

# Chemistry–A European Journal

Supporting Information

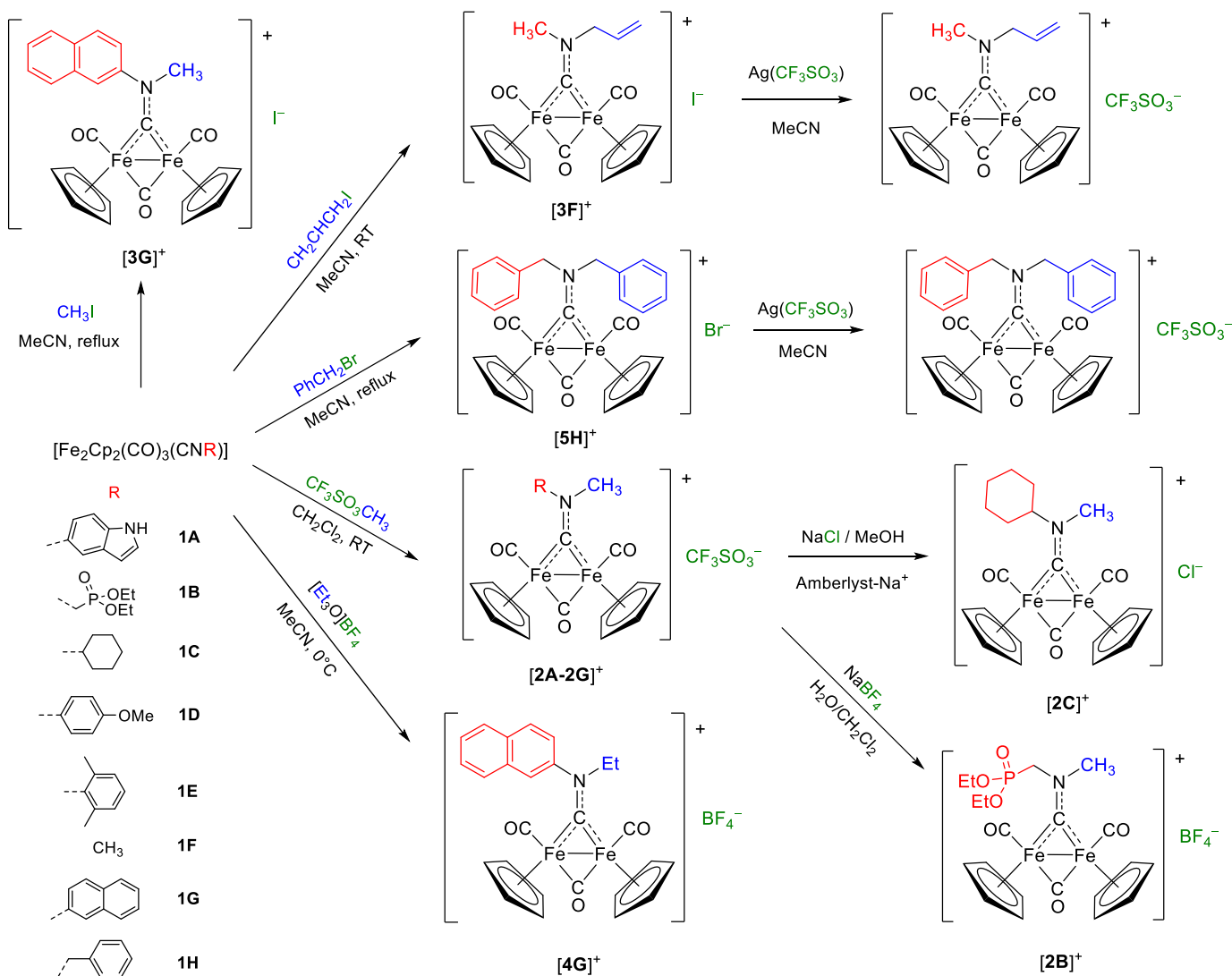
## **Easily Available, Amphiphilic Diiron Cyclopentadienyl Complexes Exhibit in Vitro Anticancer Activity in 2D and 3D Human Cancer Cells through Redox Modulation Triggered by CO Release**

Lorenzo Biancalana, Michele De Franco, Gianluca Ciancaleoni, Stefano Zacchini, Guido Pampaloni, Valentina Gandin,\* and Fabio Marchetti\*

# Supporting Information

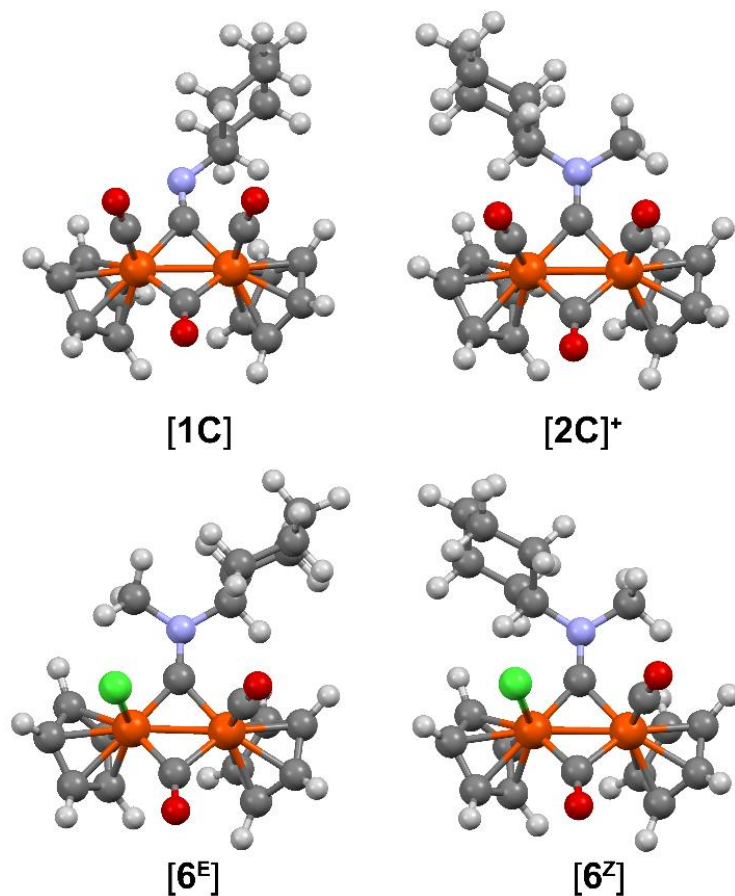
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# 1. Synthesis, X-Ray and DFT characterization.



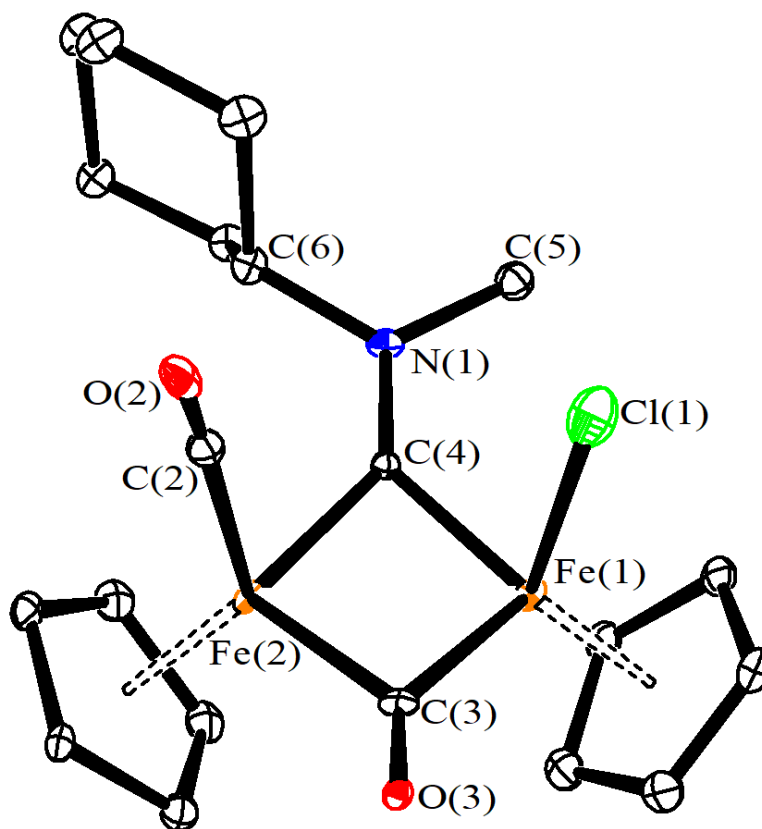
**Scheme S1.** Synthetic routes for the preparation of diiron bis-cyclopentadienyl aminocarbene complexes from  $[\text{Fe}_2\text{Cp}_2(\text{CO})_4]$ , employing different isocyanides, alkylating agents and anion exchange procedures. RT = room temperature.

**Figure S1, Table S1.** DFT-optimized geometries of selected diiron complexes, and relevant calculated bond distances (Å).



	<b>1C</b>	<b>[2C]<sup>+</sup></b>	<b>6<sup>E</sup></b>	<b>6<sup>Z</sup></b>
Fe-Fe	2.522	2.509	2.489	2.488
(CO)Fe-C <sub>carbyne</sub>	1.943	1.870	1.875	1.872
(Cl)Fe-C <sub>carbyne</sub>	=	=	1.827	1.832
C <sub>carbyne</sub> -N	1.233	1.296	1.304	1.304
μ-C-O	1.188	1.178	1.188	1.187
t-C-O	1.164	1.156	1.162	1.162

**Figure S2.** Molecular structure of **6**. Displacement ellipsoids are at the 30% probability level. H-atoms have been omitted for clarity.



## 2. IR and NMR spectra and selected spectroscopic data.

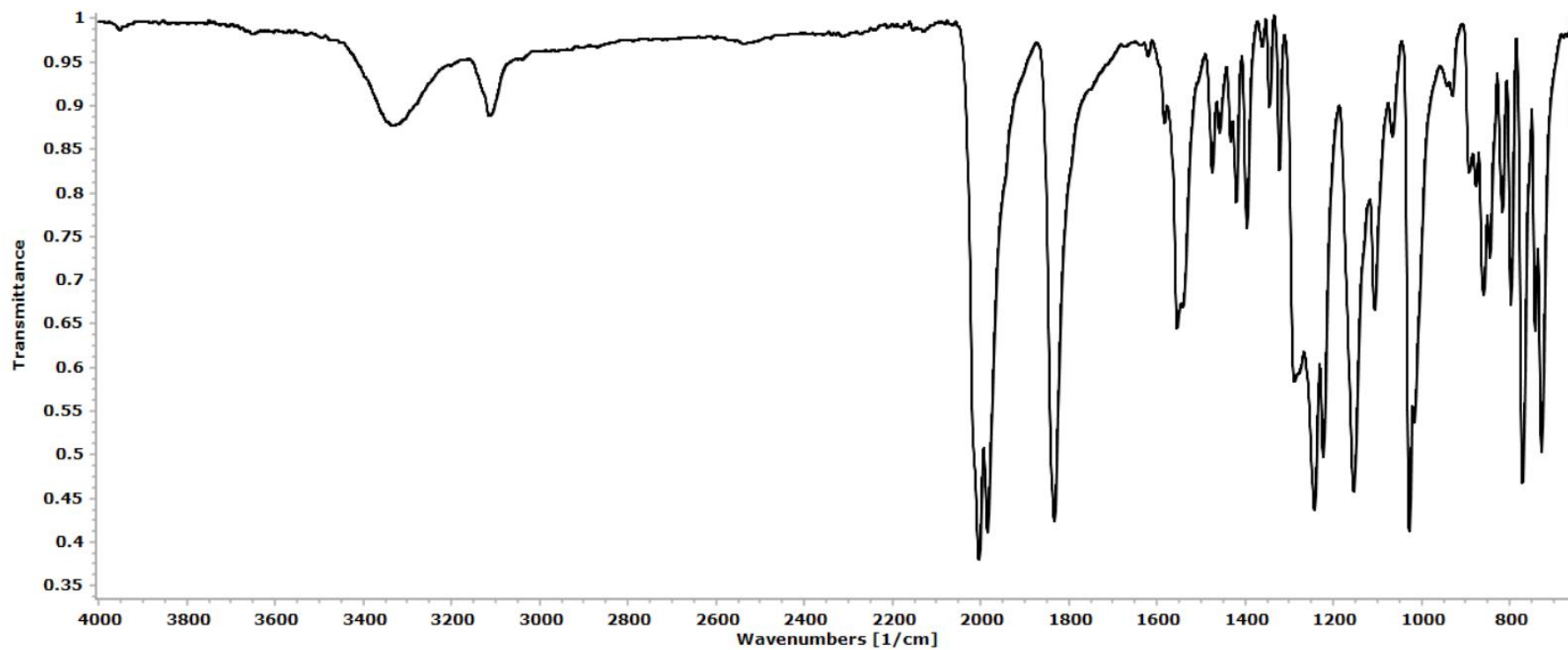
**Table S2.** Selected IR and NMR data for  $[\text{Fe}_2\text{Cp}_2(\text{CO})_2(\mu\text{-CO})\{\mu\text{-CNRR}'\}]^+$  and  $[\text{Fe}_2\text{Cp}_2\text{Cl}(\text{CO})(\mu\text{-CO})\{\mu\text{-CNMe}(\text{C}_6\text{H}_{11})\}]^+$  (**6**) complexes.

Compound	R	R'	IR ( $\text{CH}_2\text{Cl}_2$ ): $\tilde{\nu}$ / $\text{cm}^{-1}$			$^{13}\text{C}$ NMR ( $\text{CDCl}_3$ ): $\delta$ / ppm				$^1\text{H}$ NMR ( $\text{CDCl}_3$ ): $\delta$ / ppm			
			$\nu(\text{t-CO})$	$\nu(\mu\text{-CO})$	$\nu(\mu\text{-CN})$	t-CO	$\mu\text{-CO}$	$\mu\text{-CN}$	Cp	Cp	NMe	NCH	
[2A] $\text{CF}_3\text{SO}_3$ [a]	Me	Ind	2023s 1991m-sh	1836s	1542m	209.8, 209.2	255.9	324.9	91.2, 91.0	5.39, 4.65	4.53		
[2B] $\text{BF}_4$	Me	Phos	2028s 1996w	1836m	1574w	207.4, 207.1	254.7	323.7	90.3, 90.1	5.29	4.35	5.15, 4.93 ( $\text{CH}_2\text{P}$ )	
[2C] $\text{CF}_3\text{SO}_3$	Me	Cy	2020s 1988w-sh	1835m	1567w	208.5, 207.6	255.3	316.4	90.3, 90.1	5.36, 5.27	4.06	4.68 ( $\text{C}_6\text{H}_{11}$ )	
[2C] $\text{Cl}$	Me	Cy	2018s 1985w-sh	1833m	1569w	208.7, 207.8	255.8	316.2	90.7, 90.3	5.51, 5.39	4.14	4.70 ( $\text{C}_6\text{H}_{11}$ )	
[2D] $\text{CF}_3\text{SO}_3$ [c]	Me	pAnis	2021s 1989w	1836m	1540w	208.6, 207.6	254.5	324.3	90.3, 90.0	5.47, 4.82	4.57		
[2E] $\text{CF}_3\text{SO}_3$ [c]	Me	Xyl	2023s 1992w	1840m	1530w	208.6	253.9	327.8	91.3, 91.1	5.48, 4.83	4.44		
[2F] $\text{CF}_3\text{SO}_3$ [c]	Me	Me	2022s 1990w	1853m	1602w-m	209.3	257.6	315.5	90.8	5.32	4.29		
[3F] $\text{CF}_3\text{SO}_3$	Me	All	2022s 1990w	1836m	1584m	208.1, 207.8	255.3	318.1	90.20, 90.16	5.34, 5.32	4.17	5.26, 5.12 ( $\text{CH}_2$ )	
[4G] $\text{BF}_4$	Et	Naph	2021s 1989w	1836m	1538w	209.2, 208.4	254.9	323.2	90.6, 90.3	5.44, 4.65	-	5.12, 4.95 ( $\text{CH}_2\text{Me}$ )	
[5H] $\text{CF}_3\text{SO}_3$	Bn	Bn	2023s 1991m	1840m	1550w	208.2	253.7	324.4	90.6	5.40	-	5.66, 5.50 ( $\text{CH}_2\text{Ph}$ )	
<b>6</b> [c]	Me	Cy	1977s	1798s	1537m	212.5, 211.9	267.5, 268.1	334.8, 334.5	86.8, 86.6, 86.4, 86.3	4.70, 4.72, 4.64, 4.62	4.53, 4.05	5.01, 6.09 ( $\text{C}_6\text{H}_{11}$ )	

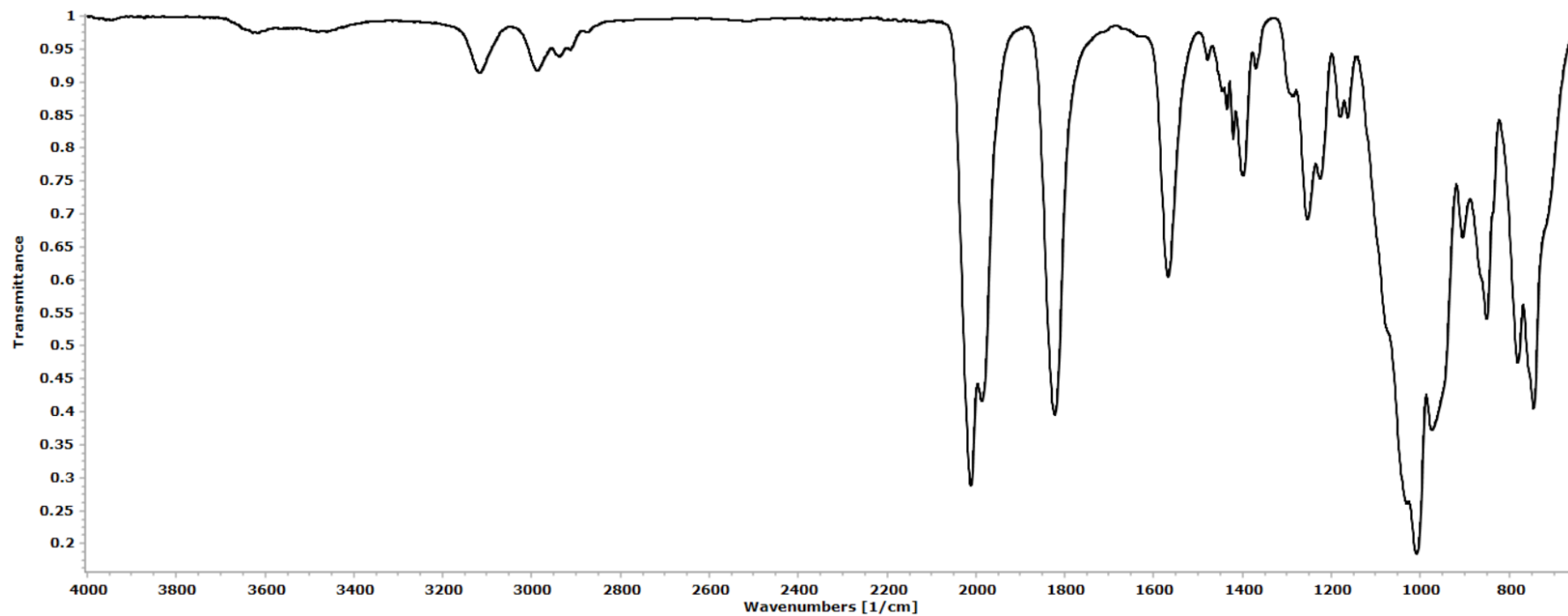
Abbreviation list. Ind = 1H-indol-6-yl; Phos =  $\text{CH}_2\text{P}(\text{O})(\text{OEt})_2$ ; Cy =  $\text{C}_6\text{H}_{11}$ ; Xyl = 2,6- $\text{C}_6\text{H}_3\text{Me}_2$ ; All =  $\text{CH}_2\text{CHCH}_2$ ; Naph = 2-naphthyl; Bn =  $\text{CH}_2\text{Ph}$ . [a] NMR data in  $\text{CD}_3\text{CN}$ . [b]  $[\mathbf{2C}]^+$   $^1\text{H}$  NMR signals in  $\text{CD}_3\text{OD}$ : 5.40, 5.36, 4.07;  $\text{D}_2\text{O}$ : 5.37, 5.35, 4.08 (identical for both salts). [c]  $^{13}\text{C}$  NMR data taken from the literature ( $\text{DMSO-d}_6$  for  $[\mathbf{2F}]\text{CF}_3\text{SO}_3$ ).<sup>[1,2]</sup> [c] NMR data given as major isomer, *minor isomer*.

**Solid-state IR spectra of diiron compounds.**

**Figure S3.** Solid-state IR spectrum (650-4000  $\text{cm}^{-1}$ ) of  $[\text{Fe}_2\text{Cp}_2(\text{CO})_2(\mu\text{-CO})\{\mu\text{-CNMe(1H-indol-6-yl)}\}] \text{CF}_3\text{SO}_3$ , **[2A]** $\text{CF}_3\text{SO}_3$ .

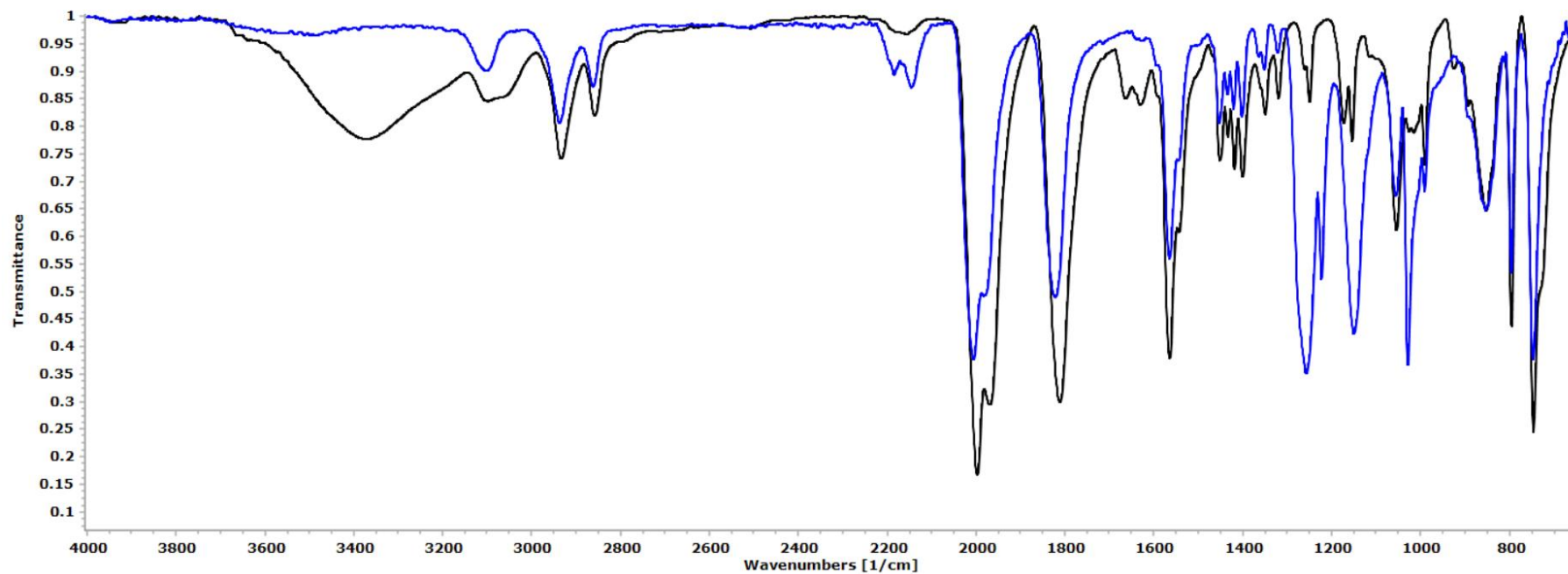


**Figure S4.** Solid-state IR spectrum (650-4000  $\text{cm}^{-1}$ ) of  $[\text{Fe}_2\text{Cp}_2(\text{CO})_2(\mu\text{-CO})\{\mu\text{-CNMe}(\text{CH}_2\text{PO}_3\text{Et}_2)\}]\text{BF}_4$ , **[2B]** $\text{BF}_4$ .

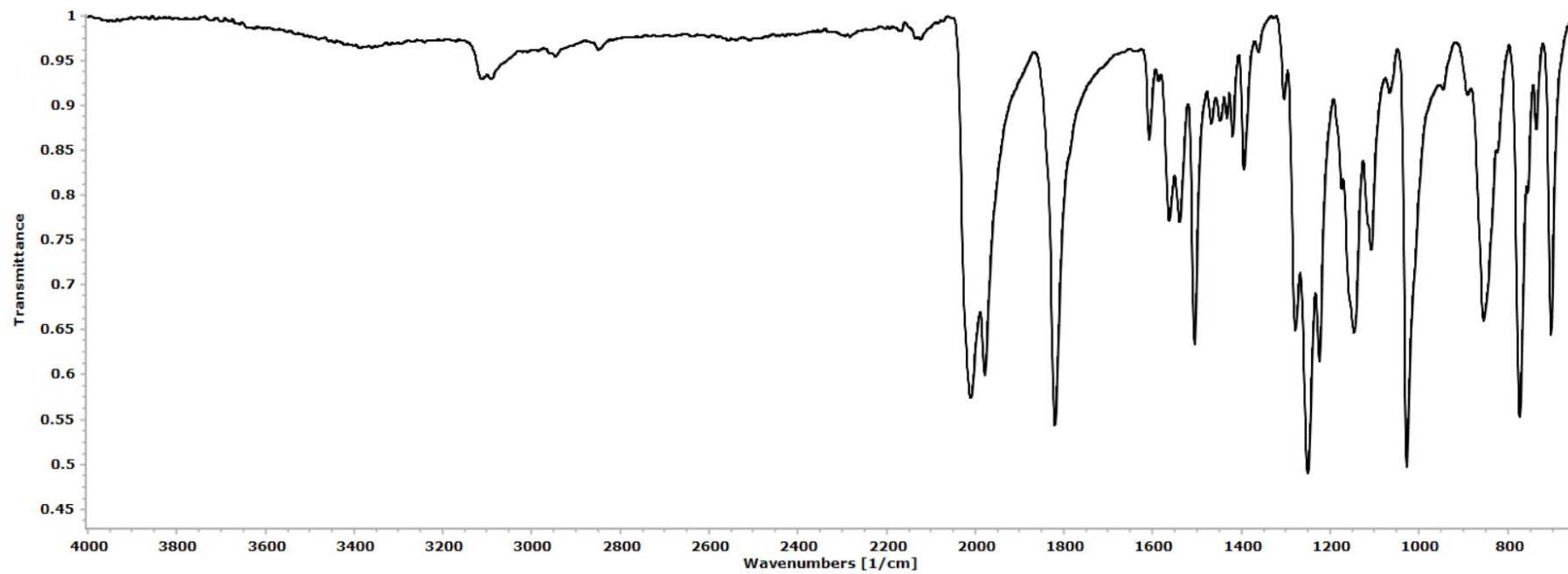




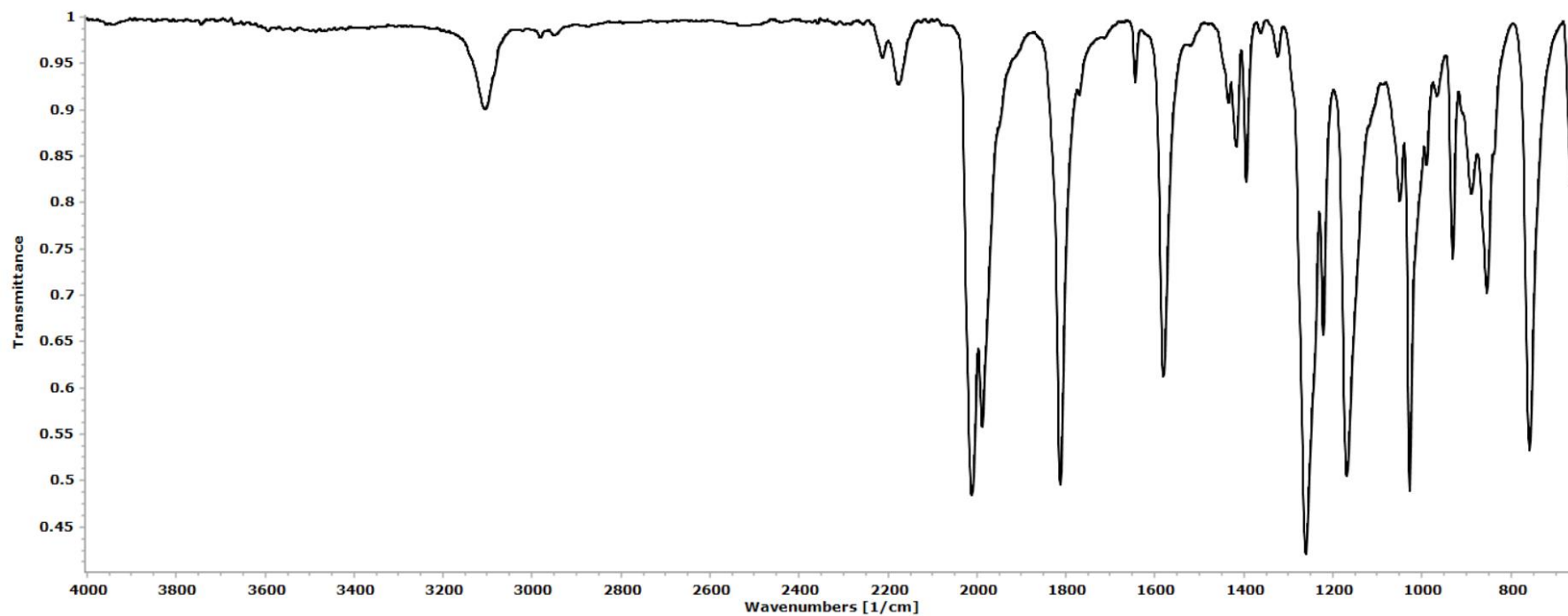
**Figure S5.** Solid-state IR spectra (650-4000  $\text{cm}^{-1}$ ) of  $[\text{Fe}_2\text{Cp}_2(\text{CO})_2(\mu\text{-CO})\{\mu\text{-CNMe}(\text{C}_6\text{H}_{11})\}]^+$ ,  $[\mathbf{2C}]^+$ , as  $\text{CF}_3\text{SO}_3^-$  (blue line) or  $\text{Cl}^-$  salt. The broad absorption at 3500  $\text{cm}^{-1}$  is due to moisture.



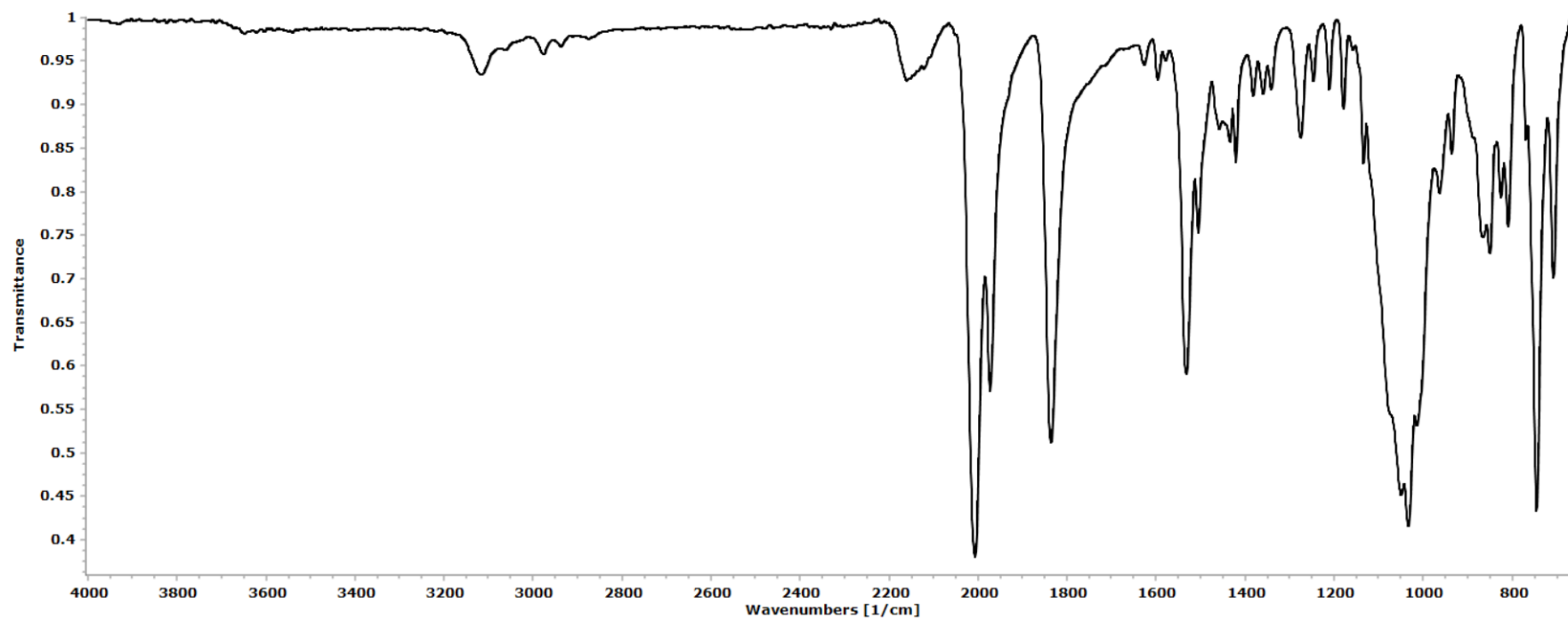
**Figure S6.** Solid-state IR spectrum (650-4000  $\text{cm}^{-1}$ ) of  $[\text{Fe}_2\text{Cp}_2(\text{CO})_2(\mu\text{-CO})\{\mu\text{-CNMe(4-C}_6\text{H}_4\text{OMe)}\}]\text{CF}_3\text{SO}_3$ , **[2D]** $\text{CF}_3\text{SO}_3$ .



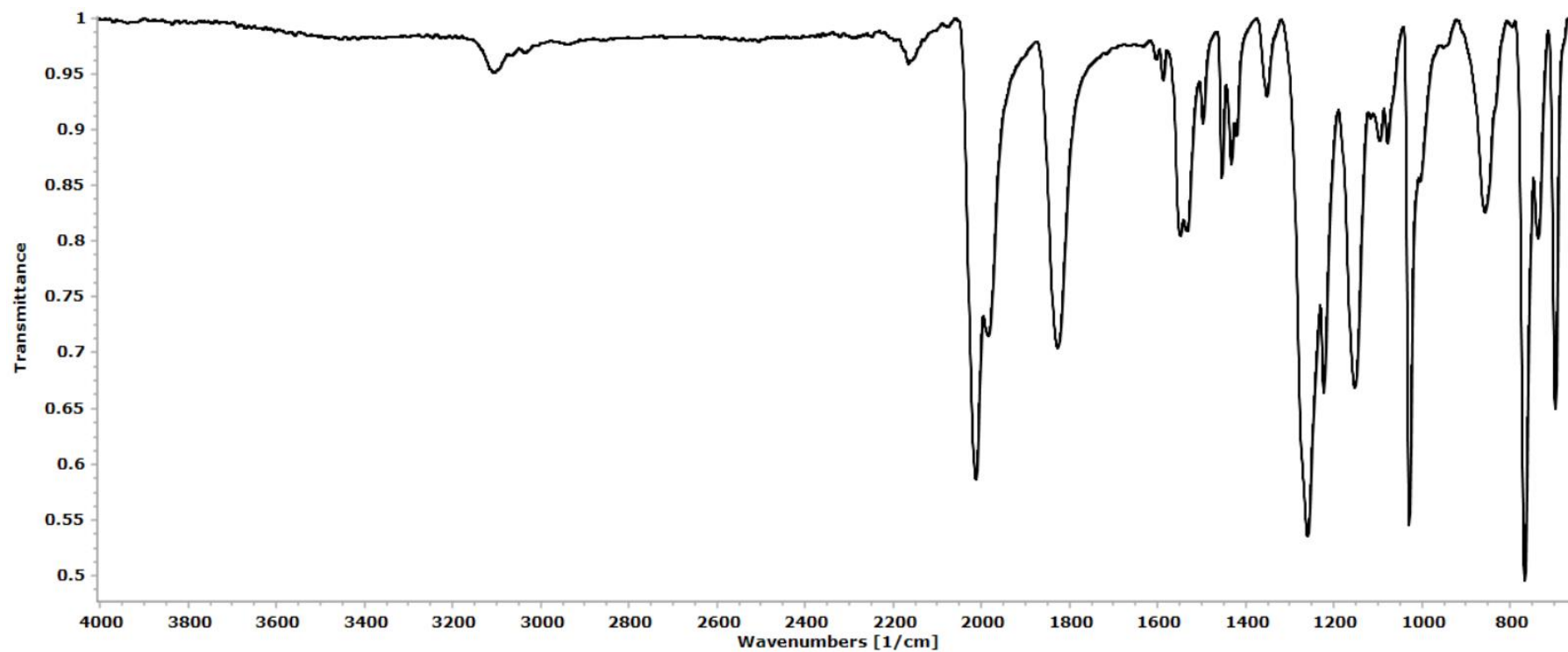
**Figure S7.** Solid-state IR spectrum (650-4000  $\text{cm}^{-1}$ ) of  $[\text{Fe}_2\text{Cp}_2(\text{CO})_2(\mu\text{-CO})\{\mu\text{-CNMe}(\text{CH}_2\text{CHCH}_2)\}] \text{CF}_3\text{SO}_3$ , **[3F]** $\text{CF}_3\text{SO}_3$ .



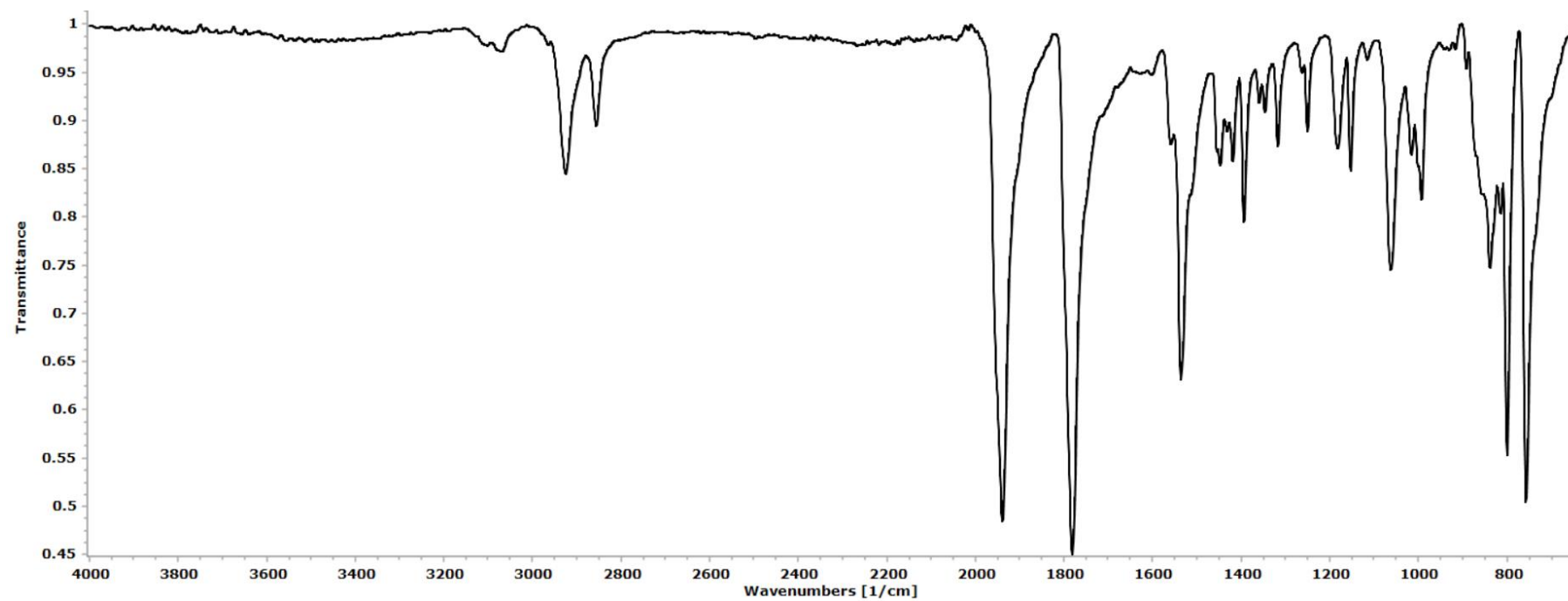
**Figure S8.** Solid-state IR spectrum (650-4000  $\text{cm}^{-1}$ ) of  $[\text{Fe}_2\text{Cp}_2(\text{CO})_2(\mu\text{-CO})\{\mu\text{-CNEt(2-naphthyl)}\}]\text{BF}_4$ , **[4G]** $\text{BF}_4$ .



**Figure S9.** Solid-state IR spectrum (650-4000  $\text{cm}^{-1}$ ) of  $[\text{Fe}_2\text{Cp}_2(\text{CO})_2(\mu\text{-CO})\{\mu\text{-CNBn}_2\}]\text{CF}_3\text{SO}_3$ , **[5H]** $\text{CF}_3\text{SO}_3$ .

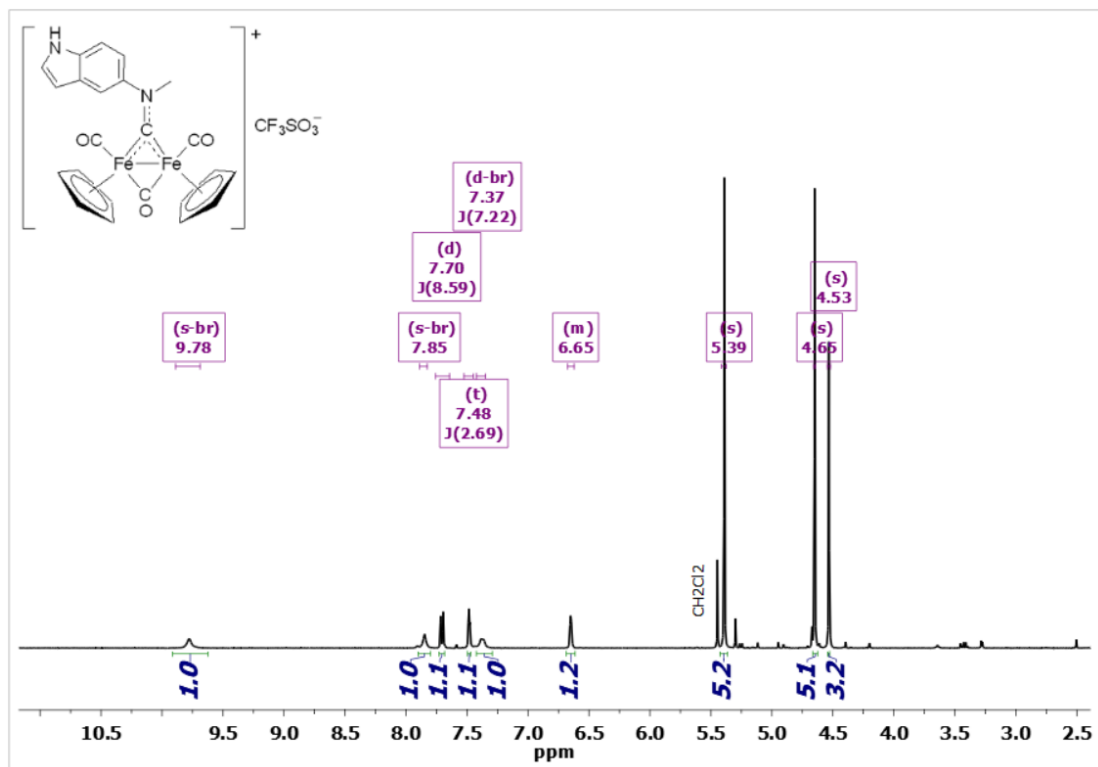


**Figure S10.** Solid-state IR spectrum ( $650\text{--}4000\text{ cm}^{-1}$ ) of  $[\text{Fe}_2\text{Cp}_2\text{Cl}(\text{CO})(\mu\text{-CO})\{\mu\text{-CNMe}(\text{C}_6\text{H}_{11})\}]$ , **6**.

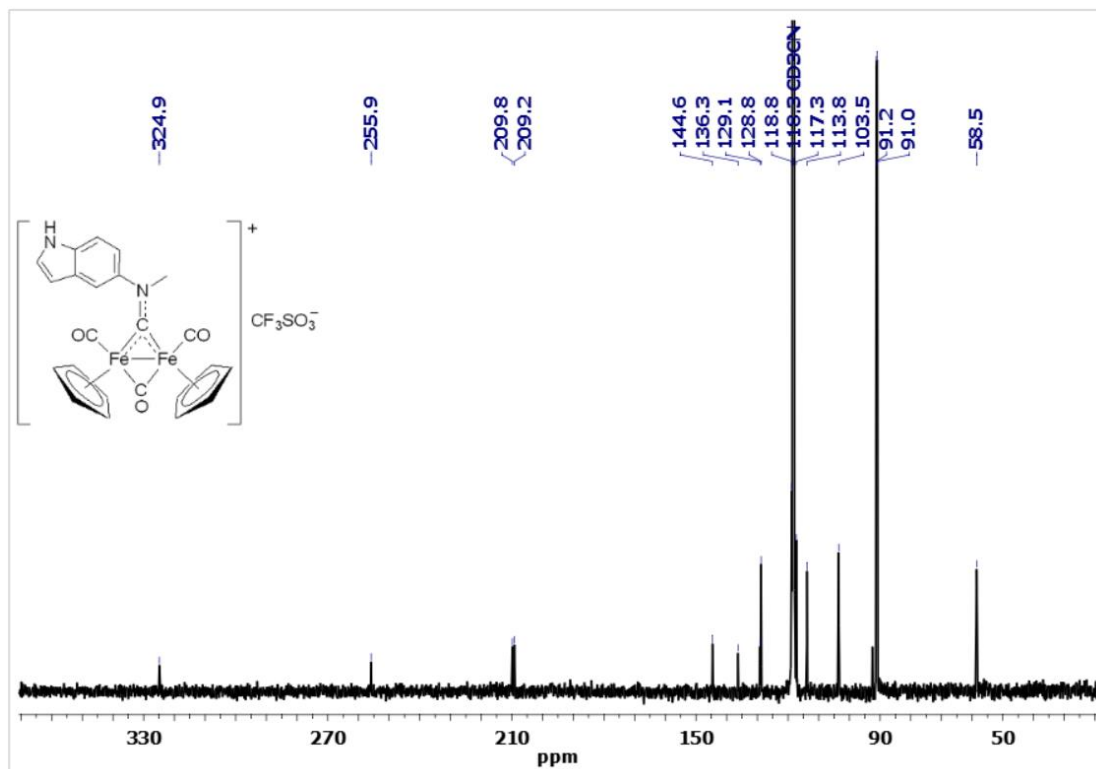


## Solution NMR spectra of diiron compounds.

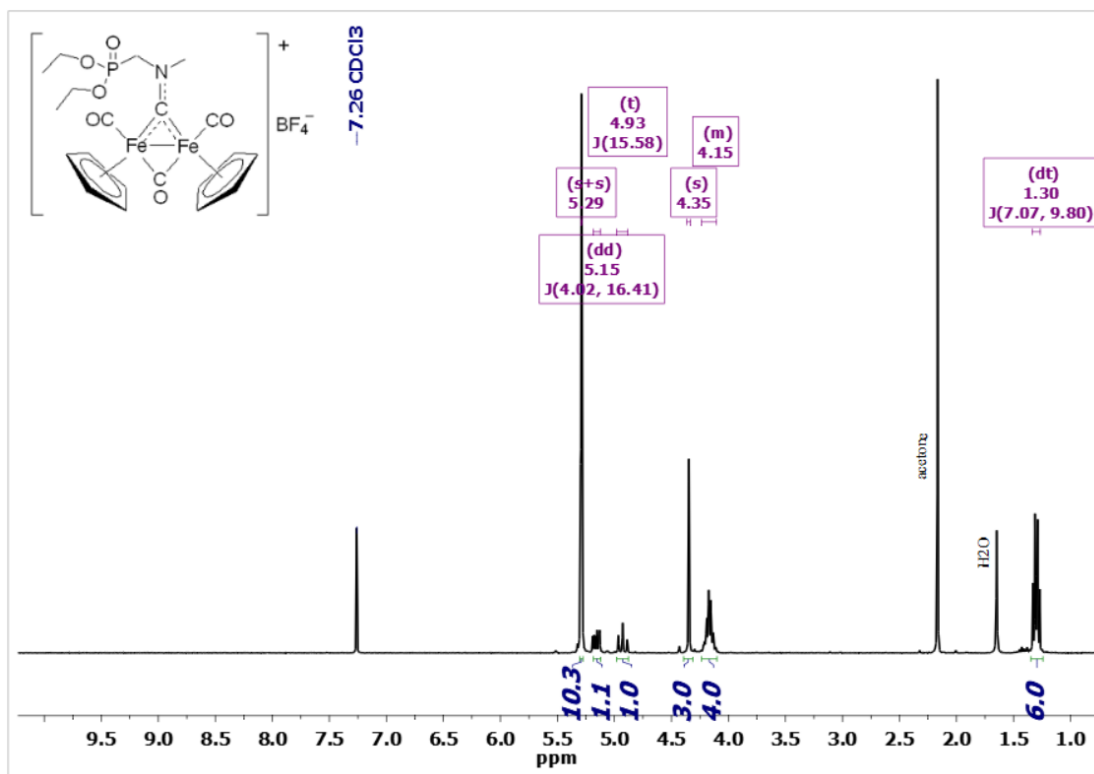
**Figure S11.**  $^1\text{H}$  NMR spectrum (401 MHz,  $\text{CD}_3\text{CN}$ ) of  $[\mathbf{2A}]\text{CF}_3\text{SO}_3$ .



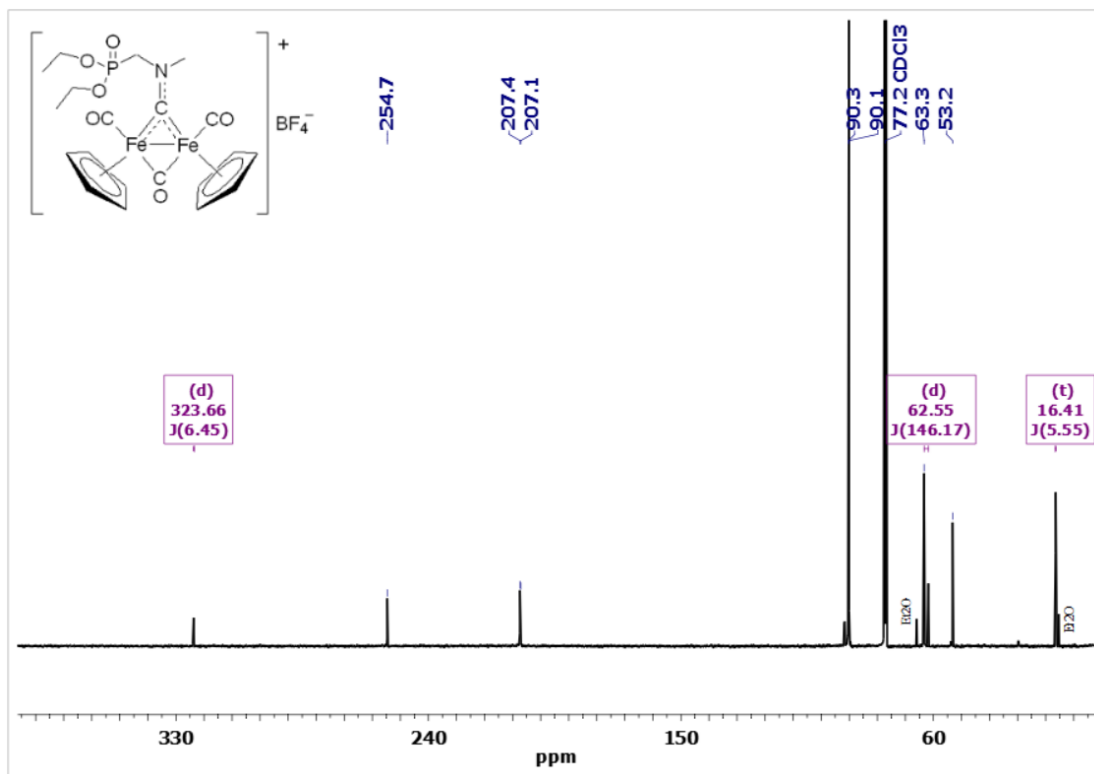
**Figure S12.**  $^{13}\text{C}\{^1\text{H}\}$  NMR spectrum (101 MHz,  $\text{CD}_3\text{CN}$ ) of  $[\mathbf{2A}]\text{CF}_3\text{SO}_3$ .



**Figure S13.**  $^1\text{H}$  NMR spectrum (401 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2B}]\text{BF}_4$ .

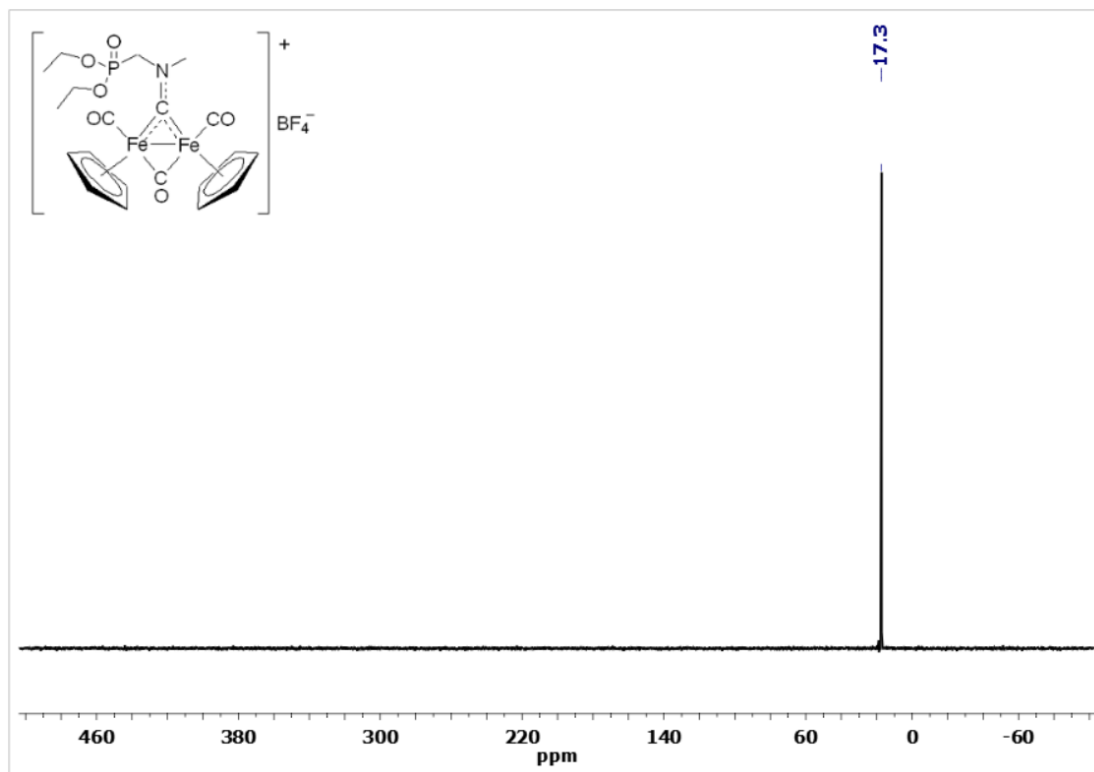


**Figure S14.**  $^{13}\text{C}\{^1\text{H}\}$  NMR spectrum (101 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2B}]\text{BF}_4$ .

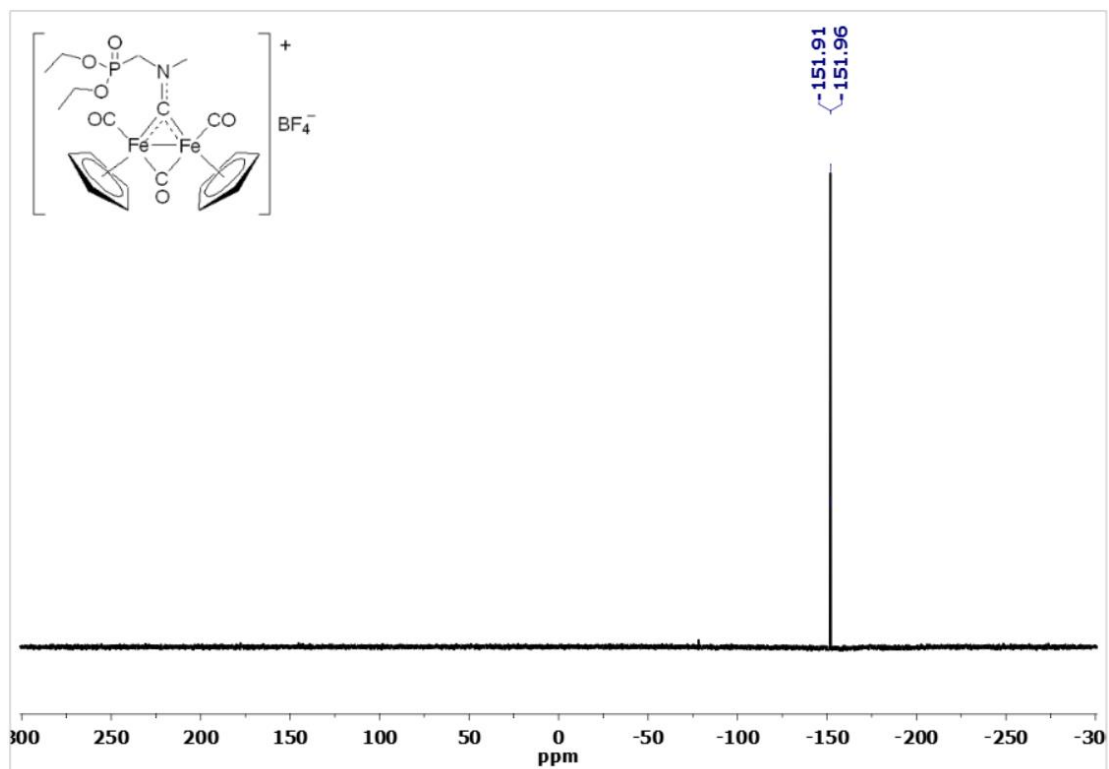




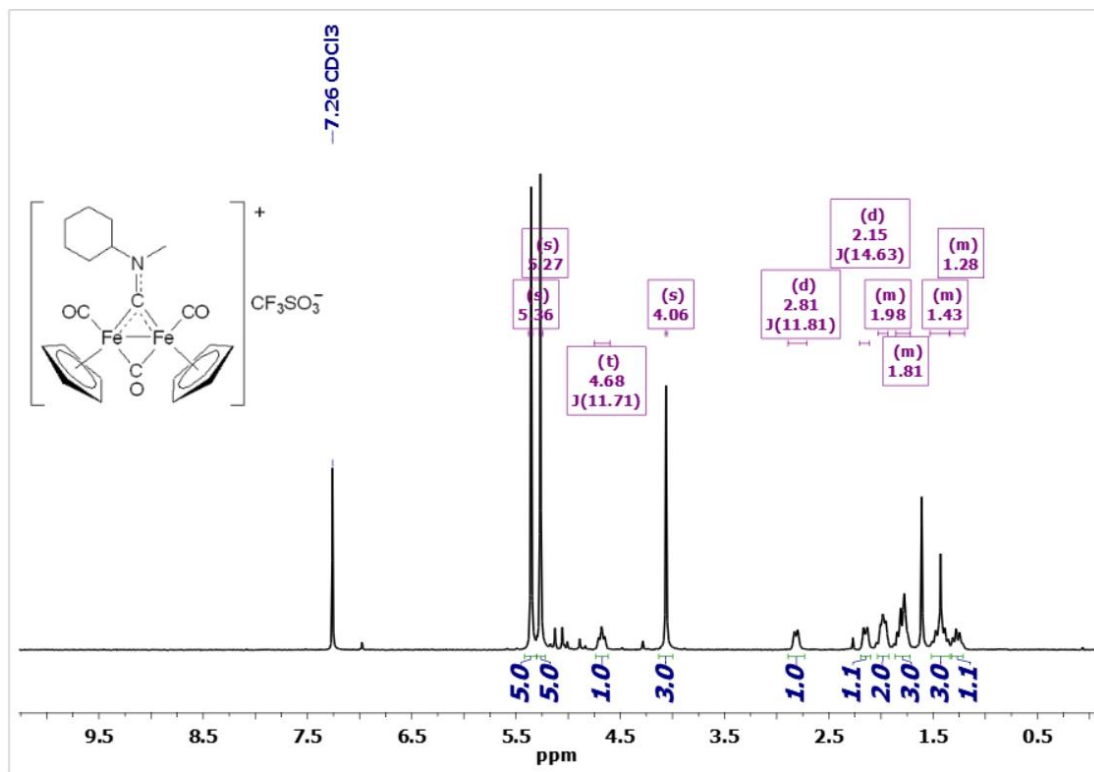
**Figure S15.**  $^{31}\text{P}\{^1\text{H}\}$  NMR spectrum (162 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2B}]\text{BF}_4$ .



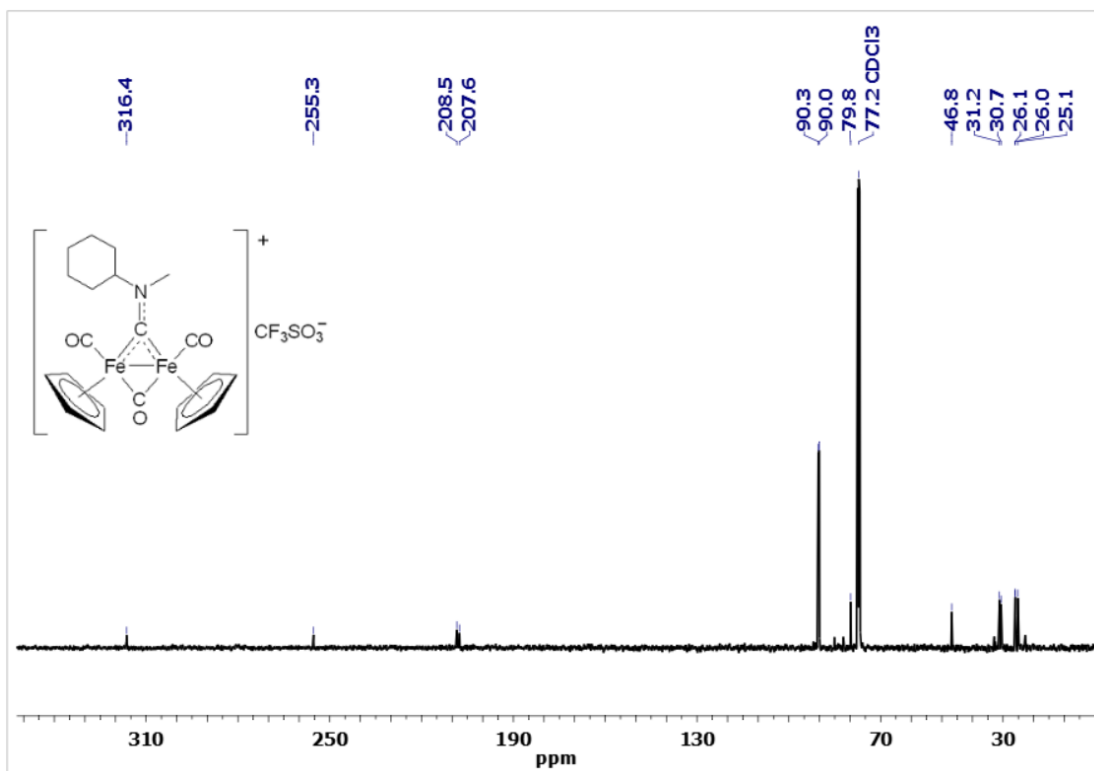
**Figure S16.**  $^{19}\text{F}\{^1\text{H}\}$  NMR spectrum (378 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2B}]\text{BF}_4$ .



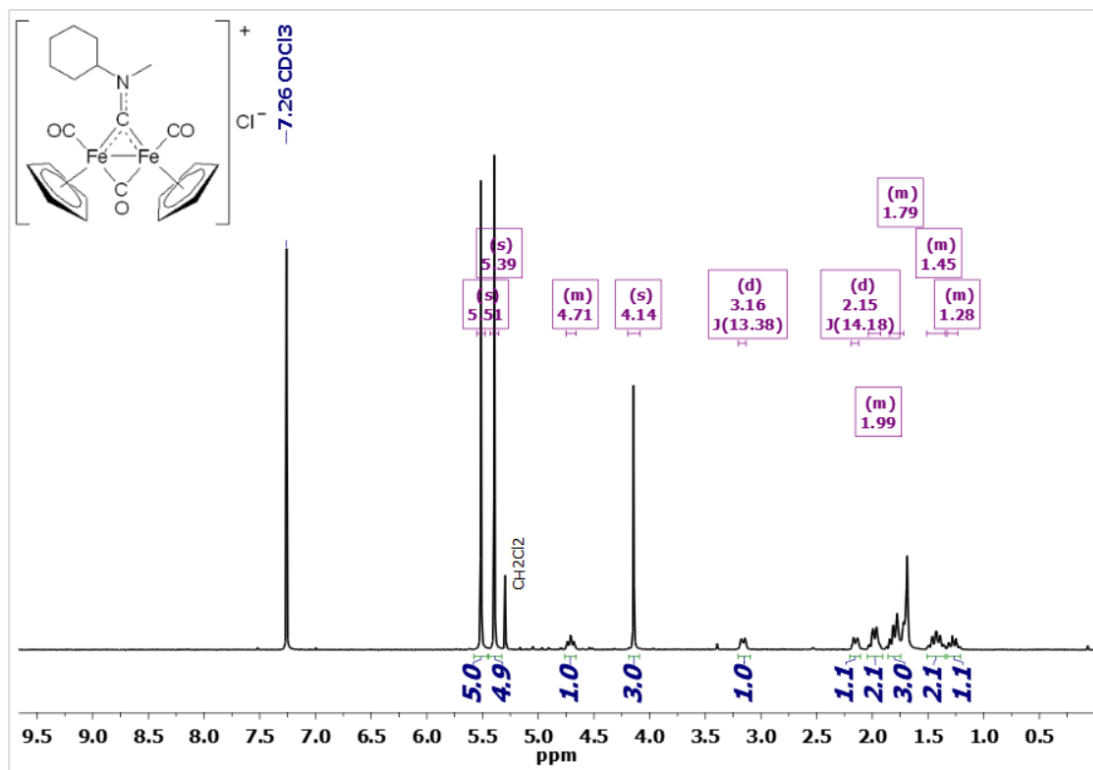
**Figure S17.**  $^1\text{H}$  NMR spectrum (401 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2C}]\text{CF}_3\text{SO}_3$ .



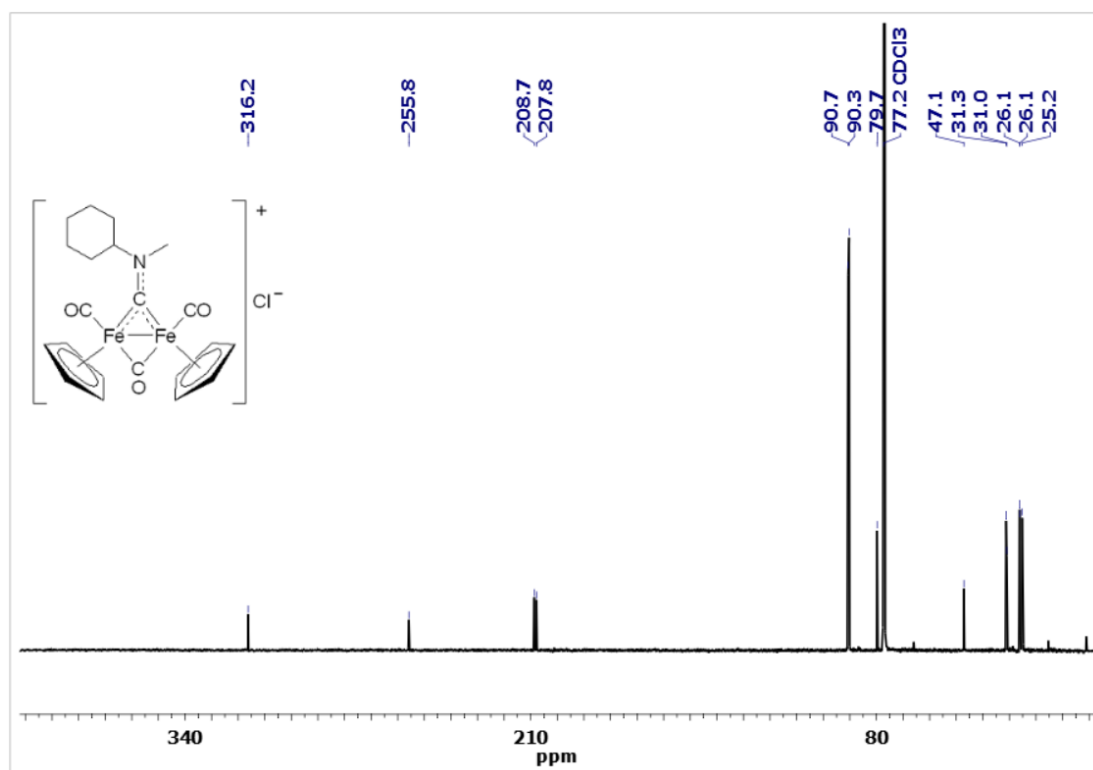
**Figure S18.**  $^{13}\text{C}\{^1\text{H}\}$  NMR spectrum (101 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2C}]\text{CF}_3\text{SO}_3$ .



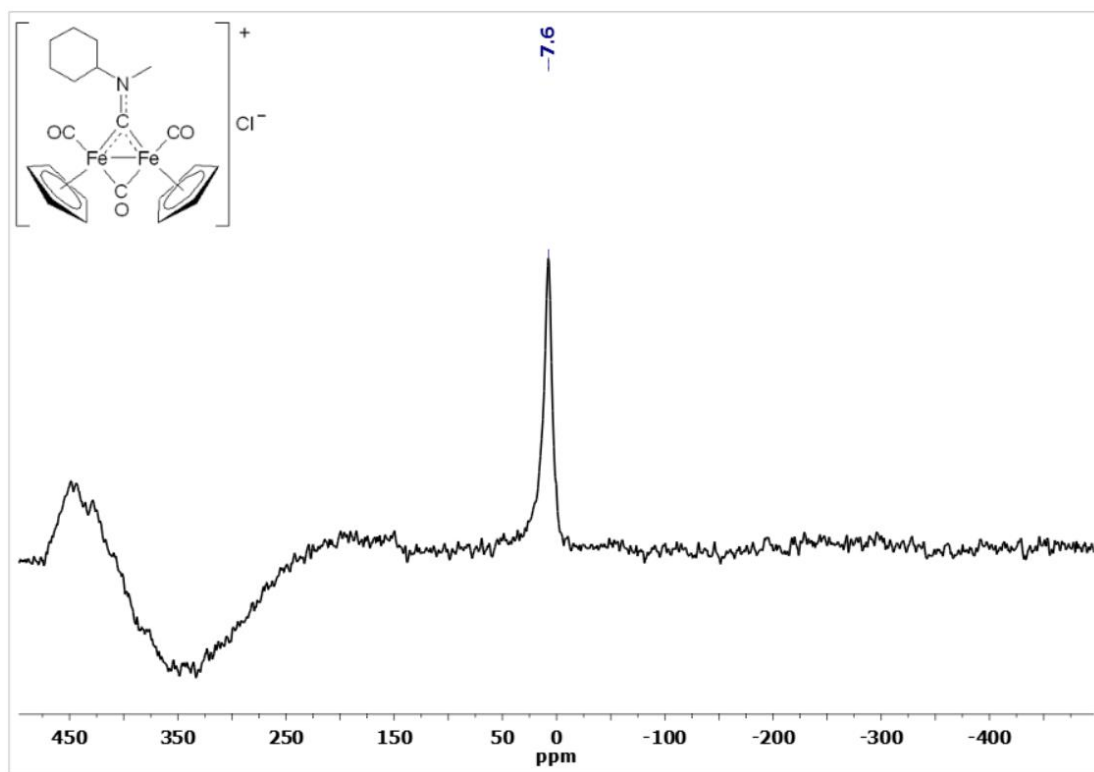
**Figure S19.**  $^1\text{H}$  NMR spectrum (401 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2C}]\text{Cl}$ .



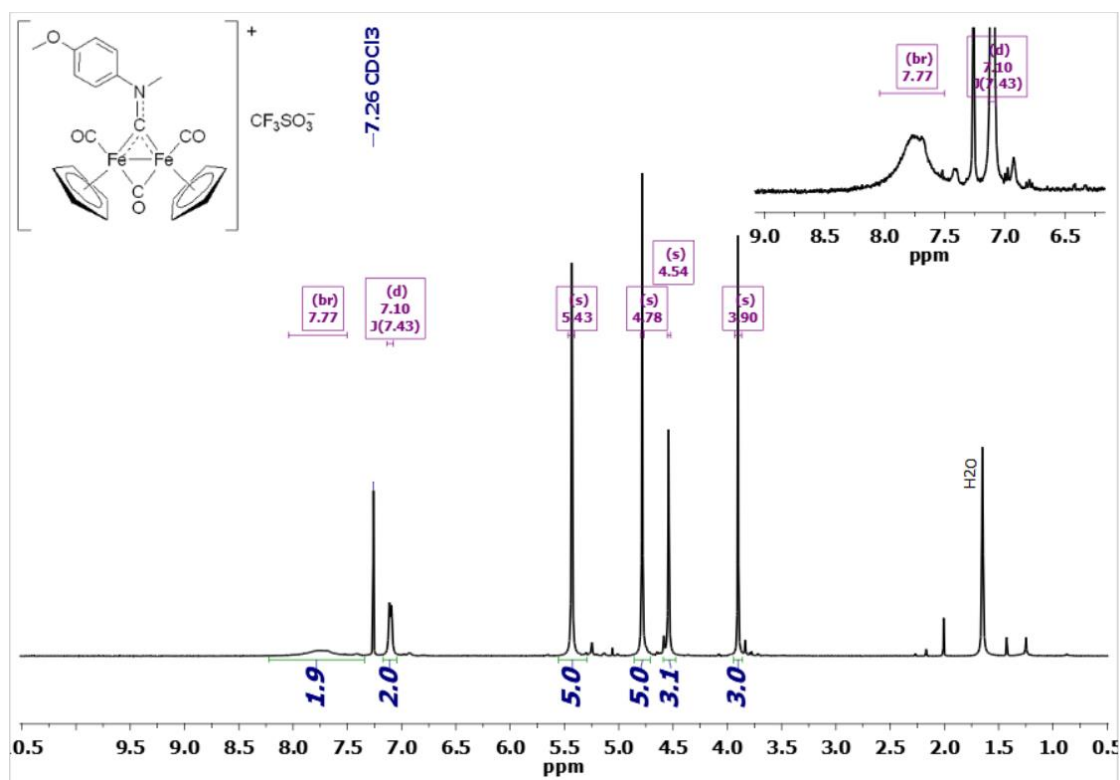
**Figure S20.**  $^{13}\text{C}\{^1\text{H}\}$  NMR spectrum (101 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2C}]\text{Cl}$ .



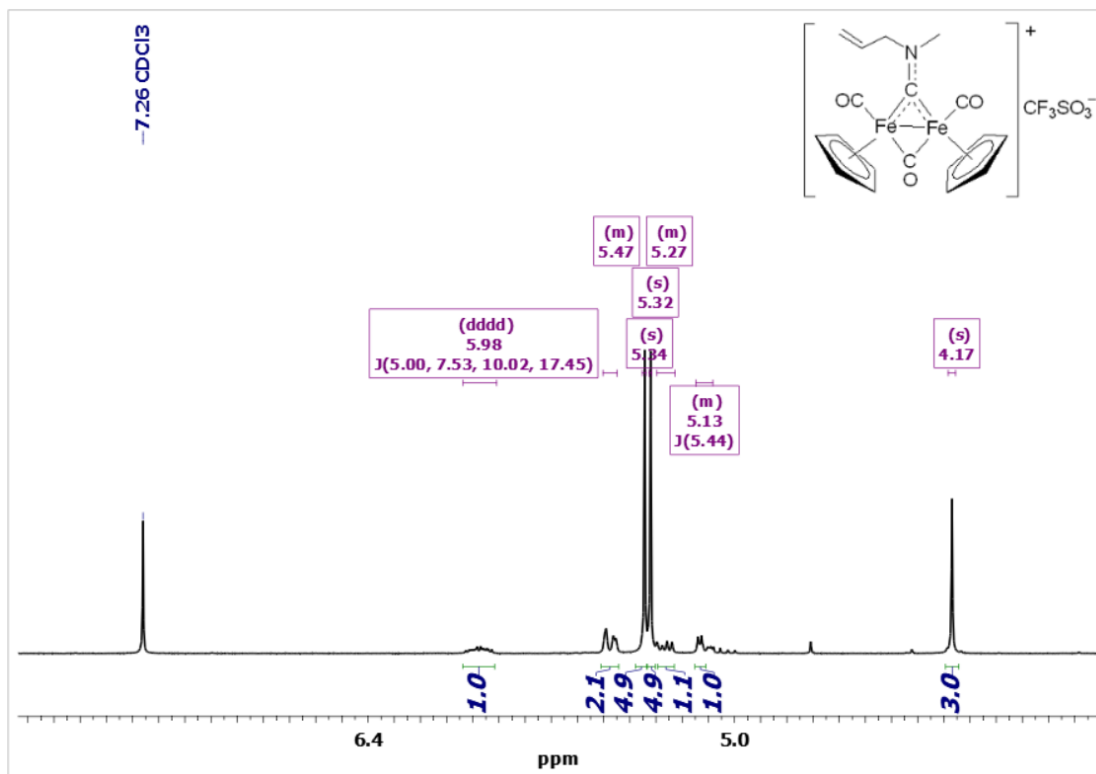
**Figure S21.**  $^{35}\text{Cl}$  NMR spectrum (39 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2C}]\text{Cl}$ .



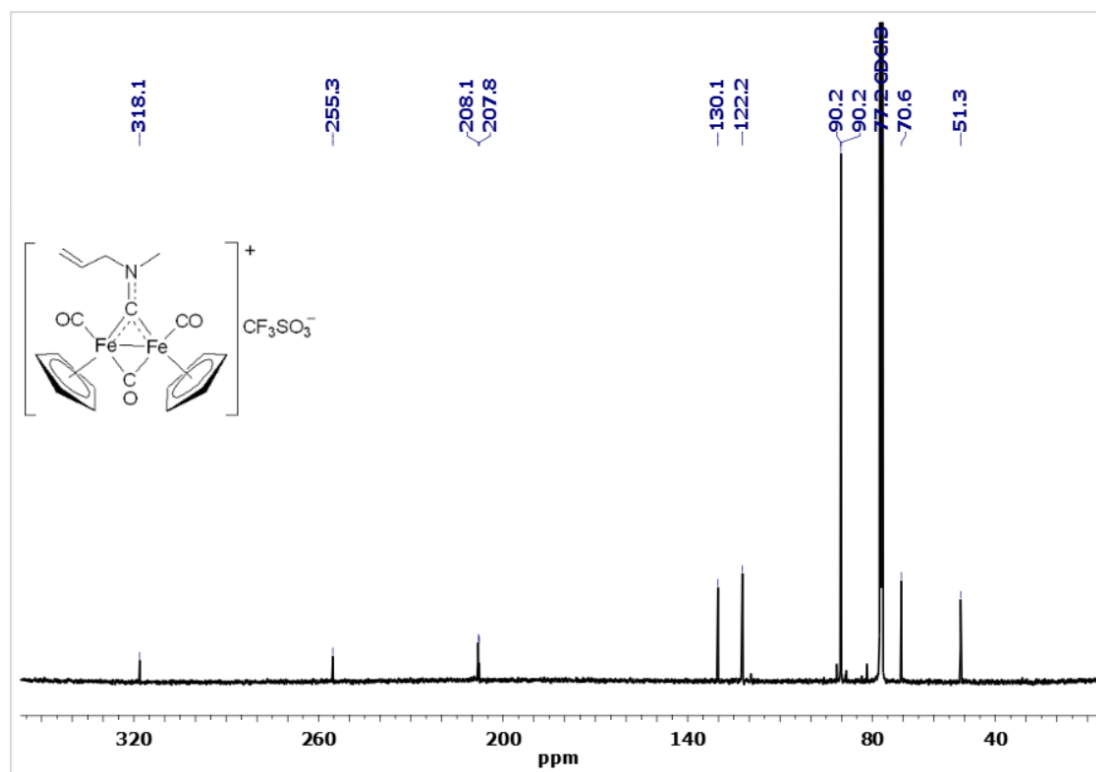
**Figure S22.**  $^1\text{H}$  NMR spectrum (401 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{2D}]\text{CF}_3\text{SO}_3$ . Inset shows the broad aromatic resonance.



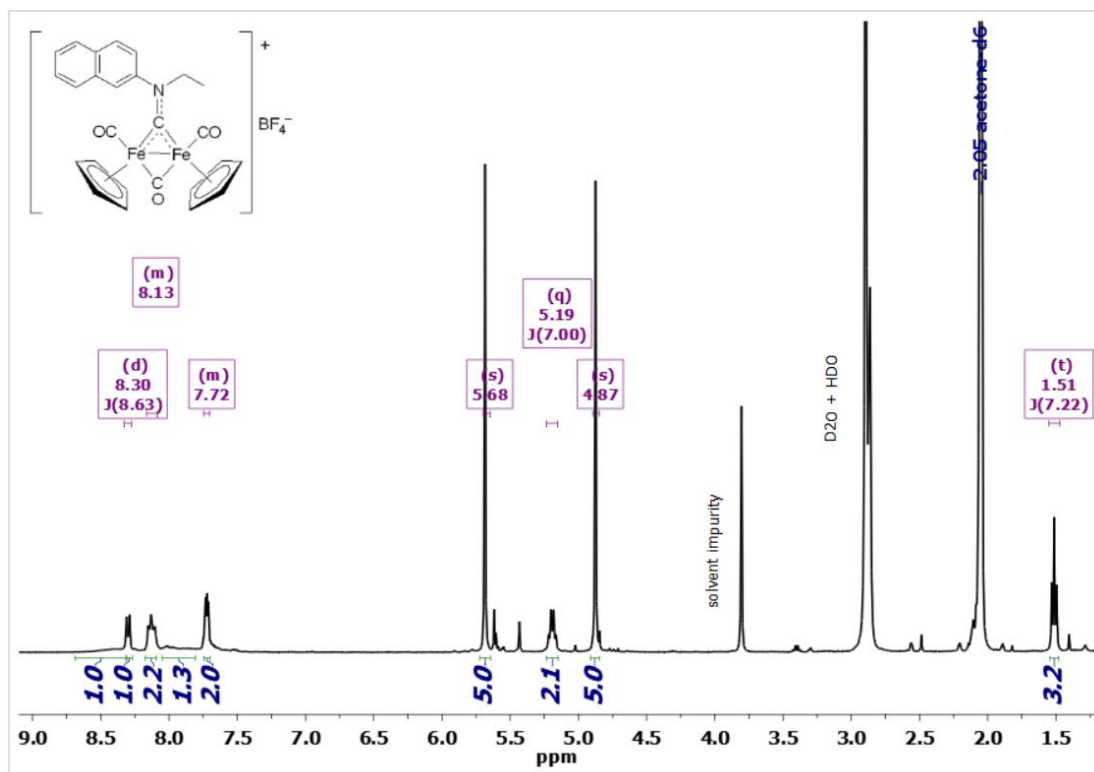
**Figure S23.**  $^1\text{H}$  NMR spectrum (401 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{3F}]\text{CF}_3\text{SO}_3$ .



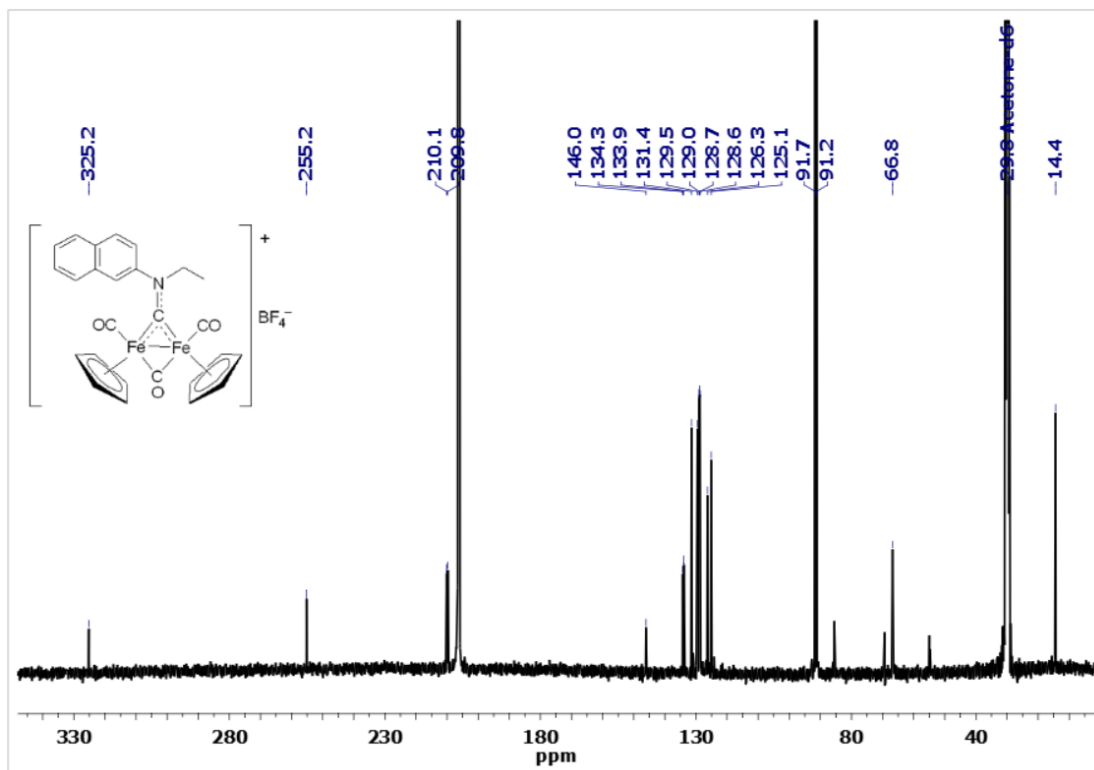
**Figure S24.**  $^{13}\text{C}\{^1\text{H}\}$  NMR spectrum (101 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{3F}]\text{CF}_3\text{SO}_3$ .



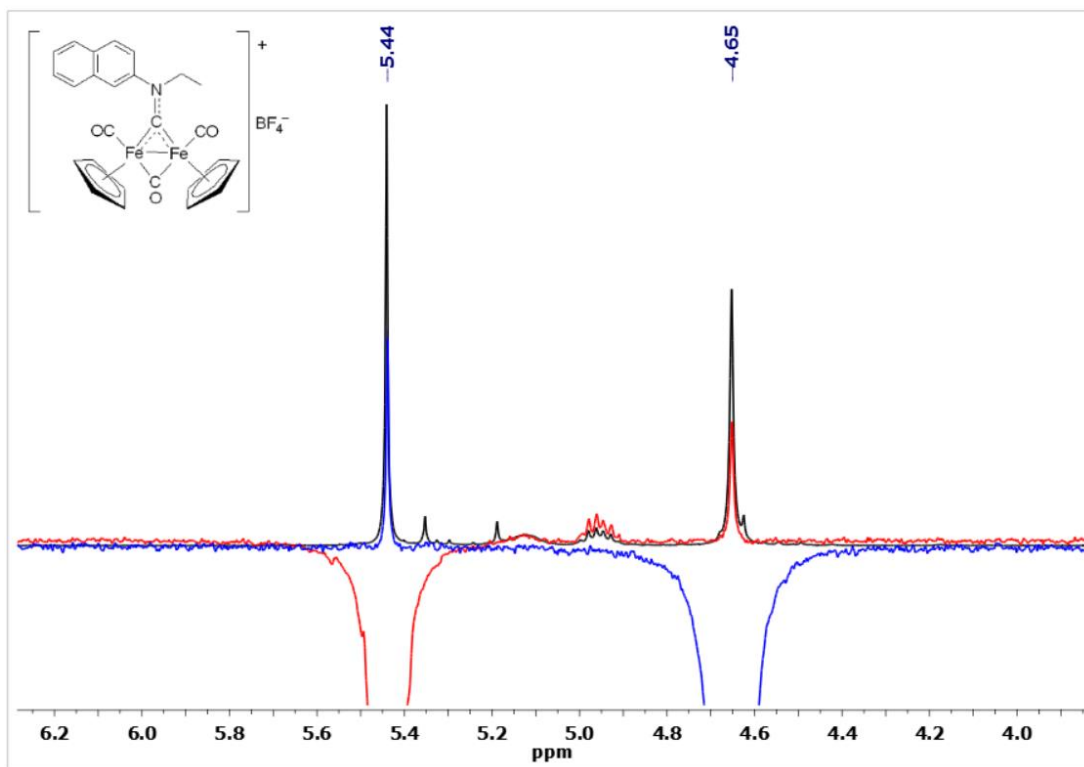
**Figure S25.**  $^1\text{H}$  NMR spectrum (401 MHz, acetone- $d_6$ ) of  $[\mathbf{4G}]\text{BF}_4$ .



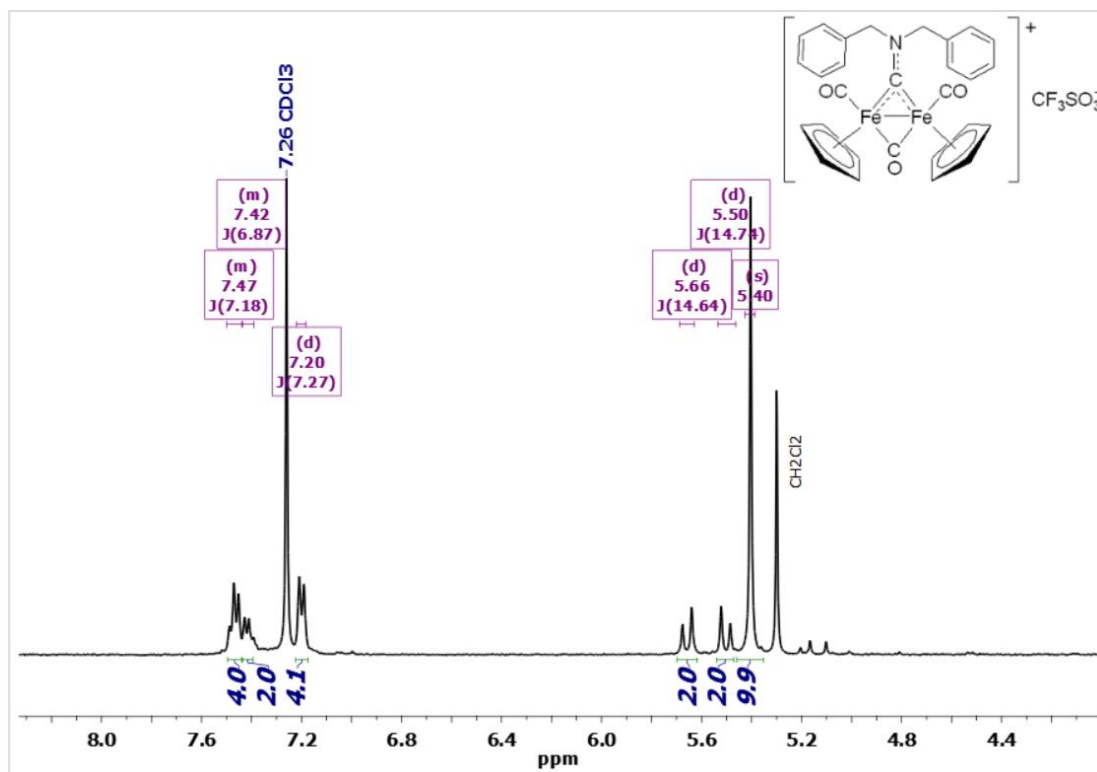
**Figure S26.**  $^{13}\text{C}\{^1\text{H}\}$  NMR spectrum (101 MHz, acetone- $d_6$ ) of  $[\mathbf{4G}]\text{BF}_4$ .



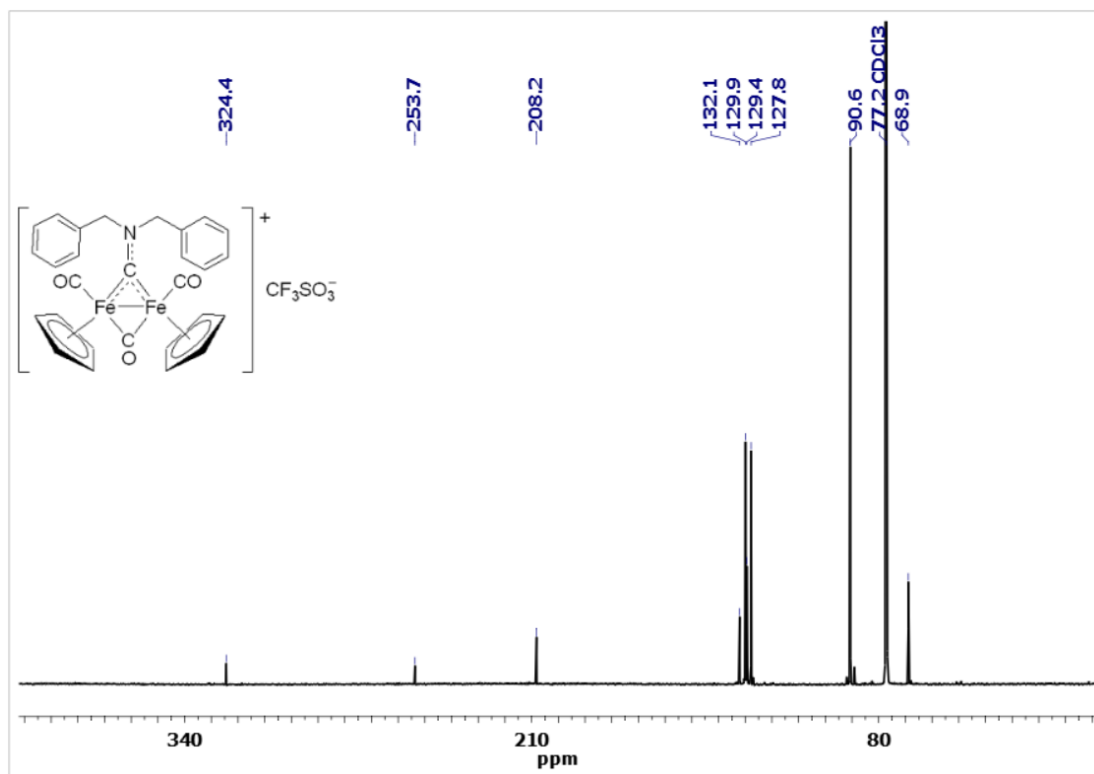
**Figure S27.**  $^1\text{H}$  (black line) and  $^1\text{H}$ -NOE (blue line: irradiation at 4.65 ppm; red line: irradiation at 5.44 ppm) NMR spectra (401 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{4G}]\text{BF}_4$ ; showing mutual NOE effect for the Cp rings.



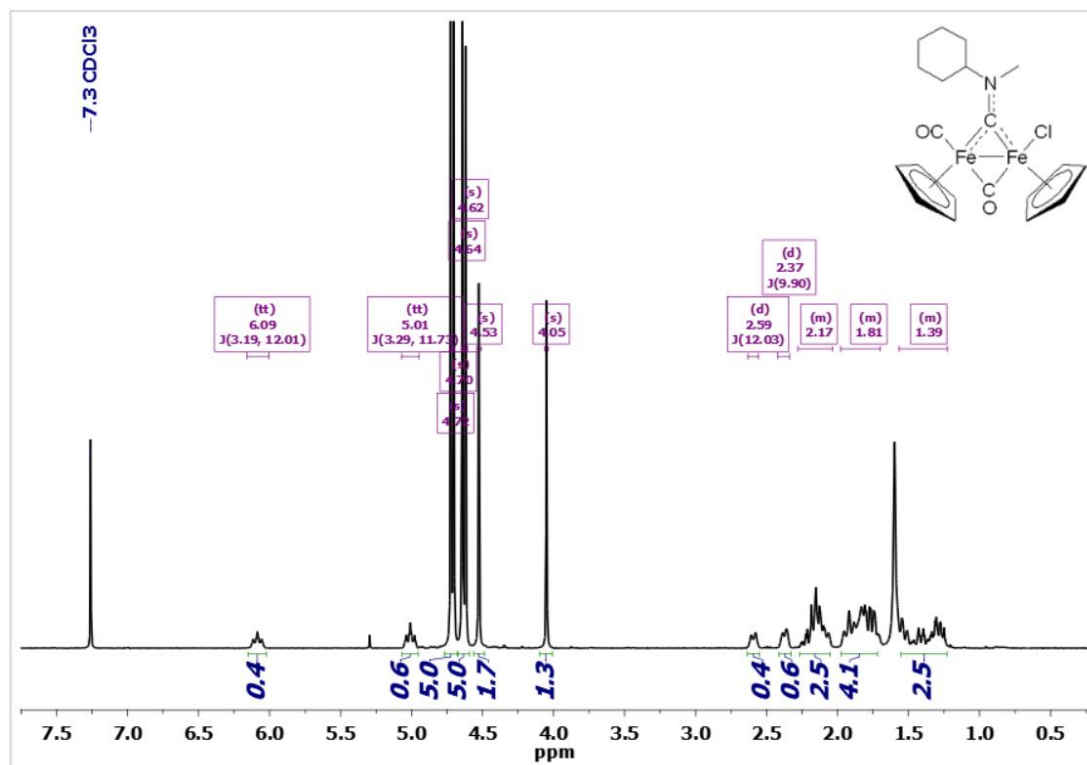
**Figure S28.**  $^1\text{H}$  NMR spectrum (401 MHz,  $\text{CDCl}_3$ ) of  $[\mathbf{5H}]\text{CF}_3\text{SO}_3$ .



**Figure S29.**  $^{13}\text{C}\{^1\text{H}\}$  NMR spectrum (101 MHz,  $\text{CDCl}_3$ ) of  $[\text{5H}]\text{CF}_3\text{SO}_3$ .

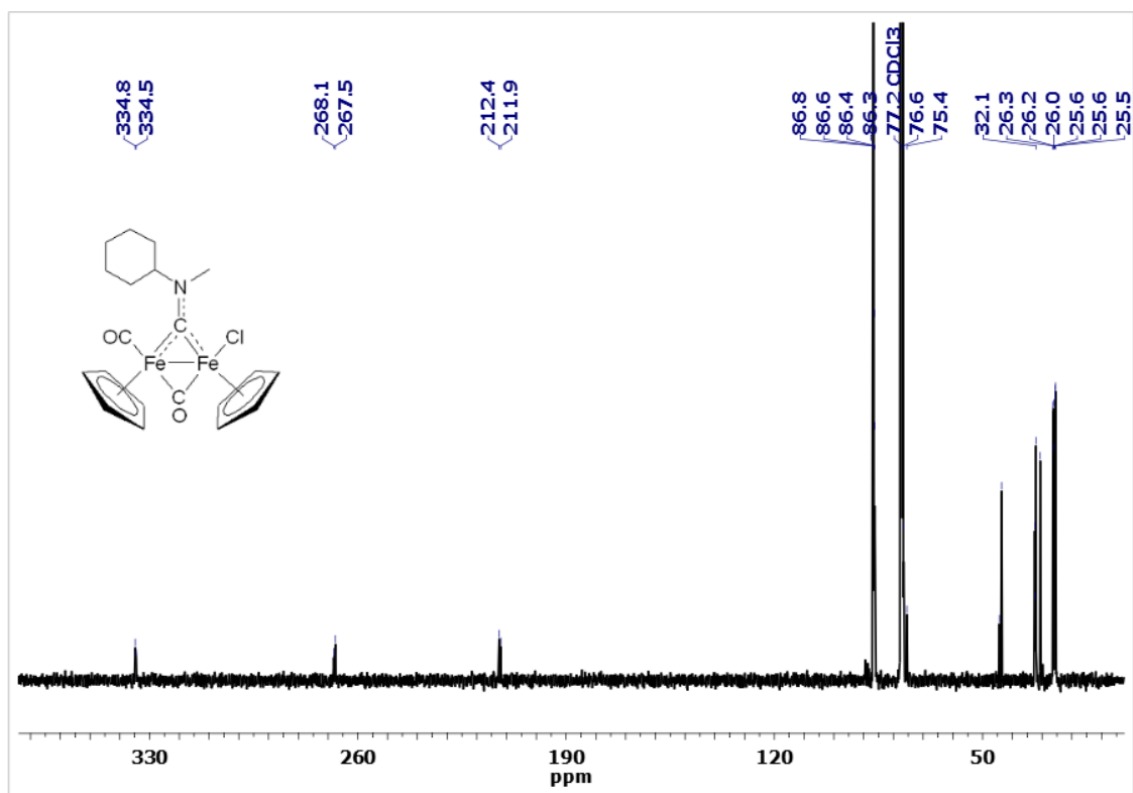


**Figure S30.**  $^1\text{H}$  NMR spectrum (401 MHz,  $\text{CDCl}_3$ ) of **6**.

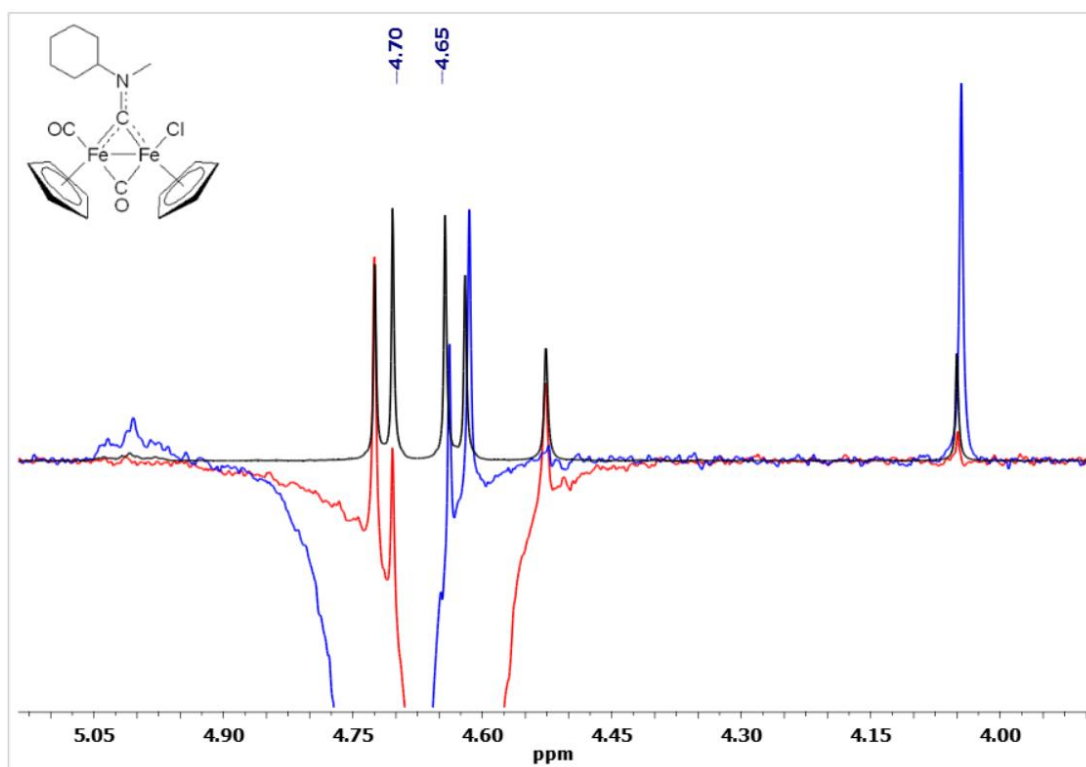




**Figure S31.**  $^{13}\text{C}\{^1\text{H}\}$  NMR spectrum (101 MHz,  $\text{CDCl}_3$ ) of **6**.



**Figure S32.**  $^1\text{H}$  (black line) and  $^1\text{H}$ -NOE (blue line: irradiation at 4.70 ppm; blue line: irradiation at 4.65 ppm) NMR spectra (401 MHz,  $\text{CDCl}_3$ ) of **6**, showing mutual NOE effect for the Cp rings (co-irradiation for the two isomers).



### 3. DFT-optimized geometries (data).

#### Compound **1C**

Fe	-2.254121	-2.477913	0.119554
Fe	-1.686951	-1.643968	-2.192534
C	-0.971463	-1.149056	-0.484094
C	-3.235820	-2.543747	-1.514854
O	-4.262692	-2.970861	-1.932563
C	-3.260842	-1.206072	0.723079
O	-3.937405	-0.373301	1.173798
C	-2.562579	-0.151513	-2.235104
O	-3.124614	0.865759	-2.283582
C	-1.785787	-3.256454	2.032191
C	-0.686764	-3.536106	1.182447
C	-1.156749	-4.356347	0.117875
C	-2.545244	-4.611242	0.324225
C	-2.944352	-3.929933	1.497821
C	0.070617	-2.787845	-2.708296
C	-1.078996	-3.416132	-3.276603
C	-1.698128	-2.494477	-4.160239
C	-0.929078	-1.288239	-4.130632
C	0.171769	-1.476240	-3.243512
H	-1.765985	-2.654878	2.930699
H	0.322945	-3.170214	1.299170
H	-0.561383	-4.735210	-0.699238
H	-3.186457	-5.191840	-0.324323
H	0.738618	-3.220470	-1.977194
H	-1.436430	-4.411368	-3.050617
H	-2.603764	-2.658593	-4.727432
H	-1.145901	-0.386891	-4.688457
H	0.916562	-0.737158	-2.984388
H	-3.938433	-3.911257	1.923466
N	-0.129122	-0.359579	-0.052156
C	0.185614	-0.104955	1.356424
C	2.036969	0.005619	3.070900
C	0.101625	1.629764	3.192602
C	1.596045	1.420523	3.464169
C	-0.251294	1.316264	1.734037
C	1.682575	-0.304294	1.611772
H	1.537374	-0.722887	3.726403
H	-0.480945	0.972488	3.854695
H	2.173998	2.152380	2.881080
H	0.254780	2.026093	1.065123
H	2.241516	0.362802	0.940813
H	-0.376910	-0.812742	1.983422
H	3.115833	-0.117250	3.226087
H	-0.191367	2.659412	3.431733
H	1.819425	1.607135	4.521991

H	-1.329079	1.420493	1.563189
H	1.967324	-1.331738	1.353393

Compound [2A]<sup>+</sup>

Fe	4.26660131890632	11.82050541092293	3.40771965512903
Fe	6.69486688127077	11.83835114403971	4.04327008242886
N	4.93173473019962	13.43369158078674	5.79813770669982
N	-0.26491961340199	15.22967095642780	6.58514123034587
H	-0.75932428048956	16.08042259003132	6.36250597149380
O	3.12598728623823	9.90564461064224	5.26782130400827
O	6.79534304200172	9.95154230240497	6.24733817843118
O	5.85974045263656	9.59605538186753	2.36016151838261
C	3.59803763490481	10.66672683277300	4.53713295632822
C	6.74035222377735	10.68998168549737	5.35992621000550
C	5.67801112072427	10.58104682817343	2.97965104328506
C	5.20368520054789	12.66495457110873	4.78552315151615
C	5.92793589922694	13.84721961917113	6.80963264955816
H	6.85048433696748	13.29267424716208	6.65650286240734
H	5.51750170605359	13.62866977709548	7.79833104625706
H	6.10956097423129	14.92183880444881	6.72325253991988
C	3.58882505824756	13.95434781261275	6.00757075362095
C	3.30566680465824	15.25557661536361	5.55458499948801
H	4.08809452553212	15.83681851186731	5.07681906707788
C	2.02808090530017	15.78535011968473	5.70301700238824
H	1.79351293864095	16.78470289924739	5.34908425690543
C	1.06010468259119	14.98367045923600	6.31434387569964
C	1.34862726871950	13.66473754075894	6.79548365617206
C	2.64726655463075	13.15533968534956	6.63717406944519
H	2.91084963423507	12.16299015519128	6.98643220490094
C	0.13478939483213	13.15176623425818	7.35980772871904
H	-0.01226659598830	12.18264117378391	7.81598516448043
C	-0.82123365045570	14.13215102001545	7.21212990513645
H	-1.86245011064640	14.14263122946145	7.50313256209453
C	4.31373994555732	12.76676909035276	1.46830598874988
H	5.20376746066459	12.90955706240193	0.87265303090475
C	3.79187742905839	13.67262258249934	2.43785692083617
H	4.22147495404992	14.62394054181794	2.71544811271865
C	2.62220368267430	13.09089761629318	2.99574185034639
H	1.99743686290352	13.51828813771042	3.76552218722092
C	2.42349468520336	11.82044529468723	2.36018395886043
H	1.61923395747341	11.12873713460242	2.57196759552522
C	3.46404054747009	11.62522176646389	1.41535919111958
H	3.60691226755053	10.75215871249872	0.79444864726678
C	7.45061830449803	13.34871682461572	2.68700224144769
H	6.85162482665897	13.99057304004299	2.05889302826015
C	7.98168554758547	12.07123153996466	2.32742477501176
H	7.82885374149848	11.56516459167466	1.38440749615890
C	8.71233250508874	11.57092001432842	3.43001716230629

H	9.22517099470939	10.62070800299406	3.48378461635056
C	8.62996684246606	12.54673959398239	4.48697644794515
H	9.08785444545442	12.45995530390683	5.46283710852819
C	7.86419849915478	13.64521470073461	4.01234046599185
H	7.62296517618766	14.54241564904401	4.56396282212509

Compound [2C]<sup>+</sup>

Fe	3.37495317234996	7.78878523058026	10.61664244838150
Fe	4.70039475512489	8.99008985228605	12.37650994566338
C	3.05928787442209	6.23834463451662	11.35441057624161
C	4.78122549031126	7.81769997824191	13.66812449439889
C	2.79557128339912	8.67537190829742	12.22017015910803
O	2.85532947337824	5.19497900345880	11.80810904932277
O	4.86115919654994	7.05265684210402	14.53151413911204
O	1.80547495841830	8.94526802041673	12.79773366591443
N	6.19074653626141	6.91781083900090	10.86437873757142
C	5.16539033979376	7.65767859606137	11.14905309440699
C	6.17121309421278	5.94915507507961	9.74907550980771
H	5.14272589083847	5.67001430339510	9.53687210491978
H	6.73622353105267	5.06226747976998	10.03333503734318
H	6.62421627222428	6.39827304030486	8.86219696805998
C	7.46842712929385	7.01262649371841	11.64187876231725
H	7.26786415364940	7.77478784281196	12.39436498309087
C	8.62937404431573	7.47642285842525	10.76221086349583
H	8.82520965611905	6.72876722446566	9.98475865718217
H	8.36089836729348	8.41224960146357	10.25664381442824
C	9.88800153083326	7.66083857712110	11.62115707556628
H	9.72375982713852	8.48348536174590	12.33160471395043
H	10.72633503135157	7.95720491995009	10.98048917813582
C	10.22808069952727	6.38320984093321	12.39773173721934
H	11.10180924679613	6.55408720146479	13.03744683173026
H	10.50003070195701	5.59174082476877	11.68561100034751
C	9.04012883731751	5.91057108920023	13.24336876640971
H	9.28247992135803	4.97237981731096	13.75558397365606
H	8.82595852450987	6.65428595825401	14.02347151059613
C	7.78687201530778	5.70924380419395	12.37955084270809
H	6.93096496836358	5.41477997998896	12.99677512649149
H	7.97561859896778	4.90067440739859	11.66450784244796
C	2.93778656034339	7.11590689605493	8.66941193719246
H	3.08536204933390	6.09714629147425	8.34018518146542
C	1.73834790508870	7.63744576442796	9.27531564282809
H	0.84293098340447	7.07236981470380	9.49485782556119
C	1.95170136835746	9.00886140024100	9.54256499636553
H	1.24978568562401	9.68084189217880	10.01583225497643
C	3.27071452922187	9.34461408045331	9.11051674051070
H	3.72531442617270	10.32111443533143	9.17489303765337
C	3.87171220578319	8.18010166659045	8.56069273802603
H	4.86879613533799	8.11621400736452	8.14924074685369

C	4.44851007223575	10.89699550607756	13.32419773790590
H	3.68108085942111	11.09090221481155	14.05978084875680
C	5.75292352436033	10.38024941541469	13.58469771557458
H	6.15555492245780	10.13549355687424	14.55815982869323
C	6.44032215585299	10.24941774593649	12.34046887821858
H	7.45855555025320	9.91448412706208	12.21134153605352
C	5.55646851337026	10.65958340030152	11.31017031502549
H	5.77953476750458	10.66242680624014	10.25278415638787
C	4.32836502924565	11.06078293280615	11.91882569974709
H	3.44721463419364	11.41247843892600	11.40063057217889

Compound **6** (E isomer)

Cl	1.52770965126137	13.50768165199517	5.78377890545806
Fe	3.54965964496935	14.41904679385791	6.56732177714066
Fe	4.06035650642669	16.09164380648336	4.79685742187595
N	4.55524800888543	13.27375315350440	4.05139080540568
O	1.84320309348211	15.93288004547722	2.94519090165462
O	2.09506706636255	16.91097090084683	6.81756252766878
C	2.70734332745656	15.95955784640397	3.72231634079512
C	2.82769850112100	16.10725222483469	6.34246588274700
C	4.21481927373499	14.22429689078015	4.87661325240674
C	4.49291978501603	11.85402867682388	4.44725607318915
H	4.27557981140354	11.24437956508770	3.56975511680655
H	5.44873723814672	11.53660619261461	4.87374713834345
H	3.69444462822050	11.73336747839224	5.17733033338965
C	4.98219430101693	13.56071698127368	2.65362533991403
H	5.02117632268412	14.64888111319130	2.59840609954042
C	6.37795251347464	13.00994505545838	2.35238687375570
H	7.09070650181930	13.37382015247722	3.10246547588301
H	6.36864260263200	11.91437230983605	2.41854374126991
C	6.80975640239331	13.43199434047023	0.94119174617928
H	7.79677971982949	13.01128469663799	0.71581939415121
H	6.91534826195364	14.52594027107586	0.91043069119268
C	5.78659114462760	12.99200005571735	-0.11263530231679
H	5.76319660287629	11.89363935020751	-0.15458199934222
H	6.09373129680854	13.34380992144480	-1.10483480998338
C	4.38238510043677	13.51133338415776	0.21985606344089
H	4.37700944642668	14.60989103004225	0.15823168411528
H	3.65471352968854	13.14930447036419	-0.51575473611076
C	3.94803842832290	13.08378246483664	1.62876847542032
H	3.85887843052390	11.99101610992778	1.65932964618602
H	2.96747584332000	13.50098427087660	1.87941653866315
C	5.34491674257676	14.02276108589139	7.57346594901914
H	6.31708914730928	14.10625724332639	7.10807791854846
C	4.54617539419091	12.84492181042301	7.64132706851189
H	4.79741906115000	11.87518417580747	7.24056202187023
C	3.34434483995877	13.17972706180511	8.32720590118963
H	2.50164621600695	12.51768306165864	8.47095240100548

C	3.40136037087420	14.53870398623377	8.72306517972905
H	2.62590689160611	15.08863923008873	9.23768951421099
C	4.63434216507804	15.07118893448989	8.24186643850893
H	4.97144638194392	16.09058525935928	8.36726522786352
C	6.12098705457607	16.43913376741097	4.25657690418599
H	6.80511743986082	15.66619352650829	3.93637448819915
C	5.24089993673031	17.18148335389727	3.42596490689090
H	5.14295648388180	17.09361588716377	2.35211081056433
C	4.50944528780361	18.09932349652207	4.26209530578490
H	3.75918771822312	18.80157391622613	3.92514374761176
C	4.93665107656201	17.90189999308764	5.59539420261237
H	4.55798289710378	18.41692162017010	6.46688638560099
C	5.92541737666294	16.87145569043253	5.59611303894098
H	6.43914353257912	16.48730269439973	6.46434319031114

Compound **6** (Z isomer)

Cl	1.58246675232065	13.45666515288726	5.80892370036800
Fe	3.59481993805352	14.40454857366358	6.57600486268605
Fe	4.08516159698514	16.08758146665192	4.80983636160339
N	4.57786797378862	13.28114665359618	4.03592418752511
O	1.89707507770019	15.85171827864091	2.93191588371996
O	2.09000643175344	16.86803126001982	6.81449941606916
C	2.74922789657583	15.91771449093597	3.71967499831235
C	2.84198005796165	16.08084859115108	6.34192390529506
C	4.25537965511809	14.22432499747925	4.87661394304902
C	4.98601484938099	13.55956310070336	2.64901261366176
H	5.85968484671835	12.95547427595773	2.39235164354105
H	4.16805630779540	13.31951296156938	1.96435693143001
H	5.23194348277674	14.61266258869622	2.55530999987362
C	4.45449157228294	11.84211643994747	4.41334340852963
H	3.90078098137387	11.85893521914055	5.35195000554674
C	3.62485945648550	11.04504493450883	3.40461011386368
H	2.65235572387930	11.53102425623089	3.27043870166134
H	4.12878124424538	11.02036897339133	2.43103865584591
C	3.44926084056532	9.60711265227054	3.91249355702739
H	2.86615120906612	9.02958891053598	3.18545904627300
H	2.86969053019350	9.62438957090867	4.84627154907546
C	4.80443453026333	8.93574648318457	4.16515681377461
H	5.33857828264159	8.83418650726695	3.20950005850069
H	4.65863081456610	7.92239133793158	4.55859083488366
C	5.66107189505010	9.75895709489836	5.13404880863683
H	5.17959154911135	9.76334527806906	6.12270113516410
H	6.64702112146040	9.29810441881025	5.26546358604197
C	5.82618155305387	11.20518151335302	4.64593546933371
H	6.40005472495225	11.20687965803759	3.71036864054997
H	6.38905043648257	11.80041848097273	5.37333112739830
C	5.39568337136720	14.05772761651653	7.59258591800547
H	6.36784927846022	14.16222069355990	7.13202004214170

C	4.62726857716430	12.86092873525086	7.66708085226911
H	4.90852441464260	11.89221297503980	7.28482345130010
C	3.41426332528201	13.16912948825817	8.34393011566139
H	2.58881965957113	12.48588591743288	8.48870857554709
C	3.43459988868032	14.53221379028050	8.73056548394823
H	2.64302622862311	15.06591119103663	9.23759749058109
C	4.65601982791653	15.09200003446258	8.25164377214650
H	4.96683869305221	16.12023772060046	8.37169766822498
C	6.14637436511493	16.46170669033470	4.29467402748594
H	6.84365291659390	15.69268237492084	3.99158470876674
C	5.26571814514988	17.18204657386657	3.44464530954153
H	5.18233237987057	17.08015066804972	2.37086124455475
C	4.50901407351212	18.09671775104502	4.26162587514516
H	3.75185557029069	18.78318193019487	3.90791775216217
C	4.92170741871584	17.92037413740827	5.60241208547836
H	4.52368659401473	18.43834491694813	6.46348618593816
C	5.92712737827115	16.90638249364078	5.62661811112278
H	6.43596656110441	16.54036017974169	6.50547137073724

#### 4. Stability studies in aqueous media.

##### Stability in water.

A mixture of the selected Fe compound (*ca.* 4 mg) and a D<sub>2</sub>O solution (0.9 mL) containing Me<sub>2</sub>SO<sub>2</sub> ( $3.36 \cdot 10^{-3}$  M) was stirred for 30 minutes then filtered over celite and transferred into an NMR tube. The orange-red solution was analyzed by <sup>1</sup>H, <sup>19</sup>F{<sup>1</sup>H}, <sup>31</sup>P{<sup>1</sup>H}, <sup>35</sup>Cl NMR then heated at 37 °C for 72 h. After cooling to room temperature, the final solution was separated from an orange-brown solid by filtration over celite, and the <sup>1</sup>H NMR spectrum was recorded (delay time = 3 s; number of scans = 20). In each case, no new {FeCp} species was identified. The amount of starting material in solution (% with respect to the initial spectrum) was calculated by the relative integral with respect to Me<sub>2</sub>SO<sub>2</sub> as internal standard [<sup>31</sup>] ( $\delta/\text{ppm} = 3.14$  (s, 6H)) (Table S2). The aqueous solution was then extracted with CDCl<sub>3</sub> (3 x 0.25 mL) and analyzed by <sup>1</sup>H and <sup>19</sup>F NMR, in order to confirm the identity of the compounds.

**[2A]CF<sub>3</sub>SO<sub>3</sub>.** <sup>1</sup>H NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = 8.47, 7.96, 7.79, 7.60, 6.76, 5.47, 4.62$ . <sup>19</sup>F NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = -78.8$ .

**[2B]BF<sub>4</sub>.** <sup>1</sup>H NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = 5.41, 5.39$  (s, 10H); 5.27–5.21 (dd, <sup>2</sup>J<sub>HP</sub> = 12.7 Hz, <sup>2</sup>J<sub>HH</sub> = 15.5 Hz, 1H), 5.13–5.03 (dd, <sup>2</sup>J<sub>HP</sub> = 13.1 Hz, <sup>2</sup>J<sub>HH</sub> = 15.7 Hz, 1H), 4.38 (app. quint, <sup>3</sup>J<sub>HH</sub> = <sup>3</sup>J<sub>HP</sub> = 7.5 Hz, 4H), 4.30 (s, 3H), 1.43 (t, *J* = 7.0 Hz, 6H). <sup>19</sup>F NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = -150.4, -150.5$ . <sup>31</sup>P NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = 19.6$ .

**[2C]CF<sub>3</sub>SO<sub>3</sub>.** <sup>1</sup>H NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = 5.37, 5.35$  (s, 10H); 4.08 (s, 3H), 2.36 (d, *J* = 10.7 Hz, 1H); 2.19–2.03 (m, 2H), 1.99–1.74 (m, 4H), 1.69–1.45 (m, 3H), 1.38–1.27 (m, 1H). <sup>19</sup>F NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = -78.8$ .

**[2C]Cl.** <sup>1</sup>H NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = 5.37, 5.35$  (s, 10 H); 4.07 (s, 3H), 2.35 (d, *J* = 12.1 Hz, 1H); 2.19–2.04 (m 2H), 1.98–1.75 (m, 4H), 1.70–1.46 (m, 2H), 1.38–1.27 (m, 2H). <sup>35</sup>Cl NMR (D<sub>2</sub>O):  $\delta/\text{ppm} = 0.35$  ( $\Delta\nu_{1/2} = 17$  Hz).



**[2D]CF<sub>3</sub>SO<sub>3</sub>.** <sup>1</sup>H NMR (D<sub>2</sub>O): δ/ppm = 7.65 (d, *J* = 8.2 Hz, 2H), 7.27 (d, *J* = 8.3 Hz, 2H); 5.45, 4.83 (s, 10H); 4.54 (s, 3H), 3.96 (s, 3H). <sup>19</sup>F NMR (D<sub>2</sub>O): δ/ppm = − 78.9.

**[3F]I.** <sup>1</sup>H NMR (D<sub>2</sub>O): δ/ppm = 6.28–6.17 (m, 1H), 5.58 (app. d, *J* = 12.4 Hz, 2H); 5.39, 5.36 (s, 10H); 5.19 (dd, *J* = 15.5, 4.8 Hz, 1H), 5.07 (dd, *J* = 15.1, 5.1 Hz, 1H), 4.16 (s, 3H).

**[5H]CF<sub>3</sub>SO<sub>3</sub>.** <sup>1</sup>H NMR (D<sub>2</sub>O): δ/ppm = 7.57–7.47 (m, 6H), 7.35–7.29 (m, 4H), 5.77 (d, *J* = 15.8 Hz, 2H), 5.65 (d, *J* = 14.8 Hz, 2H), 5.43 (s, 10H). <sup>19</sup>F NMR (D<sub>2</sub>O): δ/ppm = − 78.8.

### Isolation and identification of the precipitate from H<sub>2</sub>O.

A suspension of [2C]CF<sub>3</sub>SO<sub>3</sub> (57 mg, 0.095 mmol) in H<sub>2</sub>O (20 mL) was vigorously stirred at room temperature for 30 minutes then filtered over celite. The resulting red solution was stirred at 37 °C for 72 h, then cooled to room temperature and filtered (G4 porous filter). The resulting brown solid was washed with water and dried under vacuum (50 °C). Yield: 6 mg. The sample was analyzed by Raman spectroscopy and identified as γ-Fe<sub>2</sub>O<sub>3</sub> (maghemite, Raman shift: 711 cm<sup>−1</sup>).

### Stability in water/methanol.

A solution of the selected Fe compound in a D<sub>2</sub>O/CD<sub>3</sub>OD mixture (2:1 v/v for [2A]CF<sub>3</sub>SO<sub>3</sub>, [2B]BF<sub>4</sub>; 5:4 v/v for [5H]CF<sub>3</sub>SO<sub>3</sub>, [4G]BF<sub>4</sub>) was prepared and treated as described above. The residual amount of starting material in solution after 72 h at 37 °C was calculated with respect to Me<sub>2</sub>SO<sub>2</sub> as internal standard (Table S2).

**[2A]CF<sub>3</sub>SO<sub>3</sub>.** <sup>1</sup>H NMR (D<sub>2</sub>O/CD<sub>3</sub>OD 2:1): δ/ppm = 7.89 (s, 1H), 7.76 (d, *J* = 8.4 Hz, 1H), 7.58 (d, *J* = 3.1 Hz, 1H), 7.48–7.39 (m, 1H), 6.73 (d, *J* = 2.8 Hz, 1H); 5.48, 4.73 (s, 10H), 4.63 (s, 3H).

**[2B]BF<sub>4</sub>.** <sup>1</sup>H NMR: δ/ppm (D<sub>2</sub>O/CD<sub>3</sub>OD 2:1): δ/ppm = 5.41, 5.40 (s, 10H); 5.27–5.18 (m, 1H), 5.12–5.03 (m, 1H), 4.37 (dt, *J* = 13.9, 6.9 Hz, 1H), 4.30 (s, 3H), 1.43 (td, *J* = 7.0, 3.2 Hz, 6H). <sup>31</sup>P{<sup>1</sup>H} NMR: δ/ppm (D<sub>2</sub>O/CD<sub>3</sub>OD 2:1): δ/ppm = 20.5.

**[4G]BF<sub>4</sub>**. <sup>1</sup>H NMR (D<sub>2</sub>O/CD<sub>3</sub>OD 5:4): δ/ppm = 8.29 (d, *J* = 8.6 Hz, 1H), 8.19–7.98 (m, 3H), 7.80–7.73 (m, 3H), 5.52 (s, 5H), 5.10–5.01\* (m), 4.72 (s, 5H), 1.51 (t, *J* = 7.2 Hz, 3H). \*Over OH residual peak.

**[5H]CF<sub>3</sub>SO<sub>3</sub>**. <sup>1</sup>H NMR (D<sub>2</sub>O/CD<sub>3</sub>OD 5:4): δ/ppm = 7.56–7.49 (m, 6H), 7.29 (d, *J* = 7.2 Hz, 4H), 5.76 (d, *J* = 15.3 Hz, 2H), 5.62 (d, *J* = 15.3 Hz, 2H), 5.44 (s, 10H).

### Stability in DMSO/water.

A solution of the selected Fe compound in a DMSO-d<sub>6</sub>/D<sub>2</sub>O mixture (1:1 v/v for **[2A]CF<sub>3</sub>SO<sub>3</sub>**, **[5H]CF<sub>3</sub>CO<sub>3</sub>**; 5:3 v/v for **[4G]BF<sub>4</sub>**) was prepared and treated as described above. In this case, formation of an orange-brown precipitate was not observed. However, after 72 h at 37 °C, a new set of signals in the <sup>1</sup>H spectrum was detected, attributed to the DMSO adduct [Fe<sub>2</sub>Cp<sub>2</sub>(CO)(μ-CO){μ-CNMeR}(DMSO)]<sup>+</sup> (**[2A<sup>S</sup>]<sup>+</sup>**, **[4G<sup>S</sup>]<sup>+</sup>**, **[5H<sup>S</sup>]<sup>+</sup>**; Scheme S2).<sup>1</sup> The residual amount of starting material in solution was calculated with respect to Me<sub>2</sub>SO<sub>2</sub> as internal standard (Table S2).

**[2A]CF<sub>3</sub>SO<sub>3</sub>**. <sup>1</sup>H NMR (DMSO-d<sub>6</sub>/D<sub>2</sub>O 1:1): δ/ppm = 7.78 (s, 1H), 7.62 (d, *J* = 8.4 Hz, 1H), 7.45 (s, 1H), 7.28 (s, 1H), 6.60 (s, 1H); 5.37, 4.61 (s, 10H). <sup>19</sup>F NMR (DMSO-d<sub>6</sub>/D<sub>2</sub>O 1:1): δ/ppm = – 77.8.

**[2A<sup>S</sup>]<sup>+</sup>**. <sup>1</sup>H NMR (DMSO-d<sub>6</sub>/D<sub>2</sub>O 1:1): δ/ppm = 6.56 (s); 5.25, 5.06 (s, 5H); 4.70 (s, 3H); 4.50 (s).

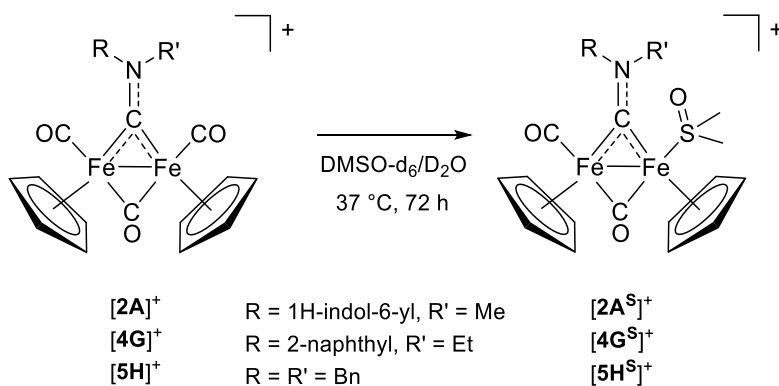
**[4G]BF<sub>4</sub>**. <sup>1</sup>H (DMSO-d<sub>6</sub>/D<sub>2</sub>O 5:3): δ/ppm = 8.14 (d, *J* = 8.7 Hz, 1H), 8.10–7.85 (m, 3H), 7.64–7.60 (m, 3H), 5.36 (s, 5H), 4.93–4.80 (m, 2H), 4.57\* (s), 1.32 (t, *J* = 7.2 Hz, 3H). <sup>19</sup>F NMR (DMSO-d<sub>6</sub>/D<sub>2</sub>O 5:3): δ/ppm = – 148.69, – 148.74.

**[4G<sup>S</sup>]<sup>+</sup>**. <sup>1</sup>H NMR (DMSO-d<sub>6</sub>/D<sub>2</sub>O 5:3): δ/ppm = 5.25, 5.06 (s, 5H); 4.47\* (s), 1.35 (m, 3H).

\*Superimposed on HDO peak.

**[5H]CF<sub>3</sub>SO<sub>3</sub>**. <sup>1</sup>H (DMSO-d<sub>6</sub>/D<sub>2</sub>O 1:1): δ/ppm = 7.44–7.34 (m, 6H), 7.16 (d, *J* = 6.7 Hz, 4H), 5.61 (d, *J* = 15.5 Hz, 2H), 5.47 (d, *J* = 15.4 Hz, 2H), 5.30 (s, 10H). <sup>19</sup>F NMR (DMSO-d<sub>6</sub>/D<sub>2</sub>O 1:1): δ/ppm = – 78.1.

**[5H<sup>S</sup>]<sup>+</sup>**. <sup>1</sup>H NMR (DMSO-d<sub>6</sub>/D<sub>2</sub>O 1:1): δ/ppm = 6.22 (d, *J* = 15.7 Hz, 1H), 5.70 (d, *J* = 15.1 Hz, 1H); 5.26, 4.97 (s, 10H).



**Scheme S2.** Formation of aminocarbene-DMSO complexes in DMSO/water solutions of  $[2A]^+$ ,  $[4G]^+$  and  $[5H]^+$  at 37 °C.

### Stability in aqueous solution (isocyanide complexes).

A solution of the selected Fe compound in an organic solvent/D<sub>2</sub>O 2:1 v/v mixture (CD<sub>3</sub>OD for **1B**; acetone-d<sub>6</sub> for **1C**) was prepared as described above and maintained at 37 °C for 24 h. After cooling to room temperature, the final solution was separated from an orange-brown solid by filtration over celite and <sup>1</sup>H/<sup>31</sup>P NMR analyses were repeated. Next, volatiles were removed under vacuum and the residue was analyzed by IR (CH<sub>2</sub>Cl<sub>2</sub>). In both cases, the starting material was identified as the major species in the final solution, along with other products.

**1B.** Red-violet solution. <sup>1</sup>H NMR (CD<sub>3</sub>OD/D<sub>2</sub>O 2:1): δ/ppm = 5.0 (br, 10H), 4.5 (br, 2H), 4.3 (br, 4H), 1.4 (br, 6H). Signal for CH hidden by HDO peak. <sup>31</sup>P{<sup>1</sup>H} NMR (CD<sub>3</sub>OD/D<sub>2</sub>O 2:1): δ/ppm = 24.6.

**1C.** Red-brown solution. <sup>1</sup>H NMR (acetone-d<sub>6</sub>/D<sub>2</sub>O 2:1): δ/ppm = 4.7, 4.6 (br, 10H); 3.4 (br, 1H), 1.5 (br, 4H), 1.2 (br, 6H). *Other species* (24 h). <sup>1</sup>H NMR (acetone-d<sub>6</sub>/D<sub>2</sub>O 2:1): δ/ppm = 6.5, 6.4, 2.6 (br, CpH); 5.3, 4.9, 2.8, 1.8, 1.7 (br). IR (CH<sub>2</sub>Cl<sub>2</sub>):  $\tilde{\nu}/\text{cm}^{-1}$  = 2201m, 2179m, 2025m, 1995m, 1771m-sh.

### Stability in cell culture medium.

**Method A.** Powdered DMEM cell culture medium (1000 mg/L glucose and L-glutamine, without sodium bicarbonate and phenol red; D2902 - Sigma Aldrich) was dissolved in D<sub>2</sub>O (10 mg/mL), according to the manufacturer's instructions. The solution of deuterated cell culture medium ("DMEM-d") was treated with Me<sub>2</sub>SO<sub>2</sub> ( $6.6 \cdot 10^{-3}$  M) and NaH<sub>2</sub>PO<sub>4</sub> / Na<sub>2</sub>HPO<sub>4</sub> (0.15 M, pD = 7.5 <sup>[4]</sup>), then stored at 4 °C under N<sub>2</sub>. The selected Fe compound (2-3 mg) was dissolved in CD<sub>3</sub>OD (0.05 mL; 0.2 mL for [2A]CF<sub>3</sub>SO<sub>3</sub>; 0.3 mL for [4G]BF<sub>4</sub> and [5H]CF<sub>3</sub>SO<sub>3</sub>) then diluted with DMEM-d up to 0.75 mL total volume ( $c_{\text{Fe2}}$  ca.  $6 \cdot 10^{-3}$  M; ca. 6.6% MeOH). The mixture was stirred for 30 minutes then filtered over celite and transferred into an NMR tube. The resulting red-orange solution was analyzed by <sup>1</sup>H and <sup>31</sup>P NMR (delay time = 3 s; number of scans = 20) then heated at 37 °C for 24 h. After cooling to room temperature, the solution was filtered over celite and NMR analyses were repeated. The amount of starting material (% with respect to the initial spectrum) was calculated by the relative integral with respect to Me<sub>2</sub>SO<sub>2</sub> as internal standard<sup>3</sup> ( $\delta/\text{ppm} = 2.95$  (s, 6H)) (Table S2).

**Method B.** The selected Fe compound (ca. 6 mg) was dissolved in DMSO (0.1 mL; 0.5 mL for [2A]CF<sub>3</sub>SO<sub>3</sub>, [4G]BF<sub>4</sub> and [5H]CF<sub>3</sub>SO<sub>3</sub>) then diluted with RPMI-1640 cell culture medium (5.0 mL; Merck; modified with sodium bicarbonate, without L-glutamine and phenol red). The orange-red solution was heated at 37 °C for 72 h then allowed to cool to room temperature. The resulting suspension was diluted with water (5 mL) and extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 x 10 mL). The combined organic extracts were dried under vacuum (40 °C) and the residue was analyzed by IR (CH<sub>2</sub>Cl<sub>2</sub>), <sup>1</sup>H and <sup>31</sup>P{<sup>1</sup>H} NMR (CDCl<sub>3</sub>; CD<sub>3</sub>CN for [2A]CF<sub>3</sub>SO<sub>3</sub>). In all cases, the starting material was identified as the major species. Its relative amount (%) with respect to other {FeCp} compounds, calculated by <sup>1</sup>H NMR on the basis of Cp and NMe resonances ( $\delta_{\text{H}}$ : 6.0–3.5 ppm), resulted  $\geq 90$  % for all the compounds except [4G]BF<sub>4</sub> and [5H]CF<sub>3</sub>SO<sub>3</sub> (ca. 75%).

**Table S3.** Stability of diiron complexes in D<sub>2</sub>O, D<sub>2</sub>O/CD<sub>3</sub>OD, DMSO-d<sub>6</sub>/D<sub>2</sub>O and cell culture medium (DMEM-d/CD<sub>3</sub>OD) solution at 37 °C calculated by <sup>1</sup>H NMR (Me<sub>2</sub>SO<sub>2</sub> internal standard).

Compound	% starting material (72 h)			% starting material (24 h) DMEM-d (6.6 % CD <sub>3</sub> OD)
	D <sub>2</sub> O	D <sub>2</sub> O/CD <sub>3</sub> OD	DMSO-d <sub>6</sub> /D <sub>2</sub> O	
[2A]CF <sub>3</sub> SO <sub>3</sub>		55 (2:1 v/v)	89 (1:1 v/v)	86
[2B]BF <sub>4</sub>	85	81 (2:1 v/v)		88
[2C]CF <sub>3</sub> SO <sub>3</sub>	75			85
[2C]Cl	79			88
[2D]CF <sub>3</sub> SO <sub>3</sub>	86			77
[2E]CF <sub>3</sub> SO <sub>3</sub> <sup>[b]</sup>	51	80 (5:2 v/v)		70
[2F]CF <sub>3</sub> SO <sub>3</sub> <sup>[b]</sup>	70	83 (5:2 v/v)		76
[3F]X <sup>[a]</sup>	60			84
[4G]BF <sub>4</sub>		51 (5:4 v/v)	42 (5:3 v/v)	85
[5H]CF <sub>3</sub> SO <sub>3</sub>	ca. 50	65 (5:4 v/v)	89 (1:1 v/v)	75

[a] X = I for the stability in D<sub>2</sub>O, X = CF<sub>3</sub>SO<sub>3</sub> for the stability in cell culture medium. [b] Stability in D<sub>2</sub>O and D<sub>2</sub>O/CD<sub>3</sub>OD taken from the literature.<sup>1</sup>

**Table S4.** Release of CO (number of equivalents) over a 24 h period as determined by GC-TCD analysis and stability of selected Fe compounds after 48 h in a 5 % MeOH aqueous solution at 37 °C.

Compound	eq. CO released <sup>[a]</sup>		% residual complex <sup>[b]</sup>
	0-24 h	24-48 h	
[2C]CF <sub>3</sub> SO <sub>3</sub>	0.25 ± 0.02	0.23 ± 0.04	83-86
[2E]CF <sub>3</sub> SO <sub>3</sub>	0.31 ± 0.07	0.29 ± 0.04	72-78
[2F]CF <sub>3</sub> SO <sub>3</sub>	0.32 ± 0.03	0.28 ± 0.06	78-82
[3F]CF <sub>3</sub> SO <sub>3</sub>	0.22 ± 0.06	0.15 ± 0.01	87-88
[5H]CF <sub>3</sub> SO <sub>3</sub>	0.29 ± 0.06	0.27 ± 0.02	80-84

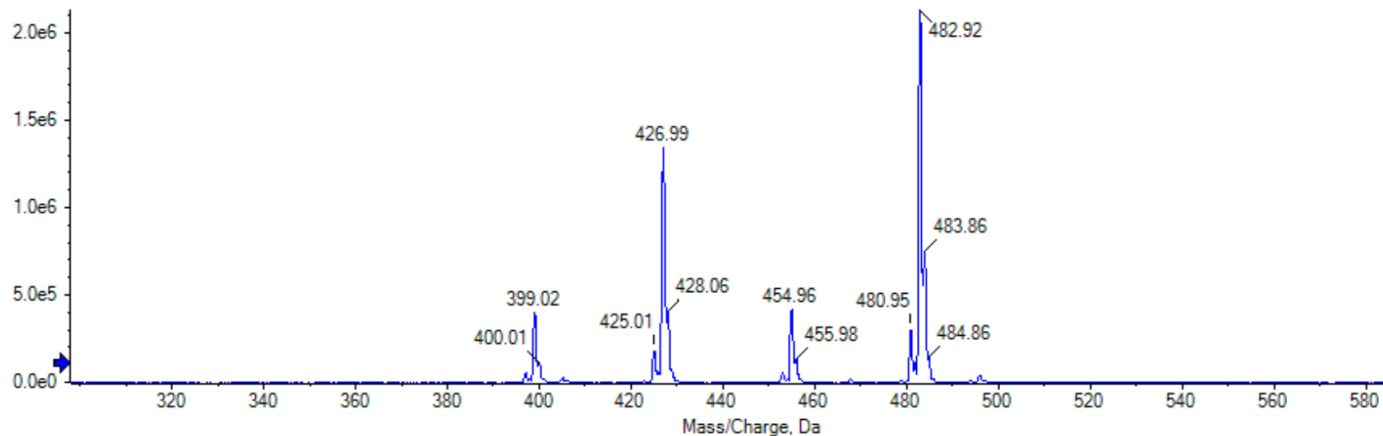
[a] eq<sub>CO</sub> = n<sub>CO</sub>/n<sub>Fe</sub>. [b] Calculated as  $[1 - \Sigma(\text{eq}_{\text{CO}})/3] \cdot 100$ .

**Table S5.** In vitro antitumor activity of diiron complexes in PSN-1 cancer cells after 24 hours. Values after 72 h are also provided for sake of comparison.

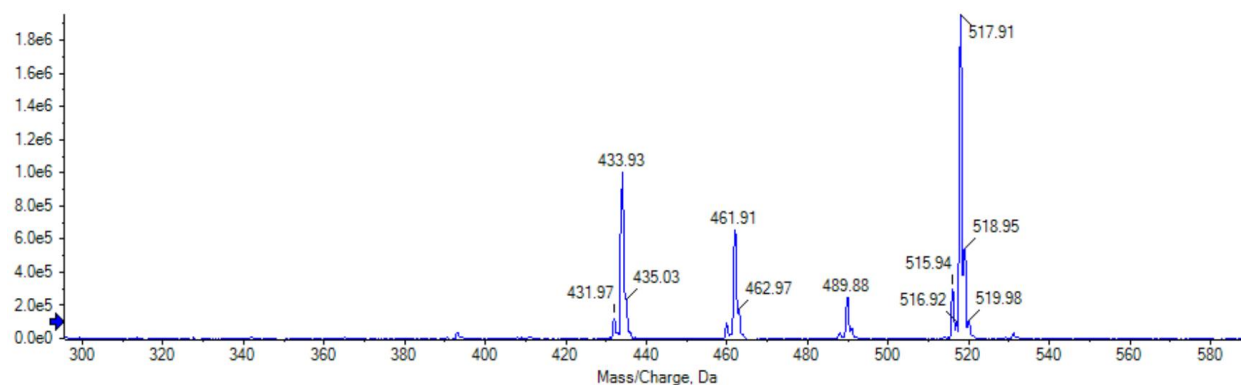
Compound	IC <sub>50</sub> (μM) ± SD	IC <sub>50</sub> (μM) ± SD
	24 h	72 h
[2A]CF <sub>3</sub> SO <sub>3</sub>	144 ± 16	54.3 ± 6.2
[2B]BF <sub>4</sub>	> 200	> 100
[2C]CF <sub>3</sub> SO <sub>3</sub>	94.2 ± 3.8	15.3 ± 2.7
[2D]CF <sub>3</sub> SO <sub>3</sub>	35.0 ± 2.0	16.2 ± 2.9
[2E]CF <sub>3</sub> SO <sub>3</sub>	156.0 ± 4.0	41.7 ± 6.3
[3F]CF <sub>3</sub> SO <sub>3</sub>	> 200	> 100
[3G]CF <sub>3</sub> SO <sub>3</sub>	136.4 ± 3.2	26.3 ± 2.8
[4G]BF <sub>4</sub>	69.5 ± 2.1	25.6 ± 4.5
[5H]CF <sub>3</sub> SO <sub>3</sub>	112.4 ± 2.4	33.7 ± 4.2

Cells ( $3 \times 10^3 \text{ mL}^{-1}$ ) were treated for 24 h with increasing concentrations of the tested compounds. The cytotoxicity was assessed by the MTT test. IC<sub>50</sub> values were calculated by a four-parameter logistic model 4-PL ( $P < 0.05$ ).

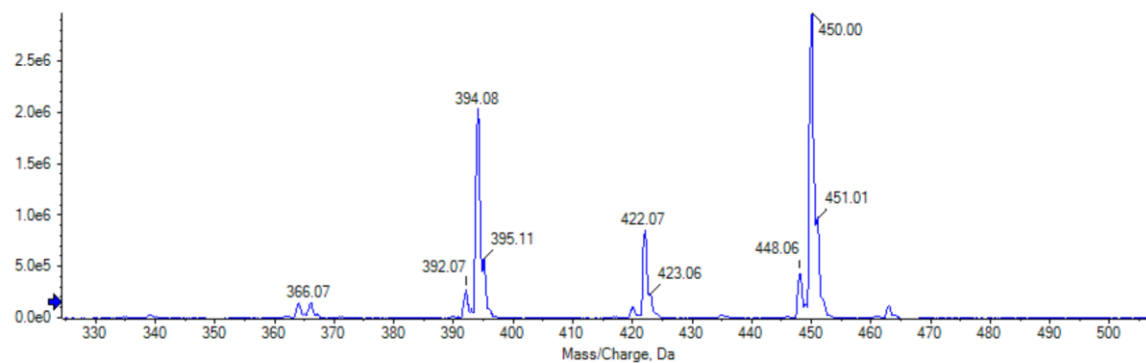
## 6. Lysozyme interaction studies (MS spectra)



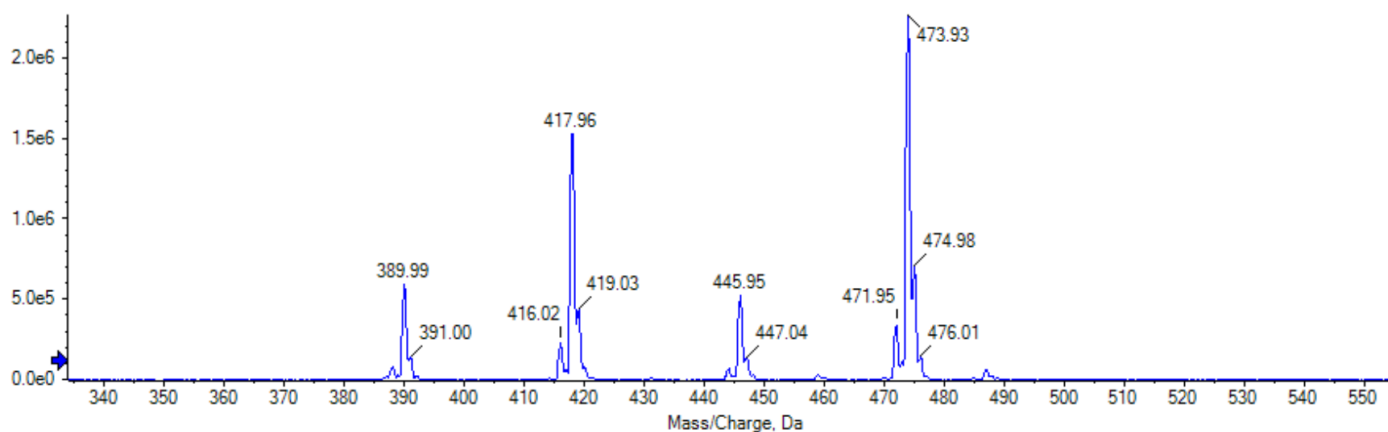
**Figure S33.** MS spectrum for  $[2A]^+$  ( $C_{23}H_{18}Fe_2N_2O_3$ ). Calcd. base peak for  $[2A]^+$ : 483.018 Da;  $[2A]^+-CO$ : 455.023 Da;  $[2A]^+-2CO$ : 427.028 Da;  $[2A]^+-3CO$ : 399.034 Da.



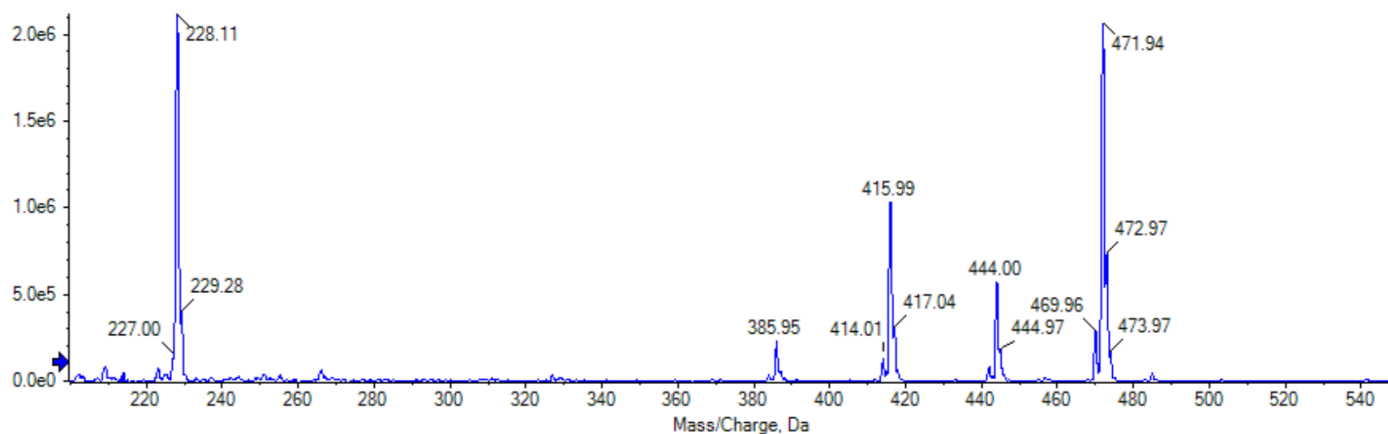
**Figure S34.** MS spectrum for  $[2B]^+$  ( $C_{20}H_{25}Fe_2NO_6P$ ). Calcd. base peak for  $[2B]^+$ : 450.054 Da;  $[2B]^+-CO$ : 422.060 Da;  $[2B]^+-2CO$ : 394.065 Da;  $[2B]^+-3CO$ : 434.036 Da.



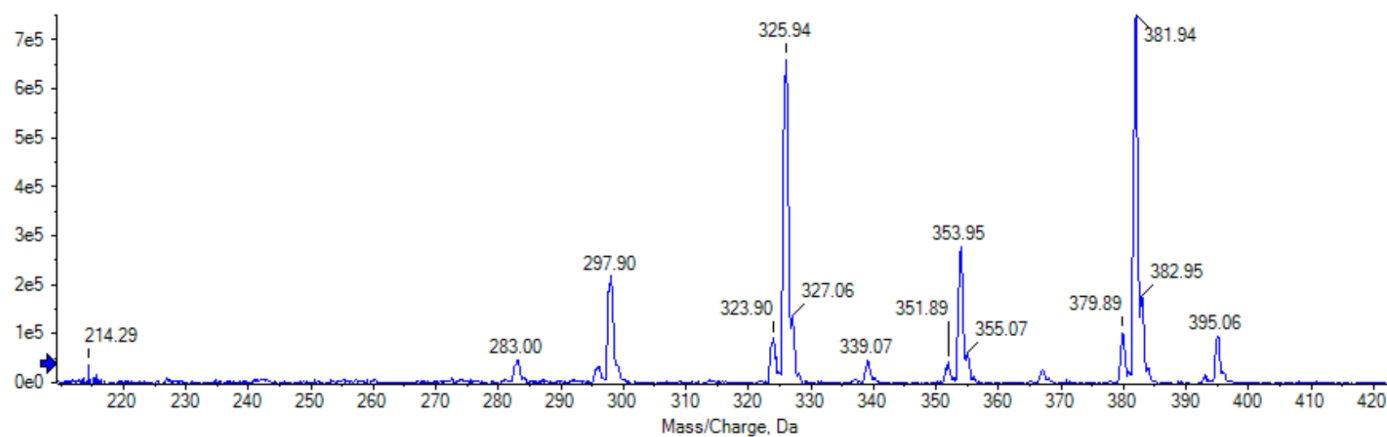
**Figure S35.** MS spectrum for  $[2C]^+$  ( $C_{21}H_{24}Fe_2NO_3$ ). Calcd. base peak for  $[2C]^+$ : 450.054 Da;  $[2C]^+-CO$ : 422.060 Da;  $[2C]^+-2CO$ : 394.065 Da.



**Figure S36.** MS spectrum for  $[2D]^+$  ( $C_{22}H_{20}Fe_2NO_4$ ). Calcd. base peak for  $[2D]^+$ : 474.018 Da;  $[2D]^+-CO$ : 446.023 Da;  $[2D]^+-2CO$ : 418.029 Da;  $[2D]^+-3CO$ : 390.034.

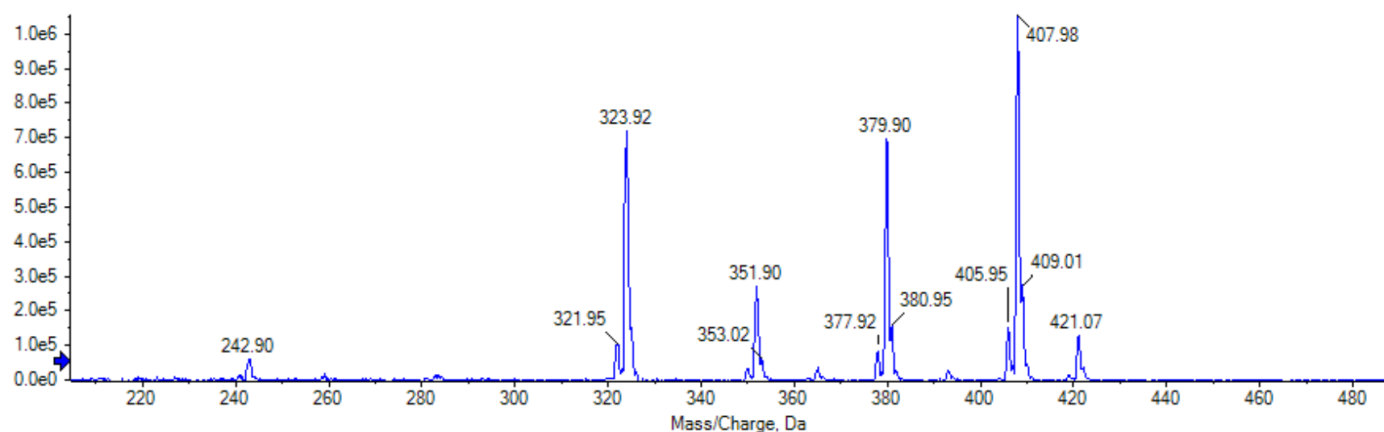


**Figure S37.** MS spectrum for  $[2E]^+$  ( $C_{23}H_{22}Fe_2NO_3$ ). Calcd. base peak for  $[2E]^+$ : 472.039 Da;  $[2E]-CO$ : 444.044 Da;  $[2E]^+-2CO$ : 416.049 Da;  $[2E]^+-3CO$ : 388.054 Da.

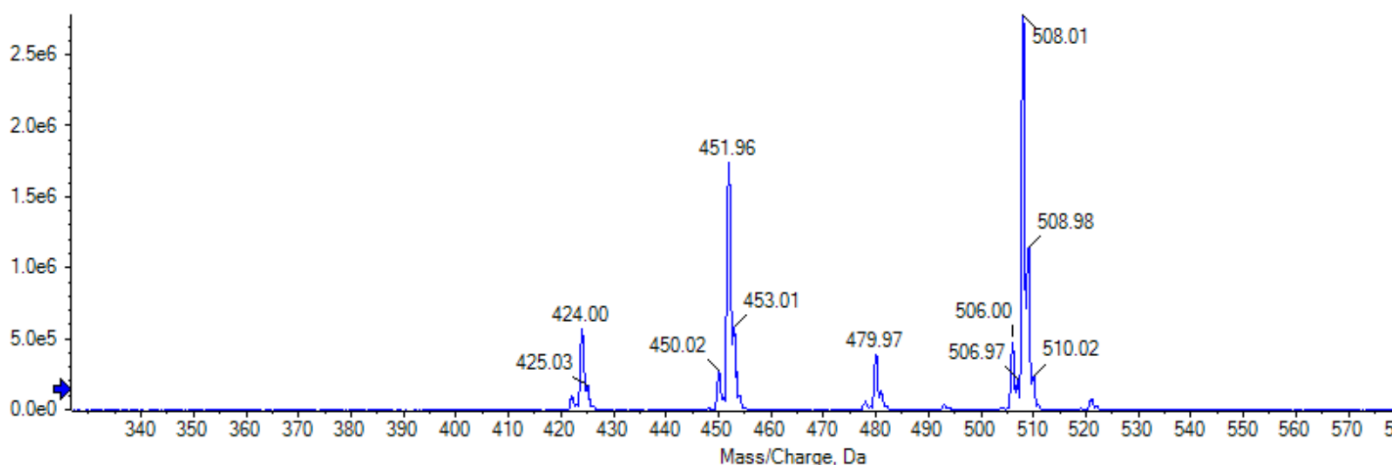


**Figure S38.** MS spectrum for  $[2F]^+$  ( $C_{16}H_{16}Fe_2NO_3$ ). Calcd. base peak for  $[2F]^+$ : 381.992 Da;  $[2F]^+-CO$ : 353.997 Da;  $[2F]^+-2CO$ : 326.002 Da;  $[2F]^+-3CO$ : 298.007 Da.

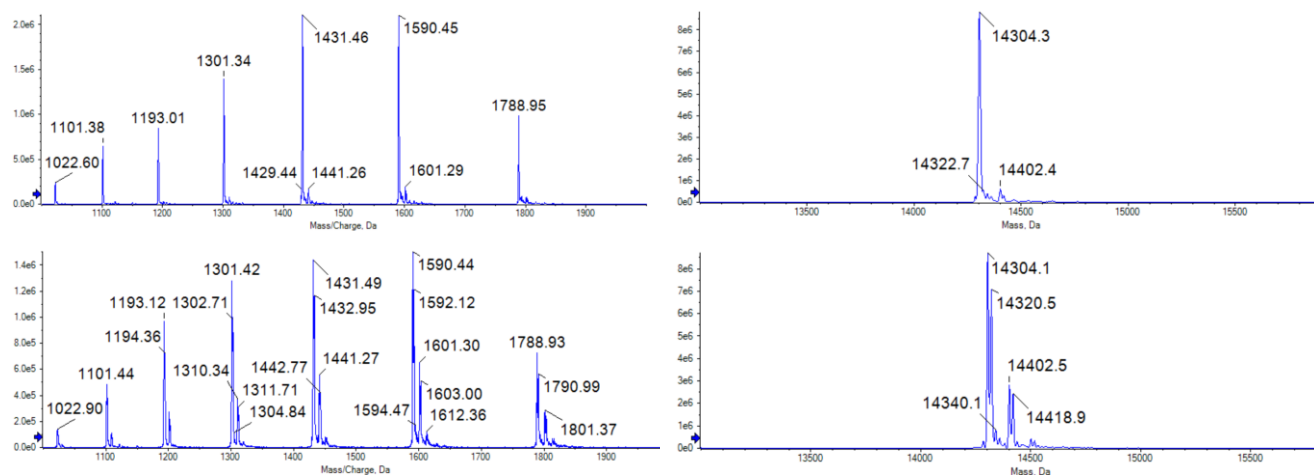




**Figure S39.** MS spectrum for  $[3F]^+$  ( $C_{18}H_{18}Fe_2NO_3$ ). Calcd. base peak for  $[3F]^+$ : 408.007 Da;  $[3F]^+-CO$ : 380.013 Da;  $[3F]^+-2CO$ : 352.018 Da;  $[3F]^+-3CO$ : 324.023 Da.



**Figure S40.** MS spectrum for  $[4G]^+$  ( $C_{26}H_{22}Fe_2NO_3$ ). Calcd. base peak for  $[4G]^+$ : 508.039 Da;  $[4G]^+-CO$ : 480.044 Da;  $[4G]^+-2CO$ : 452.049 Da;  $[4G]^+-3CO$ : 424.054 Da.



**Figure S41.** MS spectrum and peak reconstruction for lysozyme ( $M = 14304$  Da) and lysozyme- $CH_3$  ( $M = 14320$  Da).

## 7. Notes and references

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- 1 G. Agonigi, M. Bortoluzzi, F. Marchetti, G. Pampaloni, S. Zacchini, V. Zanutti, *Eur. J. Inorg. Chem.* **2018**, 960–971, and references therein.
  - 2 L. Busetto, F. Marchetti, S. Zacchini, V. Zanutti, *Organometallics* **2006**, 25, 4808-4816.
  - 3 T. Rundlöf, M. Mathiasson, S. Bekiroglu, B. Hakkarainen, T. Bowden, T. Arvidsson, *J. Pharm. Biomed. Anal.* **2010**, 52, 645–651.
  - 4 Calculated by the formula  $pD = pH^* + 0.4$ , where  $pH^*$  is the value measured for H<sub>2</sub>O-calibrated pH-meter. a) C. C. Westcott, pH Measurements; Academic Press: New York, **1978**. b) A. K. Covington, M. Paabo, R. A. Robinson, R. G. Bates, *Anal. Chem.* **1968**, 40, 700-706.