Supporting Information for

Selective NMR detection of individual reaction components hyperpolarised by reversible exchange with para-hydrogen

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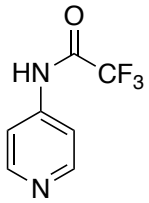
Appendix S1. Synthetic methods and characterisation

1.1 General experimental

All synthetic reactions were conducted under a nitrogen atmosphere using standard Schlenk techniques unless otherwise stated. All compounds, reagents and solvents were purchased from commercial sources and used as supplied unless otherwise stated. Petrol refers to the fraction of petroleum ether boiling in the range 40-60 °C. Flash column chromatography was carried out using ChemSupply silica gel 60 (0.04-0.06 mm, 230-400 mesh). Thin layer chromatography was carried out using Merck F_{254} aluminium-backed silica plates and visualized using UV light (254 and 365 nm) and ethanolic anisaldehyde stain. ^{1}H (400 MHz) and ^{13}C (100 MHz) NMR spectra were recorded on a Bruker AVANCE 400 instrument. Chemical shifts are quoted as parts per million (ppm) and referenced to the d_6-DMSO residual or solvent signals (δ_H 2.50, δ_C 39.52). ^{13}C NMR spectra were recorded with broadband proton decoupling. Coupling constants (J) are quoted in Hertz.

1.2 Synthesis of 4-(trifluoroacetamido)pyridine

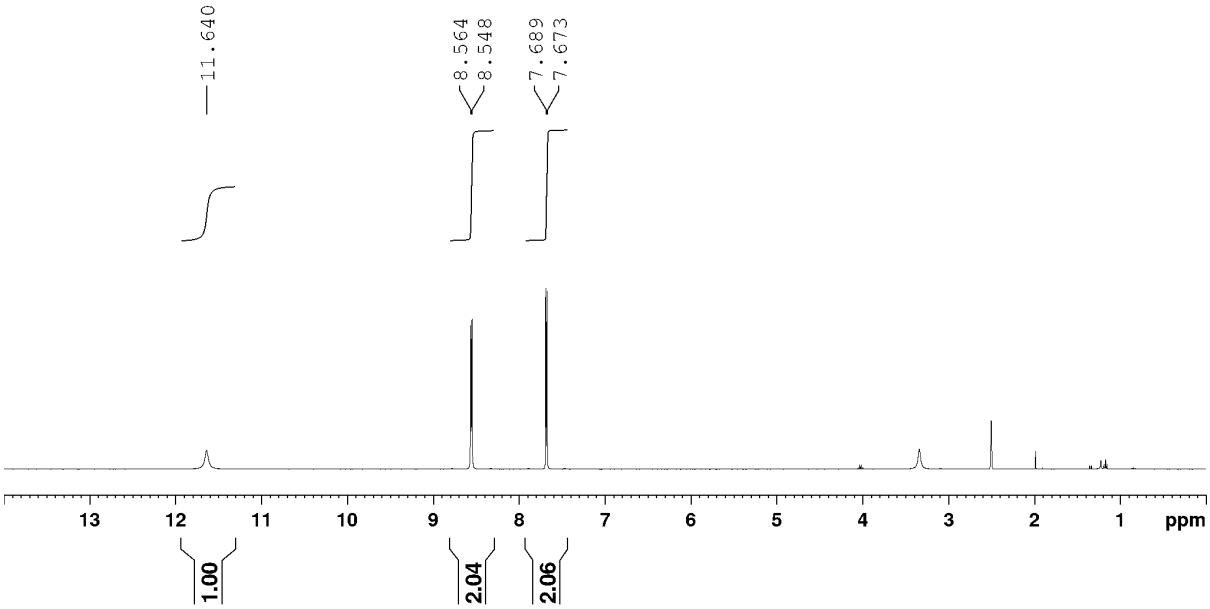


Prepared through a modification to the procedure of Bissell and Swansiger: *J. Chem. Eng. Data*, 1981, **26**, 234-235.[1]

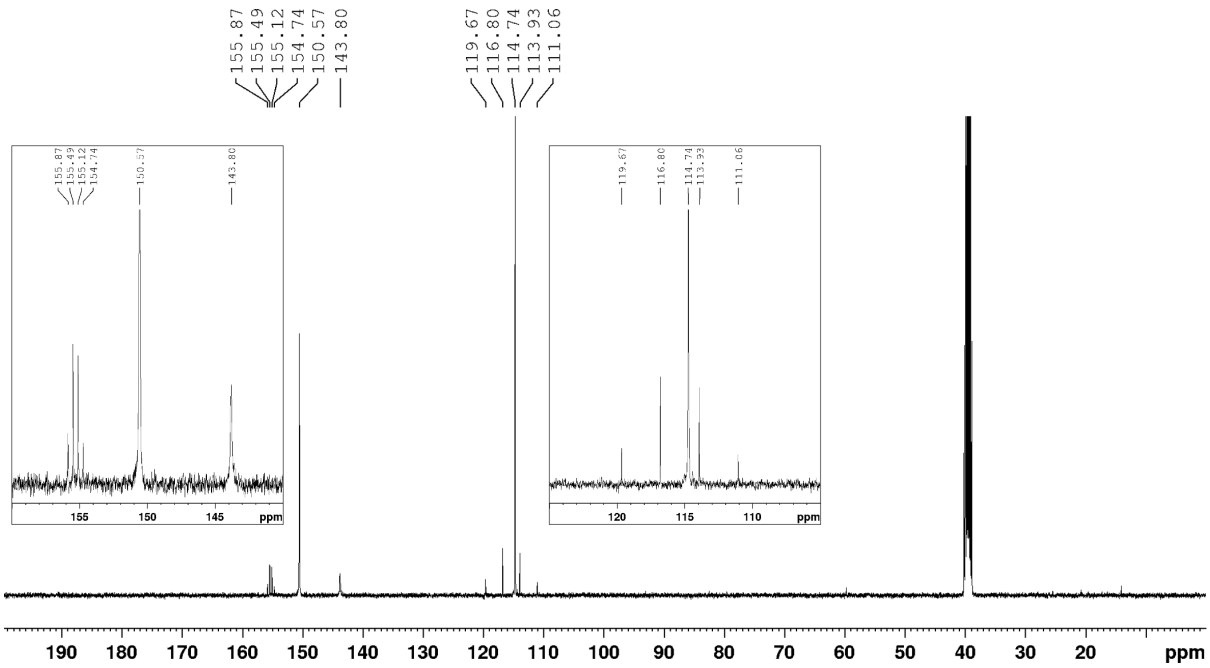
To a solution of 4-aminopyridine (200 mg, 2.1 mmol) in diethyl ether (2 mL) at 0 °C was added trifluoroacetic anhydride (290 μL, 2.1 mmol). The reaction mixture was allowed to return to room temperature and stirred overnight. Quenched with saturated aqueous $NaHCO_3$ solution (10 mL), extracted with diethyl ether (2 × 10 mL), dried over Na_2SO_4 and solvent removed under reduced pressure. Purified by flash column chromatography, eluting with 50% EtOAc-petrol increasing to 100% EtOAc, to give 4-(trifluoroacetamido)pyridine (121 mg, 30%) as a colourless crystalline solid; **^{1}H NMR** (400 MHz; d_6-DMSO) 11.64 (1 H, br s, NH), 8.56 (2 H, m, ArH_{ortho}), 7.68 (2 H, m, ArH_{meta}); **^{13}C NMR** (100 MHz, d_6-DMSO) 155.3 (q, $^{2}J_{CF}$ = 37.6 Hz), 150.6 (s), 143.8 (s), 115.4 (q, $^{1}J_{CF}$ = 288.7 Hz), 114.7 (s).

Figure S1: ^{1}H and ^{13}C NMR spectra of synthesized 4-(trifluoroacetamido)pyridine

^{1}H NMR (400 MHz; d_6-DMSO):



$^{13}C\{^{1}H\}$ NMR (100 MHz; d_6-DMSO):



Appendix S2. Details of experimental setup for SABRE

2.1 General procedure

A typical SABRE sample consisted of a solution of substrate(s) and [IrCl(COD)(IMes)] (stated concentrations) in a 1:1 mixture of CD_2Cl_2/CD_3OD (0.6 mL) in an NMR tube equipped with a J Young's valve. The samples were first degassed through a freeze-pump-thaw method using a mixture of dry ice and acetone as a cooling bath.

A Domnick Hunter 60H hydrogen generator was used as the source of >99.999% purity hydrogen gas, connected to a parahydrogen generator sourced from HyperSpin Scientific UG (http://www.hyperspin.biz). Operation of the latter at a conversion temperature of 77 K using liquid nitrogen produced ~50% *p*-H_2 enrichment levels.

The degassed sample tube was filled with *p*-H_2 up to 2 bar (gauge) pressure, then manually shaken for 10 seconds at a magnetic field strength of 65 G to facilitate polarization transfer, followed by immediate transport into the spectrometer for analysis. SABRE experiments were conducted at 298 K using a Bruker AVANCE III HD, PA BBO 400S1 BBF-H-D-05 Z probe, in a Varian MR400 magnet operating at 399.76 MHz.

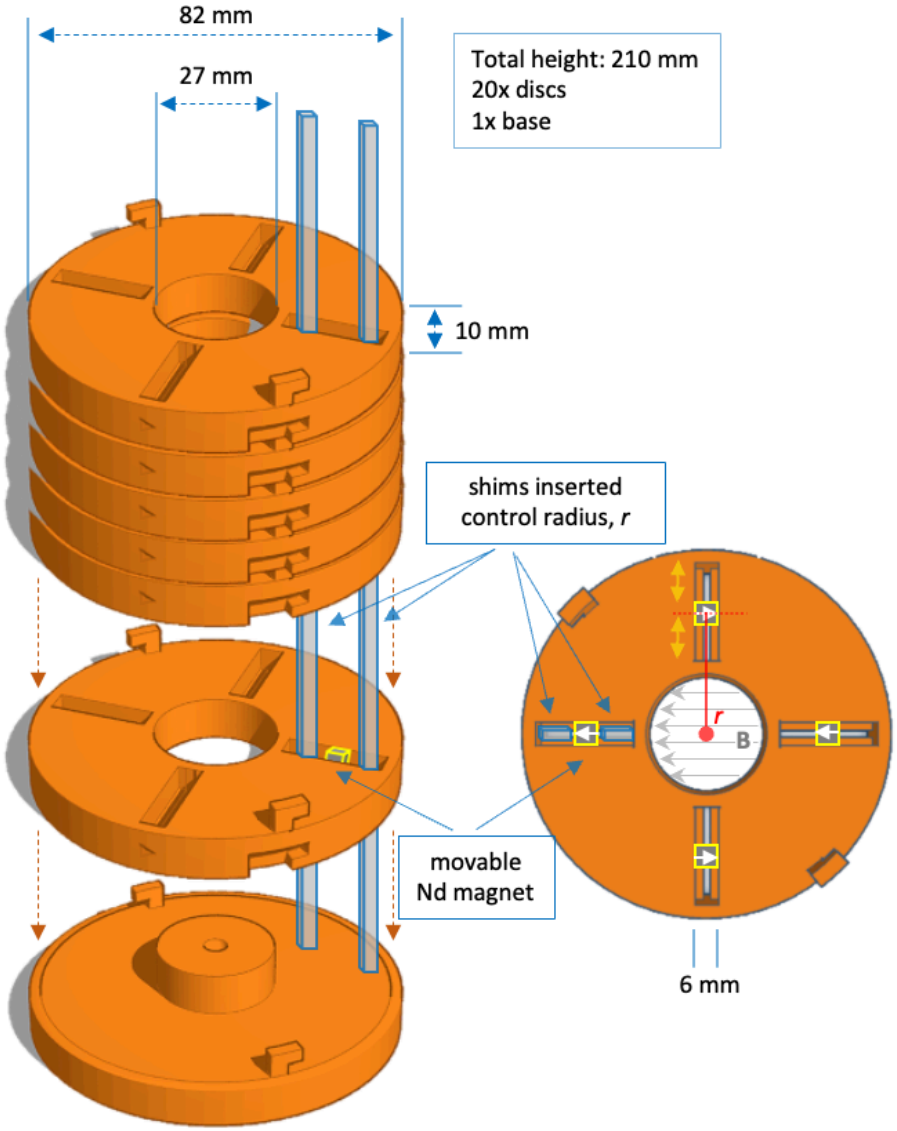
2.2 Handheld magnetic shaker

A permanent magnet array was constructed in order to create a chamber for a J Young's tap NMR tube in a standard spinner to reside at the desired polarization transfer field during SABRE experiments, based upon a design by Halse (*Magn. Reson. Chem.* 2018, **56**, 641-650).[2] A series of nickel coated NdFeB magnets (N50 grade, 5 mm x 5 mm x 5 mm) were arranged in a bespoke 3D printed ABS disc-shaped holder to create a Halbach array as depicted in Figure S2. 20 of these interlocking discs (with only every second disc containing magnets) were stacked to create a cylinder 21 cm tall, including a solid base plate. NMR tubes were pressurized with para-hydrogen, placed in the polarisation transfer field within the shaker, and the whole apparatus manually shaken for the desired time before transfer to the spectrometer for analysis.

A novel feature of this shaker design is that the magnets may reside at a variable radius from the centre of the discs. The magnets may be fixed to a given position by the addition of simple plastic shims of defined widths positioned alongside the magnets, running vertically through

the apparatus. This allows a single device to be tuned to produce any desired magnetic field inside the cylinder within the range of approximately 35 to 130 G, without the need to construct multiple different apparatus from the ground up to access different field strengths. In this design only every second disc contains magnets; even greater field strengths could easily be accomplished by the addition of magnets to all discs. A radius (*r*, Figure S2) of 26 mm was found to produce a polarization transfer field of 65±5 G (a commonly effective polarization transfer field strength for conventional SABRE systems) when measured at the central axis of the shaker using a TD8620 digital gauss meter with a transverse hall probe.

Figure S2: Handheld magnetic shaker design based on a series of vertically stacked Halbach array discs each created by 4 movable permanent magnets; their position (and thus magnetic field strength produced in the bore) controlled by the insertion of shims of a defined width.



2.3 SABRE-DREAM procedure

A sample used for conventional SABRE was hyperpolarised using the same general procedure as described in Section 2.1, transferred to the spectrometer and a single scan of an 8 scan DREAMTIME phase cycle (Simpson et al. *Angew. Chem. Int. Ed.* 2022, e202110044)[3] was collected. Hyperpolarised magnetization was reestablished in the same manner before each subsequent scan, and the overall signal enhancements compared to the thermally polarised DREAMTIME sequence using the same 8 scan phase cycle, keeping all other acquisition parameters the same, including receiver gain.

It has been demonstrated that use of a handheld magnet array as described in the previous section greatly increases reproducibility of polarization transfer during these sequences, close to that of an automated flow system (*Magn. Reson. Chem.* 2018, **56**, 641-650).[2]

Enhancement levels, ε, were calculated as the ratio of the hyperpolarised signal, $S_{polarised}$, and thermal signal, $S_{thermal}$, determined from the raw integrals of the hyperpolarised and thermal (reference) experiments respectively:

$$\varepsilon = \frac{S_{polarised}}{S_{thermal}}$$

Appendix S3. Determination of *J* coupling value in DREAMTIME sequences

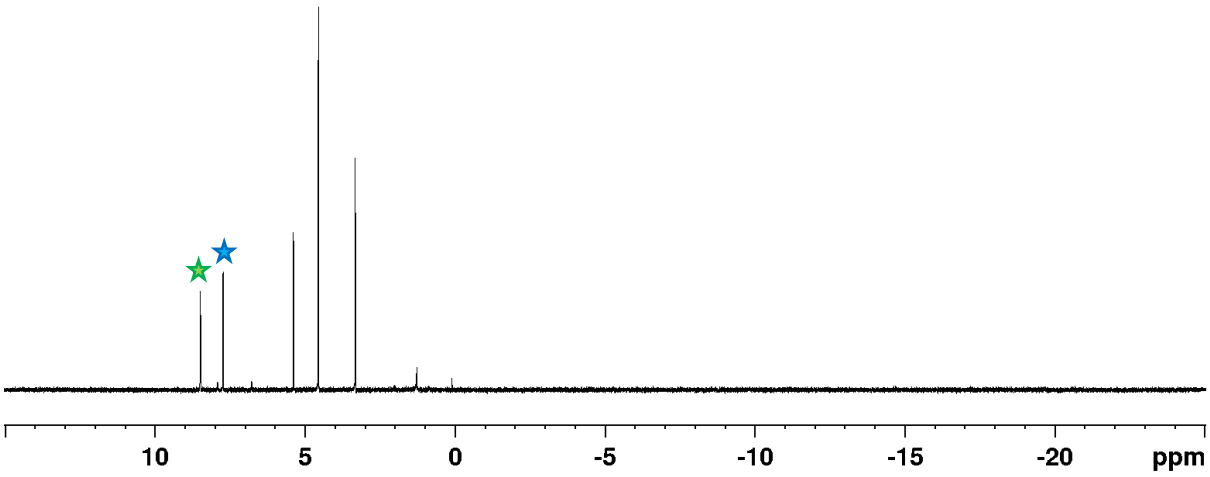
3.1 General conditions

^{1}H DREAMTIME NMR spectra were acquired at 298 K using a Bruker AVANCE III HD, PA BBO 400S1 BBF-H-D-05 Z probe, in a Varian MR400 magnet operating at 399.76 MHz, measuring from 15 to -25 ppm, DS 8, NS 16, with a 40 ms waveform encoded into the pulse sequence generated according to Simpson et. al. (*Angew. Chem. Int. Ed.* 2022, e202110044),[3] each experiment incorporating the resonant frequencies of the compound and a mutual *J* coupling as indicated.

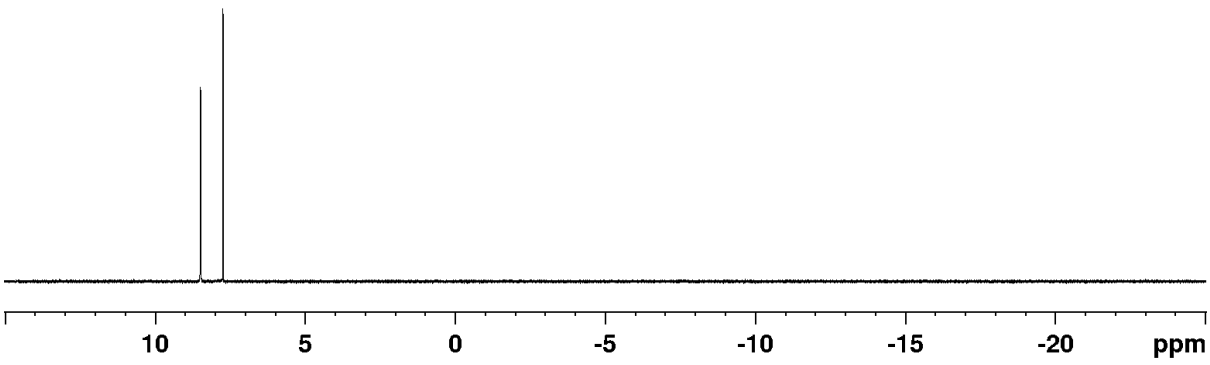
3.2 Optimisation of 4-(trifluoroacetamido)pyridine

Figure S3: amide (20 mM) in CD_2Cl_2:d_4-MeOD (1:1; 0.6 mL)

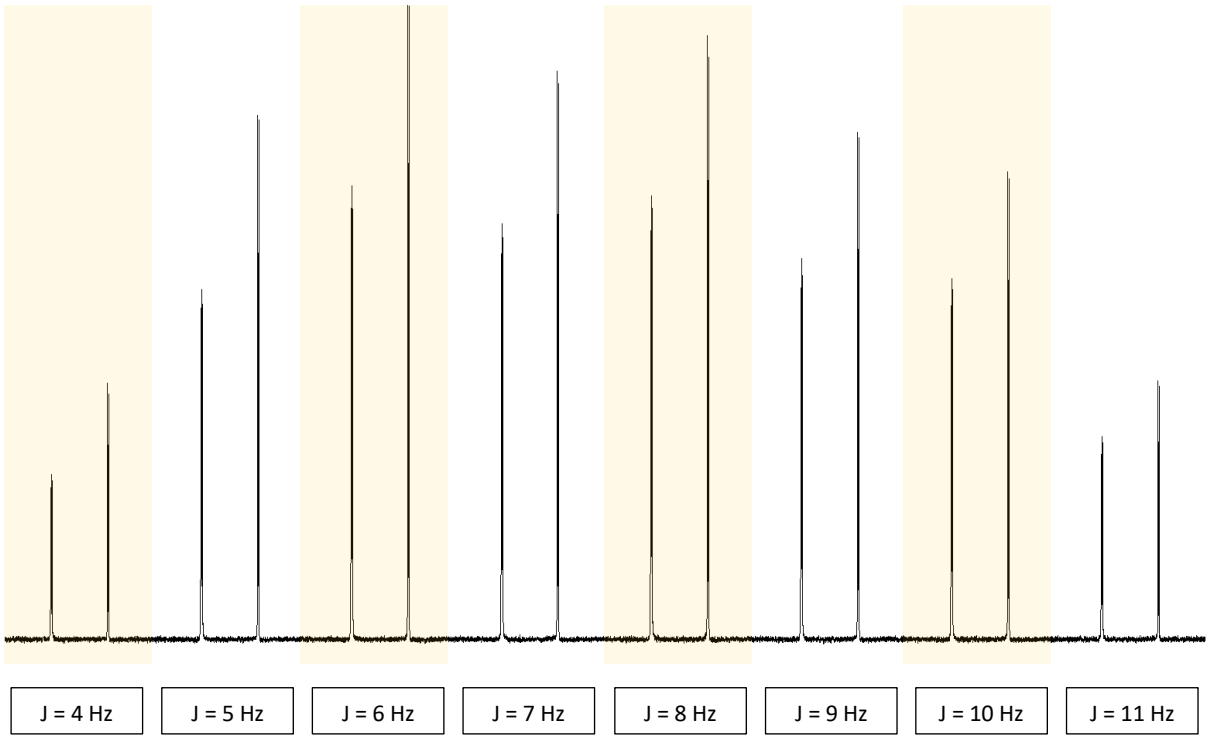
^{1}H NMR:



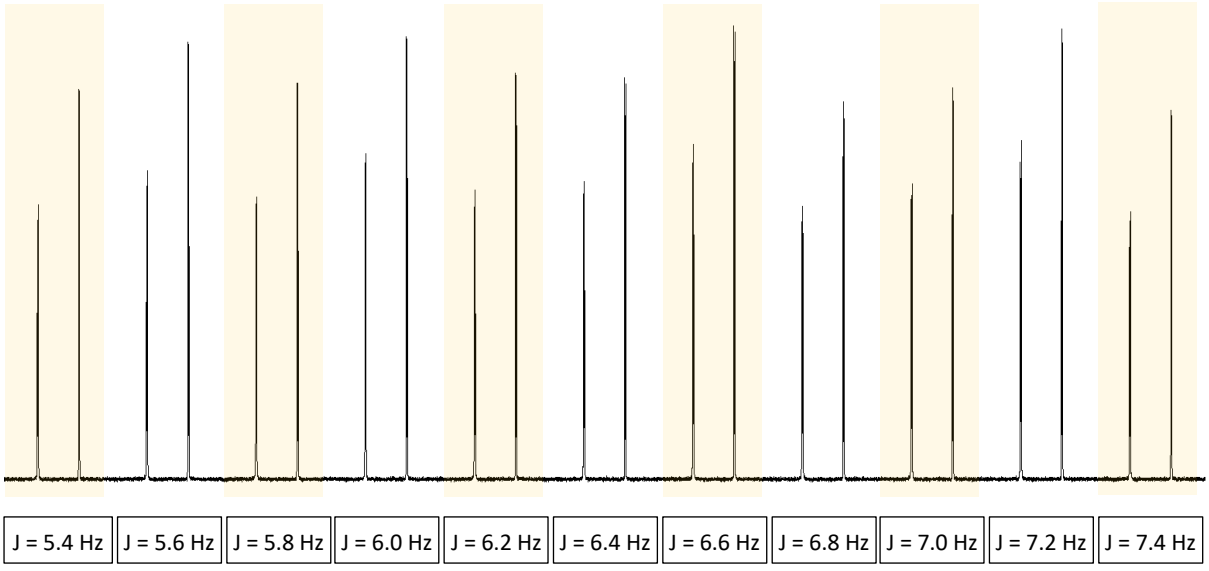
DREAMTIME NMR: *J* = 7 Hz



Parameter optimisation from J = 4 to 11 Hz:



Parameter optimisation from J = 5.4 to 7.4 Hz:

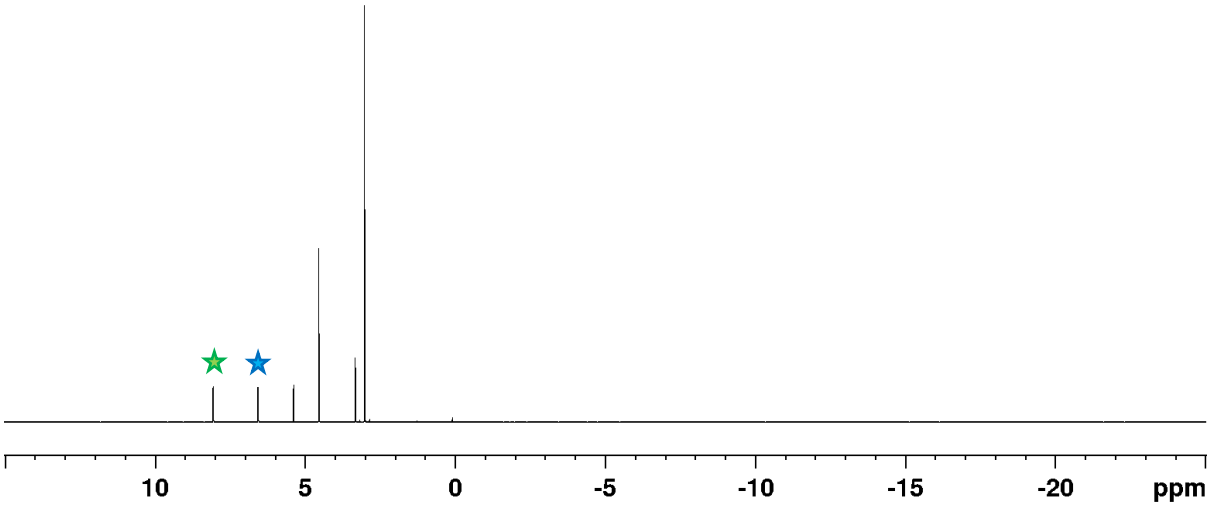


Optimised value taken as $\boldsymbol{J}$ **= 6.6 Hz**.

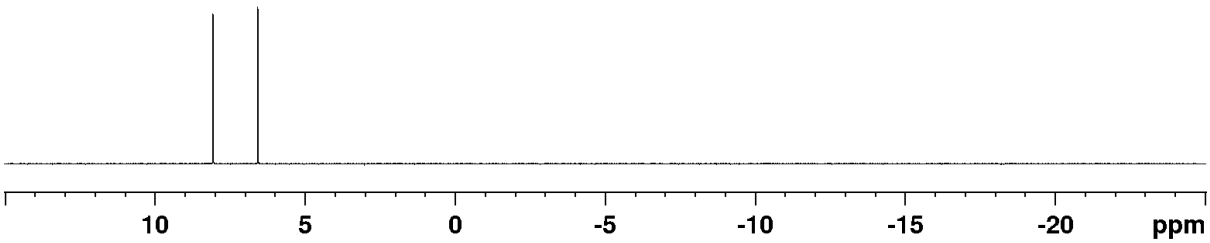
3.3 Optimisation of 4-(*N*,*N*-dimethylamino)pyridine

Figure S4: DMAP (20 mM) in CD_2Cl_2:d_4-MeOD (1:1; 0.6 mL)

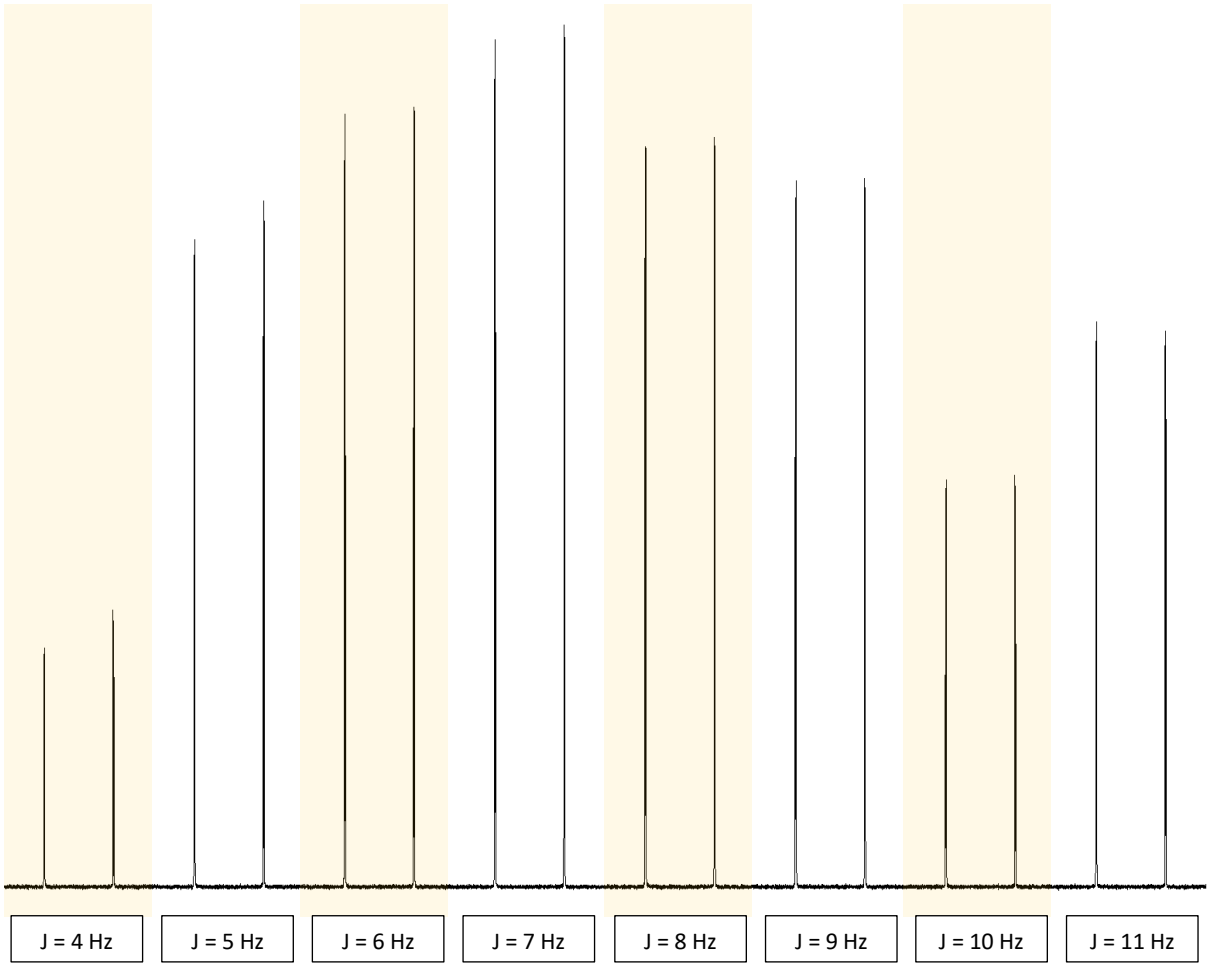
^{1}H NMR:



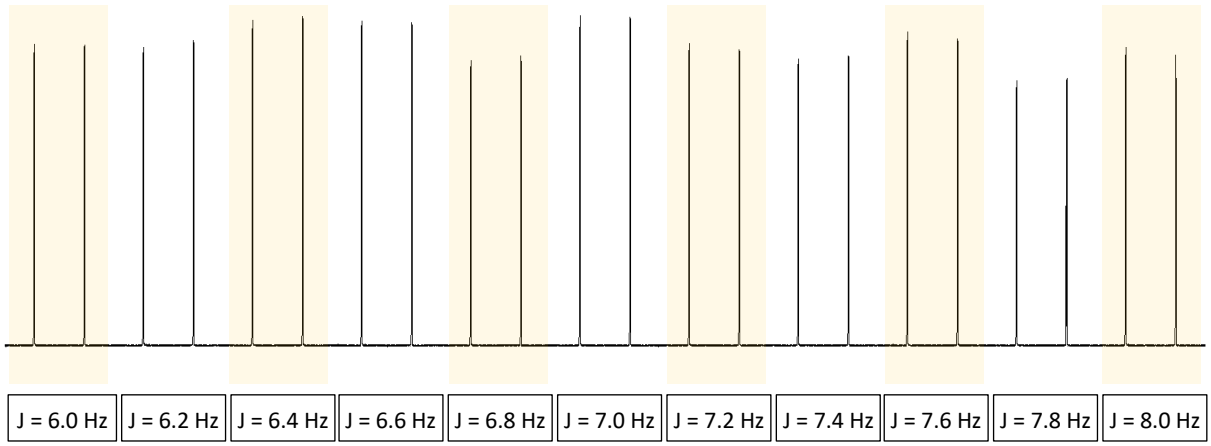
DREAMTIME NMR: J = 7 Hz



Parameter optimisation from J = 4 to 11 Hz:



Parameter optimisation from J = 6 to 8 Hz:

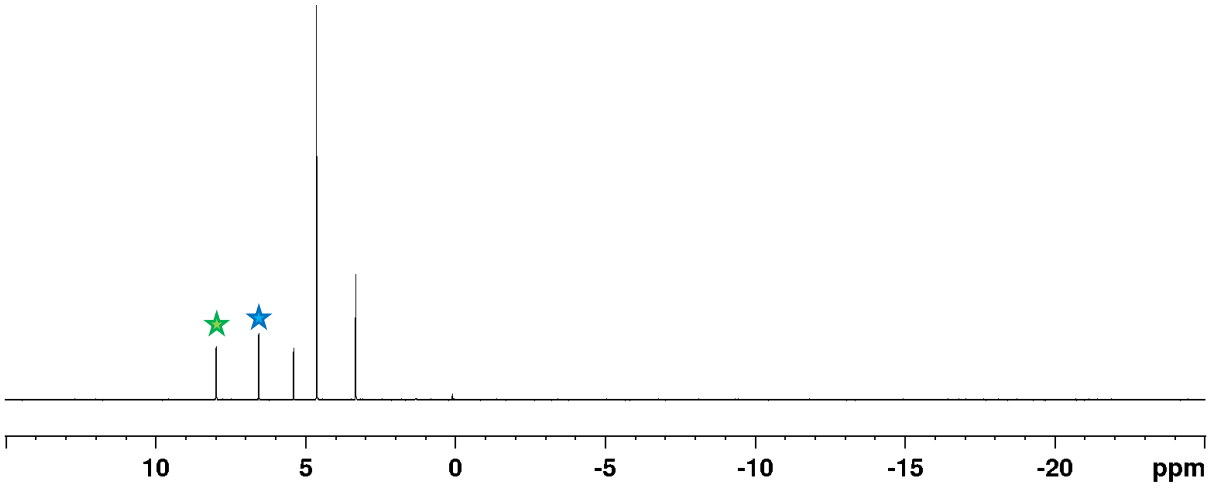


Optimised value taken as ***J* = 7.0 Hz.**

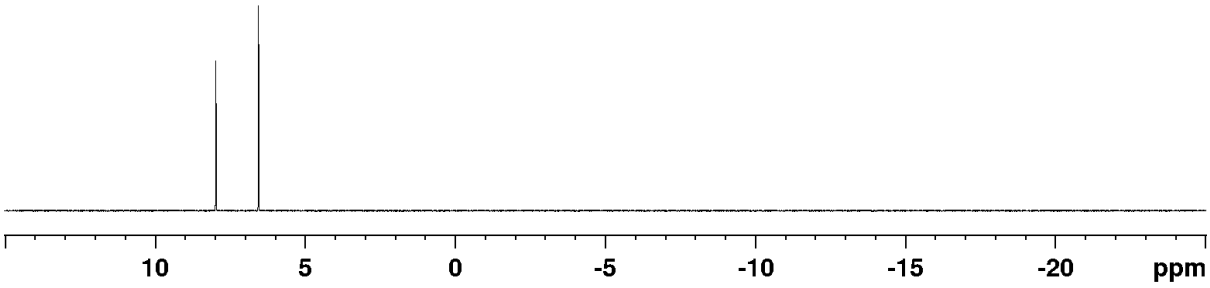
3.4 Optimisation of 4-aminopyridine

Figure S5: amine (20 mM) in CD_2Cl_2:d_4-MeOD (1:1; 0.6 mL)

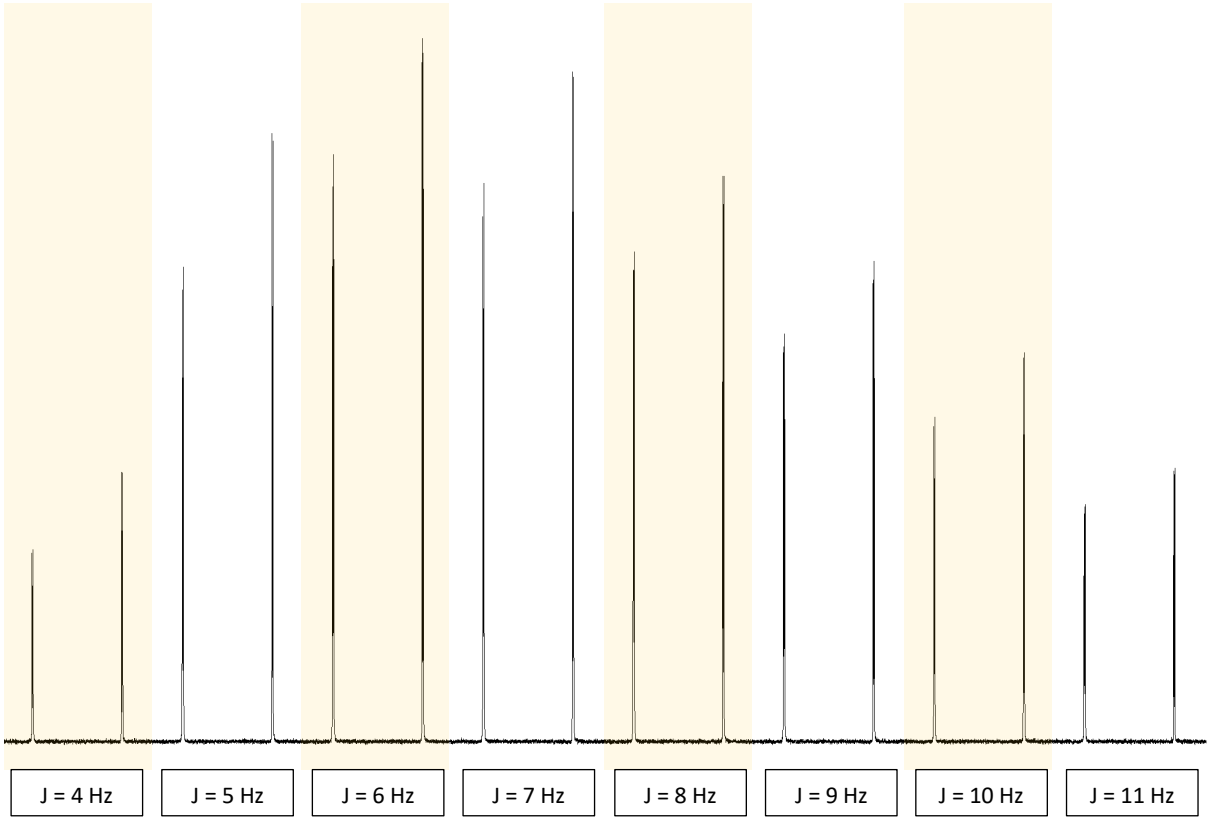
^{1}H NMR:



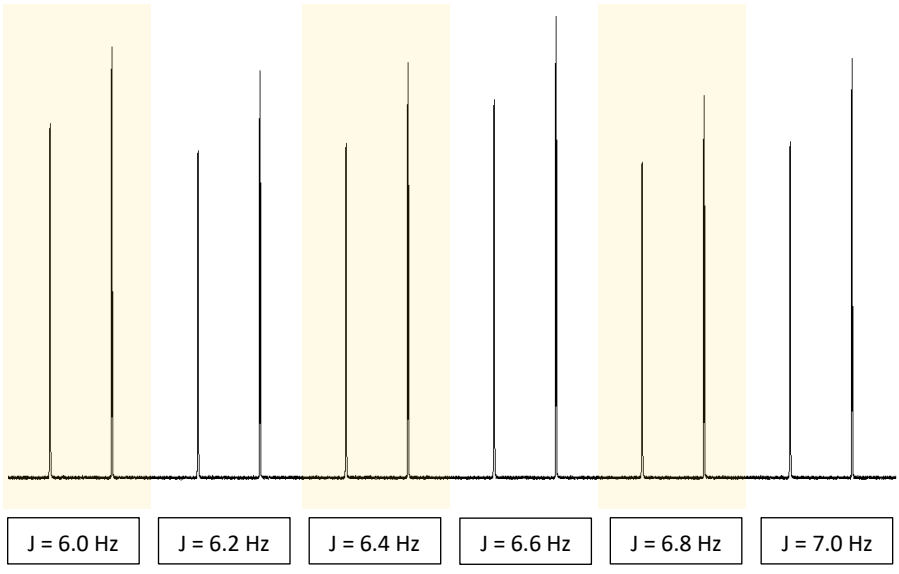
DREAMTIME NMR: J = 7 Hz



Parameter optimisation from J = 4 to 11 Hz:



Parameter optimisation from J = 6 to 7 Hz:



Optimised value taken as ***J* = 6.6 Hz**.

Appendix S4. Methanolysis of 4-(trifluoroacetamido)pyridine

4.1 Reactivity profile in the absence of DMAP

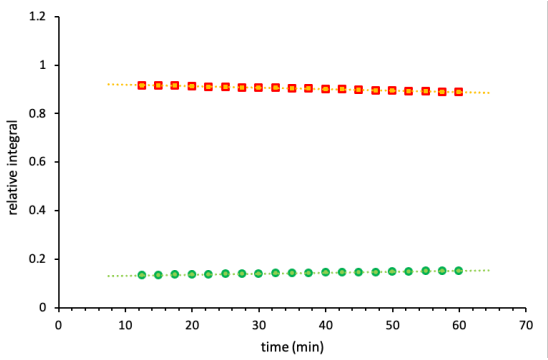
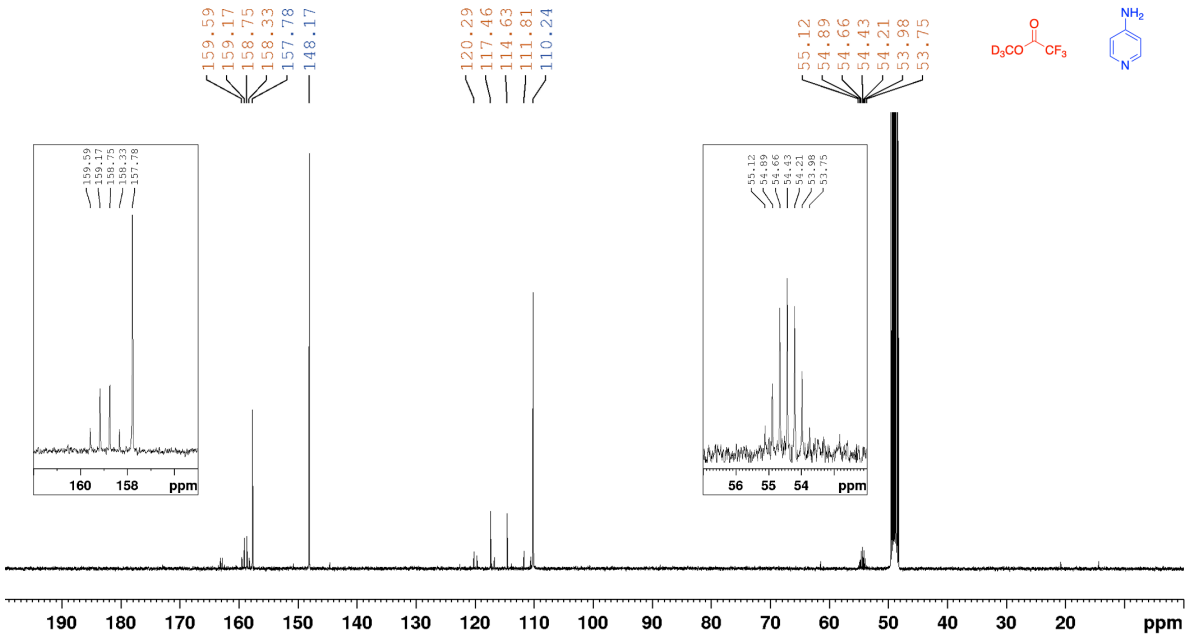


Figure S6: Relative concentrations of amide (■) and amine (●) when mixed in an approx. 9:1 ratio (60 mM with respect to the amide) in a 1:1 mixture of CD_2Cl_2/CD_3OD at 298 K.

4.2 Co-production of *d*$_3$-methyl trifluoroacetate

Figure S7: $^{13}C\{^1H\}$ NMR spectrum of a sample of 4-(trifluoroacetamido)pyridine left to react in CD_3OD for 2 days in a sealed NMR tube, revealing the production of *d*$_3$-methyl trifluoroacetate alongside 4-aminopyridine.



c.f. ***d*$_3$-methyl trifluoroacetate** generated by Jurczak *et al.*, *Org. Biomol. Chem*. 2018, **16**, 3114-3120:[4]

^{13}C (101 MHz, CD_3OD) 159.04 (q, $^2J_{CF}$ = 41.9 Hz), 116.09 (q, $^1J_{CF}$ = 284.3 Hz), 54.4 (sept, $^1J_{CD}$ = 22.7 Hz).

c.f. **4-aminopyridine** generated by Xu and Wolf, *Chem. Commun*. 2009, **21**, 3035-3037:[5]

^{13}C (100 MHz; CD_3OD) 110.3, 149.7, 156.8.

Appendix S5. SABRE-DREAM on signals with opposite SABRE enhancements

5.1 Pyridine under SABRE conditions

Figure S8: Thermal ^{1}H spectrum (and expansion) of pyridine (20 mM), [IrCl(COD)(IMes)] (5 mM) in CD_3OD (0.6 mL), reacted under 2 bar H_2 for 30 minutes.

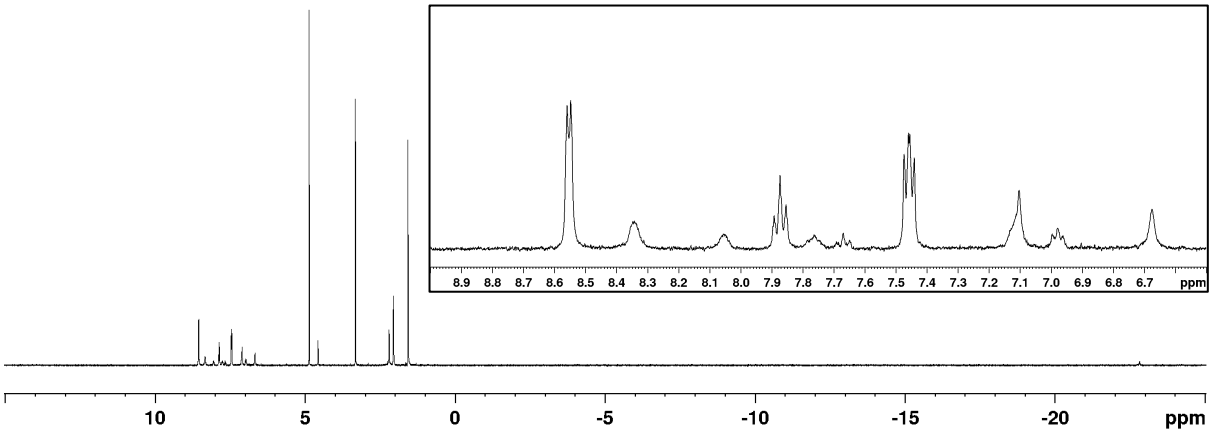
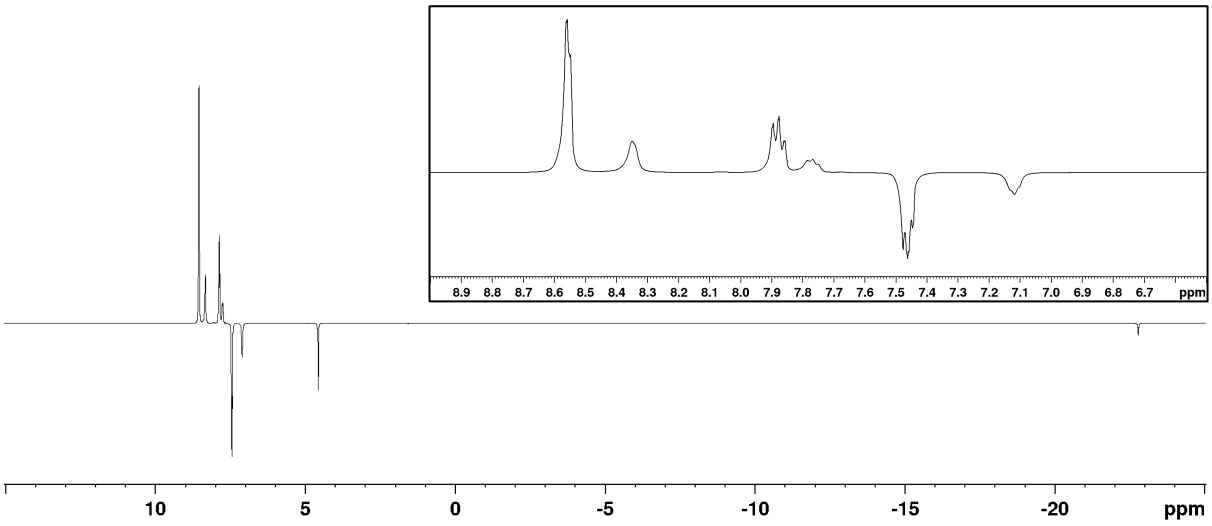


Figure S9: SABRE hyperpolarised spectrum (and expansion) obtained by shaking the tube under 2 bar p-H_2 at the earth's magnetic field (~0.5 G) for 10 seconds before transport to the spectrometer.



Note: arbitrary vertical scales used for illustration purposes only. Enhancement levels (ε) of free pyridine given below.

$\varepsilon_{(ortho)} = +243$

$\varepsilon_{(meta)} = -180$

$\varepsilon_{(para)} = +244$

5.2 Pyridine under SABRE-DREAM conditions

Figure S10: DREAMTIME spectrum (and expansion) exciting the *ortho* and *meta* resonances of free pyridine (at 8.55 and 7.46 ppm).

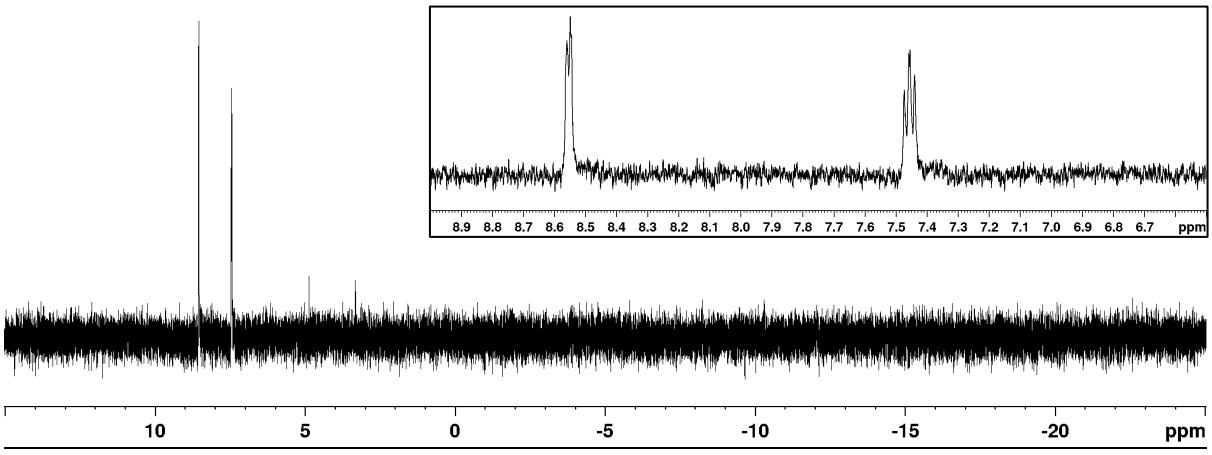
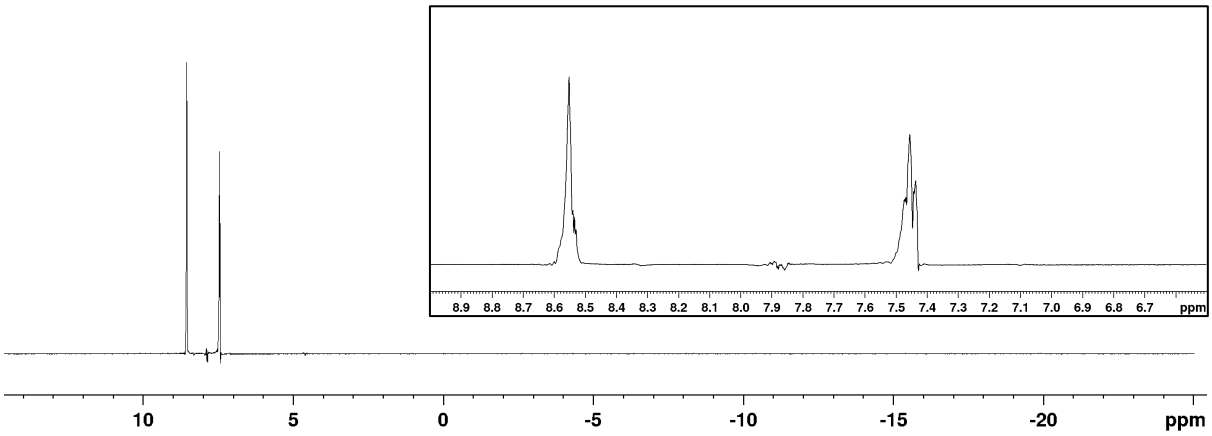


Figure S11: SABRE-DREAM spectrum (and expansion) exciting the *ortho* and *meta* resonances of free pyridine (at 8.55 and 7.46 ppm), obtained by shaking the tube under 2 bar p-H_2 at the earth's magnetic field (~0.5 G) for 10 seconds before transport to the spectrometer.



Note: arbitrary vertical scales used for illustration purposes only. Enhancement levels (ε) of free pyridine given below.

$\varepsilon_{(ortho)} = +102$

$\varepsilon_{(meta)} = +106$

References

1. E. R. Bissell and R. W. Swansiger, *J. Chem. Eng. Data*, 1981, **26**, 234-235.
2. P. M. Richardson, S. Jackson, A. J. Parrott, A. Nordon, S. B. Duckett and M. E. Halse, *Magn. Reson. Chem.*, 2018, **56**, 641-650.
3. A. Jenne, W. Bermel, C. A. Michal, O. Gruschke, R. Soong, R. Ghosh Biswas, M. Bastawrous and A. J. Simpson, *Angew. Chem., Int. Ed.*, 2022, **61**, e202110044.
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5. H. Xu and C. Wolf, *Chem. Commun.*, 2009, DOI: 10.1039/B904188E, 3035-3037.