the excitation transfer and that the rate of the energy transfer is faster than the spin lattice relaxation rate of the benzophenone triplet precursor. The experiment was therefore carried out in the time-resolved mode monitoring at the field of the $\Delta M = 2$ resonance line of the naphthalene triplet. Figure 1a shows the transient emissive polarization observed for the naphthalene triplet in glassy solution. The result suggests that a triplet precursor with a higher P_+ population will conserve this polarization during the energy transfer, thus leading to the acceptor triplet also having a higher P_+ population. It is also clear that the spin polarization conservation in the present example is independent of molecular orientation and molecular symmetry of the donor relative to the acceptor. Indeed, the benzophenone triplet precursor is n,π^* in nature while the naphthalene triplet is π,π^*.

In order to confirm the "sign" aspect of the spin polarization conservation, we have examined the system of pyruvic acid and naphthalene under identical conditions. Pyruvic acid undergoes intersystem crossing at high fields with the P_-, the lowest triplet sublevel at high fields, being the most populated.[19] Thus, the same energy transfer process between the polarized pyruvic acid triplet precursor and naphthalene should lead to the observation of a naphthalene triplet in the enhanced absorption mode. Such an observation is confirmed and shown in Figure 1b.

In the above two systems the triplet excitation transfer involved n,π^* to π,π^*. In order to establish that the excitation transfer between π,π^* and π,π^* triplets also conserves spin polarization, we have examined the systems containing coumarin. In a CW experiment irradiation of a glassy solution containing 10^{-2} M coumarin alone led to the observation of the $\Delta M = 2$ resonance line of the coumarin triplet with $D^* = 0.1249$ cm^{-1}, in good agreement with the literature value.[26] Under time-resolved conditions, the same resonance line exhibited a strong emissive polarization, thus suggesting a highly spin-selective intersystem-crossing process. Optical studies[26] have concluded that the lowest triplet state of the coumarin in polar solvents is the π,π^*, with the triplet energy at 2.56×10^2 kJ mol^{-1}. With naphthalene triplet energy at 2.54×10^2 kJ mol^{-1}, excitation transfer between coumarin and naphthalene is thus probable. When a glassy ethanol solution of coumarin and naphthalene was irradiated in the CW mode, both half-field resonance lines of the coumarin and the naphthalene triplets were simultaneously observed. The time-resolved spectra of both triplets exhibited emissive polarization. Under identical conditions, however, a system containing naphthalene alone did not yield any observation.

It should be noted that all the present experiments were carried out at a magnetic field of about 1500 G. In our early treatment of the random triplet polarization in CIDEP,[33] we have considered that the polarization magnitude may very well be field dependent. Thus, it is not unreasonable to expect that, in the triplet energy transfer processes, both the specificity and the magnitude of the polarization conserved may vary with magnetic field. A critical test for the magnetic field effect would require a variable frequency ESR spectrometer and/or a zero-field spectrometer, which are not presently available in our laboratory.

The current CIDEP results also allow an estimate of the T_1's of the triplets in the glassy solution at 77 K. In all three systems, the naphthalene triplet T_1 was about 2 μs. The T_1 of the coumarin triplet was 3.3 μs. Very few triplet T_1 values are known in literature,[27-29] and almost all of them were measured in single crystals. As a comparison, T_1's of triplet naphthalene in single crystals vary between 6.6×10^{-8} s at room temperature and 3×10^{-4} s at 4.2 K. Recently, the excitation transfer and the relaxation processes of excited triplets in condensed phases have attracted extensive theoretical attention.[30-32] It is hoped that the results reported here will add some insights into the mechanism of spin polarization conservation in excitation transfer, which heretofore has mainly been in the domain of single crystals. The understanding of the mechanism of excitation transfer is a primary step toward the understanding of the overall spin-specific photochemistry.

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Registry No. Benzophenone, 119-61-9; pyruvic acid, 127-17-3; coumarin, 91-64-5; naphthalene, 91-20-3.

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Self-Diffusion of Water at the Protein Surface: A ^{17}O Measurement

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The relative motion of solvent molecules is of considerable interest from both fundamental and applied viewpoints. In the particular case of water at the surface of a large solute particle such as a protein molecule, ideas about the surface mobility of water have spanned the range from rigid or icelike to unperturbed motion as in pure water.[1] In this communication we report an analysis of the frequency dependence of the water proton longitudinal nuclear magnetic relaxation rates measured over a wide frequency range for a protein labeled with a paramagnetic nitroxide probe molecule that provides a measure of the water diffusion coefficient at the protein surface.

The frequency dependence of the nuclear magnetic relaxation rate of water protons in a solution containing a significant concentration of a paramagnetic molecule is dominated by the intermolecular electron–nuclear dipole–dipole interaction. In the case of nitroxide solutes in water this fact has been exploited to measure the correlation times for the solute–solvent interaction.[2-4] While this earlier work may be criticized for the use of a theory less complete than that now available,[5-7] the essential features remain, namely, that the frequency dependence of the proton relaxation of the solvent molecules will provide a measure of the correlation time for the relative translational motion of the water and the nitroxide paramagnetic center. In the present experiment the nitroxide was firmly affixed to the protein by covalent bonds. The reduced mobility of the nitroxide was verified from the EPR spectra. The water proton nuclear magnetic relaxation rate was measured as a function of magnetic field strength corresponding to Larmor frequencies from 0.01 to 30 MHz. The data were analyzed in terms of the translational contribution of the water diffusing in the vicinity of the paramagnetic label on the protein to yield a good estimate for the relative diffusion coefficient for the water and the nitroxide. Since the nitroxide is constrained to translate with a much slower correlation time than that of the

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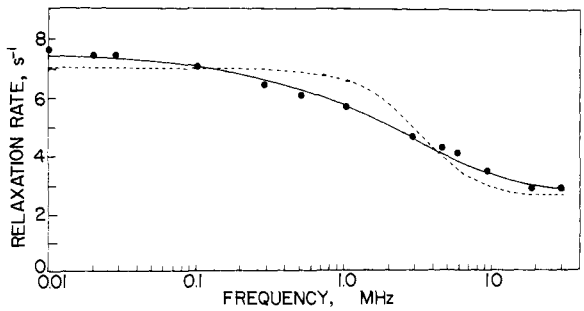

Figure 1. The water proton nuclear magnetic relaxation rate as a function of magnetic field strength plotted as proton Larmor frequency for a 0.28 mM solution of bovine serum albumin labeled with 4.6 nitroxides per protein molecule at pH 6.4 and 286 K. The solid circles are the experimental points and the solid line the calculated curve for the translational diffusion model[9] assuming the diffusion coefficient for water at the surface of $(3.0 \pm 0.6) \times 10^{-6}$ cm^2 s^{-1} and the minimum distance of approach of the spins of $(1.9 \pm 0.3) \times 10^{-8}$ cm. The dotted curve was calculated on the basis of a rotational diffusion model[2] using a rotational correlation time of $(0.6 \pm 0.2) \times 10^{-10}$ s and an average radius of the water diffusional unit of $(1.8 \pm 0.1) \times 10^{-8}$ cm. The sample contained 1 mM sodium azide.

water, the measurement provides the self-diffusion coefficient for water at the protein surface. Further, the results are not dependent upon the electron spin relaxation time of the nitroxide as long as it remains large compared with the other correlation times entering the problem such as the protein rotational correlation time or the correlation time for the relative diffusion of the nitroxide and the solvent protons. This condition will generally be satisfied for nitroxide radicals.[3] A representative dispersion plot of the data is shown in Figure 1 along with the computed fit to the data. Other models such as rotational diffusion[2,4] gave much poorer fits to the dispersion plots. The analysis, to be completely described elsewhere,[8] follows the development of Freed very closely,[9] and the solid line through the data points is based on this force-free diffusion model. The water molecule translational diffusion coefficient in the immediate vicinity of the nitroxide determined from this analysis is $(3.0 \pm 0.6) \times 10^{-6}$ cm^2 s^{-1} at 286 K. This is to be compared with a value of $(16 \pm 3) \times 10^{-6}$ cm^2 s^{-1} for pure water obtained from an aqueous solution of a spin label, which agrees with the published results of others.[10]

Though the data set is quite sufficient for this analysis, we take this result as an approximation because the theory has not specifically included the excluded volume effect required by the finite size of the surface, in this case the protein particle, and pair correlation effects between particles.[5,7] The neglect of these effects can be shown to result in a calculated diffusion constant that is somewhat smaller than the correct value and a calculated distance between the centers of the interacting particles that is larger than the correct value.[8] This result also represents an average of the diffusion coefficient over distance from the nitroxide that is initially weighted by the usual sixth power of the distance. However, the actual weighting as a function of distance from the surface is considerably weaker because the average required is over volume so that the distance dependence falls to approximately inverse third power dependence. The method, therefore, provides a characterization of water mobility at the surface defined as the first several monolayers of water. Further confirmation of our results is achieved by taking the limiting form of the equations which is independent of the model chosen[6] and obtaining the translational diffusion coefficient from the slope of a plot of relaxation rate vs. the square root of frequency. We obtain $D = (2.2 \pm 0.3) \times 10^{-6}$ cm^2 s^{-1} for the data below 0.6 MHz in Figure 1. A similar analysis for an aqueous solution of spin label gives $D = (18 \pm 4) \times 10^{-6}$ cm^2 s^{-1}.

The method employed here is perfectly general as a means for providing local relative diffusion coefficients. The same strategy should be equally useful and valid for the characterization of nonaqueous solvent mobility on any surface that could be labeled with a radical having the appropriately long electron relaxation time.

Experimental Section. Nuclear magnetic relaxation rates were made on a field cycling instrument built in this laboratory with the collaboration of Drs. Seymour Koenig and Rodney Brown, III, and is described elsewhere.[11] The spin-label samples were prepared as described by Twining et al.[12] using 3-[[(2,5-dioxo-1-pyrrolidinyl)oxy]carbonyl]-2,5-dihydro-2,2,5,5-tetramethyl-1*H*-pyrrolyl-1-oxy (Molecular Probes) which reacts with free amino groups on proteins.[13] The spin-label concentration was measured by double integration of the ESR spectra. The protein solutions contained 1 mM sodium azide as a preservative and the albumin (Sigma, essentially fatty acid free) concentration was calculated from $\epsilon_{280} = 4.34 \times 10^{4}$ L mol^{-1} cm^{-1}. The solutions were unbuffered, and the pH was adjusted by adding 0.01 N NaOH. The parameters were determined using a nonlinear least-squares simplex procedure.

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Atropisomerism in Metal Chelates. Preparation and Partial Resolution of (3,4-Diacetyl-2,5-hexanedionato)bis[((2,2′,2″-triaminotriethyl)amine)cobalt(III)] Ion

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One of the authors (Y.N.) has reported a series of optically active complexes (atropisomers), the chirality of which arises from the restricted rotation of an aromatic ring group.[1]

In the present study, we are interested in a new type of atropisomer, containing 1,1,2,2-tetraacetylethane[2] (taeH$_2$).[3] Tetraacetylethane adopts an interesting dienolic form in which the two planer acetylacetone units are perpendicularly joined back to back in the crystal.[4] The acetylacetone moieties are able to form stable

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