Journal of Inclusion Phenomena and Molecular Recognition in Chemistry 9: 277–285, 1990.

Complexation of Aliphatic Dicarboxylic Acids and Anions by alpha-Cyclodextrin

ANTHONY AVERSA, WAYNE ETTER, ROBERT I. GELB, and LOWELL M. SCHWARTZ*
Department of Chemistry, University of Massachusetts, Boston, Massachusetts 02125, U.S.A.

(Received 4 March 1989; in final form: 24 April 1989)

Abstract. Inclusion complexes are formed in aqueous solution between α-cyclodextrin and several straight-chain alkanedioic acids (ethanedioic, propanedioic, butanedioic, pentanedioic, hexanedioic, heptanedioic and octanedioic acids), several corresponding anions, several straight-chain alkenedioic acids (*cis*-butenedioic, *trans*-butenedioic and *t,t*-2,4-hexadienedioic acids) and several corresponding anions. Formation constants for these complexes were determined by measuring the effect of complexation on the pH of cyclohexaamylose/acid/base buffer equilibria. Enthalpies and entropies of complexation were calculated from the temperature dependences of the formation constants. The observed trends in the thermodynamic parameters lead to hypotheses about the structures of the complexes.

Key words. α-Cyclodextrin, aliphatic acids, aqueous solution, thermodynamic parameters.

1. Introduction

In this communication we report the results of experiments to measure the complex formation constants of inclusion complexes of a number of straight-chain aliphatic dicarboxylic acid and anion substrates by α-cyclodextrin (cyclohexaamylose) in aqueous solution. By measuring these complexation constants over a range of temperatures we determine the standard enthalpies and entropies of complexation and these results shed light on the reaction mechanisms. Our experimental methodology uses a modification of a pH potentiometric technique which we have reported previously [1]. The earlier work was restricted to a chemical system involving a solution of a monoprotic acid/base conjugate pair, which we now denote by the generic symbolism HB/B, and a complexation agent which we denote by Cy. The HB/B pair constitute a buffer solution whose pH depends on the HB acid dissociation constant and on the relative concentrations of the two conjugate species. Cy added to this solution complexes either HB or B or both and these complexations perturb the relative concentrations of uncomplexed HB and B and thus perturb the solution pH. By adding portions of solid Cy to an initial known HB/B buffer mixture (a procedure which we shall call 'titrating' with Cy), we generate a set of pH vs. solution composition data which can be analyzed by an elaborate nonlinear regression calculation to yield equilibrium constants and their standard error estimates for all complexes that form between the buffer components and the cyclodextrin.

While the previously reported methodology [1] is restricted to monoprotic HB/B

* Author for correspondence.

equilibria, we now extend the technique to include diprotic acid species. A generic diprotic acid is now denoted by H_2B and the conjugate successively deprotonated species by HB and B. For a typical uncharged dicarboxylic acid H_2B, the mono- and dianions are HB^- and B^{2-}, respectively, but we shall omit charge designations here to simplify the symbolism. From experience we know that cyclodextrin can form both binary and ternary complexes with its substrates and so we write the binary complexes as H_2BCy or HBCy or BCy and the ternary complexes as H_2BCy_2, etc. The nonlinear regression calculation is based on a simultaneous set of model equations which describe the physical and chemical properties of the solution during titration with cyclodextrin. Model equations for the monoprotic HB/B system are given in detail elsewhere [1] so that it shall suffice here to describe those equations as modified for the diprotic H_2B/HB/B system:

1. A conservation relation equates the sum of molar concentrations of all species H_2B, HB, B and their complexes to the analytical concentration of the H_2B/HB/B system.
2. A conservation relation equates the sum of molar concentrations of free Cy and of complexes (duly weighted for Cy stoichiometry) to the analytical concentration of cyclodextrin.
3. A charge balance equates the sum of all duly weighted concentrations of positively charged species including H^+ and Na^+ from NaHB and Na_2B salts to those of negatively charged species including OH^-.
4. Thermodynamic equilibrium constant expressions in terms of molar concentrations and molar activity coefficients describe the buffer and complexation equilibria. The symbols for these equilibrium constants are as follows:

   K_{a1} and K_{a2} for the first and second acidic dissociation of H_2B, respectively,

   K_{H_2BCy} for a typical binary complexation $H_2B + Cy \rightarrow H_2BCy$

   $K_{H_2BCy_2}$ for a typical stepwise ternary complexation $H_2BCy + Cy \rightarrow H_2BCy_2$

   K_w for the autoprotolysis of water.

5. Ionic activity coefficients are estimated from solution ionic strength using the Debye–Hückel correlation with temperature-dependent parameters from the Robinson and Stokes [2] tabulation. We assume values for ion size parameters as follows: a range from 0.4 to 0.9 nm for uncomplexed anions depending on the length of the carbon chain; 0.9 nm for H^+; 0.35 nm for OH^-; 1.6 nm for binary complexes and 1.8 nm for ternary complexes. Activity coefficients of uncharged species are taken as unity.
6. A small correction is applied to the measured pH because of the presence of substantial concentrations of cyclodextrin. These corrections were determined as a function of Cy concentration by measuring pH values in dilute HCl solutions as Cy was added. Another small correction is applied to the volume of solutions to compensate for the density changes as Cy is added.

Within this set of model equations we regard a number of quantities as adjustable parameters whose values are changed by the nonlinear regression in an effort to minimize the sum of squared deviations between recorded titration data and the

titration values calculated from the model equations [3]. These adjustable parameters are the equilibrium constants K_{a1} and K_{a2} and all complex formation constants. While it is not necessary to assume the existence of all possible binary and ternary complexes with all substrate species (a total of eight adjustable parameters) in all titrations in order to realize an adequate fit of the model equations to the recorded data, we have found it necessary to require as many as seven adjustable parameters in some cases. Our experience has shown that when several parameters are required in these diprotic systems, even very precise experimental data from the titration of a single buffer solution does not adequately define all the parameters. We have overcome this difficulty by merging the data from two separate titrations into a single regression analysis. One titration is made of a solution in which H_2B and HB and their complexes are the dominant species and a second titration is made in which HB and B are the dominant species. In the first titration B and its complexes are relatively minor components and in the second titration H_2B and its complexes are minor but all species are retained in the model equations describing both titrations.

2. Experimental and Data Analysis Methodology

Samples of cyclohexaamylose obtained from Aldrich Chemical Co. were equilibrated with the ambient atmosphere for at least one week and then analyzed as the hexahydrate by prolonged drying *in vacuo* at 100°C. Reagent grade samples of dicarboxylic acids were used without further purification.

Potentiometric measurements were made using an Orion Model 801 pH meter fitted with conventional glass and reference electrodes. This system was carefully equilibrated at each measurement temperature and then calibrated with 0.05 M KH_2PO_4 buffer whose temperature-dependent pH values were taken from Bates [4].

Solutions were prepared from weighed samples of the dicarboxylic acid H_2B at analytical concentrations typically 1.0–4.0 mM. Appropriate volumes of standardized NaOH were added to prepare the two buffer solutions for titration with Cy. Typically one such solution was approximately 1 mol NaOH per 2 mol H_2B and the other approximately 3 mol NaOH per 2 mol H_2B. However, in the cases of oxalic (ethanedioic) and maleic (*cis*-butenedioic) acids, which are highly dissociated in dilute solution, the low pH buffer was prepared with about 0.02 M H_2B and with no added NaOH.

In a typical experiment one of the buffer solutions was equilibrated with the electrodes for at least one hour. The titration with Cy involved adding small weighed portions of the solid hexahydrate and waiting for equilibration before recording the pH. The solid addition process was repeated eight to twelve times such that the analytical concentration of Cy in the solution increased from 0 to about 0.04 M. The thermostatted cell was scrupulously cleaned before the second buffer solution was added and the titration repeated in the same manner as with the first.

Typically the two titrations combined yielded about 20 recorded pH measurements and this constitutes an overdetermined set for a system of model equations with at most eight unknown parameters. The computerized nonlinear regression procedure for adjusting parameters to achieve a weighted least-squares fit of model equations to experimental data has been described elsewhere [3]. The computer

program allows us to fix the values of any subset of the unknown parameters while allowing the other parameters to vary in seeking the minimum sum-of-squared residuals. We take advantage of this feature to find the minimum number of parameters necessary to achieve a satisfactory fit as follows: we start by fixing all complexation constants at zero save one for a binary complex and then run the regression by adjusting only this one binary constant and K_{a1} and K_{a2}. This configuration effectively hypothesizes that only a single binary complex is formed in the two titrations. In all cases studied to date this hypothesis yields an unsatisfactory fit in that the minimum sum-of-squared residuals far exceeds what is expected from the random errors in the experimental data and the sequential pattern of these residuals is nonrandom. Then in an interactive manner we successively add other formation constants, first binaries and then ternaries, to the list of those that are adjustable. At each stage in this process the newly added parameter reduces to some extent the minimum sum-of-squared residuals. Nevertheless, we examine the adjusted value of this parameter and its standard error estimate. In some instances the value of the added parameter is within its statistical uncertainty of zero and in these cases we conclude that only a negligible concentration of the corresponding complex may exist in either solution. In these instances we then reset the value of this parameter to zero and delete it from the list of adjustable parameters for later stages in the process. The process reaches a satisfactory conclusion when the minimum sum-of-squared residuals reduces to the expected level and the sequential pattern of residuals is random. When a subset of adjustable parameters yields this satisfactory outcome, any additional parameter added to that subset turns out to have a value within statistical scatter of zero.

3. Results and Discussion

The results of this process leads to the complex formation constants given in Table I for alkanedioic substrates and in Table II for a few alkenedioic substrates. For each system at each temperature we are convinced that the set of complexation constants shown is the appropriate set in the sense that (1) each complexation constant is statistically greater than zero, that (2) any binary or ternary complexation constant not shown in the Table has been adjusted and found to be within statistical uncertainty of zero, that (3) the omission of any of the complexes shown in the Table leads to an unsatisfactory fit of the model equations to the data. We note here that the longest dicarboxylic acid in Table I is octanedioic acid. We actually attempted to study the longer acids nonanedioic and decanedioic but failed to achieve satisfactory regression fits to the data in both cases even when adjusting all eight parameters. It is possible that dimeric or other aggregate species are formed in these solutions which invalidate the chemistry assumed in the model equations.

By fitting least-squares van't Hoff lines to each equilibrium constant as a function of temperature we calculated standard enthalpies and entropies of complexation which are listed in Table III. The standard error estimates quoted for the thermodynamic parameters in these tables are based on the scatter of the individual points from the least-squares lines.

The results in Tables I and II reveal several general patterns of behavior in the

Table I. Complexation formation constants of α-cyclodextrin with alkanedioic acids and anions.

t, °C	Complex stoichiometry			
	BCy	HBCy	H_2BCy	H_2BCy_2[a]
		Oxalic (ethanedioic) acid		
15		2.4 ± 0.6[b]	35 ± 3	
20		2.6 ± 0.3	30 ± 3	
25		2.4 ± 0.3	25 ± 3	
30		2.2 ± 0.3	18 ± 2	
40		2.7 ± 0.2	14 ± 2	
		Malonic (propanedioic) acid		
15		46.5 ± 1.2	21.8 ± 1.2	
25		32.3 ± 1.0	15.6 ± 1.1	
30		27.5 ± 0.9	13.5 ± 1.1	
40		19.7 ± 0.8	10.0 ± 0.8	
50		13.7 ± 0.7	7.4 ± 0.9	
		Succinic (butanedioic) acid		
15		25.8 ± 0.3	153.7 ± 1.7	
25		17.6 ± 0.1	101.3 ± 0.9	
30		14.0 ± 0.1	79.6 ± 0.5	
40		9.8 ± 0.3	50.4 ± 0.7	
50		6.9 ± 0.4	35.7 ± 0.9	
		Glutaric (pentanedioic) acid		
20		65.7 ± 0.8	379 ± 5	
30		42.5 ± 0.8	238 ± 4	
40		26.5 ± 0.1	142 ± 3	
50		18.2 ± 0.1	93 ± 1	
		Adipic (hexanedioic) acid		
15		229 ± 1	517 ± 4	
25		152 ± 2	322 ± 3	
30		112 ± 1	239 ± 2	
40		77 ± 1	158 ± 2	
50		57 ± 1	110 ± 2	
		Pimelic (heptanedioic) acid		
15	32.1 ± 0.1	605 ± 10	1184 ± 33	4.6 ± 2.0
25	26.8 ± 0.1	395 ± 4	738 ± 17	2.9 ± 1.0
35	21.8 ± 2.1	260 ± 11	467 ± 17	2.5 ± 0.7
45	18.0 ± 1.6	174 ± 7	297 ± 11	2.1 ± 0.7
		Suberic (octanedioic) acid		
15	193 ± 7	1447 ± 41	2162 ± 63	195 ± 5
25	163 ± 3	986 ± 9	1419 ± 27	119 ± 2
35	128 ± 5	605 ± 20	873 ± 26	58 ± 2
45	100 ± 2	401 ± 8	560 ± 15	31 ± 1

[a] The formation constants $K_{H_2}BCy_2$ are for the stepwise reaction

$$Cy + H_2B{\cdot}Cy \rightarrow H_2B{\cdot}Cy_2.$$

[b] Uncertainties are standard error estimates from the nonlinear regression calculation.

Table II. Complexation formation constants of α-cyclodextrin with alkenedioic acids and anions.

t, °C	Complex stoichiometry		
	BCy	HBCy	H_2BCy
	Maleic (*cis*-butenedioic) acid		
20		76.4 ± 1.0[a]	24.7 ± 1.2
25		67.5 ± 1.3	25.3 ± 1.3
35		44.3 ± 1.0	16.6 ± 1.5
45		28.0 ± 0.1	12.1 ± 1.0
50		22.3 ± 0.8	8.3 ± 1.1
	Fumaric (*trans*-butenedioic) acid		
15		200 ± 1	3682 ± 2
25		148 ± 1	1731 ± 19
30		95.9 ± 0.1	1572 ± 5
40		60.1 ± 0.1	909 ± 5
50		40.0 ± 0.5	562 ± 5
	Muconic (*t,t*-2,4-hexadienedioic) acid		
15	86 ± 3	820 ± 1	1119 ± 26
25	67 ± 2	562 ± 9	767 ± 1
35	57 ± 1	387 ± 4	481 ± 17
45	41 ± 1	261 ± 2	334 ± 13

[a] Uncertainties are standard error estimates from the nonlinear regression calculation.

complex strength: (1) in a given substrate system at a given temperature the complex strength increases as the substrate charge increases from −2 in B to zero in H_2B; (2) in any homologous series of alkanedioic substrates (Table I) the strength of the complex increases with increasing carbon chain length at a given temperature; and (3) the strength of any given complex decreases with increasing temperature. Exceptions to pattern (1) occur with malonic acid in Table I and with maleic acid in Table II. In these two systems the Cy/monoanion complex is stronger than the corresponding dicarboxylic acid complex at each measurement temperature. Also we see that malonic acid and hydrogen malonate are out of line with the general patterns (2). Molecular malonic acid is anomalously weak and hydrogen malonate ion is anomalously strong relative to their positions in the sequence of chain lengths. Both malonic and maleic acids feature unusually strong intermolecular hydrogen bonding interactions, particularly in the monoanions, and this property may relate to the anomalous Cy complex strengths observed here. The only exception to pattern (3) appears to occur with the extremely weak Cy/hydrogen oxalate complex where the strength seems independent of temperature.

The general trend of increasing complex strength of the longer alkanedioic acids and monoanions seems to parallel the same behavior of alkane monocarboxylic acids [5]. However, the thermodynamic parameters ΔH^0 and ΔS^0 derived from the temperature-dependences of the complexation constants indicate somewhat different explanations for these trends. In Table III we see that for both the dicarboxylic

Table III. Standard enthalpies and entropies of complexation of α-cyclodextrin with aliphatic dicarboxylic substrates.

Substrate	ΔH^0, kcal mol^{-1} complex				ΔS^0, cal mol^{-1} K^{-1} complex			
	BCy	HBCy	H_2BCy	H_2BCy_2	BCy	HBCy	H_2BCy	H_2BCy_2
				Alkanedioic systems				
oxalic		0.3 ± 0.9[a]	-6.8 ± 0.6			3 ± 3	-16 ± 2	
malonic		-6.4 ± 0.1	-5.65 ± 0.05			-14.6 ± 0.5	-13.5 ± 0.2	
succinic		-7.01 ± 0.09	-7.9 ± 0.2			-17.9 ± 0.3	-17.3 ± 0.6	
glutaric		-8.1 ± 0.2	-8.9 ± 0.2			-19.5 ± 0.6	-18.5 ± 0.6	
adipic		-7.5 ± 0.3	-8.3 ± 0.3			-15.2 ± 1.1	-16.2 ± 0.8	
pimelic	-3.5 ± 0.1	-7.57 ± 0.09	-8.38 ± 0.11	-4.5 ± 0.08	-5.4 ± 0.4	-13.6 ± 0.3	-15.0 ± 0.4	-13 ± 3
suberic	-4.0 ± 0.3	-7.9 ± 0.3	-8.3 ± 0.3	-11.3 ± 0.7	-3.3 ± 1.0	-12.9 ± 1.1	-13.4 ± 0.9	-29 ± 2
				Alkenedioic systems				
fumaric		-8.55 ± 0.07	-9.97 ± 0.07			-19.1 ± 0.2	-18.3 ± 0.2	
maleic		-7.9 ± 0.4	-6.8 ± 1.0			-18.3 ± 1.3	-16.9 ± 3.2	
muconic	-4.3 ± 0.4	-6.9 ± 0.2	-7.5 ± 0.3		-6.1 ± 1.5	-10.8 ± 0.6	-12.0 ± 0.9	

[a]Uncertainties are standard error estimates based on scatter of points about the least-squares van 't Hoff lines.

acid and the monoanion complexes the exothermic enthalpic contribution increases from oxalic/oxalate to glutaric/glutarate and then decreases slightly from the maximum and remains essentially constant for longer chains. On the other hand, the complexation entropy becomes increasingly negative from oxalic/oxalate to glutaric/glutarate and then increasingly positive from glutaric/glutarate to suberic/suberate. Thus the monotonically increasing complex strength (excluding malonic/malonate) with chain length observed at any given temperature is not explained by monotonic changes in both enthalpic and entropic contributions but rather for chains longer than glutaric/glutarate the increasingly positive entropy compensates for the invariant enthalpy in causing the continuing increase in complex strength for the longer chained substrates. This enthalpic and entropic behavior contrasts with that of Cy complexation of monocarboxylic acids where in the homologous series from formic acid [1] to octanoic acid [5], ΔH^0 decreases monotonically from -0.74 ± 0.11 to -11.2 ± 0.2 kcal mol^{-1} while ΔS^0 decreases monotonically from $+0.3 \pm 0.4$ to -22.7 ± 0.8 cal mol^{-1} K^{-1}. Similarly for the monocarboxylate anions from butyrate to octanoate [5], ΔH^0 decreases monotonically from -1.39 ± 0.1 to -11.2 ± 0.2 kcal mol^{-1} and ΔS^0 decreases monotonically from $+0.8 \pm 0.3$ to -24.1 ± 0.2 cal mol^{-1} K^{-1}. These two contrasting patterns suggest different mechanisms of complexation for the longer mono- and dicarboxylic acids. The monotonically increasing complexation exothermicity with chain length for monocarboxylic acids and anions suggests that as the chain length increases, each additional methylene group enhances the bonding interaction of the substrate with the cyclodextrin. On the other hand, the observation that the dicarboxylic acid and monoanion complexation enthalpies limit at glutaric/glutarate suggests that the larger chains do not interact with Cy, that only a fixed portion of the chain is included in the Cy cavity and that the remainder of the chain extends into the solution unaffected by complexation at the other end. This interpretation is consistent with the less negative (more positive) complexation entropies observed for the dicarboxylic acids and monoanions longer than glutaric acid/glutarate. Apparently the extending portions of these substrates are not significantly constrained so that the complexed substrate molecules or ions retain more internal degrees of freedom which is reflected by the correspondingly positive contribution to the overall entropies of complexation.

This interpretation suggests further details about the complexation mechanism. We note in Table III that for all alkanoic dicarboxylic acids and monoanions from succinic/succinate to subseric/suberate, both the complexation enthalpies and entropies of the molecular acid substrates are essentially equal to the corresponding values of the conjugate monoanion substrates. Also the dianion species of these acids do not form Cy complexes at any detectable concentration. These observations together with the interpretation that only a portion of the longer chains is included in the cavity suggests that the protonated carboxylic end of the monoanion chain is included in the Cy cavity and the carboxylate end is extended into the solution. The preferred Cy inclusion of the carboxylic acid over the carboxylate group suggested here is similar to the preference observed earlier in Cy complexes of benzoic acid/benzoate as found by NMR experiments [6, 7]. Those results indicated that the Cy includes the carboxylic acid group of benzoic acid but includes the phenyl group of benzoate anion.

We now turn to a discussion of the pimelic and suberic acid substrates where both dianion complexes and ternary complexes of the molecular acids are detected. We attribute this simultaneous appearance of two new complexes in the series of alkanedioic acids and anions to the lengths of the pimelic and suberic acid molecules relative to the size of the Cy cavity. Apparently, these dicarboxylic acid molecules are sufficiently long that a single substrate can be included in two cyclodextrin molecules and so the ternary complexes become stable. At the same time the corresponding dianions are sufficiently long that the central alkane chains can be included within the Cy cavity while both dianion groups can be in contact with the solution. Apparently adipate and shorter dianions have alkane chains which are too short to allow Cy complex structures with both dianion groups in contact with the solution and so dianion complexes are not formed. Thus, it appears that the binding mechanism for the pimelate and suberate dianions is quite different from the binding mechanism for the monoanions and dicarboxylic acids of succinate/succinic acid through suberate/suberic acid which involve interaction of the Cy with the protonated carboxylic acid group. The two dianion complexes apparently rely on interactions of the Cy with the alkane chain leaving the two solvated anion groups exposed to the solvent. From Table III we see that the Cy/dianion complexation enthalpy by this mechanism is less exothermic and the complexation entropy is less negative than the corresponding Cy/monoanion and Cy/dicarboxylic acid enthalpies and entropies which involve Cy interaction with the dicarboxylic acid.

While the thermodynamic evidence reported here seems to suggest these interpretations, these must nevertheless be regarded as speculative. Further work using alternative methods of probing the structures of cyclodextrin complexes are needed for corroboration.

References

1. R. I. Gelb, L. M. Schwartz, R. F. Johnson, D. A. Laufer: *J. Am. Chem. Soc.* **101**, 1869 (1979).
2. R. A. Robinson, R. H. Stokes: *Electrolyte Solutions*, 2nd ed.; Butterworths: London (1965).
3. L. M. Schwartz and R. I. Gelb: *Anal. Chem.* **50**, 1571 (1978).
4. R. G. Bates: *Determination of pH Theory and Practice*, 2nd ed.; Wiley: New York (1973).
5. L. M. Schwartz and R. I. Gelb: *J. Incl. Phenom.* in press.
6. R. I. Gelb, L. M. Schwartz, B. Cardelino, H. Fuhrman, R. F. Johnson, D. A. Laufer: *J. Am. Chem. Soc.* **103**, 1750 (1981).
7. R. J. Bergeron, M. A. Channing, K. A. McGovern: *J. Am. Chem. Soc.* **100**, 2878 (1978).