Increasing the accuracy of structural investigations by MAS spin-echo solid-state NMR experiments

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Abstract. The use of rotor-synchronized Hahn-echo pulse train (RS-HEPT) ¹H decoupling, in a MAS (Magic Angle Spinning) ¹⁵N spin-echo NMR experiment allows the use of longer the spin-echo durations compared to the case when conventional decoupling is applied, hence enabling the accurate determination of very small J couplings down to a few Hz. Here, a thorough numerical analysis of the ¹⁵N spin-echo under RS-HEPT decoupling is performed with the purpose of characterizing the factors that still limit its efficiency. The obtained results are promising with respect to further improving the accuracy in the measured J couplings.

1. Introduction

To characterize the hydrogen bonding network formed in molecular superstructures, solid-state nuclear magnetic resonance (SS-NMR) spectroscopy emerges as a powerful technique capable to provide structural details with atomic resolution. In particular, the modulating effect of J couplings upon spin echoes is a key property [1], which enables the measurement of very weak coupling constants in SS-NMR experiments designed to provide through-bond structural characterization. Since J couplings are not normally resolved in ordinary one-dimensional SS-NMR spectra, a τ-π-τ spin-echo magic angle spinning (MAS) experiment is needed to refocus evolution due to magnetic field inhomogeneity or disorder. The use of efficient heteronuclear decoupling gives transverse dephasing times, T₂*, that are usually much longer than the decay constants of the non-refocused NMR signal, T₁*, which characterize the free-induction decay. Hence it is possible to obtain refocused line widths (Δ′ = 1/πT₂*) which are sufficiently narrow so as to observe splitting due to J couplings [2].

Previous work [3,4] has been demonstrated the use of cross-polarization (CP) spin-echo MAS experiments employing a low duty-cycle ¹H decoupling scheme based upon rotor-synchronized π pulses, namely the RS-HEPT sequence [5]. This enabled the accurate measurement of small J couplings for two synthetic deoxyguanosine derivatives by ¹⁵N MAS NMR spin-echo experiments [3]. Here, we provide a thorough numerical analysis of the ¹⁵N spin-echo under RS-HEPT decoupling with the purpose of characterizing the factors that still limit the efficiency of the proposed technique. The derived conclusions are expected to constitute the theoretical basis for further improvements in the applied heteronuclear decoupling and, consequently, reduce the magnitude of the smallest possible measured J couplings.
2. Experimental setup
Figure 1 shows the MAS spin-echo pulse sequence. $^{15}$N transverse magnetization is created using cross-polarization from $^1$H. The rotor-synchronized Hahn-echo pulse train (RS-HEPT) decoupling sequence [5], applied during the spin-echo evolution period of $2\tau$ consists of a $^1$H $\pi$ pulse every two rotor periods. This has been applied previously in one-dimensional $^{13}$C CP MAS experiments on adamantane. For each resonance in the $^{15}$N NMR spectrum acquired by the MAS spin-echo experiment, corresponding spin-echo curves have been measured by incrementing the echo time with integer multiples of the rotor periods, $\tau_R$, and observing the dependence of the corresponding line intensity as function of $2\tau$. An exponential decay was found in all cases with decay rates, $T_2'$.

3. Numerical simulations
The use of RS-HEPT decoupling in the spin-echo experiment is a key element for measuring weak $J$ coupling constants with relevance in identifying structural motifs induced by hydrogen bonding in supra-molecular systems. The decoupling performance of the sequence yields sufficiently long $T_2'$ dephasing times to allow the determination of the $J$ couplings for the non-protonated $^{15}$N resonances. It is to be noted that in the absence of $^1$H decoupling, $T_2'$ values were two to three times shorter as compared to the values determined with RS-HEPT decoupling. While more efficient than no decoupling at all, the RS-HEPT decoupling is less efficient than current high-power decoupling techniques, such as TPPM decoupling. However, the use of high-power continuous $^1$H heteronuclear decoupling, is unpractical for measuring very weak $J$ couplings because the long spin-echo duration required to identify their modulation effect upon the spin-echo decay curve can cause serious damage to the r.f. electronics.

By employing the experimental setup in figure 1, spin-echo decay times up to 200 ms could be obtained [3], which made it possible to measure weak $J$ couplings, down to 3.8 Hz (corresponding to intra-molecular $J$ couplings over two bonds). However, the measurement of even smaller $J$ coupling constants would be beneficial with respect to better structural characterization of supramolecular aggregates. To accomplish this, however, the heteronuclear decoupling efficiency during the spin-echo evolution by RS-HEPT has to be further improved. For this purpose, in the present work a thorough numerical analysis has been performed, which is aimed at identifying the main mechanisms that still limit the decoupling performances of the RS-HEPT sequence. In this study, numerical simulations of the $^{15}$N spin-echo experiment in figure 1 on extended NH7 spin systems have been performed using the SPINEVOLUTION program [6], by considering a spatial arrangement that is representative for organic compounds.
The results of the numerically simulated $^{15}$N spin-echo decay curves obtained by incrementing the spin-echo time $2 \tau$ are shown in figure 2. First, the experimental results obtained in [3] are compared with the simulated spin-echo curve (represented by thin continuous line in figure 2a), where the employed isotropic, and anisotropic, $^1$H chemical shift parameters are typical for the involved protons in deoxyguanosine, and a spinning frequency of 22.5 kHz has been considered. As can be seen, spin-echo durations up to 200 ms have been also obtained in our theoretical investigation, which agrees very well with the experimental values. One can thus conclude that the investigated spin system is sufficiently large to correctly reproduce the main experimental findings.

Next, we investigate two major mechanisms under pulsed-decoupling by RS-HEPT that prevent us from completely eliminating the influence of protons upon the evolution of low-$\gamma$ nuclear spins. In ref. [5] they have been identified as third order effects due to mixing terms between the isotropic $^1$H chemical shift, homo- and heteronuclear dipolar interaction Hamiltonians, and the interference of the latter with $^1$H chemical shift anisotropy, respectively. To obtain more insights into these effects, the spin-echo curve in the absence of proton chemical shift interactions (both, isotropic, and anisotropic) has been simulated and considered as a reference curve for ideal decoupling conditions (thick continuous line in figure 2a). As can be seen, there is almost no decay under this particular conditions, which reveals the strong influence of the $^1$H chemical shift upon the achievable decoupling efficiency. Furthermore, to identify the separate effect of the isotropic and anisotropic parts of the chemical shift interaction, two distinct computations have been performed for the spin-echo curve under the hypothetical conditions that only the isotropic chemical shifts (dashed line in figure 2a), or only the $^1$H chemical shift anisotropies (dotted line in figure 2a), are active during the spin-echo evolution time. As can be seen comparing these two spin-echo curves, their effect upon the ideal behavior are of comparable magnitudes, and manifests themselves by a strong reduction of the decoupling efficiency under RS-HEPT. Obviously, the thin continuous line in figure 2a represents the combined contribution of both, isotropic, and anisotropic, $^1$H chemical shift interactions, and this results in a further reduction of the practical decoupling performances.

Finally, practical solutions for reducing these detrimental effects of the $^1$H chemical shift interactions are analyzed in the reminder of the present work based again on numerical simulations. First, we have studied the decoupling efficiency, measured through the decay rate of the acquired $^{15}$N spin-echo curve, by using a phase alternation in the RS-HEPT sequence, by which the relative phases of consecutive pulses are shifted by an amount, $\Delta \phi$. Considering first the case of the spin-echo recorded at 22.5 kHz MAS frequency that was discussed above, the best performance was obtained for $\Delta \phi = 30^\circ$. The corresponding spin-echo decay curve is represented in figure 2b by dashed line. For comparison, also the spin-echo curve obtained without phase-alternation is shown (thin line). Next, the effect of increasing spinning frequency to 50 kHz was investigated. This has become possible in practice due to the recent progresses in the ultra-fast MAS probehead technology. The spin-echo curve obtained at 50 kHz MAS frequency is drawn in figure 2b with dotted line in the case when no phase-shifting of the RS-HEPT pulses was applied. A further improvement in the decoupling efficiency is obtained in this case compared to the characteristic efficiency under 22.5 kHz spinning frequency and phase-shifting. Interestingly, if the both changes are considered simultaneously, i.e., 50 kHz MAS frequency and phase-shifted pulses by $\Delta \phi = 30^\circ$ in the RS-HEPT sequence, one obtains a significant increase in the spin-echo decay time, of up to 800 ms (thick line in figure 2b), which is much larger than the value that would be obtained by simply adding the separate effects. This result is very promising with respect to improving the practical performances of the proposed SS-NMR spin-echo technique. For instance, the result obtained in the present study indicates the possibility of measuring very weak hydrogen-bond mediated $J$ coupling constants, of less than 1 Hz, and thus identify more subtle structural details in supramolecular aggregates of current research interest.
4. Conclusions
To further increase the accuracy of the spin-echo experiment, a numerical analysis has been performed with respect to improving the decoupling efficiency by RS-HEPT. It was found that using higher sample spinning frequencies (up to 50 kHz) combined with a suitable phase modulation of the $\pi$-pulses in the RS-HEPT sequence, could lead to almost a fourfold improvement in the characteristic spin-echo decay time. This is expected to lead to a substantial increase in the practical performances of the $^{15}$N spin-echo SS-NMR experiment.

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References
[1] Dingley A J and Grzesiek S 1998, J. Am. Chem. Soc. 120 8293
[2] Lesage A, Bardet M and Emsley L 1999 J. Am. Chem. Soc. 121 10987
[3] Pham T N, Griffin J M, Masiero S, Lena S, Gottareli G, Hodgkinson P, Filip C and Brown S P 2007 Phys. Chem. Chem. Phys. 9 3416
[4] Griffin J M, Tripon C, Samoson A, Filip C and Brown S P 2007 Magn. Reson. Chem. 45 S198
[5] Filip X, Tripon C and Filip C 2005 J. Magn. Reson. 176 239
[6] Veshtort M and Griffin R G 2006 J. Magn. Reson. 178 248.