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Local Diffusion in Paramagnetic Solutions by NMR Relaxometry at One Frequency

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Experimental studies of the unrestricted translational (t) diffusion coefficient D^t of molecules in a liquid provide information[1] about the organization of their immediate environment and a way to test the models of intermolecular forces and the theories of transport. In porous media and biological tissues, D^t is important to correlate the long-range apparent diffusion[2] of the molecules explored by pulsed gradient spin−echo (PGSE) NMR with their interactions with the fluid/matrix interfaces and/or macromolecules in a crowded environment. Here, a simple method is proposed to measure the relative diffusion coefficient of a pair of small molecules at the nanometer scale. It is easy to use on standard spectrometers and magnetic resonance (MR) imaging (I) and microscopy (M) instruments.

In a liquid solution, consider nuclear spins I on solvent or solute molecules M_I in the presence of electronic spins S on solutes M_S without long-ranged charge−charge or binding interaction with M_I. The longitudinal relaxation rate, R_1, transverse relaxation rate, R_2, and longitudinal relaxation rate, $R_{1\rho}$, in the rotating frame of a spin I can be measured in an external magnetic field $\mathbf{B}_0$ by standard NMR sequences.[3] Each rate R_α is the sum[4] $R_\alpha = R_{\alpha 0} + R_{\alpha p}$ of the value $R_{\alpha 0}$ in the diamagnetic solution without paramagnetic solutes and of the paramagnetic (p) relaxation enhancement (PRE) $R_{\alpha p}$ of the spin I due to its purely outer-sphere (OS) interactions with the spins S. The PREs R_{2p} and $R_{1\rho p}$ are indistinguishable, but are in extremely viscous solvents.[4] The mixed PRE is defined as[4]

$$R_{mix} = R_{mix2} \equiv \frac{3}{2}\left(R_{2p} - \frac{R_{1p}}{2}\right) \cong R_{mix1\rho} \equiv \frac{3}{2}\left(R_{1\rho p} - \frac{R_{1p}}{2}\right) \quad (1)$$

The relaxivity, r_α [s^{-1} mM^{-1}] ($\alpha = 1, 2, 1\rho$, mix), is defined as the PRE $R_{\alpha p}$ divided by the concentration c_S [mM] of M_S

$$r_\alpha \equiv R_{\alpha p}/c_S = (R_\alpha - R_{\alpha 0})/c_S \quad (2)$$

The relative diffusion coefficient D of M_I and M_S can be derived from three properties, $P1$, $P2$, and $P3$, of r_{mix} and r_1. The method was tested in the case of the proton PREs of the *tert*-butyl alcohol M_I = $(CH_3)_3COD$ due to the paramagnetic complex M_S = $Gd(dtpa)^{2-}$ in a D_2O solution with $(dtpa)^{5-}$ = diethylenetriamine pentaacetate. Let γ_I and γ_S be the gyromagnetic ratios of the spins I and S, and $\omega_I \equiv |\gamma_I|B_0$ and $\omega_S \equiv |\gamma_S|B_0$, their angular Larmor frequencies. The relaxation theory used to derive D involves the OS variation parameter $\lambda_D \equiv (8\pi/45)\gamma^2{}_I\gamma^2{}_S\hbar^2\, S(S+1)10^{-6}N_{Avogadro}$. To discuss $P1$−$P3$, auxiliary geometrical and dynamical molecular quantities are introduced: denote the collision diameter of M_I and M_S by b and their translational correlation time by $\tau \equiv b^2/D$. It is assumed that the longitudinal electronic time correlation function[4,5] (TCF) $G_{\|}^{nor}(t) \equiv \langle S_z(t)S_z(0)\rangle/\langle S_z(0)S_z(0)\rangle$ of the spin S is a decaying exponential with an electronic relaxation time T_{1e}. This assumption holds for nitroxide radicals and complexed paramagnetic metal ions in S states ($L = 0$), such as Mn^{2+} or Gd^{3+} at sufficiently high field values.[5]

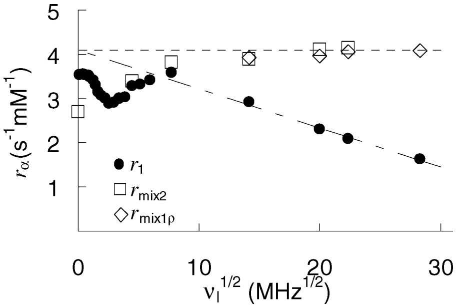

Figure 1. Relaxivities r_α ($\alpha = 1$, mix2, mix1ρ) versus $(\nu_I)^{1/2}$ in D_2O at 298 K.

(P1) Above a moderate field $B_0 \geq B_0^{indep}$ ($B_0^{indep} \geq 1.5$ T for Gd(III) spins), $r_{mix}(B_0)$ reaches the infinite-field value $r_{mix}(\infty)$

$$r_{mix}(B_0) \cong r_{mix}(\infty) \equiv r_{mix}(B_0 = \infty) \quad (3)$$

so that it becomes independent of field.

The measured proton relaxivity, r_{mix}, of M_I versus $\nu_I^{1/2}$ [MHz$^{1/2}$] is reported in Figure 1. In the frequency interval, 0−20 MHz, it grows markedly because the electronic relaxation time T_{1e} increases rapidly[5] and becomes significantly longer than τ. Then, the increase of T_{1e} has less effect. At a field $B_0^{indep} \geq$ 3−4 T (120 MHz), r_{mix} reaches a plateau, where $r_{mix} = r_{mix}(\infty) = 4.1$ s^{-1} mM^{-1} to within the experimental accuracy of 2−3%.

(P2) In the OS medium-field range defined by[4] $\omega_I\tau < 1$ and $\omega_S\tau \gg 1$, for $B_0 \geq B_0^{indep}$, the longitudinal relaxivity is

$$r_1 \cong r_{mix} - (\lambda_D/D^{3/2})\sqrt{\pi\nu_I} \cong r_{mix}(\infty) - (\lambda_D/D^{3/2})\sqrt{\pi\nu_I} \quad (4)$$

If $B_0 \geq B_0^{indep}$, the inequality $(r_{mix} - r_1) \leq 0.5 r_{mix}$ is a conservative rule ensuring that eq 4 holds.

The experimental r_1 versus $\nu_I^{1/2}$ [MHz$^{1/2}$] is shown in Figure 1. The frequency axis can be split into three field ranges: in the "low"-field range below 120 MHz, r_1 depends on the details[4−6] of the relative translational and rotational motions of the interacting species and on the electronic relaxation. Its behavior, just as that of r_{mix}, has no simple features. In the OS medium-field (mf) range from $\nu_{I\min}^{mf}$ = 120 MHz to $\nu_{I\max}^{mf}$ = 800 MHz, r_1 shows the linear decrease in $\nu_I^{1/2}$ of eq 4. The relative diffusion coefficient D = 0.88×10^{-5} cm^2 s^{-1} is readily obtained from the r_1 and $r_{mix}(\infty)$ values measured at one frequency. In the "high"-field range $\nu_I \geq \nu_{I\max}^{mf}$, r_1 is again a complicated function depending on the details of the spatial dynamics of the interacting species.

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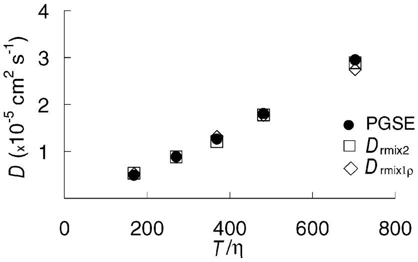

Figure 2. Relative diffusion coefficients versus T/η in D_2O.

(P3) For $B_0 \geq B_0^{\mathrm{indep}}$, D is inversely proportional to r_{mix}

$$D = k_{\mathrm{solv}}/r_{\mathrm{mix}} = D^{\mathrm{ref}} r_{\mathrm{mix}}^{\mathrm{ref}}/r_{\mathrm{mix}} \tag{5}$$

where k_{solv} is determined by the solvent, that is, the more numerous species. The solvent-related parameter k_{solv} depends very little on the temperature, pressure, and concentrations of reasonable amounts of added neutral or charged spectator species in the solution.[7] Here, spectator species are defined as molecules which do not bind to M_I or M_S for durations of the order of τ or longer. An experimental estimate of k_{solv} is $k_{\mathrm{solv}} = D^{\mathrm{ref}} r_{\mathrm{mix}}^{\mathrm{ref}}$, where D^{ref} and $r_{\mathrm{mix}}^{\mathrm{ref}}$ are the measured values for the M_I/M_S pair in a chosen reference (ref) system.

The reference system is the $(CH_3)_3COD/Gd(dtpa)^{2-}$ pair in D_2O at 298 K. The coefficient D, obtained between 283 and 343 K from eq 5, is plotted versus T/η in Figure 2, where it compares very well with its usual definition as the sum

$$D^{\mathrm{sum}} = D_I^{\mathrm{t}} + D_S^{\mathrm{t}} \tag{6}$$

of the self-diffusion coefficients D_I^{t} and D_S^{t} of $(CH_3)_3COD$ and $Lu(dtpa)^{2-}$, representing $Gd(dtpa)^{2-}$, which were measured by the PGSE NMR technique.[4,8]

The applicability of eq 5 was further tested by adding fair amounts of viscous glycerol or KCl in the reference solution. In the glycerol-d_8/D_2O mixture containing 42% w/w of glycerol, the viscosity increase by a factor of ~3.4 implies a similar reduction of the diffusion rates $D = 0.247 \times 10^{-5}$ cm^2 s^{-1} from eq 5 and $D^{\mathrm{sum}} = 0.243 \times 10^{-5}$ cm^2 s^{-1}, which are in good agreement. When adding 2.65 M of KCl, $D = 0.835 \times 10^{-5}$ cm^2 s^{-1} from eq 5 and $D^{\mathrm{sum}} = 0.854 \times 10^{-5}$ cm^2 s^{-1} change very little with respect to the reference values because the viscosity of a water solution of KCl is not altered by a rather large concentration of this salt, and the self-diffusion of $Gd(dtpa)^{2-}$ is weakly reduced by the Coulomb interactions with the surrounding ions.[1c]

Moreover, if $P1-P3$ hold, c_S and D can be simultaneously estimated as $c_S = x^3/y^2$ and $D = x^2/y^2$ with $x \equiv R_{\mathrm{mix}}/k_{\mathrm{solv}}$, $y \equiv (R_{\mathrm{mix}} - R_{1\mathrm{p}})/(\lambda_D \pi^{1/2} \nu_I^{1/2})$ (see Tables S6, S10, S14).

It was recognized[9] early that PRE studies could provide information about local diffusion at the cost of an accurate and complex treatment involving several parameters. Here, $P1-P3$ allow one to extract relative diffusion rates from experiment with straightforward algebra. The practical implementation of the method for a given M_I/M_S pair is as follows. Take a field B_0 (typically $B_0 \geq 2$ T), where r_{mix} is independent of electronic relaxation. Determine the relative diffusion coefficient D^{ref} of M_I and M_S in a reference solution, for instance, by using $P2$. Measure $r_{\mathrm{mix}}^{\mathrm{ref}}$ in this reference system. Measure r_{mix} in other solutions of M_I/M_S in the same solvent and apply $P3$ to derive D. If M_S is large enough for D_S^{t} to verify $D_S^{\mathrm{t}} \ll D_I^{\mathrm{t}}$, the method can serve to obtain the self-diffusion coefficient $D_I^{\mathrm{t}} \cong D$ of M_I. As noted previously,[4] since the molecular spatial dynamics responsible for the relaxivities takes place over distances of the order of a few nanometers, the method applies to molecular motions in confined media.[2,10] Its extension can be envisaged to large molecules,[11] in particular, if the PRE is induced by superparamagnetic particles,[6d] for which $P1$ and $P3$ hold.

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Supporting Information Available: Proofs of $P1-P3$, the raw experimental data, and complementary information are given in pp S1–S23. This material is available free of charge via the Internet at http://pubs.acs.org.

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