Coherence Transfer in Dipolar-Coupled Homonuclear Spin Systems in Solids Rotating at the Magic Angle

O. WEINTRAUB,* S. VEGA,* CH. HOELGER,† AND H. H. LIMBACH†

*Chemical Physics Department, The Weizmann Institute of Science, 76100 Rehovot, Israel; and † Institut für Organische Chemie,
Freie Universität Berlin, D-14195 Berlin, Federal Republic of Germany

Two routes for the exploitation of the t-SEDRA pulse scheme, which induces coherence transfer in dipolar-coupled homonuclear spin systems in rotating samples, are demonstrated and discussed. This sequence is utilized to deduce intramolecular connectivities by creating an initial coherence of one spin only, applying the t-SEDRA sequence, and monitoring the signal enhancement of the coupled spin. Probing the signal amplitude variations of the two spins and comparing them to simulations can also yield molecular distances. Using 2D spectroscopy, t-SEDRA can also be utilized to establish spin correlations. In this case, the t-SEDRA sequence is applied during the mixing time of a 2D dipolar-correlation experiment. These two approaches are demonstrated by performing ^{15}N CPMAS NMR experiments on a ^{15}N-doubly labeled sample of 3(5)-methyl-5(3)-phenylpyrazole. © 1994 Academic Press, Inc.

INTRODUCTION

During the past few years, various techniques have been developed for detecting the dipolar interaction in rotating samples in solid-state NMR. The detection of the dipolar interaction can provide valuable information on intra- and intermolecular bond distances as well as on the connectivity of atoms in molecules. It can also contribute to the elucidation of molecular structure in terms of bond angles. In the solid state, the utilization of cross-polarization magic-angle spinning NMR (CPMAS) (1, 2) enables us to benefit from the increased sensitivity due to CP, as well as from the spectral resolution and sensitivity enhancement due to sample rotation. Sample spinning results in partial or complete averaging of the dipolar interactions (3). To detect the effect of the dipolar interaction on the MAS spectra, one can analyze the intensities of the center- and sidebands, using the method developed by Herzfeld and Berger (4). In most cases, however, these sidebands are mainly due to the chemical-shift anisotropy (CSA) interaction. To allow for the detection of the dipolar interactions, dedicated pulse schemes have been developed. They can be divided into two main types: experiments that enable the detection of heteronuclear dipolar interaction (5–8) and methods that deal with homonuclear

interactions (9–14). In this publication we will restrict ourselves to homonuclear spin systems.

One of the successful MAS NMR methods for measuring homonuclear dipolar interactions between spins, is based on the rotational-resonance (R^2) condition (9, 10). In this technique, the spinning speed is adjusted to be equal to an integer multiple of the difference between the isotropic chemical shifts of the two interacting spins. This technique is highly selective, due to the R^2 condition, and only one preselected pair of nuclei can be monitored at one time. Other techniques rely on the interference of RF pulses with dipolar averaging, caused by the sample spinning. One of these methods is simple π-pulse excitation for the dephasing of rotational-echo amplitudes (SEDRA) (13, 14). In these experiments one π pulse is applied in the middle of each rotor period and after every two rotor periods, MAS signals can be acquired. The two pulses in a basic SEDRA unit result in the averaging of the chemical shift interactions and at the same time in the decay of the rotational-echo amplitudes of powder samples, due to the RF-driven dipolar recoupling. By monitoring the dephased signal relative to a nondephased signal, produced by the transverse-echo SEDRA (t-SEDRA) pulse sequence (14), one can obtain values of dipolar interactions. In this technique, a range of spinning frequencies can be chosen, which allows us to do a set of complementary experiments. The analogous broadband dipolar recoupling (BDR) experiment, monitoring longitudinal magnetization exchange, was performed by Sodickson et al. (15).

Another method for the detection of dipolar interactions in homonuclear spin pairs was introduced by Tycko et al. (16). Their DRAMA pulse sequence involves the application of two $\pi/2$ pulses per rotor cycle and results again in a noncomplete averaging of the dipolar interaction. Very recently Gullion et al. combined spin nutation and magic-angle spinning in order to introduce signal decays due to dipolar interactions (17).

Two-dimensional methods for establishing dipolar correlations between homonuclear spins in spinning samples were also employed. Bennett et al. applied their RF-driven

recoupling (RFDR) experiment (*18*) on fully ^{13}C-enriched alanine to generate a 2D spectrum of the dipolar-coupled spins. Other examples of 2D experiments that monitored dipolar and *J* couplings in homonuclear spin systems were also reported (*19, 20*).

In this article, experiments exploiting the t-SEDRA sequence will be reported. This pulse sequence causes coherence transfer and can be utilized in both one- and two-dimensional MAS experiments. Applications of this sequence will be shown and some comparative remarks will be made about the coherence-transfer techniques, SEDRA and t-SEDRA, and the magnetization-transfer techniques, BDR and RFDR (*15, 18*).

THE t-SEDRA HAMILTONIAN

To review the evolution of a homonuclear spin pair during a t-SEDRA experiment, we first present its pulse sequence in Fig. 1a. The basic pulse cycle is composed of π and $\pi/2$ pulses, extending over eight rotor periods. The π pulses, located in the middle of each rotor period, are responsible for the RF-induced dipolar dephasing of the MAS signal, whereas the $\pi/2$ pulse, at the fourth rotor-echo position, acts as a refocusing pulse for the induced dipolar evaluation. This can be shown by considering the effective Hamiltonians of the SEDRA and the t-SEDRA experiments. The effective Hamiltonian, describing the stroboscopically detected spin signal during a SEDRA experiment, has the form

$$\mathcal{H}_{\text{SEDRA}} = \omega_{\text{SEDRA}}\{2I_{x1}I_{x2} + 2I_{y1}I_{y2}\} + JI_{z1}I_{z2}. \quad [1]$$

This time-independent Hamiltonian was derived using both Floquet theory (*21*) and average-Hamiltonian theory (AHT) (*18*). Following the Floquet approach, the effective frequency coefficient equals

$$\omega_{\text{SEDRA}} = - \sum_{k,m=-\infty}^{\infty} d_{-k}^{22*} \omega_m^D d_{-m-k}^{33}, \quad [2]$$

where ω_m^D are the Fourier components of the dipolar term in the MAS Hamiltonian of the spin pair. This term can be written as

$$\mathcal{H}^D(t) = \sum_{n=-2}^{2} 2\omega_n^D \exp\{in\omega_R t\}[3I_{z1}I_{z2} - \mathbf{I}_1 \cdot \mathbf{I}_2] \quad [3]$$

with

$$\omega_{12} = \frac{\mu_0}{4\pi} \frac{\gamma_1 \gamma_2}{r_{12}^3}$$

$$\omega_{\pm 1}^D = \frac{\sqrt{2}}{16} \omega_{12} \sin \theta_{12} \cos \theta_{12} e^{\pm i\phi_{12}}$$

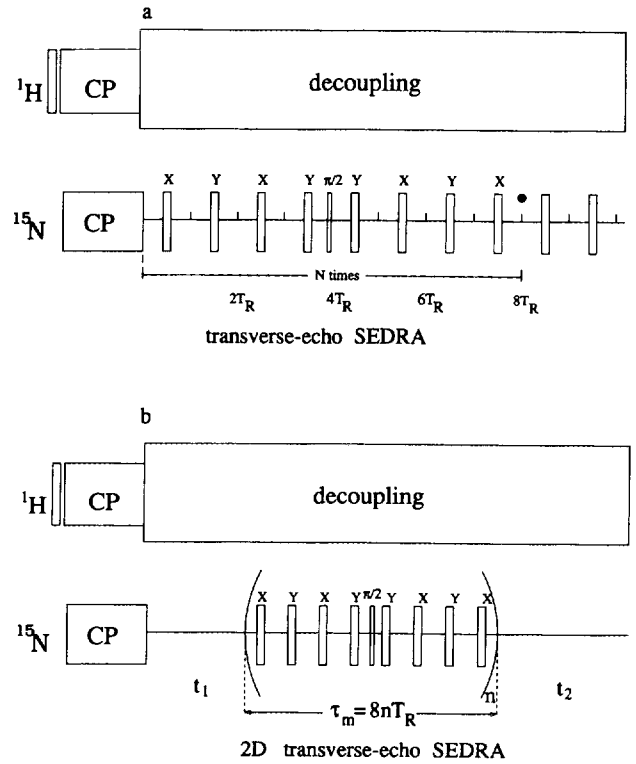$$\omega_{\pm 2}^D = \frac{1}{16} \omega_{12} \sin^2 \theta_{12} e^{\pm 2i\phi_{12}}, \quad [4]$$

FIG. 1. The basic t-SEDRA sequence is shown in (a). It is composed of 8 rotor cycles; however, to make it cyclic in the sense of AHT, 16 rotor cycles must be used with opposite phases for the two $\pi/2$ pulses. In (b) the application of t-SEDRA in the mixing time of a 2D experiment is shown. The phase of the $\pi/2$ pulse is x for t_x-SEDRA and y for t_y-SEDRA; the reason for this alternation is explained in the text.

where (θ_{12}, ϕ_{12}) are the polar angles of the dipolar vector in the frame of the rotor (*21*). The d_n^{pp} coefficients are the elements of the diagonalization matrix of the CSA-Floquet matrix. They depend on the difference between the isotropic chemical-shift values of the spins and their CSA parameters. Expressions for all the elements can be found in Ref. (*21*). If the sequence of Fig. 1a is applied twice, with the phase of the additional $\pi/2$ pulse in the opposite direction to the phase of the first $\pi/2$ pulse, the t-SEDRA experiment becomes cyclic in the sense of AHT (*22*). Using AHT, the effective Hamiltonian for the t-SEDRA experiment has the form (*14*)

$$\mathcal{H}_{\text{t-SEDRA}} = \frac{1}{2}\omega_{\text{SEDRA}}\{2I_{x1}I_{x2} + 2\mathbf{I}_1 \cdot \mathbf{I}_2\} + J\mathbf{I}_1 \cdot \mathbf{I}_2. \quad [5]$$

This is the correct form when the $\pi/2$ pulses are applied in the $+x$ and $-x$ directions.

This Hamiltonian commutes with the sum $[I_{x1} + I_{x2}]$ of the two x components of the angular momenta of the coupled spins. Hence, if the initial coherence, immediately after an excitation pulse or cross polarization, points in the x direction, i.e., the initial density matrix equals $\rho(0) = [I_{x1} + I_{x2}]$,

then, at the end of a cyclic t-SEDRA sequence, the total transverse magnetization creates an echo, $\rho(16mT_R) = [I_{x1} + I_{x2}]$. The t-SEDRA sequence does not create an echo in the regular sense, since the individual spins are not refocused. Although the AHT consideration demands a pulse cycle of 16 rotor periods, it is experimentally feasible to measure the signal every 8 rotor periods. This t-SEDRA signal is termed $S_0(8mT_R)$ and is proportional to $Tr\{\rho(8mT_R)(I_1^- + I_2^-)\}$. In a previous paper (*14*) it was shown how this quantity, together with a signal S_{SEDRA} ($8mT_R$) from the SEDRA sequence, can be used to measure dipolar interaction strengths, and how intramolecular distances can be deduced. By measuring the quantity $\Delta S/S_0$,

$$\frac{\Delta S}{S_0} = \frac{S_0(8mT_R) - S_{SEDRA}(8mT_R)}{S_0(8mT_R)}, \qquad [6]$$

and comparing it to simulated results, one can obtain the magnitude of the dipolar coupling between the spins. In this way the T_2 relaxation effects are removed from the experimental data. For a single crystallite in the powder sample this ratio will be proportional to $\{1 - \cos(8m\omega_{SEDRA}T_R)\}$. This method was demonstrated by ^{15}N MAS experiments on ^{15}N-enriched 3(5)-methyl-5(3)-phenylpyrazole (PMP) (*14*).

COHERENCE TRANSFER WITH t-SEDRA

The t-SEDRA pulse sequence induces coherence transfer between the homonuclear coupled spins. One-dimensional and 2D experiments can be constructed to establish spin correlations and to deduce dipolar-coupling strengths between low-abundance spins. One could use a singly or multilabeled molecule, selectively excite one of the labeled nuclei, apply the t-SEDRA sequence, and look for the signals of the coupled spins. A crucial step in such an experiment is the efficiency and the accuracy of the selective excitation. For example, this can be achieved by applying a selective DANTE excitation after the cross-polarization (*23*) or by taking advantage of the differences between yields of the CP processes for protonated and nonprotonated nuclei. In the first case the nonexcited nuclei retain their enhanced z magnetization, while in the second case their polarization can be strongly reduced. Although from the point of view of an ideal t-SEDRA experiment the two cases are equivalent, it is desirable to minimize the polarization of the nonexcited spins in order to avoid the excitation of their coherences, due to imperfections in the pulses composing the t-SEDRA sequence. Of course, this 1D type of experiment can be extended easily to 2D correlation experiments. However, this requires high isotopic labeling and can become very time-consuming in the case of MAS NMR. In this publication, both approaches will be discussed and demonstrated via ^{15}N CPMAS experiments on a doubly ^{15}N-labeled PMP sample.

Our 1D experiments start with 100 μs cross polarization, followed by a set of t-SEDRA sequences and the detection of the FID signal. The short CP time was chosen to excite the protonated nitrogens almost selectively. The intensity of the nonprotonated nitrogen centerband was about 6% of the intensity of the centerband of the protonated nitrogen, measured immediately after cross polarization. A set of experiments with an increasing number of t-SEDRA sequences, with lengths equal to eight rotor periods, was performed and the results are shown in Fig. 2. The strong line in these spectra decreases gradually, whereas the weak line maintains about the same intensity. The longest t-SEDRA irradiation period was 22.4 ms. The line-intensity changes are due to a combination of the coherence-transfer mechanism and the relaxation decay. In Fig. 3, the data are presented in a convenient form and compared with computer simulations. For the analysis, we recorded the intensity of each line and divided it by the sum of the two lines. This removes the effect of relaxation and allows a comparison with simulations. The experimental results are given by triangles and the simulated data by the two lines. The dotted lines correspond to a dipolar interaction of 420 Hz between the two coupled nitrogens and the dashed line corresponds to a dipolar coupling of 470 Hz. In both cases the simulated results were calculated, taking into account the finite lengths of the pulses, with intensities of 62.5 kHz, and all CSA parameters of the nitrogens in PMP (*24, 25*).

Both experimental and calculated results agree with our previously derived dipolar interaction (450 ± 50 Hz) (*14*). Thus, this experimental approach is suitable for quantitative determinations of dipolar-coupling values. However, the main purpose of such an experiment is to establish correlations between homonuclear coupled spins. This spin correlation can also be achieved by exploiting 2D spectroscopy, as will be shown below.

MAGNETIZATION-EXCHANGE EXPERIMENTS

Sodickson et al. demonstrated the application of the SEDRA sequence in their broadband dipolar-recoupling (BDR) experiments on homonuclear dipole-coupled spin pairs (*15*). They described the influence of the sequence in terms of motion of the magnetization vectors. In this experiment, the spin polarization of one of the spins was inverted selectively and the quantity $\langle I_{z1} - I_{z2}\rangle$, proportional to the difference between the inverted and noninverted z magnetization, was monitored. The signal $S^z(8mT_R)$, proportional to this difference, can be measured as a function of the number of SEDRA cycles. In this magnetization-exchange experiment, it would have been desirable to perform a $\Delta S/S_0$ measurement; however, no echo-experiment for this has yet been suggested.

In a coherence-exchange experiment, the spin system oscillates between the same coherent states during the SEDRA

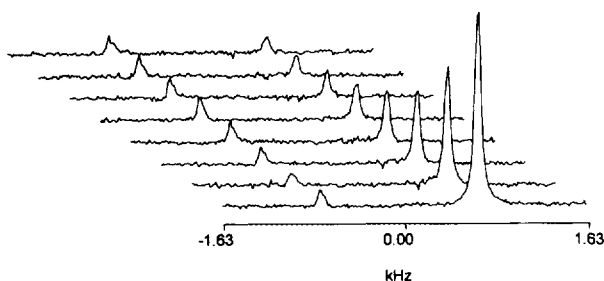-1.63   0.00   1.63

kHz

FIG. 2. Results of a set of coherence-transfer experiments. The first signal is obtained after cross polarization and each successive signal after the application of an additional t-SEDRA sequence of length $8T_R$. Acquisitions times were 22.4 ms. The initially excited nitrogen peak decreases due both to a T_2 process and to the coherence-transfer process. The peak from the nonprotonated nitrogen increases due to the coherence-transfer process; however, it is also subject to a T_2 dephasing process.

and the t-SEDRA irradiation pulses. Therefore we can expect that the T_2-relaxation effects are eliminated in the $\Delta S/S_0$ experiment. Another important effect that influences signal decay is the finite intensity of the pulses. Therefore it is important to minimize the number of additional pulses necessary in the t-SEDRA cycle. In the case of the magnetization exchange, the zero-quantum relaxation time T_{ZQ} influences the dipolar decay (10-12). Without an echo experiment that restores the decay and keeps the spin system in the same coherence manifold as the SEDRA, knowledge about the magnitude of T_{ZQ} is needed. This can usually be extracted from the spectral linewidth of the spin pair, as described by Kubo et al. (11). The effects caused by the pulse lengths can be monitored when the SEDRA sequence is applied to the equilibrium state $\rho(0) = (I_{z1} + I_{z2})$ of the spin pair. The average SEDRA Hamiltonian commutes with this spin state. However, the RF fields during the pulses, together with all interactions of the spins, will cause some destruction of this state (15). To eliminate at least this last effect, the magnitude of $\Delta S/S_0$,

$$\frac{\Delta S}{S_0} = \frac{S_0^z(8mT_R) - S^z(8mT_R)}{S_0^z(8mT_R)}, \quad [7]$$

could be monitored, where $S_0^z(8mT_R)$ is the signal resulting from the application of the SEDRA sequence to $(I_{z1} + I_{z2})$. If we ignore relaxation and pulse-length effects, this ratio is proportional to $\{1 - \cos[8m(2\omega_{SEDRA})T_R]\}$ for each microcrystal. The effective frequency of this experiment is twice as high as that for the coherence-exchange case.

The application of the RF-driven recoupling (RFDR) induced by the SEDRA π pulses on the longitudinal magnetization in the mixing time of a 2D correlation experiment was shown by Bennett et al. (18). They presented a 2D spectrum of a fully ^{13}C-enriched sample of alanine and demonstrated the possibility to perform dipolar-correlation spec-

troscopy on rotating solids. In the next section it is shown that a similar experiment can be performed with t-SEDRA in the mixing period.

t-SEDRA IN 2D CORRELATION SPECTROSCOPY

The t-SEDRA, being a coherence-transfer sequence, can also be utilized in 2D correlation experiments. This can be achieved by placing the t-SEDRA sequence in the mixing period of a 2D experiment, as shown in Fig. 1b. In order to obtain a proper spectrum, phase cycling of the $\pi/2$ pulse is necessary. To explain this, we make a distinction between a sequence with $\pi/2$ pulses in the x direction, t_x-SEDRA, and a sequence with these pulses in the y direction, t_y-SEDRA. The individual t_α-SEDRA (α = x, y) sequences refocus only the α components of the total spin coherences and lead to undesirable cross peaks.

Let us assume that immediately after the cross polarization in the experiment in Fig. 1b, the density matrix of the homonuclear spin pair equals $\rho(0) = a_1 I_{x1} + a_2 I_{x2}$. We further
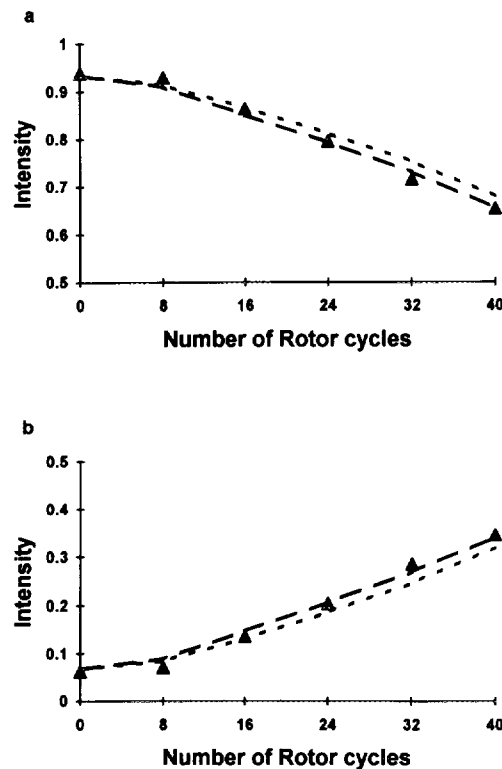

FIG. 3. Experimental results of relative signal intensities of the ^{15}N spin pair in PMP obtained after selective excitation, followed by coherence transfer induced by the t-SEDRA sequence. Simulated data, incorporating the effect of finite pulses, are shown and compared with the experimental results. The dotted line corresponds to a dipolar interaction of 420 Hz and the dashed line to dipolar interaction of 470 Hz between the nitrogen nuclei. In (a) the intensity of the centerband and sidebands of the protonated nitrogen is shown and in (b) the signal intensity for the nonprotonated nitrogen.

assume that, during the evolution and detection periods, the dipolar interaction does not affect the spins. This is a reasonable assumption for weak dipolar-coupled spins and as long as the rotational-resonance condition is not satisfied. At t_1 the spin system has a density matrix equal to

$$\boldsymbol{\rho}(t_1) = a_1 \{ \cos \Omega_1(t_1) I_{x1} + \sin \Omega_1(t_1) I_{y1} \}$$
$$+ a_2 \{ \cos \Omega_2(t_1) I_{x2} + \sin \Omega_2(t_1) I_{y2} \}, \quad [8]$$

where $\Omega_1(t_1)$ and $\Omega_2(t_1)$ are the accumulated phases of the two rotating spins due to their chemical-shift parameters. A straightforward calculation results in a transformation of the linear angular-momentum components of the spin system, due to the t_α-SEDRA sequences

$$I_{\alpha i} \xrightarrow{\quad t_\alpha\text{-SEDRA} \quad} \cos^2\theta I_{\alpha i} + \sin^2\theta I_{\alpha j}$$

$$I_{\beta i} \xrightarrow{\quad t_\alpha\text{-SEDRA} \quad} \cos\theta \cos 2\theta I_{\beta i} + \sin\theta \sin 2\theta I_{\beta j}, \quad [9]$$

where α and β equal x and y, or y and x, and $i, j = 1, 2$. The parameter θ is proportional to the length of the mixing time, which consists of m t-SEDRA cycles, and equals

$$\theta = 4m\omega_{\text{SEDRA}}T_R. \quad [10]$$

When the mixing is alternated between t_x-SEDRA and t_y-SEDRA and the signals in the detection period are added, the effective transfer of the linear coherence becomes

$$I_{xi} \xrightarrow{\quad t_x\text{-SEDRA} + t_y\text{-SEDRA} \quad} (\cos^2\theta + \cos\theta \cos 2\theta) I_{xi}$$
$$+ (\sin^2\theta + \sin\theta \sin 2\theta) I_{xj}$$

$$I_{yi} \xrightarrow{\quad t_x\text{-SEDRA} + t_y\text{-SEDRA} \quad} (\cos^2\theta + \cos\theta \cos 2\theta) I_{yi}$$
$$+ (\sin^2\theta + \sin\theta \sin 2\theta) I_{yj}.$$
$$[11]$$

With the diagonal- and cross-peak transfer coefficients

$$A_d = [\cos^2\theta + \cos\theta \cos 2\theta]$$

$$A_c = [\sin^2\theta + \sin\theta \sin 2\theta], \quad [12]$$

respectively, the signals during the detection period become

$$S(t_1, t_2) = A_d[a_1 e^{i\Omega_1(t_1+t_2)} + a_2 e^{i\Omega_2(t_1+t_2)}]$$
$$+ A_c[a_1 e^{i\Omega_1(t_1)} e^{-i\Omega_2(t_1)} e^{i\Omega_2(t_1+t_2)}$$
$$+ a_2 e^{i\Omega_2(t_1)} e^{-i\Omega_1(t_1)} e^{i\Omega_1(t_1+t_2)}], \quad [13]$$

where we use the fact that the mixing time is an integer multiple of the rotor period. The exponents in this equation can be expanded in Fourier series, with coefficients that correspond to the centerband and sideband amplitudes of the MAS CSA spectra of the two coupled spins:

$$e^{i\Omega_i(t)} = e^{i\Delta\omega_i t} \sum_{n=-\infty}^{\infty} I_{ni} e^{in\omega_R t}. \quad [14]$$

The 2D signal in Eq. [13] then becomes equal to

$$S(t_1, t_2) = A_d a_1 e^{i\Delta\omega_1(t_1+t_2)} \sum_{n=-\infty}^{\infty} I_{n1} e^{in\omega_R(t_1+t_2)}$$

$$+ A_d a_2 e^{i\Delta\omega_2(t_1+t_2)} \sum_{n=-\infty}^{\infty} I_{n2} e^{in\omega_R(t_1+t_2)}$$

$$+ A_c a_1 e^{i(\Delta\omega_1 t_1 + \Delta\omega_2 t_2)} \sum_{N,M,K=-\infty}^{\infty} I_{(N+M-K)1}$$

$$\times I^*_{M2} I_{K2} e^{i\omega_R(Nt_1 + Kt_2)} + A_c a_2 e^{i(\Delta\omega_2 t_1 + \Delta\omega_1 t_2)}$$

$$\times \sum_{N,M,K=-\infty}^{\infty} I_{(N+M-K)2} I^*_{M2} I_{K1} e^{i\omega_R(Nt_1 + Kt_2)}. \quad [15]$$

Here we have used the fact that during the mixing time the Hamiltonian is fixed; hence this time does not appear explicitly in the equations. $\Delta\omega_1$ and $\Delta\omega_2$ are the time-independent off-resonance values of spins 1 and 2, respectively.

The terms proportional to A_d result in the diagonal peaks in the double-Fourier-transformed 2D spectrum, while the terms proportional to A_c generate cross peaks between all centerbands and sidebands. The value of A_c depends on ω_{SEDRA} and the intensity of the cross peaks could be used to evaluate the strength of the dipolar interaction. Only simulations of powder averages of the spectra could provide this type of information. As was indicated before (13), the SEDRA sequence, and thus also the t-SEDRA sequence, is most efficient when $\omega_R \leqslant |\Delta\omega_1 - \Delta\omega_2| \leqslant 2\omega_R$. Therefore, when possible, one should try to choose a spinning speed that satisfies this condition for all interacting nuclei. For increasing spinning speed, the value for ω_{SEDRA} can still be significant enough to create cross peaks between the center- and sidebands of the interacting spins. However, the spinning speed should not be increased too much because then the efficiency of the coherence transfer will become zero.

Experimental 2D spectra of doubly ^{15}N-enriched PMP, obtained by the experiment shown in Fig. 1b with the proper phase cycling, is shown in Fig. 4. The mixing time in this experiment extended over 32 rotor periods and the spinning speed was $\nu_R = 2500$ Hz, which is larger than the value, 1.4 kHz, of $|\Delta\nu_1 - \Delta\nu_2|$. The ratio 0.56 between the isotropic chemical-shift difference and the spinning speed reduces the
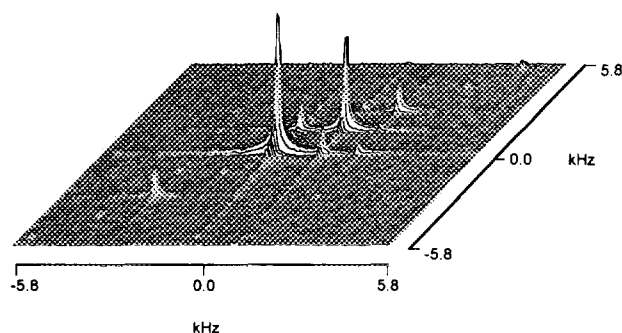
FIG. 4. Two-dimensional CPMAS correlation experiment with t$_o$-SEDRA in the mixing period. The mixing time was chosen to be 32 rotor cycles. The spinning speed was 2500 Hz and the CP contact time 5 ms. The repetition time of the experiment was 30 s and 24 signals were accumulated in the t_2 domain.

effective dipolar interaction by about 30%, which leaves enough dipolar interaction to cause a significant coherence transfer. To demonstrate the range of spinning frequencies for which the SEDRA signal decays are significant, an example of the signal intensity after SEDRA sequences of 16 ($2 \times 8T_R$) and 24 ($3 \times 8T_R$) rotor cycles is calculated and shown in Fig. 5. The intensities are evaluated for a spin pair with the CSA parameters of PMP and with changing ($\Delta\omega_1 - \Delta\omega_2$) value.

EXPERIMENTAL

All experiments were done on our homebuilt spectrometer, operating at 20.27 MHz for ^{15}N nuclei. The sample we used was solid 3(5)-methyl-5(3)-phenylpyrazole-^{15}N$_2$ (PMP). Solid PMP contains directly bonded ^{15}N spin pairs of the type -^{15}NH-^{15}N=. This molecule was previously analyzed for its intramolecular ^{15}N-^{15}N distance by various means. X-ray and neutron diffraction (26, 27) yielded an intramolecular distance of 1.36 Å. Static NMR lineshape analysis (25) gave a distance of 1.34 Å and recent CPMAS measurements, using the SEDRA sequence (14), gave an intramolecular distance of (1.38 ± 0.03) Å.

Two types of measurements were performed, using the t-SEDRA sequence. The first consisted of the coherence-transfer experiment and the second of the 2D correlation experiment. For the coherence transfer, selective excitation was achieved by a short cross-polarization time of 100 μs. This allowed an almost complete CP excitation of protonated nitrogen and hardly any excitation of the nonprotonated nitrogens. The ratio between the centerband peak intensities was 0.062. The spinning speed in the experiments was $\nu_R = 2.500$ kHz and a total of 32 signals were accumulated, with a repetition time of 30 s. Signals after up to six increments of eight rotor periods were measured, which amount to a total time of 16 ms. The intensity of the pulses during the t-SEDRA sequence was 60 kHz for the nitrogen and about

65 kHz for the proton decoupling. To avoid CP transfer during the application of the pulses on the nitrogens, the RF decoupling field strength on the protons was increased to 100 kHz during these pulses.

In the 2D correlation experiments, the spinning speed was also set to $\nu_R = 2.500$ kHz; 128 t_1 increments of 40 μs were used and the sampling time in the t_2 domain was chosen to be the same. The repetition time between signal accumulations was 30 s and the CP contact time was 5 ms. In addition to the normal CYCLOPS phase cycling, the $\pi/2$ pulses were alternately applied in the x and y directions.

Before the performance of the experiments, the t-SEDRA pulse sequence must be set accurately. For that purpose the SEDRA and t-SEDRA pulse cycles were applied to a singly labeled sample of (95%) benzamide-^{15}N and the value of $\Delta S = [S_0(32mT_R) - S_{\text{SEDRA}}(32mT_R)]$ was monitored. After a fine adjustment of all pulse lengths and an alternation of the $\pi/2$ pulses between $\pm x$, we achieved a ΔS value that was less than 3% of the S_0 ($32mT_R$) value itself.

For the theoretical curves in Fig. 3 we used a computer program (14) that takes into account all experimental data, including chemical-shift and dipolar-tensor parameters of the rotating spin pair and pulse lengths and intensities.

DISCUSSION

We have demonstrated the use of the t-SEDRA sequence and showed how it can be used to assist in extracting information about dipolar coupling parameters in systems containing homonuclear spin pairs. When measuring $\Delta S/S_0$ curves, t-SEDRA is used in order to obtain a reference signal. This eliminates contributions from noncoupled spins and corrects for relaxation effects and pulse imperfections.

Another use of the t-SEDRA sequence relies on selective initial excitation of one of the spins in the spin pair and
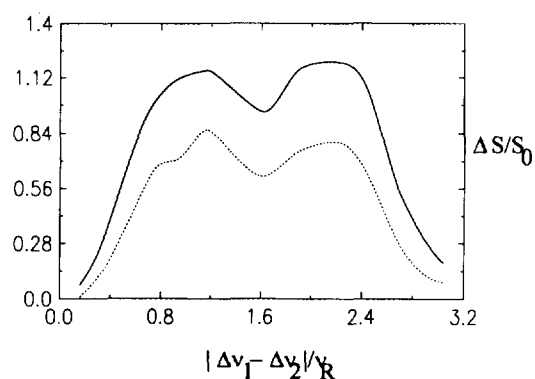

FIG. 5. The calculated intensities of spin-pair powder signals subjected to SEDRA cycles of lengths $16T_R$ and $24T_R$ are shown by the dotted and solid lines, respectively. The intensity is shown as a function of the difference between the isotropic chemical-shift values divided by the spinning frequency. The CSA parameters were choosen to be those of the PMP nitrogen nuclei.

enables the detection of the presence of dipolar-driven co-herence transfer. Experimental results can be subjected to quantitative analyses. If selective excitation is applied to a singly labeled molecule, the application of t-SEDRA could generate signals from naturally abundant spins that are coupled to the excited spin and hence provide a way of establishing multinuclear correlations. The subtraction of FID signals, obtained after SEDRA and t-SEDRA excitations, results in ΔS spectra that can be viewed as double-quantum-filtered spectra.

Two-dimensional MAS correlation experiments, with a t-SEDRA sequence in the mixing time, can also be performed in order to establish spin correlations in the same way as is done in liquid samples. For these experiments, the spinning speed should be chosen to maximize the effective dipolar interactions. The considerations of choosing the spinning speed for these experiments depend on the isotropic chem-ical-shift differences and the number of sidebands. To sim-plify the MAS sideband spectra, the spinning speed can be increased. However, this speed should not reach a level at which the ω_{SEDRA} frequency values of coupled spin pairs in the powder vanish.

The various applications of the basic SEDRA sequence in RFDR, BDR, and t-SEDRA serve the purpose of re-coupling homonuclear dipolar-coupled spin pairs rotating at the magic angle. Depending on the spin system under investigation, a choice of experiments can be made to ob-tain dipolar interactions or connectivities with the highest efficiency and best possible accuracy. In all cases, as much information as possible should be acquired on the chem-ical-shift parameters of the coupled spins, for the quan-titative analysis of the data.

REFERENCES

1. A. Pines, M. S. Gibby, and J. S. Waugh, *J. Chem. Phys.* **59,** 569 (1973).

2. J. Schaefer, E. O. Stejskal, and R. A. McKay, *J. Magn. Reson.* **25,** 569 (1977).

3. M. M. Maricq and J. S. Waugh, *J. Phys. Chem.* **70,** 3300 (1979).

4. J. Herzfeld and A. Berger, *J. Chem. Phys.* **73,** 6021 (1980).

5. M. G. Munowitz, R. G. Griffin, G. Bodenhausen, and T. H. Huang, *J. Am. Chem. Soc.* **103,** 2529 (1981).

6. T. Gullion and J. Schaefer, *Adv. Magn. Reson.* **13,** 57 (1989).

7. A. W. Hing, S. Vega, and J. Schaefer, *J. Magn. Reson.* **96,** 205 (1991).

8. M. G. Munowitz and R. G. Griffin, *J. Chem. Phys.* **76,** 2848 (1982).

9. D. P. Raleigh, M. H. Levitt, and R. G. Griffin, *Chem. Phys. Lett.* **146,** 71 (1988).

10. M. H. Levitt, D. P. Raleigh, F. Creuzett, and R. G. Griffin, *J. Chem. Phys.* **92**(11), 6347 (1990).

11. A. Kubo and C. A. McDowell, *J. Chem. Soc. Faraday Trans. 1* **84,** 3713 (1988).

12. D. P. Raleigh, F. Creuzet, S. K. Das Gupta, M. H. Levitt, and R. G. Griffin, *Am. Chem. Soc.* **111,** 4502 (1989).

13. T. Gullion and S. Vega, *Chem. Phys. Lett.* **194,** 423 (1992).

14. O. Weintraub, Ch. Hoelger, H. H. Limbach, and S. Vega, *J. Magn. Reson. A* **109,** 14 (1994).

15. D. K. Sodickson, M. H. Levitt, S. Vega, and R. G. Griffin, *J. Chem. Phys.* **98,** 6742 (1993).

16. R. Tycko and G. Dabbagh, *Chem. Phys. Lett.* **173,** 461 (1990).

17. J. M. Joers, R. Rosanke, T. Gulion, and J. R. Garbow, *J. Magn. Reson. A* **106,** 123 (1994).

18. A. E. Bennett, J. H. Ok, S. Vega, and R. G. Griffin, *J. Chem. Phys.* **96,** 8624 (1992).

19. A. Kubo and C. A. McDowell, *J. Chem. Phys.* **92,** 7156 (1990).

20. J. H. Ok, R. G. S. Spencer, A. E. Bennett, and R. G. Griffin, *Chem. Phys. Lett.* **197,** 389 (1992).

21. O. Weintraub and S. Vega, *J. Magn. Reson. A* **105,** 245 (1993).

22. U. Haeberlen, "Advances in Magnetic Resonance," Suppl. 1, Aca-demic Press, New York, 1976.

23. P. Caravatti, G. Bodenhausen, and R. R. Ernst, *J. Magn. Reson.* **55,** 88 (1983).

24. F. Aguilar-Parilla, C. Cativiela, M. D. D. de Villegas, J. Elguero, C. Foces-Foces, J. I. G. Laureiro, F. H. Cano, H. H. Limbach, J. A. A. Smith, and C. Toiron, *J. Chem. Soc. Perkin. Trans.* **2,** 1737 (1992).

25. Ch. Hoelger, F. Aguilar-Parilla, J. Elguero, and H. H. Limbach, un-published results.

26. E. N. Masien, J. R. Cannon, A. H. White, and A. C. Willis, *J. Chem. Soc. Perkin. Trans.* **2,** 1298 (1974).

27. F. H. Moore, A. H. White, and A. C. Willis, *J. Chem. Soc. Perkin. Trans.* **2,** 1068 (1975).