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Isotopomer Encapsulation in a Cylindrical Molecular Capsule: A Probe for Understanding Noncovalent Isotope Effects on a Molecular Level

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Interest in noncovalent isotope effects is on the rise.[1] These effects concern interactions of isotopomers with other molecules, such as synthetic hosts or enzymes, and the difference between the behaviors of isotopomers can be very significant. For example, Thornton et al. recently reported the separation of isotopomers as a means of evaluating their hydrophobic interactions.[2] They found that deuterated isotopomers (particularly aliphatic ones) elute faster on reverse-phase HPLC, i.e., they behave as if they were more polar. However, the interactions evaluated in this manner are not well-defined and can involve different temporary interactions between the solute, the solvent, and the stationary phase. In other examples, the binding of isotopomers of caffeine to human serum albumin was examined.[3−6] Their binding affinities vary considerably, but not in a predictable manner; again, the binding sites are not well-defined. We report here isotope effects on guest binding in the dimeric self-assembled capsule $\mathbf{1}_2$ and relate them to specific functional group interactions on the molecular level.

The cylindrical host dimer $\mathbf{1}_2$ is known to encapsulate a variety of molecules and of molecular pairs.[7−10] For example, *p*-xylene and CCl_4 are co-encapsulated as shown in Chart 1. The guests are confined to particular positions along the C_4 symmetry axis of the capsule, but are usually free to rotate around it. The tumbling of *p*-xylene within the capsule at room temperature is slow on the NMR time scale,[11] and one of its methyl groups is positioned near the resorcinarene end of the capsule. This gives rise to the specific interaction of the methyl with four aromatic rings, which is confirmed by its strong shielding in the NMR spectrum ($\delta = -2.84$ ppm, $\Delta\delta = -5.19$ ppm).

Direct competition experiments were performed with various concentrations of *p*-xylene (**2**) and *p*-xylene-d_{10} (**3**) with the dimeric capsule $\mathbf{1}_2$ (2 mM) and 10 mM of CCl_4 (Table 1) in mesitylene-d_{12}. The system equilibrates within minutes, and NMR integration allowed the concentrations of the four species A−D (Scheme 1) to be calculated in the following manner:

$$[\mathrm{B}] = 2\{\textstyle\int(\mathrm{Me}_{p-\mathrm{xylene}})/3\}/\{\int(4\mathrm{NH}_{\mathrm{capsule}})/4\}$$

$$[\mathrm{C}] = [p\text{-xylene}] - [\mathrm{B}]$$

$$[\mathrm{D}] = 2 - [\mathrm{B}]$$

$$[\mathrm{A}] = [p\text{-xylene-}d_{10}] - [\mathrm{D}]$$

The equilibrium constant K for the reaction depicted in Scheme 1 is:

$$K = \{[\mathrm{C}]\times[\mathrm{D}]\}/\{[\mathrm{A}]\times[\mathrm{B}]\}$$

Entry 5 in Table 1 does not allow the calculation of an equilibrium constant, but it adds a theoretical point at (0,0) to the graph of [C]×[D] as a function of [A]×[B] (Figure 1). The slope of the graph equals the equilibrium constant K, which is 1.32 ± 0.04. A good correlation is obtained.

Chart 1. Line Drawing of **1** (left) and a Minimized Structure of the Encapsulation Complex of *p*-Xylene and CCl_4 in the Dimeric Capsule $\mathbf{1}_2$ (right: the $C_{11}H_{23}$ Chains of the Capsule Are Shortened for Clarity)

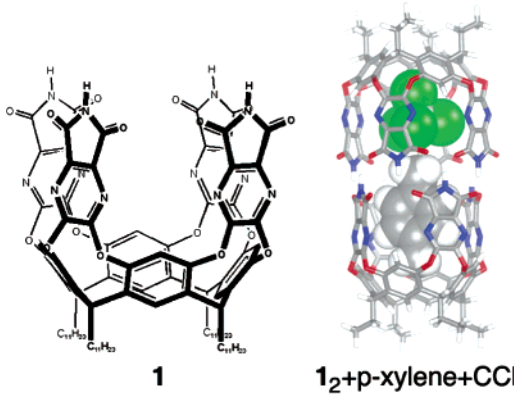
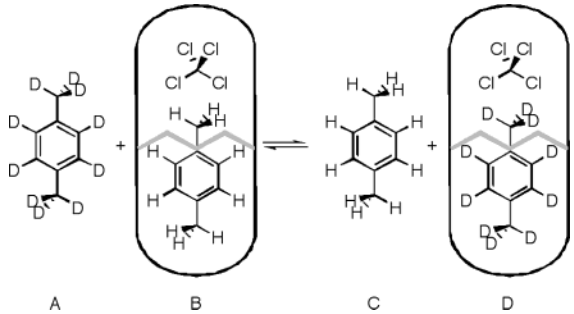


Table 1. Calculated Equilibrium Constants for the Reaction Presented in Scheme 1

entry	*p*-xylene[a]	*p*-xylene-d_{10}[a]	$\int(\mathrm{Me}_{p-\mathrm{xylene}})/\int(4\mathrm{NH}_{\mathrm{capsule}})$	*K*
1	40	10	0.568	1.30
2	30	20	0.400	1.33
3	20	30	0.252	1.33
4	10	40	0.121	1.32
5	0	50	0	

[a] In mM.

Scheme 1. *p*-Xylene and *p*-Xylene-d_{10} Are in Equilibrium between the Solution and the Encapsulated Species



Two more isotopomers, **4** and **5** (Chart 2), were tested in a similar manner to determine whether methyl deuteration or ring deuteration has a higher impact on the equilibrium constant.

Isotopomer **4** was tested against both **2** and **5**, and isotopomer **5** was tested against **3**. The equilibrium constants (Table 2) are very similar, indicating that the preference of the host is determined

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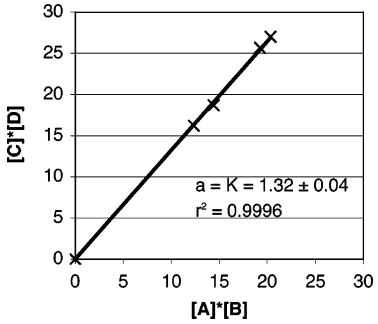


Figure 1. Calculation of the equilibrium constant for the reaction represented in Scheme 1 using a correlation between the concentrations of species [C]×[D] and that of species [A]×[B].

Chart 2. Isotopomers of *p*-Xylene

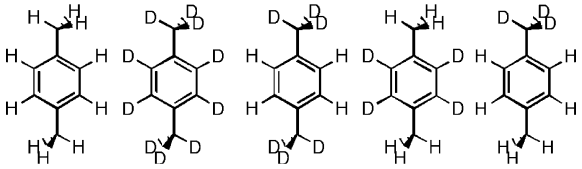


Table 2. Equilibrium Constants for Competitive Encapsulation

entry	CH_3-containing guest	CD_3-containing guest	K^a
1	2	3	1.32 ± 0.04
2	2	4	1.32 ± 0.06
3	5	3	1.26 ± 0.04
4	5	4	1.20 ± 0.05

[a] The CD_3-containing guest is always preferred by the host.

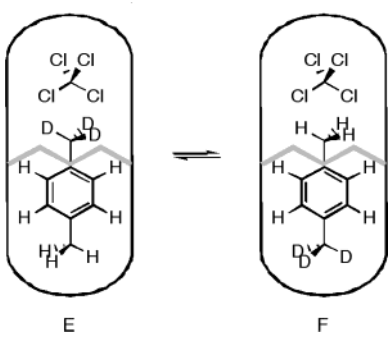
mainly by the methyl group of the guest (CH_3 vs CD_3). Since the measurement is performed by integration of the signal of the CH_3 group near the end of the capsule ($\delta = -2.84$ ppm), isotopomer pairs **3** and **4** or **2** and **5** could not be directly tested. However, calculation of these constants using ratios of the constants in Table 2 gives equilibrium constants of approximately $K = 1$.

The equilibrium constants derived from the competition experiments described in Table 2 also include contributions from the free species in solution. It could be argued, therefore, that they are affected by the preference of the different isotopomers for dissolution in the mesitylene-d_{12} solvent. Furthermore, it is not clear whether the equilibrium constants are influenced primarily by the noncovalent interactions with the capsule or with the co-guest (CCl_4). Accordingly, isotopomer **6** was prepared to resolve these issues (methyl *p*-toluate was reduced with $LiAlD_4$, the alcohol was converted to the chloride, and the final reduction used $LiEt_3BD$).[12]

Compound **6** can be encapsulated as two social isomers[8,13] (Scheme 2): in isomer E the CH_3 group faces the resorcinarene moiety of the capsule, whereas in isomer F the CD_3 group occupies that position. The equilibrium between isomers E and F is described entirely in Scheme 2, and unlike the equilibria discussed previously (Scheme 1), the solvated *p*-xylene is not part of the equation.

The concentration of E can be deduced directly from the integration ratio of the CH_3 group near the end, i.e., at -2.84 ppm, relative to the NH peaks of the capsule. An equilibrium constant of $K = [F]/[E] = 1.35 \pm 0.06$ is obtained (the error margin reflects the uncertainty in the integration itself). This result is comparable to the equilibrium constants reported in Table 2, indicating that the preference of the encapsulation of the CD_3 isotopomers over the CH_3 ones is not related to the differences in their interactions

Scheme 2. Equilibrium between the Two Social Isomers Formed upon Encapsulation of **6**



with the solvent. Rather, they are due to the interaction of the isotopomers with the capsule and not with the other guest (CCl_4). Moreover, the existence of an isotope effect for the social isomers confirms that the cause is electronic and is not related to possible differences in the size of the isotopomers (cf. crystal structures of **2**[14] and **3**[15]).

In conclusion, the capsule $\mathbf{1}_2$ provides a unique environment with well-defined contacts between the methyl group of the guest and the aromatic rings of the host. We observe a preference for the encapsulation of a CD_3 rather than a CH_3 group near the resorcinarene. It is known that the higher weight of D causes a smaller vibration amplitude and frequency for C−D relative to C−H bonds. It is probable that the close interactions of the methyl hydrogens and the surrounding π-systems in the capsule increase the force constant for stretching of the C−H and C−D bonds in the proximity of the capsule.[16,17] This possibility is addressed in the sequel.

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