Determination of the singlet state lifetime of dissolved nitrous oxide from high field relaxation measurements

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Longitudinal spin relaxation due to modulation of dipolar interactions often limits the development of hyperpolarized magnetic tracers. Recently, it has been demonstrated that transferring spin order to a singlet state significantly increases the polarization lifetimes in systems where nitrous oxide is dissolved in a liquid solvent. Additionally, previous studies have suggested that the longitudinal relaxation of nitrous oxide is largely dominated by the spin-rotation interaction. Models of spin-relaxation under Brownian motion naïvely predict the angular momentum reorienting correlation time of the spin rotation interaction to be inversely proportional to the viscosity of the solution. This dependence implies the singlet lifetime can be lengthened by increasing the dissolving solvent's viscosity—an extension which is not observed. Our work formulates a model which describes the relaxation of nitrous oxide dissolved in various solvents. We investigate the effect of altering the temperature of the solvent, as well as the effect of varying solute-solvent interactions on the singlet state as well as the longitudinal polarization lifetime. We predict the singlet lifetime for nitrous oxide dissolved in several solvents by fitting rotational and angular momentum correlation times measured at high magnetic field, and relate singlet relaxation to translational diffusion constants.

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I. INTRODUCTION

NMR and MRI can determine the spatial distribution of substrates *in vivo*. The poor sensitivity of these techniques renders detection of nuclei other than protons difficult in a clinical environment. This leaves the potential to image alternative nuclear species largely unused, despite the opportunity to visualize specific molecular species and metabolic activity using individual nuclear resonance frequencies. Recent advances in the field of hyperpolarization physics allow for nuclear spin polarization of labeled substrates *ex vivo* to levels several orders of magnitude greater than the equilibrium Boltzmann polarization.¹ Several groups have subsequently demonstrated spectroscopy and imaging of hyperpolarized nuclei *in vivo*. The enhanced sensitivity suggests methods to detect numerous diseases such as cancer, aberrant metabolic conditions, and chronic obstructive pulmonary disorders.^{2–4} Unfortunately the signal enhancement afforded by hyperpolarizing nuclei decays with time due to longitudinal relaxation of the nuclear spins. Delivery of the hyperpolarized substrate to the target organ and significant metabolic alteration must take place before the hyperpolarization has decayed.^{5–7} This limits many otherwise attractive applications of hyperpolarized substrates which take place on longer time scales.

Recently, substantial increases in polarization lifetimes have been achieved by transferring nuclear spin polarization to long lived states.^{8–10} In the present work, we investigate the relaxation processes in one class of long lived states, singlet states. We quantitatively describe the source of singlet state

relaxation, and describe conditions under which the rate can be further reduced. Although we present a model specific to nitrous oxide we expect similar models can be constructed to predict the singlet lifetime in molecules where nuclear spin relaxation is also dominated by the spin-rotation interaction at low magnetic field. We note that although the singlet state has no magnetic moment, conversion into a triplet state by adiabatic transport to an imaging-strength magnetic field results in distinct antiphase NMR lines, which makes it suitable for high sensitivity MRI.

II. THEORY

The field dependent Hamiltonian of two coupled nuclear spins is given by

$$H = \gamma B(1 + \delta_1)\vec{I}_1 + \gamma B(1 + \delta_2)\vec{I}_2 + 2\pi J\vec{I}_1 \cdot \vec{I}_2, \quad (1)$$

where $\vec{I}_1$ and $\vec{I}_2$ are the two nuclear spins, γ denotes their gyromagnetic ratio, J the scalar coupling between the nuclei, δ_1 and δ_2 are the respective chemical shifts of the two nuclei, and B is the magnetic field. In low magnetic field, the scalar coupling dominates the interaction and the singlet-triplet basis describes the system's eigenstates,¹¹

$$\begin{aligned} |S_0\rangle &= 2^{-1/2}(|\alpha\beta\rangle - |\beta\alpha\rangle) & |T_1\rangle &= |\alpha\alpha\rangle \\ |T_0\rangle &= 2^{-1/2}(|\alpha\beta\rangle + |\beta\alpha\rangle) & |T_{-1}\rangle &= |\beta\beta\rangle. \end{aligned} \quad (2)$$

Here $|\alpha\rangle$ denotes a spin aligned with the magnetic field, $|\beta\rangle$ opposed to it. S_0 denotes the singlet state with total spin $I = 0$, and $|T_\mu\rangle$ the μ th azimuthal angular momentum of the triplet state $I = 1$.

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In sufficiently high field, the chemical shift difference between the nuclei dominates the scalar coupling interaction and the eigenstates become $|\alpha\alpha\rangle, |\alpha\beta\rangle, |\beta\alpha\rangle, |\beta\beta\rangle$.¹¹ Subsequent transport of the system from high field to low field results in a transfer of quantum level populations from one eigenbasis to the other, described as⁸

$$|S_0\rangle \leftrightarrow |\beta\alpha\rangle \quad |T_1\rangle \leftrightarrow |\alpha\alpha\rangle \quad |T_0\rangle \leftrightarrow |\alpha\beta\rangle \quad |T_{-1}\rangle \leftrightarrow |\beta\beta\rangle, \quad (3)$$

where J and γ have the same sign ($|S_0\rangle$ and $|T_0\rangle$ change places in Eq. (3) otherwise).

Furthermore, this transfer is reversible. Selectively populating the $|\beta\alpha\rangle$ state in a high magnetic field, and adiabatically transferring the system to a region of low magnetic field results in an increased population of $|S_0\rangle$ in low field.^{12,13} The experiments described here utilize this technique to generate and measure relaxation of the singlet state separately from the longitudinal relaxation of the individual nuclei.

Intra-molecular dipolar interactions typically dominate nuclear spin relaxation in the dissolved or liquid state. Because of its antisymmetry, the singlet state is immune to relaxation by the symmetric, intra-pair dipolar interaction,¹⁴ and also by the symmetric component of the chemical shift anisotropy (CSA) and intermolecular dipolar interactions.^{14–16} Relaxation of the singlet state of dissolved $^{15}\text{N}_2\text{O}$ is therefore dominated by the largest remaining interactions with an antisymmetric component, the spin-rotation interaction, and if present with sufficient concentration, the interaction with paramagnetic dissolved species.¹⁰ The relaxation period T_S of the singlet state due to the spin-rotation interaction at low field ($\gamma B(\delta_1 - \delta_2) \ll J$) under these conditions is given by¹⁴

$$\frac{1}{T_S^{sr}} = \frac{4k_B T (C_{\perp 1} - C_{\perp 2})^2 I_{\perp} \tau_c^{sr}}{3\hbar^2}, \quad \tau_c^{sr} = H \frac{I_{\perp}}{8\pi\eta r_h^3}. \quad (4)$$

In the above, k_B denotes the Boltzmann constant, T the absolute temperature, and $C_{\perp}$ the nonvanishing components of the spin rotation tensors for the two ^{15}N nuclei. The spin-rotation correlation time is denoted by τ_c^{sr} , and its form arises from a Stokes-Einstein model of Brownian motion, with the commonly employed correction H for solvent molecule size and solvent-solute van der Waals interactions. We assume that upon dissolution of nitrous oxide molecules in liquid of viscosity η , the individual nitrous oxide molecules interact with a solvation shell of solvent molecules; the moment of inertia of the nitrous oxide-solvent complex is denoted by $I_{\perp}$. The hydrodynamic radius of the idealized sphere molecule is denoted by r_h .

Except in simple cases of minimally interacting solutions, H and $I_{\perp}$ are difficult to calculate from first principles, although Eq. (4) can be used to predict the variation of T_S with temperature and viscosity. Furthermore, high-field measurements of longitudinal relaxation rates can be used to constrain these quantities and verify the model of molecular rotational dynamics. In this publication, we investigate the above strategy for predicting and increasing the singlet lifetime.

III. MATERIALS AND METHODS

Doubly ^{15}N -labeled N_2O gas (98% + ^{15}N fraction, Cambridge Isotope Laboratories, Inc.) was dissolved in a variety of solvents in 5 mm NMR tubes (Wilmad Glass). Solutions were deoxygenated under N_2 prior to introduction of N_2O . The solvents were mixed with Chelex (Biorad, Chelex 100 resin) for 24 h in order to chelate paramagnetic impurities. Subsequent removal of the Chelex granules using filter paper was intended to reduce overall impurity concentration, and particularly that of paramagnetic metal salts. Several samples were then mixed with 100 μM of ethylenediaminetetraacetic acid (EDTA) to further chelate any remaining paramagnetic impurities. N_2O was then introduced in a two-step process: (1) the gas was released from a lecture bottle into a pumping manifold with a calibrated volume and (2) a section of the NMR tube was then placed in contact with liquid N_2 using a custom dewar. This process condensed the N_2O gas into the cold section of the NMR tube. The amount of gas condensed inside the tube was monitored using a pressure gauge on the calibrated volume. The tube was then flame-sealed and the custom dewar was removed. The tube was allowed to warm up to room temperature and inverted several times, after which the dissolved N_2O was in equilibrium with a gas pressure of 7–40 bar. Individual tube pressures were estimated using the calibrated pressure drop, the volume of the sealed tube, and published N_2O solubilities. Relative sample pressures were verified by comparing the integrated NMR line spectra scaled by published N_2O solubilities.

NMR experiments were performed on a Varian 11.7 Tesla vertical bore system. After the sample was fully thermally relaxed to the 11.7 T field, the N_2O singlet state was populated using a two-step process. First, a selective long (3.3 ms) square π pulse centered on the NMR frequency of the terminal ^{15}N spectral doublet was applied. The pulse length was chosen to have a spectral node (the eleventh) at the frequency of the adjacent central ^{15}N doublet, enabling selective excitation of the terminal ^{15}N nucleus. The tube was then adiabatically transported by hand from the bore of the 11.7 T magnet to the center of a custom-built low-field μ -metal shielding cylinder, with cap (8.75" diameter, 17.5" length). This process increased the $m = 0$ singlet state relative to the triplet state populations. The field in the μ -metal shielding cylinder is less than 0.1 Gauss. After a predetermined time interval, the sample was reintroduced into the bore of the 11.7 T magnet. A hard, non-selective $\pi/2$ pulse was applied to both nuclei to determine their polarization before significant relaxation to thermal polarization had taken place. The singlet lifetime was determined by fitting the antiphase population, measured at high field, to a bi-exponential; the fast time constant corresponds to equilibration of the triplet populations (low-field T_1), and the longer decay constant to the lifetime of the singlet state.⁸ In practice, the time-constants differ by approximately an order of magnitude and are easily separated, see Fig. 1.

Determination of the singlet lifetime required accurate estimation of the amplitude of the antiphase NMR signals after long delays. When investigating solvents with low N_2O solubility, these spectra exhibited correspondingly low SNR,

FIG. 1. Polarization decay at short time periods correspond to interconversion of the triplet states, while the behavior at long time intervals is dominated by relaxation of the singlet state.

and an unavoidable splitting due to the other ^{15}N . Phasing ambiguity and non-Lorentzian lineshapes were often encountered and were handled by integrating correlations between points in the spectra separated by the well-measured J-coupling prior to fitting to a single Lorentzian lineshape. In other words, antiphase amplitude I in the complex spectrum $S(f)$ with splitting J and doublet center frequencies f_0 and f_1 was characterized by integrals over regions $\delta \gg J$,

$$I = \sqrt{\int_{f_0-\delta}^{f_0+\delta} S(f)S^*(f+J)df} - \sqrt{\int_{f_1-\delta}^{f_1+\delta} S(f)S^*(f+J)df}. \quad (5)$$

Only spectra exhibiting clear anti-phase behavior were used. This correlation avoids difficulty in phasing low-signal spectra and, unlike use of the magnitude spectrum, averages to zero in the absence of signal.¹⁰

In addition to low-field singlet lifetimes, the longitudinal relaxation times were measured at high field with a standard Carr-Purcell inversion-recovery pulse sequence. The bore temperature air intake vent for the sample was supplied with nitrogen gas to avoid water condensation on the magnet electronics for measurements recorded below the ambient lab temperature. Sample temperatures were determined by utilizing the pre-installed temperature sensor on the Varian NMR probe.

Additionally, the longitudinal relaxation time was measured in four solvents, H_2O , D_2O , neat isopropanol, and carbon disulfide as a function of temperature. These solvents were chosen as representative of a variety of varying physical characteristics; H_2O and D_2O are polar solvents, and differences in measured N_2O relaxation rates discern the effect of protonation of the solvent. Carbon disulfide is an aprotic, non-polar, spinless, and low-viscosity solvent; relaxation in CS_2 is expected to be minimally affected by direct interaction with the solvent molecules. Finally, the effect of solubility on relaxation was investigated by utilizing neat isopropanol. Nitrous oxide is highly soluble in isopropanol, as compared to the other solvents studied. A single measurement of nitrous oxide dissolved in deuterated isopropanol was also performed to further constrain relaxation effects related to the protonation of the solvent, as opposed to solubility effects. A longitudinal spin relaxation model was developed by incorporat-

ing the effects of dipolar relaxation, chemical shift anisotropy, spin rotation, and solvent viscosity.^{14,17,18}

$$\frac{1}{T_1} = R_{sr}\tau_c^{sr} + R_{csa}\tau_c^{rot} + R_{dipole}\tau_c^{rot}, \quad (6a)$$

$$R_{sr} = \frac{4k_B T C_{\perp}^2 I_{\perp}}{3\hbar^2}, \quad R_{csa} = \frac{2}{15}(\gamma B(\Delta\sigma))^2$$

$$R_{dipole} = \frac{3}{2}\left(\frac{\mu_0}{4\pi}\right)^2 \gamma^4 \hbar^2 r_{N-N}^{-6}, \quad \tau_c^{rot} = \frac{I_{\perp}}{6k_B T \tau_c^{sr}}. \quad (6b)$$

In Eq. (6), τ_c^{rot} denotes the correlation time. This is identical for both the chemical shift anisotropy and dipole interaction since both correlation times arise from the same molecular randomization mechanism. Chemical shift anisotropy correlation time γ denotes the gyromagnetic ratio of the nitrogen nuclei, $\Delta\sigma$ the chemical shift anisotropy, $C_{\perp}$ is the relevant nuclear spin rotation coupling constant, and r_{N-N} denotes the nitrogen–nitrogen bond length in N_2O . The expressions of Eq. (6) are well-known from NMR relaxation theory and applicable in the absence of additional relaxation mechanisms and rapid collisional reorientation, a condition that is well met here.^{17,18} Correlations between mechanisms of similar angular dependence (e.g., CSA and dipole–dipole) are not necessary to consider in this case due to the small dipolar interaction. The relationship between angular and J-reorienting correlation times expressed in Eq. (6b) is the result of very general rotational diffusion arguments expressed in Ref. 17 and hereafter referred to as the Hubbard model. We postulate that deviations from the Hubbard model arise from microscopic dynamics which are more accurately described by the reorientation of a solvent-solute complex with a different moment of inertia than that of the free solvent molecule. It can be shown that utilizing the assumptions of the Hubbard model concerning the relationship between the angular momentum and J-reorienting correlation times, one is able to derive both Eq. (6) for the longitudinal relaxation rates at high field as well as the relaxation of the singlet state at low field given in Eq. (4) (Ref. 14).

Accurate measurements of the molecular constants found in Eq. (6) can be found in the literature; the spin-rotation tensors of the linear N_2O take a simple form and can be described by a single value for each nucleus ($C_{\perp, \text{central}} = 3.35 \text{ kHz}$, $C_{\perp, \text{terminal}} = 2.48 \text{ kHz}$ ¹⁹), and the CSA tensors can be characterized by a single value $\Delta\sigma = \sigma_{\parallel} - \sigma_{\perp}$: $\Delta\sigma_{\text{central}} = 505 \text{ ppm}$, $\Delta\sigma_{\text{terminal}} = 366 \text{ ppm}$.²⁰ The size of the through-space dipolar coupling can be calculated and agrees well with a measured value of 865 Hz from powder-pattern solid state spectra (data not shown). Because CSA exceeds the dipolar coupling by over an order of magnitude, intramolecular dipolar relaxation can be neglected and is not discussed further.

IV. RESULTS

Typical singlet lifetime measurements are shown in Fig. 1. The singlet character metric of Eq. (5) is plotted as a function of holding time in the mu-metal chamber and shows

FIG. 2. Predictions and experimental data of the T_1 of the terminal and central nitrogen nuclei in nitrous oxide over a wide temperature range are given in the left and center panels. Predictions and measurements of the variation of the singlet state lifetime with temperature are shown in the right panel. H_2O and D_2O T_1 data are plotted in blue circles and triangles, CS_2 in red circles, and isopropanol in green circles. The closed circles describe the measurements made of the terminal nitrogen nucleus, and the open circles describe the measurements on the central nitrogen nucleus. H_2O T_S data are plotted in blue squares, CS_2 in red squares, and isopropanol in green squares.

a characteristic biexponential decay; the longer time constant represents T_S .

As can be seen from Figs. 2(a) and 2(b), the model accurately describes the temperature dependence of the relaxation rates, although the solvents differ substantially with respect to molecular reorientation dynamics; thus, the overall magnitude and even the trend with temperature may be qualitatively dissimilar. The leftmost and center panels of Fig. 2 show the longitudinal lifetimes of nitrous oxide dissolved in water and deuterium oxide (blue circles and triangles), isopropanol (green circles), and carbon disulfide (red circles) over a range of temperatures. The left panel of Fig. 2(a) depicts the temperature dependence of the T_1 of the terminal nitrogen nuclei of nitrous oxide (closed data points), and the center panel shows the dependence of the central nitrogen nuclei (open data points). The rightmost panel summarizes the temperature dependence of the singlet state lifetime of nitrous oxide dissolved in water (open blue square), isopropanol (open green square), and carbon disulfide (open red square). The longest T_1 occurs at a characteristic temperature, for which CSA and spin-rotation mechanisms have comparable contributions to the relaxation rate. The fits shown in the figure are to the functional form of Eq. (6) in which $I_\perp$ and Hr_h^{-3} , assumed temperature-independent, are allowed to vary. The fits appear shifted with respect to the data. We postulate this may be the result of assuming $I_\perp$ and Hr_h^{-3} are temperature independent parameters. This point is elaborated on in Sec. V. The resulting fit values are summarized in Table I.

Correspondence between T_S predicted by Eq. (4) and the measured values are shown in (Table I). The table shows accurate agreement between the predicted T_S and that measured experimentally, as well as the ratio T_S/T_1 at 298 K. The magnitudes of $I_\perp$ are within a factor of 2 of the moment of inertia of free nitrous oxide ($41.6 \text{ amu}\cdot\text{\AA}^2$).

Among the relaxation mechanisms modeled in Eq. (6), only the antisymmetric component of the spin-rotation interaction is expected to cause significant relaxation of the singlet-like state. As is seen in the high-temperature limit of the T_1 data, this contribution is expected to diminish as the

temperature is decreased, primarily due to increased solvent viscosity. The measured temperature dependence of the singlet lifetime is shown in Fig. 2(c), juxtaposed with the dependence calculated from Eq. (4) and the hydrodynamic variables $I_\perp$ and Hr_h^{-3} derived from the high-field T_1 data.

V. DISCUSSION

Based on the success of this model, we believe that the dominant relaxation mechanism depopulating the singlet state in nitrous oxide is the spin-rotation interaction, and that spin-lattice relaxation is predominantly caused by a combination of the spin-rotation interaction and CSA. We note when N_2O is dissolved in alcohol and water/ D_2O solvents, a substantial degree of deviation from the Stokes-Einstein model of molecular dynamics is required in order to reproduce the measured relaxation characteristics. In the case of CS_2 , however, this is not the case; the best fit $I_\perp = 42.1 \text{ amu}\cdot\text{\AA}^2$ is very close to the

TABLE I. Prediction of singlet and longitudinal relaxation times from high magnetic field measurements. The first two rows compare the measured singlet lifetime with the prediction of the Hubbard model, in four different solvents. The third and fourth rows compare the measured extension of the singlet state with respect to the uncorrelated longitudinal lifetime for the terminal and central nitrogen nuclei. T_1 represents the average nuclear relaxation rate, i.e., $1/T_1 = 1/2 (1/T_1^N + 1/T_1^C)$, where T_1^N denotes the terminal nitrogen, and T_1^C the central nitrogen. The fifth and sixth row contain the measured and fitted T_1 values. The seventh and eighth row contain the fitted values of the moment of inertia of the solute-solvent complex, and the product of the hydrodynamic volume and the microviscosity stick-slip condition correction factor.

	H_2O	D_2O	CS_2	Isopropanol
T_S (min., 298 K)	22.8 ± 0.9	21.5 ± 1.9	8.5 ± 1.2	11.1 ± 0.5
T_S (theory)	22.7	24.8	9.8	10.0
T_S/T_1 (measured)	12.9	11.1	10.6	11.7
T_S/T_1 (theory)	14.4	14.8	11.6	12.1
T_1 (s, 298 K)	106	116	48	57
T_1 (s, theory)	95	87	44	55
$I_\perp$ ($\text{amu}\cdot\text{\AA}^2$)	100.2	100.2	42.1	137.8
Hr_h^{-3} ($\text{\AA}^{-3}$)	0.40	0.40	2.1	1.1

41.6 amu-Å² calculated based on the known bond distances, and the corresponding HR_h^{-3} is reproduced if one assumes the geometrically calculated hydrodynamic radius of a linear molecule ($r_h = 1.46$)²¹ and the $H = 6$ predicted by the Gierer-Wirtz model of a small, non-interacting solvent molecule re-oriented by solvent of similar size.²²

Although the other solvents do not reproduce the Hubbard model of rotational diffusion is not surprising, discrepancies of an order of magnitude or more, particularly in viscous, self-interacting, or solute-interacting solvents, are commonly observed.^{23,24} Our choice of fit parameters is an attempt to give physical interpretation to the discrepancies observed, although several equivalent choices are possible.^{22,25} Noting the differing dependencies of the two correlation times on the fit parameters HR_h^{-3} and $I_\perp$, our optimization is equivalent to independent optimization of τ_c^{sr} and τ_c^{rot} to fit the high-field T_1 data. The same is true of the other formulations as well.

It is important to note that interaction with paramagnetic impurities can bias relaxation rate measurements, including those of singlet-like states.²⁶ In addition to the removal of paramagnetic dissolved oxygen, we tested for the presence of a gaseous paramagnetic impurity in nitrous oxide lecture bottle itself (both NO and O₂ are paramagnetic and might be present in a N₂O sample). However, two samples, differing by a factor of four in filling pressure, yielded the same T_1 and T_s despite what would be expected to be gaseous impurity levels in the same ratio. The effect of paramagnetic relaxation due to impurities present in the solvents was investigated by comparing the T_1 and T_s in untreated, once-chelexed and 4×-chelexed samples. No significant difference was observed in either H₂O or isopropanol relaxation characteristics. Furthermore, subsequent addition of EDTA to the filtered chelexed samples did not result in different relaxation lifetimes. In order to rule out the potential for relaxation of the singlet state due to interactions with the NMR tube wall, a sample tube was constructed with a polytetrafluoroethylene liner to prevent contact of the solvent with the glass NMR tube wall—this did not increase either the T_1 or T_s . Additionally, the singlet lifetime in NMR tubes with different diameters was measured. We expected the T_s to be lower in tubes with smaller internal diameter if a significant contribution to the overall relaxation could be attributed to the interaction of the nitrous oxide with the tube wall—this, however, was not observed.

The similar magnitudes of T_1 and T_s in the H₂O and D₂O samples would imply that if paramagnetic impurities were a significant cause of relaxation then nearly identical impurity concentration contaminated both samples. Additionally, the similarity of T_1 and T_s in the protonated and deuterated isopropanol samples would again require identical paramagnetic impurity concentrations in the two samples. Furthermore, the similar ratio of T_1 of the terminal and central nitrogen nuclei in both the deuterated and protonated water and isopropanol samples would imply the ratio of the magnitude of the random fields at the location of the two nitrogen nuclei nuclear sites due to paramagnetic impurities is nearly identical in all four samples, and is similar to the ratio determined from the

ratio of spin rotation and chemical shift anisotropy tensors at the two nuclear sites. Although these set of coincidences are a possibility individually, taken together they seem highly unlikely. The relatively low contribution of relaxation arising from paramagnetic impurities at high magnetic fields is consistent with measurements of transport of hyperpolarized nuclei which imply relaxation of T_1 from paramagnetic impurities is typically only significant in low magnetic fields.²⁷

The effect of the deoxygenating procedure on the solvent relaxivity was investigated as well. The study's samples were deoxygenated with N₂ gas prior to filling with N₂O. To determine the efficiency of the deoxygenating procedure, the singlet lifetime was compared to that of an additional sample, which underwent four freeze pump thaw cycles to remove residual dissolved oxygen. Ascorbic acid was also added to the solvent to reduce any superoxide (O₂⁻), hydroperoxide (HOO·), and dioxygen radicals in the solvent.²⁶ Neither procedure yielded a difference in the singlet lifetimes.

Modification of the sample temperature (Fig. 2(c)) was the only procedure that was found to significantly change the relaxivity of the solvent. From these tests, we conclude that interactions with species other than the solvent were not significant in determining either T_s or T_1 .

In order to determine if intermolecular interactions affect the relaxation characteristics, we compared T_s and T_1 measurements in H₂O and D₂O. As is seen in Fig. 2 and Table I, no significant difference can be seen between the protonated and deuterated solvents. The relatively small contribution to the overall relaxation rates due to intermolecular relaxation was exhibited in the T_1 and T_s measurements of protonated and deuterated isopropanol as well. The T_1 of the central and terminal nitrogen of nitrous oxide dissolved in protonated isopropanol at 303 K are 40 s and 83 s as compared to 46 s and 87 s for the sample dissolved in deuterated isopropanol. Additionally, the singlet lifetimes at 298 K were not significantly different in the protonated versus deuterated isopropanol samples and were measured to be 11.1 ± 0.5 min, and 11.8 ± 0.9 min, respectively. Furthermore, we note that the spinless CS₂ has the poorest relaxation characteristics of the solvents studied, indicating that although solvent-solute interactions do determine both T_1 and T_s , it is not primarily through interaction with the solvent nuclear spin.

We note that diffusion or flow of singlet molecules out of the sensitive region of the detection coil might be misinterpreted as singlet relaxation. However, the expected diffusion distance (<2 mm during the longest T_s studied) is not sufficient to introduce significant error in these studies. Furthermore, a larger, custom RF coil was built and used to transfer polarization to the singlet state in a sample region much greater (~10 cm) than the receive coil dimension. This yielded identical magnitudes of the singlet lifetimes, consistent with the assertion that the singlet lifetime is minimally influenced by diffusion of nitrous oxide out of the sensitive region of the coil over the time course of these experiments.

Although the model of singlet state relaxation in dissolved N₂O presented here captures the essential features of the measurements, including the peaked temperature dependence of T_1 , the increased T_s at low temperature, and the T_s/T_1 enhancement factor, two significant limitations are ap-

parent. First, the ratio of high-field T_1 measurements taken at the two nitrogen positions in the high-temperature (spin-rotation dominated) regime is not equal to the predicted $(C_{\perp, \text{terminal}}/C_{\parallel, \text{terminal}})^2 = 1.82$, but is instead 1.51 ± 0.02 . This discrepancy may indicate an error in the published spin-rotation tensor values, or the presence of a relaxation mechanism of unknown origin. In addition, comparison of the theoretical and measured T_1 and T_S values in Fig. 2 reveals a small but significant discrepancy. In particular, the temperature of the maximum high-field T_1 and the extent of the T_S increase at low temperature differ significantly from the predicted values in water and isopropanol. We believe that these discrepancies represent a temperature-dependence of the microviscous interactions as parameterized by Hr_h^{-3} and $I_{\perp}$.

It is interesting to note that although each solvent exhibits increased N_2O T_S as its viscosity increases, as predicted by the model, the more viscous isopropanol has generally shorter N_2O T_S than H_2O does. This surprising effect is duplicated in the measured translational diffusivity trends in the three solvents ($D = 1.9, 3.0$, and 5.2×10^{-9} m²/s in H_2O , isopropanol, and CS_2), highlighting the effect of microviscous characteristics on both diffusive processes and the inadequacy of the unmodified Stokes-Einstein model for interacting solvents. It is more accurate to regard T_S as increasing monotonically with D^{-1} , a trend which holds for all of the measurements described here. We therefore expect that measurement of the translational diffusion constants is the best predictor of which solute-solvent systems exhibit long singlet lifetimes when the relaxation is dominated by spin-rotation at low magnetic fields.

In conclusion, we have presented a model to describe relaxation of the singlet state of nitrous oxide dissolved in various solvents, using a classical model of rotation diffusion and correction factors describing the microviscous characteristics of the solvent which are determined using high-field T_1 measurements. We believe that the relaxation of the singlet state of N_2O in low field is dominated by the spin-rotation interaction. The model predicts that low-field lifetimes of the longitudinal polarization and the singlet state will increase at lower temperatures, primarily because the influence of the spin-rotation interaction lessens with increasing solvent viscosity. This trend is confirmed experimentally. We expect that application of the model to other molecular species will be sufficiently accurate to justify custom synthesis of dilabeled molecules, with future applications in long-lifetime biological imaging agents.

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