ChemComm RSCPublishing

SUPPLEMENTARY INFORMATION

Hydrophobic Radicals Embedded in Neutral Surfactants for Dynamic Nuclear Polarization of Aqueous Environments at 9.4 Tesla.

Moreno Lelli,[a] Aaron J. Rossini,[a] Gilles Casano,[b] Olivier Ouari,[b] Paul Tordo,[b] Anne Lesage,[a] and Lyndon Emsley[a]

[a] Centre de RMN à Trés Hauts Champs, Institut de Sciences Analytiques (CNRS/ENS Lyon/UCB Lyon 1), Université de Lyon, 69100 Villeurbanne, France.
[b] Aix-Marseille Universite, CNRS, ICR UMR 7273, 13397 Marseille, France.

*To whom correspondence should be addressed. E-mail: Lyndon.Emsley@ens-lyon.fr

This PDF file includes:

SUPPLEMENTARY INFORMATION

Materials and Methods.

General Information. The surfactants Tween-80®, Brij® C10 (Brij-56), IGEPAL® CO-630, and sodium dodecyl sulphate (SDS) were purchased from Sigma-Aldrich. L-Alanine was purchased from ACROS Organics, α-D-Glucose was purchased from Sigma-Aldrich, Uniformly ^{13}C-labelled L-proline was purchased from Cambridge Isotope Laboratories. The radical BDPA was purchased from Sigma-Aldrich, while bTbK,[1] bCTbK,[2] and TEKPol[3] were prepared following the synthesis already reported in the literature.

DNP-NMR Methods. All DNP-enhanced NMR experiments were performed using a solid-state 400 MHz DNP-NMR spectrometer designed by Bruker-Biospin[4]. This system consists of a wide-bore 9.4 T magnet ($\omega_H/(2\pi)$ = 400.3 MHz, $\omega_C/(2\pi)$ = 100.7 MHz) with a Bruker Avance III spectrometer console, and is equipped with a double/triple resonance 3.2 mm low-temperature CP-MAS probe. DNP is achieved by irradiating the sample with high-power microwaves at a frequency of 263 GHz that are generated by a gyrotron and are delivered to the sample by a corrugated wave-guide (~5 W of power reaching the sample). The gyrotron operates continuously during the DNP-enhanced experiments (stability of better than ±1%). Sapphire rotors (with ZrO_2 caps) were used for optimal microwave penetration. Spinning frequencies were regulated to 8.0 kHz ± 2 Hz. The sample temperatures were ≈ 105 K. The chemical shifts are referenced to TMS at 0 ppm.

^{1}H 1D direct excitation experiments were acquired with a rotor synchronized spin echo in order to suppress the background signals. The pulse sequence was: $\pi/2 — \tau — \pi — \tau —$ acquisition, $\pi/2$ and π hard pulses were calibrated at 2.5 μs and 5.0 μs (100 kHz), respectively. The τ echo-delays were set to 1 rotor period. Experiments were acquired over a spectral window of 200 kHz, with an acquisition time of 5.12 ms, and a recycle delay of 3.0 s. Standard cross-polarization (CP) was used for the acquisition of 1D ^{13}C spectra, the recycle delay between scans was 2.0 s in all experiments. The ^{1}H $\pi/2$ pulse length was 2.5 μs

(ν_1 = 100 kHz). A linear amplitude ramp (from 100% to 50% of the nominal RF field strength) was used for the ^{1}H channel, with a 2.0 ms contact time (τ_{CP}), and a nominal RF-field amplitude (ν_1) of 88.4 kHz for ^{1}H and 58.1 kHz for ^{13}C. SPINAL-64[5] proton decoupling was applied during the acquisition of the ^{13}C signal with ν_1 = 100 kHz. The 1D fid ^{13}C acquisition time was 25.3 ms for 1024 complex points. 1D spectra were processed using exponential window functions with a linebroadening of 200 Hz for ^{13}C and 400 Hz for ^{1}H spectra.

Solution-NMR Methods. Liquid state NMR spectra were acquired on a 600 MHz Bruker instrument (14.1 T) equipped with a ^{1}H/^{13}C/^{15}N cryoprobe and an Avance III Bruker console. The samples were prepared in aqueous environment using H_2O/D_2O 90/10 as solvent. The temperature was stabilized to 298 K.

The ^{1}H 1D spectra were acquired with an excitation sculpting water suppression sequence,[6] with selective π pulses on water of 2000 μs, non-selective π pulses of 20.4 μs and a short $\pi/2$ excitation pulse of 1.0 μs. Each spectrum was recorded accumulating 128 scans with 852 ms of acquisition time and 1.15 s of recycle delay. The spectra were processed with an exponential windows function of 0.3 Hz.

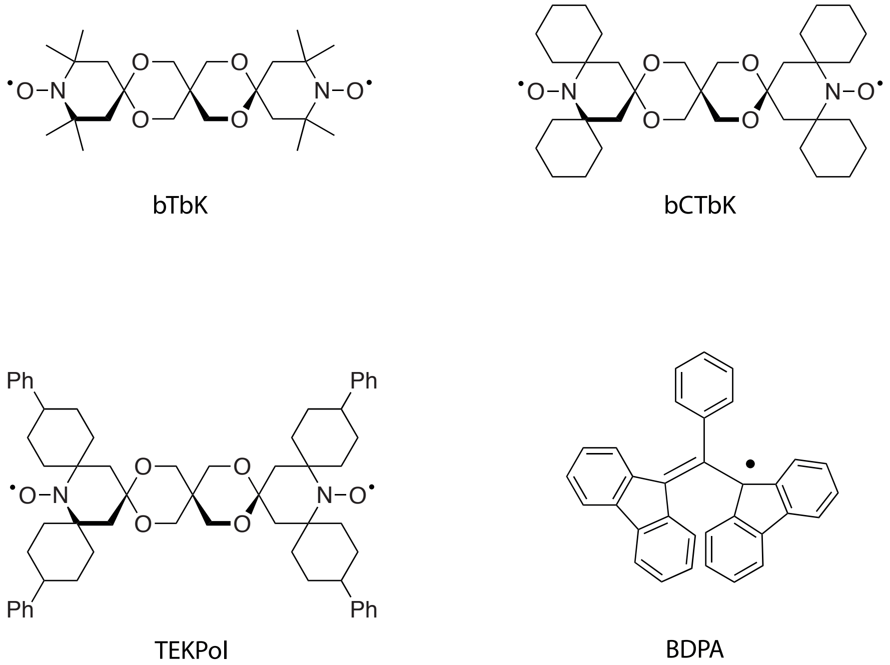
The ^{13}C 1D spectra were acquired with a direct excitation double-echo experiment to suppress the probe background and to remove the baseline distortion. The sequence was $\pi/2$ — τ — π — 2τ — π — τ — acquisition: the $\pi/2$ and π hard pulses were calibrated at 12.0 μs and 24.0 μs, respectively; the τ echo-delays were optimized to 50 μs each. Waltz-16 ^{1}H-decoupling at 3.12 kHz was applied during acquisition and a weaker ^{1}H-decoupling at ~2 kHz was kept during the recycle delay. Each experiment was acquired accumulating 128 scans with 865 ms of acquisition (64 k real points) and 2.0 s of recycle delay. The spectra were processed with an exponential windows function of 0.3 Hz.

Sample Preparation.

bTbK dissolved in sodium dodecyl sulphate aqueous solutions. A solution of 100 mM sodium dodecyl sulphate (SDS) and 50 mM α-D-Glucose solution was prepared in H_2O/D_2O 90/10. The dissolution of bTbK was performed by directly stirring a weighted amount of bTbK powder with the previously prepared 100 mM SDS solution until complete dissolution.

bTbK dissolved in glycerol/water/SDS solution. A solution of 400 mM sodium dodecyl sulphate (SDS) and 50 mM L-alanine was prepared by dissolving the weighted powder in a d_8-Glycerol:D_2O:H_2O 60:30:10 mixture. bTbK was than dissolved by directly stirring the radical powder in the above prepared glycerol/water/SDS solution by gently heating the mixture at 40-50 °C.

Radicals dissolved in glycerol/water/surfactant solution. The used radical (bTbK, bCTbK, TEKPol, or BDPA) was first dissolved in the pure surfactant (Tween-80®, Brij® 56, IGEPAL® CO-630) by stirring the radical powder in the melted amphiphile gently heated at around 40-50 °C. Radical solution with a concentration of ~50 mM can thus be obtained in such a way. Then this solution was diluted up to the desired radical concentration by addition of the d_8-Glycerol:D_2O:H_2O 60:30:10 mixture. Uniformly ^{13}C-labelled L-proline was added to the solution in order to have a final concentration of 12 mM in proline to monitor the ^{13}C enhancement in CP experiments. For the case of Tween-80 and a radical concentration of 12 mM, a final surfactant concentration of ~190 mM was used.



bTbK

bCTbK

TEKPol

BDPA

Scheme S1. Molecular structures of the radicals.

SUPPLEMENTARY INFORMATION

Table S1. DNP Enhancements observed for several water-insoluble radicals solubilized in different surfactant solutions. The ^{1}H (ε_{H}) and the ^{13}C (ε_{C_CP}) enhancements are measured comparing experiment with and without microwave irradiation in direct excitation and ^{1}H-^{13}C CP experiments, respectively (the conditions are reported in the Materials and Methods section).

Radical	Amphiphile	Conc. (mM)	ε_{H}	ε_{C_CP}
bTbK	SDS	10	17	17
bTbK	Tween-80	12	42	44
bCTbK	Tween-80	12	22	24
BDPA	Tween-80	14	4.8	-
TEKPol	Tween-80	7.5	58	59
TEKPol	Brij-56	10	8.9	10
TEKPol	IGEPAL CO-630	10	15	16

References

1. Y. Matsuki, T. Maly, O. Ouari, H. Karoui, F. Le Moigne, E. Rizzato, S. Lyubenova, J. Herzfeld, T. Prisner, P. Tordo, and R. G. Griffin, *Angew. Chem. Int. Ed.*, 2009, **48**, 4996–5000.
2. 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. Copéret, A. Lesage, P. Tordo, and L. Emsley, *J. Am. Chem. Soc.*, 2012, **134**, 2284–2291.
3. A. Zagdoun, G. Casano, O. Ouari, M. Schwarzwälder, A. J. Rossini, F. Aussenac, M. Yulikov, G. Jeschke, C. Copéret, A. Lesage, P. Tordo, and L. Emsley, *J. Am. Chem. Soc.*, 2013, **135**, 12790–12797.
4. 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, and W. E. Maas, *Phys. Chem. Chem. Phys.*, 2010, **12**, 5850–5860.
5. B. M. Fung, A. K. Khitrin, and K. Ermolaev, *J. Magn. Reson.*, 2000, **142**, 97–101.
6. T. L. Hwang and A. J. Shaka, *J. Magn. Reson., A*, 1995, **112**, 275–279.