Sensitizing solid state nuclear magnetic resonance of dilute nuclei by spin-diffusion assisted polarization transfers

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Recent years have witnessed efforts geared at increasing the sensitivity of NMR experiments, by relying on the suitable tailoring and exploitation of relaxation phenomena. These efforts have included the use of paramagnetic agents, enhanced $^1$H–$^1$H incoherent and coherent transfers processes in 2D liquid state spectroscopy, and homonuclear $^{13}$C–$^{13}$C spin diffusion effects in labeled solids. The present study examines some of the opportunities that could open when exploiting spontaneous $^1$H–$^1$H spin-diffusion processes, to enhance relaxation and to improve the sensitivity of dilute nuclei in solid state NMR measurements. It is shown that polarization transfer experiments executed under sufficiently fast magic-angle-spinning conditions, enable a selective polarization of the dilute low-$\gamma$ spins by their immediate neighboring protons. Repolarization of the latter can then occur during the time involved in monitoring the signal emitted by the low-$\gamma$ nuclei. The basic features involved in the resulting approach, and its potential to improve the effective sensitivity of solid state NMR measurements on dilute nuclei, are analyzed. Experimental tests witness the advantages that could reside from utilizing this kind of approach over conventional cross-polarization processes. These measurements also highlight a number of limitations that will have to be overcome for transforming selective polarization transfers of this kind into analytical methods of choice. © 2011 American Institute of Physics. [doi:10.1063/1.3643116]

I. INTRODUCTION

Sensitivity remains a—if not the—main concern in the scope of applications that nuclear magnetic resonance (NMR) spectroscopy can tackle. Sensitivity is given by a number of independent factors including sample availability within the coil volume, the degree of spin polarization, and the influence and character of background noise. Another important factor in improving NMR’s effective signal-to-noise ratio (SNR) can stem from methods that increase the rates at which the spins’ resonance frequencies can be measured. Foremost among these advances was the introduction of Fourier transform (FT) principles, which by compressing the data acquisition process brought about multi-fold improvements in the values that SNR could achieve per unit time (SNR$_t$). Additional strategies to achieve such aim shift their focus from the data acquisition process, to the shortening of the otherwise long recycle delays that might be needed in FT NMR. While initially relying on the use of Ernst-angle excitations and on natural $T_1$ relaxation processes to maximize the SNR, recent years have witnessed the introduction of more proactive approaches to assist in such rebuilding of magnetizations. Particularly successful protocols employed to such aim include the use of paramagnetic relaxation agents in NMR studies of solid and liquid proteins, repeated repolarizations of targeted $^{13}$C moieties in biosolids via selective homonuclear excitation and recoupling sequences, selective excitation of only those resonances to be targeted in solution biomolecular NMR experiments, and active repolarization of $^{13}$C-bound protons in solution-phase experiments by means of homonuclear mixing sequences. The last of these alternatives are is most closely relevant to the present work, whose aim is to explore how the combination of the principles that underlie selective $^1$H excitation experiments could aid in the acquisition of solid state NMR spectra from dilute spins.

Solid state NMR observations of low-$\gamma$ nuclei $\delta$ like $^{13}$C or $^{15}$N, are normally based on transfer processes, such as cross-polarization [CP (Ref. 15–18)] from $^1$H nuclei, or on the INEPT three-pulse scheme. These sequences can transfer spin order from an ensemble of protons $I$ to the targeted spin $\delta$, enhancing the latter’s polarization by a factor of $\approx \gamma_I/\gamma_\delta$: they also enable one to repeat the signal averaging process using a recycle time delay dictated by the usually faster $I$-spin proton $T_1$ relaxation. Still, in cases dealing with dilute low-$\gamma$ nuclei, this enhancement amounts to only a fraction of the full gain that could potentially be obtained. In principle, this should encompass an additional factor $\approx \sqrt{(N_I/N_\delta)}$, accounting for the fact that the number $N_I$ of polarized $^1$Hs exceeds the target’s abundance by a population difference that is on the order of this ratio. It is known that such transfer is not enabled by unitary manipulations, leaving a substantial pool of potential sensitivity untapped. This work explores whether non-unitary mechanisms underlying relaxation-enhancing experiments that have recently emerged in the field of solution-phase NMR could be of use to exploit this untapped potential. Specifically, we explore whether $^1$H–$^1$H spin diffusion processes occurring spontaneously even under fast magic-angle-spinning (MAS) conditions can be combined with selective excitation processes to

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idealized spin-dynamic simulations for L-tyrosine’s geometry, spontaneous $^1$H–$^1$H spin-diffusion processes that arise in such powdered networks could then rapidly reinstate the expended $^1$H polarization. Even under MAS conditions, such homogenizing processes could take place in timescales equal (or shorter) to those required for implementing a normal $^{13}$C free induction decay (FID) acquisition process. This is a potential source of SNR, enhancement, which is here explored.

The redistribution of magnetizations illustrated in Figure 1 among protons that transferred their polarization to their bonded $^{13}$C and those $^1$Hs that are distant to the latter, amounts to a spontaneous shortening of the effective longitudinal relaxation time characterizing the $^{13}$C-bound protons. This is akin to processes that have been reported in solution-phase selective excitation experiments. This would also mean that a scan involving a neighbor-specific polarization transfer process could be repeated at rates that are much higher than those involved in conventional acquisitions, leading, in turn, to an increase in the effective spectral SNR. Such gain would effectively exploit, via an intermediate transfer step involving the $S$-bound $^1$Hs, the large $N_I/N_S$ polarization reservoir that cannot be efficiently transferred via coherent heteronuclear unitary processes—thus, materializing to some extent the maximum gain one could expect for this process based on thermodynamic arguments. Variations in the quantitative aspects of this scenario would arise if considering experiments on other nuclei possessing different natural abundances than $^{13}$C, or on labeled biomolecules possessing varying degrees of isotopic substitution. But in any case it is clear that if the concept just described could be realized, improvements in the sensitivity of a NMR experiment per unit of signal averaging time would materialize.

While the spin-diffusion processes illustrated by the solid state simulations of Figure 1 appear to proceed significantly faster than their solution-state counterparts, translating their potential into increased $^{13}$C sensitivities poses a number of challenges. One of these stems from the relative lack of specificity of the $H \rightarrow C/N$ polarization transfer, a scenario that would break the local-transfer assumption underlying the hypotheses above. For instance, ancillary experiments reveal that, even if executed with relatively short contact times (on the order of 0.05–0.1 ms), the depletion of the bulk proton magnetization under the action of cross-polarization sequences was not sufficiently localized to justify the execution of these experiments. Further complications arise from the need to execute some form of RF-driven high-power heteronuclear decoupling, for achieving high resolution in this kind of solid state NMR experiments. This is a need which can often be obviated in liquid-phase experiments; for instance, by relying on spin-state selective sequences, or by using indirectly detected schemes involving the action of a single $\pi$-pulse on the $^1$Hs over the course of the whole low-$\gamma$ evolution period. By contrast, an active reliance on heteronuclear decoupling—with ensuing $^1$H saturation effects—cannot be avoided in solid-state $^{13}$C/$^{15}$N studies. In order to deal with these two issues we decided to try and mimic “liquid-like” conditions like those present in the above-mentioned experiments by (i) implementing tests at the

**II. THEORETICAL CONSIDERATIONS**

As mentioned, most solid state NMR experiments on $^{13}$C or $^{15}$N involve a polarization transfer from $^I = 1$Hs to the targeted $S$ nuclei; commonly a CP step under MAS-modified (e.g., ramped or adiabatic) Hartmann-Hahn conditions. The contact time defining the length of this CP is usually set sufficiently long to maximize the $S$-spin polarization, without much concern on whether this transfer comes mostly from a specific $^1$H or not. By contrast, and in parallel with the liquid-state experiments alluded to earlier, we shall focus in this study on cases where the $\approx \gamma_I/\gamma_S$ $S$-spin polarization enhancement factor comes mostly from neighboring $^1$Hs, while attempting to leave all other protons in the ensemble untouched. In a natural-abundance sample, where $^{13}$C makes up roughly 1% of the actual molecular skeletons, such neighbor-specific polarization transfer process would then deplete spin order located in the directly bonded protons; but it would leave $\approx 99\%$ of the remaining $^1$Hs with their magnetizations nearly fully relaxed. As Figure 1 illustrates with
highest MAS spinning rates available—in our case on the order of 40 kHz—so as to maximize the locality of the heteronuclear transfer and alleviate the needs of the dipolar decoupling; (ii) replacing the cross-polarization transfer block by a dipole-driven, rotor-synchronized, INEPT-like block, tuned to maximize the single-bond $I \rightarrow S$ transfer; (iii) appending to this modified INEPT an additional storage pulse for flipping all non-bonded $^1$H magnetizations back to the longitudinal $z$ axis; and (iv) replacing the usual high-power forms of heteronuclear decoupling by an acquisition that incorporates an even-numbered rotor-synchronized train of $(\pi)^{13}$ pulses, implemented using the highest available RF field strengths to reach the scenario illustrated in Figs. 1(b) and 1(d). All these processes would combine to return the $^{12}$C-bound $^1$H magnetizations back to the $z$ axis at the conclusion of a hetero-decoupled $^{13}$C acquisition, while allowing a low duty cycle and, hence, short recycling delays. A pulse sequence resulting from incorporating all these considerations is illustrated in Scheme 1. Section III provides further details on the operation of the resulting sequence, which, henceforth, we refer to as the SPIn-diffusion assisted polarizing (SPIDAP) experiment, together with technical aspects of its implementation.

### III. EXPERIMENTAL METHODS

NMR experiments were performed on a Varian VNMR S® 600 MHz NMR spectrometer using a triple-resonance probe (double-tuned to $^1$H and $^{13}$C operation) and a 1.6 mm spinner capable of spinning samples stably up to 40 kHz speeds. Typical $\pi/2$ pulse lengths for this system were 1.5 $\mu$s for $^{13}$C, and $^1$H $\pi/2$'s ranging from 0.7 to 1.6 $\mu$s, respectively, at $\approx$100–500 W power levels. For every sample that was examined, three complementary 1D NMR data sets were usually recorded. One of these involved a conventional $^{13}$C Cross Polarization Magic Angle Spinning (CPMAS) experiment with Two Pulse Phase Modulation (TPPM) $^1$H decoupling during the acquisition, and contact conditions optimized to the specific spinning rate employed. The remaining experiments assessed were based on the refocused INEPT illustrated in Scheme 1. This experiment includes a canonical three-pulse $(\pi/2)^{1H}_x – \tau – (\pi/2)^{1H}_y, (\pi/2)^{1C}_x – \tau –$ Freeman-Morris transfer, with both $\tau$ values set to single rotor period $T_r$. In order to prevent the MAS averaging of the dipolar interaction during the course of these periods, $(\pi)^{1H}$ pulses were placed mid-point of these $^1$H $\rightarrow$ $^{13}$C transfer periods $T_r$, leading to a Transferred Echo Double Resonance (TEDOR)-like transfer. As the spinning rates used ($\approx$30–40 kHz) were on the order of the one-bond heteronuclear dipolar coupling strengths ($\approx$25 kHz), this enabled the $^{13}$C-bound protons to execute a specific transfer that would otherwise be averaged away by the sample spinning. Then, in order to keep the polarization of the remaining $^{12}$C-bound proton reservoir available for a subsequent scan, this specific transfer was concluded by a $(\pi/2)^{1H}_x$ pulse. This pulse had only a limited effect on the protons that had transferred their polarization to the $^{13}$C, while acting as storage of all the remaining unused magnetizations back along the main magnetic field. $^1$Hs that were involved in the polarization transfer to the $^{13}$C nuclei, could thus be assisted in regaining back their polarizations by spin diffusing from this “unused” proton reservoir.

Following this transfer the $^{13}$C signal was monitored. This signal acquisition process had to be executed using a heteronuclear decoupling that would lead to sharp $^{13}$C lines, but would not significantly affect the stored $^{12}$C-bound proton magnetization, and which would not require high-power decoupling conditions that are ill-posed to fast repetition rates. Simulations on small spin systems revealed that fast spinning could suffice towards this end; experimentally, however, line widths in excess of 1 kHz were observed in the absence of any form of $^1$H irradiation. A number of decoupling sequences based on the application of rotor-synchronized $(\pi)^{1H}$ pulse-trains were, thus, assayed. The behavior of these decoupling sequences was investigated both numerically and experimentally as a function of rotor synchronization, radiofrequency strength, and $\pi$-pulse phases. The best overall results in terms of $^{13}$C line width and preservation of the proton reservoir were obtained when utilizing the shortest possible $\pi$ pulses, one pulse per rotor cycle, and phases corresponding to either XY-4 (x-y-x-y) (Ref. 31) or to the x-y-x-y-x-y-x-cycle. For the sake of completion, a third set of experiments based on the use of CP- or INEPT-based transfer and TPPM decoupling, was also implemented.

### IV. RESULTS

Before exploring the fast recovery features introduced in Figure 1, attention was focused on optimizing the means of achieving suitable decoupling with a low duty cycle. Figure 2 presents a series of $^{13}$C NMR spectra acquired at MAS frequencies of 35–40 kHz on natural-abundance sucrose and L-tyrosine powders, comparing the decoupling performance observed using high-power TPPM with that obtained upon performing decoupling with one proton $\pi$ pulse every $T_r$ rotor period. For the 30–40 kHz MAS speeds here investigated, the decoupling performance observed with these two alternatives was similar: line widths of $\approx$100 Hz resulted in both cases. Other approaches including the use of a single $^1$H $\pi$ pulse...
FIG. 2. Comparison between the $^1$H decoupling performances afforded by the rotor-synchronized $\pi$-pulse supercycled train given in the experimental (red traces) vs TPPM (black). Experiments were performed on L-tyrosine [(a), (b)] and on sucrose [(c), (d)] at MAS speeds of 35 kHz [(a), (c)] and 40 kHz [(b), (d)]. $^1$H $\pi$ pulses were set to 3.2 (tyrosine) and 1.5 (sucrose) $\mu$s; TPPM RF strengths were set at 94 kHz (tyrosine) and 179 kHz (sucrose), respectively. INEPT was used in conjunction with the $\pi$ pulse decoupling, whereas cross-polarization was employed in combination with TPPM (spectra were scaled to equal maximum peak heights, and are not meant to compare sensitivities).

every two or three rotor periods yielded worse line shapes, and therefore their use was not pursued despite the advantages that may have resulted from their lower duty cycle (vide infra). When comparing the absolute spectral SNRs of short-contact-time ($\approx 50$ $\mu$s) CP experiments incorporating TPPM decoupling against those based on a single-$T_r$ INEPT-based enhancement sequence, similar sensitivities per unit time resulted. All further comparisons are, thus, based on this heteronuclear polarization transfer protocol.

Next, the $^1$H($^{12}$C) $\rightarrow$ $^1$H($^{13}$C) polarization transfer hypothesis was compared in experiments involving the rotor-synchronized $\pi$-based decoupling introduced in Scheme 1. To evaluate this homonuclear $^1$H polarization transfer feature, experiments were done with the last $^1$H ($\pi/2$) flip-back pulse of the INEPT module ($-x$ phase in Scheme 1) enabled and disabled. Also monitored in these tests was the performance obtained when the INEPT transfer was followed by TPPM decoupling. SNR$_s$ were then determined from proton-bearing carbons for different recycle delays—in all instances for the same total acquisition time for each experiment. Results arising from these comparisons for the aromatic sites of tyrosine, are shown in Figure 3. As expected, the maximum SNR$_s$ shifts towards lower recycle delays in the flip-back cases, evidencing a faster apparent $T_1$ relaxation. A sensitivity enhancement of around 40% was observed when the ($\pi/2$)$^1$H flip-back pulse was added to the $\pi$-pulse decoupling sequence. Still, as the proton relaxation times $T_1$ measured for tyrosine were about 1 s, the maximum SNR for the other two experiments was not significantly affected by the presence or absence of a flip-back pulse. For the maximum recycle delay assayed (4 s) one sees that, in fact, SNRs are very close with and without the flip-back variants, witnessing the fact that at such recycle delay protons are relaxed almost fully to equilibrium and further polarization recoveries through spin-diffusion become superfluous.

Although reaching SNR$_s$ that were only a fraction of their INEPT-based counterparts, we consider it worth to investigate the effects that flip-back pulses could have upon using CP variants of the experiment—both for the $\pi$-pulse decoupling and the standard TPPM decoupling. These measurements generally focused on short contact times, to ensure a selectivity of the polarization transfer and not disturbing most of the protons in the sample. These results are summarized in Figure 4. Although for the shortest contact times a shift of the maximum SNR$_s$ akin to that evidenced in the INEPT case is also here observed, contact times of 200 $\mu$s or longer blur these effects. This is likely reflecting a decreased selectivity of the CP process at longer contact times. These measurements also evidence, via the SNR$_s$ build-up at shorter times, the higher efficiency of the $\pi$-pulse based decoupling strategy for recycling magnetization, over continuous irradiation approaches which surely affect the bulk $^1$H magnetization more severely.
FIG. 4. $^{13}$C SNR$_t$'s experiments akin to those in Figure 3, recorded at a spinning frequency of 35 kHz as a function of recycle delays, but starting from cross polarizations (CP) with the indicated contact times. After these polarization transfers either a $\pi$-based (red) or TPPM (black) were used for $^1$H decoupling; for the former cases a $^1$H 90° flipback pulse was applied after the CP contact. (The duration of each acquisition was here set to 5 min and all plots are adjusted to a long-term CP TPPM SNR$_t$ of unity.)

A second set of tests was performed for sucrose. The proton relaxation times $T_1$ of sucrose are approximately an order of magnitude longer than those of tyrosine, and thus one expect standard experiments to yield maximal SNR$_t$ when using recycle delays of 30–60 s. This is indeed observed, as evidenced by a significant shift of the maximum SNR per unit time towards smaller recycle delays (Figure 5). A significant sensitivity enhancement is also easily noticed in these data upon including a flip-back pulse.

V. DISCUSSION

The results just presented evidence the feasibility of relying on spin-diffusion processes among the $^{12}$C/$^{13}$C-bonded protons in a solid, to transfer underutilized polarization from the former proton ensemble to the latter. Both in terms of the shortening of the effective relaxation times and of the higher SNR per unit time that can be achieved, these effects display a clear parallel with relaxation enhancement processes that have been observed and exploited in solution state NMR experiments of biomolecules. Still, both for the case of the tyrosine and sucrose samples hereby explored (and presumably for other typical rigid solids as well), the repolarization process also ended up involving an apparent time scale that was on the order of $\approx$1 s. This is a much longer delay than what we had originally anticipated, and places a limit on the usefulness of the approach as hereby described.

To further understand the source of this discrepancy, a series of ancillary measurements and calculations were made. The former were aimed at finding out how much of the $^1$H bulk polarization is actually preserved at the conclusion of the SPIDAP flip-backed acquisition. The latter were aimed at elucidating to what extent could the quantum-mechanical simulations displayed in Figure 1 for intra-molecular, small-cluster, spin-diffusion phenomena be extended to the much larger $^1$H spin networks that will have to be involved when exploiting the use of these phenomena for collecting natural-abundance $^{13}$C spectra. Figure 6(a) presents representative results summarizing the first of these issues: it displays the $^1$H

FIG. 5. Representative $^{13}$C SNR$_t$'s measured for sucrose as function of the recycle delay. Acquisition conditions were similar as in Fig. 3 ($\pi$-pulse decoupling without/with flipback pulse for red/blue traces and TPPM decoupling for black trace), and differed among the panels as follows. [(a), (b)]: one $\pi$-pulse decoupling per $T_r$; [(c), (d)]: one $\pi$-pulse decoupling per 2$T_r$. [(a)–(c)]: 5 ms total acquisition; (d) 10 ms total acquisition time. (a) MAS at 35 kHz; [(b)–(d)] MAS at 40 kHz. SNR$_t$'s were normalized to unity at long recycle delay times, as determined for the pyranose carbons in experiments lasting in all cases for 80 min.
signal intensity observed upon implementing a SPIDAP sequence akin to that introduced in Figure 1, but which upon conclusion was followed with a single \((\pi/2)^1\mathrm{H}\) and by a simple \(^1\mathrm{H}\)-detected acquisition under MAS. Varied in these set of measurements was the time that would be devoted to the \(^{13}\mathrm{C}\) NMR signal acquisition—in essence, varying the number of rotor-synchronized \((\pi)^{1}\mathrm{H}\) decoupling pulses that would be used. If all these (even-numbered) \(\pi\)-flips would be acting ideally and instantaneously, the ensuing \(^1\mathrm{H}\) signal intensity should not depend on this parameter. By contrast, it can be clearly seen that as the \(^{13}\mathrm{C}\) signal acquisition goes into the ms regime and the number of \((\pi)^{1}\mathrm{H}\) decoupling pulses increases up to several hundreds, a progressive saturation of the bulk proton magnetization sets in. We attempted to remedy this deficiency by reducing the decoupling duty cycle to fewer rotor periods, but the ensuing reduction of the acquisition times ended up leading to a doubling of the \(^{13}\mathrm{C}\) line widths. Alternative routes involving different phase-cycling of the pulses are currently being sought to deal with this complication, so as to further enhance the sensitivity performance of this approach.

It follows from Figure 6(a) that the \(\pi\)-pulse proton decoupling train used in the experiments presented in Figs. 3–5, which employed some 200 decoupling cycles, only left \(\approx 1/2\) of the original \(^1\mathrm{H}\) bulk magnetization in a steady state. (As mentioned, higher or lower levels of bulk \(^1\mathrm{H}\) polarization could be reached by tailoring the overall \(^{13}\mathrm{C}\) acquisition time.) It is interesting to consider how would the kind of repolarization profiles introduced in Figure 1 change, under such varying steady-state conditions. To investigate this, we considered a \(^1\mathrm{H}\) reservoir made of three “different” kinds of protons: those which would be directly connected to the \(^{13}\mathrm{C}\) and whose polarization would be entirely expended over the course of the INEPT transfer; those next nearest \(^1\mathrm{H}\) neighbors which, as illustrated in Figure 1, would quickly repolarize these expended polarizations due to strong intramolecular (or short-distance intermolecular) dipolar couplings; and the bulk of the proton reservoir which would, in turn, have to repolarize this strongly coupled sphere but would do so on a slower timescale. Calling these three reservoirs \(A\), \(B\), and \(C\), respectively, we decided to model these transfers with a set of simple exchange equations:

\[
\begin{align*}
\frac{dM_A}{dt} &= -N_BR_{AB}M_A + R_{AB}M_B + 0 \times M_C - (M_A - M_{40})/T_1, \\
\frac{dM_B}{dt} &= N_BR_{AB}M_A - R_{AB}M_B - N_CM_{BC}M_B + N_BR_{BC}M_C - (M_B - M_{80})/T_1, \\
\frac{dM_C}{dt} &= 0 \times M_A + N_CM_{BC}M_B - N_BR_{BC}M_C - (M_C - M_{60})/T_1.
\end{align*}
\]

Here, \(N_{A,B,C}\) represent here the “sizes” of the three domains. For the sake of meaningfulness these were set to \(N_A = 0.01\), \(N_B = 0.05\), \(N_C = 0.94\), respectively, so as to account for the very different populations that these spin reservoirs would entail in a typical natural abundance organic solid. In the solution of these equations it was then assumed that \(M_A(\text{time} = 0) = 0\) which would account for the full initial transfer from \(A\) to the bonded \(^{13}\mathrm{C}\). As for the exchange rates between the different reservoirs these were assumed to be of the order \(R_{AB} = 2-4\) kHz for the intramolecular exchange (values based on the average build-up curves predicted by the simulations in Fig. 1), and \(R_{BC} = 0.5-1\) kHz for the weaker intermolecular exchange (which is further slowed down by the fast MAS).

A common \(T_1 = 1\) s value was assumed for all protons, and deviations from full relaxation originating from the effects of the \((\pi)^{1}\mathrm{H}\) pulse train (i.e., the effect observed in Fig. 6(a)) were accounted for by manipulating the size of \(N_C\). Figures 6(b) and 6(c) illustrate the kind of build-up curves that are then predicted as a function of progressive saturation of the \(^1\mathrm{H}\) bulk polarization, focusing on the \(^{13}\mathrm{C}\) SNR expected per unit time as can be derived from the degree of site’s \(A\) polarization. These curves show, overall, a close resemblance to the kind of behavior that is observed experimentally: the rapid buildup predicted by the small-cluster simulations in Figure 1 is replaced by a slower repolarization that is essentially dictated by a cross relaxation rate related to the bulk \(^1\mathrm{H}\) reservoir; the effects of progressive saturation are also clearly evidenced by the maximum SNR gain that the steady-state repetition of this experiment can achieve.

It follows from all these considerations that improvements in the performance of conventional solid state \(^{13}\mathrm{C}\) or \(^{15}\mathrm{N}\) NMR acquisitions can benefit from enhancing spin-diffusion processes among the various \(^1\mathrm{H}\) spin reservoirs—something which, in turn, is possible only by using pulse sequences which, like the one introduced in this paper, are optimized to this effect. At the same time it appears that excessive pulsing—particularly, as related to the high-power sequences usually employed to achieve heteronuclear decoupling—could impair the gains of these

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**FIG. 6.** (a) Experimental \(^1\mathrm{H}\) longitudinal magnetization loss during a \(\pi\)-pulse train sequence akin to that in Scheme 1 for decoupling purposes, as a function of the number of intervening \((\pi)^{1}\mathrm{H}\) pulses. (b) Simulation for \(R_{AB}=4\) kHz, \(R_{BC}=1\) kHz. (c) Long-term normalized \(^{13}\mathrm{C}\) SNR's calculated with Eq. (1) as function of the recycle delay, assuming that the kind of losses illustrated in panel (a) amounted to 0% (red), 20% (cyan), 50% (blue), and 100% (black) of the bulk \(^1\mathrm{H}\) reservoir \(N_C\). The diffusion rates indicated in the figure correspond to the rates in Eq. (1), and a longitudinal relaxation time \(T_1 \approx 1\) s was assumed in all cases.
approaches, owing to a progressive destruction of the $^1\text{H}$ bulk polarization. Methods to deal simultaneously with these conflicting requirements were here presented; further optimizations of these conditions are currently being pursued.

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