when compared with those of benzoic acid.

Descending the vertical axis of Fig. 3 is equivalent to increasing solution pH so that the host's charge decreases with decreasing protonation and its hydrophobicity increases as a consequence. Correspondingly, the guest becomes negatively charged with the probable result that its hydration increases and its hydrophobicity decreases. On the horizontal axis, host hydrophobicity increases and the charge separation in the diprotonated host increases as *n* in $\beta CDNH(CH_2)_nNH_2$ increases. Thus, in the upper left hand corner of Fig. 3, the host has a dipositive charge and the guest has either no charge or a negative charge, and it appears that either a charge–dipole or a charge–charge interaction stabilises the complex. The intensity of this interaction probably diminishes as *n* increases and the charges in $\beta CDNH_2(CH_2)_nNH_3^{2+}$ move further apart so that the stabilities of the complexes diminish in the upper right hand corner of Fig. 3. (The orientation of the carboxylic acid or carboxylate guest within the annulus may also change with variation of $\beta CDNH(CH_2)_nNH_2$ charge as its protonation changes as indicated by the molecular modelling studies discussed below.)

At the lower left hand corner of Fig. 3, the host is uncharged, the guest is negatively charged and no complexes are observed. (This is consistent with the observation that the analogous complexes formed between βCD and $\beta CDNH_2$ and the same carboxylates as those studied here are characterised by stability constants in the range 13–110 $dm^3\ mol^{-1}$ while the complexes formed by βCD, $\beta CDNH_2$ and $\beta CDNH_3^+$ with the analogous acids are characterised by stability constants in the range 340–1680 $dm^3\ mol^{-1}$.[2]) However, as hydrophobicity increases with *n* as Fig. 3 is traversed from left to right, complexation of the carboxylate guest strengthens to give more stable complexes in the lower right hand corner of Fig. 3. While it is possible that the increase in host hydrophobicity is the main source of complex stabilisation, it also appears (from the NMR and molecular modelling studies discussed below) that the $-NH(CH_2)_6NH_2$ substituent enters the annulus in basic solution both in $\beta CDNH(CH_2)_6NH_2$ and in its carboxylate complexes.[21] As the $\beta CDNH(CH_2)_6NH_2$ carboxylate complexes are some of the more stable species, it seems that this latter effect stabilises the host–guest complex while the short $-NH(CH_2)_2NH_2$ substituent is less effective in this role. Similar self-complexations are observed for the pendant naphthyl group and pendant dansyl group of 6^A-*N*-[*N*′-(5-dimethylamino-1-naphthylsulfonyl)diaminoethane]-6^A-deoxy-β-cyclodextrin and 3^A-*O*-naphthylmethyl-β-cyclodextrin, respectively.[22,23] The charged $-NH_2(CH_2)_6NH_3^{2+}$ substituent does not appear to enter the $\beta CDNH_2(CH_2)_6NH_3^{2+}$ annulus as is discussed below.

Superimposed on these effects is that of hydration. The hydration of $\beta CDNH(CH_2)_nNH_2$ and its protonated analogues probably has two main components: water occupying the annulus but interacting weakly with the methylene, methine and ether oxygen moieties defining the hydrophobic centre of the annulus, and water hydrogen bonding with the hydroxy groups at either end of the annulus. (From 6–6.5 H_2O have been observed in the βCD annulus in solid state neutron and X-ray diffraction studies.[24–27]) The displacement of water from the annulus by the hydrophobic moiety of the guest during complexation represents a major hydration change and probably makes a significant contribution to the free energy of complexation.[1,3,6,15,16] Depending on whether it is $\beta CDNH(CH_2)_nNH_2$ or one of its protonated analogues which acts as the host, and whether the carboxylic acid or its carboxylate acts as the guest, the extent of hydration may either increase or decrease on formation of the complex. The complete or partial cancellation of host charge by that of the guest is likely to produce a decrease in the overall hydration of the complex compared with that of its charged components, and a consequent lessening of complex stability. This occurs in the centre of Fig. 3 and is exemplified by equilibria (2) and (3), (6) and (7), (10) and (11), and (14) and (15) in Table 1. Hydration effects on stability are likely to be less for equilibria (1), (4), (5), (8), (9), (12), (13) and (16) where no change in overall charge occurs on complexation. Thus, a degree of complex stabilisation may be achieved through minimising the interactions of the hydrophobic moieties of the host and guest with water through partial encapsulation of the hydrophobic guest moiety in the hydrophobic annulus of the host while retaining the hydration of their hydrophilic moieties.

There is no apparent systematic stereochemical effect of the variation of *n* in $\beta CDNH(CH_2)_nNH_2$ and its conjugate acids and of the stereochemistry of the carboxylic acids and the conjugate bases other than those subsumed into the discussion of factors i)–iv) above. However, there is a small chiral discrimination in equilibria (9) and (13) when $n = 2$ and 3, where $[\beta CDNH_2(CH_2)_nNH_3{\cdot}(R)$-2-phenylpropanoic acid$]^{2+}$ is more stable than its (*S*)-analogue when $n = 2$ and *vice versa* when $n = 3$. Although small, these differences are consistent with *n* of the $-NH_2(CH_2)_nNH_3^{2+}$ substituent influencing guest orientation as is also the case for the $-NH(CH_2)_nNH_2$ substituent where $n = 3$ and 6 in equilibria (12) and (16).

NMR structural studies

The detailed assignment of the ^1H NMR spectrum of $\beta CDNH(CH_2)_6NH_2$ at pH ≥ 11.5 (Fig. 4) is presented in the Experimental section, and the cross-peaks observed in the ROESY spectrum (Table 2) provide structural information. The cross-peaks arising from interaction between Hb–Hf of the $-NH(CH_2)_6NH_2$ substituent and H3 and H5 of the βCD annulus and Ha–Hc and $H5^A$ are consistent with complexation of $-NH(CH_2)_6NH_2$ inside the annulus as shown schematically in Fig. 5. These cross-peaks are absent from the ROESY spectrum obtained after acidification of the sample solution to pH 1 with hydrochloric acid consistent with the protonated $-NH_2(CH_2)_6NH_3^{2+}$ substituent being excluded from the annulus as a result of its decreased hydrophobicity.

The titration data discussed above indicate that the $[\beta CDNH(CH_2)_6NH_2{\cdot}$4-methylbenzoate$]^-$ complex constitutes 85% of the total $[\beta CDNH(CH_2)_6NH_2]$ and [4-methylbenzoate] at the pH ≥ 11.5 of the NMR study. The ^1H chemical shifts and the spectral resolution of the $-NH(CH_2)_6NH_2$ substituent methylene protons of $\beta CDNH(CH_2)_6NH_2$ differ from those observed for the $[\beta CDNH(CH_2)_6NH_2{\cdot}$4-methylbenzoate$]^-$ complex (Fig. 5 and Experimental) consistent with the methylene chain of $-NH(CH_2)_6NH_2$ being inside the βCD annulus and parallel to the face of the aromatic ring where they experience an anisotropic field arising from the high π electron density of the guest. Cross-peaks between Ha–Hf of $-NH(CH_2)_6NH_2$ and H3 and H5, and H*o* and H*m* of the 4-methylbenzoate and H3 and H5 (Fig. 6 and Table 2) are consistent with the simultaneous complexation of both entities in the βCD annulus. However, there are no cross-peaks due to interactions between the $-NH(CH_2)_6NH_2$ substituent and 4-methylbenzoate. These data are consistent with either a single complex where the carboxylate protrudes from either the primary or the secondary face (Fig. 5) of the βCD annulus (respectively delineated by primary and

Table 2 ^{1}H NMR cross-peaks[a] observed for βCDNH$_2$(CH$_2$)$_6$NH$_2$ and its 4-methylbenzoate and (*S*)-2-phenylpropanoate complexes

	βCDNH$_2$(CH$_2$)$_6$NH$_2$					
Annular protons	6-Aminohexyl substituent protons					
	Ha	Hb	Hc	Hd	He	Hf
H3		+	++	++	++	++
H5	+	++	++	++	++	+
H5^A	++	+	+			

	[βCDNH$_2$(CH$_2$)$_6$NH$_2$·4-methylbenzoate]$^-$								
Annular protons	6-Aminohexyl substituent and 4-methylbenzoate protons								
H	Ha	Hb	Hc	Hd	He	Hf	H*o*	H*m*	Me
H3	++	++	++	++	++	++	+	++	+
H5	++	++	++	++	++	+	+	++	+

	[βCDNH$_2$(CH$_2$)$_6$NH$_2$·(*S*)-2-phenylpropanoate]$^-$								
Annular protons	6-Aminohexyl substituent and (*S*)-2-phenylpropanoate protons								
	Ha	Hb	Hc	Hd	He	Hf	H*o*,	H*m*	Me
H3		+	+	+	+	++		+	
H5	+	+	+	+	+	+			
H5^A	++								

[a] The intensity of the cross peaks increases from + to ++. The concentrations of βCDNH$_2$(CH$_2$)$_6$NH$_2$ and either 4-methylbenzoate or (*S*)-2-phenylpropanoate, when present, were 0.06 mol dm^{-3}.

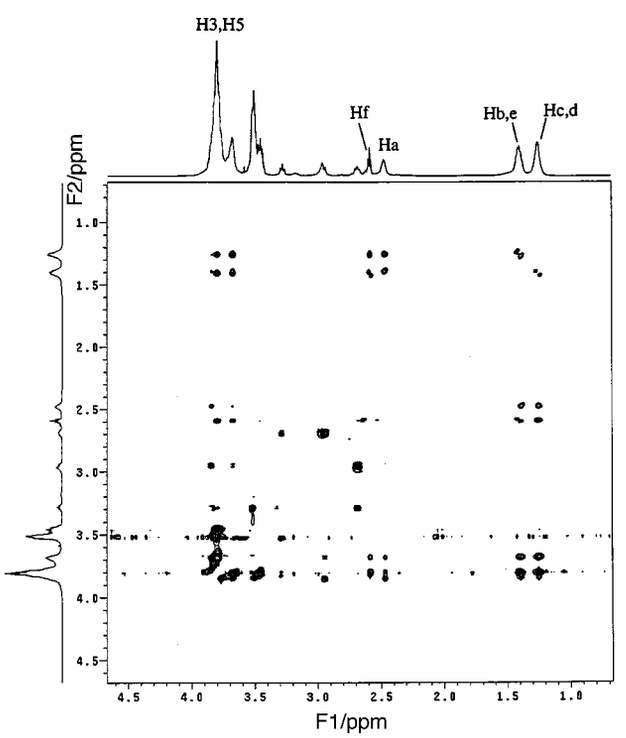

Fig. 4 The ^{1}H 600 MHz ROESY NMR spectrum of βCDNH-(CH$_2$)$_6$NH$_2$. Cross-peaks are formed between Hb–Hf and H3, Ha–Hf and H5 and Ha–Hc and H5^A.

secondary hydroxy groups), or the coexistence of both complex isomers.

The titration data above also indicate that the [βCDNH-(CH$_2$)$_6$NH$_2$·(*S*)-2-phenylpropanoate]$^-$ complex constitutes 90% of the total [βCDNH(CH$_2$)$_6$NH$_2$] and [(*S*)-2-phenylpropanoate] at the pH ≥ 11.5 of the NMR study. In the ROESY spectrum cross-peaks between Ha–Hf of -NH(CH$_2$)$_6$NH$_2$ and H3 and H5, and H*o* and H*m* of (*S*)-2-phenylpropanoate and H3 and H5 (Table 2) are consistent with the simultaneous complexation of both entities in the βCD annulus. However, there are no cross-peaks due to interactions between the -NH(CH$_2$)$_6$-NH$_2$ substituent and (*S*)-2-phenylpropanoate. The cross-peak between H*o*, H*m* and H3 and the lack of cross-peaks between the aryl protons of (*S*)-2-phenylpropanoate and H5 is consistent with shallow complexation of the aromatic ring in the βCD annulus. No cross-peaks are observed between the methyl group of the (*S*)-2-phenylpropanoate and either H3 or H5 consistent with the (*S*)-2-phenylpropanoate moiety protruding from the secondary face of the β-CD annulus. A very similar spectrum was recorded for the [βCDNH(CH$_2$)$_6$NH$_2$·(*R*)-2-phenylpropanoate]$^-$ complex.

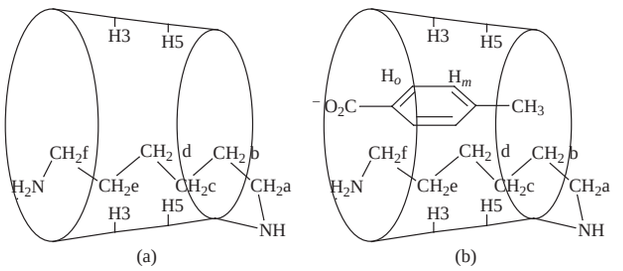

Fig. 5 Schematic representations of the structures of (a) the intramolecular complex formed by βCDNH(CH$_2$)$_6$NH$_2$ and (b) the intermolecular [βCDNH(CH$_2$)$_6$NH$_2$·4-methylbenzoate]$^-$ complex.

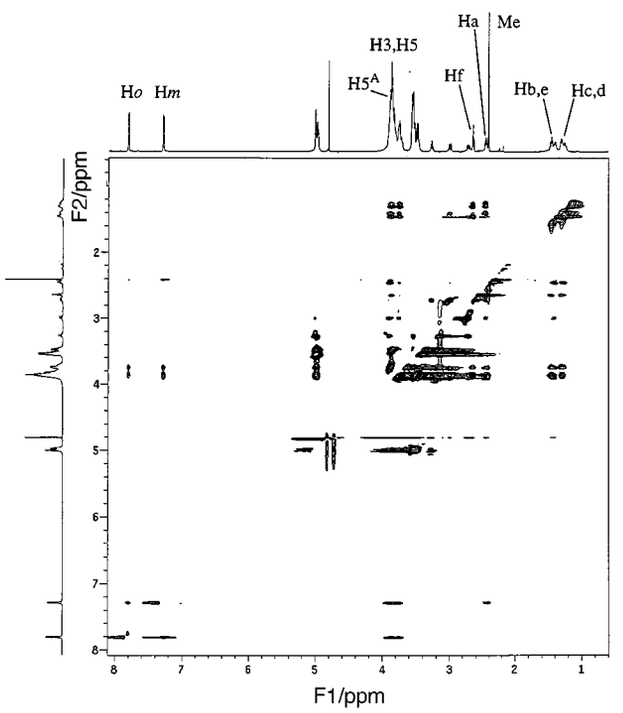

Fig. 6 The ^{1}H 600 MHz ROESY NMR spectrum of the [βCDNH(CH$_2$)$_6$NH$_2$·4-methylbenzoate]$^-$ complex showing cross-peaks formed between Ha to Hf and H3 and H5, H*o* and H*m* of 4-methylbenzoate and H3 and H5, and Me of 4-methylbenzoate and H3 and H5.

Molecular modelling

Gas phase force-field molecular modelling[20] produced the global energy minimised (877.1 kJ mol^{-1}) structure of βCDNH-(CH$_2$)$_6$NH$_2$ with the -NH(CH$_2$)$_6$NH$_2$ substituent complexed inside the βCD annulus as shown in Fig. 7. Similar modelling showed that the -NH$_2$(CH$_2$)$_6$NH$_3^{2+}$ substituent of βCDNH$_2$-(CH$_2$)$_6$NH$_3^{2+}$ does not enter the βCD annulus. Both of these

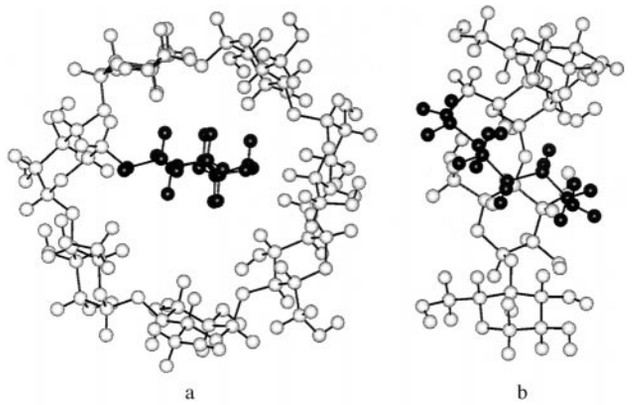

Fig. 7 The global energy minimised structure of $\beta CDNH(CH_2)_6NH_2$ viewed from a) the primary face of the annulus and b) from the side with three glucopyranose units cut away. The $-NH(CH_2)_6NH_2$ substituent is shown in dark shading.

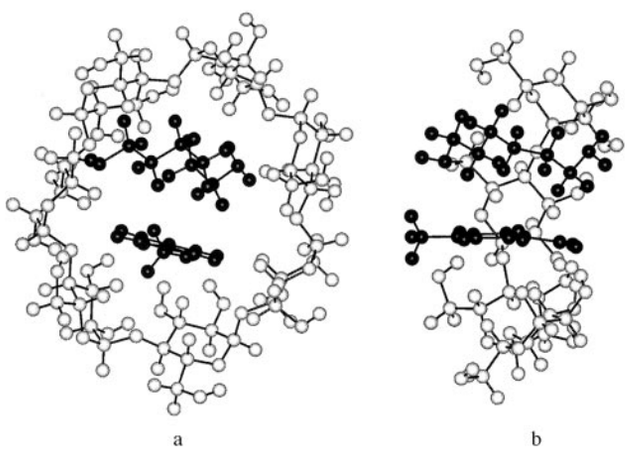

Fig. 8 The global energy minimised structure of $[\beta CDNH(CH_2)_6NH_2 \cdot 4\text{-methylbenzoate}]^-$ viewed from a) the primary face of the βCD annulus and b) from the side with three glucopyranose units cut away. The $-NH(CH_2)_6NH_2$ substituent and 4-methylbenzoate are shown in dark shading.

models are consistent with the deductions made from the NMR data discussed above.

Modelling the $[\beta CDNH(CH_2)_6NH_2 \cdot 4\text{-methylbenzoate}]^-$ complex produced a global energy minimum (797.4 kJ mol^{-1}) for the structure shown in Fig. 8 where 4-methylbenzoate is oriented with its carboxylate group towards the secondary face of the βCD annulus and the $-NH(CH_2)_6NH_2$ substituent is also complexed inside the βCD annulus consistent with the NMR data. This orientation of the carboxylate towards the secondary face is also found in adamantane-1-carboxylate complexes of αCD and βCD[10] and the cyclohexanecarboxylate complex of βCD.[11] When the 4-methylbenzoate orientation is reversed so that the carboxylate group is oriented towards the primary face the complex energy is 870.4 kJ mol^{-1} consistent with this being a less favoured orientation. The modelled structure of the $[\beta CDNH_2(CH_2)_6NH_3 \cdot 4\text{-methylbenzoate}]^+$ complex shows the 4-methylbenzoate guest to have its carboxylate group in the vicinity of the primary face of the βCD annulus. This reversal of orientation, compared with that in the $[\beta CDNH(CH_2)_6NH_2 \cdot 4\text{-methylbenzoate}]^-$ complex, is consistent with the charge of the $-NH_{(1+m)}(CH_2)_2NH_{(2+n)}^{(m+n)+}$ substituent (where $m = 0$ or 1 and $n = 0$ or 1) substantially influencing guest orientation through electrostatic interactions. This has also been found to occur in modelling studies of the 4-methylbenzoate complex of protonated heptakis(6-amino-6-deoxy)-β-cyclodextrin[12] and also its amino acid complexes.[14] While our modelling studies show the probable orienting effects of charge in $[\beta CDNH_2(CH_2)_6NH_3 \cdot 4\text{-methylbenzoate}]^+$ and $[\beta CDNH(CH_2)_6NH_2 \cdot 4\text{-methylbenzoate}]^-$, the latter complex was not detected in solution as discussed above.

Molecular modelling also shows that both the $-NH(CH_2)_6NH_2$ substituent and (*S*)-phenylpropanoate are complexed within the βCD annulus in the $[\beta CDNH(CH_2)_6NH_2 \cdot (S)\text{-phenylpropanoate}]^-$ complex. The carboxylate group is oriented towards the secondary face of the βCD annulus. The $[\beta CDNH(CH_2)_6NH_2 \cdot (R)\text{-phenylpropanoate}]^-$ complex is found to have a similar structure to that of its (*S*)-analogue with some differences in orientation of the guest within the βCD annulus. The globalised energy minima of the $[\beta CDNH(CH_2)_6NH_2 \cdot (S)\text{-phenylpropanoate}]^-$ complex and its (*R*)-analogue are 738.5 and 812.3 kJ mol^{-1}, respectively, showing the (*S*)-diastereomer to be the more stable in the gas phase.

Conclusion

The stabilities of the host–guest complexes formed between the 6^A-(ω-aminoalkylamino)-6^A-deoxy-β-cyclodextrin hosts and their protonated forms [where the ω-aminoalkylamino groups are $-NH(CH_2)_nNH_2$ and $n = 2$, 3, 4 and *6*] and the guests, benzoic acid, 4-methylbenzoic acid and (*R*)- or (*S*)-2-phenylpropanoic acid and their conjugate bases, vary significantly. This is consistent with the charge and hydrophobicity of the host and the guest generating significant secondary interactions which affect complex stability. The ^1H NMR studies show that the $-NH(CH_2)_6NH_2$ substituent of $\beta CDNH(CH_2)_6NH_2$ self-complexes inside the βCD annulus, and that in $[\beta CDNH(CH_2)_6NH_2 \cdot 4\text{-methylbenzoate}]^-$ and its (*S*)-2-phenylpropanoate analogue both the guest and the $-NH(CH_2)_6NH_2$ substituent are simultaneously complexed within the βCD annulus. Gas phase force-field modelling also show this to occur for $\beta CDNH(CH_2)_6NH_2$ and these two complexes, where in the latter two cases the carboxylate group is oriented towards the secondary face of the annulus. The entry of the $-NH(CH_2)_6NH_2$ substituent into the βCD annulus may significantly affect complex stability, as may also be the case for the $-NH(CH_2)_nNH_2$ substituents where $n = 2$, 3 and 4 if they also enter the βCD annulus. In contrast the fully protonated $-NH_2(CH_2)_nNH_3^{2+}$ substituent does not enter the βCD annulus according to the ^1H NMR and molecular modelling studies.

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