Pergamon

Tetrahedron Letters 40 (1999) 171–174

TETRAHEDRON LETTERS

Influence of Remote Intramolecular Hydrogen Bonds on the Thermodynamics of Molecular Recognition of cis-1,3,5-Cyclohexanetricarboxylic Acid.

Pablo Ballester, Antoni Costa, Pere M. Deyà, Manuel Vega, Jeroni Morey,

Departament de Química. Universitat de les Illes Balears 07071 Palma de Mallorca, Spain.

and Ghislain Deslongchamps

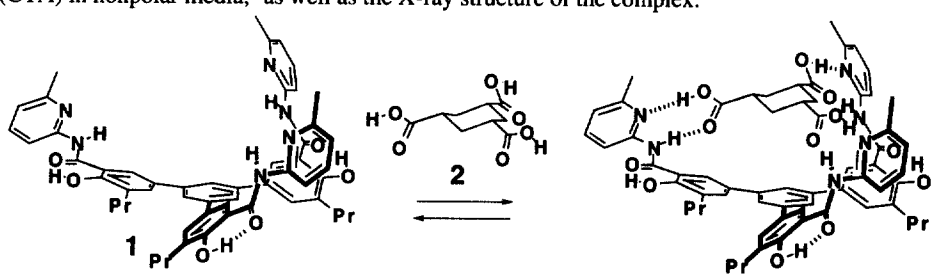
Department of Chemistry, University of New Brunswick, Fredericton, N.B., Canada E3B 6E2.

Received 15 July 1998; revised 19 October 1998; accepted 26 October 1998

Abstract: Variable temperature binding studies and isothermal titration microcalorimetry were used to probe the thermodynamics of molecular recognition of *cis*-1,3,5-cyclohexanetricarboxylic acid by tripodal hosts. Remote intramolecular hydrogen bonds, used to restrict conformationally one of the hosts, exhibit a strong influence on the thermodynamic functions for the binding process ΔH and ΔS, with little effect on ΔG. This suggests that the conformational lock imposed by the intramolecular hydrogen bonds organizes the receptor in a conformation that is not optimal for the binding of the triacid.

The complexation of organic molecules by abiotic hosts in organic solvents, where the hydrogen bond (H-bond) is the principal binding force, is characterized typically by an enthalpic benefit greater than the entropic cost.[1] The enthalpy change mainly arises from the energy release that accompanies the pairwise interaction of the H-bonding groups upon complexation. The entropic cost is a consequence of the formation of an ordered and fairly rigid bimolecular complex from two less ordered components, with the associated loss in degrees of freedom.[2] Therefore, constraining a host to a conformation close to that found in the complex (usually referred to as preorganization[3]) should be an important factor in reducing the entropy loss, thus improving the binding free energy.

We have reported the synthesis of tripodal host **1**, its recognition of *cis*-1,3,5-cyclohexanetricarboxylic acid **2** (CTA) in nonpolar media,[4] as well as the X-ray structure of the complex.[5]



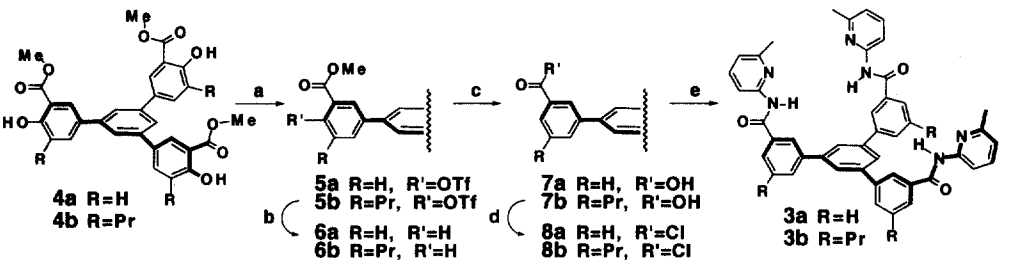
The receptor design incorporated an intramolecular H-bond between phenol and amide carbonyl groups on each of the three arms, restricting rotation about the three aryl-amide single bonds.[6] The premise was that these H-bonds would aid in preorganizing the host into an endoreceptor conformation that is optimized for triacid recognition and therefore confer an entropic advantage for the targeted complexation. However, preorganizing a

0040-4039/99/$ - see front matter
PII: S0040-4039(98)02306-5

receptor into a fixed geometry can be detrimental if the conformation is not perfectly complementary to the guest, which can lead to a reduced enthalpy of binding.

To probe these various preorganizational aspects of host-guest chemistry, we wish to report the synthesis and binding properties of the analogous tripodal receptors **3a-b** which are devoid of intramolecular H-bonded conformational restrictors, and to compare them with host **1** using a full complement of spectroscopic and thermodynamic binding analyses, including isothermal titration calorimetry.

Hosts **3a-b** were synthesized from the previously reported triesters **4a-b** which were obtained through $SiCl_4$ catalyzed cyclotrimerization of a suitably functionalized acetophenone derivative.[3] As illustrated in Scheme 1, the phenolic hydroxyls were removed by converting triesters **4a-b** to the corresponding tris aryl triflates **5a-b** followed by ring deoxygenation mediated by $Cl_2Pd(PPh_3)_2$ to afford triesters **6a** and **6b** in 81% and 63% overall yield, respectively. Hydrolysis of triesters **6a-b** afforded the triacids **7a-b** which were converted to triacyl chlorides **8a-b** and coupled with 6-methyl-2-aminopyridine to produce tripodal receptors **3a** and **3b** in 73% and 61% overall yield, respectively, from triesters **6a-b**.[7]



Scheme 1. (a) Tf_2O, CH_2Cl_2; (b) $Cl_2Pd(PPh_3)_2$, $Ph_2P(CH_2)_3PPh_2$, Bu_3N, HCO_2H/DMF, 90°; (c) NaOH, H_2O-THF; (d) $SOCl_2/CH_2Cl_2$; (e) 6-methyl-2-aminopyridine, CH_2Cl_2.

As an initial assessment of the relative binding properties of hosts **1** and **3a-b**, ^1H NMR titrations were carried out in 20% MeOH-d4/$CDCl_3$ with CTA **2**. Unexpectedly, all three receptors gave similar association constants[8] (K_a) for CTA recognition (**1**: 466 ± 8 M^{-1}, **3a**: 410 ± 14 M^{-1}, **3b**: 549 ± 51 M^{-1}). The K_a ratio between hosts **1** and **3a** was corroborated by competitive binding experiments ($CDCl_3$, 260K) which gave a K_a**1**/K_a**3a** ratio of 1.14. Apparently, the preorganization of host **1** through intramolecular H-bonding confers no clear advantage for the association of CTA.

In order to gain insight on the thermodynamic functions of the complexation process, van't Hoff analyses of the **1**-CTA and **3a**-CTA pairs using variable-temperature (VT) ^1H NMR were carried out. In accord with the guidelines established by Wilcox[1b], a full titration of the **3a**-CTA system was performed out at each temperature to determine the K_a values. The K_a values from 277 to 314K gave a linear van't Hoff plot and were retained for the ΔH and ΔS determinations. ΔH and ΔS values of -9.06±0.9 kcal/mol and -18.8±3.1 eu (R^2=0.997) were obtained. Similar results were obtained for the same system using simpler "single-point" variable temperature van't Hoff experiments (ΔH=-9.42±0.9 kcal/mol, ΔS=-19.8±3 eu, R^2=0.996), which is justified since the limiting chemical shifts of the aromatic hydrogens monitored during the titrations were almost temperature independent. To speed up the analyses, the same "single-point" VT method was used to study the **1**-CTA system, yielding values of ΔH=-6.95±0.3 kcal/mol and ΔS=-10.8±1 eu (R^2=0.999).[9,10]

These results support the view that the structural constraints imposed by the intramolecular H-bonds in host **1** have the desired effect of producing a more ordered host but, unfortunately, not in a conformation which is optimal for CTA recognition.

There is concern that ΔH values of low accuracy may be produced from VT studies.[11] A versatile and accurate alternative for the determination of all thermodynamic binding data is isothermal titration calorimetry (ITC).[12] In our case, the host concentration was adjusted to return a "c" value of 6, high enough for a reliable calorimetric determination of K_a and ΔH while maintaining concentrations as close as possible to the NMR

titration experiments.[13] Accordingly, hosts **1**, **3a**, and **3b** were titrated by ITC as ≈10 mM solutions in 20%MeOH/$CHCl_3$ with CTA. The isothermal titration calorimetry results are reported in Table 1.

Table 1. Isothermal titration calorimetry for hosts **1** and **3a-b** with CTA **2** in 20% MeOH/$CHCl_3$ at 294 K.

Host	K_a (M^{-1})	ΔH (kcal/mol)	ΔS (eu)
1	582±24	-6.90±0.1	-10.72
3a	485±33	-9.46±0.2	-19.82
3b	563±25	-10.54±0.2	-23.18

The thermodynamic values K_a and ΔH, obtained by ITC, are in general agreement with the VT NMR results. Thus, for the systems under study, the total enthalpy measured by ITC appears to be representative of the intrinsic binding enthalpy, which is not necessarily the case for other host-guest systems.[11c]

The collective results indicate clearly that the less preorganized hosts **3a-b** promote more optimal H-bonding interactions with CTA than host **1**. The conformational changes induced in host **1** upon CTA binding reduce the exothermicity of the association by ≈2.56–3.64 kcal/mol compared to **3a-b** which must correspond to the gain in energy due to the distortion of the intra- and intermolecular H-bonds in **1**-CTA. Analyzing the ^1H NMR signals of the H-bonded protons at 260K in $CDCl_3$ before and after CTA complexation provided experimental support for this hypothesis. Indeed, the phenol protons in free **1** appear at a concentration-invariant value of 12.4 ppm and shift upfield to 11.8 ppm upon CTA complexation, a sign of weakened H-bonding (assuming that the binding of an acid to an amidopyridine arm has a negligible anisotropic effect on the remote H-bonded phenol proton). In addition, both the amide NH and CTA carboxyl protons in the **1**-CTA complex are shifted upfield compared to those found in the **3a**-CTA complex (≈0.2 ppm for both) a further indication of weaker H-bonded interactions.

Although the entropy of complexation, is affected by the polar nature of the solvent, there is a clear difference in the behaviour of the two systems[2]: the ΔS values for hosts **3a-b** are larger than for host **1** by about 10 eu. This could be due to differences in desolvation on binding but since the hydroxyl groups are on the outside of the cavity, a more likely explanation is the difference in preorganization of the receptors.[14] The more flexible hosts **3a-b** can achieve better complementarity with CTA while maximizing the H-bonding interactions at the expense of an entropy loss. These systems are a beautiful abiotic example of Koshland's induced-fit hypothesis.[15,16]

In conclusion, we rationalize our resuls as follows: the rigidification of host **1** through intramolecular H-bonding has been achieved. Disappointingly, the improper preorganization of receptor **1** was indeed found to be detrimental to CTA binding, as the conformation was not perfectly complementary to the guest, leading to a reduced enthalpy of binding. Studies on the selectivity of receptors **1** and **3a-b** towards the binding of several flexible triacids are in course and will be reported in due time.

Acknowledgment. We thank the DGICYT (Projects PB96-0143 and PB95-0341) and NSERC (Canada) for financial support. This study is part of a collaborative program on molecular recognition between UIB (Spain) and UNB (Canada) funded by a NATO Collaborative Research Grant (SRG-930949).

References and Notes

1. For examples, see (a) Searle, M.S.; Westwell, M.S.; Williams, D.H. *J. Chem. Soc., Perkin Trans 2* **1995**, 141-151. (b) Adrian, J.C.; Wilcox, C.S. *J. Am. Chem. Soc.* **1991**, *113*, 678-680. (c) Williams, K.; Askew, B.; Ballester, P.; Buhr, C.; Jeong, K.-S.; Jones, S.; Rebek, J. Jr. *J. Am. Chem. Soc.* **1989**, *111*, 1090-1094. (d) Sheridan, R.E.; Whitlock, H.W. Jr. *J. Am. Chem. Soc.* **1988**, *110*, 4071-4074. (e) van

Staveren, C.J.; Aarts, V.M.L.J.; Grootenhuis, P.D.J.; van Eerden, J.; Harkema, S.; Reinhoudt, D.N. *J. Am. Chem. Soc.* **1986**, *108*, 5271-5276.

2. The entropy cost for the association of two molecules of low molecular mass (100-300 Da) has a limiting value of 50-60 kJ mol^{-1} ($T\Delta S^{\circ}$ at 298 K). The observed values of $T\Delta S^{\circ}$ are generally much lower due to residual motion and/or desolvation. See Williams, D.H.; Westell, M.S. *Chem. Soc. Rev.*, **1998**, *27*, 57-63 and references cited therein.
3. Cram, D.J. *Angew. Chem. Int. Ed. Engl.* **1986**, *25*, 1039-1134.
4. Ballester, P.; Costa, A.; Deyà, P.M.; González, J.F.; Rotger, M.C.; Deslongchamps, G. *Tetrahedron Lett.* **1994**, *35*, 3813-3816.
5. Ballester, P., Costa, A., Deyà, P.M., Deslongchamps, G., Mink, D., Decken, A., Prohens, R. Tomàs, S., Vega, M., *J. Chem. Soc. Chem. Commun.* **1997**, 357-358.
6. For another example of H-bonds as host conformational restrictors, see: López d la Paz, M.; Ellis, G.; Penadés, S.; Vicent, C. *Tetrahedron Lett.* **1997**, *38*, 1659-1662.
7. All compounds were characterized by a full complement of high resolution analyses.
8. HOSTEST v5.1, Wilcox, C.S.; Glagovich, N.M. Department of Chemistry, University of Pittsburgh, Pittsburgh, PA, **1994**.
9. The temperature range for the single-point VT analyses of **1**-CTA and **3a**-CTA was 274 to 299K in 5 degree increments. Wider temperature intervals gave rise to curved van't Hoff plots due to heat capacity effects or other temperature-dependent phenomena.
10. The uncertainties in ΔH and ΔS were calculated for 95% confidence levels. The small uncertainties seen here suggest negligeable ΔCp values. Walpole, R.E.; Myers, R.H. *Probabilidad y Estadística*, McGgraw-Hill/Interamericana de México, 3rd Ed., Naucalpan de Juárez, **1992**.
11. Izatt, R.M.; Bradshaw, J.S.; Pawlak, K.; Bruening, R.L.; Tarbet, B.J. *Chem. Rev.* **1992**, *92*, 1261-1354.
12. (a) Wiseman, T.; Williston, S.; Brandts, J.F.; Lin, L.-N. *Anal. Biochem.* **1989**, *179*, 131-137. (b) Freire, E.; Mayorga, O.L.; Straume, M. *Anal. Chem.* **1990**, *62*, 950A-959A. (c) Bundle, D.R.; Sigurskjold, B.W. *Methods in Enzymology*, **1994**, *247*, 288-305.
13. The "c" parameter ($c=K_a M_{tot} n$, M_{tot}=total host concentration, n=number of binding sites) should be between 5 and 500 for the accurate simultaneous determination of ΔH, K_a and n. *Micro Calorimetry System User's Manual*, MicroCal ITC manual, pp. 46-47.
14. Calorimetric studies performed in a mixture of 20% THF/$CHCl_3$ also gave ΔS values for hosts **3a-b** larger than host **1** by 10 eu. The fact that the same difference in entropy is obtained in a solvent with different hydrogen bonding capabilities suggests that this phenomenon is not related to a difference in solvation.
15. Koshland, D.E. Jr. *Proc. Natl. Acad. Sci. USA* **1958**, *44*, 98.
16. For an early example of induced-fit in a H-bonded host-guest system, see Shinkai, S.; He, G.-X.; Matsuda, T.; Hamilton, A.D.; Rosenzweig, H.S. *Tetrahedron Lett.* **1989**, *30*, 5895-5898.