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Dynamic Response to Hydration in a Superabsorbing Polymer Studied by ^{13}C NOE and Spin–Lattice Relaxation Times[†]

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ABSTRACT: The magic angle sample spinning (MASS) technique has been used to study dynamic response to hydration in a superabsorbing polymer, hydrolyzed starch-*g*-poly(acrylonitrile). ^{13}C NOE and spin–lattice relaxation times have been measured at different hydration levels. The steady-state NOE strongly depends upon the degree of hydration and is found to be greater than 100% at a level of 8.0 g/g. Protonated carbons show a much greater enhancement (ca. 140%) than other carbons. Transient NOE behavior is consistent with the process of preferential hydration in this polymer. It is shown that MASS affords selective proton irradiation for the observation of NOE within a given polymer segment. These experiments additionally provide evidence for intersegment dipolar relaxation interactions. ^{13}C T_1 measurements show that at high hydrations the cooperative effects of ^{13}C–^{1}H cross-relaxation and proton spin-diffusion inhibition cause a nonexponential decay for the ^{13}C magnetization. The correlation time for hydration-induced polymer motions is estimated to be (0.2–0.5) × 10^{-9} s at 8.0 g/g. A comparison of steady-state and transient NOE data in D_2O and H_2O reveals an additional dipolar relaxation pathway between the polymer carbons and the hydration reservoir.

Introduction

The dynamical characterization of a macromolecule is often done through the measurement of ^{13}C spectral and relaxation parameters.[1] Of specific interest is the nuclear Overhauser enhancement (NOE),[2] wherein the ^{13}C signal intensity is enhanced upon continuous radio-frequency irradiation of protons, to which the ^{13}C is dynamically coupled. The enhancement is given by

$$\eta = (\gamma_H/\gamma_C)(W_2 - W_0)/(W_0 + W_1 + W_2) \qquad (1)$$

Here η is the NOE factor, γ_H and γ_C are the magnetogyric ratios of the ^{1}H and ^{13}C, and W_0, W_1, and W_2 are the zero, single, and double quantum transition rates, respectively. η is transient[3] with respect to the motional correlation time τ_c. It yields a maximum value of 1.988 when the extreme narrowing condition ($\omega\tau_c \ll 1$) is met. This is a feature which has been extensively exploited in liquid-state[4] NMR and also in solid-state NMR[5] where fast internal motions (for example, methyl rotations) are present.

The gel state of a macromolecule represents an interesting intermediate phase where lack of fluidity rules out the possibility of molecular tumbling. It has been shown that the primary dynamic response to diluent addition is a dramatic increase in the local segmental mobility, in both synthetic polymers[6] and biopolymers.[7] In gels, the mobility is often restricted and the associated NMR interactions are incompletely averaged. However, line-narrowing techniques such as magic angle sample spinning (MASS)[8] together with dipolar decoupling,[9] when desired, can be effectively employed to observe high-resolution ^{13}C NMR spectra in the gel state.[10] The measurement of ^{13}C spectral and relaxation parameters,[11] such as T_1, T_2, and NOE, at the individual carbon sites, therefore, become possible. This aids in a more complete dynamical characterization of the polymer in the gel state.

In this paper we have studied the dynamic response to hydration in a superabsorbing polymer, namely, hydrolyzed starch-*g*-poly(acrylonitrile) (HSPAN), by ^{13}C nuclear Overhauser enhancements and spin–lattice relaxation times. The superabsorbing property of this polymer arises from the strong ionic repulsions from the ionizable groups attached to the grafted pendant chains, the low degree of cross-linking, and the presence of various hydrophilic groups, such as carboxylate, amide, and hydroxyl, which act as hydration sites.[12] In the hydrated state, the polymer is essentially a hydrogel and therefore represents a semirigid macromolecular system. MASS alone is often sufficient as a line-narrowing technique since spectral broadening due to the heteronuclear ^{13}C–^{1}H dipolar interactions is averaged to a great extent. This obviates the need to employ dipolar decoupling. It therefore becomes experimentally convenient to use low-power radio-frequency irradiation for the continuous saturation of ^{1}H spins. The good ^{13}C signal resolution due to MASS enables one to inspect the chemically distinguishable carbon sites in the repeat unit of the polymer for individual Overhauser and relaxation behavior. Furthermore, it becomes possible to measure NOE under conditions of complete proton saturation or selective saturation of certain protons within a given polymeric segment, the latter being facilitated by the averaging of ^{1}H–^{1}H homonuclear dipolar interactions by MASS.

Experimental Section

The steady-state and transient NOE experiments were performed at ambient probe temperature (293–295 K) on a Bruker MSL-300 FT NMR spectrometer at a ^{13}C resonance frequency of 75.48 MHz. Proton irradiation was applied at 300.13 MHz either by strong dipolar decoupling at 40 kHz or by continuous broad-band (BB) decoupling using low radio-frequency power. Steady-state NOEs were measured directly by comparison of integrated intensities from gated and inverse-gated spectra recorded under identical spectrometer conditions. Steady-state NOE was built up by proton saturation for 6–10 s. For observing transient NOE, the ^{13}C magnetization was followed by simply varying the recycle delay of ^{13}C acquisition. Proton irradiation was employed only during data acquisition, and this was typically 20–30 ms. For spin–lattice relaxation time measurements of ^{13}C and ^{1}H, a π–τ–(π/2) inversion–recovery sequence was used, except for ^{13}C T_1 of dry polymer, where a cross-polarization inversion–recovery sequence was used.[13] The π/2 pulse length was typically 4.2 μs for ^{13}C and 8.0 μs for ^{1}H. For selective irradiation experiments, ratio-frequency power levels were carefully calibrated to ensure identical pulse behavior in different experiments

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where the radio frequency had to be routed through a directional coupler. MASS was usually kept at around 1.5–2.0 kHz.

The polymer was synthesized in our laboratory by using the standard graft copolymerization technique. The details of the synthesis are given elsewhere.[14] For NMR measurements the polymer samples were hydrated using 99.9% D_2O (obtained from Bhabha Atomic Research Centre, Bombay, India). Different hydration levels could be conveniently achieved by a controlled exposure to a relative humidity of 100% in a desiccator. Hydration levels were estimated gravimetrically and expressed in terms of grams of water absorbed per gram of dry polymer (g/g).

Theoretical Section

The general theory of NOE was originally developed by Solomon[15] and is fully described by Noggle and Schirmer.[2a] For the sake of completeness, the theory for the time development of ^{13}C magnetization, appropriate to our study, is sketched below. Although MASS is employed to improve spectral resolution, sample spinning effects[5b] are not considered explicitly here. This consideration could become important at very low levels of hydration (less than 0.8 g/g), not employed in the present study, where residual anisotropies in the ^{13}C spectra could be large.[16] When S (^{13}C) and I (^1H) spins are coupled with each other, the rate of change of magnetization of each spin system can be described by a set of coupled differential equations[15] written in the general form

$$\frac{dI}{dt} = -\frac{1}{T_1^{II}}(I - I_0) - \frac{1}{T_{IS}}(S - S_0) \qquad (2)$$

$$\frac{dS}{dt} = -\frac{1}{T_1^{SS}}(S - S_0) - \frac{1}{T_{SI}}(I - I_0) \qquad (3)$$

Here T_1^{II} is the spin–lattice relaxation time of I spins and includes not only I–I interactions but also I–S interactions. T_1^{SS} is the spin–lattice relaxation time of S spins and T_{IS} and T_{SI} are cross-relaxation times between I and S spins. It may be noted that T_{IS} and T_{SI} are in general not equal, and also it is difficult to distinguish these experimentally.[5b]

The solution of eq 2 is cast in the most general form

$$S_z(t) = S_0 + m_1 e^{-\lambda_1 t} + m_2 e^{-\lambda_2 t} \qquad (4)$$

with

$$\lambda_{1,2} = \frac{1}{2}\left[\frac{1}{T_1^{II}} + \frac{1}{T_1^{SS}}\right] \pm \left[\left[\frac{1}{T_1^{II}} - \frac{1}{T_1^{SS}}\right]^2 + 4\left[\frac{1}{T_{IS}}\right]^2\right]^{1/2} \qquad (5)$$

The coefficients m_1 and m_2 in eq 4 determine the actual time evolution of ^{13}C magnetization in a given experiment for either growth or decay in the presence or absence of proton irradiation. When the protons are continuously irradiated, by either low-power continuous broad-band irradiation or pulsed high-power dipolar decoupling, $\langle I_z \rangle = 0$. The ^{13}C magnetization enhances to a steady-state value according to

$$S_z(t) = S_0[(1 + \eta) + e^{-t/T_1^{SS}}] \qquad (6)$$

where η is the NOE given by

$$\eta = (\gamma_I/\gamma_S)(T_1^{SS}/T_{IS}) \qquad (7)$$

For the steady-state NOE to be built up, the proton irradiation must be maintained for a duration which is several times the longest ^{13}C T_1 as prescribed in eq 6.[17] η as given eq 7 is the same as that formulated in the liquid state and is independent of the number of interacting protons.

The transient behavior of ^{13}C magnetization is conveniently traced by varying the recycle delay of the inverse-gated decoupler experiment.[18] Since protons are saturated only during the fixed acquisition period and not during the variable recycle delay, $\langle I_z \rangle \neq 0$ always. Thus the growth of ^{13}C magnetization to equilibrium is determined not only by ^{13}C and ^1H T_1s but also by the ^{13}C–^1H cross-relaxation rate. Equations 2 and 3 have to be solved with the boundary conditions

$$\langle I_z(0) \rangle = 0; \quad \langle S_z(0) \rangle = S_z(t) = S_0(1 - e^{-AQT/T_1^{SS}})$$

Here AQT is the acquisition time. The coefficients in eq 4 are calculated as

$$m_1 = \frac{1}{\lambda_1 - \lambda_2}\left[\left[\frac{1}{T_1^{SS}} - \lambda_2\right][S_z(t) - S_0] - \frac{1}{T_{IS}} I_0\right] \qquad (8)$$

$$m_2 = S_z(t) - S_0 - m_1 \qquad (9)$$

with λ_1 and λ_2 are given by eq 5. Clearly, eqs 4 and 8 show that ^{13}C magnetization does not grow single exponentially but rather exhibits a transient Overhauser behavior in its approach to Boltzmann equilibrium. The magnetization growth depends not only on T_1^{SS} but also on T_{IS} and T_1^{II}. However, when $1/T_{IS} \ll 1/T_1^{SS}$, the ^{13}C magnetization builds up single exponentially without the NOE.

We consider the measurement of the ^{13}C spin–lattice relaxation time (T_1^{SS}) by the π–τ–(π/2) pulse sequence, without proton irradiation during the variable delay τ. The magnetization decay is not a single exponential now and is given by eq 4 together with

$$m_1 = \frac{2S_0}{\lambda_1 - \lambda_2}\left[\frac{1}{T_1^{SS}} - \lambda_2\right] \qquad (10)$$

$$m_2 = -(2S_0 + m_1) \qquad (11)$$

However, if $(1/T_1^{II} - 1/T_1^{SS})^2 \gg (1/T_{IS})^2$, the decay is approximated by a single exponential. Similarly, if proton spins are saturated during the variable delay τ, T_1^{SS} is a single exponential.[2]

Results and Discussion

(a) Steady-State NOE. We show in Figure 1 the ^{13}C MASS spectra of hydrolyzed starch-*g*-poly(acrylonitrile) at three representative hydration levels without (a–c) and with (d–f) proton presaturation. The nuclear Overhauser enhancements obtained are under steady-state conditions as we have employed both proton preirradiation and recycle delay for a duration that is several times the longest ^{13}C and ^1H T_1s. The Overhauser enhancements were determined directly by the ratio of the integrated peak areas in the gated and inverse-gated spectra; $\eta = (S - S_0)/S_0$, where S is the area of a peak obtained in the gated decoupling mode of measurement and S_0 is the area of the same peak in the inverse-gated decoupling mode of measurement.

We observe that the NOEs measured in the hydrated polymer, especially of the protonated aliphatic carbons, for which C–H dipolar relaxation would be the sole mechanism, span a range within the theoretical limits of 0.153 and 1.988. As observed earlier from cross-polarization and Bloch decay ^{13}C spectra, the hydration at the various hydrophilic sites in this polymer proceeds rather preferentially.[19] Since the hydration at the carboxylate and the amide sites is essentially favored when hydration is 0.8

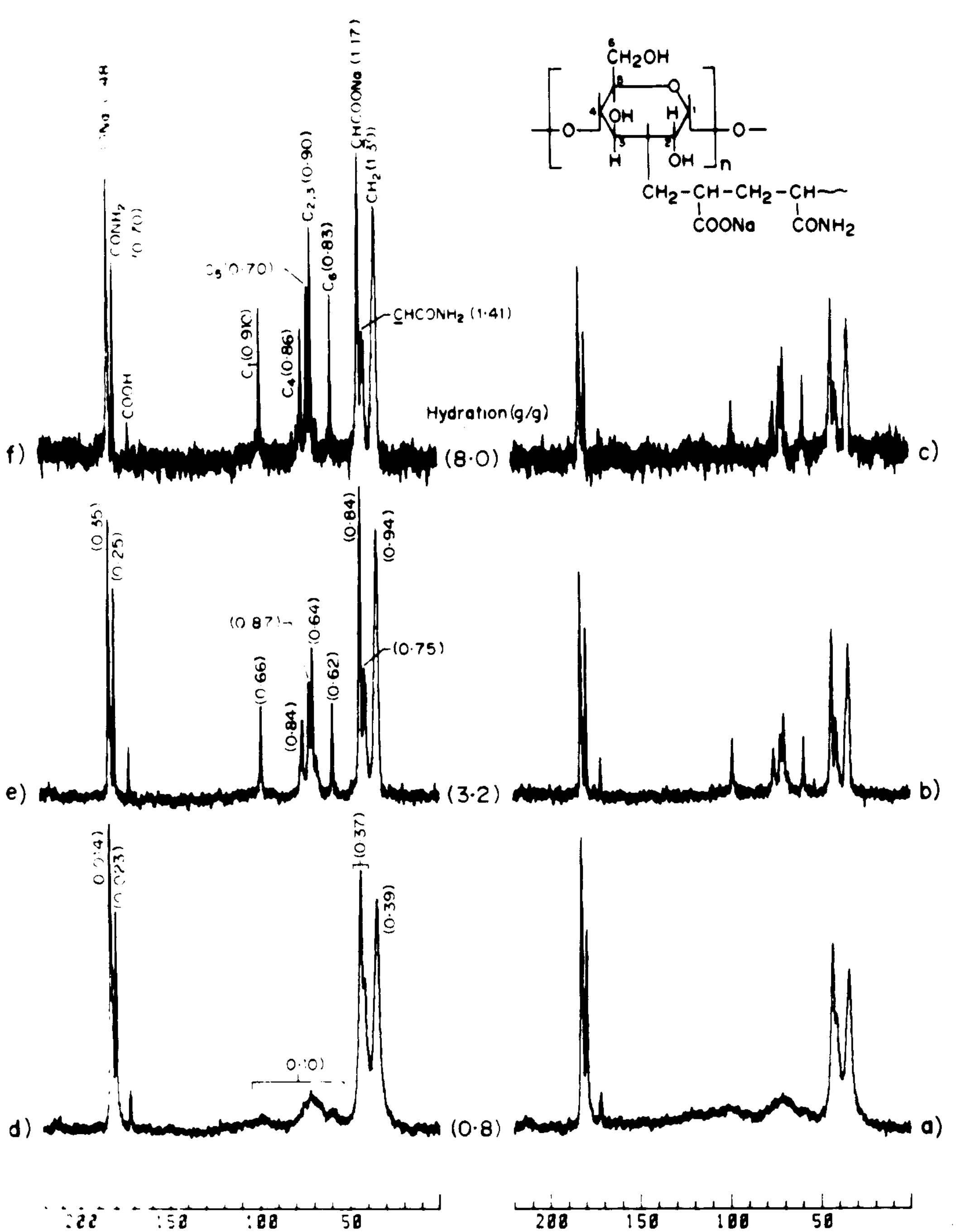

Figure 1. 75.5-MHz ^{13}C MASS (2.3 kHz) NMR spectra demonstrating the nuclear Overhauser effect in a superabsorbing polymer, hydrolyzed starch-*g*-poly(acrylonitrile), at different indicated hydration levels (grams of water per gram of polymer). Spectra without enhancement are shown in a–c, and the corresponding spectra with NOE are shown in d–f. Signal assignments and the corresponding NOEs (η), estimated from the integral area, are also shown. The number of transients were 800 for a, b, d, and e and 1000 for c and f. An exponential line broadening of 10 Hz was used before Fourier transformation, and all the spectra are plotted in the absolute intensity mode.

g/g, an increase in the motional flexibility of the pendant chains leads to ca. 40% NOE to be measured for the CH and CH_2 groups. The starch units, on the other hand, are unhydrated (or less hydrated), and their carbon NOEs are marginal (10%), lying close to the theoretical minimum. When the hydration level is increased to 3.2 g/g, the glucose units of the starch backbone are hydrated as well and the associated carbon NOEs build up, although they are far from maximal. When the hydration is further increased to 8.0 g/g, the enhancements exceed 100% and are found to be the greatest for CH and CH_2 of pendant chains.

Another interesting observation in this highly hydrated state is that when all the protons are saturated by broadband excitation, the maximum steady-state NOE that we have measured is independent of the number of protons (pendant CH, CH_2, 140%; glucose C_6, others, 90%). This is a feature which was observed earlier only in the liquid state.[4b] It ought to be noted, however, that the maximum steady-state NOE values of 140% are short of the theoretically predicted maximum of 198%. In this polymeric gel there is a lack of molecular tumbling. Therefore, it is the constrained segmental motion of the polymer chains that provides the dipolar relaxation pathways necessary for NOE buildup. Since Hartmann–Hahn cross-polarization is virtually eliminated and the residual scalar C–H couplings could be revealed in the ^{13}C spectra,[14] the segmental mobility must be faster than the strength of C–H dipolar interactions (ca. 25 kHz). With no fast internal motions present (e.g., methyl groups), the fulfillment of the extreme narrowing condition ($\omega\tau_c \ll 1$) is

Table I
^{13}C and ^{1}H Spin-Lattice Relaxation Times[a]

	relaxation time[b] (ms)						
	pendant			starch			
hydration (g/g)	COO$^-$/CONH$_2$	CH	CH$_2$	C$_1$	C$_4$	C$_{2,3,5}$	C$_6$
0.0 (dry)	35 × 10^3	30 × 10^3	30 × 10^3	30 × 10^3	30 × 10^3	30 × 10^3	30 × 10^3
0.8		200 (353)	175 (353)	—	—	—	—
1.6	1.5 × 10^3	160 (460)	108 (460)	—	—	—	—
3.2	3.0 × 10^3	202 (394)	143 (328)	220 (690)	234 (690)	258 (690)	120 (690)
8.0	1.4 × 10^3	243 (385)	136 (312)	289 (1000)	259 (811)	240 (565)	115 (1000)

[a] Values in parentheses denote the T_1 values for protons measured in the ^1H MASS spectrum. The absence of T_1 data in certain columns indicates that the signal resolution in the MASS spectra was not adequate to make independent measurements. [b] The T_1s were estimated by a linear least-squares fit of the data.

therefore unlikely to be met precisely for any of the carbon atoms. This is so even when the polymer is highly hydrated. This is validated by the value for τ_c calculated from ^{13}C T_1 data, as discussed in a later section. A distribution of motional correlation times would also contribute to a reduction in the maximum observable NOEs.[20] However, in view of an earlier observation in this polymeric gel, where cross-polarization data could be fitted essentially to a single correlation time model,[14] this is unlikely to be the dominant cause.

(b) Selective Irradiation. It was shown earlier by us that, in the hydrated state of this superabsorbing polymer, the static proton line is inhomogeneously broadened, and the application of the MASS technique causes spectral narrowing.[21] As a result, the proton resonances of the various functional groups within the monomeric unit could be resolved even at slow spinning speeds (1–2 kHz). The solvent-induced motions are very effective in averaging the various intragroup ^1H–^1H dipolar interactions. An important consequence is that the process of rapid spin diffusion is considerably inhibited, if not totally eliminated. The frequency isolation of coupled protons causes the proton relaxation of the pendant chains and the backbone starch units to proceed independently. This is indeed found to be so as indicated by independent ^1H T_1 measurements (Table I). The observed proton MASS spectral resolution suggests that selective saturation of certain proton populations would therefore be feasible. This will enable one to measure NOE selectively within a strongly dipolar coupled C–H spin network.

We show in Figure 2e the ^1H MASS spectrum of 8.0 g/g hydrated HSPAN and in parts b and c the enhancement due to selective irradiation of the aliphatic and starch protons, respectively, using a soft radio-frequency pulse.[22] The spectra taken without (Figure 2a) and with (Figure 2d) broad-band proton presaturation are also shown for comparison. Within each of the selective irradiation experiments, the enhancement is by and large confined to dipolar relaxation interaction within the irradiated network. It may be noted that NOE factors are somewhat smaller in the selective irradiation experiments (Figure 2b,c) than in the broad-band preirradiation scheme (Figure 2d). In this hydrated state, the polymer system can be thought of as a "three-spin" system. This is in complete analogy to an AMX spin system,[4b] where X represents the observed ^{13}C spins and A and M correspond to the two distinct MASS-resolved proton populations, namely, pendant and starch protons, respectively. In view of the importance of dipolar interaction between the observed carbon (X) and unirradiated protons (A or M), a three-spin mechanism would tend to reduce η. That such an effect is strongly operating in our system is depicted in Figure 2. Special attention is drawn to experiments for starch carbons (Figure 2), where a high degree of ^1H selective supression could be achieved (Figure 2c,g). Another important feature of these selective irradiation experiments is the observation that for most carbons the biselective NOE (i.e., when both groups are saturated) is larger than the sum of the selective NOEs, in accordance with the theory.[4d]

(c) Transient NOE. In addition we have carried out transient NOE experiments to investigate the time development of ^{13}C spin magnetization as a function of hydration. The results are shown in Figures 3 and 4, for representative carbons in the repeat unit at two hydration levels (1.6 and 8.0 g/g). In each case, the magnetization behavior can be satisfactorily fitted to eq 4, using eqs 5, 8, and 9. As noted earlier, in the recycle-dependent experiments that we have employed, the ^{13}C magnetization builds up under the collective influence of T_{IS}, T_1^{II}, and T_1^{SS} processes.

We note from Figure 3a,b that the transient behavior at the two hydration levels is similar for the CH$_2$ carbons of the pendant chain, whereas the same is not true for the carbons of the starch backbone. In the 1.6 g/g hydrated polymer, the magnetization buildup is rapid for pendent chain CH$_2$ (Figure 3a), which exhibits nonexponentiality and an overshoot of its equilibrium value. This is far less pronounced for the C$_{2,3,5}$ resonances of the starch unit (Figure 3c), in accordance with the preferential hydration of the associated sites. Upon further hydration (8.0 g/g), C$_{2,3,5}$ assumes a faster transient response (Figure 3d). An analysis of the data shows that the η values deduced are similar to those measured from steady-state NOE experiments.

A distinct feature of the transient NOE for the carboxylate/amide carbon (Figure 4a,b) is that the behavior is simply an exponential approach to Boltzmann equilibrium, dictated mainly by T_1^C at all hydration levels. As seen, the ^{13}C magnetization never overshoots its equilibrium value. In a previous section it is indicated that the ^{13}C magnetization behavior in the transient NOE experiment is a single exponential when $1/T_{C\text{-}H} \gg 1/T_1^C$. It is therefore clear that the smaller η values measured for these groups in the steady state are not due to improper waiting times in the gated/inverse-gated decoupling experiments.[17] These are rather due to the dominance of nondipolar relaxation mechanisms for the weakly dipolar-coupled carboxylate carbon, ^{13}C relaxation via anisotropic chemical shielding can be significant due to a large anisotropy at our 7.1-T field (6–9 kHz).[23]

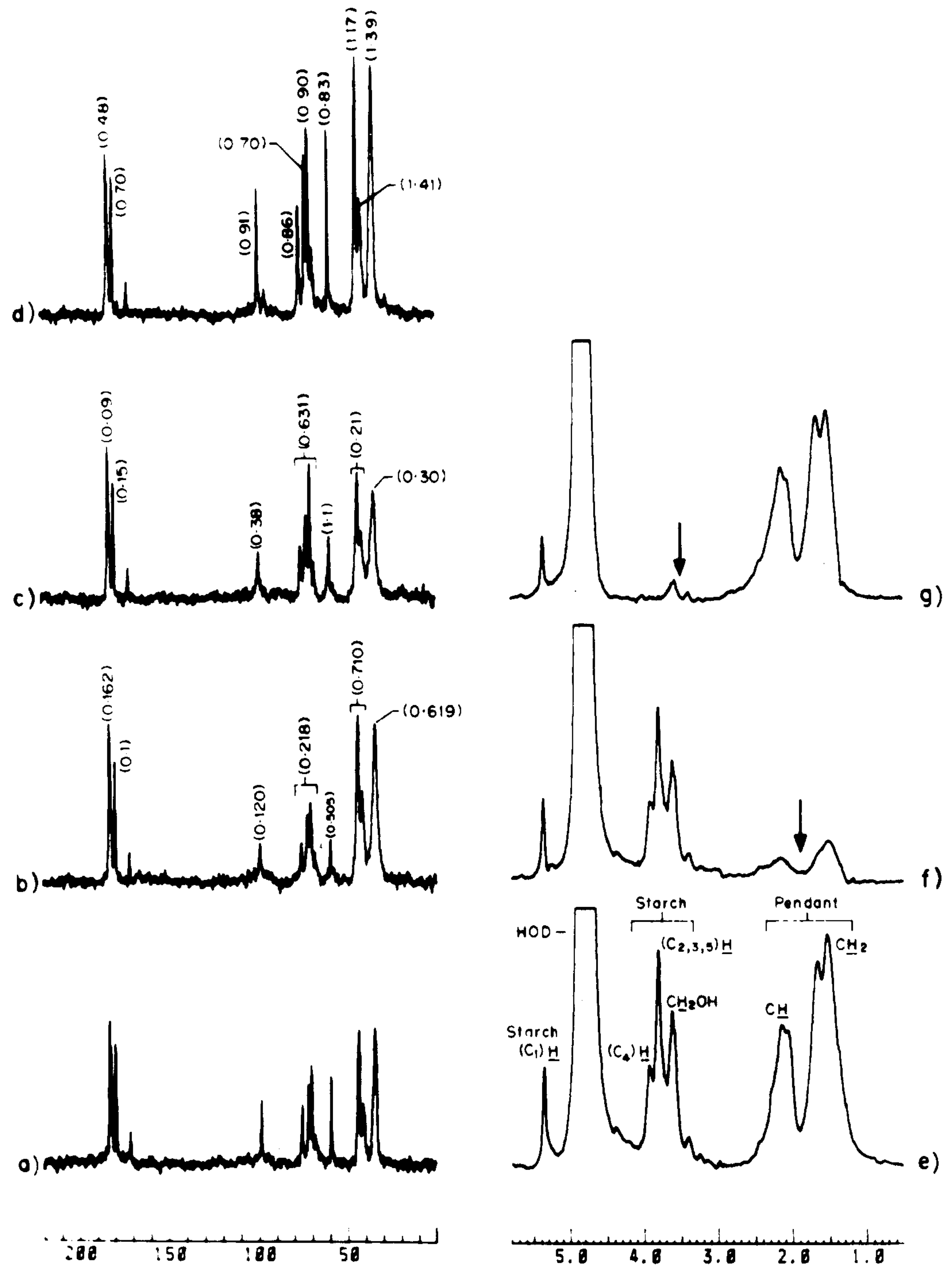

Figure 2. 75.5-MHz ^{13}C MASS NMR spectra of hydrolyzed starch-*g*-poly(acrylonitrile): (a) without proton preirradiation, (b) with selective aliphatic proton presaturation, (c) with starch proton presaturation, (d) complete proton presaturation using noise modulation. 300-MHz ^{1}H MASS NMR spectra: (e) without preirradiation, (f) presaturation of protons of pendant chain, (g) presaturation of starch protons except C_1H. The position of the radio-frequency carrier is indicated by the arrow.

(d) ^{13}C Spin–Lattice Relaxation. The carbon spin–lattice relaxation times, T_1^C, provide additional information about the hydration-induced dynamic response of the polymer in the vicinity of the ^{13}C Larmor frequency. We have carried out carbon T_1 measurements with proton saturation so that the decay can be fitted to a single exponential. The results are summarized in Table I. In the dry polymer, resolution of all the carbons in the repeat unit is not quite the same as in the hydrated samples. However, in the CP–MAS spectrum the carbon resonances of the pendant chains and starch units are distinctly resolved. In the dry state, polymer mobility is too slow to promote an efficient spin–lattice relaxation. The polymer system is essentially in the long correlation limit ($\omega\tau_c \gg 1$), and the carbon relaxation lies on the low-temperature side of the carbon T_1 minimum.

In contrast to the dry polymer, the T_1 values in the hydrated polymer are reduced by more than 2 orders of magnitude, lying in the range of 100–300 ms. A dominant feature of the relaxation, when hydrated with D_2O, is that water-induced changes in the carbon T_1 are associated with the changes in the local motions of the polymer itself.

An inspection of the T_1 data, especially for the protonated carbons, shows that the ratios of T_1^C (CH) to T^C (CH_2) are found to be 1.14, 1.41, 1.48, and 1.78, for hydration at the level of 0.8, 1.6, 3.22, and 8.0 g/g, respectively. Ideally, if the carbon is relaxed only by the directly bonded protons, the ratio should be 2.0. This is not found to be so. A similar discrepancy in the T_1 ratio was noticed earlier in bulk polymers.[24] This was thought

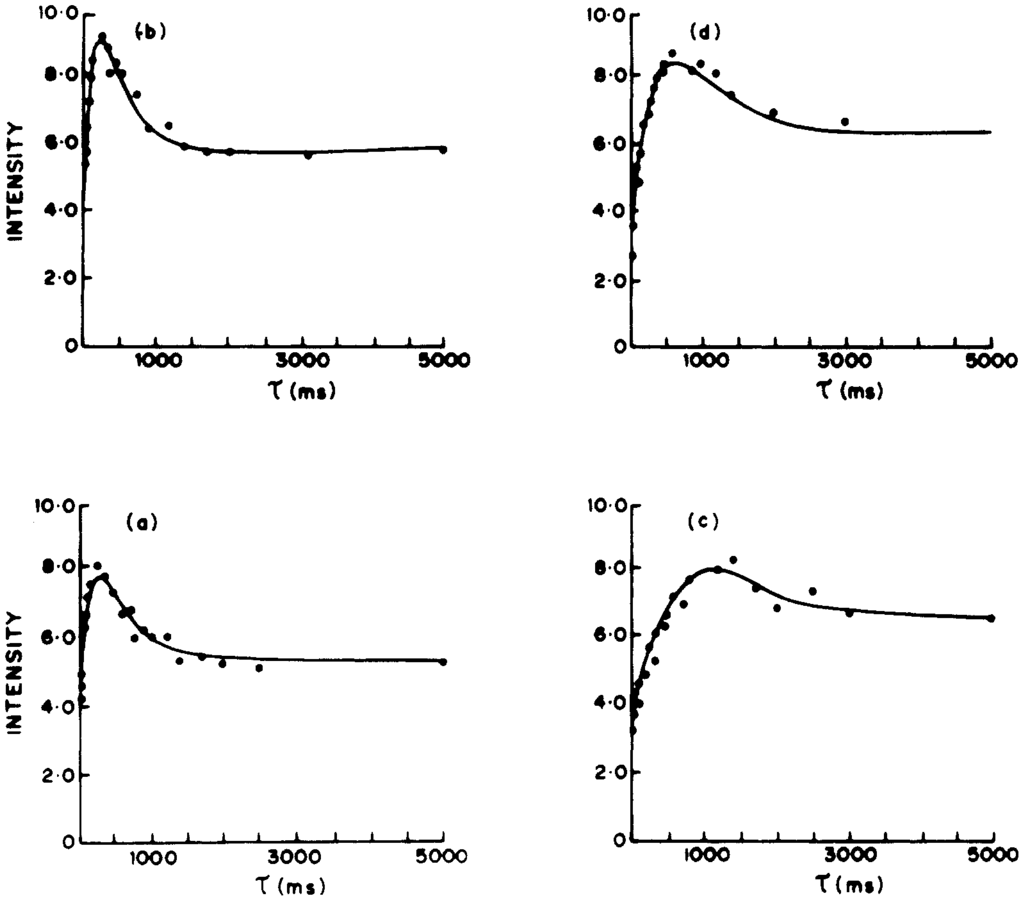

Figure 3. Time evolution of ^{13}C magnetization showing the transient NOE behavior at 1.6 g/g (bottom row) and 8.0 g/g (top row) hydrations. Experimental data (●) for protonated pendant [CH_2 (a,b)] and starch [$C_{2,3,5}$ (c,d)] carbons are shown. In each case the data are fitted to eq 4 as discussed in the text with relaxation parameters given in Table I and η shown in Figure 1.

to arise from the inequivalent reorientation of the C–H vector of CH and CH_2 groups.

In bulk polymers the local motions are thermally activated. In the polymeric gel, on the other hand, the plasticization action causes an enhancement in mobility. It has been found that the dynamic response to diluent addition is to average the various tensorial interactions (chemical shielding, dipolar).[6a,25] The nature and extent of averaging is very much dependent upon the type of diluent and the level of hydration. For hydrated HSPAN it was found that the static ^{13}C spectra exhibited residual powder pattern anisotropy at very low levels of hydration (<0.8 g/g).[14] Moreover, ^1H–^{13}C cross-polarization spectra could be obtained[19] in the matched Hartmann–Hahn experiment.[26] The ^{13}C resonances were, however, isotropically averaged at higher hydration levels (>1.8 g/g).[19] These spectral features are further supported by NOE data presented here. Considerations of nonequivalent motions could apply, at best, in our case only at hydrations 0.8 g/g and below.

We first consider that a given carbon (CH or CH_2) experiences both intra- and intergroup C–H dipolar interactions, modulated by the same chain motion. Interchain interaction between the pendant chain and the distant starch units can be neglected due to attenuation by longer internuclear distances. Taking C–H (CH) = 1.08 Å, C–H (CH_2) = 1.09 Å, and C–C = 1.54 Å[27] and tetrahedral bond angles and considering the various C–H dipolar interactions from both bonded and nonbonded protons within the pendant unit, we calculate the ratio $T_1(CH)/T_1(CH_2)$ = 1.811. The noticeable disagreement at 0.8 g/g hydration is likely to be caused by the nonequivalent motions of the CH and CH_2 groups. This is in line with the spectral observations presented above. We find that as the hydration level is increased the experimental T_1 ratio tends to approach the value computed above. In fact, the two are in very good agreement at 8.0 g/g hydration. Since at this hydration level the pendant chains undergo fast isotropic motions, considerations of inequivalent CH and CH_2 reorientations do not apply. The T_1 ratio must therefore be determined by dipolar relaxation interactions with bonded and nonbonded protons.

The T_1 measurements for the starch unit at two representative levels, 3.2 and 8.0 g/g, are also summarized in Table I. In the case of starch carbons, we could measure the spin–lattice relaxation time distinctly at each carbon site only when hydrated to 8.0 g/g (Table I). At 3.2 g/g hydration level we could measure the T_1 for the starch unit as a whole and not at single sites. Below this level poor signal resolution precluded any T_1 measurement being made.

In the dry polymer the relaxation time is long (30 s) and is similar to the T_1 of pendant carbons. Clearly, the polymer motions are too slow to provide an efficient relaxation mechanism at the applied Larmor frequency. There is a dramatic decrease in T_1 when the polymer is hydrated. We note that, at levels 3.2 and 8.0 g/g, the starch units are hydrated as well.[19] As observed earlier,[14] the hydration-induced local mobility provides an efficient averaging of anisotropic C–H dipolar and ^{13}C chemical shielding interactions in the static ^{13}C spectra. The mobility at the starch sites must therefore be sufficiently fast compared to the frequency strength of the interactions being averaged. This is also reflected in the steady-state NOE measurements, as discussed earlier. On the basis of

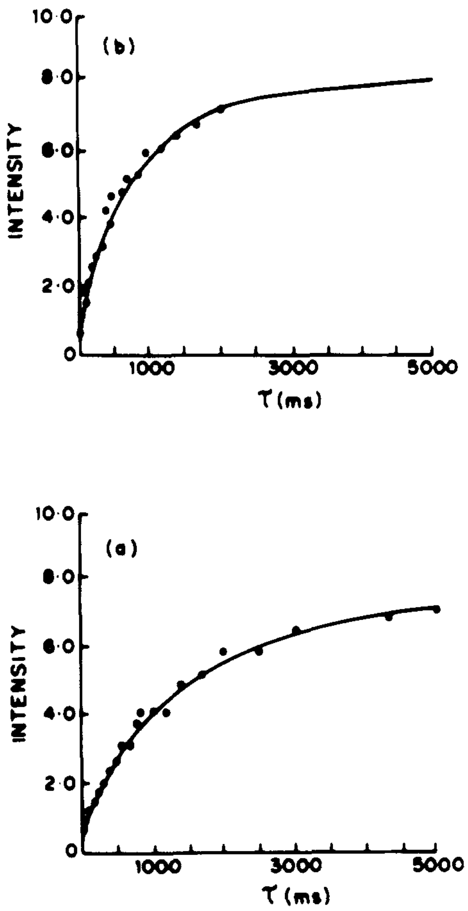

Figure 4. Time evolution of ^{13}C magnetization at 1.6 g/g (bottom) and 8.0 g/g (top) hydrations for the unprotonated carbons. Experimental points (●) are fitted to eq 4 as discussed in the text with relaxation parameters given in Table I and η shown in Figure 1.

the spectral and NOE data, one might expect that, in the highly hydrated state (8.0 g/g), the correlation times for the pendant and starch motions must be similar.

Proton relaxation times have been used for the dynamical characterization of water in this superabsorbing polymer.[28] It was found that water exists in both bound and free environments, especially at the level 8.0 g/g. When free water is present in the system, one must additionally consider the dynamic exchange at the hydroxyl sites, mediated by water. A correct description for the starch carbons would involve both these motional processes. However, in our case we could discount the latter on the grounds that the medium is essentially 99% deuterated and that the dipolar relaxation would be attenuated greatly by the low magnetogyric ratio of ^2H and a longer distance to the interacting carbon. While the T_1 values are determined by the enhanced mobility of the starch backbone due to hydration, they, however, do not lead us to estimate the relative importance of these modes of local mobility.

We also note that ^{13}C T_1s are seen to be considerably shorter (see Table I) than the proton T_1s, measured at the same sites. Long proton T_1s are normally associated with a spin-diffusion mechanism.[29] In our case this was found to be unimportant. The differences can therefore be rationalized by considering that the inverse sixth power of distances enters the relaxation interaction. For protons in the CH_2 group, for example, the H–H distance (1.27 Å) is 15% longer than the C–H distance (1.1 Å). The ratio of the inverse sixth power of the H–H and C–H distance works out to 2.37, in fair agreement with the experimentally measured T_1^H/T_1^C of 2.29 for the CH_2 group in the 8.0 g/g hydrated polymer. These data support the view that the same chain motion dominates both the ^1H and ^{13}C spin relaxation. Finally, we observe that for the carboxylate/amide carbons the ^{13}C T_1s are longer than that of the protonated carbons. This is directly attributable to the attenuation of the C–H dipolar couplings with distant protons and to the effects of deuterium exchange (amide groups).

In order to estimate the motional correlation time τ_c from the carbon T_1 values, we first consider the effects of hydration on the static ^{13}C line shapes. It was noted earlier that when all sites are hydrated, no apparent features of powder pattern anisotropy due to chemical shielding or the proton–carbon dipolar coupling could be discerned from the static ^{13}C cross-polarization and Bloch decay spectra.[19] We therefore consider a simple motional model in which isotropic random reorientations of the C–H vectors occur. ^{13}C T_1 and the NOE factor $(1 + \eta)$ are given by[3,30]

$$\frac{1}{T_1} = \frac{1}{10}\gamma_H^2\gamma_C^2\hbar^2\sum r_i^{-6}\left[\frac{\tau_c}{1 + (\omega_H - \omega_C)^2\tau_c^2} + \frac{3\tau_c}{1 + \omega_C^2\tau_c^2} + \frac{6\tau_c}{1 + (\omega_H + \omega_C)^2\tau_c^2}\right] \quad (12)$$

$$\mathrm{NOE} = 1 + \eta = 1 + \frac{\gamma_H}{\gamma_C}\left[\frac{6\tau_c}{1 + (\omega_H + \omega_C)^2\tau_c^2} - \frac{\tau_c}{1 + (\omega_H - \omega_C)^2\tau_c^2}\right] \Big/ \left[\frac{\tau_c}{1 + (\omega_H - \omega_C)^2\tau_c^2} + \frac{3\tau_c}{1 + \omega_C^2\tau_c^2} + \frac{6\tau_c}{1 + (\omega_H + \omega_C)^2\tau_c^2}\right] \quad (13)$$

Substituting in these equations the carbon T_1 of pendant CH_2 (Table I) and assuming that the carbon is relaxed by both directly bonded and adjacent protons, we obtain for the correlation time a value of 0.2×10^{-9} s from T_1 (eq 12) and 0.46×10^{-9} s from NOE (eq 13). This estimate for τ_c places the ^{13}C relaxation in the vicinity of the T_1 minimum ($\omega\tau_c \sim 1$), showing that the extreme narrowing condition is not reached. It is therefore not surprising that the steady-state NOE values do not quite reach the theoretical maximum of 198% even at this 8.0 g/g hydration level.

We have also investigated the effect of proton presaturation on the carbon T_1 decay. The experimentally obtained plot of the decay of the ^{13}C magnetization of the aliphatic carbon of 0.8 g/g hydrated HSPAN is shown in Figure 5a and further compared with a more hydrated sample (8.0 g/g; Figure 5b). The magnetization decay in the presence and absence of proton presaturation is also shown. This observation is in accord with eqs 4 and 5 and eqs 10 and 11. It brings out the importance of cross relaxation in the observed ^{13}C spin–lattice decay, especially at higher hydrations. The observed nonexponentiallity is the greatest for the 8.0 g/g hydrated sample, where cross relaxation is most efficient (large NOE) and operates on an equal footing with ^{13}C T_1. The carbon signal decay for the 0.8 g/g hydrated polymer, on the other hand, is very nearly exponential due to an inefficient cross-relaxation mechanism (smaller NOE). Such nonexponential decays are observed when protons are few and isolated.[5b] In this hydrated polymer it is notable that the spin-diffusion inhibition renders protons to behave isolatedly, rather than collectively, thereby leading to a revelation of the nonexponentiality.

(e) Comparison of NOE in H_2O and D_2O. All the NOE measurements discussed so far were carried out on

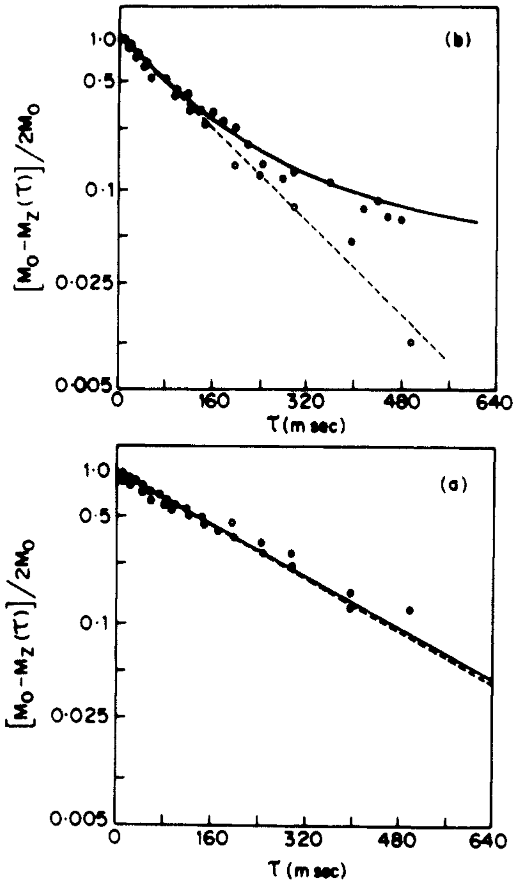

Figure 5. Decay of ^{13}C magnetization in spin-lattice relaxation measurements for the pendant CH_2 carbon at 0.8 g/g (a) and 8.0 g/g (b). Experimental data without (●) and with (○) a proton saturating pulse during the variable τ delay are shown. The solid line is calculated using eqs 4, 10, and 11 with the following values for the relaxation parameters: (a) T_1^C = 200 ms, T_1^H = 340 ms, η = 0.4; (b) T_1^C = 115 ms, T_1^H = 380 ms, η = 1.35. The dashed line shows the exponential decay due to a T_1^{SS} process only using T_1^C values above.

samples hydrated with D_2O. This way the contributions from interdipolar relaxation between the polymer carbons and water to the ^{13}C NOE and T_1 could be neglected. Of considerable fundamental interest is the dynamic coupling, if any, between the water of hydration and the macromolecular system. This has been explored to a limited extent by steady-state and transient NOE measurements, the results of which are shown in Figure 6. We observe that the hydration by H_2O causes an overall increase in the steady-state NOE values for most of the carbons, the maximum NOE enhancement of ca. 60% being measured for the pendant CH_2 (Figure 6b). A dipolar relaxation interaction between polymer carbons and the hydration reservoir (H_2O) would tend to an increase in the steady-state NOE, since the motional correlation time for water molecules would invariably be shorter than those of the polymer segments. This is indeed noticed in the steady-state NOE values that we have measured for most of the carbons in the repeat unit. The importance of the dynamics of water in the ^{1}H and ^{13}C spin relaxation of synthetic[25] and biopolymers[31] has been recognized.

We also observe an increase in the Overhauser enhancement for the COO^- and $CONH_2$ group carbons upon going from D_2O to H_2O. However, the changes in NOE enhancements are small (8–10%) as compared to those of protonated carbons such as CH_2 groups (50–60%). This is in accord with the dominance of the dipolar relaxation mechanism for the latter and the influence of the nondipolar relaxation mechanism for the former.

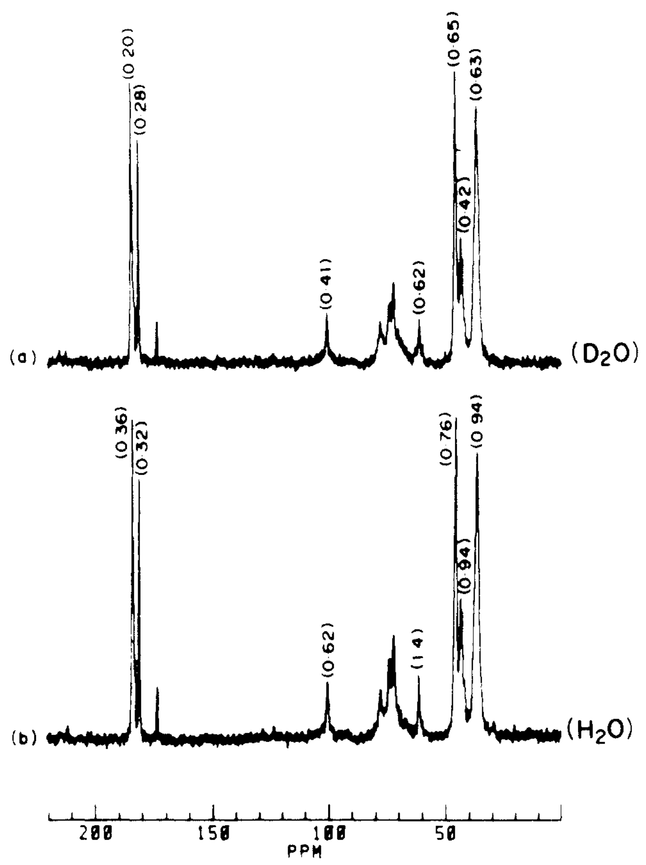

Figure 6. Steady-state NOE observed for 1.6 g/g hydrated polymer in D_2O (a) and H_2O (b). The Overhauser enhancements (η) are shown on top of each of the ^{13}C resonances in the monomeric unit.

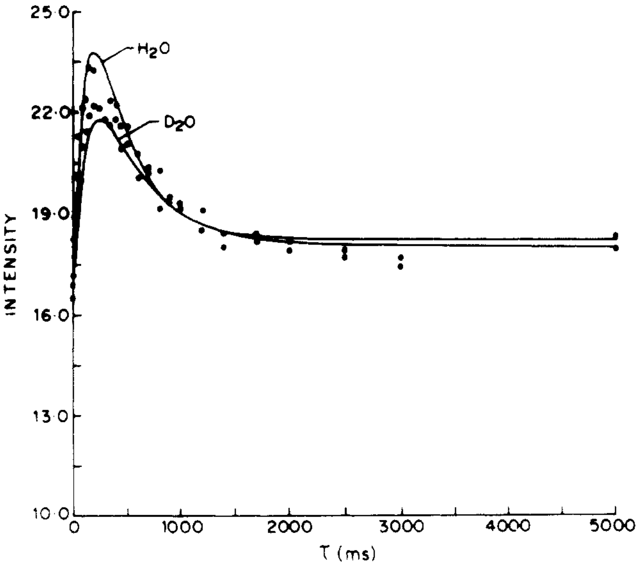

Figure 7. Transient NOE response for 1.6 g/g hydrated polymer in D_2O (●) and H_2O (○). Solid lines were calculated using eqs 4, 8, and 9 with the following values for the relaxation parameters. D_2O: T_1^C = 108 ms, T_1^H = 460 ms, η = 0.35. H_2O: T_1^C = 100 ms, T_1^H = 320 ms, η = 0.55.

In Figure 7, we compare the transient NOE behavior for the pendant CH_2 carbon in the polymer hydrated with D_2O and H_2O. These curves distinctly separate the magnetization evolution in each case. Analysis of the data shows that while ^{13}C T_1s are unchanged between H_2O and D_2O, both T_1^H and η values are significantly different. Equality of ^{13}C T_1s is not surprising because ^{13}C relaxation essentially proceeds through intradipolar interactions within a given group, including its immediate neighbor as discussed earlier. Interaction with water of hydration would be weaker due to a longer internuclear distance. On the other hand, NOE buildup does not depend on the number of interacting protons or its proximity, when the entire proton system is saturated and steady-state conditions are established. This is evident in the Overhauser enhancement for the aliphatic carbons (ca. 60%). We therefore observe that the dynamic coupling between the polymer and the hydration (H_2O) reservoir is far more effective in determining the observed NOEs than by the hydration-induced enhancement in local mobility alone.

Conclusions

We have found that in a superabsorbing polymer, such as hydrolyzed starch-*g*-poly(acrylonitrile), steady-state and transient NOE and spin-lattice relaxation time measurements bring out the dynamic response to hydration. The NOE factor is seen to be sensitive to the hydration level. At low hydrations the NOE behavior is seen to be consistent with the process of preferential hydration in this polymer. It is also found that the NOE does not reach the theoretical maximum even when hydrated to 8.0 g/g. Selective ^{1}H irradiation experiments with MASS bring out the importance of interchain interactions between the pendant groups and starch units. The carbon spin-lattice relaxation time measurements also demonstrate the importance of dipolar relaxation coupling to nonbonded protons. Evidence for dipolar relaxation of polymer carbons by the water protons is provided by NOE experients in H_2O. Since cross-polarization methods become ineffective, the observed Overhauser enhancements offer considerable scope for ^{13}C spectral sensitivity under MASS.

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