

Complexation Chemistry of Cyclohexaamyloses. 2. p-Methylcinnamate Anion¹

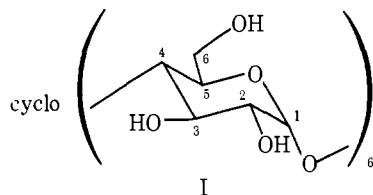
Robert I. Gelb, Lowell M. Schwartz, and Daniel A. Laufer*

Contribution from the Department of Chemistry, University of Massachusetts at Boston, Boston, Massachusetts 02125. Received March 17, 1978

Abstract: Formation constants at 30 °C of cyclohexaamylose-p-methylcinnamate anion complexes with 1:1 and 2:1 stoichiometries are estimated from conductometric and ¹³C NMR spectrometric data. Analysis of the ¹³C NMR data indicates direct binding of the carboxylate terminal of p-methylcinnamate by cyclohexaamylose in the 1:1 complex. The structure of the 2:1 complex involves occlusion of the anion between two cyclohexaamylose molecules in a head-to-head orientation.

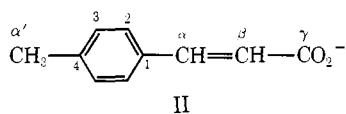
Introduction

Cyclohexaamylose (I), which we will denote as Cy, forms inclusion complexes with a variety of molecules and ions in aqueous solutions. These complexes are often assumed to be binary, i.e., 1:1 host-guest. However, there are some examples of ternary, i.e., two Cy-one guest structures, involving n-heptane,² cyclohexane,² and orange I.³ Complexes with 3:1



structures have been suggested with long-chain carboxylic acid derivatives.^{4,5} Little thermodynamic and structural information regarding ternary and higher complexes of Cy is available in the literature. Suspecting that formation of these higher complexes is more likely than previously supposed, we began to study the reactions of appropriate substrates with Cy. As an example, we have found that the 4-biphenylcarboxylate anion forms a dominant 2:1 adduct with Cy in aqueous solution.¹ Conductometric measurements of corresponding equilibrium constants at five temperatures between 20 and 45 °C yield values of -28 ± 2 kcal mol⁻¹ and -72 ± 6 cal mol⁻¹ K⁻¹ for the standard enthalpy and entropy changes, respectively, of the complexation reaction. The displacements of ¹³C resonances of Cy, which are the result of the complexation reaction, indicate a bitoroidal model in which the larger Cy rims containing the C₂ and C₃ carbon atoms assume a "face-to-face" orientation.

Given the dominance of ternary complex formation with the 4-biphenylcarboxylate anion, we have extended our investigation to derivatives of *trans*-cinnamic acid as substrates. Although the molecular dynamics⁶ and catalytic properties⁷ of some of these complexes have been studied, data concerning their stoichiometries, stabilities, and structures are presently sparse. We now report results of stoichiometric, thermodynamic, and structural studies with *trans*-p-methylcinnamate anion (II).



¹³C NMR Stoichiometric Analysis

The complexation of the p-methylcinnamate ion (A⁻) by cyclohexaamylose markedly displaces several of the Cy ¹³C NMR resonances. To determine the overall stoichiometry of complex formation we measured these displacements in 0.050

F Cy solutions containing up to 0.084 F Na⁺A⁻. No additional peaks other than those ascribed to Cy and A⁻ were observed.

As shown in an earlier work,¹ a plot of observed Cy displacement $\Delta\delta_{obsd}$ vs. $a/V^\circ F_{Cy}$ consists of two linear segments which intersect where $1/n = a/V^\circ F_{Cy}$. F_{Cy} denotes the initial concentration of Cy in V° mL and a is the number of Na⁺A⁻ moles which were successively added to the Cy solution. The stoichiometric coefficient n pertains to the overall complex reaction: $nCy + A^- = Cy_nA^-$. Resonance displacements of C₁ and C₃ Cy carbons are plotted in Figure 1 and the results of least-squares treatments of the line segments are given in Table I. Simultaneous solution of the line equations yielded n values of 2.00 ± 0.03 and 2.09 ± 0.03 for the C₁ and C₃ plots, respectively, where the uncertainties quoted are standard error estimates. Uncertainties in the form of line parameter variances were obtained from the least-squares analysis, and the corresponding n -value uncertainties were calculated from these by propagation of variance procedures. An estimate of the maximum probable error in n was made on the basis of estimated error limits as previously reported.¹ The results were $n = 2.00 \pm 0.3$ based on C₁ data and 2.09 ± 0.3 based on C₃ data.

Conductance Measurements of Complex Formation Constants

We have recently shown how the complex formation constants for the reaction of Cy with an ionic species can be measured by an electrolytic conductance technique.¹ In that study conductance measurements were made of an aqueous solution of lithium 4-biphenylcarboxylate into which successive aliquots of Cy solution were added. In the present work, the only procedural modification was to add solid rather than liquid portions of Cy. Since we must know the total solution volume after any such addition, the dependency of solution volume on Cy concentration must be determined by a separate set of density measurements. This set yielded the following empirical equation for the volume of solution formed by mixing 50 mL of water and m_{Cy} mmol of Cy:

$$V = 50 + 0.73m_{Cy} \quad (1)$$

This equation (1) together with eq 5-11 in ref 1 form the set of model equations which represent the behavior of the aqueous solutions with respect to stepwise formation constants K_1 and K_2 , conductance parameters λ°_0 , λ°_1 , and λ°_2 , and species concentrations. These are solved numerically by a nonlinear regression technique described briefly in ref 1 and in more detail in ref 8. In this latter reference, we discuss how statistical uncertainties in the parameters can be obtained from the matrix formulation of the regression technique.

In the present work, the conductance measurements were made at 30 °C on solutions of 0.014 F lithium p-methylcinn-

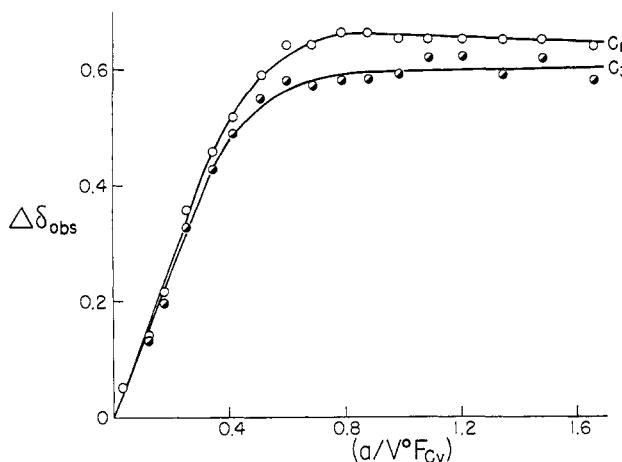


Figure 1. Chemical shift displacements of C_1 and C_3 carbons of Cy upon addition of A^- .

namate and Cy concentrations from 0 to $\sim 0.032 F$. The nonlinear regression calculation yielded the results shown in Table II. The quality of the fit of the model equations to the experimental data was reflected in (a) a standard deviation of residuals of 0.25% between calculated and experimental conductance values and (b) a completely random pattern of residuals in successive points. The standard deviation of residuals agrees with our independent estimate of 0.2% of the precision of the conductance measurements found by replicate determinations with the same apparatus. The random pattern of residuals indicates that no systematic error exists.

Our value of λ°_2 is in reasonable agreement with the value of 14.3 for λ° obtained for the ternary complex of Cy with 4-biphenylcarboxylate.¹ These λ° values would be expected to be similar because of the comparable size and structure of the two complexes.

We attempted to fit the data with model equations based on formation of only a binary complex and also based on formation of only a ternary complex. In both cases the data could not be adequately fit within the precision of measurement and examination of the residuals showed marked trends. The analysis confirmed the existence of substantial concentrations of both binary and ternary complexes.

¹³C NMR Studies of CyA⁻ and Cy₂A⁻ Structures and Complex Formation Constants

The ¹³C NMR spectrum of a D₂O solution containing Cy, *p*-methylcinnamate ion (A⁻), and their binary and ternary complexes consists of resonance lines corresponding to eight nonequivalent carbons on A⁻ (structure II) and six non-equivalent carbons on Cy (structure I). As in our previous study, we denote a given carbon resonance by both a superscripted symbol, e.g. (Cy2) referring to C₂ carbon of Cy or (A⁻β) referring to C_β carbon of A⁻, and a subscripted symbol, 0, 1, or 2 referring to the uncomplexed species, the binary complex, and the ternary complex, respectively.

Observed resonances of *p*-methylcinnamate anion were assigned to specific carbons using the following comparative criteria: (1) symmetry factors, (2) relative relaxation rates of protonated and nonprotonated carbons, and (3) best fit of measured p-CH₃ substituent effects in the *p*-methylcinnamate/cinnamate system with corresponding effects in the *p*-methylstyrene/styrene system.⁹ ¹³C NMR assignments of Cy carbons have been previously reported.¹⁰

Since all species involved in the complexation reactions undergo rapid exchange relative to the NMR time scale, the observed chemical shifts $\delta_{obsd}^{(A^-)}$ from a given A⁻ carbon and $\delta_{obsd}^{(Cy)}$ from a given Cy carbon are weighted averages of chemical shifts of the individual species in solution containing

Table I. Least-Squares Slope and Intercept Values for Line Segments in Figure 1 with Standard Error Estimates of Uncertainties

carbon	line segment $\langle a/V^\circ F_{Cy} \rangle$	intercept, ppm	slope
1	0.0	0.4	-0.003 ± 0.012
1	0.8	1.6	0.673 ± 0.005
3	0.0	0.4	-0.002 ± 0.011
3	0.8	1.6	0.580 ± 0.030

Table II. Results of Nonlinear Regression Calculations on Conductance Measurements of Lithium *p*-Methylcinnamate and Cy Aqueous Solutions

parameters	values	std error estimates ^a
$\log K_1$	2.2	0.4
$\log K_2$	2.5	0.2
λ°_0 ^b	29.4	0.1
λ°_1 ^b	15	4
λ°_2 ^b	12.3	0.4

^a Based on a standard deviation of residuals of 0.002 conductance units (μmho) which corresponds to $\pm 0.25\%$. ^b mho cm^2 equiv⁻¹.

A⁻ and Cy, respectively. This averaging process is expressed as

$$\delta_{obsd}^{(A^-)} = (\delta_0^{(A^-)}[A^-] + \delta_1^{(A^-)}[\text{CyA}^-] + 2\delta_2^{(A^-)}[\text{Cy}_2\text{A}^-])/F_{A^-} \quad (2)$$

for any given carbon of A⁻ and as

$$\delta_{obsd}^{(Cy)} = (\delta_0^{(Cy)}[\text{Cy}] + \delta_1^{(Cy)}[\text{CyA}^-] + 2\delta_2^{(Cy)}[\text{Cy}_2\text{A}^-])/F_{Cy} \quad (3)$$

for any given carbon of Cy. In these equations, [] denote the molar concentration of enclosed species, F_{A^-} and F_{Cy} , the formal (analytical) concentrations of A⁻ and Cy, and the factor 2 in the third term of eq 3 reflects the stoichiometry of the Cy₂A⁻ complex. The molar concentrations of species in these equations depend on the complex formation constants and the solution compositions as expressed by the following equations:

conservation of A⁻

$$F_{A^-} = [A^-] + [\text{CyA}^-] + [\text{Cy}_2\text{A}^-] \quad (4)$$

conservation of Cy

$$F_{Cy} = [\text{Cy}] + [\text{CyA}^-] + 2[\text{Cy}_2\text{A}^-] \quad (5)$$

binary complex formation equilibrium

$$K_1 = \gamma_1[\text{CyA}^-]/\gamma_0[\text{Cy}][A^-] \quad (6)$$

ternary (stepwise) complex formation equilibrium

$$K_2 = \gamma_2[\text{Cy}_2\text{A}^-]/\gamma_1[\text{Cy}][\text{CyA}^-] \quad (7)$$

activity coefficient correlation (Debye-Hückel)

$$\log \gamma = -B_1 \sqrt{I}/(1 + B_2 a_i \sqrt{I}) \quad (8)$$

where γ_0 , γ_1 , and γ_2 refer to A⁻, CyA⁻, and Cy₂A⁻, respectively. γ in eq 8 refers to each of these when the ion-size parameter a_i is taken as 0.75, 1.6, and 1.8 nm for A⁻, CyA⁻, and Cy₂A⁻, respectively. The Debye-Hückel parameters at 30 °C, $B_1 = 0.5161$ and $B_2 = 3.301$ are from the Robinson and Stokes tabulation.¹¹ While these values refer to an H₂O solvent, they can hardly be in substantial error for the D₂O solvent employed in our ¹³C NMR measurements. In any case the errors introduced here are small compared with the uncertainty in our results for K_1 and K_2 . The ionic strength, I , is F_{A^-} .

Table III. *p*-Methylcinnamate (PMC) and α -Cyclodextrin Chemical Shifts, Their Standard Errors, and Their Displacements in the Complexes Relative to the Uncomplexed Species. ^a Comparison with Shift Displacements in the 4-Biphenylcarboxylate (BPC) System^b

PMC carbon	$\delta_0(A^-)$	$\delta_1(A^-)$	$\delta_2(A^-)$	$\Delta\delta_1(A^-)$	$\Delta\delta_2(A^-)$	BPC carbon	$\delta_0(A^-)$	$\Delta\delta_2(A^-)$
γ	176.89	174.0	173.94	-2.9	-2.95	α	176.33	-3.3
β	124.24	125.8	127.03	1.6	2.79	4	136.22	0.9
α	141.71	142.2	140.00	0.5	-1.71	3	130.17	2.8
						2	128.10	-1.5
						1	144.12	0.35
1	133.39	132.2	134.71	-1.2	1.32	1'	140.92	1.8
2	130.59	130.5	129.52	-0.1	-0.07	2'	127.72	-1.6
3	128.72	129.7	128.11	1.0	-0.61	3'	130.54	1.4
4	141.31	141.9	135.55	0.6	-1.76	4'	129.12	-1.4
α'	21.43	22.4	21.15	1.0	-0.28			
Standard error	± 0.01	± 0.2	± 0.01	± 0.2	± 0.02			
$\log K_1 = 1.7 \pm 0.4; \log K_2 = 3.3 \pm 0.4$								
Cy carbon	$\delta_0(Cy)$	$\delta_1(Cy)$	$\delta_2(Cy)$	$\Delta\delta_1(Cy)$	$\Delta\delta_2(Cy)$		$\Delta\delta_2(Cy)$	
						($A^- = BPC$)		
1	102.37	102.8	103.09	0.5	0.72		0.75	
2	72.74	72.9	73.06	0.2	0.32		0.39	
3	74.30	74.7	74.97	0.4	0.67		0.43	
4	82.23	82.7	82.15	0.5	-0.07		0.08	
5	72.99	73.2	72.99	0.2	0.00		0.12	
6	61.45	61.2	61.40	-0.2	-0.05		0.00	
Standard error	± 0.01	± 0.1	± 0.01	± 0.1	± 0.02			
$\log K_1 = 2.0 \pm 0.5; \log K_2 = 3.0 \pm 0.5$								

^a Chemical shifts are given in parts per million relative to Me₄Si. Upfield displacements are indicated by negative signs. ^b Reference 1.

Our purpose in doing these studies is to extract the unknown parameters K_1 , K_2 , $\delta_0(A^-)$, $\delta_1(A^-)$, $\delta_2(A^-)$, $\delta_0(Cy)$, $\delta_1(Cy)$, and $\delta_2(Cy)$ from $\delta_{obsd}(A^-)$ and $\delta_{obsd}(Cy)$ vs. F_{A^-} and F_{Cy} data. The number of parameters involved here is 2 plus $3 \times 8 = 24$ chemical-shift parameters for A^- carbons plus $3 \times 6 = 18$ chemical-shift parameters for Cy carbons. Consequently, we must measure at least 44 δ_{obsd} shifts and, since each solution which contains both A^- and Cy provides 14 such data values, we require at least four solutions. In practice, we made ^{13}C NMR measurements on far more than the minimum since we wish to extract statistical uncertainty estimates from the data also. A total of 42 solutions were prepared with F_{A^-} varying up to 0.15 F and F_{Cy} up to 0.10 F and with a wide range of Cy/ A^- mole ratios. The unknown parameters were calculated from the data by the following strategy. Two separate computations were made, one utilizing $\delta_{obsd}(A^-)$ measurements and eq 2 and the other utilizing $\delta_{obsd}(Cy)$ and eq 3. In both of these computations K_1 and K_2 were regarded as unknown parameters to be determined by nonlinear regression of the model eq 2 and 4-8 for A^- data or eq 3-8 for Cy data. This nonlinear regression procedure is described in detail in ref 8. Briefly, one begins with initial K_1 and K_2 estimates and successively refines these by a matrix formulation procedure. Optimal K_1 and K_2 are accepted when these fail to change on successive iteration. These optimal values minimize the sum of squares of the deviations between the δ_{obsd} values predicted by the model equations and the experimentally recorded δ_{obsd} values. The matrix calculation also yields standard error estimates for K_1 and K_2 . The δ_{obsd} values calculated from the model equations and the current set of K_1 and K_2 proceeds as follows. (1) In each solution, activity coefficients are calculated from F_{A^-} using eq 8. (2) Having eliminated $[A^-]$, $[CyA^-]$, and $[Cy_2A^-]$ from eq 4-7 by substitution, there remains a nonlinear equation involving $[Cy]$, K_1 , K_2 , and activity coefficients. This is solved for $[Cy]$ in each solution by the method of successive approximations. (3) All other species concentrations in all solutions are calculated from eq 4-7. (4) Either eq 2 or 3, when written for all carbons in all solutions, represents a set of overdetermined linear equations for the unknown shift parameters δ_0 , δ_1 , and δ_2 . The set of eq 2 for A^- involves 24 such parameters

and the set of eq 3 for Cy involves 18. Either set must be solved by linear multiple regression¹² and, if the matrix formulation is used, this procedure yields both the parameter values and standard error estimates. (5) Having δ_0 , δ_1 , and δ_2 for each carbon, either eq 2 or 3 is used to calculate δ_{obsd} values for all carbons in all solutions and the sum of squared deviations of these values from the recorded δ_{obsd} data serves as the fitting function for K_1 , K_2 iteration. (6) K_1 and K_2 are then adjusted as indicated by the matrix formulation to obtain the minimum in the fitting function.

Inherent in our treatment of these ^{13}C NMR data is the implicit assumption that the various ^{13}C NMR resonances of Cy, A^- , and the complexes are each independent of concentration and ionic strength. To verify this behavior for Cy, we obtained spectra at five concentrations between 0.005 and 0.10 F and in NaCl solutions up 0.5 F . In no case were any of the Cy resonances perturbed to a measurable extent. That is, variations in the data appeared to be random and did not exceed ± 0.02 ppm. We also found the A^- resonances to be essentially independent of concentration between 0.02 and 0.15 F . These results lead us to believe that the Cy A^- and Cy₂ A^- resonances are concentration independent as well. The results of our calculations appear in Table III.

We examined the residuals ($\delta_{calcd} - \delta_{obsd}$) and these appeared to be randomly scattered and showed no systematic trends. They rarely exceeded 0.03 ppm and gave a root-mean-square deviation of about ± 0.02 ppm, which is consistent with the resolution of the ^{13}C NMR spectrometer employed. The values of $\log K_1$ and $\log K_2$ represent best values determined from Cy data and A^- data independently. These values which refer to a D₂O solvent are in approximate agreement with one another and with the conductometric log K values, obtained with an H₂O solvent.

Discussion

Interpretation of the ^{13}C NMR data listed in Table III indicates some of the structural features of the ternary and binary complexes. In this table we have included ^{13}C NMR displacements of the ternary Cy-4-biphenylcarboxylate complex as a means of comparison. Our discussion is divided

into four parts, the first two of which deal with the Cy data and the last two with perturbations in *p*-methylcinnamate resonances.

Ternary Cy Displacements. The $\Delta\delta_2^{(\text{Cy})}$ values reflect little perturbation at the C₅ and C₆ carbons (0.00 and -0.05 ppm, respectively), whereas the C₂ and C₃ carbons are significantly perturbed (0.32 and 0.67 ppm, respectively). The latter secondary carbon atoms are located on the larger rim of the Cy torus and we have termed it the "head" of the molecule. We consider the pattern of C₂, C₃ vs. C₅, C₆ displacements as evidence favoring an antiparallel, "head-to-head" association of the two Cy constituents in Cy₂A⁻. A space-filling model of a Cy₂A⁻ complex with this orientation was assembled. In this model the A⁻ guest is coaxially occluded in a cavity zone defined by the duplicate bands of glycosidic oxygens (I, C₁-O-C₄). The model suggests that the points of closest approach among the two Cy and one A⁻ constituents would result in the substantial perturbations at C₂ and C₃ carbons which are observed. On the other hand, the C₅ and C₆ carbons are located on the "tails" of the "head-to-head" model. These sites are remote from the A⁻ ion and this is consistent with the data. The anomeric C₁ carbons are significantly deshielded (0.72 ppm) and this may be due in part to macrocyclic conformational changes associated with the complexation as well as direct interaction with the substrate. The pattern and magnitude of ¹³C NMR resonance displacements is substantially the same as those observed in the formation of the 4-biphenylcarboxylate complex with Cy.

Binary Cy Displacements. The pattern of Cy displacements in the binary complex is similar to that of the ternary complex. The C₅ and C₆ carbons are perturbed to a lesser extent than the C₃ carbon (0.2 and -0.2 vs. 0.4 ppm). These displacements are consistent with a binary model where the A⁻ ion is partially occluded in the cavity zone surrounded by the wider rim of Cy. We note that the magnitude of the C₃ displacement is smaller in the binary complex compared with the ternary complex in which the environment of the C₃ carbon located at the head of the molecule is perturbed by both Cy-Cy and Cy-A⁻ interactions (0.4 vs. 0.67 ppm). Since the Cy-Cy interactions must be absent in the binary complex, the comparison of Cy displacements in the binary and ternary complexes appears reasonable.

Ternary A⁻ Displacements. The parameters of this set indicate perturbations of all A⁻ carbon atoms as expected from a ternary model where the guest molecule is entirely occluded. The C_γ and C_{α'} carbons as well as the C₃ and C₄ phenylene carbons are shielded, reflecting the transfer of these terminal atoms from D₂O solution into a less polar environment of the Cy cavity. The similarity of displacements of carboxylate carbons here and in the ternary 4-biphenylcarboxylate complex suggests that a displacement of about -3 ppm is characteristic of this environmental change.

A ternary complex in which the substrate is completely occluded between two Cy molecules is supported by the work of Behr and Lehn.⁶ They measured ²H nuclear quadrupole relaxation times, T_q , of Cy complexes of A⁻ in which the α and α' positions of A⁻ had been deuterated.

T_q for both deuterons decreased markedly with addition of Cy to A⁻ solutions, reflecting the interaction of both "ends" of the A⁻ substrate with Cy. Our equilibrium constant data indicate that the Cy₂A⁻ complex is dominant under the experimental conditions used in their work, [Cy₂A⁻]/[CyA⁻] varying between ~2 in the more dilute solutions up to ~20 in the more concentrated Cy mixtures. Interaction of the Cy molecule with both the α and α' A⁻ sites seems to indicate complete occlusion of A⁻ in the ternary complex, as suggested above.

Binary A⁻ Displacements. Several comparisons between displacements in the binary and ternary complexes are evident;

these are (1) the magnitude and direction of the C_γ (carboxylate) displacements are strikingly similar in binary and ternary complexes (-2.9 and -2.95 ppm, respectively); (2) the C_α and particularly the C_β carbons on the acrylate side chain are perturbed in the binary complex (0.5 and 1.6 ppm, respectively) but experience larger perturbations (-1.71 and 2.79 ppm) in the ternary complex; (3) the terminal methyl carbon (C_{α'}) along with the C₃ and C₄ phenylene carbons are substantially shielded upon inclusion in the ternary complex but these are deshielded in the binary complex.

These observations seem consistent with a binary complex in which the carboxylate terminal of *p*-methylcinnamate is preferentially bound within the Cy cavity. However, we were not able to rule out the existence of an isomeric binary complex in which the *p*-tolyl terminal is bound, but we feel that the relative proportion of such a complex, if it exists at all, is small.

Our reasoning is as follows. (1) The similarity of C_γ displacements in both binary and ternary complexes strongly suggests that the carboxylate terminal experiences a similar environment in both complexes and, as a result, appears to be bound in a Cy cavity in the binary complex. (2) The binary and ternary displacements observed for C_α and C_β may result from both direct interaction with the Cy cavity and interaction with the complexed carboxylate group. The increased displacements observed upon formation of the ternary complex suggest tighter binding in the ternary adduct. However, increased resonance, π polarization, and field effects brought about by complexation of the phenylene group in Cy₂A⁻ may play a role as well. (3) The shieldings of C_{α'}, C₃, and C₄ upon formation of the ternary complex represent upfield perturbations of 1.3, 1.6, and 2.4 ppm, respectively, compared with these resonances in the binary complex. This seems consistent with insertion of the *p*-tolyl terminal into the Cy cavity in the ternary but not binary complex.

In summary, the ¹³C NMR data seem to indicate a binary complex in which the "charge rich" carboxylate terminal is included in the Cy cavity as the dominant structural form. The possible existence of a minor binary adduct where the *p*-tolyl moiety enters the Cy cavity cannot be eliminated since it would always be present as a fixed proportion of the total concentration of binary complex in our experiments. Presumably such a complex, if it exists, might be detected by measurement of the temperature dependence of $\Delta\delta_1^{(A^-)}$ values. Because the formation constant of an isomeric binary adduct would probably have a different temperature dependence from that of the dominant form observed at 30 °C, the values of $\Delta\delta^{(A^-)}$ at different temperatures would reflect the differing isomeric proportions. We are currently undertaking such studies.

In any case, addition of a second Cy results in complete occlusion of the *p*-methylcinnamate ion and the complex appears to be stabilized in part by intermacrocyclic interactions at the wider Cy rims. The interactions are evidenced by increased perturbations of C₂ and C₃ sites of Cy.

The preferential binding of a charged group observed in this work seems at variance with the often quoted concept of a hydrophobic cavity in cyclohexaamylose. However, similar effects have been noted in this laboratory and elsewhere for comparable systems. The unusual stability and exclusive formation of a 2:1 complex from two molecules of Cy and one 4-biphenylcarboxylate anion imply that a binary adduct with a bound phenyl moiety is relatively unfavorable in that system.¹ ¹H NMR studies of the 1:1 complexes of Cy with *p*-nitrophenol and *p*-nitrophenolate anion indicate that in both cases the *p*-nitro substituents, isoelectronic with carboxylate anion, are preferentially bound in the Cy cavity.¹³

The specific binding modes which have been noted thus far indicate that cyclohexaamylose is a more selective host than previously supposed. We are continuing our investigation of

the structural and thermodynamic features of ternary and binary cycloamylose complexes. A further understanding of the factors involved in aqueous Cy complex formations and, in particular, selectivity in these reactions, appears central to the role of cycloamyloses as models for biological complexations.

Experimental Section

Lithium and sodium salts of *p*-methylcinnamic acid were prepared by treatment of portions of the parent acid, obtained from the Aldrich Chemicals Co., with an excess of Li₂CO₃ or NaOH in hot aqueous solution. The crude salt was collected after cooling and was twice recrystallized from boiling water. Portions of the sodium salt were analyzed by titration with standard HCl and were employed for all ¹³C NMR measurements. Solutions of the lithium salt were employed for conductance measurements after appropriate analysis with standard HCl. In both cases, the precipitation of *p*-methylcinnamic acid during titration with HCl precluded the usual analysis of the titration data obtained before or near the equivalence point. The end points were determined by a suitable Gran plot method.

Cyclohexaamylose was obtained from the Aldrich Chemicals Co. and was used without further purification. Subsequent ¹³C NMR experiments gave resonances which could only be ascribed to cyclohexaamylose. No additional signals above the normal noise level could be detected under high signal/noise conditions.

¹³C NMR experiments employed a Varian CFT-20 spectrometer equipped with a 10-mm sample tube maintained at 30 ± 2 °C. In a typical series of experiments, spectra were recorded after each addition of a microweighed portion of sodium *p*-methylcinnamate to an 8.00-mL aliquot of α -cyclodextrin solution contained in the sample tube. Instrument settings were as follows: 30° tip angle; 2-s delay time; 1-s acquisition time. A spectral width of 4000 Hz was employed with accumulation of 8192 data points so that the resolution was 0.02 ppm throughout. Several thousand cycles were acquired for each spectrum.

Proton-decoupled ¹³C chemical shifts were measured in parts per million downfield from external Me₄Si.

Conductance measurements employed a General Radio 1608A impedance bridge equipped with dip-type conductance cell. The experiments involved addition of weighed portions of α -cyclodextrin to a sample solution contained in a vessel thermostated at 30 ± 0.1 °C. The conductance was measured upon each addition after allowing appropriate time for equilibration.

Acknowledgments. We thank Professor Elkan R. Blout for providing access to the CFT-20 spectrometer at the Department of Biological Chemistry, Harvard Medical School. This work was supported in part by a research grant from the National Institute of General Medical Sciences of the National Institutes of Health, U.S. Public Health Service (GM-19645).

References and Notes

- (1) The previous paper in this series is R. I. Gelb, L. M. Schwartz, C. T. Murray, and D. A. Laufer, *J. Am. Chem. Soc.*, **100**, 3553 (1978).
- (2) A. Wishnia and S. J. Lappi, *J. Mol. Biol.*, **82**, 77 (1974).
- (3) A. Harada, M. Furue, and S. Nozakura, *Macromolecules*, **9**, 701 (1976).
- (4) R. Bergeron, Y. Machida, and K. Bloch, *J. Biol. Chem.*, **250**, 1223 (1975).
- (5) H. Schlenk and D. M. Sand, *J. Am. Chem. Soc.*, **83**, 2312 (1961).
- (6) J. P. Behr and J. M. Lehn, *J. Am. Chem. Soc.*, **98**, 1743 (1976).
- (7) (a) R. Breslow, *Adv. Chem. Ser.*, **No. 100**, 21 (1971); (b) R. Breslow and L. E. Overman, *J. Am. Chem. Soc.*, **92**, 1075 (1970).
- (8) L. M. Schwartz and R. I. Gelb, *Anal. Chem.*, in press.
- (9) G. K. Hamer, I. R. Peat, and W. F. Reynolds, *Can. J. Chem.*, **51**, 897 (1973).
- (10) P. Colson, H. J. Jennings, and I. C. P. Smith, *J. Am. Chem. Soc.*, **96**, 8081 (1974).
- (11) R. A. Robinson and R. H. Stokes, "Electrolyte Solutions", 2nd ed (revised), Butterworths, London, 1965.
- (12) O. L. Davis and P. L. Goldsmith, "Statistical Methods in Research and Production", 4th ed (revised), Oliver and Boyd, Edinburgh, 1972.
- (13) R. J. Bergeron, M. A. Channing, G. J. Gibely, and D. M. Pillor, *J. Am. Chem. Soc.*, **99**, 5146 (1977), and references cited therein.

On the Amide Hydrogen Bond and the Anomalous Packing of Adipamide

A. T. Hagler*^{1a} and L. Leiserowitz*^{1b}

Contribution from the Department of Chemical Physics and the Department of Structural Chemistry, The Weizmann Institute of Science, Rehovot, Israel. Received August 26, 1977

Abstract: A methodology for the study of the role of hydrogen bonding and other energetic factors in determining the symmetry and secondary and tertiary structures of hydrogen-bonded crystals is presented. Lattice energy minimization techniques are extended to the study of "hypothetical" crystal packing modes in order to carry out this analysis. The comparison of these hypothetical crystal structures with observed structures is also proposed as a useful tool for testing the validity of potential functions. The model system chosen for this initial study is adipamide. The observed packing mode of adipamide is anomalous with respect to the family of primary amides in that it does not involve hydrogen-bonded rings. The hydrogen-bonding structure of adipamide appears to be less stable than the standard packing motif. A total of 16 hypothetical crystals, comprising various space groups, were constructed. These include the three typical modes of hydrogen-bonded networks (secondary structures) related by various symmetries to form structures common to the family of primary amides. The observed structure is calculated to be more stable than all hypothetical structures considered. Analysis of intra- and interlayer energies indicates that adipamide, by foregoing the most favorable hydrogen-bonding arrangement, achieves a better interlayer packing which more than compensates for the poorer hydrogen bonding. Several hypothetical structures are found to be within a kilocalorie or two of the experimental structure. This result is related to the tendency to form the maximum number of hydrogen bonds and polymorphism in crystals. Finally, the results are discussed in terms of symmetry considerations, both with regard to the experimental observation that amides rarely pack in orthorhombic and higher symmetries, and the need to relax symmetry constraints when studying hypothetical models.

Introduction

There have been many studies of the lattice energetics of organic molecules carried out in the past few years.²⁻⁸ These have been mainly directed towards using the precise infor-

mation available in the crystalline state to determine and test intermolecular potential functions. In these studies the common practice has been to minimize the crystal energy with respect to some subset of the crystalline degrees of freedom in