Direct Evidence for the Role of Transient Complexation in Spin-Label–Alcohol Interactions

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A simple method by which to identify the dominant motional component governing dipolar coupling in transient interactions between solvent molecules and paramagnetic solutes is described and applied to the interactions between a nitroxide spin probe (Tempone or 4-oxo-Tempo) and trifluoroethanol and pentafluorophenol. The method involves examining the paramagnetic species-enhanced relaxation of the solvent nuclei as a function of solvent concentration in an inert cosolvent. The component of the interaction governed by the rotational motion of the transient complex can be separated from the translational motion of the molecules that are free in solution by comparing the slope and intercept of the straight line obtained when $[(R_{obsd} - R_b)/N_e]$ is plotted vs. $[1/(c_{OH}^0 + (1/K_c))]$, where R_{obsd} and R_b are the observed and bulk nuclear relaxation rates, N_e is the concentration of paramagnetic species added to the solution, c_{OH}^0 is the alcohol concentration, and K_c is the formation constant for the solvent:solute complex. These studies show that the behavior of each of the ^{1}H and ^{19}F on CF_3CH_2OH and C_6F_5OH exhibits a mixture of the two components. The rotation of the transient complex is clearly the dominant motion governing dipolar coupling for the OH protons in each case. Radical–nucleus distances in the complex for the other nuclei on CF_3CH_2OH increase as one proceeds down the carbon chain from the OH, and the *relative* importance of the translational component of the interspecies dipolar coupling increases in the sequence ^{1}H (OH) < ^{1}H (CH_2) < ^{19}F (CF_3).

Introduction

A very powerful method by which to characterize the molecular dynamics of transient solvent–solute interactions involves the coupled use of the dynamic nuclear polarization (DNP)[1,2] of solvent nuclei by paramagnetic species with the nuclear relaxation enhancement (NRE, of which PRE or proton relaxation enhancement is a special case)[3-8] of the same solvent nuclei to provide direct information about the intermolecular coupling of the spins. The strength of the technique lies in the very pronounced effects that even a small amount of free radical can have on solvent nuclei. Moreover, paramagnetic species shift the sensitivity of NMR methods to motions on the 10^{-10}- to 10^{-11}-s time scale by raising the relevant frequencies for the intermolecular relaxation component by a factor of 660 or more.

The interaction of the unpaired electron spin on the paramagnetic solute and the nuclear spin on the solvent molecule can occur by both dipolar and scalar mechanisms, either of which can be modulated by more than one type of motion of the interacting species. For the dipolar mechanism, either translational diffusion of the interacting molecules or rotational diffusion of the associated species can be effective.[2] In previous studies of the molecular dynamics of interactions in systems containing nitroxide free radicals, identification of the dominant type of motion has been difficult. Often the dominant motion has been assumed,[9-12] with the support of circumstantial evidence. Methods that involve a detailed study of NRE,[6-8] DNP,[13,14] or NMR relaxation dispersion[15] as a function of magnetic field to identify the subtle dif- ferences in spectral density functions attributable to these two mechanisms are cumbersome and retain significant uncertainty when a mixture of the two motions is present.

A simple method to identify the *dominant* motional component governing dipolar coupling in transient interactions is given in this paper. The method also provides a reliable measure of the partitioning of the interaction between these two major mechanisms. This study offers direct evidence of strong, stereospecific interactions between nitroxide spin probes and fluorinated alcohols by characterizing the change in interspecies interaction as a function of solvent mole fraction in an inert cosolvent. Results for ^{1}H and ^{19}F nuclei on CF_3CH_2OH and C_6F_5OH interacting with the spin probe 4-oxo-2,6,6-tetramethyl-1-piperidinyloxy (Tempone, also commonly called 4-oxo-Tempo) are presented.

Theoretical Background

The relaxation transitions that couple a solvent nucleus and solute unpaired electron are shown in Figure 1. In addition to the scalar transition shown as *c*, dipolar transitions labeled *p*, *q*, *r*, and *s* are effective in coupling each pair of states. At low fields these dipolar components are in the ratio $q:r:s = 3:2:12$. The total paramagnetic species-induced relaxation is the sum of the scalar and dipolar components as $2q + r + s + c$. When the local dipolar relaxation rates are governed by rotational diffusion,[16] the total dipolar rate is given by

$$R_{dr} = (3/10)\gamma_e^2\gamma_n^2\hbar^2\tau_c d_r^{-6}[n_p N_e N_p^{-1}][J_r(\omega_I) + (7/3)J_r(\omega_S)] \tag{1}$$

where $J_r(\omega_i) = 1/(1 + \omega_i^2\tau_c^2)$ and τ_c is the dipolar correlation time for rotational diffusion; d_r is the average pair radius for the rotating adduct; N_e and N_p refer to the unpaired electron and nuclear concentrations, respectively; n_p is the number of receptor nuclei bound near each electron; and $J_r(\omega)$ is the rotational spectral density function which depends on the frequency appropriate for the particular transition.

When the dipolar coupling is given by translational diffusion, the total dipolar rate is given by[17]

$$R_{dt} = (2\pi/5)\hbar^2\gamma_e^2\gamma_n^2\tau_d d_t^{-3}N_e[J_t(\omega_I) + (7/3)J_t(\omega_S)] \tag{2}$$

where

$$J_t(\omega_i) \sim [1 + 0.9(\omega_i\tau_d)^{1/2} + 1.5(\omega_i\tau_d)^{3/2}]^{-1} \tag{3}$$

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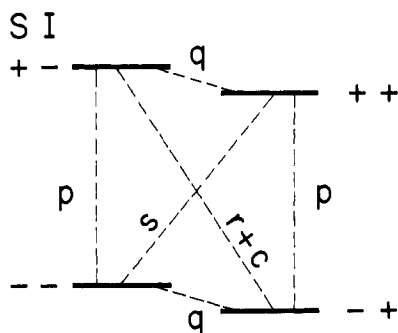(15) B. Borah and R. G. Bryant, *J. Chem. Phys.*, **75**, 3297–3300 (1981).

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Figure 1. Diagram showing four combined spin states for a simple spin $S = {}^1/_2$ unpaired electron spin interacting with an $I = {}^1/_2$ solvent nuclear spin.

Figure 2. A plot of the spin-probe-induced component of the inverse of the T_1 time for the OH protons on CF_3CH_2OH as a function of the reciprocal of the CF_3CH_2OH concentration (●). Also shown is the plot for the reciprocal of the CF_3CH_2OH concentration corrected for the instantaneous fraction tied up in transient complexes according to eq 6 (■) with the least-squares line fitting the data.

Previous attempts to identify the dominant component have been based on fitting the entire spectral density curve[13-15] or on the existence of the plateau present in the composite spectral density function $J(H)$ for the rotational mechanism that is absent in the translational case, and subsequent curve fitting.[7,8] When two motional components are present for the same solvent nucleus, the uncertainty in such a separation can be very large.

Experimental Section

Proton and fluorine spin–lattice relaxation measurements were performed by using $180°-\tau-90°$ pulse sequences on a Bruker WH-90 pulsed FT-NMR spectrometer at three frequencies as described previously.[6,8]

Solvents and solutes were commercial samples (C_6D_6 and Tempone from Aldrich; CF_3CH_2OH and C_6F_5OH from PCR) that were used without further purification. Radical solutions in the concentration range of 5×10^{-4} to 5×10^{-5} M for ^1H and in the range of 2.5×10^{-4} to 1×10^{-2} M for ^{19}F were made by successive dilution using solvent mixtures of the appropriate mole fractions of C_6D_6 and the fluorinated species. C_6D_6 was used to provide a deuterium lock for the NMR spectrometer and to minimize changes in viscosity of the samples as the mole fraction is varied. Viscosities were checked on representative samples with a thermostated Oswald viscometer, and effects of the small viscosity changes were determined to be minimal compared to changes in observed rates. The samples were sealed under vacuum by the freeze–pump–thaw technique in order to eliminate dissolved oxygen.

Results

CF_3CH_2OH. Paramagnetic species-enhanced relaxation rates for ^1H and ^{19}F nuclei on CF_3CH_2OH with Tempone in 1:1 mixtures of the alcohol with C_6D_6 have been published previously.[6-8] The plot of observed relaxation rate vs. spin probe concentration gives a straight line, the slope of which is the paramagnetic species-induced component of the solvent nuclear relaxation rate, given as $R_{reduced} = (R_{obsd} - R_b)/N_e$. When this value is measured for the same nucleus with the same spin probe in a series of mixtures of the same pair of solvents with varying mole fractions, a straight line is obtained for each nucleus for each mole fraction. However, significant changes are observed in the slopes for the OH protons, while the CH_2 protons and fluorine nuclei show relatively little variation.

If, as has been determined by DNP measurements to be true for all three nuclei under observation on CF_3CH_2OH with Tempone, the scalar component for each nucleus is negligible or very small,[6,7] $R_{reduced}$ is simply the sum of the contributions to the intermolecular coupling of the nuclear and electron spins arising from the reduced rotational (R_{dr}/N_e) and translational (R_{dt}/N_e) contributions. The simplest interpretation of the results, then, is that the marked increase in $R_{reduced}$ as the alcohol mole fraction decreases indicates dominance by the rotational mechanism for the OH protons, while the translational motion is dominant for the other two nuclei. The basis for this is the observation that the simple form for R_{dr}/N_e contains the term n_p/N_p, which is the ratio of the fraction of protons bound near the unpaired electron
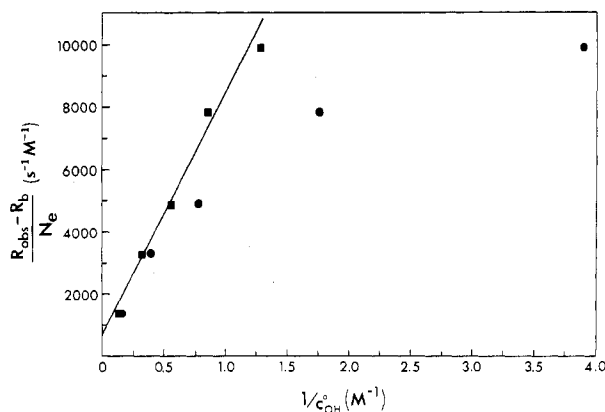
in the complex to the total available nuclear concentration in solution. This implies there is a dependence on the concentration of the solvent involved in complex formation. There is no corresponding dependence for the R_{dt}/N_e expression.

Quantitatively, however, this breaks down. A plot of $R_{reduced}$ vs. $1/N_p$ fails to produce a linear relationship for any of the three nuclei. The OH data do not give a straight line passing through the origin that would characterize pure rotational dominance nor even a straight line with a finite intercept that might be characteristic of some mixture of rotational and translational components. In addition, neither the CH_2 nor the CF_3 plot is linear. A plot for the OH protons is shown in Figure 2.

By realizing that the nonlinearity arises because the fraction of the spin probe molecules involved in complexes varies significantly in the mole fraction range used in this study, straight-line plots can be obtained. The fraction of spin probe tied up in complexes at any instant giving rise to the rotational component and the fraction free in solution giving rise to the component reflecting the translational motion of the two species must be incorporated directly in order to separate the rotational and translational contributions to the NRE for each nucleus. As a first step, the expression given by Müller-Warmuth and co-workers is[18]

$$(R_{obsd} - R_b) = x_{complex}R_{dr} + (1 - x_{complex})R_{dt} \qquad (4)$$

Rewriting $x_{complex}$, the instantaneous fraction of alcohol molecules tied up in a complex, in terms of the formation constant for the complex K_c (which can be measured by ESR techniques and has a value of 1.9 m^{-1} for this system)[8] and $c_{OH}°$, the amount of proton donor added, yields[5]

$$x_{complex} = \frac{c_{complex}}{c_{OH}°} = \frac{K_c c_{OH}°}{(1 + K_c c_{OH}°)} \frac{N_e}{c_{OH}°} = \frac{K_c N_e}{1 + K_c c_{OH}°} \qquad (5)$$

The reduced paramagnetic species-induced relaxation rate becomes

$$R_{reduced} = \frac{1}{(c_{OH}° + 1/K_c)}[R_{dr} - R_{dt}*N_e] + R_{dt}* \qquad (6)$$

where $R_{dt}* = R_{dt}/N_e$. Thus a plot of $R_{reduced}$ vs. $1/[c_{OH}° + 1/K_c]$ rather than simply vs. $1/c_{OH}°$ should produce an intercept that gives the reduced translational component and a slope that is proportional to R_{dr}. (The $R_{dt}*N_e$ term is very small for all samples examined because N_e is typically 10^{-3} M or less.) These plots for each of the three CF_3CH_2OH nuclei with Tempone are shown in Figures 2 and 3, and the slopes and intercepts that result are given in Table I.

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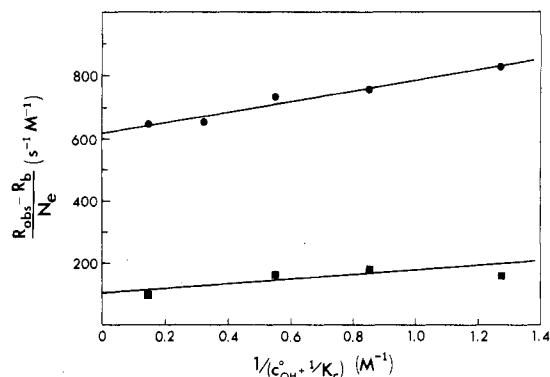
Figure 3. Plots of the spin-probe-induced component of the inverse of the T_1 time for the CH_2 protons (●) and CF_3 fluorines (■) on CF_3CH_2OH vs. the inverse CF_3CH_2OH concentration corrected according to eq 6 with the least-squares lines fitting the data.

TABLE I: Results for Fit of Reduced Relaxation Rates vs. Reduced Complex Mole Fraction

solvent	nucleus	slope[a]	intercept[a]
CF_3CH_2OH	OH ^1H	7600 ± 600	700 ± 400
CF_3CH_2OH	CH_2 ^1H	166 ± 20	620 ± 20
CF_3CH_2OH	CF_3 ^{19}F	73 ± 28	104 ± 23
C_6F_5OH	OH ^1H	35000 ± 1300	3900 ± 1700
C_6F_5OH	o-F	1120 ± 170	330 ± 130
C_6F_5OH	m-F	280 ± 40	200 ± 50
C_6F_5OH	p-F	56 ± 8	101 ± 9

[a] ± values are one standard deviation in the appropriate variable.

C_6F_5OH. The T_1 rates for OH protons and the three types of fluorines on C_6F_5OH also give a linear dependence when plotted as a function of Tempone concentration. The reduced paramagnetic species-induced relaxation rates determined from the slopes of such plots in solutions of different C_6F_5OH mole fractions give the slope and intercept in Table I. (The value of $K_c = 4.4$ M^{-1} has been determined previously from ESR measurements.)[8] Results for the ^{19}F in each of the three positions are shown in Figure 4.

Discussion

Plots of reduced relaxation rates vs. $[c_{OH}^0 + 1/K_c]^{-1}$ produce straight lines by which the rotational and translational motional components of the dipolar relaxation rate can be separated. The data can be checked as it is being taken by plotting observed rates vs. spin probe concentration to ensure that straight lines characteristic of well-behaved systems are obtained. As an alternative, eq 4 can be used to treat all the data obtained for various solvent mole fractions and Tempone concentrations. However, such plots prove unreliable compared to plots dictated by eq 6 based on the slopes of the relaxation rate vs. probe concentration, especially when a mixture of the two motional components is present.

The slopes and intercepts obtained provide effectively the rotational and translational contributions in these cases because the $[R_{dt}*N_e]$ term in the slope is very small compared to R_{dr}. When these slopes and intercepts are used, the translational and rotational contributions to the total rate for any given sample can be determined. Such results are shown in Table II for solutions con-
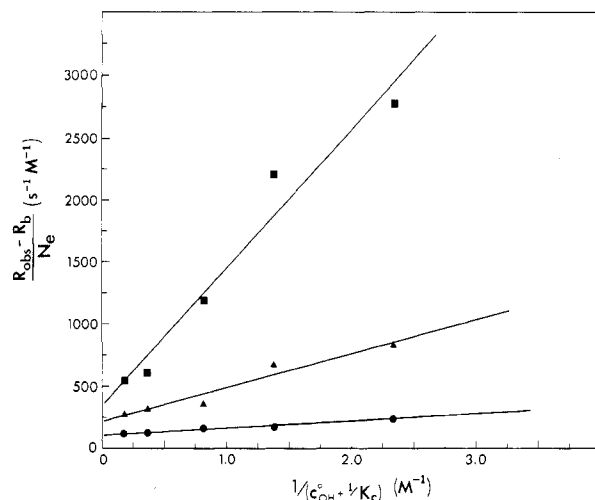

Figure 4. Plot of the spin-probe-induced component of the inverse of the T_1 time for the o-F (■), m-F (▲), and p-F (●) on C_6F_5OH vs. the inverse C_6F_5OH concentration corrected according to eq 6 with the least-squares lines fitting the data.

taining 0.001 M Tempone and either 50% or 2% of the proton donor. As is indicated in Table II, the behavior of each of the four nuclei considered is governed by a mixture of a rotational and translational component, rather than any one nucleus being completely dominated by one mechanism or the other. However, the rotational tumbling of the complex is clearly the major component in the cases of each of the OH protons. The translational component is effective only because such a small fraction of the proton donor is available in a complex at any instant. In the 2% solutions where 0.13% of the CF_3CH_2OH is tied up in a complex, the rotational component accounts for approximately 90% of the OH proton relaxation. On the other hand, translational diffusion of the two species is the major component for the CH_2 protons, which, when the CF_3CH_2OH is tied up in a complex, are considerably removed from the binding site.

The results indicate clearly that information concerning the behavior of systems in which the motion of the associated species is important is best obtained in solutions of low mole fraction of the proton donor. This includes systems dominated by dipolar coupling of the two spins and those for which a scalar component governed by sticking is dominant. For the protons in these samples, low field dynamic nuclear polarization (DNP) measurements have shown that scalar coupling of the nuclear and unpaired electron spins can be neglected. DNP measurements have also shown that the scalar component for the fluorines on CF_3CH_2OH is very small. If these 21.1-kG results could be used directly to predict the expected degree of scalar enhancement in a low-field DNP experiment, one would obtain, assuming that the rotational component was solely arising from a scalar component, that the ultimate enhancement would be −210, which is of the same order as that observed. However, at these higher fields, the scalar component which depends only on the frequency of the transition governed by the ESR frequency of the spin probe (see Figure 1) should be significantly reduced from its low-field value and an interpretation based on a rotational dipolar component seems more plausible.

TABLE II: Motional Components of the Relaxation of CF_3CH_2OH and C_6F_5OH by 0.001 M Tempone

solvent	nucleus	% solvent	$X_{complex}$	rotational rate, s^{-1}	translational rate, s^{-1}	% rot	% trans
CF_3CH_2OH	OH ^1H	50	1.4×10^{-4}	1.01	1.11	48	52
CF_3CH_2OH	OH ^1H	2	1.3×10^{-3}	9.22	1.11	89	11
CF_3CH_2OH	CH_2 ^1H	50	1.4×10^{-4}	0.026	0.597	4	96
CF_3CH_2OH	CH_2 ^1H	2	1.3×10^{-3}	0.237	0.596	28	72
CF_3CH_2OH	^{19}F	50	1.4×10^{-4}	0.012	0.077	13	87
CF_3CH_2OH	^{19}F	2	1.4×10^{-3}	0.109	0.077	59	41
C_6F_5OH	OH ^1H	50	1.9×10^{-4}	6.8	3.4	67	33
C_6F_5OH	OH ^1H	2	2.3×10^{-3}	84.2	3.4	96	4

These results prompt a reexamination of the results of the study of the field-dependent relaxation rates in these systems.[7,8] If one can assume the correlation time determinations made in those measurements remain approximately valid even though these results arise from a mixture of components, the rotational rates can be used to calculate the values of d_r, the distance between the interacting spins in the complex. The results for CF_3CH_2OH are $d_r(OH) = 0.28$ nm, $d_r(CH_2) = 0.46$ m, $d_r(CF_3) = 0.52$ nm, and for C_6F_5OH, $d_r(OH) = 0.25$ nm. This trend in interaction distances is far more acceptable for a nonlinear complex formed between the nitroxide group of the spin probe and OH proton on an unbranched alcohol that is free to rotate, than the initial results reported previously. These suffered from the inability to separate the two contributions to the dipolar relaxation because the mechanism had to be deduced solely from the shape of the spectral density curve. These data for each nucleus were analyzed previously as if it were all of one mechanism or the other.

An additional interesting comparison to make involves the different translational rates for the nuclei studied. Clearly the rate is largest for the OH protons on each of the proton donors, with the fluorine rate being considerably smaller. The larger OH proton rates may arise from the proximity of the OH protons to the unpaired spin probe electron as the complex is broken and the two species begin to diffuse apart, an observation consistent with previously hypothesized increased dipolar coupling in samples with significant scalar components.[19] This would not account for the decreased fluorine component. However, though the smaller γ^2 term for fluorine nuclei has a limited effect, the reduced accessibility of the electron-rich site on the spin probe to the larger CF_3 group may result in the diminished rate in this case, or the preferred orientation of the alcohol in solvating the spin probe may inhibit the ability of the F to approach closely.

Conclusion

Several important points result from the work described in this paper. First, the use of variable mole fractions proves to be an effective technique by which to separate rotational and translational components of dipolar interactions between unpaired electron and nuclear spins. The most effective means by which to characterize the nature of the transient complex between the two species is in samples of low proton donor mole fraction; the translational component is more accessible in samples of high donor mole fraction. Care in selecting the inert cosolvent is necessary, with a solvent of similar viscosity most appropriate to minimize changes in correlation times and thus position on the spectral density curve as the solvent mixture is varied. For the OH protons on both CF_3CH_2OH and C_6F_5OH, the rotational motion of the complex is clearly the dominant dipolar relaxation mechanism. The CH_2 protons and the fluorines on the alcohol have much smaller rotational components, a feature ascribed to the much longer interaction distances in the transient complex, such that the translational motion of the free solvent and solute species becomes the dominant dipolar component in solutions of high alcohol mole fraction. This method of separating the major components of the motions responsible for the paramagnetic species-induced relaxation of solvent nuclei when used in conjunction with studies of these rates as a function of magnetic field promises to be an important means by which to characterize the molecular dynamics of solvent–solute interactions.

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Sphere–Rod Transition of Surfactant Micelles and Size Distribution of Rodlike Micelles

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A theory of the salt- or temperature-induced sphere–rod transition of surfactant micelles and the reversible linear aggregation of surfactant into rodlike micelles has been developed on the basis of a simple treatment of the law of mass action, and its implication is given in terms of statistical thermodynamics. The sphere–rod conversion is described by the introduction of an initiation parameter, which is strongly dependent on the salt concentration and temperature near their threshold values. The stepwise association of surfactant molecules or ions beyond the spherical micelles is analyzed by considering the configurational degeneracy of rodlike micelles. The association constant of monomer addition to the rodlike micelles is found to be inversely proportional to the aggregation number, in contrast to the equal association constant for isodesmic association. The present theory leads to the Poisson distribution of length of rodlike micelles, whose maximum is located at an aggregation number, independently of the surfactant concentration.

Introduction

Surfactant molecules or ions aggregate reversibly into micelles above a certain critical concentration in aqueous solutions. Ionic surfactants generally form spherical or globular micelles in solution, but they can further aggregate into rodlike or wormlike micelles with increasing surfactant concentration, when a simple salt is present beyond a certain threshold concentration.[1] In the presence of excess concentrations of salt, the electrostatic effect of charged micelles would be sufficiently suppressed. Indeed, nonionic surfactants having a weakly hydrophilic group or a highly hydrophobic group can also form rodlike micelles,[2,3] possibly above a certain threshold temperature.

Spherical micelles are formed cooperatively at the critical micelle concentration; the hydrophobic groups of molecules or ions are incorporated into a micelle in such a way that they are exposed to aqueous media to the least extent.[4,5] The spherical micelle

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