Inclusion Complexes Oriented in Thermotropic Liquid-Crystalline Solvents Studied with Carbon-13 NMR

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The inclusion complex of cryptophane-A and chloroform dissolved in two nonchiral liquid-crystalline environments was investigated via ^{13}C NMR. Stable solutions of oriented complexes were prepared using aromatic (ZLI 1132) and aliphatic (ZLI 1695) thermotropic nematic liquid crystals as solvents; ordering of the complexes was manifested by the $^{1}H-^{13}C$ dipolar splitting of the ^{13}C resonance of labeled chloroform. In both solutions, the dipolar splitting for the bound ligands was substantially larger than that obtained for the free ligands, indicating a significant increase in ligand ordering upon complexation despite the absence of direct contact with the oriented solvent molecules. A similar enhancement in ordering was observed for complexed ligands compared to that for free ligands in both liquid-crystalline solvents. Also, the application of heteronuclear decoupling to the ZLI 1695 solution resulted in a reduced line width for the bound ^{13}C chloroform resonance, suggesting that a significant component of the observed line broadening may originate from intermolecular couplings between host and guest molecules. These results demonstrate the potential for using restored dipolar couplings to investigate structural and dynamical aspects of inclusion complexes in solution.

I. Introduction

A number of synthetic organic molecules have been designed to incorporate small neutral guest species within their engineered cavities, thereby forming inclusion complexes via reversible trapping processes (see, for example, refs 1−7). Correspondingly, there has been considerable interest in studying the structural and dynamical aspects of ligand binding in these systems. Moreover, because the behavior of organic inclusion complexes is governed by various enthalpic and entropic contributions of more general importance, including van der Waals interactions, solvent effects, relative orientation and motional freedom of the guest, and structural reorganization of the host, such systems may also provide useful models of ligand binding in larger (e.g., biological) molecules.

One such complex-forming molecule is cryptophane-A. Roughly spherical in shape, cryptophane-A is composed of two bowl-shaped cyclotriveratrylene subunits connected by three aliphatic [$(CH_2)_2$] linker groups, forming an interior cavity (Figure 1). The complexes between cryptophane-A and various guest species have been studied previously with NMR, including those between cryptophane-A and methane,[5] chloroform,[5,6] and xenon.[7−9] These complexes form readily in tetrachloroethane and exhibit association constants of 131 M^{-1}, 860 M^{-1},[7] and 3900 M^{-1},[9] respectively, at 278 K.

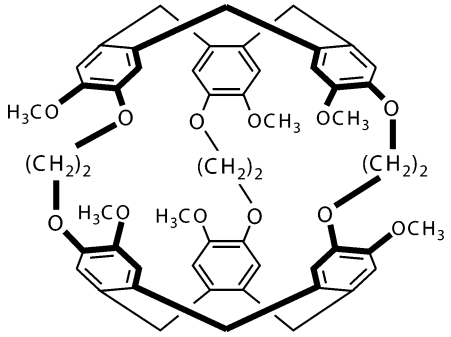

Figure 1. Chemical structure of cryptophane-A.

Here the preparation and preliminary magnetic resonance investigation of the inclusion complex of cryptophane-A and $^{13}CHCl_3$ in two nonchiral thermotropic nematic liquid-crystalline environments are reported.[19] In the absence of isotropic rotational diffusion (hindered by the anisotropic environment of the liquid crystals[10]), dipolar couplings within the inclusion complexes were partially restored. In the liquid-crystalline solutions, a dramatic increase in the dipolar splitting of the ^{13}C resonance for chloroform bound within cryptophane-A was observed (compared to that of uncomplexed chloroform in the liquid-crystalline solvent). These results demonstrate the potential for exploiting reintroduced dipolar couplings to probe the structure and dynamics of ligand binding and complex ordering in such systems.

Recent work by Péchiné et al.[11] has investigated the use of (cyclodextrin-based) chiral hosts oriented in aqueous (nonchiral) liquid-crystalline environments to perform enantiomeric discrimination of chiral ligands. In this work, it was demonstrated that differential quadrupolar splittings could be observed for (*R*) and (*S*) ligands using ^{2}H NMR spectroscopy; the observed

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splittings for bound ligands were reduced compared to that of free ligands in the liquid-crystalline solutions.

II. Experimental Section

Two liquid-crystalline solutions, stable for up to 7 days, were prepared by employing organic thermotropic liquid crystals ZLI 1695 and ZLI 1132 (EMD Chemicals, Inc.) as the solvent. ZLI 1695 and ZLI 1132 exhibit nematic phases from 13 to 72 °C and from −40 to 71 °C, respectively. ZLI 1695 has a negative magnetic susceptibility anisotropy, and the liquid-crystal directors align uniformly in the plane perpendicular to the applied magnetic field. ZLI 1132 has a positive magnetic susceptibility anisotropy, and the liquid-crystal directors align uniformly parallel to the field. The dissolution of cryptophane-A (synthesized by the A. Collet group, Ecole Normale Supérieure de Lyon) in the liquid-crystalline solvents was mediated using d_2-1,1,2,2-tetrachloroethane (Aldrich). Up to 7 mg of cryptophane-A was dissolved in 25 μL of d_2-tetrachloroethane ($C_2D_2Cl_4$). Larger quantities of cryptophane-A gave liquid-crystalline solutions with poorer stability. The cryptophane-A/$C_2D_2Cl_4$ solution was then mixed with ∼0.4 g of the liquid crystals. For different runs, $^{13}CHCl_3$ was added to the sample either prior to or after the cryptophane-A/$C_2D_2Cl_4$ solution had been mixed with the liquid crystals.

NMR experiments were performed on a Varian Unity Inova 300 spectrometer equipped with a 5-mm PFG auto-switchable liquid-state probe and on a Chemagnetics Infinity 500 spectrometer equipped with a 7.5-mm MAS solid-state probe. The undecoupled ^{13}C spectra were recorded at 75.3 MHz with a simple pulse-acquire sequence with an rf field strength of 35.7 kHz. The spectra were recorded in nonspinning mode with a recycle delay of 6 s (no field-frequency lock was employed). For the variable-temperature study, the sample was allowed to equilibrate at each temperature for 30 min. The ^{13}C spectra obtained under CW ^1H heteronuclear decoupling (38 kHz) were recorded at 125.6 MHz; sample spinning and the field-frequency lock were not employed. A recycle delay of 20 s was used to guard against the possibility of sample heating.

III. Results and Discussion

The inclusion complex of cryptophane-A and chloroform has been studied previously in isotropic solutions.[5] In the present work, it is shown that this complex also forms readily in liquid-crystalline environments. Information about the complex is obtained from the observed ^{13}C NMR spectra. Figure 2 shows ^{13}C NMR spectra obtained from two solutions containing $^{13}CHCl_3$, cryptophane-A, and $C_2D_2Cl_4$ dissolved in liquid crystals ZLI 1695 (Figure 2A) and ZLI 1132 (Figure 2B). Both spectra display two sets of doublets and one triplet. The triplet "impurity" arises from partially restored $^2H-^{13}C$ (naturally abundant) couplings in $C_2D_2Cl_4$ species aligned in the liquid crystals. $C_2D_2Cl_4$ is too large to fit within the binding cavity of cryptophane-A, preventing complexation. However, the presence of two sets of doublets in each spectrum confirms the existence of two environments for the $^{13}CHCl_3$ species: that of "free" $^{13}CHCl_3$ in the bulk liquid crystals and that of $^{13}CHCl_3$ "trapped" within the cavity of cryptophane-A.

Generally, the ordering of trapped and free ligands will not be identical. Thus, orientationally dependent magnetic resonance interactions (e.g., chemical shift anisotropy (CSA) and dipolar couplings) can be significantly different for ligands in the two environments. The centers of the doublets do not align, indicating differences in the experimental chemical shift for free and trapped chloroform of 218 Hz (2.91 ppm) and 424 Hz (5.65 ppm) for ZLI 1695 and ZLI 1132 solutions at 25 °C, respectively. The higher-intensity doublets (with splittings of 1184 and 2970 Hz in Figure 2A and B, respectively) are characteristic of the splittings that were observed for $^{13}CHCl_3$ dissolved in the pure ZLI 1695 and ZLI 1132 liquid crystals (not shown) and were assigned to free chloroform in both solutions. Correspondingly, the smaller, broader doublet in each spectrum was the result of $^{13}CHCl_3$ trapped within cryptophane-A. The splittings for trapped chloroform (2915 and 6321 Hz for Figure 2A and B, respectively) were significantly enhanced compared to that of the free chloroform in both solutions. The relative integrals of the peaks of free and trapped chloroform are consistent with complete equilibration of the solutions given the cryptophane-A/chloroform association constant (230 M^{-1} at 298 K[5]).

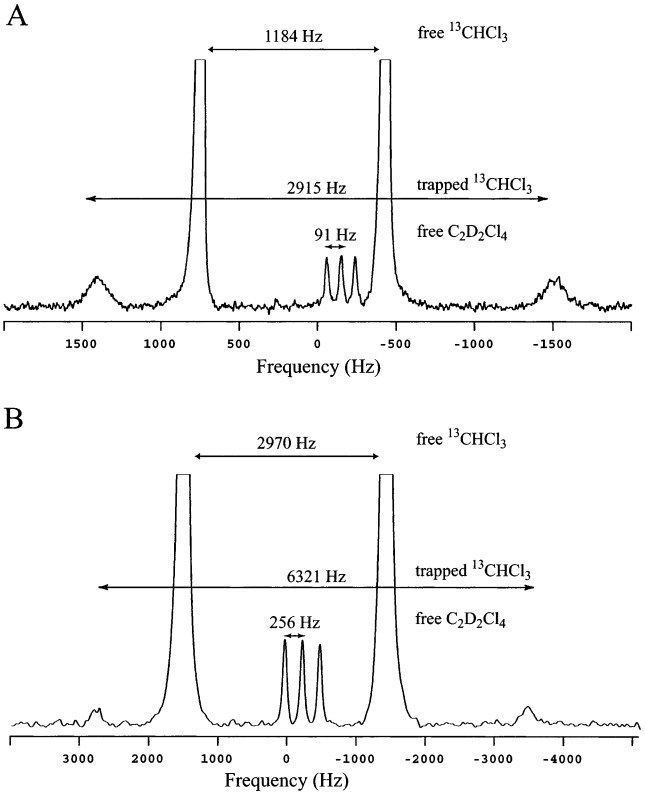

Figure 2. ^{13}C NMR spectra of ^{13}C-labeled chloroform dissolved in cryptophane-A/$C_2D_2Cl_4$/ZLI 1695 (A) and in cryptophane-A/$C_2D_2Cl_4$/ZLI 1132 (B) solutions at 25 °C. The spectra were recorded with 2048 and 8192 scans, respectively; spectra shown in A and B were recorded with spectral widths of 15 and 20 kHz, respectively, and are shown baseline-corrected with 5- and 50-Hz line broadening, respectively.

The observed splitting for $^{13}CHCl_3$ in an anisotropic environment is the sum of the scalar coupling and the reintroduced dipolar coupling and is given by

$$\Delta\nu_{^{13}CH} = J_{^{13}CH} + 2D_{^{13}CH} \quad (1)$$

where J^{13}_{CH} is the indirect coupling constant (approximately +215 Hz for chloroform) and D^{13}_{CH} is the strength of the direct dipole−dipole interaction given by[12]

$$D_{^{13}CH} = -\left(\frac{\mu_0}{8\pi^2}\right)\frac{\gamma_H\gamma_{^{13}C}\hbar}{r^3}S_{^{13}C-H} \quad (2)$$

where r is the internuclear distance (0.1084 nm for chloroform[13]), γ_H and γ^{13}_C are the gyromagnetic ratios for H and ^{13}C, respectively, μ_0 is the vacuum permeability constant, $\hbar$ is Planck's constant divided by 2π, and S^{13}_{C-H} is the order

parameter in the direction of the C−H vector relative to the magnetic field. The sign of D^{13}_{CH} is determined by the sign of S^{13}_{C-H}, which in turn has been obtained previously for chloroform dissolved in different calamitic liquid crystals with negative and positive magnetic susceptibility anisotropy, $\Delta\chi$.[14] Specifically, chloroform is known to orient preferentially such that its C_3 axis lies roughly perpendicular to the liquid-crystal director, supported by values of $S^{13}_{C-H} > 0$ for liquid crystals with $\Delta\chi < 0$ (e.g., ZLI 1695) and $S^{13}_{C-H} < 0$ for liquid crystals with $\Delta\chi > 0$ (e.g., ZLI 1132). As a consequence, D^{13}_{CH} is negative for free chloroform in ZLI 1695 and positive in ZLI 1132.

The sign of D^{13}_{CH} for trapped chloroform in the ZLI 1695 solution is established using the gradient method.[15] In this method, the sign of S^{13}_{C-H} can be determined from a series of measurements performed in the mesophase with varying temperature using the equation

$$\delta^{exp} = \delta^{iso} - \frac{2}{3}\Delta\sigma S_{^{13}C-H} \qquad (3)$$

where δ^{exp} is the experimental chemical shift, δ^{iso} is the isotropic chemical shift, and $\Delta\sigma$ is the ^{13}C shielding anisotropy. For free $^{13}CHCl_3$, the use of positive S^{13}_{C-H} values gives $\Delta\sigma = -55$ ppm (in good agreement with expected values based on previous work, which determined $\Delta\sigma$ to be ca. −50 ppm in such cases[13]). The isotropic chemical shift was determined to be −36.89 ppm, which is very close to our experimental value of −36.66 ppm. For trapped $^{13}CHCl_3$, the use of positive S^{13}_{C-H} values gives $\Delta\sigma = -46.5$ ppm (a reasonable value) and an isotropic chemical shift of −40.65 ppm. Hence, the sign of the dipole−dipole interaction for $^{13}CHCl_3$ trapped in cryptophane-A is the same as that for free $^{13}CHCl_3$ in ZLI 1695 (i.e., negative); correspondingly, it is expected that D^{13}_{CH} for free and trapped $^{13}CHCl_3$ in ZLI 1132 will also share the same (positive) sign.

The amount of restored dipolar coupling depends on the degree of solute ordering in the liquid-crystalline environment. Thus, knowledge of the signs of D^{13}_{CH} and J^{13}_{CH} allows the magnitude of S^{13}_{C-H} to be obtained from the observed line splitting using eq 1 and inverting eq 2. The values for the order parameters of $^{13}CHCl_3$ free in the bulk liquid-crystalline environment of ZLI 1695 and ZLI 1132 are respectively 0.029 and −0.056 at 25 °C. The apparent 2-fold difference in order can be primarily attributed to the difference in the alignment of the liquid crystals toward the magnetic field. (Naturally, this factor is not exactly 2 because the environment is not the same; consequently, the orienting forces are different.) Correspondingly, the values for the order parameters of $^{13}CHCl_3$ trapped in cryptophane-A in ZLI 1695 and ZLI 1132 solutions are 0.066 and −0.125, respectively, at 25 °C. Thus, an ∼2.2-fold enhancement of S^{13}_{C-H} for trapped versus free chloroform is observed for both liquid-crystalline solvents. The effect of temperature on S^{13}_{C-H} was studied for chloroform in the ZLI 1695 solution (Figure 3), with increasingly enhanced ordering observed for both environments at lower temperatures.

The high degree of ordering observed for the trapped chloroform is consistent with the tight fit expected for the ligand within the host's cavity. (The signs obtained for S^{13}_{C-H} for free and trapped chloroform indicate that the enhancements do not primarily result from simple geometrical factors alone but instead reflect an increased ordering of the guest within the complex.) The van der Waals volume of chloroform is ∼72 Å^3,[5] whereas the corresponding cavity volume for cryptophane-A is ∼95 Å^3 when the linker groups are in an extended anti configuration.[9] Moreover, the large stabilizing enthalpy of formation and large destabilizing entropy of formation ($\Delta H = -34.3$ kJ mol^{-1} and $\Delta S_0 = -67$ J mol^{-1} K^{-1}, respectively[5]) determined previously for this complex suggest a highly rigid, "crystal-like" structure.[7] It has also been reported[7] that X-ray crystallographic studies of the analogous complex between chloroform and cryptophane-A6 (in which ethoxy groups are attached to the benzene rings of the cyclotriveratrylene subunits instead of methoxy groups) indicate a highly rigid, closely packed structure, with the 3-fold axis of chloroform aligned with that of the host. Indeed, recent crystallographic results have confirmed the same structural arrangement for chloroform and cryptophane-A.[16] Because chloroform tends to have a preferred orientation within cryptophane-A, it follows that knowledge of the sign of S^{13}_{C-H} of trapped chloroform may also provide information regarding the orientation of the cryptophane-A host

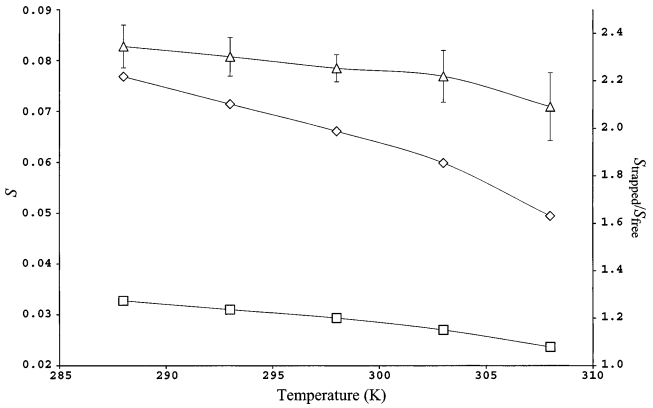

Figure 3. Temperature dependence of the order parameters for the $^{13}C-H$ bond in chloroform dissolved in a cryptophane-A/$C_2D_2Cl_4$/ZLI 1695 solution relative to the magnetic field. Order parameter for trapped chloroform (◇), order parameter for free chloroform (□), and ratio of the order parameters for trapped and free chloroform (△). Error bars are estimated from uncertainties in the peak splittings (which in turn are based upon the line widths). The lines are meant to guide the eye.

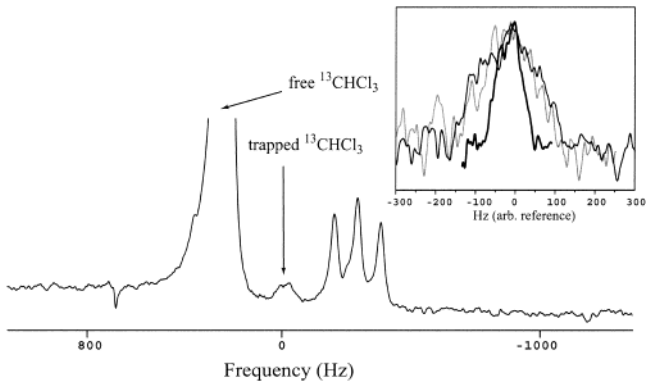

Figure 4. ^{13}C spectrum under 38-kHz CW ^1H heteronuclear decoupling of ^{13}C-labeled chloroform dissolved in a cryptophane-A/$C_2D_2Cl_4$/ZLI 1695 solution. The 125.6-MHz carbon spectrum was acquired with 2048 scans and is shown with 10-Hz line broadening applied during data analysis. The inset shows an overlay of intensity-normalized peaks for the bound resonances from spectra acquired with and without heteronuclear decoupling. The magnitude of the line width reduction upon decoupling (roughly 2-fold) was estimated from Lorentzian fits of the peaks using a commercial software package (Origin 7.0). Multiple fits of each line were performed (varying the point ranges and baseline correction methods). Because of the spectral noise, the range in line widths obtained from the various fits of a given peak was generally greater than the statistical fitting uncertainty: Decoupled peak (bold line): fwhm ≈ (75−90) ± 4 Hz; undecoupled, low-frequency peak (black line): fwhm ≈ (170−205) ± 5 Hz; undecoupled, high-frequency peak (light-gray line): fwhm ≈ (142−160) ± 5 Hz (values include 10-Hz line broadening).

in a liquid-crystalline matrix. If so, then the positive value for S^{13}_{C-H} observed for the trapped guest in ZLI 1695 may indicate that the complex tends to order such that its primary axis lies roughly perpendicular to the liquid-crystal director.

The effects of decoupling were also investigated in the ZLI 1695 solution. Under ^{1}H broadband heteronuclear decoupling, the doublets collapsed into two single lines (Figure 4). Also, whereas the line width of the free chloroform was essentially unchanged by the decoupling, the line width of trapped chloroform was reduced significantly (roughly by a factor of 2; see Figure 4 (inset)). It is likely that this line-narrowing was primarily the result of the loss of *intermolecular* dipolar coupling between host and guest species, although other effects such as the presence of a distribution of order parameter values (caused, for example, by slight variations in the host environment across the sample) may have also contributed to the observed effect.

IV. Conclusions

In summary, the preparation of two stable liquid-crystalline matrices containing inclusion complexes of chloroform and cryptophane-A has been demonstrated. Clear differences in the orientationally dependent spectral features were observed between free and trapped ligands in both solutions, including significantly larger dipolar splittings for trapped ligands corresponding to an ~2.2-fold increase in the order parameter for the C−H bond of the guest molecule. The larger-order parameters for trapped chloroform are consistent with a higher degree of spatial ordering and reduced motion compared to that of free chloroform in the bulk liquid-crystalline solutions. Determining the relative importance of the different contributions to the observed effects (e.g., relative orientation and motional freedom of the guest and the structure, dynamics, and overall ordering of the host) remains an intriguing problem. Future studies will include the investigation of other ligands of different sizes (and more coupled spins) as well as other hosts with different cage properties in both thermotropic and (aqueous) lyotropic liquid-crystalline environments. Finally, the presence of significant intermolecular dipolar couplings induced by the liquid-crystalline environment presents an alternative route for magnetization transfer between host and guest spins,[17] with an efficiency rivaling that of the Overhauser-type methods exploited within inclusion complexes in isotropic solutions.[8,18]

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References and Notes

(1) Cram, D. J.; Tanner, M. E.; Knobler, C. B. *J. Am. Chem. Soc.* **1991**, *113*, 7717−7727.
(2) Robbins, T. A.; Knobler, C. B.; Bellew, D. R.; Cram, D. J. *J. Am. Chem. Soc.* **1994**, *116*, 111−122.
(3) Branda, N.; Grotzfeld, R. M.; Valdés, C.; Rebek, J., Jr. *J. Am. Chem. Soc.* **1995**, *117*, 85−88.
(4) Valdés, C.; Spiz, U. P.; Toledo, L. M.; Kubik, S. W.; Rebek, J., Jr. *J. Am. Chem. Soc.* **1995**, *117*, 12733−12745.
(5) Garel, L.; Dutasta, J. P.; Collet, A. *Angew. Chem., Int. Ed. Engl.* **1993**, *32*, 1169−1171.
(6) Canceill, J.; Lacombe, L.; Collet, A. *J. Chem. Soc., Chem. Commun.* **1987**, *3*, 219−221.
(7) Bartik, K.; Luhmer, M.; Dutasta, J.-P.; Collet, A.; Reisse, J. *J. Am. Chem. Soc.* **1998**, *120*, 784−791.
(8) Luhmer, M.; Goodson, B. M.; Song, Y. Q.; Laws, D. D.; Kaiser, L.; Cyrier, M. C.; Pines, A. *J. Am. Chem. Soc.* **1999**, *121*, 3502−3512.
(9) Brotin, T.; Dutasta, J.-P. *Eur. J. Org. Chem.* **2003**, 973−984.
(10) Emsley, J. W.; Lindon, J. C. *NMR Spectroscopy Using Liquid Crystal Solvents*; Pergamon Press: Oxford, England, 1975.
(11) Péchiné, J.-M.; Meddour, A.; Courtieu, J. *Chem. Commun.* **2002**, *16*, 1734−1735.
(12) Frech, C. B.; Fung, B. M.; Schadt, M. *Liq. Cryst.* **1988**, *3*, 713−722.
(13) Vaara, J.; Oikarinen, K. Jokisaari, J. Lounila, J. *Chem. Phys. Lett.* **1996**, *253*, 340−348.
(14) Jokisaari, J.; Hiltunen, Y. *J. Magn. Reson.* **1984**, *60*, 307−319.
(15) Lounila, J.; Jokisaari, J. *Prog. Nucl. Magn. Reson. Spectrosc.* **1982**, *15*, 249−290.
(16) Dutasta, J.-P. Personal communication.
(17) Goodson, B. M. Nuclear Magnetic Resonance of Laser-Polarized Noble Gases in Molecules, Materials, and Organisms. Ph.D. Thesis, University of California, Berkeley, CA, 1999.
(18) Song, Y.-Q.; Goodson, B. M.; Taylor, R. E.; Laws, D. D.; Navon, G.; Pines, A. *Angew. Chem., Int. Ed. Engl.* **1997**, *36*, 2368−2370.
(19) Results presented in part at the 43rd Experimental Nuclear Magnetic Resonance Conference, April 14−19, 2002, Pacific Grove, CA.