NMR Spectroscopy

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Highly Efficient, Water-Soluble Polarizing Agents for Dynamic Nuclear Polarization at High Frequency**

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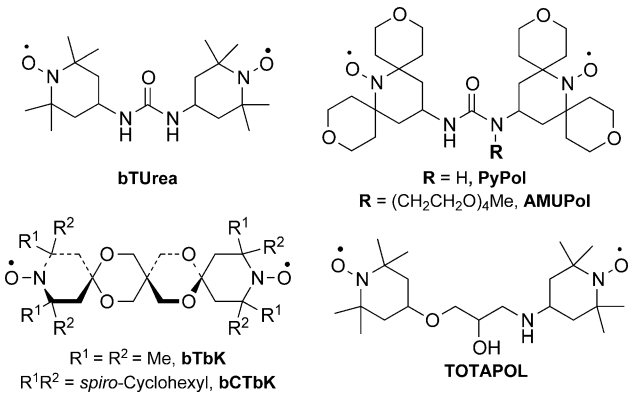
Dynamic nuclear polarization coupled to solid-state NMR spectroscopy (DNP/ssNMR) is emerging as a very powerful technique to overcome the limitations resulting from the intrinsic poor sensitivity of NMR spectroscopy. The DNP method exploits the microwave-driven transfer of polarization from the electron spin of a paramagnetic center (polarizing agent) to surrounding nuclei and has been demonstrated to produce uniformly polarized samples. First discovered in the 1950s with low magnetic fields,[1] high-field DNP NMR experiments are now possible with the introduction of gyrotron sources[2] which are capable of delivering high-power high-frequency microwaves (MW) up to 527 GHz and NMR probes that operate at approximately 100 K. DNP experiments have also been performed at up to 263 GHz in the 4–80 K temperature range with lower power sources.[3] DNP experiments that achieve even a fraction of the theoretical maximum sensitivity enhancement (658 for ^{1}H, 2617 for ^{13}C) can allow the low sensitivity of NMR spectroscopy to be overcome and to achieve breakthroughs in the use of the technique to investigate previously inaccessible systems. In this respect, in the past few years DNP-enhanced solid-state NMR spectroscopy under magic-angle spinning (MAS) conditions has made great progress, and DNP signal enhancement factors of up to approximately 50 and up to around 100 have been reported (at 9.4 T, 263 GHz, 100 K) for biological solids[4] and hybrid or inorganic materials,[5] respectively.

Initial high-field DNP experiments were performed with cross-effect (CE) polarization transfer mechanism and solutions doped with high concentrations of mono-radical nitroxide species (40 to 100 mM), such as TEMPO.[6] In 2004, Hu[7] et al. using a series of bis-TEMPO-*n*-ethyleneglycol (BT*n*E) dinitroxides demonstrated that biradical polarizing agents yielded significant improvements in the CE polarization efficiency. Then, Song[8] et al. introduced the water-soluble biradical TOTAPOL (Scheme 1) which has good solubility and stability in aqueous media containing glycerol and salts at concentrations typically found in protein solutions. Outstanding results were obtained with TOTAPOL, which is today the most commonly employed exogenous polarizing agent in DNP/ssNMR applications to biological systems.[4] TOTAPOL was shown, however, to have a relatively flexible structure[9] which is detrimental to the frequency-matching conditions required for an efficient CE mechanism, and the DNP enhancements observed with TOTAPOL ($\varepsilon(^{1}H) < 80$ on standard samples at 9.4 T, 263 GHz EPR, 100 K) remain far from the theoretical limit, and decrease rapidly when $T > 100$ K.[10] In addition, since CE DNP enhancements scale relatively to the applied magnetic field as approximately B_0^{-1}, TOTAPOL is expected to yield even lower signal enhancements[11] on high-field DNP/SSNMR spectrometers operating at 14.1 ($\omega_{0S} = 395$ GHz) or 18.8 T ($\omega_{0S} = 527$ GHz), which are required for optimal resolution.



Scheme 1. Structures of bTUrea, PyPol, AMUPol, bTbK, bCTbK, and TOTAPOL.

In 2009, in collaboration with Griffin's group (MIT), we achieved another step forward with the introduction of bTbK[12] (Scheme 1), a biradical in which the two TEMPO moieties are linked by a rigid tether inducing a quasi-orthogonal relative orientation of the TEMPOs g tensors. For a glassy solution of bTbK, the ^{1}H frequency-matching

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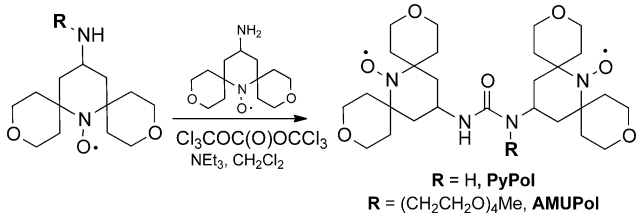
condition for the CE mechanism is closely approached, and it was demonstrated that bTbK provides DNP enhancement factors 1.4 times higher than TOTAPOL under similar conditions. Recently, in collaboration with Emsley's group (ENS Lyon) we developed a series of bTbK analogues with increased molecular weights and longer relaxation times T_{1e} and T_{2e}, and we showed[5e] that in organic solvent glasses the DNP enhancements obtained with bCTbK are 3 to 4 times higher than those obtained with bTbK. However, bTbK and bCTbK are insoluble in glycerol/water mixtures and only sparingly soluble in DMSO/water, hence their use is almost limited to DNP/ssNMR applications carried out in organic solvents.[5d,e,i]

Two bTbK derivatives (bTbtk[13] and bTbtk-py[14]) showing better water solubility and retaining the desirable orientation of bTbK have been reported. However, the increase in the DNP signal enhancements observed with these polarizing agents compared to TOTAPOL remains relatively modest ($\varepsilon_{bTbtk}/\varepsilon_{TOTAPOL} \approx 1.1$ and $\varepsilon_{bTbtk\text{-}py}/\varepsilon_{TOTAPOL} \approx 1.2$). In the course of our search of new CE DNP polarizing agents[5e,12,13,15] we investigated the influence of various parameters (e–e dipolar interaction, g tensor orientations, rigidity of the molecule, electron spin relaxation times) on the DNP efficiency. Among the different dinitroxides that we examined, bTUrea (Scheme 1) retained our attention. bTUrea, first described in 1965,[16] has been tested as DNP polarizing agent on a model system by Hu[9] et al. in DMSO/water 60/40. It was described to be three times more efficient than TEMPO but less efficient than TOTAPOL, poor solubility was noted, and its use in DNP/ssNMR applications has not been reported. We reexamined the performance of bTUrea and TOTAPOL in DMSO/water and glycerol/water solutions and noticed that at the same concentration bTUrea was more efficient than TOTAPOL. We then synthesized a series of bTUrea derivatives with the goals of improving solubility in aqueous solution and also further enhancing DNP efficiency. We report herein on two new biradical polarizing agents PyPol and AMUPol (Scheme 1) which show high water solubility and greatly enhanced DNP efficiency.

The general method to prepare PyPol and AMUPol is shown in Scheme 2, and details on the synthesis are given in the Supporting Information. DNP signal enhancements were measured with AMUPol and PyPol with a proline standard sample in water/glycerol at 263 and 395 GHz (400 and 600 MHz ^{1}H frequency respectively), 97 K sample temperature. Further instrumentation and experimental details are given in the Supporting Information. DNP signal enhancements $\varepsilon(^1\text{H})$, polarization build-up times (T_{DNP}), and $T_1(^1\text{H})$ values are shown in Table 1. A very strong signal enhancement is observed (Figure 1) with these two new bTUrea derivatives, with ε of 235 and 207 at 263 GHz and 128 (both) at 395 GHz for AMUPol and PyPol, respectively. The shorter T_{DNP} for AMUPol allows for faster signal averaging making it the more favorable biradical for DNP/ssNMR applications.



Scheme 2. Synthesis of PyPol and AMUPol.

Table 1: Values of ε (^{1}H), polarization build-up time (T_{DNP}), T_1 (^{1}H), T_{1e} and T_{2e} for bTUrea, TOTAPOL, PyPol and AMUPol.

Compound	$\varepsilon(^1\text{H})$[a]	T_{DNP}[b] [s]	$T_1(^1\text{H})$ [s]	T_{1e} [μs][e]	T_{2e} [μs][e]	MW[d] [g mol^{-1}]
bTUrea[c]	62 ± 3	16.5	17.3	306	1.8	368
TOTAPOL	70 ± 3	5.6	7.1	342	3.2	399
	(30±2)	*(7.7)*	*(9.6)*			
PyPol	207 ± 7	5.7	6.5	470	12	536
	(128±9)	*(8.7)*	*(9.5)*			
AMUPol	235 ± 5	3.5	3.3	460	10	726
	(128±8)	*(4.9)*	*(5.1)*			

[a] DNP measurements in [D_8]glycerol/D_2O/H_2O (60/30/10 volume ratio) at 263 and 395 (in parenthesis) GHz (400 and 600 MHz ^{1}H NMR respectively), 97 K, 8 kHz MAS, 10 mM biradical concentration (except for bTUrea, less than 3 mM) and 0.25 M U-^{13}C-^{15}N proline. $\varepsilon(^1\text{H})$ measured by ^{13}C CP-MAS. [b] DNP polarization build-up time measured with standard saturation recovery experiment with microwave irradiation. [c] bTUrea sample was prepared without proline and DNP enhancement measured on glycerol ^{13}C natural abundance signal. [d] Molecular weight. [e] At 9 GHz EPR frequency and 97 K with 0.1 mM biradical concentration.

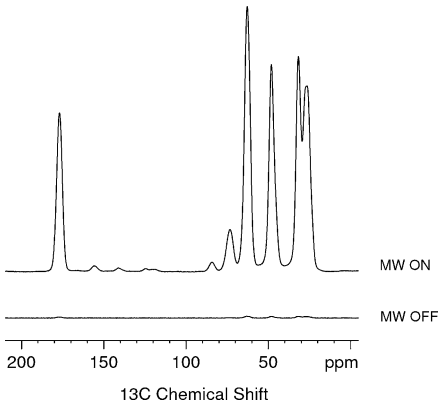


Figure 1. ^{13}C CPMAS spectra of 0.25 M U-^{13}C-^{15}N proline with 10 mM AMUPol in [D_8]glycerol/D_2O/H_2O (60/30/10 volume ratio) with (top trace) and without (bottom trace) microwave irradiation at 14 kHz MAS, 97 K sample temperature, 8 scans, 1 dummy scan, 6.4 s recycle delay, 9 W microwave power at end of probe waveguide.

A TOTAPOL sample was prepared and measured under identical conditions for comparison and showed ε of 70 and 30 at 263 and 395 GHz, respectively. The DNP efficiency for AMUPol was investigated as a function of sample temperature (Figure 2) and MAS spinning frequency (Supporting Information). The temperature dependence of AMUPol is almost linear from 100 K to over 180 K ($\varepsilon = 83$ at 160 K) whereas TOTAPOL shows a much steeper temperature dependence ($\varepsilon < 10$ at 160 K), especially below 130–140 K.[10] This reduced temperature dependence allows for DNP experiments with AMUPol at sample temperatures that are significantly higher than typical DNP experiments and up

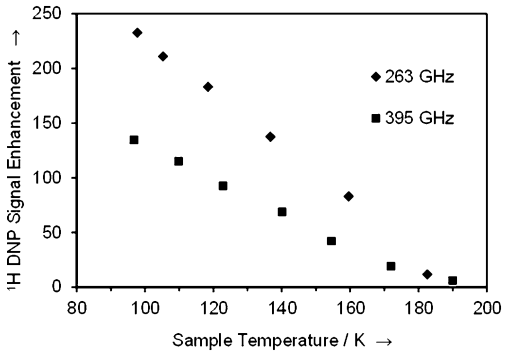


Figure 2. Temperature dependence of ^{1}H DNP signal enhancement for AMUPol measured at 263 and 395 GHz. ^{1}H DNP signal enhancement measured by ^{13}C CP-MAS experiment on 0.25 M U-^{13}C-^{15}N proline with 10 mM AMUPol in [D_8]glycerol/D_2O/H_2O (60/30/10 volume ratio) with and without microwave irradiation for each data point. Microwave power level optimized for each data point. 8 scans, 1 dummy scans, recycle delay = $1.3 \times T_{DNP}$ for each data point. Sample temperature calibrated with KBr T_1 measurements[21] in same rotor and active volume as DNP sample.

to sample temperatures where spectral resolution may be improved. The MAS dependence of the DNP signal enhancement (Supporting Information) is relatively flat up to 14 kHz spinning frequency, again in contrast to published data for TOTAPOL which shows a noticeable drop in DNP efficiency at MAS frequency above 3 kHz.[10a,c] This result is highly encouraging for the development of faster spinning MAS DNP probes and applications. Electron T_{1e} and T_{2e} relaxation times were measured at 9 GHz EPR frequency and 97 K (Table 1), and their temperature dependence as well as that of T_1(^{1}H), are also shown in Supporting Information.

Many factors influence the efficiency of a dinitroxide CE DNP polarizing agent, to name but a few: the e–e dipole interaction, the electron relaxation times, the relative orientation of the two g tensors. Furthermore the dinitroxide must have a good solubility in the solvent system used for the experiment and must be compatible with the formation of a good glass at low temperatures. AMUPol and PyPol exhibit good solubility (up to 30 mM) in glycerol/water mixtures (60/40 v/v), and good frozen glasses are formed ($T < 220$ K). It has been shown that lengthening T_{1e} and T_{2e} allows for better saturation of the electron spins and results in higher CE efficiency.[5e,17] For nitroxides in glassy organic solvents around 100 K, on one hand it has been shown that T_{1e} depends on the molecular weight[18] of the radicals: the heavier the radical, the slower the relaxation, on the other hand, replacing the rotating methyl groups of TEMPO moieties with spirocyclohexyl groups lengthen T_{2e}.[5e,19]

Thus, AMUPol and PyPol are expected to have electron relaxation times, significantly longer than those of bTUrea and TOTAPOL. This assumption is confirmed with the EPR measurements showing especially significant increase in T_{2e} for AMUPol and PyPol (Table 1). Preliminary DFT calculations (Supporting Information) show that for the major conformer of bTUrea, PyPol and AMUPol, the relative orientation of the TEMPO moieties and the e–e dipole coupling (estimated from the point dipole approximation) are similar. Furthermore for the bTUrea derivatives the magnitude of the e–e dipole coupling (ca. 34 MHz) is larger than for TOTAPOL (ca. 23 MHz). Therefore, we conclude that the much higher efficiency of PyPol and AMUPol in comparison with TOTAPOL results from their longer electron relaxation times and larger e–e dipole coupling.[10c,20] In addition, owing to the presence of the urea linker, the molecular structure of bTUrea and its derivatives is much more rigid than that of TOTAPOL[9] and more molecules are expected to closely approach the CE frequency matching.

DNP on and off measurements were performed with $1.3 \times T_{DNP}$ as the recycle delay which provides optimal sensitivity in a multi-scan experiment. Since T_{DNP} is not exactly equal to T_1, this can have a slight effect on the reported DNP signal enhancement but best reflects to conditions under which a DNP-enhanced solid-state NMR experiment would be run.

In conclusion, our experiments demonstrate the superior performance at 263 and 395 GHz of the biradicals PyPol and AMUPol when compared to TOTAPOL. The enhancement factors obtained with AMUPol are about 3.5 times (at 263 GHz) and 4 times (at 395 GHz) larger than for TOTAPOL under identical experimental conditions. These new polarizing agents push back the barriers to the broadband applicability of DNP/ssNMR to increasingly complex biological systems or water soluble materials, currently not amenable to NMR characterization.

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[1] a) A. W. Overhauser, *Phys. Rev.* **1953**, *92*, 411–415; b) T. R. Carver, C. P. Slichter, *Phys. Rev.* **1953**, *92*, 212–213; c) T. R. Carver, C. P. Slichter, *Phys. Rev.* **1956**, *102*, 975–980.

[2] L. R. Becerra, G. J. Gerfen, R. J. Temkin, D. J. Singel, R. G. Griffin, *Phys. Rev. Lett.* **1993**, *71*, 3561–3564.

[3] a) K. R. Thurber, W.-M. Yau, R. Tycko, *J. Magn. Reson.* **2010**, *204*, 303–313; b) B. D. Armstrong, D. T. Edwards, R. J. Wylde, S. A. Walker, S. Han, *Phys. Chem. Chem. Phys.* **2010**, *12*, 5920–5926.

[4] a) D. A. Hall, D. C. Maus, G. J. Gerfen, S. J. Inati, L. R. Becerra, F. W. Dahlquist, R. G. Griffin, *Science* **1997**, *276*, 930–932; b) M. Rosay, A.-C. Zeri, N. S. Astrof, S. J. Opella, J. Herzfeld, R. G. Griffin, *J. Am. Chem. Soc.* **2001**, *123*, 1010–1011; c) V. S. Bajaj, M. L. Mak-Jurkauskas, M. Belenky, J. Herzfeld, R. G. Griffin, *Proc. Natl. Acad. Sci. USA* **2009**, *106*, 9244–9249; d) E. Salnikov, M. Rosay, S. Pawsey, O. Ouari, P. Tordo, B. Bechinger, *J. Am. Chem. Soc.* **2010**, *132*, 5940–5941; e) Ü. Akbey, W. T. Franks, A. Linden, S. Lange, R. G. Griffin, B.-J. van Rossum, H. Oschkinat, *Angew. Chem.* **2010**, *122*, 7971–7974; *Angew. Chem. Int. Ed.* **2010**, *49*, 7803–7806; f) P. C. A. van der Wel, K.-N. Hu, J. Lewandowski, R. G. Griffin, *J. Am. Chem. Soc.* **2006**, *128*, 10840–10846; g) G. T. Debelouchina, M. J. Bayro, P. C. A. van der Wel, M. A. Caporini, A. B. Barnes, M. Rosay, W. E. Maas, R. G. Griffin, *Phys. Chem. Chem. Phys.* **2010**, *12*, 5911–5919; h) M. J. Bayro, G. T. Debelouchina, M. T. Eddy, N. R. Birkett, C. E. MacPhee, M. Rosay, W. E. Maas, C. M. Dobson, R. G. Griffin, *J. Am. Chem. Soc.* **2011**, *133*, 13967–13974; i) M. Renault, S. Pawsey, M. P. Bos, E. J. Koers, D. Nand, R.

Tommassen-van Boxtel, M. Rosay, J. Tommassen, W. E. Maas, M. Baldus, *Angew. Chem.* **2012**, *124*, 3053 – 3056; *Angew. Chem. Int. Ed.* **2012**, *51*, 2998 – 3001; j) H. Takahashi, I. Ayala, M. Bardet, G. De Paëpe, J.-P. Simorre, S. Hediger, *J. Am. Chem. Soc.* **2013**, *135*, 5105 – 5110; k) L. Reggie, J. J. Lopez, I. Collinson, C. Glaubitz, M. Lorch, *J. Am. Chem. Soc.* **2011**, *133*, 19084 – 19086; l) E. Ravera, B. Corzilius, V. K. Michaelis, C. Rosa, R. G. Griffin, C. Luchinat, I. Bertini, *J. Am. Chem. Soc.* **2013**, *135*, 1641 – 1644.

[5] a) A. Lesage, M. Lelli, D. Gajan, M. A. Caporini, V. Vitzthum, P. Miéville, J. Alauzun, A. Roussey, C. Thieuleux, A. Mehdi, et al., *J. Am. Chem. Soc.* **2010**, *132*, 15459 – 15461; b) M. Lelli, D. Gajan, A. Lesage, M. A. Caporini, V. Vitzthum, P. Miéville, F. Héroguel, F. Rascón, A. Roussey, C. Thieuleux, et al., *J. Am. Chem. Soc.* **2011**, *133*, 2104 – 2107; c) O. Lafon, M. Rosay, F. Aussenac, X. Lu, J. Trébosc, O. Cristini, C. Kinowski, N. Touati, H. Vezin, J.-P. Amoureux, *Angew. Chem.* **2011**, *123*, 8517 – 8520; *Angew. Chem. Int. Ed.* **2011**, *50*, 8367 – 8370; d) A. J. Rossini, A. Zagdoun, M. Lelli, J. Canivet, S. Aguado, O. Ouari, P. Tordo, M. Rosay, W. E. Maas, C. Copéret, et al., *Angew. Chem.* **2012**, *124*, 127 – 131; e) A. Zagdoun, G. Casano, O. Ouari, G. Lapadula, A. J. Rossini, M. Lelli, M. Baffert, D. Gajan, L. Veyre, W. E. Maas, M. Rosay, R. T. Weber, C. Thieuleux, C. Coperet, A. Lesage, P. Tordo, L. Emsley, *J. Am. Chem. Soc.* **2012**, *134*, 2284 – 2291; f) A. Zagdoun, A. J. Rossini, M. P. Conley, W. R. Grüning, M. Schwarzwälder, M. Lelli, W. T. Franks, H. Oschkinat, C. Copéret, L. Emsley, et al., *Angew. Chem.* **2013**, *125*, 1260 – 1263; *Angew. Chem. Int. Ed.* **2013**, *52*, 1222 – 1225; g) A. J. Rossini, A. Zagdoun, F. Hegner, M. Schwarzwälder, D. Gajan, C. Copéret, A. Lesage, L. Emsley, *J. Am. Chem. Soc.* **2012**, *134*, 16899 – 16908; h) D. Lee, H. Takahashi, A. S. L. Thankamony, J.-P. Dacquin, M. Bardet, O. Lafon, G. D. Paëpe, *J. Am. Chem. Soc.* **2012**, *134*, 18491 – 18494; i) A. Zagdoun, A. J. Rossini, D. Gajan, A. Bourdolle, O. Ouari, M. Rosay, W. E. Maas, P. Tordo, M. Lelli, L. Emsley, et al., *Chem. Commun.* **2012**, *48*, 654 – 656.

[6] a) D. S. Wollan, *Phys. Rev. B* **1976**, *13*, 3671 – 3685; b) D. S. Wollan, *Phys. Rev. B* **1976**, *13*, 3686 – 3696.

[7] K.-N. Hu, H. Yu, T. M. Swager, R. G. Griffin, *J. Am. Chem. Soc.* **2004**, *126*, 10844 – 10845.

[8] C. Song, K.-N. Hu, C.-G. Joo, T. M. Swager, R. G. Griffin, *J. Am. Chem. Soc.* **2006**, *128*, 11385 – 11390.

[9] K.-N. Hu, C. Song, H. Yu, T. M. Swager, R. G. Griffin, *J. Chem. Phys.* **2008**, *128*, 052302.

[10] a) M. Rosay, L. Tometich, S. Pawsey, R. Bader, R. Schauwecker, M. Blank, P. M. Borchard, S. R. Cauffman, K. L. Felch, R. T. Weber, R. J. Temkin, R. G. Griffin, W. E. Mas, *Phys. Chem. Chem. Phys.* **2010**, *12*, 5850 – 5860; b) P. Mieville, V. Vitzthum, M. A. Caporini, S. Jannin, S. Gerber-Lemaire, G. Bodenhausen, *Magn. Reson. Chem.* **2011**, *49*, 689 – 692; c) F. Mentink-Vigier, U. Akbey, Y. Hovav, S. Vega, H. Oschkinat, A. Feintuch, *J. Magn. Reson.* **2012**, *224*, 13 – 21.

[11] a) A. B. Barnes, E. Markhasin, E. Daviso, V. K. Michaelis, E. A. Nanni, S. K. Jawla, E. L. Mena, R. DeRocher, A. Thakkar, P. P. Woskov, et al., *J. Magn. Reson.* **2012**, *224*, 1 – 7; b) Y. Matsuki, K. Ueda, T. Idehara, R. Ikeda, I. Ogawa, S. Nakamura, M. Toda, T. Anai, T. Fujiwara, *J. Magn. Reson.* **2012**, *225*, 1 – 9.

[12] a) Y. Matsuki, T. Maly, O. Ouari, H. Karoui, F. Le Moigne, E. Rizzato, S. Lyubenova, J. Herzfeld, T. Prisner, P. Tordo, R. G. Griffin, *Angew. Chem.* **2009**, *121*, 5096 – 5100; *Angew. Chem. Int. Ed.* **2009**, *48*, 4996 – 5000; b) M. Gafurov, S. Lyubenova, V. Denysenkov, O. Ouari, H. Karoui, F. L. Moigne, P. Tordo, T. Prisner, *Appl. Magn. Reson.* **2010**, *37*, 505 – 514.

[13] E. L. Dane, B. Corzilius, E. Rizzato, P. Stocker, T. Maly, A. A. Smith, R. G. Griffin, O. Ouari, P. Tordo, T. M. Swager, *J. Org. Chem.* **2012**, *77*, 1789 – 1797.

[14] M. K. Kiesewetter, B. Corzilius, A. A. Smith, R. G. Griffin, T. M. Swager, *J. Am. Chem. Soc.* **2012**, *134*, 4537 – 4540.

[15] a) C. Ysacco, E. Rizzato, M.-A. Virolleaud, H. Karoui, A. Rockenbauer, F. L. Moigne, D. Siri, O. Ouari, R. G. Griffin, P. Tordo, *Phys. Chem. Chem. Phys.* **2010**, *12*, 5841 – 5845; b) C. Ysacco, H. Karoui, G. Casano, F. L. Moigne, S. Combes, A. Rockenbauer, M. Rosay, W. Maas, O. Ouari, P. Tordo, *Appl. Magn. Reson.* **2012**, *43*, 251 – 261.

[16] E. G. Rozantsev, V. A. Golubev, M. V. Neiman, Y. V. Kokhanov, *Izv. Akad. Nauk Arm. SSR Khim. Nauki* **1965**, 572 – 573.

[17] L. B. Casabianca, A. I. Shames, A. M. Panich, O. Shenderova, L. Frydman, *J. Phys. Chem. C* **2011**, *115*, 19041 – 19048.

[18] H. Sato, V. Kathirvelu, A. Fielding, J. P. Blinco, A. S. Micallef, S. E. Bottle, S. S. Eaton, G. R. Eaton, *Mol. Phys.* **2007**, *105*, 2137 – 2151.

[19] a) V. Kathirvelu, C. Smith, C. Parks, M. Abdul Mannan, Y. Miura, K. Takeshita, S. S. Eaton, G. R. Eaton, *Chem. Commun.* **2009**, 454 – 456; b) A. Rajca, V. Kathirvelu, S. K. Roy, M. Pink, S. Rajca, S. Sarkar, S. S. Eaton, G. R. Eaton, *Chem. Eur. J.* **2010**, *16*, 5778 – 5782.

[20] K. R. Thurber, R. Tycko, *J. Chem. Phys.* **2012**, *137*, 084508.

[21] K. R. Thurber, R. Tycko, *J. Magn. Reson.* **2009**, *196*, 84 – 87.

