Available online at www.sciencedirect.com
ScienceDirect
Mendeleev Commun., 2005, 15(1), 38–40

Mendeleev Communications

Calorimetric and spectrophotometric study on the interaction of hydroxypropyl-β-cyclodextrin with ascorbic acid

Irina V. Terekhova* and Natalya A. Obukhova

Institute of Solution Chemistry, Russian Academy of Sciences, 153045 Ivanovo, Russian Federation.
Fax: +7 0932 33 6237; e-mail: ivt@isc-ras.ru

DOI: 10.1070/MC2005v015n01ABEH001901

Hydroxypropyl-β-cyclodextrin forms a 1:1 molecular complex with ascorbic acid and exhibits slight affinity to the ascorbate anion in aqueous solutions, as found by calorimetry and UV spectrophotometry.

Cyclodextrins (CDs) are cyclic oligosaccharides composed of six, seven or eight glucose units. They have an external hydrophilic surface and a hydrophobic cavity capable of including different guest molecules with appropriate shape and size.[1] The ability of CDs to form inclusion complexes causes practical applications of CDs in pharmaceutical, food and cosmetic industries, separation technology and enzymatic catalysis.[2] Chemically modified CDs are especially interesting due to their improved physico-chemical properties (such as higher water solubility), lower toxicity and higher selectivity in complexation with guest molecules.[3]

It is well known that ascorbic acid is easily oxidised to inactive species.[4] The bioavailability and stability of ascorbic acid can be enhanced due to encapsulation by CDs. The pro-

tective action of CDs was studied.[5–8] In particular, the inclusion of ascorbic acid in β-CD increases the stability of the guest molecule to oxidising agents in acidic (0.5 M H_2SO_4) and neutral solutions.[5,6] Hydroxypropyl-β-cyclodextrin (HP-β-CD) acts as a secondary antioxidant preventing the oxidation of ascorbic acid.[8] Previously,[9,10] we reported on the thermochemical investigations of interactions between ascorbic acid and natural α- and β-CDs and their monomer units such as glucose and maltose. Here we consider the thermodynamics of the interaction of HP-β-CD with ascorbic acid in aqueous solutions.

HP-β-CD from Aldrich was used (water content of 27.6%; the average molar substitution for each glucopyranose residue was 0.6). Ascorbic acid was additionally purified by recrystallization from water–ethanol mixtures. All solutions were prepared in twice-distilled water.

The enthalpies of solution of HP-β-CD in pure water and freshly prepared aqueous solutions of ascorbic acid were obtained at 298.15 K using a calorimeter of solution. The concentration of HP-β-CD was constant (1.00±0.05)×10^{-3} mol kg^{-1}, while the ascorbic acid concentration was changed from 0 to 0.65 mol kg^{-1} to obtain a saturation curve and maximal yields of complexes. Experimental pH values (pH 2.2–2.5) and the calculations of the ascorbic acid equilibrium concentrations using the RRSU computer program[11–13] showed that, in the concentration range under study, the undissociated form of ascorbic acid (H_2A) is predominant. Potassium hydroxide was used for adjusting pH 7.5 at which the content of an ionised species (HA^-) is maximal.

The experimental enthalpies of solution of HP-β-CD in pure water and aqueous ascorbic acid solutions formed the basis for the calculation of the enthalpy of transfer ($\Delta_{tr}H$). The thermodynamic parameters of complex formation were estimated using the HEAT computer program,[14] in which the search of unknown parameters lg K and Δ_cH^0 is reduced to the numerical minimization of functional F:

$$F = \sum_i^N w_i(\Delta H_{i\,\mathrm{exp}} - \Delta H_{i\,\mathrm{calc}})^2, \quad (1)$$

where ΔH_i is the thermal effect of i-th reaction, N is the number of experiments, w_i is the weight factor calculated as $w_i = A/(\delta\Delta H_i)^2$ (where A is the coefficient chosen from the condition $\sum w_i = N$, and $\delta\Delta H_i$ is the absolute error of the measurement of ΔH_i).

In the case of weak interactions (without complex formation), enthalpic virial coefficients were obtained according to the MacMillan–Mayer theory:[15,16]

$$\Delta_{tr}H/m_y = 2h_{xy} + 3h_{xyy}m_y + 3h_{xxy}m_x + \ldots, \quad (2)$$

where m_x and m_y are the molalities of HP-β-CD and ascorbic acid, respectively; h_{xy}, h_{xyy} and h_{xxy} are the enthalpic coefficients of pair and triplet interactions.

The UV spectra were recorded at room temperature (273 K) in the range 210–300 nm using a Specord M40 spectrophotometer. Quartz cells with a path length of 1 cm were employed. The spectrophotometric investigation was carried out at a constant concentration of ascorbic acid (7×10^{-5} mol kg^{-1}), a variable concentration of HP-β-CD (from 0 to 0.097 mol kg^{-1}) and pH 2 (HCl) in order to reach the prevalence of the undissociated acid H_2A. Under these conditions, ascorbic acid has a UV band with λ_{max} = 244 nm and ε = 10000 kg mol^{-1} cm^{-1} obtained from the Lambert–Beer dependence.

Figure 1 summarises new experimental calorimetric data in combination with our previous data.[9,10] The linear dependence $\Delta_{tr}H$ (m_{AA}) [Figure 1(*a*)] corresponds to systems in which weak interactions without complex formation occur. For these systems, the enthalpic coefficients of pair interactions h_{xy} were calculated. Coefficients h_{xy} reflect all energetic changes resulting from solute–solute and solute–solvent interactions.[17] As it can be seen in Figure 1(*a*), the interaction of undissociated ascorbic acid with β-CD is weak and characterised by a negative value of h_{xy} (h_{xy} = –1800±900 J kg mol^{-2})[9] caused by a dominant contribution from the most probable van der Waals interactions and weak H-bonding. In spite of the fact that the interaction of β-CD with ascorbic acid is enthalpically favourable, a complex is not formed.

HP-β-CD has flexible hydroxypropyl groups, which can participate in additional interactions with hydrophilic parts of the guest molecule and enhance the selectivity of complexation and the stability of host–guest complexes.[18,19] The opposite behaviour was also found when the hydroxypropyl groups served as sterical hindrances for the inclusion of the guest molecule.[20] As follows from Figure 1(*b*), HP-β-CD forms

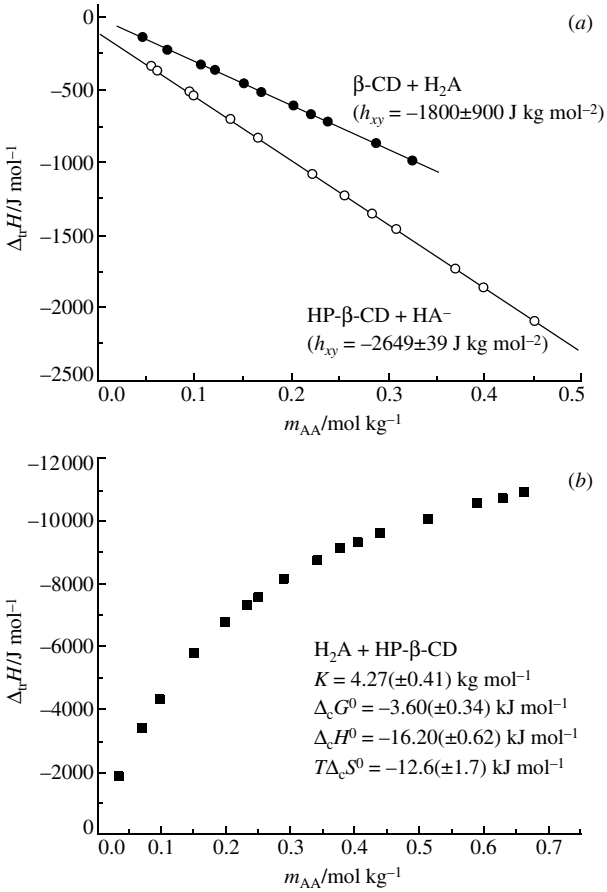

Figure 1 Enthalpy of the transfer of cyclodextrins from water to aqueous solutions of ascorbic acid *vs*. ascorbic acid concentration (T = 298.15 K).

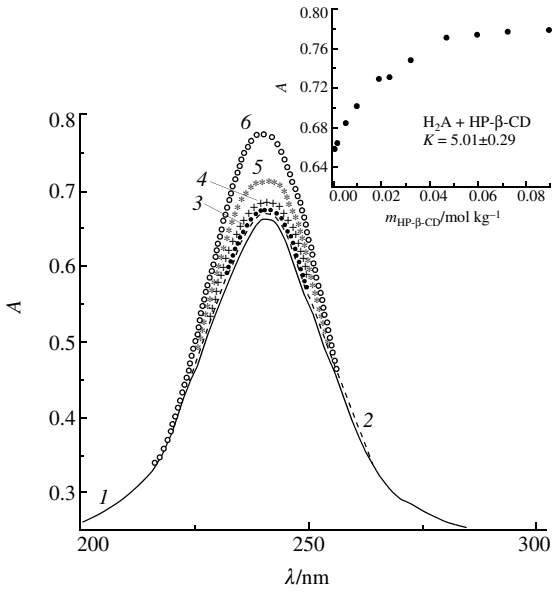

Figure 2 UV spectra of ascorbic acid with the addition of HP-β-CD: (*1*) 0; (*2*) 5.5×10^{-4}; (*3*) 1.0×10^{-3}; (*4*) 5.5×10^{-3}; (*5*) 1.0×10^{-2}; (*6*) 5.6×10^{-2} and 9.7×10^{-2} mol kg^{-1} (pH 2, T 293 K). Inset: absorbance of aqueous ascorbic acid solutions as a function of HP-β-CD concentration (λ = 244 nm).

a very weak molecular complex with ascorbic acid (K = = 4.27±0.41 kg mol^{-1}). The complex formation is characterised by a negative enthalpy of complexation ($\Delta_c H^0$ = = –16.20±0.62 kJ mol^{-1}) and a negative entropy of complexation ($T\Delta_c S^0$ = –12.6±1.7 kJ mol^{-1}). Thus, the HP-β-CD/H_2A complex is stabilised by an enthalpy contribution. A large negative value of $\Delta_c H^0$ can be attributed to cohesion interactions and possible additional hydrogen-bonding between the polar groups of ascorbic acid and the hydroxypropyl groups of CD. A negative entropic term demonstrates the formation of more structured species in the test system.

The formation of the HP-β-CD/H_2A complex was confirmed by the spectrophotometric results presented in Figure 2. Figure 2 shows the effect of increasing amounts of HP-β-CD on the spectrum of ascorbic acid. The absorbance A at 244 nm increases with HP-β-CD concentration and then reaches a constant value (saturation curve presented in the insert of Figure 2). Spectrophotometric measurements were performed at constant pH. Therefore, the variation of absorbance corresponds to complex formation.[5] The molar absorptivity (ε) of ascorbic acid is changed due to the transfer of ascorbic acid from polar aqueous media to the apolar HP-β-CD cavity.[21] Calculations of the stability constant were performed for 1:1, 2:1 and 1:2 binding models using the FTMT computer program.[11] The determination of lg K is based on the statistical principle of maximal probability and is reduced to the searching of a minimum of the function F reflected the deviation of experimental A from calculated data. The very good agreement between the calculated and experimental values gives a strong indication of the existence of 1:1 complexes. The apparent stability constant of the complex (K = 5.01±0.29 kg mol^{-1}) is in accordance with the K obtained by calorimetry.

The complex formation between ascorbic acid and HP-β-CD is very sensitive to pH values. It is well known that ascorbic acid dissociates with pK_{a_1} = 4.0 and pK_{a_2} = 11.3,[22] and the molecular form H_2A (99–100%) is predominant at pH ≤ 2, while at pH 6–10 the ionised form HA^- dominates (> 95%). It can be clearly seen in Figure 1(*a*) that the ionised form HA^- does not form complexes with HP-β-CD; the interaction is weak and characterised by an exothermic effect (h_{xy} = –2649±39 J kg mol^{-2}) caused by prevailing H-binding and van der Waals interactions. Thus, HP-β-CD exhibits slight affinity to the ascorbate anion. Similar results were found for the interactions of CDs with some negatively and positively charged species, and they can be attributed to the effect of a lower hydrophobicity of charged guest molecules and relatively large and strong hydration shells in comparison with neutral molecules.[18,23]

This work was supported by the Russian Foundation for Basic Research (grant no. 03-03-96411) and the President of the Russian Federation (grant no. MK 1060.2003.03). We are grateful to O. N. Ivanova for her assistance in spectrophotometric experiments.

References

1 K. A. Connors, *Chem. Rev.*, 1997, **97**, 1325.
2 A. R. Hedges, *Chem. Rev.*, 1998, **98**, 2035.
3 M. V. Rekharsky and Y. Inoue, *Chem. Rev.*, 1998, **98**, 1875.
4 R. J. Wilson, A. E. Beezer and J. C. Mitchell, *Thermochim. Acta*, 1995, **264**, 27.
5 M. I. Manzanares, V. Solís and R. H. de Rossi, *J. Electroanal. Chem.*, 1996, **407**, 141.
6 M. I. Manzanares, V. Solís and R. H. de Rossi, *J. Electroanal. Chem.*, 1997, **430**, 163.
7 H. H. Tonnesen, *Int. J. Pharmac*., 2001, **225**, 1.
8 E. Núñez-Delicado, A. Sánchez-Ferrer and F. Garsía-Carmona, *J. Agric. Food Chem.*, 1997, **45**, 2830.
9 I. V. Terekhova and O. V. Kulikov, *Mendeleev Commun*., 2002, 111.
10 I. V. Terekhova, O. V. Kulikov and E. S. Titova, *Thermochim. Acta*, 2004, **412**, 121.
11 V. P. Vasil'ev, V. A. Borodin and E. V. Kozlovsky, *Primenenie komp'yuterov v khimiko-analiticheskikh vychisleniyakh* (*Application of Computers in Chemico-Analytical Calculations*), Vysshaya Shkola, Moscow, 1993 (in Russian).
12 A. A. Bugaevsky and B. A. Dunay, *Zh. Anal. Khim.*, 1971, **26**, 205 [*J. Anal. Chem. USSR* (*Engl. Transl.*), 1971, **26**, 183].
13 V. O. Kruglov and A. A. Bugaevsky, in *Matematika v khimicheskoi termodinamike* (*Mathematics in Chemical Thermodynamics*), Nauka, Novosibirsk, 1980, p. 36 (in Russian).
14 V. A. Borodin, E. V. Kozlovsky and V. P. Vasil'ev, *Zh. Neorg. Khim*., 1982, **27**, 2169 (*Russ. J. Inorg. Chem.*, 1982, **27**, 1224).
15 W. G. MacMillan and J. E. Mayer, *J. Chem. Phys*., 1945, **13**, 276.
16 J. E. Desnoyers, G. Perron, L. Avedikian and J. P. Morel, *J. Solution Chem*., 1976, **5**, 631.
17 J. Fernandez and T. H. Lilley, *J. Chem. Soc., Faraday Trans*., 1992, **88**, 2503.
18 E. Junquera and E. Aicart, *J. Incl. Phenom. Mol. Rec. Chem.*, 1997, **29**, 119.
19 Y. Dotsikas and Y. L. Loukas, *J. Biochem. Biophys. Methods*, 2002, **52**, 121.
20 B. Cappello, C. di Miao and M. Iervolino, *J. Incl. Phenom. Macrocycl. Chem*., 2002, **43**, 251.
21 Y. Dotsikas and Y. L. Loukas, *J. Biochem. Biophys. Methods*, 2002, **52**, 121.
22 C. Creutz, *Inorg. Chem.*, 1981, **20**, 4449.
23 M. V. Rekharsky and Y. Inoue, *J. Am. Chem. Soc*., 2000, **122**, 4418.

Received: 27th January 2004; Com. 04/2227