ELSEVIER

Thermochimica Acta 399 (2003) 203–208

thermochimica acta

www.elsevier.com/locate/tca

The formation of amino acid and dipeptide complexes with α-cyclodextrin and cucurbit[6]uril in aqueous solutions studied by titration calorimetry

H.-J. Buschmann [a,*], E. Schollmeyer [a], L. Mutihac [b]

[a] *Deutsches Textilforschungszentrum, Nord-West e.V., Adlerstrasse 1, D-47798 Krefeld, Germany*
[b] *Department of Analytical Chemistry, Faculty of Chemistry, University of Bucharest, 4-12 Regina Elisabeta Blvd., 703461 Bucharest, Romania*

Received 3 June 2002; received in revised form 14 August 2002; accepted 16 August 2002

Abstract

The complex stabilities and the thermodynamic data for the complexation of α-cyclodextrin and cucurbit[6]uril with some amino acids (glycine, L-alanine, L-valine, L-phenylalanine, 6-amino hexanoic acid, 8-amino octanoic acid, 11-amino undecanoic acid) and dipeptides (glycyl–glycine, glycyl–L-valine, glycyl–L-leucine and glycyl–L-phenylalanine) have been determined in aqueous solution by calorimetric titrations. The complex formation with α-cyclodextrin is mainly favoured by entropic contributions due to the release of water molecules from the cavity of the ligand. The values of the reaction enthalpies are small with the exception of 11-amino undecanoic acid. In case of the ligand cucurbit[6]uril, ion–dipole interactions between the protonated amino groups of the amino acids and the carbonyl groups take place. By steric reasons these interactions are lowered for native amino acids because the polar carboxylic groups are always located outside the hydrophobic cavity of cucurbit[6]uril. The complexes of both ligands with dipeptides in water are characterised by hydrophobic interactions and in case of cucurbit[6]uril by additional ion–dipole interactions.

Keywords: Cyclodextrin; Cucurbit[6]uril; Calorimetry; Titration; Amino acids; Dipeptides

1. Introduction

α-Cyclodextrin is formed during the enzymatic degradation of starch [1,2]. It is a polysaccharide built from six D-glucose units. These D-glucose units are covalently linked at the carbon atoms C_1 and C_4 to form a torus shaped molecule with a rigid cavity. In this cavity different organic guest molecules can be enclosed [3–8]. The distribution of hydrophilic and hydrophobic groups of cyclodextrins confers them specific properties with many applications.

Cucurbit[6]uril is a high symmetric molecule like α-cyclodextrin. It is formed from urea, glyoxal and formaldehyde during an acid-catalysed reaction [9]. Six glycoluril units form a rigid molecule with a cavity. The structures of both ligands is shown in Fig. 1. Both the natural and the synthetic ligand are closely related like brother or sisters as can be shown from Table 1. The most important difference is their solubility in aqueous solution. The solubility of cucurbit[6]uril increases in the presence of salts or acids [9]. Both ligands are able to enclose a large number of

* Corresponding author. Tel.: +49-2151-843-210; fax: +49-2151-843-143.
E-mail address: buschmann@dtnw.de (H.-J. Buschmann).

0040-6031/02/$ – see front matter
PII: S0040-6031(02)00462-8

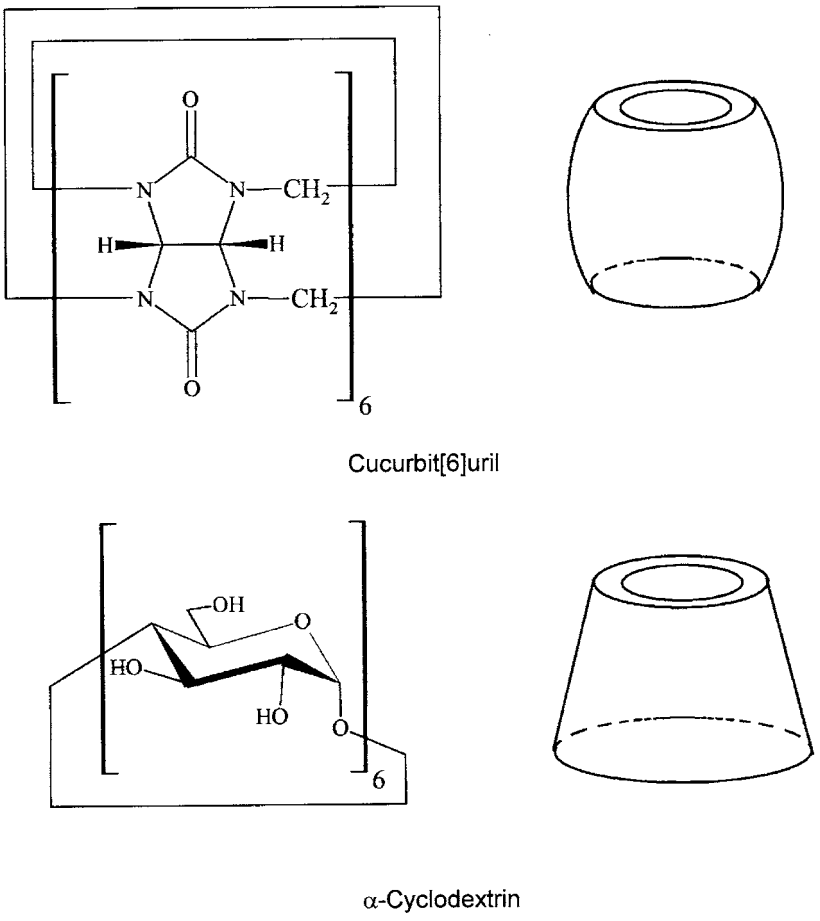

Fig. 1. Structures of α-cyclodextrin and cucurbit[6]uril.

Table 1
Properties of α-cyclodextrin [16] and cucurbit[6]uril

Properties	α-Cyclodextrin	Cucurbit[6]uril
Total formula	$C_{36}H_{60}O_{30}$	$C_{30}H_{36}N_{24}O_{12}$
Number of monomer units	6	6
Molecular weight	972	997
Solubility in water ($g\,l^{-1}$) at 25 °C	145	0.018
Cavity diameter (nm)	0.47–0.53	0.55
Height of molecule (nm)	0.79	0.6
Water content of the crystals (mol)	6.0–7.5	10–12

guest molecules within their cavities. For example, the formation of nobel gas complexes with cyclodextrins has been already reported many years ago [10]. The corresponding xenon complex with cucurbit[6]uril has been described recently [11]. Both ligands form complexes with e.g. amines [12–20], aliphatic alcohols [21–23] and carboxylic acids [23,24]. Even large dye molecules are complexed by cyclodextrins [25] and cucurbit[6]uril [26]. The number of cyclodextrin complexes studied during the last decades is enormous [27,28].

Up to now much less is known about the complex formation with cucurbit[*n*]urils. For a better understanding of the factors influencing the complex formation with α-cyclodextrin and cucurbit[6]uril, we

studied the complexation of some amino acids and peptides with both ligands.

2. Experimental

The α-cyclodextrin (pharmaceutical grade) was purchased from Wacker (Germany). The ligand cucurbit[6]uril was synthesised and purified according to published procedures [17]. The following amino acids: glycine (gly), L-alanine (L-ala), L-valine (L-val), L-phenylalanine (L-phe), 6-amino hexanoic acid, 8-amino octanoic acid and 11-amino undecanoic acid (all Fluka) were of the highest purity commercially available. The glycyl–glycine (gly–gly), glycyl–L-leucine (gly–leu), glycyl–L-valine (gly–val) and glycyl–L-phenylalanine (gly–phe) were obtained from Fluka. All amino acids and peptides were used without further purification.

The complexation of amino acids and peptides with cucurbit[6]uril was carried out in aqueous formic acid (50% (v/v), Merck) and with α-cyclodextrin in bidistilled water as solvent. The stability constants and reaction enthalpies were determined by calorimetric titrations using a Tronac Model 450 calorimeter (TRONAC, Orem Utah, USA).

During these titrations a solution of the corresponding ligand (0.04–0.08 mol l^{-1}) was added continuously to a solution of the amino acid or peptide ((2–5) × 10^{-3} mol l^{-1}). The measured heat Q is related to the reaction enthalpy ΔH and the number of moles of the complexes formed Δn by the following equation:

$$Q = \Delta H\, \Delta n$$

The Δn depends on the stability of the complex. The calculation of the stability constants from the titration curves has been described in the literature in detail [29]. Two typical titration curves are shown in Fig. 2. The reaction of cucurbit[6]uril with L-val is exothermic and the complexation of the dipeptide gly–val with α-cyclodextrin endothermic.

The accuracy of the calorimeter used for these titrations was checked from the reaction of 18-crown-6 with $BaClO_4$ in aqueous solution. The observed stability constant ($\log K = 3.55 \pm 0.03$) and reaction enthalpy ($\Delta H = -31.9 \pm 0.5$ kJ mol^{-1}) is in accordance with published results [30].

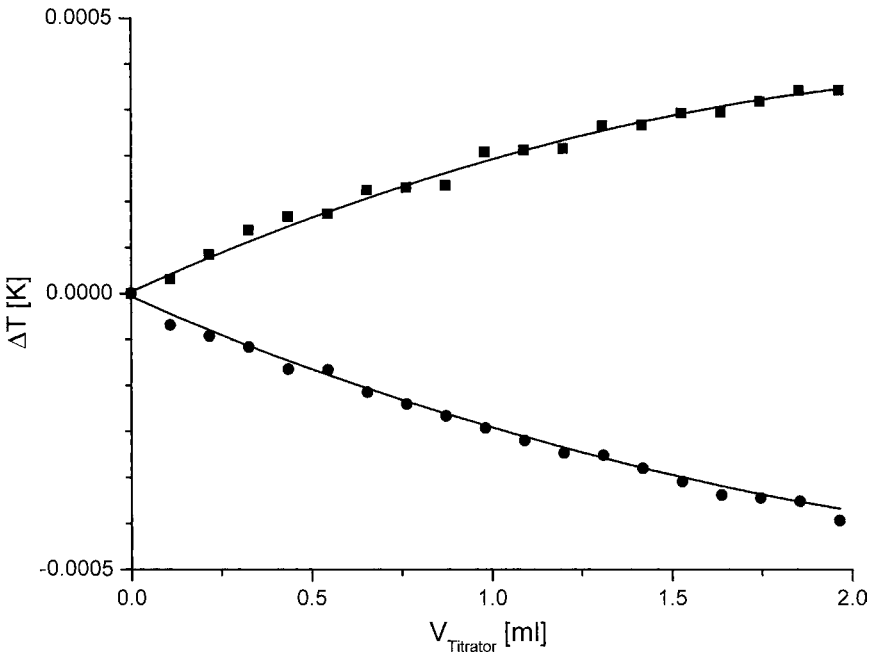

Fig. 2. Temperature changes during the titration of cucurbit[6]uril with L-val (■) in aqueous formic acid (50% (v/v)) and of α-cyclodextrin with gly–val (●) in aqueous solution at 25 °C.

3. Results and discussion

The values of stability constants and thermodynamic parameters for the complex formation between α-cyclodextrin and cucurbit[6]uril with amino acids and dipeptides in water or in aqueous formic acid are given in Tables 2 and 3.

It is well known that at neutral pH, the amino acids exists as the zwitterionic form bearing amino, carboxyl and hydrophobic groups. From the studies of complexation of β-cyclodextrin with L-isoleucine in aqueous solution at different pH, the same results at low pH range and in the neutral pH range have been reported. At high values of pH the amino acid is in anionic form differing values of the complex stabilities have been observed. In this case other reactions between the host and the guest may take place [31].

Table 2
Stability constants $\log K$ (K in $\mathrm{l\,mol^{-1}}$) and thermodynamic values ΔH and $T\Delta S$ ($\mathrm{kJ\,mol^{-1}}$) for the complex formation of amino acids with α-cyclodextrin in aqueous solution and with cucurbit[6]uril in formic acid (50% (v/v)) at 25 °C (together with literature data)

Amino acid	Value	α-Cyclodextrin	Cucurbit[6]uril
L-Phe	$\log K$	2.41 ± 0.03, 1.18[a]	3.16 ± 0.01
	$-\Delta H$	1.6 ± 0.9, 7.0[a]	6.7 ± 1.7
	$T\Delta S$	12.1 ± 1.1, −0.3[a]	11.3 ± 1.7
Gly	$\log K$	2.75 ± 0.05	–[b]
	$-\Delta H$	2.0 ± 0.6	–
	$T\Delta S$	13.6 ± 0.9	–
L-Ala	$\log K$	3.05 ± 0.02	3.02 ± 0.04
	$-\Delta H$	8.0 ± 0.4	7.0 ± 0.8
	$T\Delta S$	9.3 ± 0.5	10.2 ± 1.0
L-Val	$\log K$	3.21 ± 0.03	3.15 ± 0.05
	$-\Delta H$	5.8 ± 0.7	4.5 ± 0.8
	$T\Delta S$	12.4 ± 0.9	13.4 ± 1.1
6-Amino hexanoic acid	$\log K$	3.18 ± 0.02, 1.34[a]	3.57[c]
	$-\Delta H$	4.0 ± 0.6, 1.5[a]	12.0[c]
	$T\Delta S$	14.1 ± 0.7, 6.0[a]	8.4[c]
8-Amino octanoic acid	$\log K$	2.99 ± 0.04, 1.88[a]	3.27[c]
	$-\Delta H$	4.7 ± 0.5, 14.8[a]	12.2[c]
	$T\Delta S$	12.3 ± 0.7, −4.1[a]	6.4[c]
11-Amino undecanoic acid	$\log K$	3.16 ± 0.03, 3.34[a]	1.93[c]
	$-\Delta H$	30.0 ± 0.8, 26.6[a]	5.2[c]
	$T\Delta S$	−12.1 ± 1.0, −4.6[a]	5.8[c]

[a] [12].
[b] Heat effect too small for the calculation of stability constants and reaction enthalpies.
[c] [23].

Table 3
Stability constants $\log K$ (K in $\mathrm{l\,mol^{-1}}$) and thermodynamic values ΔH and $T\Delta S$ (in $\mathrm{kJ\,mol^{-1}}$) for the complex formation of dipeptides with α-cyclodextrin in aqueous solution and with cucurbit[6]uril in formic acid (50% (v/v)) at 25 °C

Dipeptide	Value	α-Cyclodextrin	Cucurbit[6]uril
Gly–phe	$\log K$	–[a]	3.05 ± 0.04
	$-\Delta H$		9.6 ± 0.8
	$T\Delta S$		7.7 ± 0.9
Gly–gly	$\log K$	–[a]	2.90 ± 0.02
	$-\Delta H$		14.9 ± 0.5
	$T\Delta S$		1.6 ± 0.6
Gly–leu	$\log K$	2.42 ± 0.09	2.57 ± 0.07
	$-\Delta H$	0.9 ± 0.1	6.9 ± 0.3
	$T\Delta S$	12.9 ± 0.7	7.7 ± 0.7
Gly–val	$\log K$	2.45 ± 0.01	3.19 ± 0.04
	$-\Delta H$	−1.0 ± 0.2	1.7 ± 0.8
	$T\Delta S$	14.9 ± 0.2	16.4 ± 1.1

[a] Heat effect too small for the calculation of stability constants and reaction enthalpies.

All amino acids examined form complexes with α-cyclodextrin and with the exception of glycine also with cucurbit[6]uril. Only very few results for the reaction with α-cyclodextrin are available from the literature. They have been included in Table 2 for comparison. With the exception of 11-amino undecanoic acid the published stability constants are at least one order of magnitude smaller than those measured in this work. No data for the complexation of the peptides used have been published up to now.

The stabilities of the complexes formed with the ligands α-cyclodextrin and cucurbit[6]uril are similar. However, the values of the reaction enthalpies and entropies are different for both ligands. Thus, different contribution to the measured thermodynamic parameters have to be considered. In the cavity of α-cyclodextrin, two water molecules are included [32]. These water molecules are not able to form hydrogen bonds as in the bulk phase and therefore they are named “high-energy water” by Saenger [32]. The release of these water molecules from the cavity should result in favourable enthalpic and entropic contributions. No strong additional interactions between the amino acids and α-cyclodextrin are expected during the complex formation. As a result, small values of the reaction enthalpies are observed. Only in case of 11-amino undecanoic acid a high value of the

reaction enthalpy is found. The positive and nearly constant values of the reaction entropies can be attributed to the release of two water molecules from the cavity. The molar entropy of fusion of water is $T\Delta S = 6.6\,\mathrm{kJ\,mol^{-1}}$ [33]. Obviously, other possible contributions as e.g. changes in the solvation or decrease of the molecular freedom of the host and guest molecule have only minor contributions to the overall reaction entropy.

In the cavity of cucurbit[6]uril, three water molecules are enclosed and additional water molecules are located at the carbonyl groups of the ligand [34]. As discussed for α-cyclodextrin, the release of water molecules from the cavity only results in small enthalpic contributions. The main contribution to the reaction enthalpies results from ion–dipole interactions between the protonated amino group of the amino acids and the carbonyl groups of cucurbit[6]uril. In case of the natural amino acids, these interactions are lowered for steric reasons because the polar carboxylic groups are always located outside the hydrophobic cavity of cucurbit[6]uril. The different contributions to the reaction entropies are comparable to those for α-cyclodextrin. However, due to the interactions between the protonated amino groups and the carbonyl groups additional solvent molecules are released during the complex formation.

The complex formation of α-cyclodextrin with dipeptides is only favoured by the reaction entropy. Again the release of two water molecules from the cavity seems to be the most important contribution. The values of the reaction enthalpies are close to zero. The formation of complexes with cucurbit[6]uril is favoured as well by enthalpic as by entropic contributions. The ion–dipole interactions are the reason for this behaviour.

Both α-cyclodextrin and cucurbit[6]uril are able to form complexes with amino acids and dipeptides. Their chemical structures look very similar. Even the stabilities of the complexes formed are of the same order of magnitude. Thus, one may expect these ligands really to act nearly identical. At the end different enthalpic and entropic contributions to the overall reaction enthalpies and entropies have to be considered for both ligands. In case of α-cyclodextrin, the hydrophobic interactions are responsible for the observed values. For cucurbit[6]uril, ion–dipole interactions are the important factor.

Acknowledgements

Financial support of this research project by the NATO under the research grant no. LST.CLG 977861 is gratefully acknowledged.

References

[1] A. Villiers, Compt. Rend. Acad. Sci., Paris 112 (1891) 435.
[2] F. Schardinger, Z. Untersuch. Nahrungsm. Genussm. 6 (1903) 865.
[3] F. Cramer, Einschlussverbindungen, Springer, Heidelberg, 1954.
[4] F. Cramer, W. Saenger, H. Spatz, J. Am. Chem. Soc. 89 (1967) 14.
[5] J. Szejtli, Cyclodextrin Technology, Kluwer Academic Publishers, Dordrecht, 1988.
[6] D. Duchêne, Cyclodextrin and Their Industrial Uses, Editions de Santé, Paris, 1987.
[7] H.-J. Buschmann, D. Knittel, E. Schollmeyer, J. Incl. Phenom. 40 (2001) 169.
[8] J.-M. Lehn, Supramolecular Chemistry, Concepts and Perspectives, VCH, Weinheim, 1995.
[9] R. Behrend, E. Meyer, F. Rusche, Liebigs Ann. Chem. 339 (1904) 1.
[10] F. Cramer, F.M. Henglein, Chem. Ber. 90 (1957) 2572.
[11] M. El Haouaj, Y.H. Ko, M. Luhmer, K. Kim, K. Bartik, J. Chem. Soc., Perkin Trans. 2 (2001) 2104.
[12] G. Castronuovo, V. Elia, D. Fessas, A. Giordano, F. Velleca, Carbohydr. Res. 272 (1995) 31.
[13] W.L. Mock, N.-Y. Shih, J. Org. Chem. 48 (1983) 3618.
[14] W.L. Mock, N.-Y. Shih, J. Org. Chem. 51 (1986) 4440.
[15] H.-J. Buschmann, E. Cleve, E. Schollmeyer, Inorg. Chim. Acta 193 (1992) 93.
[16] R. Hoffmann, W. Knoche, C. Fenn, H.-J. Buschmann, J. Chem. Soc., Faraday Trans. 90 (1994) 1507.
[17] K. Jansen, H.-J. Buschmann, A. Wego, D. Döpp, C. Mayer, H.-J. Drexler, H.-J. Holdt, E. Schollmeyer, J. Incl. Phenom. 39 (2001) 357.
[18] C. Meschke, H.-J. Buschmann, E. Schollmeyer, Thermochim. Acta 297 (1997) 43.
[19] H.-J. Buschmann, E. Cleve, K. Jansen, A. Wego, E. Schollmeyer, J. Incl. Phenom. 40 (2001) 117.
[20] H.-J. Buschmann, L. Mutihac, K. Jansen, J. Incl. Phenom. 39 (2001) 1.
[21] S. Andini, G. Castronuovo, V. Elia, E. Gallota, Carbohydr. Res. 217 (1991) 87.
[22] M.V. Rekharsky, F. Schwarz, Y.B. Tewari, R.N. Goldberg, J. Phys. Chem. 98 (1994) 10282.
[23] H.-J. Buschmann, K. Jansen, E. Schollmeyer, Thermochim. Acta 317 (1998) 95.
[24] G. Castronuovo, V. Elia, D. Fessas, F. Velleca, G. Viscardi, Carbohydr. Res. 287 (1996) 127.
[25] H.-J. Buschmann, E. Schollmeyer, J. Incl. Phenom. 14 (1992) 91.

[26] H.-J. Buschmann, E. Schollmeyer, J. Incl. Phenom. 29 (1997) 167.
[27] M.V. Rekharsky, Y. Inoue, Chem. Rev. 98 (1998) 1875.
[28] P.D. Ross, M.V. Rekharsky, Biophys. J. 71 (1996) 2144.
[29] H.-J. Buschmann, Inorg. Chim. Acta 195 (1992) 51.
[30] H.-J. Buschmann, E. Schollmeyer, Thermochim. Acta 333 (1999) 49.
[31] T. Ugawa, S. Nishikawa, J. Phys. Chem. A 105 (2001) 4248.
[32] W. Saenger, M. Noltemeyer, P.C. Manor, B. Hingerty, B. Klar, Bioorg. Chem. 5 (1976) 187.
[33] A.A. Bondi, Physical Properties of Molecular Crystals, Liquids and Glass, Wiley, New York, 1968.
[34] P. Germain, J.M. Letoffe, M.P. Merlin, H.-J. Buschmann, Thermochim. Acta 315 (1998) 87.