Journal of Inclusion Phenomena and Molecular Recognition in Chemistry 7: 537–543, 1989.

Complexation of Adamantane-Ammonium Substrates by Beta-Cyclodextrin and its *O*-methylated Derivatives

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

(Received: 29 August 1988; in final form: 1 January 1989)

Abstract. A spectrophotometric method is used to determine formation constants of complexes of β-cyclodextrin, the 2,6-di-*O*-methylated and 2,3,6-tri-*O*-methylated derivatives of β-cyclodextrin as hosts with adamantan-1-ylammonium, adamantan-2-ylammonium and adamantan-1-ylmethylammonium as substrate species. The spectrophotometric method uses methyl orange anion and acid forms as indicator species. Complexes of the cyclodextrins with these species are determined as well as with the adamantane derivatives. Standard enthalpies and entropies of formation of all complexes are calculated from the temperature variation of the equilibrium constants. β-Cyclodextrin and its 2,6-*O*-methyl derivative have comparable complex strengths with adamantaneammonium substrates and these strengths are about two orders of magnitude stronger than the corresponding complexes of the permethylated derivative. Thermodynamic parameters are interpreted in terms of differing intramolecular properties of the cyclodextrin complexes.

Key words. Cyclodextrin, formation constant, uv-visible spectrophotometry, cationic guests.

1. Introduction

Thermodynamic properties of cycloamylose inclusion complexes have been widely studied in recent years. Enthalpies and entropies of complex formation are now available for cyclohexaamylose and cycloheptaamylose with a variety of substrate species [1–7]. However, relatively little has appeared on the complexation equilibria of derivatized cycloamyloses. We now report an investigation of the thermodynamic properties of some *O*-methylated cycloheptaamylose complexes with adamantane derivatives. Specifically, we compare complexation properties of cycloheptaamylose, here denoted as Cy, with those of heptakis(2,6-di-*O*-methyl)-β-cyclodextrin and heptakis(2,3,6-tri-*O*-methyl)-β-cyclodextrin denoted as DMCy and TMCy, respectively. The substrate species are adamantan-1-ylammonium ion, adamantan-2-ylammonium ion, and adamantan-1-ylmethylammonium ion symbolized, 1-A, 2-A and AMA, respectively. Structural formulas for these species are shown in Figure 1. In addition we have measured complex formation constants of these cycloamyloses with acidic and basic forms of methyl orange, HMO and MO, respectively. We employ a previously reported [8] spectrophotometric method to obtain the complexation constants. Briefly, the method relies on cycloamylose complexation with a photometric acid-base indicator system (here methyl orange) buffered near the pK_a of the indicator. The absorbance is measured at a wavelength chosen to maximize

* Author for correspondence.

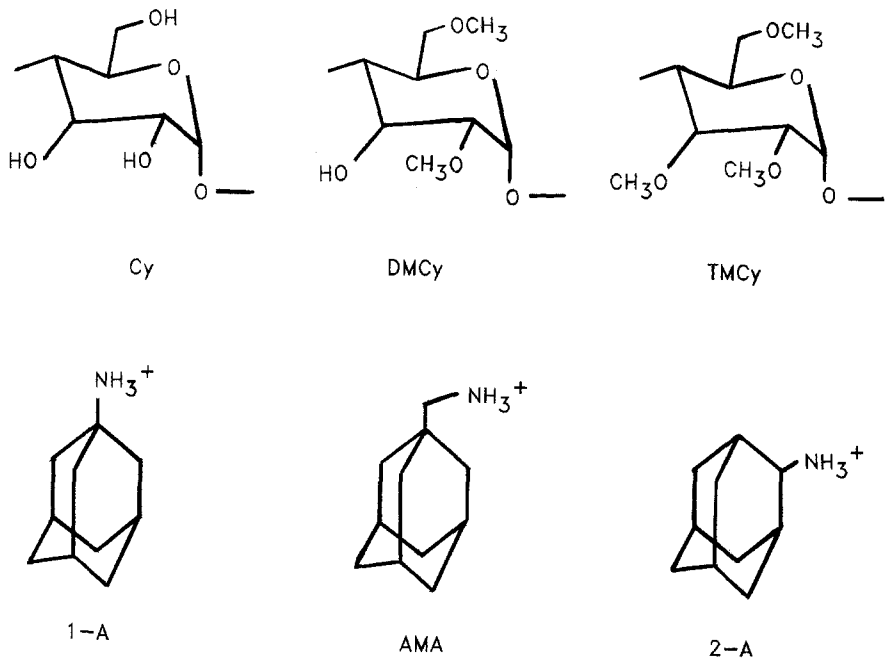

Fig. 1. Structural formulas of amylose units of β-cyclodextrin or cyclohexaamylose (Cy), heptakis (2,6-di-*O*-methyl)-β-cyclodextrin (DMCy) and heptakis (2,3,6-tri-*O*-methyl)-β-cyclodextrin (TMCy). Each β-cyclodextrin is a cyclic oligomer of seven amylose units. Also shown are structural formulas of the substrates adamantan-1-ylammonium ion (1-A), adamantan-1-ylmethylammonium ion (AMA) and adamantan-2-ylammonium ion (2-A).

the difference in absorptivity of protonated and unprotonated indicator species. Cycloamylose forms complexes with both indicator species but these complexes are of unequal strength and have absorptivities different from those of the free indicator species. As a consequence of these properties, the addition of cycloamylose to the buffered indicator causes marked changes in absorbance. Thus, appropriate analysis of absorbance versus composition data yields cycloamylose complexation constants with both protonated and unprotonated indicator species as well as molar extinction coefficients of both complexed and uncomplexed indicator species. In the present experiment we also add varying concentrations of adamantane derivatives as substrate species. Neither the substrates nor their cycloamylose complexes absorb at the wavelength chosen. Nevertheless, their presence in solution perturbs the indicator/proton/cycloamylose equilibrium which is reflected in significant absorbance changes. By this perturbation the cycloamylose/substrate complexation constants are determined.

2. Experimental and Calculational Methods

Methyl orange (*p*-dimethylamino-phenylazobenzene-*p*-sulfonic acid) as the sodium salt, the cycloamyloses and adamantane derivatives all were obtained from Aldrich Chemical Co. Cycloheptaamylose samples were twice recrystallized from water but

methylated derivatives were used without further preparation. Stock solutions were prepared from weighed portions of the anhydrous substances obtained by vacuum drying at 100°C for at least one day. Adamantan-1-ylamine obtained as the reagent grade hydrochloride was used without further purification. Other adamantane derivatives were dissolved in dry hexane and treated with HCl gas to obtain the hydrochloride salts which were then weighed into stock solutions without further purification.

The solutions to be measured were prepared with reagents in the following concentration ranges: 3.0×10^{-5} M methyl orange; HCl between 1.7 and 13 mM or buffered at pH = 7 with phosphate; cycloamylose, 0–2 mM; and substrate species, 0–5 mM.

Absorbance measurements were made with a Beckman Acta CIII uv-visible spectrophotometer equipped with a thermostatted 1.000 cm cuvet that was used throughout. Special care was taken to assure thermal equilibrium of each solution. Absorbance readings at 530 nm ranged between 0.1 and 1.2 with replication of 0.001. Absorbance versus composition data were analyzed by a nonlinear regression procedure. Initial estimates of cycloamylose complexation constants with indicator and substrate species were adjusted to obtain a minimum weighted sum-of-squares deviation between observed and calculated absorbance values. Calculated absorbance values were derived from a set of model equations based on the following assumptions:

1. Beer's Law applies to each absorbing species, i.e. MO and HMO and their cycloamylose complexes. Each species has an independent extinction coefficient.
2. Thermodynamic equilibrium constant expressions describe the indicator acid–base behavior and various cycloamylose complexations.
3. Ionic activity coefficients are estimated by the Debye–Hückel correlation which is quite accurate in these solutions where the ionic strength never exceeded 0.02 M. Temperature-dependent coefficients in the correlation are taken from Robinson and Stokes [9] and ion-size parameters of 0.9, 1.0, 0.9 and 1.6 nm are assumed for H^+, MO anion, adamantanyl cation species and cycloamylose complexes, respectively.
4. Uncharged species activity coefficients have unit value.
5. Charge balance and mass conservation equations relate analytical concentrations of MO, cycloamylose and substrate to their respective species concentrations.
6. Adamantanyl amine substrates are present exclusively as the protonated ammonium ions in their solutions having pH near 2.5.

The results of the nonlinear regression analysis yields complexation constants of cycloamylose with MO, HMO and the substrate species, pK_a for the indicator and extinction coefficients for the indicator species and their cycloamylose complexes. In addition, the rms fit between observed and calculated absorbance values and standard error estimates for each calculated parameter are output. Rms fit values were always acceptably small, i.e. $\leqslant 0.001$. Differences between calculated and measured absorbance values never exceeded 0.002 and appeared to be free from systematic trends. Because our principal intent here is to determine cycloamylose constants, we do not report the various temperature dependent extinction

Table I. Complexation constants of Cy, DMCy, and TMCy with MO, HMO, 1-A, 2-A and AMA substrates

	Substrate				
t, °C	MO	HMO	1-A	2-A	AMA
			Cy Complexes		
17.0	$5.3(\pm0.4) \times 10^3$	$0.24(\pm0.04) \times 10^3$	$10.8(\pm1.2) \times 10^3$	$9.0(\pm1.0) \times 10^3$	$38(\pm8) \times 10^3$
25.0	$4.5(\pm0.4) \times 10^3$	$0.23(\pm0.04) \times 10^3$	$8.9(\pm1.1) \times 10^3$	$7.4(\pm0.9) \times 10^3$	$30(\pm6) \times 10^3$
37.0	$3.3(\pm0.3) \times 10^3$	$0.20(\pm0.04) \times 10^3$	$6.1(\pm0.7) \times 10^3$	$5.1(\pm0.6) \times 10^3$	$21(\pm4) \times 10^3$
49.5	$2.3(\pm0.2) \times 10^3$	$0.16(\pm0.04) \times 10^3$	$3.9(\pm0.5) \times 10^3$	$3.7(\pm0.4) \times 10^3$	$19(\pm3) \times 10^3$
			DMCy Complexes		
17.0	$42.7(\pm1.7) \times 10^3$	$0.98(\pm0.09) \times 10^3$	$8.9(\pm0.3) \times 10^3$	$8.4(\pm0.3) \times 10^3$	$38.2(\pm1.6) \times 10^3$
25.0	$37.6(\pm1.5) \times 10^3$	$0.95(\pm0.10) \times 10^3$	$8.0(\pm0.3) \times 10^3$	$7.6(\pm0.3) \times 10^3$	$35.2(\pm1.5) \times 10^3$
37.0	$28.6(\pm0.9) \times 10^3$	$0.86(\pm0.09) \times 10^3$	$6.5(\pm0.2) \times 10^3$	$6.5(\pm0.2) \times 10^3$	$28.4(\pm1.0) \times 10^3$
49.5	$20.7(\pm0.5) \times 10^3$	$0.78(\pm0.10) \times 10^3$	$5.3(\pm0.1) \times 10^3$	$5.1(\pm0.1) \times 10^3$	$21.5(\pm1.0) \times 10^3$
			TMCy Complexes		
17.0	$6.2(\pm0.3) \times 10^3$		58 ± 6	90 ± 7	377 ± 18
25.0	$5.1(\pm0.2) \times 10^3$		57 ± 6	84 ± 7	368 ± 20
37.0	$3.6(\pm0.1) \times 10^3$		50 ± 5	73 ± 6	319 ± 16
49.5	$2.3(\pm0.1) \times 10^3$		41 ± 5	59 ± 6	248 ± 15

[a] Quoted $\pm$ uncertainties are standared error estimates.

Table II. ΔH^0 and ΔS^0 of complex formation of Cy, DMCy, and TMCy with MO, HMO, 1-A, 2-A and AMA substrates

Substrate	ΔH^0, kcal mol^{-1}	ΔS^0, cal mol^{-1} K^{-1}
	Cy Complexes	
1-A	-5.9 ± 0.5	-1.8 ± 1.5
2-A	-5.2 ± 0.2	$+0.2 \pm 0.6$
AMA	-4.1 ± 0.7	$+6.9 \pm 2.2$
MO	-4.7 ± 0.4	$+1.2 \pm 1.2$
HMO	-1.9 ± 0.7	$+3.0 \pm 2.0$
	DMCy Complexes	
1-A	-3.0 ± 0.2	$+7.7 \pm 0.5$
2-A	-2.8 ± 0.3	$+8.4 \pm 1.0$
AMA	-3.3 ± 0.4	$+9.5 \pm 1.2$
MO	-4.1 ± 0.4	$+6.8 \pm 1.3$
HMO	-1.2 ± 0.3	$+9.5 \pm 1.0$
	TMCy Complexes	
1-A	-2.0 ± 0.5	$+1.4 \pm 1.5$
2-A	-2.4 ± 0.3	$+0.6 \pm 1.0$
AMA	-2.4 ± 0.5	$+3.4 \pm 1.6$
MO	-5.8 ± 0.4	-2.4 ± 1.4

[a] Quoted $\pm$ uncertainties are standard error estimates.

coefficients for methyl orange species. However, the values of these extinction coefficients obtained with the various series of experiments involving different adamantanyl substrates were always consistent to within their uncertainty estimates. In other words, absorbance measurements of approximately twelve solutions containing HMO, MO, Cy and substrate 1-A, for example, yielded extinction coefficients for various MO species that differed by less than one standard deviation from values obtained from a different series of solutions using substrate 2-A or ADA. Similarly, MO and HMO complexation constants obtained from different series of experiments were consistent to within one standard deviation and so we report an average value for these constants in Table I. Complexation constants and their uncertainties for each cycloamylose-substrate system are also shown in Table I. Standard enthalpies and entropies of formation of the various complexes are calculated by fitting least squares van't Hoff lines to the temperature-dependent equilibrium constant values. Standard error estimates are calculated from the scatter of points about the regression line. These results are shown in Table II.

3. Discussion

We begin by focusing on cycloamylose complexations of the three cationic adamantanyl substrates. The results shown in Table I indicate that corresponding complexes of these substrates with Cy and DMCy have nearly identical strengths while corresponding complexes with TMCy are about two orders of magnitude weaker.

Several conclusions seem clear from these results. First, methylation of the primary hydroxyl rim (6-OH, the 'narrow' rim) has little effect on complex strength which suggests that the narrow rim does not play a significant role in these cycloamylose complexations. Second, complete methylation of Cy to form TMCy results in much weaker complexation properties and indicates that secondary hydroxyl groups present at the Cy and DMCy wide rim play an important role in complex formation. Next, we note that AMA complexes of the three cycloamyloses are consistently stronger than with 1-A. AMA complexes with Cy and DMCy are about four times stronger at 25°C than 1-A complexes and the AMA · TMCy complex is about six times stronger than the 1-A complex. The two substrates differ only by a methylene group inserted between the ammonium and adamantanyl sites and we interpret the enhanced AMA complexation as resulting from closer interaction between ammonium and the cycloamylose wide rim. This view seems to be supported by the ΔS^0 results. AMA complexes have consistently more positive ΔS^0 values than the 1-A or 2-A complexes and this may result from expulsion of outer sphere solvation water from ammonium upon complexation. This would presumably increase system disorder and lead to more positive ΔS^0 of complexation. This interpretation is also consistent with the observation that ΔS^0 for AMA complexation with Cy, which is about 9 cal mol^{-1} K^{-1} more positive than for the 1-A complex, decreases to a value near 2 cal mol^{-1} K^{-1} for the less polar methylated DMCy and TMCy derivatives. In any case, it seems clear that substrate/cycloamylose interactions in these complexes is signifigantly dependent on the geometric features of the substrate as well as on cycloamylose properties.

ΔH^0 values for adamantanyl complexations are more exothermic with Cy than with its methylated derivatives, with the principal changes resulting from methylation at 2-OH. This may be a result of steric interference between substituent methoxyl groups and the substrate if we assume similar cycloamylose conformations in the complexes. (X-ray crystallographic evidence indicates that uncomplexed Cy and TMCy have similar conformations at least in the solid phase [10, 11].) Steric strain in the methylated complexes provides a positive contribution to ΔH^0 and may account for the substantial difference in complexation enthalpies between Cy and DMCy.

Less exothermic complexation ΔH^0 values of the methylated cycloamyloses may also be related to a loss of interglucosyl hydrogen-bonding in their complexes. Such interactions have been shown to exist in Cy [12, 13] and these may be enhanced by conformational changes due to complexation. However, interglucosyl hydrogen bonding is almost certainly much weaker in the methylated cycloamyloses and so little or no bonding enhancement can occur upon complexation. This would result in less negative ΔH^0 values for DMCy and TMCy complexations than for Cy complexations, as observed. Several other interpretations are also possible. These rely on differing solvent interactions of the primary 6-OH versus 6-OCH_3 groups in the methylated derivatives as well as possible differences in solvent interactions of the 3-OH groups present in both Cy and DMCy. Definitive explanations of these cycloamylose complexations will require a much larger body of data than is presently available. Nevertheless, it is apparent that inter- and intramolecular hydrogen bonding properties of Cy and its methyl-derivatives play a significant role in the complexation properties of these compounds.

We now turn to a brief discussion of methyl orange complexations and note that Cy, DMCy and TMCy complexes with the anion indicator form have formation constants in approximate ratios of 1 : 10 : 1, respectively. Both Cy and DMCy form complexes with HMO, the DMCy complexes being about five times stronger, but no complex between TMCy and HMO could be detected. This latter observation means that a TMCy · HMO complex, if it forms at all, is at least 100 times weaker than the anion complex.

Interpretation of these results is complicated by the complex properties of the MO and HMO substrate species. MO and HMO are capable of *cis–trans* azo group isomerism and HMO may also exist in at least two acid–base tautometric forms. Several differing equilibrium distributions of the tautometric and isomeric forms of methyl orange in their cycloamylose complexes are indicated by differing species extinction coefficients, not shown here. A few examples will suffice to illustrate the complex behavior we have observed. On the one hand, we observe that the molar extinction coefficient for uncomplexed MO is near $5.2 \times 10^3\ cm^{-1}\ M^{-1}$ at 17°C and decreases slowly to a value about 3% less near 50°C. On the other hand, Cy complexed MO has an extinction coefficient of about $2.5 \times 10^3\ cm^{-1}\ M^{-1}$ which increases by about 25% at 50°C. In contrast, MO complexed with DMCy has a temperature-independent extinction coefficient near $2.0 \times 10^3\ cm^{-1}\ M^{-1}$, while MO complexes with TMCy has an absorptivity whose value of $2.5 \times 10^3\ cm^{-1}\ M^{-1}$ at 17°C decreases slowly with increasing temperature. HMO complexes of Cy and DMCy have extinction coefficients whose values are about 60% and 30%, respectively, of that of uncomplexed HMO and have large and differing temperature coefficients. It seems clear that this unusual behavior reflects differing distributions of *cis–trans* isomers in MO complexed and uncomplexed species and both isomeric and tautomeric behavior in HMO species. ΔH^0 and ΔS^0 data reflect the differing cycloamylose interactions with each of the several substrate species and thus are difficult to interpret in simple terms.

References

1. M. L. Bender and M. Komiyama: *Cyclodextrin Chemistry*, Springer-Verlag: New York (1978) and references therein.
2. K. Takeo and T. Kuge: *Stärke* **24**, 331 (1972).
3. E. A. Lewis and L. D. Hansen: *J. Chem. Soc., Perkin Trans.* 2, 2081 (1973).
4. M. R. Eftink and J. C. Harrison: *Bioorg. Chem.* **10**, 388 (1981).
5. L. A. Selvidge and M. R. Eftink: *Anal. Biochem.* **154**, 400 (1986).
6. T. Matsui, D. H. Evans, T. Osa, and N. Kabayashi: *J. Am. Chem. Soc.* **107**, 3411 (1985).
7. R. I. Gelb, L. M. Schwartz, B. Cardelino, H. Fuhrman, R. F. Johnson, and D. A. Laufer: *J. Am. Chem. Soc.* **103**, 1750 (1981).
8. R. I. Gelb, L. M. Schwartz, B. Cardelino, and D. A. Laufer: *Anal. Biochem.* **103**, 362 (1980).
9. R. A. Robinson and R. H. Stokes: *Electrolyte Solutions*, 2nd ed., Butterworths: London (1965).
10. B. Casu, M. Reggiani, G. G. Gallo, and A. Vigevani: *Tetrahedron* **24**, 803 (1968).
11. B. Casu, M. Reggiani, G. G. Gallo, and A. Vigevani: *Carbohyd. Res.* **12**, 157 (1970).
12. B. Casu, M. Reggiani, G. G. Gallo, and A. Vigevani: *Tetrahedron* **22**, 3061 (1966).
13. M. St. Jaques, P. R. Sundarrajan, K. J. Taylor, and R. H. Marchessault: *J. Am. Chem. Soc.* **98**, 4386 (1976).