

Original Research Article

Complexation of Natural and Methylated β -cyclodextrins with Long-chain Carboxylic Acids in Aqueous Solutions. Calorimetric Studies at 298 K

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Abstract

The formation of complexes between β -cyclodextrin or methyl- β -cyclodextrin and monocarboxylic acids from C₅ to C₁₂ has been studied calorimetrically at 298 K in aqueous buffer phosphate, pH 11.3. When a complex forms, calorimetry enables the calculation of both the enthalpy and the association constant, from which the Gibbs free energy and the entropy of the process can be obtained. The forces involved in the association process are then discussed in the light of the signs and values of the thermodynamic parameters obtained. For β -cyclodextrin, 1:1 inclusion complexes form, characterized by an irregular variation in the values of enthalpies, entropies and association constants at increasing alkyl chain length. A model is proposed to explain this unusual behaviour for acids longer than ten carbon atoms. The association involving methyl- β -cyclodextrin shows, instead, a regular variation of the thermodynamic parameters up to the C₁₂ term. An elongation of the cavity effect is discussed: the cavity behaves as it were deeper than that of the natural cyclodextrins. Association is characterized prevalently by hydrophobic interactions; for the longer terms, the high and almost invariant entropic contribution determines the large association constants, notwithstanding the positive, unfavorable enthalpic contribution. This is an indication that the relaxation of water molecules from the hydrophobic hydration shells of the external methyl groups is the contribution that mainly determines the association process.

Introduction

Natural cyclodextrins (CDs), the most suitable host molecules for the recognition in aqueous media of hydrophobic guest molecules, are cyclic oligomers of α -D-glucose. They are characterized by a fairly polar exterior and by a cavity which is nonpolar relative to the exterior and to the usual external aqueous environments. The most important property of CDs is their ability to form complexes with a great variety of organic substances either in solution or in the solid state [1–5]. The formation of a complex is a process determined by a variety of noncovalent intermolecular forces: hydrophobic interactions, hydrogen bonds, van der Waals interactions, conformational energy, dipole-dipole and ion-dipole interactions [1, 6–8]. Upon association, another contribution originates from the rearrangement of water molecules originally surrounding both cyclodextrin and guest molecule [9–11]. Complexation alters the physicochemical properties of the included substances:

because of that, cyclodextrins are widely used for the enhancement of the aqueous solubility, stability and bioavailability of apolar drugs with serious problems of side effects [12–15]. In the last years, chemically modified cyclodextrins have been prepared with the aim of providing versatile carrier and delivery systems for drug molecules [6, 15]. Obviously, undesired biological effects (for example, hemolysis) of the employed CD derivatives in solution should be eliminated or minimized.

Complexes of cyclodextrins are extensively described in the literature: however, it is still a matter of debate which of the afore-mentioned contributions is mainly responsible for the overall free energy of association. In preceding papers, we have reported on the smallest cyclodextrin, α CD, having six glucose units, and its interaction with various hydroxyalkylated substances [9, 16–19], in water and in mixed solvents. The thermodynamics of the interaction of β CD, hydroxypropyl- β -cyclodextrin and methyl- β -cyclodextrin with alkanols [20, 21], in water and in mixed solvents was studied, too. The present work continues our program aimed at understanding the determining factors in the formation

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of the complexes. The interaction between β -cyclodextrin or the chemically modified methyl- β -cyclodextrin and monocarboxylic acids from C_5 to C_{12} in aqueous solution, pH 11.3, where they exist prevalingly as charged ions, will be investigated. In this way, the role of the methyl substitution on the β -cyclodextrin will be analyzed together with the effect of the length of the alkyl chain of the guest in the formation of a complex. The association process will be followed through a microcalorimetric method at 298 K. As well as the detection of the thermal effect, this technique shows whether an association process occurs, and allows the evaluation of its equilibrium constant. From that, the Gibbs energy and entropy of the process can be derived. Knowing the values and signs of the thermodynamic parameters obtained for both cyclodextrins makes possible to propose hypotheses about the forces involved in the interaction between CDs and the examined guest molecules. That can be particularly useful to design new modified cyclodextrins having more suitable characteristics to include specific drugs.

Experimental

Materials

β -cyclodextrin (β CD) and methyl- β -cyclodextrin (M β CD) were purchased from Sigma: they were both cell culture tested products. The mean substitution of the latter is 10.5–14.7 as determined by NMR.

Monocarboxylic acids were purchased from Sigma or Aldrich: their purity is 98%–99% or more. All substances were used as received, without further purification. Aqueous solutions of known molalities were prepared by mass, using doubly distilled water. For solutions at pH 11.3, a 0.5 mol kg⁻¹ NaH₂PO₄-NaOH buffer was employed. The choice of this buffer is determined by the need to avoid anions interfering with the inclusion process. It is reported in the literature that phosphate and sulfate anions satisfy this requirement in the pH range 2–11 [22]. The initial concentration of carboxylic acids varied between 8.1×10^{-2} and 3.3×10^{-4} mol kg⁻¹ passing from pentanoic to dodecanoic acid. The initial concentration of β CD varied between 1.4×10^{-2} and 8.5×10^{-4} mol kg⁻¹, while that of M β CD was between 2.9×10^{-2} and 2.6×10^{-2} mol kg⁻¹.

Calorimetry

Measurements of the experimental heats of mixing, $\Delta_{\text{mix}}H$, of two binary solutions containing any one of the solutes, were determined at 298 K using a Thermal Activity Monitor (TAM) from Thermometric, equipped with a titration vessel. A microcomputer controlled the injections and collected the titration data. Approximately 30 injections of the titrating solution were made in each experiment, and at least two experiments were performed for each substance. Enthalpies of dilution of

the added substance in water were determined, using the same number of injections and concentrations as in the titration experiments, and were subtracted from the enthalpies of the mixing process. The dilution of the component present in the cell (usually the carboxylic acid) was considered to be negligible. The estimated uncertainties in the molalities are 1% and in the heat effects 2%.

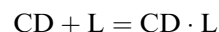
Treatment of the data

The values of the experimental heats of dilution can be used to fit the following power-law expansion: [23–25]

$$\Delta H_{\text{dil}}(m^i \rightarrow m^f) = h_{\text{xx}}m^f(m^f - m^i) + h_{\text{xxx}}m^f(m^{f2} - m^{i2}) + \dots \quad (1)$$

where ΔH_{dil} (J kg⁻¹) is the heat of dilution of a solute from the initial (m^i) to the final molality (m^f). The enthalpic interaction coefficients, h , appearing in Eq. (1) are used to determine the contributions to the total enthalpy changes originating from the dilution of each of the solutes during the mixing process.

Assuming that a 1:1 complex is formed when mixing two binary solutions, the association process can be represented as follows:



where CD indicates the cyclodextrin and L a guest (carboxylic acid) molecule. The enthalpy of formation of a complex, or in general the enthalpy of interaction between solutes, ΔH^* , is related to the heat of mixing two binary solutions, $\Delta_{\text{mix}}H$, and to the heats of dilution experienced by the two solutes, $\Delta_{\text{dil}}H$, as follows:

$$\Delta H^* = \Delta_{\text{mix}}H\{(m_{\text{CD}}^i)(m_{\text{L}}^i)\} \rightarrow (m_{\text{CD}}^f, m_{\text{L}}^f) - \Delta_{\text{dil}}H(m_{\text{CD}}^i \rightarrow m_{\text{CD}}^f) - \Delta_{\text{dil}}H(m_{\text{L}}^i \rightarrow m_{\text{L}}^f) \quad (2)$$

where m_{CD}^i , m_{L}^i , m^f , and m_{L}^f are the initial and final molalities of the cyclodextrin and carboxylic acid. ΔH^* , normalized to the total molality of the guest, m_{L} , can be related to the actual molality of the cyclodextrin host molecule, m_{CD} , to the standard molar enthalpy of association, $\Delta_a H^{\circ}$, and to the apparent association constant, K_a' , as follows [26]:

$$\Delta H^*/m_{\text{L}} = (\Delta_a H^{\circ} K_a' m_{\text{CD}}^f)/(1 + K_a' m_{\text{CD}}^f) \quad (3)$$

Equation (3) can be rewritten in a linear form, more useful for fitting the data, as follows:

$$m_{\text{L}}/\Delta H^* = 1/\Delta_a H^{\circ} + 1/(\Delta_a H^{\circ} K_a' m_{\text{CD}}^f) \quad (4)$$

For each value of ΔH^* , the actual concentration of the host molecule is given by:

$$m_{\text{CD}}^f = m_{\text{CD}} - [\Delta H^*/\Delta H^*(\text{sat})]m_{\text{L}} \quad (5)$$

where m_{L} is the total stoichiometric molality of the guest. The standard enthalpy and the constant are

Table 1. Pairwise enthalpic interaction coefficients for α -Cyclodextrin (α CD), β -cyclodextrin (β CD), and methyl- β -cyclodextrin (M β CD) in water and in phosphate buffer 0.5 mol kg⁻¹, pH 11.3, at 298 K.

Substance	h_{xx}^a	Solvent medium
α CD	-3920 ± 65^b	water
α CD	1365 ± 80^c	buffer
β CD	-2754 ± 200^d	water
β CD	4202 ± 142	buffer
M β CD	28412 ± 470	water
M β CD	67569 ± 1200	buffer

Errors reported are the 95% confidence limits. ^a J kg mol⁻². ^b Ref. 9. ^c Ref.18. ^d Ref.20.

obtained from Eqs. (4) and (5) by an iterative least square method. The iterations are continued until two successive values of $\Delta_a H^{\circ'}$ differ by less than 2%. The values of the free energy and entropy are then obtained through the usual thermodynamic relations. The absence of any information about the activity coefficients leads to the evaluation of association parameters that are thermodynamically not exactly defined. Only an apparent constant, K'_a , can be determined, and consequently the standard Gibbs energy and entropy, $\Delta_a G^{\circ'}$ and $\Delta_a S^{\circ'}$, suffer of the same limitations.

If the simple association model discussed above does not describe the studied systems, using Eqs. (4) and (5) gives thermodynamic parameters that lead to a large discrepancy between calculated and experimental curves.

Results

In Table 1, the enthalpic self-interaction coefficients are reported for methyl- β -cyclodextrin (M β CD) in water and in phosphate buffer 0.5 mol kg⁻¹, pH 11.3: coefficients are positive and very large in both solvents. In the same Table coefficients for the natural α - and β -cyclodextrins are shown: they are negative in water and positive in buffer.

In Table 2, the association constant, enthalpy, Gibbs energy and entropy are reported for the interaction of

the natural β CD with monocarboxylic acids from C₅ to C₁₂ in phosphate buffer, pH 11.3. The association parameters have been obtained through Equations (4) and (5), under the hypothesis that only one type of adduct forms with a 1:1 stoichiometry. At increasing length of the alkyl chain, association enthalpies pass from positive (+1.9 kJ mol⁻¹ for pentanoic acid) to negative, starting with nonanoic acid (-0.75 kJ mol⁻¹). It is not possible to evaluate the association parameters for hexanoic to octanoic acid, being the association heat very small, almost null, hence not detectable. Constants increase at increasing length of the alkyl chain up to decanoic acid, and association is driven mainly by the relevant positive entropic contribution. Passing to undecanoic acid, a jump is noted in both enthalpy and entropy. Enthalpy becomes large and negative and entropy almost zero: that makes the association constant to decrease. However, it increases again for dodecanoic acid mainly because of the relevant favorable enthalpic contribution.

The interaction between β -cyclodextrin and several carboxylic acids has been studied previously by other authors using mainly calorimetry [7]. and potentiometry [27]. The data reported are rather different from those given in the present work probably because of the different experimental conditions here employed. For instance, for hexanoic, octanoic and decanoic acids, measurements reported in the literature were carried out in water [27], namely around pH 6, where the equilibrium exists between the charged and uncharged forms of the acid. The same occurs for pentanoic and heptanoic acids studied at pH 6.9 [7]. On the contrary, here we work at high pH, so that equilibrium is heavily shifted toward the charged form. The charged and uncharged forms of carboxylic acids behave very differently towards the interaction with a cyclodextrin, [16]. hence it is important to control the pH for only one of the two forms be predominant.

In Table 3, the thermodynamic association parameters are reported for the interaction between M β CD and the same carboxylic acids, in phosphate buffer, pH 11.3. Enthalpies are positive, decreasing at increasing alkyl chain length. Constants increase: in correspondence of

Table 2. Thermodynamic parameters for the association between β -cyclodextrin and monocarboxylic acids, in Na₂HPO₄ 0.5 mol kg⁻¹, pH 11.3, at 298 K.

Acid	$K^{a,b}$	$\Delta_a H^{\circ'}$ ^{b,c}	$-\Delta_a G^{\circ'}$ ^{c,d}	$T\Delta_a S^{\circ'}$ ^{c,e}
Pentanoic	$(3.1 \pm 0.7) 10$	1.9 ± 0.2	8.5 ± 0.6	10.4 ± 0.8
Hexanoic		ND ^f		
Heptanoic		ND ^f		
Octanoic		ND ^f		
Nonanoic ¹	$(2.6 \pm 0.6) 10^2$	-0.75 ± 0.08	13.7 ± 0.6	13.0 ± 0.7
Decanoic	$(1.9 \pm 0.1) 10^3$	-2.5 ± 0.1	18.6 ± 0.2	16.1 ± 0.3
Undecanoic	$(5 \pm 1) 10^2$	-13 ± 2	15.1 ± 0.7	2 ± 3
Dodecanoic	$(2.4 \pm 0.5) 10^3$	-21.1 ± 0.6	19.3 ± 0.5	-2 ± 1

^a kg/mol. ^b Errors reported are the standard deviations as obtained by fitting the data to Eqs. (4) and (5). ^c kJ/mol. ^d Errors are half the range of $\Delta_a G^{\circ'}$ calculated from the upper and lower error in K'_a . ^e Errors are the sum of the errors on free energy and enthalpy. ^f ND means that association heat is almost null, not detectable: the mixing enthalpy is similar to the dilution enthalpy.

Table 3. Thermodynamic parameters for the association between methyl- β -cyclodextrin and monocarboxylic acids, in Na_2HPO_4 0.5 mol kg^{-1} , pH 11.3, at 298 K.

Acid	K'_a ^{a,b}	$\Delta_a H^{\circ}$ ^{b,c}	$-\Delta_a G^{\circ}$ ^{c,d}	$T\Delta_a S^{\circ}$ ^{c,e}
Pentanoic		NA ^f		
Hexanoic	$(3.4 \pm 0.9) 10$	11 ± 2	8.7 ± 0.7	20 ± 3
Heptanoic	$(2.7 \pm 0.1) 10^2$	7.8 ± 0.1	13.9 ± 0.1	21.7 ± 0.2
Octanoic	$(1.26 \pm 0.03) 10^3$	6.8 ± 0.1	17.7 ± 0.1	24.5 ± 0.2
Nonanoic	$(1.0 \pm 0.1) 10^4$	5.4 ± 0.1	22.9 ± 0.4	28.3 ± 0.5
Decanoic	$(1.2 \pm 0.2) 10^4$	4.6 ± 0.1	23.2 ± 0.4	27.8 ± 0.4
Undecanoic	$(1.5 \pm 0.7) 10^4$	2.6 ± 0.1	24 ± 1	26 ± 1
Dodecanoic	$(1.7 \pm 0.9) 10^4$	0.7 ± 0.1	24 ± 1	25 ± 1

^a kg/mol. ^b Errors reported are the standard deviations as obtained by fitting the data to Eqs. (4) and (5). ^c kJ/mol. ^d Errors are half the range of $\Delta_a G^{\circ}$ calculated from the upper and lower error in K'_a . ^e Errors are the sum of the errors on free energy and enthalpy. ^f Means that measurements have been performed, but association does not occur.

nonanoic acid a jump toward high values is noted, prevalingly determined by the positive and large entropic term. For pentanoic acid, it is not possible to derive the association constant for the system behaves as a non-associating one.

Figure 1 shows the complete thermodynamic framework for the association of monocarboxylic acids with the natural and methylated cyclodextrins.

Discussion

The enthalpic self-interaction coefficient for $M\beta CD$ determined in water is about 2.8×10^4 J kg mol^{-2} , while for the natural βCD it is much smaller and negative, about -2.7×10^3 J kg mol^{-2} . The coefficient for the former cyclodextrin is among the highest reported in the literature: it accounts for the relaxation of many structured water molecules from the cavity and from the hydrophobic hydration shells of the external methyl groups when two hydrated molecules approach each other. Instead, the coefficient for βCD is negative, but

less negative than that for αCD [9], thus indicating that the larger macrocycle is a structure breaker less effective than the smaller one. It is reported that αCD undergoes a tense \rightarrow relaxed transition upon the formation of a complex [28]. On the contrary, βCD presents a relaxed conformation even in the absence of a guest, with the large cavity more exposed to bulk solvent. This behaviour in water is different from that of other saccharides, which are characterized by positive coefficients, and similar to that of some higher members of polyols, whose coefficients change sign depending on the stereochemistry of the solute [29]. In that case, the hypothesis was that the separation of the hydrophilic and hydrophobic domains in the solute molecule determines sign and values of the pairwise enthalpic interaction coefficients. For α - and β -cyclodextrin in water, the sign and magnitude of the coefficients are probably a consequence of being molecules hydrophilic at the exterior and hydrophobic inside. The pairwise enthalpic interaction coefficient should be composed of two contributions, one originating from the overlapping of the hydrated hydrophilic exterior and the other one

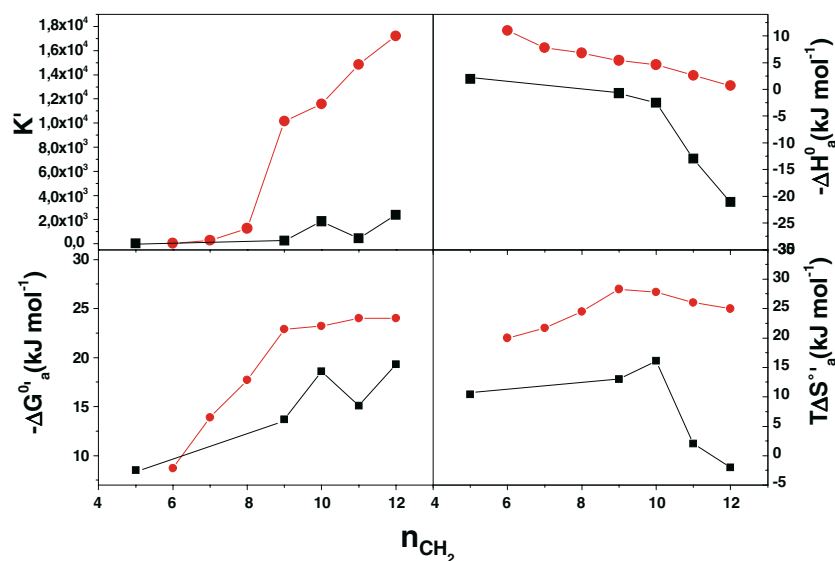


Figure 1. Thermodynamic association parameters – enthalpy, Gibbs free energy, constant and entropy- vs the number of carbon atoms in the alkyl chain, n_C , for monocarboxylic acids interacting with β -cyclodextrin (■) and methyl- β -cyclodextrin (●) at 298 K.

from the displacement of structured water present in the hydrophobic cavity. The former contribution is negative for the prevalence of hydrophilic-hydrophilic interactions, while the latter is positive. For α CD, the negative contribution is prevailing, while for β CD the positive contribution makes less negative the pairwise interaction coefficient notwithstanding the larger size of the macrocycle. For the methylated β -cyclodextrin, the exterior is prevalently hydrophobic: then, the interaction between two hydrated molecules is characterized by a pairwise coefficient composed of two positive contributions which determine its large value.

In phosphate buffer, the coefficient for $M\beta$ CD becomes even larger, about $6.8 \times 10^4 \text{ J kg mol}^{-2}$, while those for both the natural cyclodextrins pass from negative to positive. That reproduces the behaviour shown by glycine, formamide and other hydrophilic solutes, whose pair enthalpic interaction coefficients become positive or less negative in the presence of urea [30]. According to preceding studies on ternary aqueous solutions of hydroxylated substances, urea interacts mainly with hydrophilic domains, as the hydroxyl groups, and the consequent steric hindrance leads to attenuated hydrophilic interactions, thus reducing the exothermic contribution. The positive coefficients of the natural cyclodextrins, and the increased coefficient for $M\beta$ CD are an indication that phosphate ions behave as urea. The large and positive value of the pairwise enthalpic coefficient for methyl- β -cyclodextrin makes to provide that the interaction with an alkylated substance is a process determined mainly by hydrophobic interactions, hence an entropy-driven process.

According to the commonly accepted view, complexation of an alkylated guest molecule with a cyclodextrin occurs through the inclusion of the alkyl chain into the prevalently hydrophobic cavity. The functional group forms hydrogen bonds with the external hydroxyl groups on the rim of the macrocycle cavity, thus preventing the further penetration of the alkyl chain [17]. The present data relative to the interaction between the natural or modified β -cyclodextrin and monocarboxylic acids as guests show that association occurs through the same mechanism. However, the presence of the external methyl groups in methyl- β -cyclodextrin cause the balance of the forces upon association to be different from that acting when the natural cyclodextrin is involved.

In the association between the natural β CD and monocarboxylic acids, enthalpies are positive or almost zero for the first terms of the series. The positive value of enthalpy would result from several contributions, among them the endothermic contribution due to the disruption of hydrogen bonds between water molecules in the cavity, the endothermic dehydration of the including hydrophobic guest molecule, and the exothermic contribution stemming from the van der Waals interactions between the cyclodextrin cavity and the guest. The cavity diameter of β -cyclodextrin is approximately 6.2 \AA [1]: that means a loose adaptation of an alkyl chain upon the formation of a complex, and

consequently a small negative contribution from the interactions with the cavity. That effect is particularly evident for the lower-molecular-mass acids as, for instance, pentanoic acid whose association is characterized by a small constant and by a positive and small enthalpic term. As can be seen from Table 2, only for an alkyl chain longer than eight carbon atoms the exothermic contribution of the binding interactions with the cavity overcomes all other endothermic contributions. For hexanoic to octanoic acids, thermodynamic parameters cannot be evaluated because the association heat is almost null. That is, the endothermic contributions balance the exothermic contributions due to the ameliorated interactions of the guest with the cavity. Enthalpy becomes negative starting with nonanoic acid and then there is a jump passing from decanoic to undecanoic acid (from -2.5 to -13 kJ mol^{-1}). A discontinuity occurs in the constant, too, being this parameter for undecanoic acid much smaller than that for decanoic acid: after that, constant increases again for dodecanoic acid. Entropy, large and positive for pentanoic acid, increases at increasing alkyl chain length. Upon inclusion water molecules relax from an ordered microenvironment, namely the cavity, and from the ordered hydrophobic hydration shells of the guests to a more disordered bulk. The β CD cavity is rather large towards the inclusion of an alkyl chain that needs to be dehydrated only partially to be included. Hence, the contribution due to the dehydration of the guest is small, and the entropic term should be mainly determined by the relaxation of water molecules from the cavity, as it occurs in the case of alkanols, too [20]. Also for entropy a sudden change occurs, passing from strongly positive for decanoic acid (16.1 kJ mol^{-1}) to almost zero for undecanoic acid ($2 \pm 3 \text{ kJ mol}^{-1}$) and becoming negative for dodecanoic acid. These association data have been obtained imposing that a simple 1:1 model holds that postulates the presence of only one adduct. However, on the basis of the jump in the enthalpies in correspondence of undecanoic acid, together with the irregular variation in the association constants and the sudden decrease in entropy, for longer alkyl chains it is necessary to assume a more complex model to rationalize the data obtained. The hypothesis is that only one adduct forms for alkyl chains formed by about ten CH_2 groups. For longer terms, there must be a change in the interaction mechanism. One model could provide one guest molecule interacting with two β CD; then, 1:2 complexes could form in addition to the 1:1 complexes. This is a reasonable hypothesis, but it does not hold here, because it provides a complexation occurring through the penetration of the charged carboxyl group in one of the two cavities, an event excluded by all data reported in the literature. In preceding studies, instead, we found that formation of 2:1 complexes is possible only for the uncharged form of the carboxylic group [16, 31]. An alternative model might take into consideration that long chains can pass through the cyclodextrin ring and protrude from the other side. The protruding part

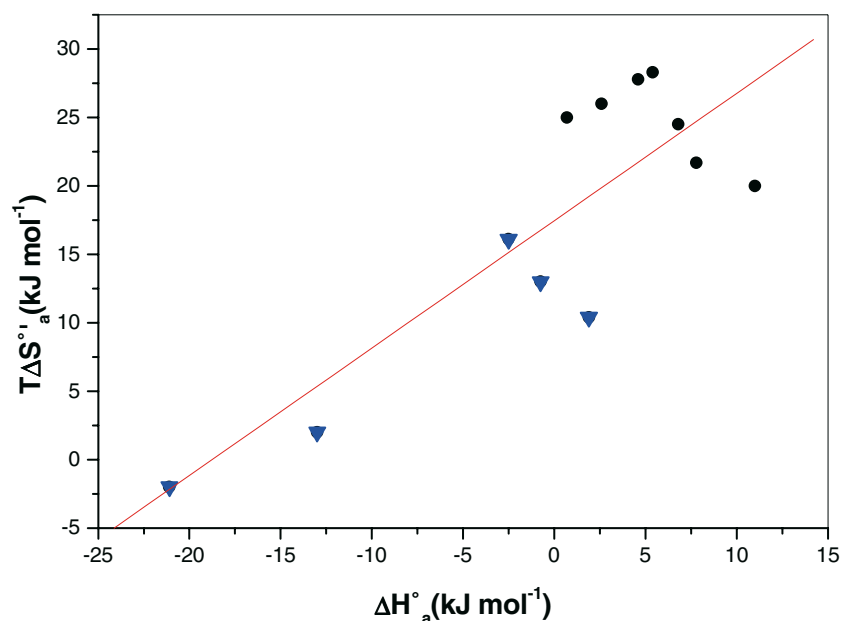


Figure 2. Enthalpy-entropy compensation plot for the formation of complexes between β -cyclodextrin (▼) or methyl- β -cyclodextrin (●) and the monocarboxylic acids reported in the present work.

would, then, be caged by water molecules. However, it would not account for the increasing enthalpies at increasing alkyl chain length. Enthalpy, in fact, should remain almost constant since the negative contribution for the formation of the water cage would be balanced by the positive contribution originating from the dehydration of the guest molecule when passing through the cyclodextrin. How an alkyl chain longer than the height of the cavity can, then, be included? It is useful to remember that hydroxypropyl- β -cyclodextrin forms complexes with various cycloalkanols through the inclusion of the ring into the cavity [21]. Beyond that, the rather strong association of β - and methyl- β -cyclodextrin with 4-cyclohexylbutan-1-ol, [21], is an evidence that a long alkyl chain can bend in the cavity of a β -cyclodextrin. Then, the present data could be explained according to the hypothesis that, because of the flexibility of the alkyl chain, two adducts form simultaneously, bearing the included alkyl residue in a bent or in an extended form, whose concentration ratios depend on the alkyl chain length. For alkyl chains shorter than ten or longer than eleven carbon atoms, only one adduct would be present, in the extended or bent form, respectively, while for intermediate alkyl chains, both adducts would form. This model has been already proposed for α -cyclodextrin interacting with monocarboxylic acids at the same pH used here [32]. There, it was inferred that for alkyl chains shorter than eight or longer than ten carbon atoms only one adduct is present having the included chain in the extended or bent form, respectively. Hence, going from the smaller to the wider cyclodextrin, a shift occurs in the term up to which only the adduct bearing the extended alkyl chain forms, passing from C₈ to C₁₀. Since the height of the cavity is the same for both cyclodextrins, then the determining factor should be the larger width of the

cavity. In the β CD cavity, the alkyl chain has more space to bend with less strain, so that a longer alkyl chain is necessary to detect the jump in the thermodynamic parameters, primarily enthalpy, that reflect the variation in the forces governing the association.

Methyl- β -cyclodextrin has a mean substitution degree of 10–14, meaning that, in the average, 10–14 methyl groups substitute 10 hydrogen atoms. As expected on the basis of the positive and large enthalpic pairwise interaction coefficient, the parameters characterizing the association with monocarboxylic acids indicate that hydrophobic interactions are mainly responsible for the complex formation, being the enthalpies positive and the entropies positive and much larger than those relative to the interaction of β CD with the same substances. Enthalpies become less positive at increasing alkyl chain length, reaching a very small value for dodecanoic acid (0.7 kJ mol⁻¹). This trend is dictated by two effects: the dehydration of the external methyl groups and the alkyl chain of the guest, an endothermic process, and the formation of van der Waals interactions between the included alkyl chain and the cavity, an exothermic process. The increasingly stronger interactions of the alkyl chain with the cavity determine enthalpies becoming increasingly less positive. For what concerns entropy, the very large and positive contribution is due to an additional effect, peculiar of this modified cyclodextrin, namely the relaxation to the bulk of water molecules from the hydrophobic hydration shells of the external methyl groups upon inclusion. This contribution can be assumed to be the predominant one, being entropy large and almost invariant at increasing alkyl chain length. Acids having an alkyl chain shorter than seven carbon atoms form weak complexes characterized by small association constants, that are, probably, mainly determined by the interaction of the alkyl chain

with the hydrophobic external methyl groups. As respect to the natural β -cyclodextrin there is no jump in the values of enthalpies, suggesting that, up to the C₁₂ term, only one type of adduct forms, the one having the guest in its extended form. Then, the methyl substitution makes the cavity to behave as it were deeper than that characterizing the natural cyclodextrin, and longer alkyl chains are required to fit it in their extended form. Even longer alkyl chains would be needed for the formation of the second adduct, detected by a jump in the thermodynamic properties.

Finally, it is interesting to compare the processes described here with the association of the smallest cyclodextrin, α CD, with alkylated substances. The internal diameter of α CD (about 4.9 Å) is much smaller than that of β CD (about 6.2 Å), so the shape-matching between the host cavity and the guest inserting group is the major factor determining the association between the interacting molecules. The association is ruled prevalently by enthalpy, while entropy becomes negative at increasing alkyl chain length, an indication of the prevalence of van der Waals interactions [16, 17]. between the alkyl chain and the cavity.

Conclusions

The formation of a complex between a cyclodextrin and a prevalently hydrophobic guest molecule is a process ruled by the changes experienced by the solvent water upon association: dehydration of the guest molecule, desolvation of the cavity, formation of a hydration shell for the complex. For the methylated cyclodextrin, the relaxation of water molecules to the bulk, as a consequence of the interaction of the alkylated guest molecule with the hydrophobic exterior, is the major effect that determines the values of the association constants.

For most complexes reported in the literature, a roughly linear trend is obtained when reporting ΔH° vs ΔS° : this compensatory enthalpy-entropy relationship exists [1–4, 16, 33–35]. for all processes dominated by aquation phenomena and ascribed to the modifications experienced by the solvent in the hydration cospheres of the interacting substances. Another quantitative correlation of the enthalpy-entropy compensation is performed by plotting $T\Delta S^\circ$ vs ΔH° [35,36]. For a large number of complexes, the trend is linear. As shown in Fig. 2, it is very hard to recognize even a trend in such scattered plot, as usually occurs when a limited number of data is used. However, it is very descriptive of the processes examined. In fact, the first five points refer to the systems involving β CD, with a jump passing from undecanoic and dodecanoic acids to the smaller ones, a consequence of the probable change of inclusion characteristics. The last six points, at high positive enthalpy and entropy, refer to M β CD. If a line is forced to pass through all points (correlation coefficient = 0.84), the $T\Delta S^\circ$, at $\Delta H^\circ = 0$, and slope are 17 ± 2 kJ mol⁻¹ and

0.9 ± 0.2 , respectively, values not far from those reported in the literature for complexes formed by modified cyclodextrins with flexible sidearms [1]. Accordingly, the slope indicates to what extent the enthalpic gain is cancelled by entropic loss, while the positive intercept suggests that the complex is stabilized even in the absence of enthalpic contributions. The large entropic term confirms the conclusions already drawn: the association process is determined mainly by the changes experienced by water in the hydration shells of the interacting hydrated molecules.

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