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Long live the singlet state!

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ABSTRACT

The field of long-lived states in NMR is reviewed. The relationship of long-lived-state phenomena to those associated with spin isomerism is discussed. A brief overview is given of key developments in the field of long-lived states, including chemical symmetry-switching, the role of magnetic equivalence and magnetic inequivalence, long-lived coherences, hyperpolarized NMR involving long-lived states, quantum-rotor-induced polarization, and parahydrogen-induced hyperpolarization. Current application areas of long-lived states are reviewed, and a peer into the crystal ball reveals future developments in the field.

It is now 15 years since our group published two papers demonstrating that it is possible to store nuclear spin order in solution for times much longer than the magnetization relaxation time T_1[1,2]. Both papers involved molecular systems containing well-isolated pairs of proton spins-1/2 in chemically inequivalent sites, embedded in molecules lacking a high degree of symmetry. A long-lived mode of spin order called *singlet order*, which corresponds to the population difference between the nuclear singlet and triplet states, was populated. In the first case this was done by applying a radiofrequency pulse sequence to a sample in thermal equilibrium in high magnetic field, followed by transport of the sample out of the NMR magnet into a low magnetic field region [1]. In the second case the excitation of singlet order was accomplished directly in high magnetic field [2]. In both experiments, the singlet order was allowed to evolve while suppressing singlet-triplet transitions induced by the chemical shift difference between the protons. In the first paper, chemical shift suppression was achieved by removing the sample from the NMR magnet [1]. In the second paper, the sample was retained in the high-field NMR magnet, while the chemical shift difference was suppressed by applying a resonant radiofrequency field [2]. Singlet order relaxes more slowly than ordinary magnetization, since it is immune to the dominant intra-pair dipole-dipole relaxation mechanism. It was demonstrated that the nuclear singlet order relaxes with a time constant T_S which is many factors larger than T_1.

In retrospect, the most novel feature of these experiments was *not* the demonstration that certain types of nuclear spin order may persist for times much longer than T_1. Such phenomena have been long known in the context of nuclear spin isomerism, which occurs in small, highly symmetrical molecules with a high degree of rotational freedom, *ortho*- and *para*-hydrogen being the seminal examples [3]. In spin isomerism, the Pauli principle, which constrains the overall symmetry of allowed quantum states, entangles the spatial wavefunctions and spin states, such that nuclear energy levels are invested with energy differences associated with the *spatial* quantum mechanics of the rotating molecules. Such energies may be many orders of magnitude larger than nuclear Zeeman splittings. For example, the splitting between the rotational ground states of parahydrogen (nuclear spin singlet) and orthohydrogen (nuclear spin triplet) is ~ 3.6 THz, which is about 4 orders of magnitude larger than typical nuclear Zeeman energies. This large energy difference makes it possible to generate large population differences between the nuclear singlet and triplet state simply by bringing the hydrogen gas into contact with a cold magnetic material which catalyses the singlet-triplet transitions. The population difference between the singlet and triplet state persists for very long times (up to days or weeks) when the sample is removed from the catalyst, allowing the convenient production of hydrogen gas enriched in the nuclear singlet state (parahydrogen spin isomer). The inherent long-lived singlet order in *para*-enriched hydrogen was famously exploited by Weitekamp and co-workers who showed that it may be unlocked by a chemical hydrogenation reaction, generating greatly enhanced NMR signals [4–6]. Related effects occur in compounds such as 4-methylpyridine (γ-picoline), in which the methyl (CH_3) group has a very low rotational barrier at cryogenic temperatures. In this case the splitting

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between the spatial quantum levels is called a tunneling splitting and is of the order of ~ 125 GHz, as measured by neutron scattering [7]. As in the case of parahydrogen, a large population difference can be generated between the nuclear symmetry species by cooling the sample. When the temperature is raised, strongly enhanced NMR signals may be induced, both in the solid state [8], and in solution after dissolution [9–13].

The real novelty in our 2004 experiments was the demonstration that long-lived modes of nuclear spin order are not limited to systems with high degrees of symmetry and extreme rotational freedom and which display spin isomerism, but may be exhibited by rather "ordinary" molecules, and may be accessed using relatively conventional radiofrequency pulse sequences, supplemented in some cases by changes in the static magnetic field. It is now clear that such long-lived state phenomena are relatively widespread for clusters of coupled nuclear spins. Spin isomerism is a special case, in which the Pauli-principle entanglement of spin and spatial modes for freely rotating symmetrical moieties allows large amounts of order to be deposited in long-lived states by thermal means alone (Fig. 1).

In the years since 2004, our group and several others have researched intensively on long-lived states (LLS) and related phenomena. Some key developments are as follows:

- It was shown that *chemical symmetry switches* may be used to access long-lived spin order modes, as an alternative and supplement to magnetic field procedures. The local symmetry of the nuclear spin hamiltonian is broken by chemical and physical transformations of the sample, allowing access to the long-lived modes of spin order. This can provide access to long-lived states even in cases of complete magnetic equivalence [19–26].
- The use of magnetic inequivalence, as opposed to chemical equivalence, to access long-lived states in systems of more than 2 coupled spins-1/2 [27–29]. Most demonstrations of this principle have required that the degree of magnetic inequivalence (as defined in Ref. [30]) is very small. However, for some molecular geometries, a class of LLS persists even for strong magnetic inequivalence [30].
- The design and synthesis of molecular systems which support extremely long-lived states. For example, a $^{13}C_2$-labelled naphthalene derivative exhibits a T_S value exceeding 1 h in room-temperature solution, in low magnetic field [31]. Compounds called diazirines, which contain a three-membered ring formed by two nitrogen atoms and one carbon, are particularly promising [25,32].
- Demonstrations of long-lived coherences (LLCs) with very long lifetimes, and which in some cases oscillate coherently for tens of minutes [33–40].
- The use of long-lived states for the storage of hyperpolarized nuclear spin order [15,18,19,41–43]. Dynamic nuclear polarization (DNP) sometimes generates long-lived spin order directly, as a by-product of strong nuclear polarization [15–18,44].
- The development of methods for converting nuclear magnetization into long-lived spin order, and back again [45,46], including radiofrequency pulse sequences combined with field cycling [1,15,33,40,43,46–50], pulse sequences that operate in the weakly-coupled (strong inequivalence) regime [2,46,51,52], the M2S/S2M (magnetization-to-singlet and singlet-to-magnetization) pulse sequences and related methods for the near-equivalence regime [29,46,49,53–57], the simple and elegant SLIC (spin-lock-induced crossing) method [54,58–60], and the robust schemes which exploit adiabatic passage through avoided level crossings [39,40,61–63].
- The identification of long-lived states in rapidly rotating methyl groups, and their involvement in nuclear hyperpolarization phenomena observed when certain substances are warmed rapidly from the cryogenic solid state to ambient temperature [9–11].
- The study of heteronuclear long-lived states in ultralow-field NMR [64].
- The use of minuscule chemical shift differences to access long-lived singlet order, including the chemical shift differences between CH_2D protons in some chiral molecules [65–67], and mass-induced shift differences in compounds labelled with spin-0 isotopes such as ^{18}O[68].
- The elucidation of relaxation mechanisms for long-lived-states [31,69–76], including unusual mechanisms such as spin-rotation and spin-internal-motion [31,33,48,71,75], the antisymmetric components of the chemical shielding tensors [31,74], and scalar relaxation of the second kind [73,76].
- The use of long-lived states for the preparation of hard-to-access spin isomers, such as para-$^{15}N_2$[77].

In addition, promising proof-of-concept applications have been reported in several areas, including:

- Ligand binding and screening in biomolecular NMR [78–81].
- The study of diffusion, transport and chemical exchange [51,82–87].
- Quantum computation [88].
- Nuclear hyperpolarization using parahydrogen as a singlet-polarized source [20,23–25,32,54,57,77,89–96].

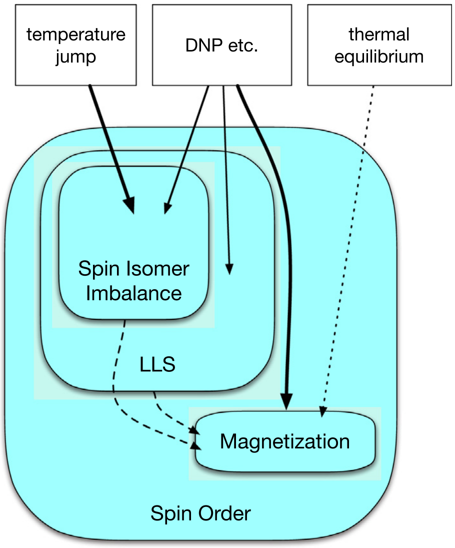

Fig. 1. The relationships between spin isomerism, long-lived states, hyperpolarization methods and thermal equilibrium. Magnetization and long-lived spin order (LLS) are both types of nuclear spin order. A population imbalance between spin isomers is a special type of long-lived spin order which may arise in symmetrical moieties with high rotational freedom, such that the Pauli principle constrains the allowed combinations of spin and spatial quantum states. Spin-isomer population imbalances may be generated by temperature changes, as in parahydrogen-induced hyperpolarization (PHIP) [4–6] and quantum-rotor-induced polarization (QRIP) [9–13]. Hyperpolarization techniques such as dynamic nuclear polarization (DNP) generate enhanced magnetization [14], but may also directly generate long-lived spin-order [15], including spin-isomer population imbalances [16–18]. Long-lived spin order may be converted into magnetization by radiofrequency pulse sequences or relaxation and/or chemical transformations (dashed arrows). Most NMR experiments exploit the small amount of thermal equilibrium magnetization in a strong magnetic field at non-cryogenic temperatures (dotted line).

- Hyperpolarized NMR sensors for molecular imaging and spectroscopy [32,40,77,97,98].

There are even speculations that long-lived nuclear spin states might be involved in human cognition [99,100].

In my view, there have been two big disappointments in the long-lived state field:

- No important metabolite has yet been found that supports a long-lived state with a really substantial lifetime in biologically relevant conditions, and which could be used to transport hyperpolarized nuclear spin order in biomolecular imaging applications. Although $^{13}C_2$-labelled pyruvate does support a long-lived singlet state with a longer lifetime that the $^{13}CT_1$ in low magnetic field [101], the extension of lifetime is not observed in magnetic fields high enough for typical imaging experiments, and is not significant enough to compensate for the increased complexity and increased losses of NMR experiments involving long-lived states. The recent demonstration that singlet-hyperpolarized fumarate may be generated by a direct catalytic reaction of parahydrogen [57] holds out somewhat more hope in this direction.
- The substance $^{15}N_2O$ ($^{15}N_2$-labelled nitrous oxide) initially seemed to be a promising target for hyperpolarized long-lived state NMR. The two ^{15}N spins-1/2 are isolated, possess a chemical shift difference (providing access to singlet order from magnetization). Nitrous oxide is biocompatible and widely used as an anaesthetic and for recreational purposes. $^{15}N_2O$ displays a long T_S of up to 20 min in solution [48], and 7 min in blood [102], and may be hyperpolarized in frozen solution by DNP [103]. Unfortunately attempts to exploit this material have so far been defeated by the very efficient nuclear spin-rotation relaxation of $^{15}N_2O$ in the gas phase.

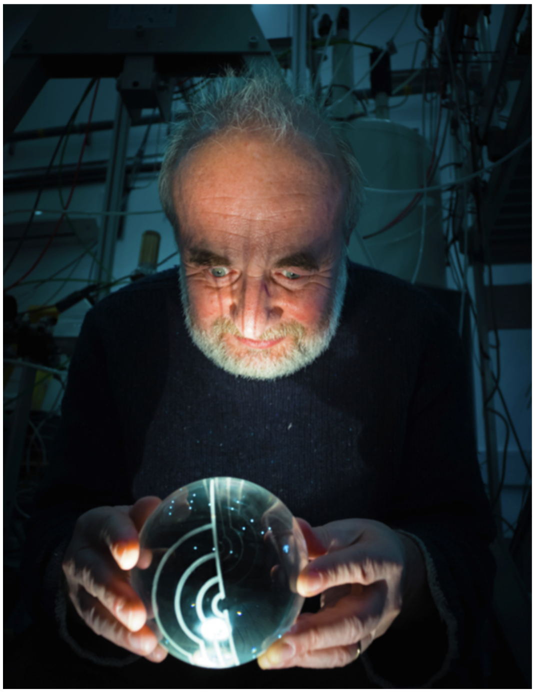

Fig. 2. My crystal ball was given to me in 2019 as a wonderful present from my research group, and is decorated with an occult inscription resembling magnetization vector trajectories for a composite pulse [104]. Unfortunately its picture quality has deteriorated – possibly through overuse (I am writing this in England in March 2019). Photograph by Karel Kouřil.

So where do we go from here?.

Despite the limited picture quality of my crystal ball (Fig. 2), the outline of future developments may be guessed.

- It is likely that the promising applications for long-lived states, as sketched above, will continue to develop. In particular the use of LLS phenomena for the characterization of ligand binding looks close to applications in realistic contexts [78–81].
- Long-lived states have always held promise as a way of extending the lifetime of hyperpolarized spin order, potentially extending the reach of hyperpolarization techniques such as dissolution-DNP in biomolecular imaging. Time has gone by and this potential has not yet been fully realised, despite some promising demonstrations. In this respect, long-lived states resemble nuclear fusion: A technology which has a great future, and which will always have a great future. Nevertheless long-lived states might still realise their potential in molecular imaging, if a hyperpolarized substance with a sufficiently long lived state is identified which has real significance as a metabolite or as a tag for functional molecular imaging. A lot of progress has been made [32,40,77,97,98], but we are not there yet.
- The fields of long-lived states and parahydrogen-induced hyperpolarization have a natural synergy, since parahydrogen is itself a long-lived nuclear singlet state. Insights from LLS spin dynamics have already played a major role in elucidating the mechanisms of important hyperpolarization methods such as SABRE (Signal Amplification by Reversible Exchange) [91,92,105]. The close contact between the fields is likely to continue into the future, specially in the context of low-field and ultra-low-field NMR [64,96,106].
- Long-lived states have also strengthened the productive contact between NMR and the related fields of quantum rotors and spin isomers. For example the powerful theories of molecular symmetry [107,108] have a clear relationship with the theory of long-lived states.
- In my mind, one of the most robust achievements of long-lived nuclear spin states is the convergence of language and concepts between the study of nuclear spins and the study of electron spins. The language of singlets and triplets is commonplace in electronic spectroscopy and molecular quantum mechanics but was quite rarely used in NMR before the demonstrations of long-lived singlet states – with some notable exceptions, such as the treatment of magnetically equivalent spin systems [109,110]. This is because the prevailing conceptual language of NMR has been largely determined by the “weak coupling” approximation, which has broad validity in high-field solution NMR. Key conceptual tools such as the product operator formalism [111,112] were developed within the context of the weak-coupling approximation and have become part of everyday NMR language. Singlet and triplet spin states do not fit well into this formalism.

  The opposite is true in multi-electron systems where the large electron exchange interaction is dominant. Electronic singlets and triplets are everyday concepts in molecular quantum mechanics and electronic spectroscopy, and are taught at undergraduate level.

  The study of nuclear singlet and triplet states establishes a productive analogy with electron-spin phenomena which involve electron singlet and triplet spin dynamics. This facilitates a two-way transfer of information and experience between ordinary NMR and phenomena in related fields including chemically-induced dynamic electron polarization (CIDEP) [113], chemically-induced dynamic nuclear polarization (CIDNP) [114,115], electrically-detected magnetic resonance

[116], and spintronics [117].
Despite the annoying flicker of my crystal ball, the continuation of this synergy into the future is plain.

To conclude: Ladies and gentlemen – be upstanding, and raise your glasses: Long live the singlet state!

Acknowledgments

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References

[1] M. Carravetta, O.G. Johannessen, M.H. Levitt, Beyond the T1 limit: singlet nuclear spin states in low magnetic fields, Phys. Rev. Lett. 92 (15) (2004) 153003.
[2] M. Carravetta, M.H. Levitt, Long-lived nuclear spin states in high-field solution NMR, J. Am. Chem. Soc. 126 (2004) 6228–6229.
[3] A. Farkas, Orthohydrogen, Parahydrogen and Heavy Hydrogen, Cambridge University Press, Cambridge, 1935.
[4] C.R. Bowers, D.P. Weitekamp, Transformation of symmetrization order to nuclear-spin magnetization by chemical reaction and nuclear magnetic resonance, Phys. Rev. Lett. 57 (1986) 2645–2648.
[5] C.R. Bowers, D.P. Weitekamp, Parahydrogen and synthesis allow dramatically enhanced nuclear alignment, J. Am. Chem. Soc. 109 (1987) 5541–5542.
[6] M.G. Pravica, D.P. Weitekamp, Net NMR alignment by adiabatic transport of parahydrogen addition products to high magnetic field, Chem. Phys. Lett. 145 (1988) 255–258.
[7] M. Prager, A. Heidemann, Rotational tunneling and neutron spectroscopy: a compilation, Chem. Rev. 97 (8) (1997) 2933–2966.
[8] J. Haupt, A new effect of dynamic polarization in a solid obtained by rapid change of temperature, Phys. Lett. A 38 (6) (1972) 389–390.
[9] M. Icker, S. Berger, Unexpected multiplet patterns induced by the Haupt-effect, J. Magn. Reson. 219 (2012) 1–3.
[10] B. Meier, J.-N. Dumez, G. Stevanato, J.T. Hill-Cousins, S.S. Roy, P. Håkansson, S. Mamone, R.C.D. Brown, G. Pileio, M.H. Levitt, Long-lived nuclear spin states in methyl groups and quantum-rotor-induced polarization, J. Am. Chem. Soc. 135 (2013) 18746–18749.
[11] J.-N. Dumez, P. Håkansson, S. Mamone, B. Meier, G. Stevanato, J.T. Hill-Cousins, S.S. Roy, R.C.D. Brown, G. Pileio, M.H. Levitt, Theory of long-lived nuclear spin states in methyl groups and quantum-rotor induced polarisation, J. Chem. Phys. 142 (4) (2015) 044506.
[12] B. Meier, Quantum-rotor-induced polarization, Magn. Reson. Chem. 56 (7) (2018) 610–618.
[13] B. Meier, K. Kouřil, C. Bengs, H. Kouřilová, T.C. Barker, S.J. Elliott, S. Alom, R.J. Whitby, M.H. Levitt, Spin-isomer conversion of water at room temperature and quantum-rotor-induced nuclear polarization in the water-endofullerene H2O@C60, Phys. Rev. Lett. 120 (26) (2018) 266001.
[14] J.H. Ardenkjaer-Larsen, B. Fridlund, A. Gram, G. Hansson, L. Hansson, M.H. Lerche, R. Servin, M. Thaning, K. Golman, Increase in signal-to-noise ratio of >10,000 times in liquid-state NMR, Proc. Natl. Acad. Sci. U.S.A. 100 (18) (2003) 10158–10163.
[15] M.C.D. Tayler, I. Marco-Rius, M.I. Kettunen, K.M. Brindle, M.H. Levitt, G. Pileio, Direct enhancement of nuclear singlet order by dynamic nuclear polarization, J. Am. Chem. Soc. 134 (18) (2012) 7668–7671.
[16] D. Mammoli, B. Vuichoud, A. Bornet, J. Milani, J.-N. Dumez, S. Jannin, G. Bodenhausen, Hyperpolarized para-Ethanol, J. Phys. Chem. B 119 (10) (2015) 4048–4052.
[17] A. Jhajharia, E.M.M. Weber, J.G. Kempf, D. Abergel, G. Bodenhausen, D. Kurzbach, Communication: dissolution DNP reveals a long-lived deuterium spin state imbalance in methyl groups, J. Chem. Phys. 146 (4) (2017) 041101, wOS:000394520200002.
[18] J.-N. Dumez, B. Vuichoud, D. Mammoli, A. Bornet, A.C. Pinon, G. Stevanato, B. Meier, G. Bodenhausen, S. Jannin, M.H. Levitt, Dynamic nuclear polarization of long-lived nuclear spin states in methyl groups, J. Phys. Chem. Lett. 8 (15) (2017) 3549–3555.
[19] W.S. Warren, E. Jenista, R.T. Branca, X. Chen, Increasing hyperpolarized spin lifetimes through true singlet eigenstates, Science 323 (5922) (2009) 1711–1714.
[20] Y.N. Zhang, P.C. Soon, A. Jerschow, J.W. Canary, Long-lived 1H nuclear spin singlet in dimethyl maleate revealed by addition of thiols, Angew. Chem. Int. Ed. 53 (13) (2014) 3396–3399.
[21] A. Bornet, X. Ji, D. Mammoli, B. Vuichoud, J. Milani, G. Bodenhausen, S. Jannin, Long-lived states of magnetically equivalent spins populated by dissolution-DNP and revealed by enzymatic reactions, Chem. Eur. J. 20 (51) (2014) 17113–17118.
[22] P. Saul, S. Mamone, S. Glöggler, Nuclear singlet multimers (NUSIMERs) with long-lived singlet states, Chem. Sci. 10 (2) (2019) 413–417.
[23] Y. Zhang, K. Basu, J.W. Canary, A. Jerschow, Singlet lifetime measurements in an all-proton chemically equivalent spin system by hyperpolarization and weak spin lock transfers, Phys. Chem. Chem. Phys. 17 (37) (2015) 24370–24375.
[24] Y. Zhang, X. Duan, P.C. Soon, V. Sychrovský, J.W. Canary, A. Jerschow, Limits in proton nuclear singlet-state lifetimes measured with para-hydrogen-induced polarization, ChemPhysChem 17 (19) (2016) 2967–2971.
[25] B. Procacci, S.S. Roy, P. Norcott, N. Turner, S.B. Duckett, Unlocking a diazirine long-lived nuclear singlet state via photochemistry: NMR detection and lifetime of an unstabilized diazo-compound, J. Am. Chem. Soc. 140 (48) (2018) 16855–16864.
[26] S. Yang, J. McCormick, S. Mamone, L.-S. Bouchard, S. Glöggler, Nuclear spin singlet states in photoactive molecules: from fluorescence/NMR bimodality to a bimolecular switch for spin singlet states, Angew. Chem. Int. Ed. 58 (9) (2019) 2879–2883.
[27] K. Claytor, T. Theis, Y. Feng, J. Yu, D. Gooden, W.S. Warren, Accessing long-lived disconnected spin-1/2 eigenstates through spins >1/2, J. Am. Chem. Soc. 136 (43) (2014) 15118–15121.
[28] Z. Zhou, K. Claytor, W.S. Warren, T. Theis, Accessing long lived 1H states via 2H couplings, J. Magn. Reson. 263 (2016) 108–115.
[29] Y. Feng, R.M. Davis, W.S. Warren, Accessing long-lived nuclear singlet states between chemically equivalent spins without breaking symmetry, Nat. Phys. 8 (11) (2012) 831–837.
[30] G. Stevanato, S. Singha Roy, J. Hill-Cousins, I. Kuprov, L.J. Brown, R.C.D. Brown, G. Pileio, M.H. Levitt, Long-lived nuclear spin states far from magnetic equivalence, Phys. Chem. Chem. Phys. 17 (8) (2015) 5913–5922.
[31] G. Stevanato, J.T. Hill-Cousins, P. Håkansson, S.S. Roy, L.J. Brown, R.C.D. Brown, G. Pileio, M.H. Levitt, A nuclear singlet lifetime of more than one hour in room-temperature solution, Angew. Chem. Int. Ed. 54 (2015) 3740–3743.
[32] T. Theis, G.X. Ortiz, A.W.J. Logan, K.E. Claytor, Y. Feng, W.P. Huhn, V. Blum, S.J. Malcolmson, E.Y. Chekmenev, Q. Wang, W.S. Warren, Direct and cost-efficient hyperpolarization of long-lived nuclear spin states on universal 15N2-diazirine molecular tags, Sci. Adv. 2 (3) (2016) e1501438.
[33] G. Pileio, M. Carravetta, M.H. Levitt, Extremely low-frequency spectroscopy in low-field nuclear magnetic resonance, Phys. Rev. Lett. 103 (8) (2009) 083002.
[34] R. Sarkar, P. Ahuja, P.R. Vasos, G. Bodenhausen, Long-lived coherences for homogeneous line narrowing in spectroscopy, Phys. Rev. Lett. 104 (5) (2010) 053001.
[35] A. Bornet, S. Jannin, J.A. Konter, P. Hautle, B. van den Brandt, G. Bodenhausen, Ultra high-resolution NMR: sustained induction decays of long-lived coherences, J. Am. Chem. Soc. 133 (39) (2011) 15644–15649.
[36] S. Chinthalapalli, A. Bornet, T.F. Segawa, R. Sarkar, S. Jannin, G. Bodenhausen, Ultrahigh-resolution magnetic resonance in inhomogeneous magnetic fields: two-dimensional long-lived-coherence correlation spectroscopy, Phys. Rev. Lett. 109 (2012) 047602.
[37] A. Sadet, L. Fernandes, F. Kateb, R. Balzan, P.R. Vasos, Long-lived coherences: improved dispersion in the frequency domain using continuous-wave and reduced-power windowed sustaining irradiation, J. Chem. Phys. 141(5).
[38] M. Singh, S. Chinthalapalli, G. Bodenhausen, Lifetimes of long-lived states in inhomogeneous magnetic fields, Chem. Phys. Lett. 623 (2015) 113–116.
[39] K.F. Sheberstov, A.S. Kiryutin, C. Bengs, J.T. Hill-Cousins, L.J. Brown, R.C.D. Brown, G. Pileio, M.H. Levitt, A.V. Yurkovskaya, K.L. Ivanov, Excitation of singlet-triplet coherences in pairs of nearly-equivalent spins, Phys. Chem. Chem. Phys. 21 (11) (2019) 6087–6100.
[40] A.S. Kiryutin, M.S. Panov, A.V. Yurkovskaya, K.L. Ivanov, G. Bodenhausen, Proton relaxometry of long-lived spin order, ChemPhysChem 20 (5) (2019) 766–772.
[41] P.R. Vasos, A. Comment, R. Sarkar, P. Ahuja, S. Jannin, J.-P. Ansermet, J.A. Konter, P. Hautle, B. van den Brandt, G. Bodenhausen, Long-lived states to sustain hyperpolarized magnetization, Proc. Natl. Acad. Sci. U.S.A. 106 (44) (2009) 18469–18473.
[42] P. Ahuja, R. Sarkar, S. Jannin, P.R. Vasos, G. Bodenhausen, Proton hyperpolarisation preserved in long-lived states, Chem. Commun. 46 (43) (2010) 8192–8194.
[43] G. Pileio, S. Bowen, C. Laustsen, M.C.D. Tayler, J.T. Hill-Cousins, L.J. Brown, R.C. D. Brown, J.H. Ardenkjaer-Larsen, M.H. Levitt, Recycling and imaging of nuclear singlet hyperpolarization, J. Am. Chem. Soc. 135 (13) (2013) 5084–5088.
[44] K.L. Ivanov, T. Kress, M. Baudin, D. Guarin, D. Abergel, G. Bodenhausen, D. Kurzbach, Relaxation of long-lived modes in NMR of deuterated methyl groups, J. Chem. Phys. 149 (5) (2018) 054202.
[45] M.H. Levitt, Singlet nuclear magnetic resonance, Annu. Rev. Phys. Chem. 63 (1) (2012) 89–105.
[46] G. Pileio, Singlet NMR methodology in two-spin-1/2 systems, Prog. Nucl. Magn. Reson. Spectrosc. 98–99 (2017) 1–19.
[47] M. Carravetta, M.H. Levitt, Theory of long-lived nuclear spin states in solution nuclear magnetic resonance. I. Singlet states in low magnetic field, J. Chem. Phys. 122 (2005) 214505.
[48] G. Pileio, M. Carravetta, E. Hughes, M.H. Levitt, The long-lived nuclear singlet state of 15N-nitrous oxide in solution, J. Am. Chem. Soc. 130 (38) (2008) 12582–12583.
[49] G. Pileio, M. Carravetta, M.H. Levitt, Storage of nuclear magnetization as long-lived singlet order in low magnetic field, Proc. Natl. Acad. Sci. U.S.A. 107 (40) (2010) 17135–17139.

[50] S.J. Elliott, P. Kadeřávek, L.J. Brown, M. Sabba, S. Glöggler, D.J. O'Leary, R.C.D. Brown, F. Ferrage, M.H. Levitt, Field-cycling long-lived-state NMR of 15N2 spin pairs, Mol. Phys. 1–7 (2018).

[51] R. Sarkar, P.R. Vasos, G. Bodenhausen, Singlet-state exchange NMR spectroscopy for the study of very slow dynamic processes, J. Am. Chem. Soc. 129 (2) (2007) 328–334.

[52] G. Pileio, M.H. Levitt, Theory of long-lived nuclear spin states in solution nuclear magnetic resonance. II. Singlet spin locking, J. Chem. Phys. 130 (21) (2009) 214501–214514.

[53] M.C.D. Tayler, M.H. Levitt, Singlet nuclear magnetic resonance of nearly-equivalent spins, Phys. Chem. Chem. Phys. 13 (13) (2011) 5556–5560.

[54] J. Eills, G. Stevanato, C. Bengs, S. Glöggler, S.J. Elliott, J. Alonso-Valdesueiro, G. Pileio, M.H. Levitt, Singlet order conversion and parahydrogen-induced hyperpolarization of 13C nuclei in near-equivalent spin systems, J. Magn. Reson. 274 (2017) 163–172.

[55] G. Stevanato, J. Eills, C. Bengs, G. Pileio, A pulse sequence for singlet to heteronuclear magnetization transfer: S2hM, J. Magn. Reson. 277 (2017) 169–178.

[56] G. Stevanato, Alternating Delays Achieve Polarization Transfer (ADAPT) to heteronuclei in PHIP experiments, J. Magn. Reson. 274 (2017) 148–162.

[57] B. Ripka, J. Eills, H. Kouřilová, M. Leutzsch, M.H. Levitt, K. Münnemann, Hyperpolarized fumarate via parahydrogen, Chem. Commun. 54 (86) (2018) 12246–12249.

[58] S.J. DeVience, R.L. Walsworth, M.S. Rosen, Preparation of nuclear spin singlet states using spin-lock induced crossing, Phys. Rev. Lett. 111 (17) (2013) 5.

[59] T. Theis, Y. Feng, T. Wu, W.S. Warren, Composite and shaped pulses for efficient and robust pumping of disconnected eigenstates in magnetic resonance, J. Chem. Phys. 140 (1) (2014) 014201.

[60] D.A. Barskiy, O.G. Salnikov, A.S. Romanov, M.A. Feldman, A.M. Coffey, K.V. Kovtunov, I.V. Koptyug, E.Y. Chekmenev, NMR Spin-Lock Induced Crossing (SLIC) dispersion and long-lived spin states of gaseous propane at low magnetic field (0.05 T), J. Magn. Reson. 276 (2017) 78–85.

[61] A.S. Kiryutin, A.V. Yurkovskaya, N.N. Lukzen, H.-M. Vieth, K.L. Ivanov, Exploiting adiabatically switched RF-field for manipulating spin hyperpolarization induced by parahydrogen, J. Chem. Phys. 143 (23) (2015) 234203.

[62] A.N. Pravdivtsev, A.S. Kiryutin, A.V. Yurkovskaya, H.-M. Vieth, K.L. Ivanov, Robust conversion of singlet spin order in coupled spin-1/2 pairs by adiabatically ramped RF-fields, J. Magn. Reson. 273 (2016) 56–64.

[63] B.A. Rodin, K.F. Sheberstov, A.S. Kiryutin, J.T. Hill-Cousins, L.J. Brown, R.C.D. Brown, B. Jamain, H. Zimmermann, R.Z. Sagdeev, A.V. Yurkovskaya, K.L. Ivanov, Constant-adiabaticity radiofrequency pulses for generating long-lived singlet spin states in NMR, J. Chem. Phys. 150 (6) (2019) 064201.

[64] M. Emondts, M.P. Ledbetter, S. Pustelny, T. Theis, B. Patton, J.W. Blanchard, M. C. Butler, D. Budker, A. Pines, Long-lived heteronuclear spin-singlet states in liquids at a zero magnetic field, Phys. Rev. Lett. 112 (7) (2014) 077601.

[65] S.J. Elliott, L.J. Brown, J.-N. Dumez, M.H. Levitt, Long-lived nuclear spin states in monodeuterated methyl groups, Phys. Chem. Chem. Phys. 18 (27) (2016) 17965–17972.

[66] S.J. Elliott, L.J. Brown, J.-N. Dumez, M.H. Levitt, Long-lived nuclear spin states in rapidly rotating CH2D groups, J. Magn. Reson. 272 (2016) 87–90.

[67] S.J. Elliott, B. Meier, B. Vuichoud, G. Stevanato, L.J. Brown, J. Alonso-Valdesueiro, L. Emsley, S. Jannin, M.H. Levitt, Hyperpolarized long-lived nuclear spin states in monodeuterated methyl groups, Phys. Chem. Chem. Phys. 20 (15) (2018) 9755–9759.

[68] M.C.D. Tayler, M.H. Levitt, Accessing long-lived nuclear spin order by isotope-induced symmetry breaking, J. Am. Chem. Soc. 135 (6) (2013) 2120–2123.

[69] P. Ahuja, R. Sarkar, P.R. Vasos, G. Bodenhausen, Molecular properties determined from the relaxation of long-lived spin states, J. Chem. Phys. 127 (13) (2007) 134112–134116.

[70] M.C.D. Tayler, S. Marie, A. Ganesan, M.H. Levitt, Determination of molecular torsion angles using nuclear singlet relaxation, J. Am. Chem. Soc. 132 (24) (2010) 8225–8227.

[71] G. Pileio, Relaxation theory of nuclear singlet states in two spin-1/2 systems, Prog. Nucl. Magn. Reson. Spectrosc. 56 (3) (2010) 217–231.

[72] G. Pileio, Singlet state relaxation via intermolecular dipolar coupling, J. Chem. Phys. 134 (21) (2011) 214505–214509.

[73] G. Pileio, Singlet state relaxation via scalar coupling of the second kind, J. Chem. Phys. 135 (17) (2011) 174502.

[74] G. Pileio, J.T. Hill-Cousins, S. Mitchell, I. Kuprov, L.J. Brown, R.C.D. Brown, M.H. Levitt, Long-lived nuclear singlet order in near-equivalent 13C spin pairs, J. Am. Chem. Soc. 134 (42) (2012) 17494–17497.

[75] P. Håkansson, Prediction of low-field nuclear singlet lifetimes with molecular dynamics and quantum-chemical property surface, Phys. Chem. Chem. Phys. 19 (16) (2017) 10237–10254.

[76] S.J. Elliott, C. Bengs, L.J. Brown, J.T. Hill-Cousins, D.J. O'Leary, G. Pileio, M.H. Levitt, Nuclear singlet relaxation by scalar relaxation of the second kind in the slow-fluctuation regime, J. Chem. Phys. 150 (6) (2019) 064315.

[77] J. Bae, Z. Zhou, T. Theis, W.S. Warren, Q. Wang, 15N4-1,2,4,5-tetrazines as potential molecular tags: integrating bioorthogonal chemistry with hyperpolarization and unearthing para-N2, Sci. Adv. 4 (3) (2018) eaar2978.

[78] N. Salvi, R. Buratto, A. Bornet, S. Ulzega, I. Rentero Rebollo, A. Angelini, C. Heinis, G. Bodenhausen, Boosting the sensitivity of ligand-protein screening by NMR of long-lived states, J. Am. Chem. Soc. 134 (27) (2012) 11076–11079.

[79] R. Buratto, A. Bornet, J. Milani, D. Mammoli, B. Vuichoud, N. Salvi, M. Singh, A. Laguerre, S. Passemard, S. Gerber-Lemaire, S. Jannin, G. Bodenhausen, Drug screening boosted by hyperpolarized long-lived states in NMR, ChemMedChem 9 (11) (2014) 2509–2515.

[80] R. Buratto, D. Mammoli, E. Chiarparin, G. Williams, G. Bodenhausen, Exploring weak ligand-protein interactions by long-lived NMR states: improved contrast in fragment-based drug screening, Angew. Chem. Int. Ed. 53 (42) (2014) 11376–11380.

[81] R. Buratto, D. Mammoli, E. Canet, G. Bodenhausen, Ligand-protein affinity studies using long-lived states of fluorine-19 nuclei, J. Med. Chem. 59 (5) (2016) 1960–1966.

[82] S. Cavadini, J. Dittmer, S. Antonijevic, G. Bodenhausen, Slow diffusion by singlet state NMR spectroscopy, J. Am. Chem. Soc. 127 (2005) 15744–15748.

[83] R. Sarkar, P. Ahuja, P.R. Vasos, G. Bodenhausen, Measurement of slow diffusion coefficients of molecules with arbitrary scalar couplings via long-lived spin states, ChemPhysChem 9 (16) (2008) 2414–2419.

[84] P. Ahuja, R. Sarkar, P.R. Vasos, G. Bodenhausen, Diffusion coefficients of biomolecules using long-lived spin states, J. Am. Chem. Soc. 131 (22) (2009) 7498–7499.

[85] G. Pileio, J.-N. Dumez, I.-A. Pop, J.T. Hill-Cousins, R.C.D. Brown, Real-space imaging of macroscopic diffusion and slow flow by singlet tagging MRI, J. Magn. Reson. 252 (2015) 130–134.

[86] G. Pileio, S. Ostrowska, Accessing the long-time limit in diffusion NMR: the case of singlet assisted diffusive diffraction q-space, J. Magn. Reson. 285 (2017) 1–7.

[87] M.C. Tourell, I.-A. Pop, L.J. Brown, R.C.D. Brown, G. Pileio, Singlet-assisted diffusion-NMR (SAD-NMR): redefining the limits when measuring tortuosity in porous media, Phys. Chem. Chem. Phys. 20 (20) (2018) 13705–13713.

[88] S.S. Roy, T.S. Mahesh, Initialization of NMR quantum registers using long-lived singlet states, Phys. Rev. A 82 (5) (2010) 052302.

[89] M.B. Franzoni, L. Buljubasich, H.W. Spiess, K. Münnemann, Long-lived 1H singlet spin states originating from para-hydrogen in Cs-symmetric molecules stored for minutes in high magnetic fields, J. Am. Chem. Soc. 134 (25) (2012) 10393–10396.

[90] L. Buljubasich, M.B. Franzoni, H.W. Spiess, K. Münnemann, Level anti-crossings in parahydrogen induced polarization experiments with Cs-symmetric molecules, J. Magn. Reson. 219 (2012) 33–40.

[91] T. Theis, M. Truong, A.M. Coffey, E.Y. Chekmenev, W.S. Warren, LIGHT-SABRE enables efficient in-magnet catalytic hyperpolarization, J. Magn. Reson. 248 (2014) 23–26.

[92] T. Theis, M.L. Truong, A.M. Coffey, R.V. Shchepin, K.W. Waddell, F. Shi, B.M. Goodson, W.S. Warren, E.Y. Chekmenev, Microtesla SABRE enables 10% nitrogen-15 nuclear spin polarization, J. Am. Chem. Soc. 137 (4) (2015) 1404–1407.

[93] S.S. Roy, P. Norcott, P.J. Rayner, G.G.R. Green, S.B. Duckett, A hyperpolarizable 1H magnetic resonance probe for signal detection 15 minutes after spin polarization storage, Angew. Chem. Int. Ed. 55 (50) (2016) 15642–15645.

[94] S.S. Roy, P.J. Rayner, P. Norcott, G.G.R. Green, S.B. Duckett, Long-lived states to sustain SABRE hyperpolarised magnetisation, Phys. Chem. Chem. Phys. 18 (36) (2016) 24905–24911.

[95] D. Graafen, M.B. Franzoni, L.M. Schreiber, H.W. Spiess, K. Münnemann, Magnetic resonance imaging of 1H long lived states derived from parahydrogen induced polarization in a clinical system, J. Magn. Reson. 262 (2016) 68–72.

[96] Z. Zhou, J. Yu, J.F.P. Colell, R. Laasner, A. Logan, D.A. Barskiy, R.V. Shchepin, E.Y. Chekmenev, V. Blum, W.S. Warren, T. Theis, Long-lived 13C2 nuclear spin states hyperpolarized by parahydrogen in reversible exchange at microtesla fields, J. Phys. Chem. Lett. 8 (13) (2017) 3008–3014.

[97] S. Glöggler, S.J. Elliott, G. Stevanato, R.C.D. Brown, M.H. Levitt, Versatile magnetic resonance singlet tags compatible with biological conditions, RSC Adv. 7 (55) (2017) 34574–34578.

[98] S. Mamone, S. Glöggler, Nuclear spin singlet states as magnetic on/off probes in self-assembling systems, Phys. Chem. Chem. Phys. 20 (35) (2018) 22463–22467.

[99] M.P.A. Fisher, Quantum cognition: the possibility of processing with nuclear spins in the brain, Ann. Phys. 362 (2015) 593–602.

[100] M.P.A. Fisher, Are we quantum computers, or merely clever robots?, Int J. Mod. Phys. B 31 (07) (2017) 1743001.

[101] I. Marco-Rius, M.C.D. Tayler, M.I. Kettunen, T.J. Larkin, K.N. Timm, E.M. Serrao, T.B. Rodrigues, G. Pileio, J.H. Ardenkjaer-Larsen, M.H. Levitt, K.M. Brindle, Hyperpolarized singlet lifetimes of pyruvate in human blood and in the mouse, NMR Biomed. 26 (12) (2013) 1696–1704.

[102] R.K. Ghosh, S.J. Kadlecek, J.H. Ardenkjaer-Larsen, B.M. Pullinger, G. Pileio, M.H. Levitt, N.N. Kuzma, R.R. Rizi, Measurements of the persistent singlet state of N (2)O in blood and other solvents-potential as a magnetic tracer, Magn. Reson. Med. 66 (4) (2011) 1177–1180.

[103] N.N. Kuzma, P. Håkansson, M. Pourfathi, R.K. Ghosh, H. Kara, S.J. Kadlecek, G. Pileio, M.H. Levitt, R.R. Rizi, Lineshape-based polarimetry of dynamically-polarized 15N2O in solid-state mixtures, J. Magn. Reson. 234 (2013) 90–94.

[104] M.H. Levitt, R. Freeman, NMR population inversion using a composite pulse, J. Magn. Reson. 33 (2) (1979) 473–476.

[105] R.W. Adams, J.A. Aguilar, K.D. Atkinson, M.J. Cowley, P.I.P. Elliott, S.B. Duckett, G.G.R. Green, I.G. Khazal, J. Lopez-Serrano, D.C. Williamson, Reversible interactions with para-hydrogen enhance NMR sensitivity by polarization transfer, Science 323 (5922) (2009) 1708–1711.

[106] T. Theis, P. Ganssle, G. Kervern, S. Knappe, J. Kitching, M.P. Ledbetter, D. Budker, A. Pines, Parahydrogen-enhanced zero-field nuclear magnetic resonance, Nat. Phys. 7 (7) (2011) 571–575.

[107] P.R. Bunker, P. Jensen, Fundamentals of Molecular Symmetry, Institute of Physics, Bristol, 2005.
[108] P.R. Bunker, P. Jensen, Molecular Symmetry and Spectroscopy, second ed., NRC Research Press, Ottawa, 2006.
[109] J.A. Pople, W.G. Schneider, H.J. Bernstein, High-Resolution Nuclear Magnetic Resonance, McGraw-Hill, New York, 1959.
[110] P.L. Corio, Structure of High-Resolution NMR Spectra, Academic, New York, 1966.
[111] O.W. Sørensen, G.W. Eich, M.H. Levitt, G. Bodenhausen, R.R. Ernst, Product operator formalism for the description of NMR pulse experiments, Prog. Nucl. Magn. Reson. Spectrosc. 16 (1984) 163–192.
[112] R.R. Ernst, G. Bodenhausen, A. Wokaun, Principles of Nuclear Magnetic Resonance in One and Two Dimensions, Clarendon Press, Oxford, 1987.
[113] C. Blättler, F. Jent, H. Paul, A novel radical-triplet pair mechanism for chemically induced electron polarization (CIDEP) of free radicals in solution, Chem. Phys. Lett. 166 (4) (1990) 375–380.
[114] R. Kaptein, Chemically induced dynamic nuclear polarization. VIII. Spin dynamics and diffusion of radical pairs, J. Am. Chem. Soc. 94 (18) (1972) 6251–6262.
[115] M.G. Zysmilich, A. McDermott, Photochemically induced dynamic nuclear polarization in the solid-state 15N spectra of reaction centers from photosynthetic bacteria rhodobacter sphaeroides R-26, J. Am. Chem. Soc. 116 (18) (1994) 8362–8363.
[116] C. Boehme, K. Lips, Theory of time-domain measurement of spin-dependent recombination with pulsed electrically detected magnetic resonance, Phys. Rev. B 68 (24) (2003) 245105.
[117] S.A. Wolf, D.D. Awschalom, R.A. Buhrman, J.M. Daughton, S. von Molnár, M.L. Roukes, A.Y. Chtchelkanova, D.M. Treger, Spintronics: a spin-based electronics vision for the future, Science 294 (5546) (2001) 1488–1495.