

Supplementary Information for

Perfluorinated Iridium Catalyst for Signal Amplification by Reversible Exchange Provides Metal-Free Aqueous Hyperpolarized [1-¹³C]-Pyruvate

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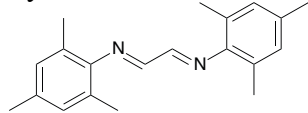
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1. Experimental Section:

- a. Synthesis of perfluorinated -SABRE ligand
based on published literature¹

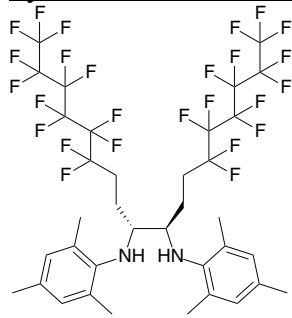
Synthesis of *N,N'*-1,2-Ethanediyldienebis[2,4,6-trimethylbenzenamine], I



Glyoxal (12.1 mL; 107 mmol) is added to a stirring solution of 2,4,6-trimethylaniline (30 mL; 214 mmol) in methanol (85 mL) at room temperature. After the solution had been stirred for 5 min few drops of formic acid (5 drops) is added to the reaction mixture. The solution is stirred for an additional three hours at room temperature. A yellow precipitate was formed, collected by filtration and wash with methanol (3x20 mL) and dry under vacuum to obtain *N*1, *N*2-dimesitylethane-1,2-diimine. The material was used directly in the following step without further purification.

¹H NMR: (400 MHz, CDCl₃) 8.10 (s, 2H), 6.91 (s, 4H), 2.29 (s, 6H), 2.16 (s, 12H). Data in accordance with those previously reported.¹

Synthesis of 1*H*,1*H*,2*H*,2*H*-perfluorinated octyl-*N,N'*-bis(2,4,6-trimethylphenyl)-9,10-diamine, II



A 1.7 M solution of tert-butyl lithium in pentane (5 mL, 9 mmol, 8 eq.) was added to a solution of 1*H*,1*H*,2*H*,2*H*-perfluorinated octyl iodide (2 g, 4 mmol, 4 eq.) in dry Et₂O (60 mL) at -78 °C. After the mixture had been stirred for 20 min at -78 °C, the solid *N,N'*-dimesitylethanediiimine (0.30 g, 1.03 mmol, 1 eq.) was added portion wise. The reaction mixture was stirred for 4 h. The reaction was slowly warmed to -30 °C and quenched with a saturated solution of ammonium chloride (0.6 mL).

Water (20 mL) was added, the organic layer was separated, and the aqueous layer was extracted with diethyl ether (3 × 15 mL). The combined organic layers were dried over anhydrous MgSO₄, and concentrated. The crude product was purified by flash column chromatography (4:1 hexane/dichloromethane (DCM)) to yield diamine, diimine, and threo-diamine (0.6 g, 60% yield, white solid).

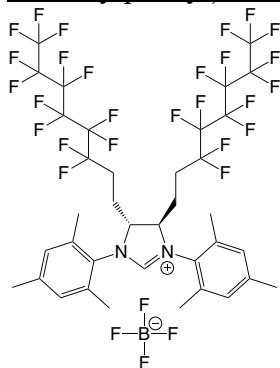
¹H NMR (400 MHz, CDCl₃) δ 1.64-1.82 (m, 2H), 2.00 (s, 12H), 2.07-2.27 (m, 4H), 2.33 (s, 6H), 2.67-2.91 (m, 2H), 2.95 (br s, 2H), 3.23 (br d, ³J_{H-H} = 10 Hz, 2H), 6.82 (s, 4H) ppm.

¹⁹F NMR (282.23 MHz, CDCl₃) δ -83.8 (t, ³J_{F-F} = 10 Hz, 6F), -114.4 (m, 4F), -121.9 (m, 4F), -122.8 (m, 4F), -123.4 (m, 4F), -126.1 (m, 4F) ppm.

¹³C NMR (400 MHz, CDCl₃) δ 18.3, 20.3, 21.7, 29.2, 57.1, 107-115, 117.4, 118.7, 128.6, 129.9, 131.4, 140.4 ppm.

MS (ESI), *m/z*: 990 [M+H]⁺.

Synthesis of trans-4,5-bis(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl)-1,3-bis(2,4,6-trimethylphenyl)-4,5-dihydroimidazolium tetrafluoroborate, **III**



A mixture of 1*H*,1*H*,2*H*,2*H*-Perfluorinated octyl-*N,N'*-bis(2,4,6-trimethylphenyl)-9,10-diamine (0.250 g, 0.25 mmol) from Example 1, ammonium tetrafluoroborate (~10% molar excess) (0.030 g, 0.25 mmol), and triethyl orthoformate (0.5 mL) was heated to 125 °C and stirred for 15 h. After cooling to room temperature, the solution was evaporated and the solid was triturated with diethyl ether (6 × 3 mL). The residue was redissolved in acetone, filtered, and concentrated to yield to the dihydroimidazolium tetrafluoroborate. (0.2 g, 70% yield, yellowish solid).

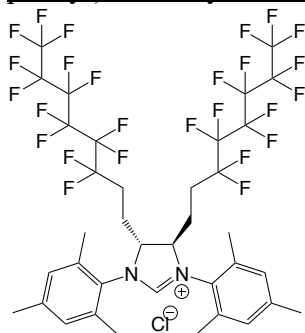
¹H NMR (400 MHz, acetone-*d*₆) δ 2.15-2.70 (m, 8H), 2.34 (s, 6H), 2.51 (s, 6H), 2.94 (br s, 2H), 5.16 (m, 2H), 7.17 (s, 4H), 9.06 (s, 1H) ppm.

¹⁹F NMR (400 MHz, acetone-*d*₆) δ -81.6 (t, ⁴J_{F-F}=10 Hz, 6F), -115.1 (m, 4F), -122.5 (m, 4F), -123.6 (m, 4F), -124.5 (m, 4F), -126.9 (m, 4F), -150.9 (4F, BF₄) ppm.

¹³C NMR (400 MHz, acetone-*d*₆) δ 17.4, δ 17.7, 19.9, 24.4, 26.2, 67.9, 105-118, 118, 118.9, 129.5, 130.1, 130.5, 135.8, 136, 140.8, 159.7, ppm.

MS (ESI) *m/z*: 999 [M+H]⁺.

Synthesis of trans-4,5-Bis(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl)-1,3bis(2,4,6-trimethylphenyl)-4,5-dihydroimidazolium chloride, **IV**



Dihydroimidazolidium tetrafluoroborate salt (0.5 g, 0.46 mmol) was dissolved in MeOH (1.5 mL) and passed through a short column of ion exchange resin Amberlite 400. The column was washed with MeOH until no spot was visible via TLC under UV. The solvent was removed, and the resulting yellowish solid was dried with a vacuum pump to yield product (0.48 g, 99%).

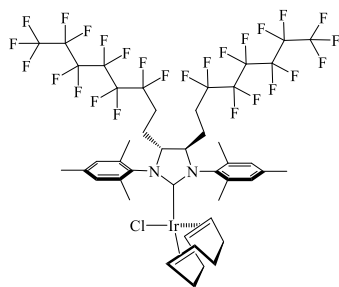
¹H NMR (400 MHz, acetone-*d*₆) δ 2.15-2.70 (m, 8H), 2.34 (s), 2.51 (s, 6H), 2.94 (br s), 5.16 (m, 2H), 7.17 (s, 4H), 9.06 (s, 1H) ppm.

¹⁹F NMR (400 MHz, acetone-*d*₆) δ -81.8 (t, ⁴J_{F-F}=10 Hz, 6F), -115.1 (m, 4F), -122.5 (m, 4F), -123.6 (m, 4F), -124.5 (m, 4F), -126.9 (m, 4F) ppm.

¹³C NMR (100 MHz, acetone-*d*₆) δ 17.4, δ 17.7, 19.9, 24.4, 26.2, 67.9, 105-118, 118, 118.9, 129.5, 130.1, 130.5, 135.8, 136, 140.8, 159.7 ppm.

MS (ESI) *m/z*: 999 [M+H]⁺.

b. Synthesis of the perfluorinated-SABRE catalyst, V



Potassium tert-butoxide (112 mg, 1.00 mmol, 2.5 eq.) was added to a stirred solution of trans-4,5-bis(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl)-1,3-bis(2,4,6-trimethylphenyl)-4,5-dihydroimidazolium chloride (320 mg, 0.88 mmol, 2.2 eq.) in tetrahydrofuran (10 mL) at room temperature in a glove box. The resulting suspension was stirred for 30 minutes. A solution of [Ir(COD)Cl]₂ (268 mg, 0.40 mmol, 1.0 eq.) was added and the resulting solution was stirred at room temperature overnight (Cowley et al., *J. Am. Chem. Soc.*, 133, 6134–6137 (2011)). The solvent was removed under reduced pressure to give the crude product and dried overnight under vacuum. This sample was purified by flash chromatography with a 5:4 EtOAc:Hexane to give 104 mg (40% yield).

¹H NMR (400 MHz, CDCl₃) δ 1.04, 1.18, 1.49, 1.76, 2.13, 2.22, 2.25, 2.4, 2.48, 2.66, 2.94, 3.73, 4.01, 4.2, 6.84, 6.91 6.97 ppm.

¹⁹F NMR (400 MHz, CDCl₃) δ -81.08, -114.33, -122.05, -123.07, -124.02, -126.39 ppm.

¹³C NMR (100 MHz, CDCl₃) δ 18.7, 20.95, 21.07, 21.34, 26.45, 27.21, 29.83, 30.74, 31.42, 49.93, 55.12, 68.11, 68.47, 83.26, 86.69, 108.32, 111.0, 112.95, 115.85, 117.75, 118.69, 129.02, 129.3, 130.36, 130.55, 134.71, 134.8, 135.32, 136.33, 137.92, 138.28, 138.45, 138.5, 206.72 ppm.

MS (ESI) m/z: 1299 [M+H]⁺.

c. ¹H, ¹³C, ¹⁹F, COSY, HMBC, HSQC NMR spectrum

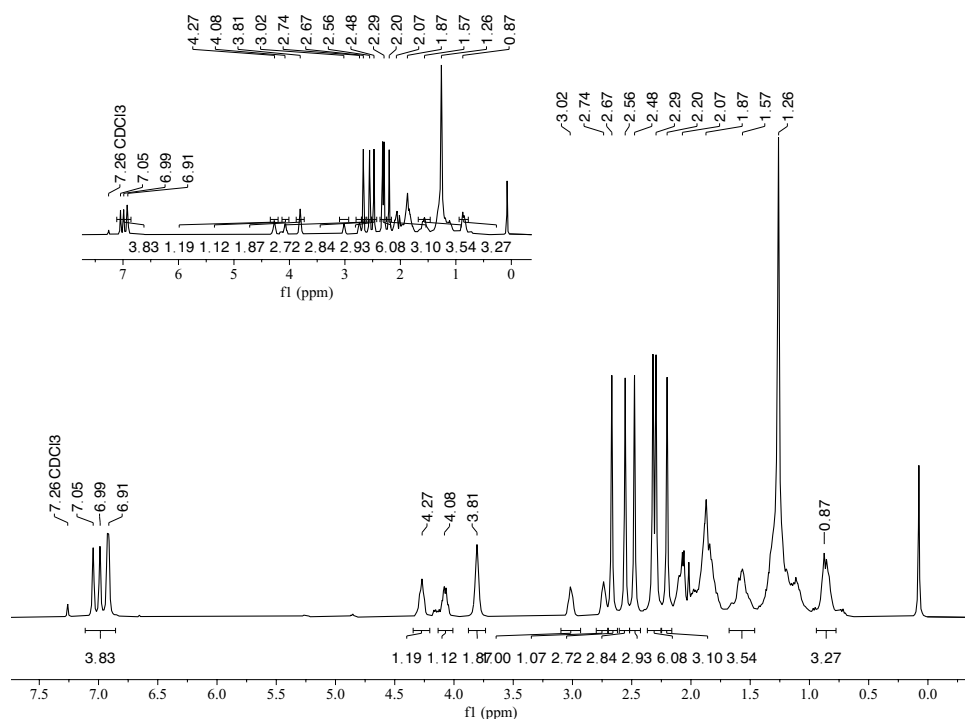


Fig S1: ¹H NMR perfluorinated-SABRE pre-catalyst

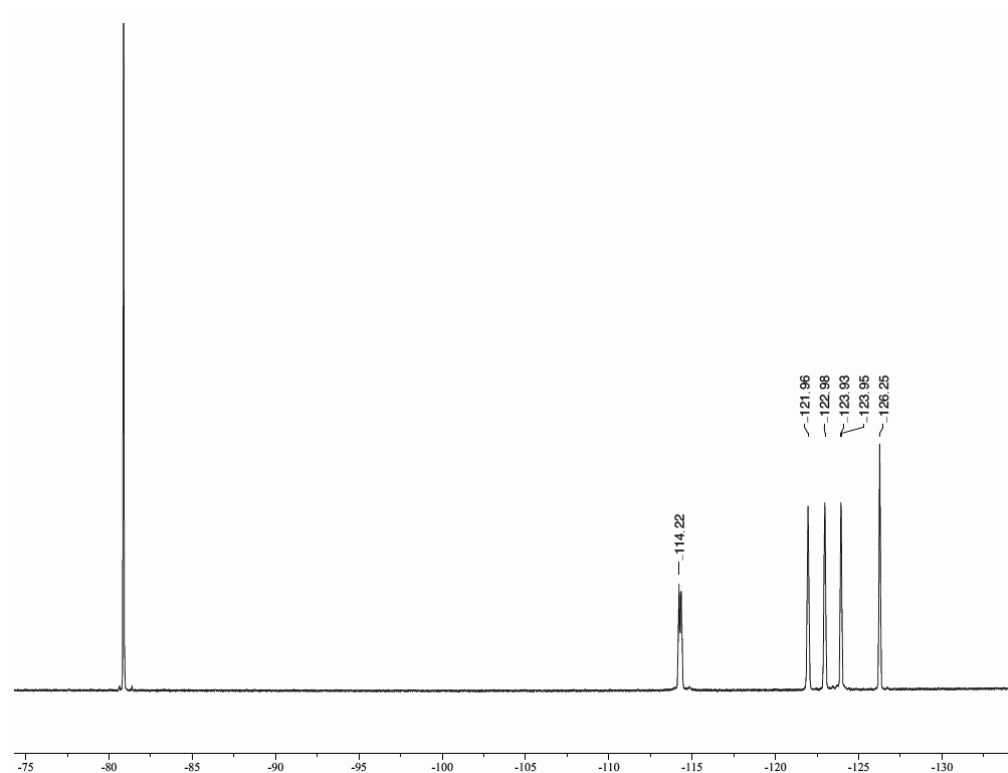


Fig S2: ^{19}F NMR perfluorinated-SABRE pre-catalyst

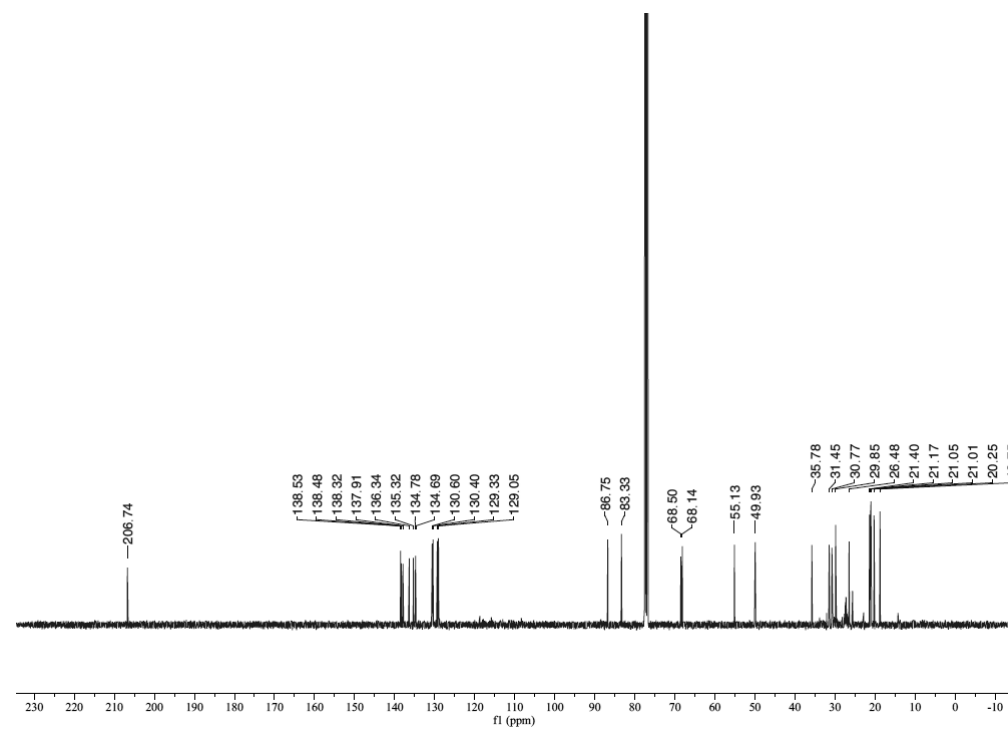


Fig S3: ^{13}C NMR perfluorinated-SABRE pre-catalyst

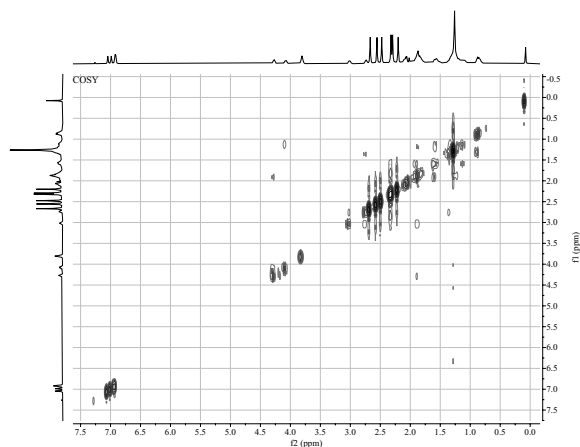


Fig S4: COSY NMR perfluorinated-SABRE pre-catalyst

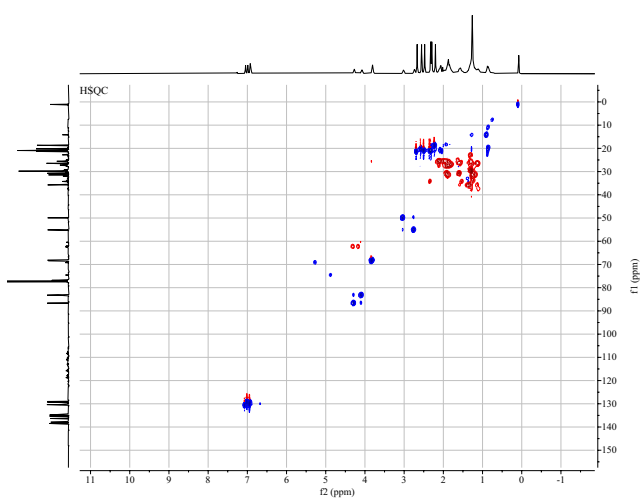


Fig S5: HSQC NMR perfluorinated-SABRE pre-catalyst

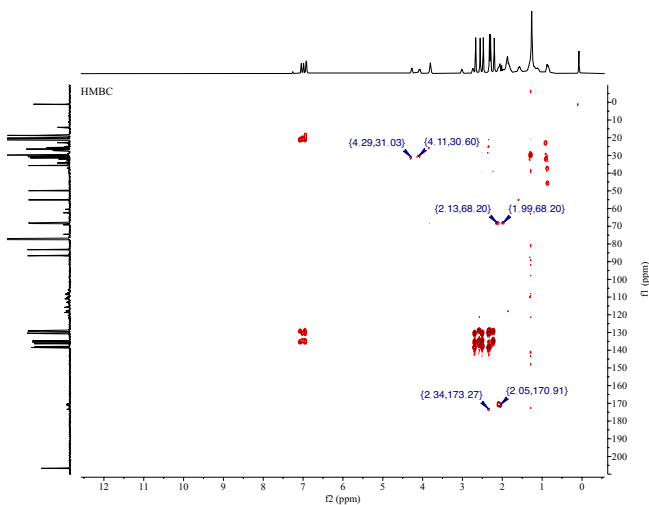


Fig S6: HMBC NMR perfluorinated-SABRE pre-catalyst

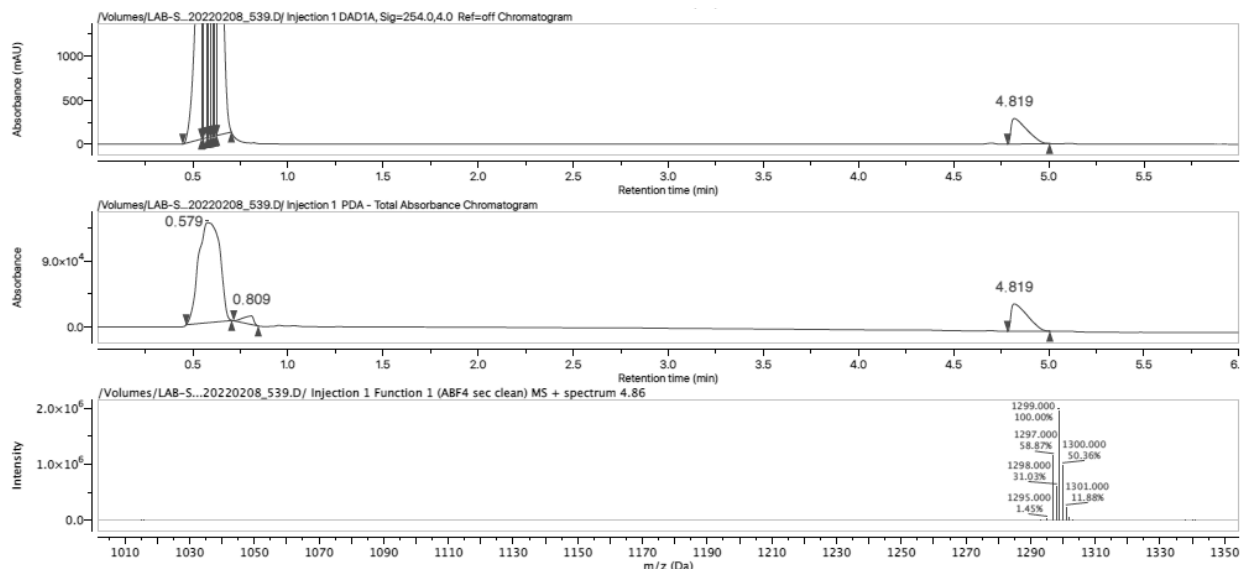


Fig S7: LCMS perfluorinated-SABRE pre-catalyst

d. Sabre sheath apparatus and experiments

SABRE hyperpolarization setup:

The experimental setup has been used previously.^{3,4} *p*-H₂ was produced by a distribution system reported earlier⁵ and stored in aluminum cylinder (1-20 days). The *p*-H₂ percentage used in this study varied from 95% to 70% enrichment. The *p*-H₂ bubbling had a flow rate set at 90 standard cubic centimeters per minute (sccm). The *p*-H₂ flow was directed via Polytetrafluoroethylene (PTFE) tubing using the MFC (Sierra Instruments SmartTrak 100 series) and directed to a conventional 5 mm medium-wall NMR tube (Norell) to allow for *p*-H₂ bubbling through the sample. The setup for bubbling parahydrogen through the sample tubes consisted of a mass flow controller, which regulated the flow of pressurized parahydrogen, a safety valve which released pressure above 100 psi from the setup, and a combination of manual valves. The polarization transfer magnetic field was established with an apparatus consisting of a solenoid coil placed inside a three-layered mu-metal shield (6 in. ID & 15 in. in length, part number ZG-206, Magnetic Shield Corp., Bensenville, IL). The magnetic field was created using a custom-built solenoid coil (41 mm diameter (40mm core, 20 cm long windings with 220 turns AWG20 (0.9 mm) Cu wire and with 220 Ω resistor in series) and a triple independent channel DC power supply (KEITHLEY 2231A-30-3). The solenoid coil was driven with a variable-resistance decade box in series to provide finer control of the internal magnetic field inside the shield.

Activation of the catalyst took place for 15 min at ambient temperature and magnetic field. For polarization build up, if not stated otherwise, the sample was placed in the 0.4 μT field and an isopropanol/dry ice bath or water/ice for the desired temperature.

Samples Preparation: The SABRE samples were prepared in CD₃OD using the stated concentration of sodium pyruvate (in most cases 20 or 30 mM [1-13 C]-labeled), the perfluorinated iridium catalyst at 6 mM concentration, synthesized as described below, with 40 mM dimethyl sulfoxide (DMSO). Samples were prepared in a glove box to avoid any oxygen contamination. The solvents (methanol, ethyl acetate and D₂O) were previously degassed before use. The samples were filled into medium wall NMR tubes for all SABRE-SHEATH experiments.

^{13}C polarization detection and quantification: 1.81 T benchtop NMR system (Spinsolve 80 Carbon, Magritek, Germany) was used to measure the hyperpolarized and thermal NMR signals of the samples. The data acquisition parameters for Magritek Spinsolve 13C (Expert Software) were set at 329 ppm (~ 6.6 kHz) spectral width, 3500 ms repetition time, 20 μs pulse acquisition delay 90-degree pulse angle using -6.76 dB pulse amplitude and 90 μs pulse length, 2 μs pre-acquisition delay, 16 dB receiver gain, single scan with 8192 data points, 150 μs dwell time and zero fill factor of 2.

The ^{13}C signal enhancement was computed by comparing the HP signal to the external ^{13}C signal thermal signal reference (4 M sodium [$1\text{-}^{13}\text{C}$]acetate) using eq 1

$$\varepsilon(^{13}\text{C}) = \frac{S_{\text{HP}}}{S_{\text{REF}}} \cdot \frac{C_{\text{REF}}}{C_{\text{HP}}} \cdot \frac{A_{\text{REF}}}{A_{\text{HP}}} \quad (1)$$

where S_{HP} and S_{REF} are ^{13}C signals from HP [$1\text{-}^{13}\text{C}$]pyruvate and thermal signal reference [$1\text{-}^{13}\text{C}$]acetate, respectively, C_{REF} and C_{HP} are concentrations of thermal signal reference [$1\text{-}^{13}\text{C}$]acetate (4 M) and HP [$1\text{-}^{13}\text{C}$]pyruvate, respectively, and A_{REF} and A_{HP} are effective cross sections of the NMR tubes for thermal signal reference [$1\text{-}^{13}\text{C}$]acetate and HP [$1\text{-}^{13}\text{C}$]pyruvate samples, respectively. The percentage of ^{13}C polarization ($\%P^{13}\text{C}$) was computed by multiplying the signal enhancement ($\varepsilon^{13}\text{C}$) by thermal ^{13}C nuclear spin polarization at 1.81 T ($1.5681 \times 10^{-4} \%$) in accordance with Equation S2: $\%P^{13}\text{C} = \varepsilon(^{13}\text{C}) \times 1.56181 \times 10^{-6} \times 100$.

e. High-resolution 600 MHz ^1H NMR spectroscopy of hydride spectral region

A solution employed for ^{13}C SABRE-SHEATH hyperpolarization was taken to a 400 MHz NMR spectrometer for acquisition of a proton spectrum after initial activation and detection of ^{13}C SABRE-SHEATH hyperpolarization. The solution was prepared in a pressure NMR tube and contained [$1\text{-}^{13}\text{C}$]pyruvate (30 mM), DMSO (48 mM), 75% enriched $p\text{-H}_2$, and perfluorinated-SABRE catalyst (6 mM) in CD_3OD at room temperature $^\circ\text{C}$,

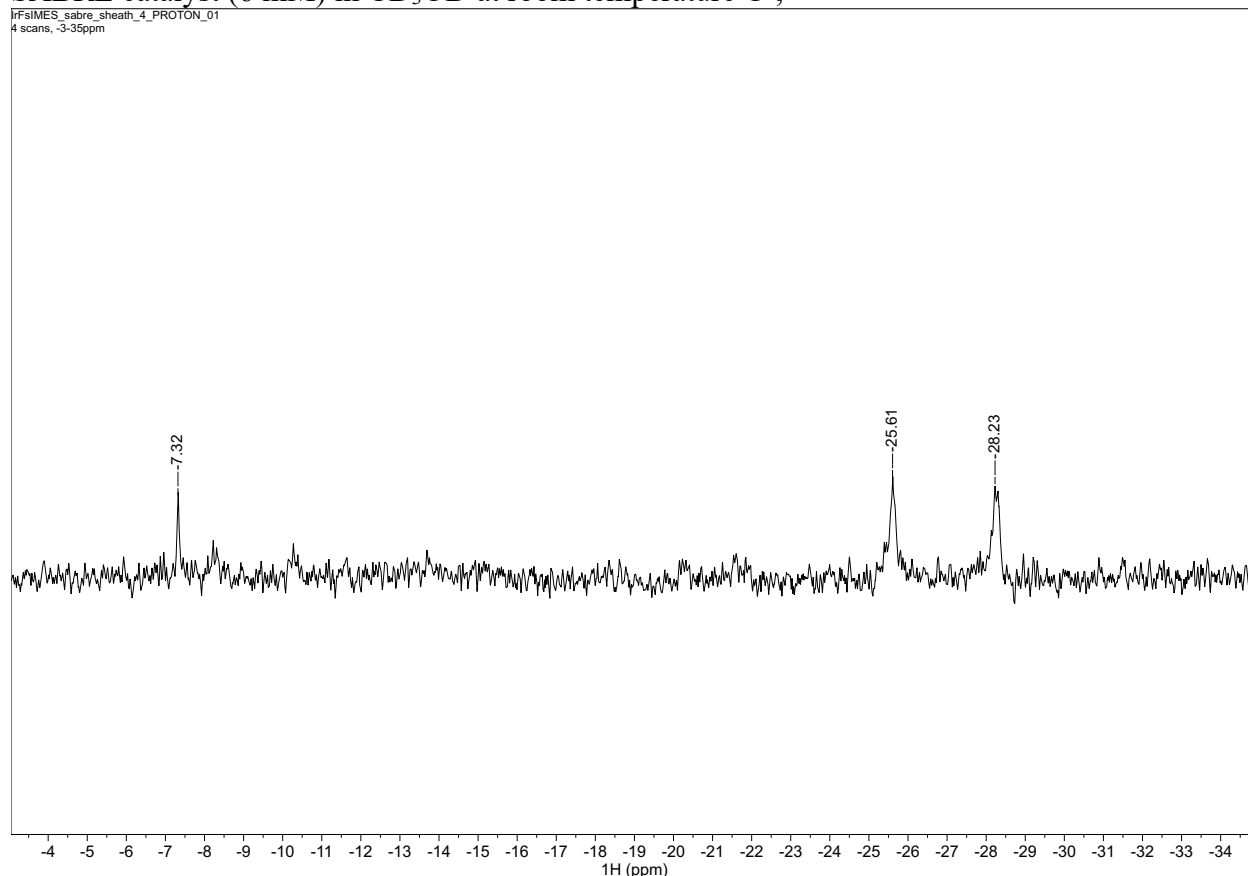


Fig S8: ^1H spectrum of hydride region was acquired approximately 15 seconds after SABRE-SHEATH hyperpolarization. Acquisition parameters: 400 MHz, number of scans = 4, RF probe temperature = 25 °C.

f. Perfluorinated-SABRE catalyst efficiency on SABRE-SHEATH experiments

i. Activation time

The perfluorinated-SABRE catalyst activation (<15 mins) is performed by bubbling ~95% $p\text{-H}_2$ at a flow rate of 90 standard cubic centimeters per minute (sccm) at 8 atm $p\text{-H}_2$ partial pressure

Activation time

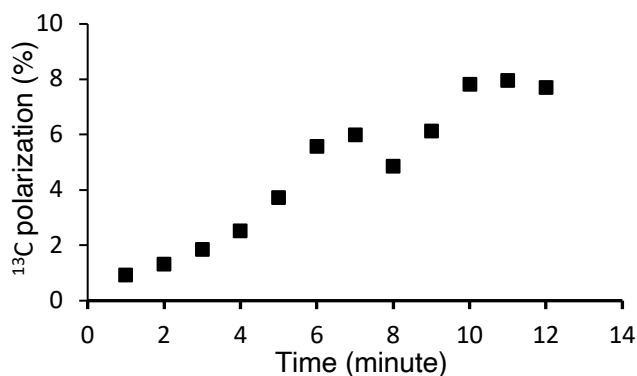


Fig S9: Activation time

ii. Pressure and flow

Hyperpolarization of $[1\text{-}^{13}\text{C}]\text{pyruvate}$ was performed using SABRE in SHield Enables Alignment Transfer to Heteronuclei (SABRE-SHEATH) was optimized by studying the effects of parahydrogen pressure and flow rate as well as the effect of magnetic transfer field, temperature, and concentration of the fluorinated catalyst and DMSO. The relaxation dynamics of the $[1\text{-}^{13}\text{C}]\text{pyruvate}$ was studied as well.

Optimization of $p\text{-H}_2$ parameters such as the pressure and flow rate are evaluated. The NMR samples consisted in 30 mM sodium $[1\text{-}^{13}\text{C}]\text{pyruvate}$, 2.6 mM fluorinated SABRE catalyst, and 40 mM dimethyl sulfoxide (DMSO), the mixing field was at 0.4 μT and temperature at 0°C.

Parahydrogen Flow

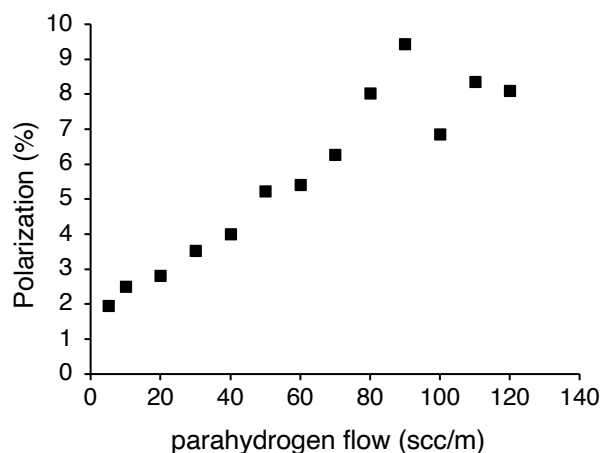


Fig S10: ParaH₂ flow

Parahydrogen pressure

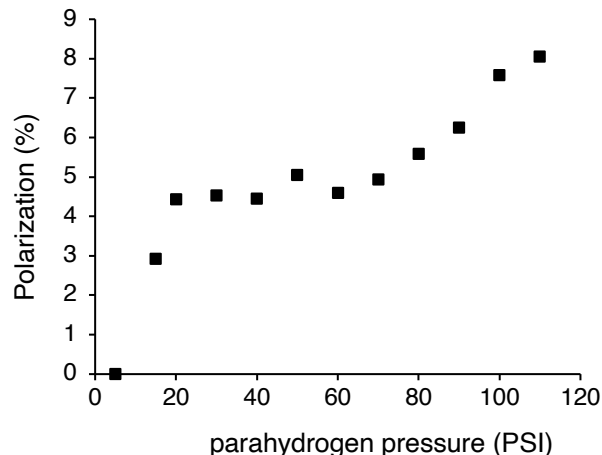


Fig S11: ParaH₂ pressure

iii. Catalyst concentration and DMSO concentration

The catalyst and DMSO concentration were optimized based on the optimum conditions established above, temperature 0°C, magnetic transfer field at 0.4 μ T, *p*-H₂ flow and pressure 90 ssc/m and 110 PSI in the same way and showed here.

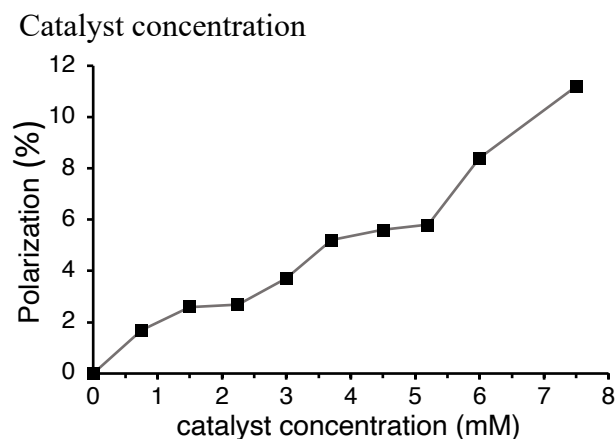


Fig S12:Catalyst concentration

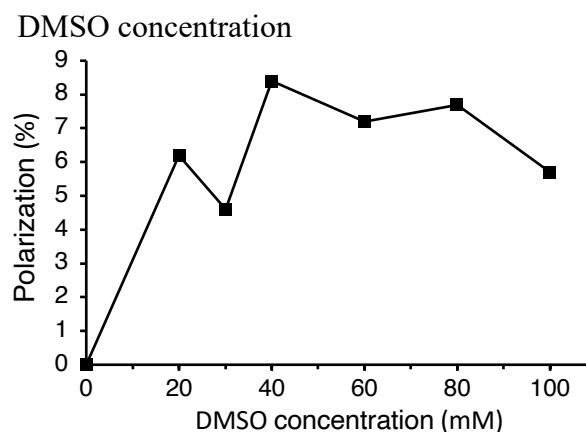


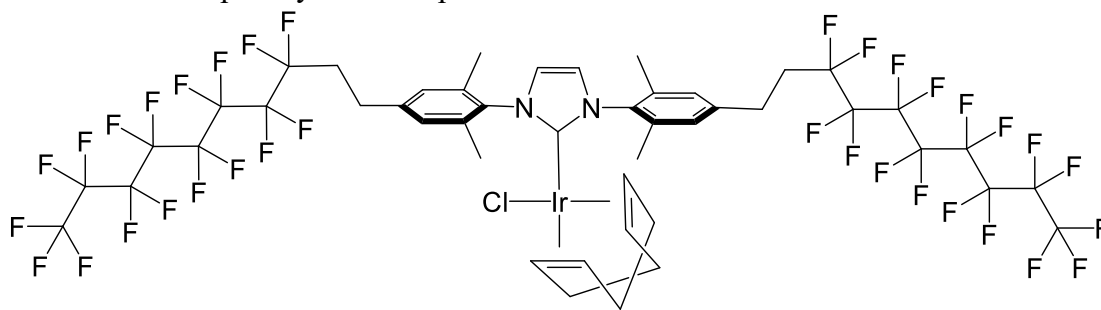
Fig S13:DMSO concentration

g. Alternative perfluorinated-SABRE catalysts, [IrCl(COD)(f-IMes)] and their SABRE-SHEATH activity.

i. 1,3-bis(2,6-dimethyl-4-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-tridecafluorooctyl)phenyl)-1H-imidazol-2-ylidene as ligand

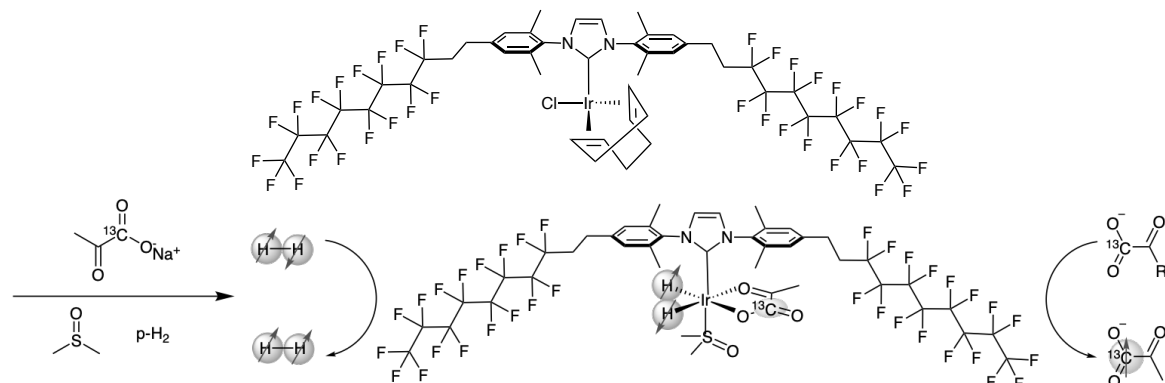
The ligand synthesis is based on published literature¹

The Iridium complex synthesis is performed as follow:



Potassium tert-butoxide (2.5 eq.) was added to a stirred solution of 1,3-bis(2,6-dimethyl-4-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-tridecafluorooctyl)phenyl)-1H-imidazol-3-ium-2-ide, chloride (2.2 equiv) in tetrahydrofuran at room temperature in a glove box. The resulting suspension was stirred for 30 min. A solution of [Ir(COD)Cl]₂ (1.0 eq.) was added, and the resulting solution was stirred at room temperature for 2 hours. The solvent was removed under reduced pressure to give the crude product and dried overnight under vacuum. This sample was purified with flash chromatography DCM/hexane (4:1) to obtain the fluorinated SABRE catalyst. MS (ESI) *m/z* (%): 1469 [M-Cl]⁺ (100).

The hyperpolarization of sodium pyruvate using the PERFLUORINATED -SABRE catalyst [IrCl(COD)(f-IMes)] with f-IMes = 1,3-bis(2,6-dimethyl-4-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-tridecafluorooctyl)phenyl)-1H-imidazol-2-ylidene is shown in Scheme 1.



Scheme S1: Schematic of Hyperpolarization of [1-¹³C]pyruvate using the PERFLUORINATED - SABRE catalyst [IrCl(COD)(f-IMes)] with f-IMes = 1,3-bis(2,6-dimethyl-4-(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-tridecafluorooctyl)phenyl)-1H-imidazol-2-ylidene with dimethyl sulfoxide as co-ligand

The SABRE samples were prepared in CD₃OD, using 40 mM sodium [1-¹³C]pyruvate, 6.6 mM perfluorinated SABRE catalyst, and 50 mM dimethyl sulfoxide (DMSO). The SABRE samples were exposed to the SABRE-SHEATH hyperpolarization conditions with the same set-up and optimum conditions described in the main text of the manuscript. The NMR tubes were pressurized (110 psi, i.e., approximately 8 bar total pressure) with *p*-H₂ bubbling through the solution at a flow of 90 scc/m to activate the catalyst and to hyperpolarize the sodium [1-¹³C]pyruvate solution. Activation of the catalyst took place for about 15 minutes at ambient temperature and magnetic field. For polarization build-up, the sample was placed in the magnetic field (typically about 0.4 μT) and a water bath between -5 °C and 5 °C to regulate. The spectrum was acquired immediately following manual sample transfer to a 1.8 T benchtop NMR after 5 seconds, and the results are set forth in the top spectrum below. In addition, the bottom spectrum shows a single-scan thermally polarized ¹³C signal from 4 M sodium [1-¹³C] acetate using similar acquisition parameters. The signal enhancement is ε~16900 and polarization is about $P(^{13}\text{C}) \sim 2.17\%$.

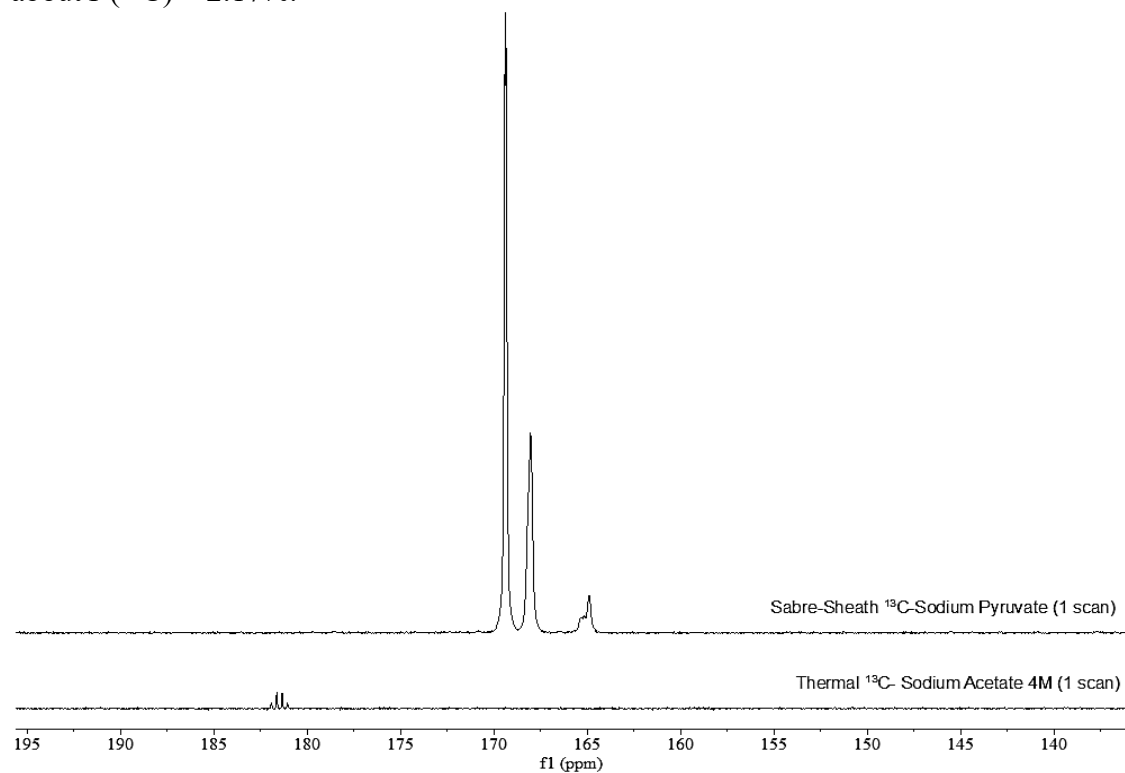
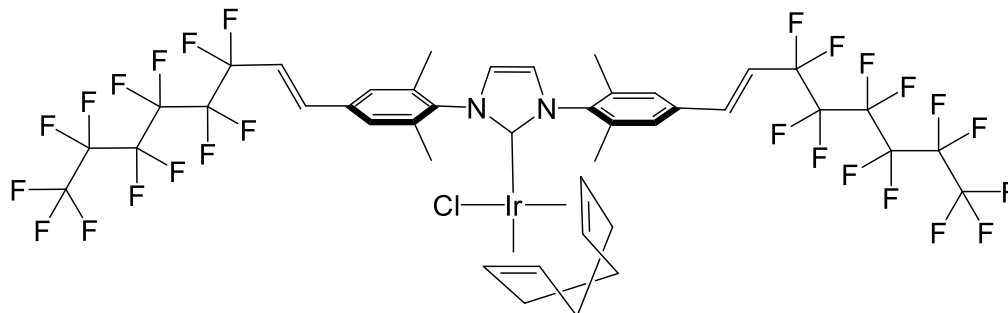


Fig S14: Comparison of a representative spectrum of ^{13}C -hyperpolarized $[1-^{13}\text{C}]$ -pyruvate with signal enhancement ε of ~ 16900 fold, corresponding to $P_{^{13}\text{C}}$ of $\sim 2.17\%$, as evidenced by the top spectrum as compared to the bottom spectrum showing a single-scan thermally polarized ^{13}C signal from 4 M sodium $[1-^{13}\text{C}]$ acetate using similar acquisition parameters

ii. 1,3-bis(2,6-dimethyl-4-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooct-1-en-1-yl)phenyl)-1H-imidazol-3-ium-2-ide as ligand

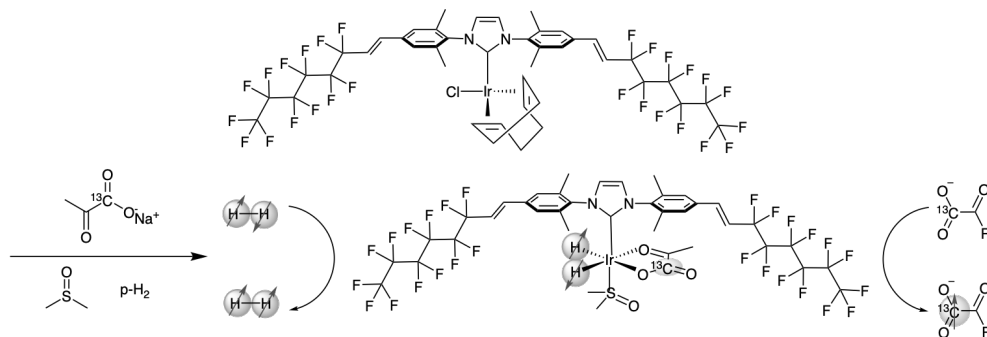
The ligand synthesis is based on published literature¹

The Iridium complex synthesis is performed as follow:



Potassium tert-butoxide (2.5 eq.) was added to a stirred solution of 1,3-bis(2,6-dimethyl-4-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooct-1-en-1-yl)phenyl)-1H-imidazol-3-ium-2-ide, chloride (2.2 equiv) in tetrahydrofuran at room temperature in a glove box. The resulting suspension was stirred for 30 min. A solution of $[\text{Ir}(\text{COD})\text{Cl}]_2$ (1.0 eq.) was added and the resulting solution was stirred at room temperature for 2 hours. The solvent was removed under reduced pressure to give the crude product and dried overnight under vacuum. This sample was purified with flash chromatography DCM/hexane (4:1) to obtain the fluorinated SABRE catalyst. MS (ESI) m/z (%): 1266 $[\text{M}-\text{Cl}]^+$ (100).

The hyperpolarization of sodium pyruvate using the perfluorinated-SABRE catalyst $[\text{IrCl}(\text{COD})(\text{f-IMes})]$ with $\text{f-IMes} = 1,3\text{-bis}(2,6\text{-dimethyl-4-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooct-1-en-1-yl)phenyl)-1H-imidazol-3-ium-2-ide}$ is shown in Scheme 2. Scheme 2. Hyperpolarization of $[1-^{13}\text{C}]$ pyruvate using the shown perfluorinated SABRE catalyst with dimethyl sulfoxide as co-ligand.



Scheme S2: Schematic of Hyperpolarization of $[1-^{13}\text{C}]$ pyruvate using the perfluorinated-SABRE catalyst $[\text{IrCl}(\text{COD})(\text{f-IMes})]$ with $\text{f-IMes} = 1,3\text{-bis}(2,6\text{-dimethyl-4-(3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooct-1-en-1-yl)phenyl)-1H-imidazol-3-ium-2-ide}$ with dimethyl sulfoxide as co-ligand

In the reaction scheme above, the efficient hyperpolarization transfer from $p\text{-H}_2$ -derived hydrides to the ^{13}C nuclear spin of $[1-^{13}\text{C}]$ pyruvate was attained by performing SABRE in sub-microtesla magnetic fields using SABRE in SHield Enables Alignment Transfer to Heteronuclei (SABRE-

SHEATH) using a solution mixture of $[\text{IrCl}(\text{H})_2(\text{DMSO})_2(\text{F-IMes})]$, $[1\text{-}^{13}\text{C}]\text{pyruvate}$ and p-H_2 in deuterated methanol.

The SABRE samples were prepared in CD_3OD , using 20 mM sodium $[1\text{-}^{13}\text{C}]\text{pyruvate}$, 7.6 mM perfluorinated SABRE catalyst shown in Scheme S2, and 50 mM dimethyl sulfoxide (DMSO). The SABRE samples were exposed to the SABRE-SHEATH hyperpolarization conditions with the same set-up and optimum conditions described in the main text. The NMR tubes were pressurized (110 psi, i.e., approximately 8 bars total pressure) with p-H_2 bubbling through the solution at a flow of 90 scc/m to activate the catalyst and to ^{13}C -hyperpolarize the sodium $[1\text{-}^{13}\text{C}]\text{pyruvate}$ solution. Activation of the catalyst took place for about 15 minutes at ambient temperature and magnetic field. For polarization build-up, the sample was placed in the magnetic field (typically about $0.4\text{ }\mu\text{T}$) and a water bath set between $-5\text{ }^\circ\text{C}$ and $+5\text{ }^\circ\text{C}$ to regulate the reaction temperature. The spectrum was acquired immediately following manual sample transfer to a 1.8 T benchtop NMR after 5 seconds, and the results are set forth in the top spectrum below. In addition, the bottom spectrum shows a single-scan thermally polarized ^{13}C signal from 4 M sodium $[1\text{-}^{13}\text{C}]\text{acetate}$ using similar acquisition parameters. As is apparent from the results set forth, the signal enhancement is $\varepsilon \sim 19000$ and polarization is about $P(^{13}\text{C}) \sim 4.91\%$.

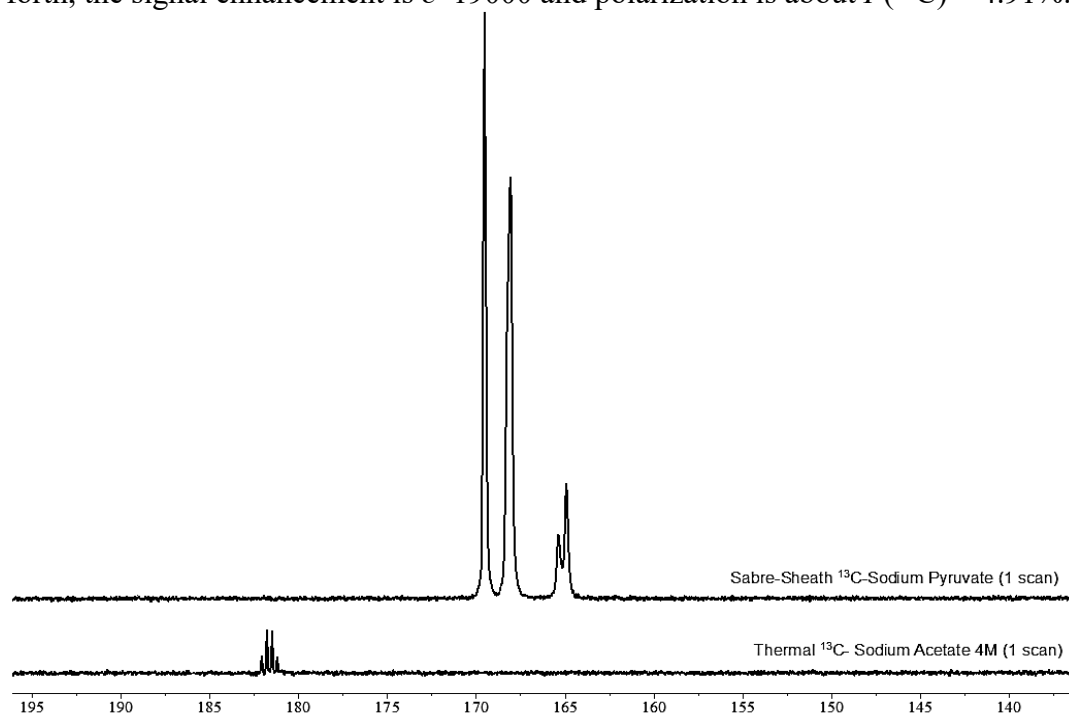


Fig S15: Comparison of a representative spectrum of ^{13}C -hyperpolarized $[1\text{-}^{13}\text{C}]\text{-pyruvate}$ with signal enhancement ε of ~ 19000 fold, corresponding to $P_{^{13}\text{C}}$ of $\sim 4.91\%$, as evidenced by the top spectrum as compared to the bottom spectrum showing a single-scan thermally polarized ^{13}C signal from 4 M sodium $[1\text{-}^{13}\text{C}]\text{acetate}$ using similar acquisition parameters.

2. Solution quantification by absolute qNMR with External Calibration (EC)²

Once the Sabre sheath- ReDiss samples were prepared and measured on the benchtop NMR for hyperpolarized signal, the samples were collected and measured for ^1H NMR spectra acquisition using a 400MHz NMR system. This method allowed us to quantify in millimolar concentration the residual solvent of the thermal samples (e.g., CH_3OD , CH_3OH , EtOAc , etc.). Using an external standard reference sample (5 mM benzoic acid acid), the concentrations of the residual solvent, the pyruvate concentration were determined. The proton spectrum was acquired under quantitative

conditions to ensure the samples gained thermal equilibrium polarization between scans: acquisition time and recycle delay of 8.4 and 30s, respectively.

The qNMR quantification process was repeated 3 times on different samples and gave consistent results.

Table S1: NMR analysis of the aqueous solution after re-D.

Molecule	Sample 1	Sample 2	Sample 3	Average	deviation
Ethyl Acetate	808.67	854.17	598.84	753.89	136.19
DMSO	6.53	12.56	9.59	9.56	3.015112
Pyruvate	3.00	4.92	3.48	3.80	0.9992

*All concentrations are reported in mM and were calculated via an external calibrant of 5 mM Benzoic Acid qNMR standard prepared by Cambridge Isotopes Laboratories.

3. Solution quantification by LC-MS and GC-MS.

All solvents were purchased from commercial sources and were LCMS grade or better. Sodium pyruvate ($^{13}\text{C}_3$, 99%) was purchased from Cambridge Isotope Laboratories.

Quantitation of pyruvate: Liquid chromatography/mass spectrometry analysis was performed on a Waters Acquity UPLC® coupled to a Waters Xevo Q-ToF quadrupole time of flight mass spectrometer operating in electrospray ionization (ESI) in negative mode. The capillary and sampling cone voltages were set to 1000 and 10 V, respectively. Source and desolvation temperatures were set to 120 °C and 450 °C, respectively, and the cone and desolvation gas flows were set to 50 and 800 L/hour, respectively. To maintain mass accuracy, leucine enkephalin at a concentration of 2 ng/mL in 50% acetonitrile/water containing 0.1% formic acid and injected at a rate of 10 $\mu\text{L}/\text{min}$. Data was acquired in profile mode from 50 to 1200 m/z. The quantitation ion was either M-H or 2M-H. Skyline⁶ was used for mass spectrometry data processing.

The analytes were analyzed by either HILIC chromatography or reverse phase chromatography.

HILIC Method: The analytes were separated by HILIC chromatography on a Waters ACQUITY UPLC BEH amide (1.7 μm , 2.1 X 100 mm) column. Chromatographic separation was achieved with 90:10 water:acetonitrile (solvent A) and 90:10 acetonitrile:water (solvent B) each containing 10 mM NH_4OAc , pH 9. Gradient elution, with a flow rate of 0.500 mL/min, began with an initial hold of 85% B for 0.67 minutes, decreased to 50%B over 1.63 minutes, decreased to 5%B over 1.37 minutes, hold for 0.66 minutes at 5%, then return to initial conditions (85% B) in 0.04 minutes. The column was held at 85%B for 10 minutes before the next injection. The column temperature was maintained at 40 °C in a column oven. The injection volume was 3 μL and the autosampler temperature was held at 8 °C.

Reverse Phase Method: The analytes were separated by reverse phase chromatography on a Phenomenex Synergi-Hydro RP (2.5 μm , 2.1 X 100 mm) column. Chromatographic separation was achieved with water (solvent A) and acetonitrile (solvent B) each containing 0.1% formic acid. Gradient elution, with a flow rate of 0.400 mL/min, began with an initial hold of 5% of 0.30 minutes, increased to 100% over 6 minutes, hold for 0.7 minutes, then a return to initial conditions (5%) in 0.10 minutes. The total run time was 9.30 minutes. The injection volume was 3 μL and the autosampler temperature was held at 8 °C.

Preparation of Calibration Curves for pyruvate quantitation: The linearity of the method was assessed by a calibration curve in the range of 0.01– 10 mg/mL of analyte. The curves were fitted to a linear in log space least-squares linear regression method by the measurement of the peak-area ratio of the analyte to the internal standard (sodium pyruvate- $^{13}\text{C}_3$). The acceptable criterion for the calibration curve was a correlation of determination (R^2) of 0.98 or better, and that each back-calculated standard concentration must be within 20% deviation from the nominal value.

LCMS analysis of the aqueous solution after re-D.

	aqueous phase						Organic phase	
	Pyruvate- $^{13}\text{C}_1$		methanol				Pyruvate- $^{13}\text{C}_1$	
sample name	cal conc mM	SD	cal conc mM	SD	methanol %	SD	cal conc mM	SD
Sample 1	3.51	0.41	1933.5	61.64	6.2	0.2	0.01	0.00
Sample 2	5.67	1.47	2831.8	83.74	9.1	0.27	0.02	0.01
Sample 3	4.48	1.24	326.3	17.9	1	0.06	0.09	0.01
Sample 4	6.00	1.49	4288.1	144.82	13.7	0.46	0.07	0.03
Sample 5	6.64	1.65	4899.7	182.11	15.7	0.58	0.02	0.01
Sample 6	4.80	1.21	4451.7	138.64	14.3	0.44	0.12	0.08
Sample 7	2.42	0.70	6046.3	234.01	19.4	0.75	0.10	0.04
Sample 8	0.85	0.24	4272.4	132.27	13.7	0.42	0.13	0.03
Sample 9	0.69	0.19	2054.0	95.48	6.6	0.31	0.10	0.03

4. ICP-MS data.

Sample Name	Ir (ppb)
Sample 1- aqueous phase	177
Sample 2 - aqueous phase	301
Sample 3 - aqueous phase	228
Sample 4 - aqueous phase	294

Iridium used per experiment is 1mg/100 μL , the Iridium content represents 14.39% which is 1.439 g/L. The residual Iridium is 177 $\mu\text{g/L}$, which is an 8130-fold decrease in Iridium content.

5. References Cited in the Supporting Information

- (1) Hošek, J.; Rybáčeková, M.; Čejka, J.; Cvačka, J.; Kvíčala, J. Synthesis of Heavy Fluorous Ruthenium Metathesis Catalysts Using the Stereoselective Addition of Polyfluoroalkyllithium to Sterically Hindered Diimines. *Organometallics* **2015**, *34* (13), 3327–3334. <https://doi.org/10.1021/acs.organomet.5b00325>.
- (2) Pauli, G. F.; Chen, S.-N.; Simmler, C.; Lankin, D. C.; Gödecke, T.; Jaki, B. U.; Friesen, J. B.; McAlpine, J. B.; Napolitano, J. G. Importance of Purity Evaluation and the Potential of Quantitative ^1H NMR as a Purity Assay. *J. Med. Chem.* **2014**, *57* (22), 9220–9231. <https://doi.org/10.1021/jm500734a>.
- (3) Adelabu, I.; TomHon, P.; Shah Hafez Kabir, M.; Nantogma, S.; Abdulmojeed, M.; Mandzhieva, I.; Ettedgui, J.; E. Swenson, R.; C. Krishna, M.; M. Goodson, B.; Theis, T.; Y

- Chekmenev, E. Order-Unity ^{13}C Nuclear Polarization of $[1-^{13}\text{C}]$ Pyruvate in Seconds and the Interplay of Water and SABRE Enhancement. <https://doi.org/10.1002/cphc.202100839>.
- (4) Chapman, B.; Joalland, B.; Meersman, C.; Ettedgui, J.; Swenson, R. E.; Krishna, M. C.; Nikolaou, P.; Kovtunov, K. V.; Salnikov, O. G.; Koptug, I. V.; Gemeinhardt, M. E.; Goodson, B. M.; Shchepin, R. V.; Chekmenev, E. Y. Low-Cost High-Pressure Clinical-Scale 50% Parahydrogen Generator Using Liquid Nitrogen at 77 K. *Anal. Chem.* **2021**, 93 (24). <https://doi.org/10.1021/acs.analchem.1c00716>.
- (5) Adelabu, I.; Ettedgui, J.; Joshi, S. M.; Nantogma, S.; Chowdhury, M. R. H.; McBride, S.; Theis, T.; Sabbasani, V. R.; Chandrasekhar, M.; Sail, D.; Yamamoto, K.; Swenson, R. E.; Krishna, M. C.; Goodson, B. M.; Chekmenev, E. Y. Rapid ^{13}C Hyperpolarization of the TCA Cycle Intermediate α -Ketoglutarate via SABRE-SHEATH. *Anal. Chem.* **2022**.
- (6) Henderson, C. M.; Shulman, N. J.; MacLean, B.; MacCoss, M. J.; Hoofnagle, A. N. Skyline Performs as Well as Vendor Software in the Quantitative Analysis of Serum 25-Hydroxy Vitamin D and Vitamin D Binding Globulin. *Clinical chemistry*. England February 2018, pp 408–410. <https://doi.org/10.1373/clinchem.2017.282293>.