Organometallics

Supporting Information

A Biomimetic Model for the Active Site of [Fe]-H_2ase Featuring a 2-Methoxy-3,5-dimethyl-4-phosphato-6-acylmethylpyridine Ligand

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Contents:

1. Experimental details

(1) General considerations

All reactions were carried out using standard Schlenk and vacuum-line techniques under an atmosphere of highly purified nitrogen. MeCN, CH_2Cl_2 and N, N-dimethylformamide (DMF) were distilled under N_2 from CaH_2, while THF from sodium/benzophenone ketyl. *t*-Butyldimethylsilyl chloride (TBSCl), *n*-Bu_4NF (1 M in THF), *n*-BuLi (1.6 M in hexane), MeI, Ag_2CO_3, I_2, $O=P(PhO)_2Cl$, and some other materials are available commercially and used without further purification. 4-hydroxy-3,5,6-trimethyl-2-pyridone (**A**),[1] 6-methyl-2-mercaptopyridine,[2] and $Na_2Fe(CO)_4 \cdot (1,4\text{-dioxane})_{1.5}$[3] were prepared according to the published methods. Column chromatography was performed on silica gel (100–200 mesh) using a proper eluent. While the solid state IR spectra were recorded on a Bio-Rad FTS 135 infrared spectrophotometer, the ^1H, $^{13}C\{^1H\}$, and $^{31}P\{^1H\}$ NMR spectra in solution were obtained on a Bruker Avance 400 NMR spectrometer. Elemental analyses were performed on an Elementar Vario EL analyzer. Melting points were determined on a SGW X-4 microscopic melting point apparatus and were uncorrected.

(2) Synthesis and spectroscopic data of compounds 1–7

Preparation of 4-TBSO-3,5,6-trimethyl-2-pyridone (1). A solution consisting of 4-hydroxy-3,5,6-trimethyl-2-pyridone (**A**) (1.53 g, 10.0 mmol), TBSCl (1.51g, 10.0 mmol), Et_3N (1.70 mL, 12.0 mmol), and DMF (20 mL) was stirred at room temperature for 24 h and then a large excess of ice-water mixture (60 mL) was added. After the resulting mixture was extracted with ethyl acetate (10 mL×3), the combined

organic layers were washed with saturated aqueous NaCl (50 mL) and then was dried over anhydrous Na_2SO_4. After removal of Na_2SO_4 and solvent ethyl acetate, **1** (2.30 g, 86%) was obtained as a white solid. Mp 102−103 °C (dec). Anal. Calcd for $C_{14}H_{25}NO_2Si$: C, 62.87; H, 9.42; N, 5.24. Found: C, 62.65; H, 9.42; N, 5.17. IR (KBr disk): $\nu_{C=O}$ 1644 (vs); $\nu_{Si\text{-}CH3}$ 831 (s) cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$): 0.14 (s, 6H, $(CH_3)_2Si$), 0.98 (s, 9H, $(CH_3)_3C$), 1.87 (s, 3H, 5-CH_3 of C_5N), 1.96 (s, 3H, 6-CH_3 of C_5N), 2.26 (s, 3H, 3-CH_3 of C_5N), 13.24 (s, 1H, NH) ppm. $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): −3.1 (s, $(CH_3)_2Si$), 10.8 (s, 5-CH_3 of C_5N), 11.8 (s, 6-CH_3 of C_5N), 17.2 (s, 3-CH_3 of C_5N), 18.7 (s, *C*$(CH_3)_3$), 26.0 (s, C(*C*$H_3)_3$), 108.8, 111.7, 139.5, 162.2, 166.0 (5s, C_5N) ppm.

Preparation of 2-Methoxy-4-TBSO-3,5,6-trimethylpyridine (2). A heterogeneous solution consisting of pyridone derivative **1** (0.53 g, 2.0 mmol), Ag_2CO_3 (0.83 g, 3.0 mmol) and MeI (1.25 mL, 20.0 mmol), and CH_2Cl_2 (10 mL) was stirred at room temperature for 10 h in the dark. The resulting mixture was first filtered through celite and then washed the celite using CH_2Cl_2 (5 mL×3) to give a filtrate. Solvent was removed from the filtrate at reduced pressure to leave a residue, which was subjected to column chromatography (silica gel). Elution with ethyl acetate/petroleum ether (v:v = 1:20) and then removal of ethyl acetate/petroleum ether from the eluate produced **2** (0.430 g, 76%) as a colorless oil; R_f = 0.32 [TLC, SiO_2, ethyl acetate/petroleum ether (v:v = 1:20)]. Anal. Calcd for $C_{15}H_{27}NO_2Si$: C, 64.01; H, 9.67; N, 4.98. Found: C, 64.00; H, 9.68; N, 4.81. IR (KBr disk): $\nu_{Si\text{-}CH3}$ 832 (s) cm^{-1}.

^{1}H NMR (400 MHz, $CDCl_3$): 0.17 (s, 6H, $(CH_3)_2Si$), 1.04 (s, 9H, $(CH_3)_3C$), 2.00 (s, 3H, 3-CH_3 of C_5N), 2.03 (s, 3H, 5-CH_3 of C_5N), 2.35 (s, 3H, 6-CH_3 of C_5N), 3.91 (s, 3H, OCH_3) ppm. $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): −3.1 (s, $(CH_3)_2Si$), 10.3 (s, 3-CH_3 of C_5N), 13.0 (s, 5-CH_3 of C_5N), 18.8 (s, *C*$(CH_3)_3$), 22.6 (s, 6-CH_3 of C_5N), 26.1 (s, C(*C*$H_3)_3$), 53.3 (s, OCH_3), 107.5, 116.0, 151.3, 160.0, 161.0 (5s, C_5N) ppm.

Preparation of 2-Methoxy-4-TBSO-3,5-dimethyl-6-chloromethylpyridine (3). While a stirred solution of pyridine derivative **2** (2.82 g, 10.0 mmol) in THF (20 mL) was cooled to 0 °C, *n*-BuLi (7.5 mL, 12.0 mmol) was dropwise added and then the mixture continued to be stirred at this temperature for 1 h to give a deep red solution. After the deep solution was slowly added to a solution of hexachloroethane (2.84 g, 12.0 mmol) in THF (10 mL) at −78 °C, the mixture was warmed to 0 °C and then was stirred at this temperature for 2 h. The resulting mixture was washed with 100 mL of saturated aqueous $NaHCO_3$ and then extracted with Et_2O (30 mL×3). The combined organic layer was dried over anhydrous $MgSO_4$. After removal of $MgSO_4$, the organic layer was evaporated at reduced pressure to give a residue. The residue was subjected to column chromatography (silica gel). Elution with ethyl acetate/hexane (v:v = 3:100) afforded **3** (2.88 g, 91%) as a yellow oil; R_f = 0.45 [TLC, SiO_2, ethyl acetate/petroleum ether (v:v = 3:100)]. Anal. Calcd for $C_{15}H_{26}ClNO_2Si$: C, 57.03; H, 8.30; N, 4.43. Found: C, 57.28; H, 8.17; N, 4.24. IR (KBr disk): $\nu_{Si\text{-}CH3}$ 830 (vs) cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$): 0.18 (s, 6H, $Si(CH_3)_2$), 1.04 (s, 9H, $C(CH_3)_3$), 2.02 (s, 3H, 3-CH_3 of C_5N), 2.18 (s, 3H, 5-CH_3 of C_5N), 3.92 (s, 3H, OCH_3), 4.58 (s, 2H, CH_2)

ppm. $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): −3.0 (s, $(CH_3)_2Si$), 10.5 (s, 3-CH_3 of C_5N), 12.2 (s, 5-CH_3 of C_5N), 18.8 (s, $C(CH_3)_3$), 26.1 (s, $C(CH_3)_3$), 46.6 (s, CH_2), 53.6 (s, OCH_3), 110.8, 118.1, 148.8, 160.5, 161.4 (5s, C_5N) ppm.

Preparation of (2-MeO-4-TBSO-3,5-Me_2-6-$COCH_2C_5N$)$Fe(CO)_3I$ (4). A mixture of pyridine derivative **3** (0.316 g, 1.0 mmol), $Na_2Fe(CO)_4 \cdot (1,4\text{-dioxane})_{1.5}$ (0.346 g, 1.0 mmol), and NaI (1.50 g, 10 mmol) in MeCN (15 mL) was stirred at room temperature for 4 h. After the resulting mixture was cooled to 0 °C, I_2 (0.254 g, 1.0 mmol) was added and then the new mixture was stirred at room temperature for 1 h. Volatiles were removed at reduced pressure, the residue was subjected to column chromatography (silica gel). Elution with CH_2Cl_2 developed a red band, from which **4** (0.316 g, 55%) was obtained as brown-red solid. Mp 103−104 °C (dec). Anal. Calcd for $C_{19}H_{26}FeINO_6Si$: C, 39.67; H, 4.56; N, 2.43. Found: C, 39.55; H, 4.68; N, 2.62. IR (KBr disk): $\nu_{C\equiv O}$ 2081 (vs), 2034 (vs), 2000 (vs); $\nu_{CH2C=O}$ 1668 (s); $\nu_{Si\text{-}CH3}$ 830 (s) cm^{-1}. ^1H NMR (400 MHz, acetone-d_6): 0.31 (s, 6H, $(CH_3)_2Si$), 1.09 (s, 9H, $(CH_3)_3C$), 2.19 (s, 3H, 3-CH_3 of C_5N), 2.29 (s, 3H, 5-CH_3 of C_5N), 4.09 (s, 3H, OCH_3), 4.28, (d, J = 20.8 Hz, 1H of CH_2CO), 4.78 (d, J = 20.8 Hz, 1H of CH_2CO) ppm. $^{13}C\{^1H\}$ NMR (100 MHz, acetone-d_6): −2.9 (s, $(CH_3)_2Si$), 12.0 (s, 3-CH_3 of C_5N), 14.3 (s, 5-CH_3 of C_5N), 19.2 (s, $C(CH_3)_3$), 26.1 (s, $C(CH_3)_3$), 61.3 (s, OCH_3), 67.3 (s, $CH_2C{=}O$), 117.4, 121.4, 158.0, 164.7, 165.7 (5s, C_5N), 200.7, 210.4, 211.2 (3s, C≡O), 257.4 (s, $CH_2C{=}O$) ppm.

Preparation of (2-MeO-4-TBSO-3,5-Me_2-6-$COCH_2C_5N$)$Fe(CO)_2$(η^2-6-Me-2-SC_5H_3N) (5). A mixture of iodide complex **4** (0.120 g, 0.20 mmol), 6-methyl-2-mercaptopyridine (0.275 g, 0.22 mmol), Et_3N (30.6 μL, 0.22 mmol), and CH_2Cl_2 (10 mL) was stirred at room temperature for 1 h. After volatiles were removed at reduced pressure, the residue was subjected to column chromatography (silica gel). Elution with CH_2Cl_2/acetone (v:v = 15:1) developed a yellow band, from which **5** (0.104 g, 91%) was obtained as a yellow solid. Mp 60–61 °C (dec). Anal. Calcd for $C_{24}H_{32}FeN_2O_5SiS$: C, 52.94; H, 5.92; N, 5.14. Found: C, 52.89; H, 6.05; N, 4.87. IR (KBr disk): $\nu_{C\equiv O}$ 2023 (vs), 1956 (vs); $\nu_{CH2C=O}$ 1646 (s); $\nu_{Si\text{-}CH3}$ 830 (s) cm^{-1}. ^1H NMR (400 MHz, acetone-d_6): 0.29 (s, 6H, $(CH_3)_2Si$), 1.07 (s, 9H, $(CH_3)_3C$), 2.11 (s, 3H, 3-CH_3 of C_5N), 2.17 (s, 3H, 5-CH_3 of C_5N), 2.58 (s, 3H, 6-CH_3 of C_5H_3N), 3.43 (s, 3H, OCH_3), 3.71 (d, J = 20.0 Hz, 1H of CH_2CO), 4.24 (d, J = 20.0 Hz, 1H of CH_2CO), 6.51 (d, J = 8.0 Hz, 1H, 5-H of C_5H_3N), 6.82 (d, J = 7.6 Hz, 1H, 3-H of C_5H_3N), 7.30 (t, J = 7.8 Hz, 1H, 4-H of C_5H_3N) ppm. $^{13}C\{^1H\}$ NMR (100 MHz, acetone-d_6): –4.2 ($(CH_3)_2Si$), 10.7 (3-CH_3 of C_5N), 12.4 (5-CH_3 of C_5N), 17.9 ($C(CH_3)_3$), 21.5 (6-CH_3 of C_5H_3N), 24.8 ($C(CH_3)_3$), 59.9 (OCH_3), 61.3 (CH_2C=O), 114.9–175.2 (C_5H_3N, C_5N), 210.8, 213.5 (2s, C≡O), 263.1 (CH_2C=O) ppm.

Preparation of (2-MeO-4-HO-3,5-Me_2-6-$COCH_2C_5N$)$Fe(CO)_2$(η^2-6-Me-2-SC_5H_3N) (6). A mixture of complex **5** (0.280 g, 0.5 mmol), n-Bu_4NF (0.6 mL, 0.6 mmol), acetic acid (34.3 μL,

0.6 mmol), and THF (25 mL), was stirred at room temperature for 1 h. After volatiles were removed at reduced pressure, the residue was subjected to column chromatography (silica gel). Elution with CH_2Cl_2/acetone (v:v = 5:1) developed a yellow band, from which **6** (0.157 g, 73%) was obtained as a yellow solid. Mp 122−123 °C (dec). Anal. Calcd for $C_{18}H_{18}FeN_2O_5S·H_2O$: C, 48.23; H, 4.50; N, 6.25. Found: C, 48.43; H, 4.66; N, 6.05. IR (KBr disk): ν_{OH} 3737 (m); $\nu_{C\equiv O}$ 2026 (vs), 1959 (vs); $\nu_{CH2C=O}$ 1633 (s) cm^{-1}. ^1H NMR (400 MHz, acetone-d_6): 2.13 (s, 3H, 3-CH_3 of C_5N), 2.18 (s, 3H, 5-CH_3 of C_5N), 2.57 (s, 3H, 6-CH_3 of C_5H_3N), 3.42 (s, 3H, OCH_3), 3.71 (d, J = 19.6 Hz, 1H of CH_2CO), 4.23 (d, J = 19.6 Hz, 1H of CH_2CO), 6.50 (d, J = 8.0 Hz, 1H, 5-H of C_5H_3N), 6.81 (d, J = 7.6 Hz, 1H, 3-H of C_5H_3N), 7.29 (t, J = 7.6 Hz, 1H, 4-H of C_5H_3N), 9.19 (s, 1H, OH) ppm. $^{13}C\{^1H\}$ NMR (100 MHz, acetone-d_6): 10.4 (s, 3-CH_3 of C_5N), 12.2 (s, 5-CH_3 of C_5N), 22.9 (s, 6-CH_3 of C_5H_3N), 61.4 (s, OCH_3), 62.5 (s, *C*H_2C=O), 111.4−176.7 (m, C_5H_3N, C_5N), 212.3, 214.9 (2s, C≡O), 264.7 (s, CH_2*C*=O) ppm.

Preparation of (2-MeO-3,5-Me_2-4-OP=O$(OPh)_2$-6-$COCH_2C_5N$)Fe$(CO)_2$(η^2-6-Me-2-SC_5H_3N) (7). A stirred solution of complex **6** (0.086 g, 0.20 mmol) in CH_2Cl_2 (10 mL) was cooled to −40 °C, and then Et_3N (34 μL, 0.24 mmol) and O=P$(PhO)_2$Cl (50 μL, 0.24 mmol) were dropwise added. After the resulting mixture was warmed to room temperature, it was continuously stirred at this temperature for 4 h. Volatiles were removed at reduced pressure and the residue was subjected to column chromatography (silica gel).

Elution with CH_2Cl_2/acetone (v:v = 20:1) developed a yellow band, from which **7** (0.120 g, 92%) was obtained as an orange solid. Mp 93–94 °C (dec). Anal. Calcd for $C_{30}H_{27}FeN_2O_8PS$: C, 54.40; H, 4.11; N, 4.23. Found: C, 54.71; H, 4.29; N, 4.16. IR (KBr disk): $\nu_{C\equiv O}$ 2018 (vs), 1957 (vs); $\nu_{CH2C=O}$ 1648 (s); $\nu_{P=O}$ 1301 (s) cm^{-1}. ^1H NMR (400 MHz, acetone-d_6): 2.21 (s, 3H, 3-CH_3 of C_5N), 2.28 (s, 3H, 5-CH_3 of C_5N), 2.58 (s, 3H, 6-CH_3 of C_5H_3N), 3.48 (s, 3H, OCH_3), 3.82 (d, J = 20.0 Hz, 1H of CH_2CO), 4.33 (d, J = 20.0 Hz, 1H of CH_2CO), 6.54 (d, J = 8.0 Hz, 1H, 5-H of C_5H_3N), 6.83 (d, J = 7.6 Hz, 1H, 3-H of C_5H_3N), 7.28–7.46 (m, 11H, 4-H of C_5H_3N, $2C_6H_5$) ppm. $^{13}C\{^1H\}$ NMR (100 MHz, acetone-d_6): 11.6 (s, 3-CH_3 of C_5N), 13.4 (s, 5-CH_3 of C_5N), 22.9 (s, 6-CH_3 of C_5H_3N), 61.9 (s, OCH_3), 62.7 (s, CH_2C=O), 118.0–176.4 (m, C_5H_3N, C_5N, C_6H_5), 211.8, 214.7 (2s, C≡O), 263.4 (s, CH_2C=O) ppm. $^{31}P\{^1H\}$ NMR (162 MHz, acetone-d_6, 85% H_3PO_4): −18.7 (s, P=O) ppm

2. X-ray crystal structure determinations of 4 and 7.

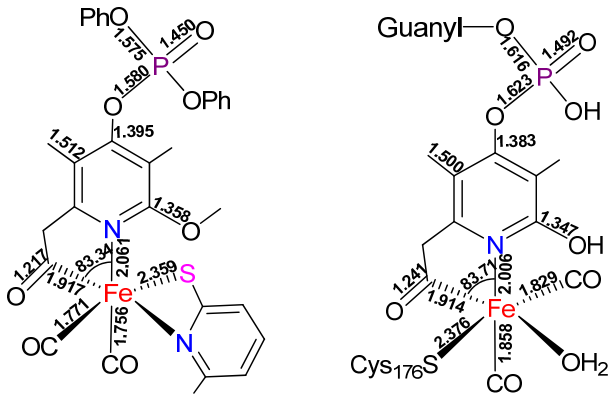
Single crystals of **4** suitable for X-ray diffraction analysis were grown by slow diffusion of hexane into its CH_2Cl_2 solution at −10 °C, while those of **7** by slow diffusion of hexane into its acetone solution at −20 °C, respectively. A single crystal of **4** was mounted on a SuperNova diffractometer equipped with an AtlasS2 accessory, and data were collected using a confocal monochromator with Cu Kα radiation (λ = 1.54184 Å) in the ω scanning mode at 100 K. A single crystal of **7** was mounted on a Rigaku MM-007 (rotating anode) diffractometer equipped with a CCD plate, and data were collected using a confocal monochromator with Mo Kα radiation (λ = 0.71073 Å) in the ω-φ scanning mode at 113 K. Data collection, reduction, and absorption correction were performed by the CRYSTALCLEAR program[4], The structures were solved by direct methods using the SHELXS program[5] and refined by Olex2 program[5c] with full-matrix least-squares techniques (SHELXL)[6] on F^2. Hydrogen atoms were located by using the geometric method. Details of crystal data, data collections, and structure refinements are summarized in Table S1.

Table S1. Crystal data and structure refinement details for 4 and 7

	4	**7**
mol formula	$C_{19}H_{26}FeINO_6Si$	$C_{30}H_{27}FeN_2O_8PS$ $\cdot C_3H_6O$
mol wt	575.25	720.49
cryst syst	triclinic	triclinic
space group	P-1	P-1
a /Å	6.7011(2)	9.2203(18)
b /Å	13.6909(7)	9.4569(19)
c /Å	13.7031(7)	20.857(4)
α /deg	104.516(5)	85.51(3)
β /deg	101.534(4)	89.66(3)
γ /deg	92.720(3)	67.10(3)
V/Å^3	1186.15(10)	1669.6(7)
Z	2	2
D_c/g·cm^{-3}	1.611	1.433
abs coeff/mm^{-1}	16.084	0.619
F(000)	576.0	748.0
index ranges	−8≤h≤5	−12≤h≤12
	−16≤k≤17	−12≤k≤12
	−15≤l≤17	−27≤l≤27

no. of reflns	9093	20145
no. of indep reflns	4648	7919
$2\theta_{max}$ /deg	148.156	55.696
R	0.0416	0.0409
R_w	0.1030	0.1105
goodness of fit	1.051	0.978
largest diff peak, hole/e Å^{-3}	2.99/–1.15	0.50/–0.48

3. Comparison of some bond lengths (Å) and angles (deg) between target model 7 and the active site of [Fe]-H_2ase [7]



Scheme S1. Comparison of some bond lengths (Å) and angles (deg) between target model **7** (left) and the active site of [Fe]-H_2ase (right)

4. IR and NMR spectra of 1–7 (Figures S1–S22)

(1) IR and ^{1}H (^{13}C) NMR spectra of **1**

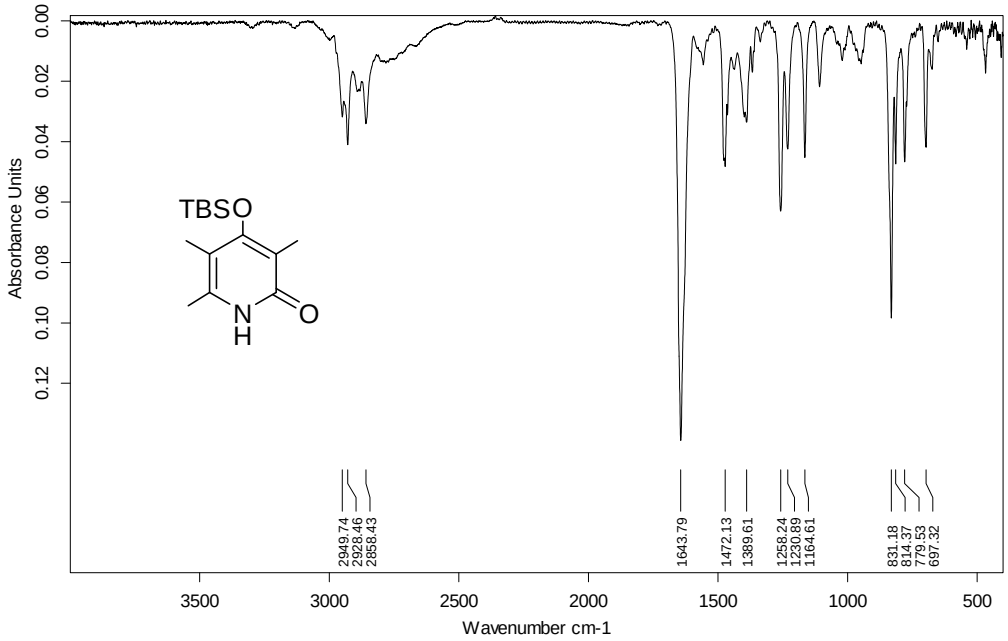


Figure S1. IR spectrum of **1**

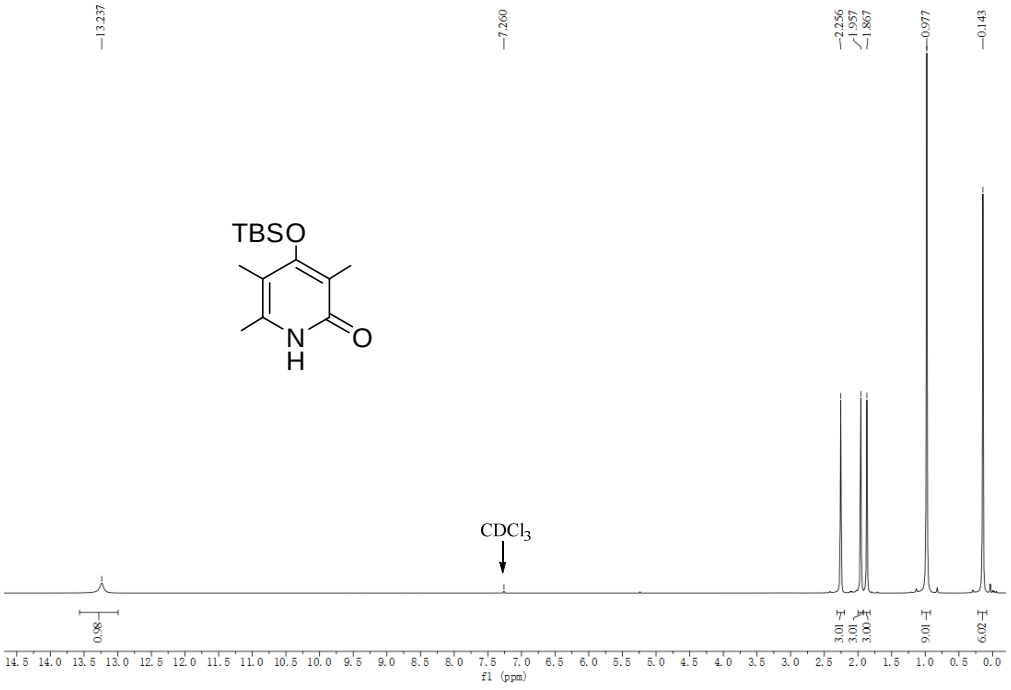


Figure S2. ^{1}H NMR spectrum of **1**

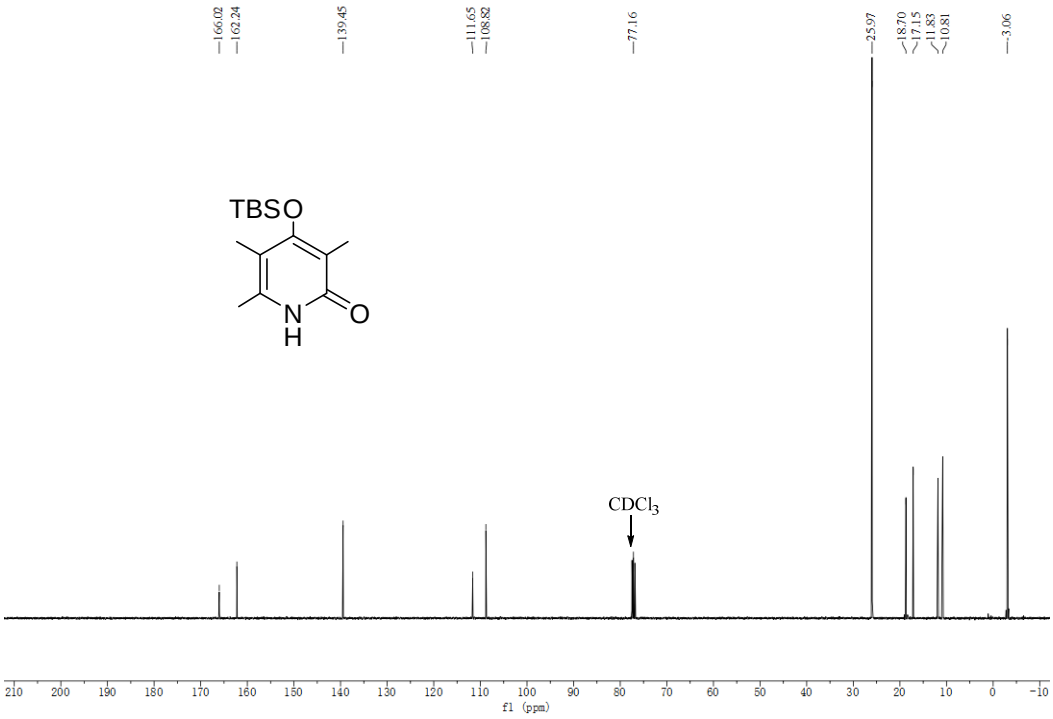


Figure S3. ^{13}C NMR spectrum of **1**

2. IR and ^{1}H (^{13}C) NMR spectra of **2**

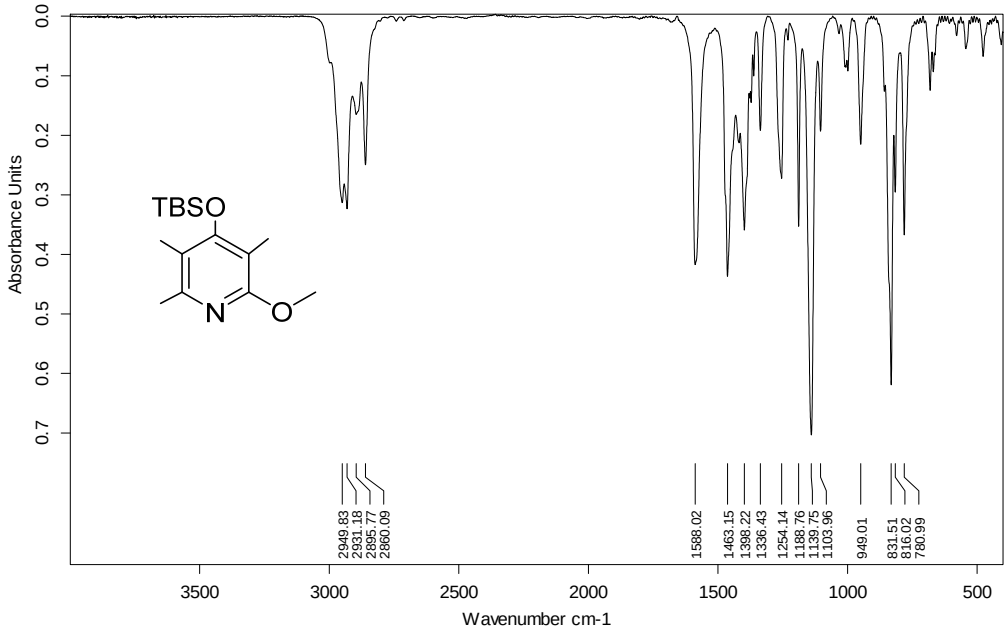


Figure S4. IR spectrum of **2**

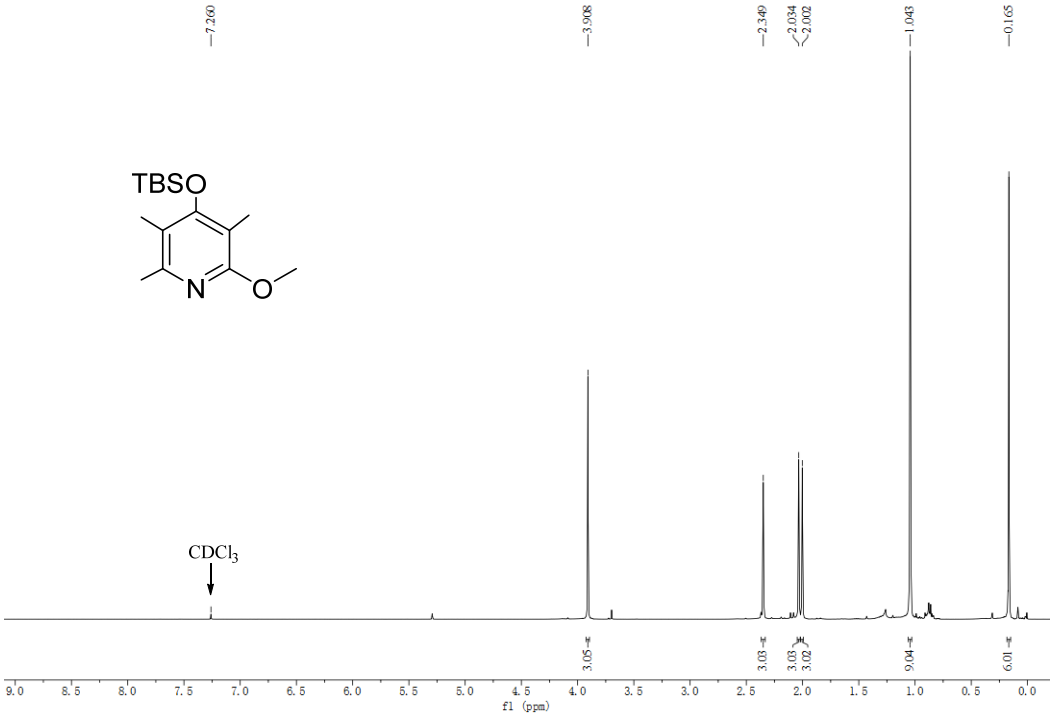


Figure S5. ^{1}H NMR spectrum of **2**

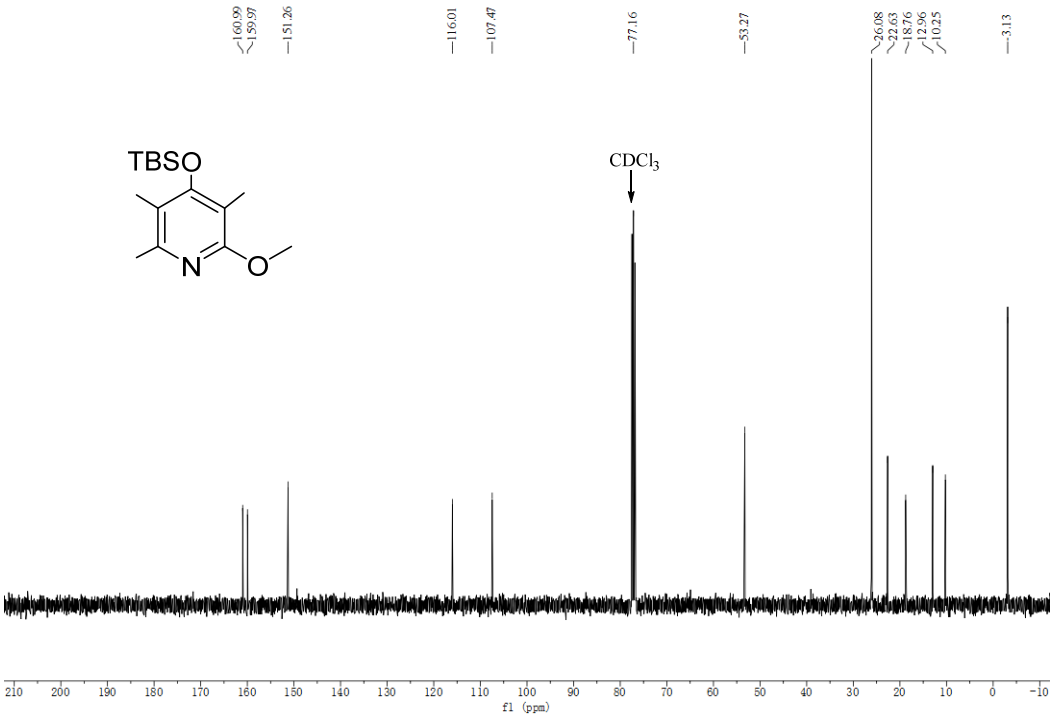


Figure S6. ^{13}C NMR spectrum of **2**

3. IR and ^{1}H (^{13}C) NMR spectra of **3**

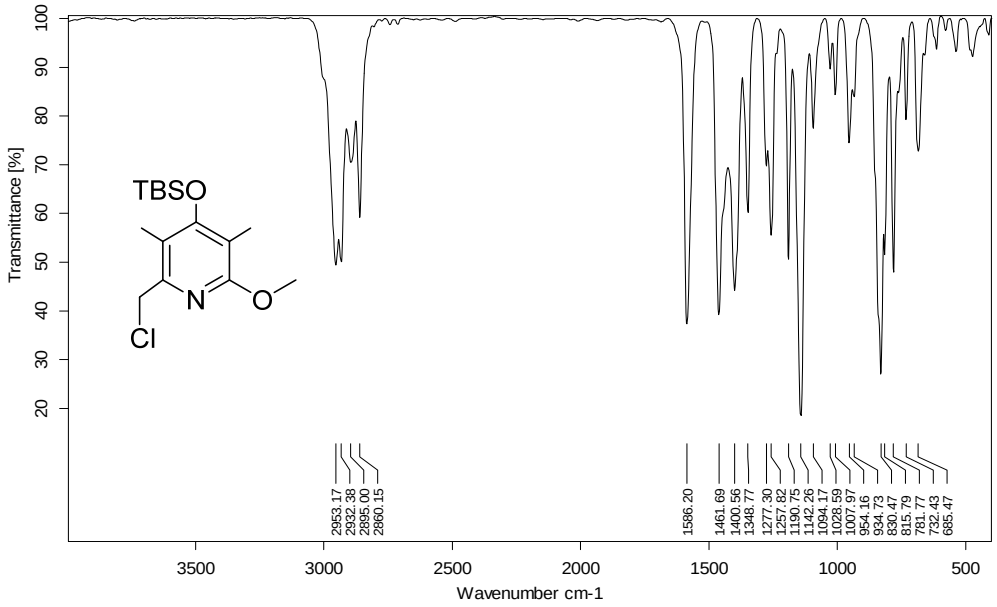


Figure S7. IR spectrum of **3**

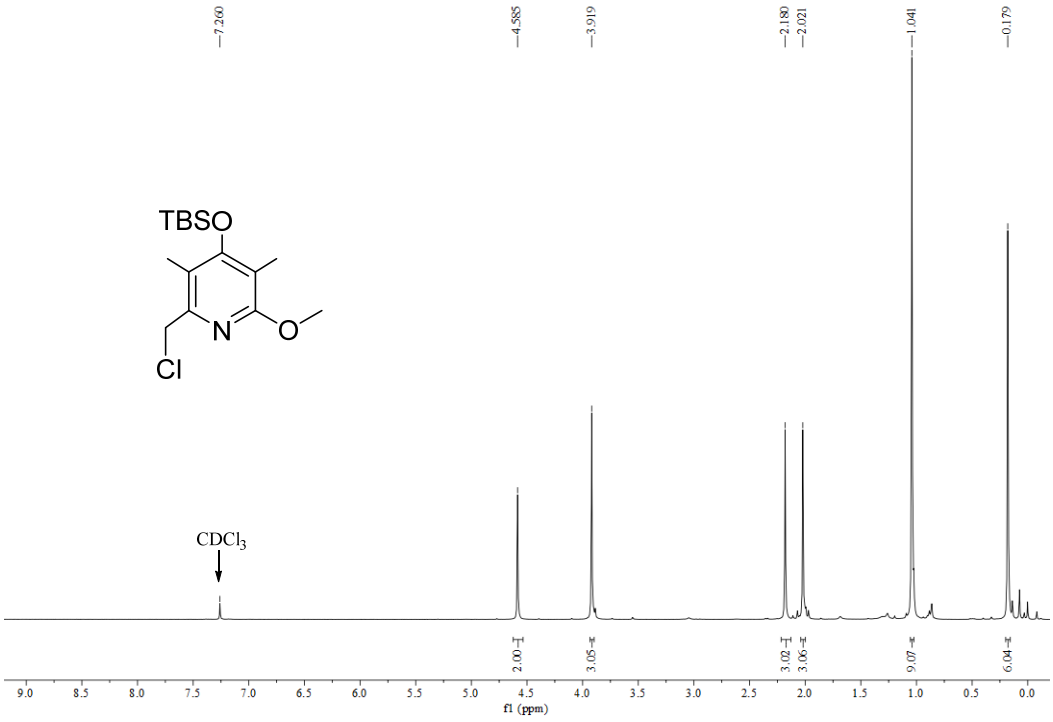


Figure S8. ^{1}H NMR spectrum of **3**

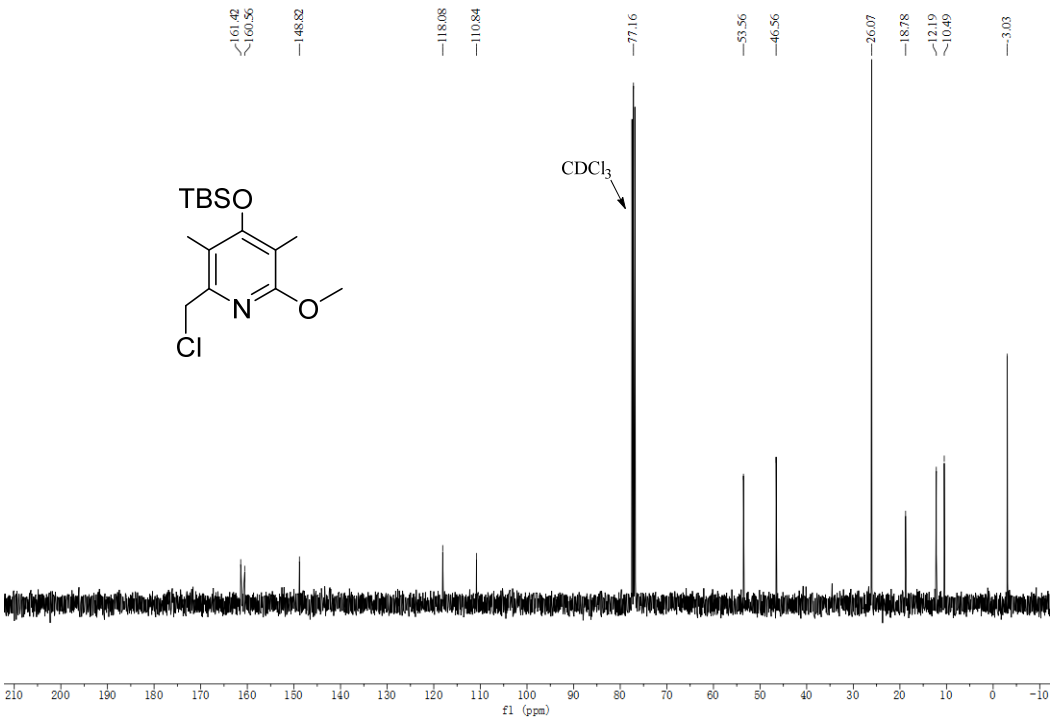


Figure S9. ^{13}C NMR spectrum of **3**

4. IR and ^{1}H (^{13}C) NMR spectra of **4**

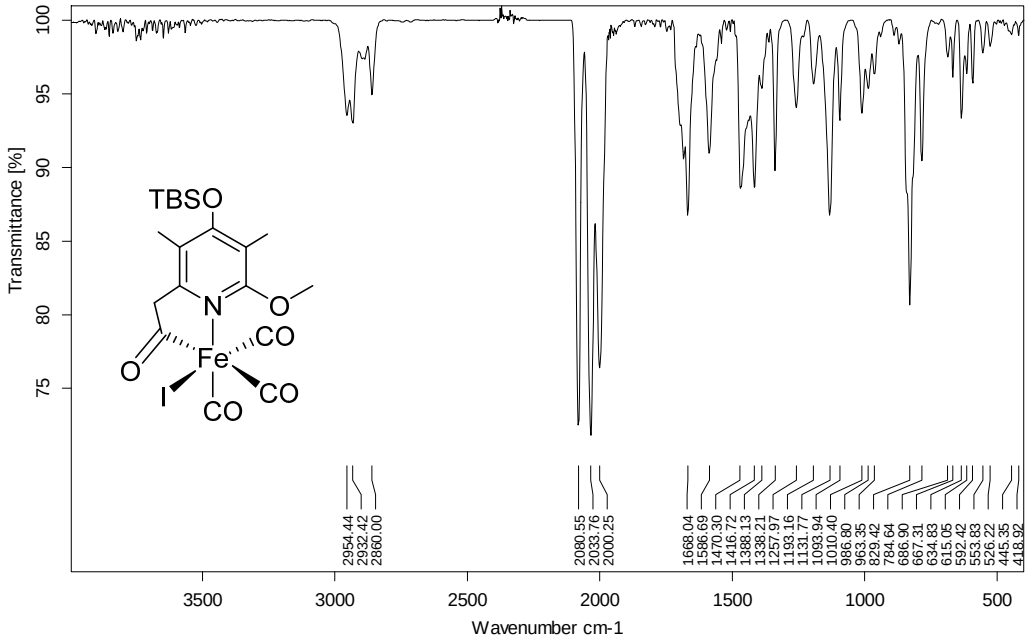


Figure S10. IR spectrum of **4**

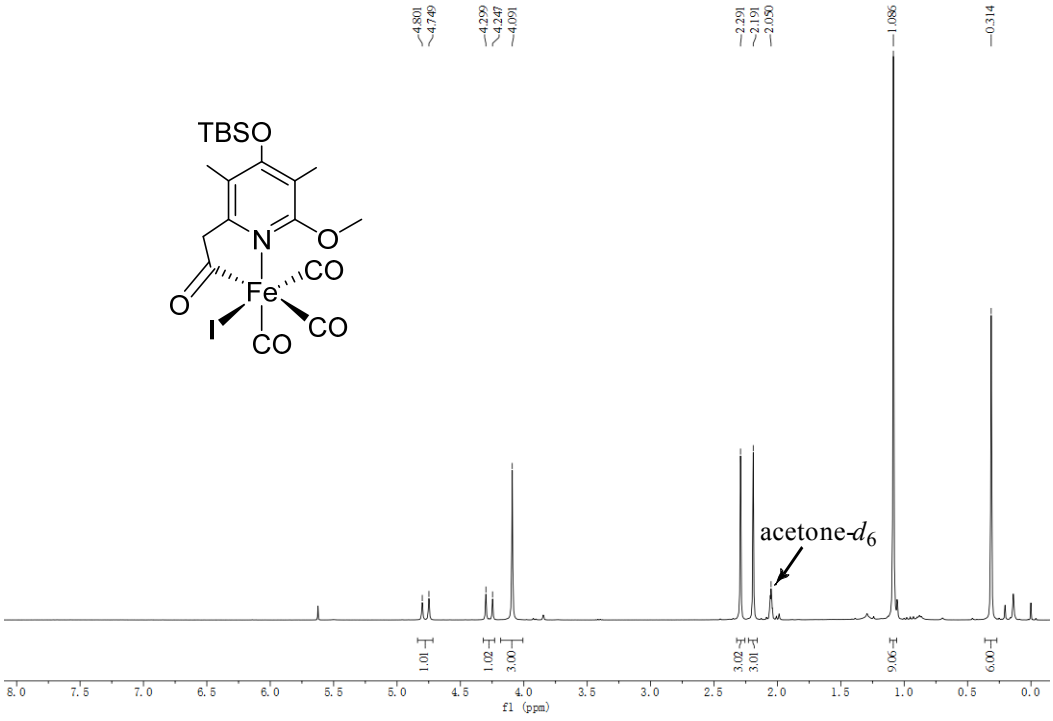


Figure S11. ^{1}H NMR spectrum of **4**

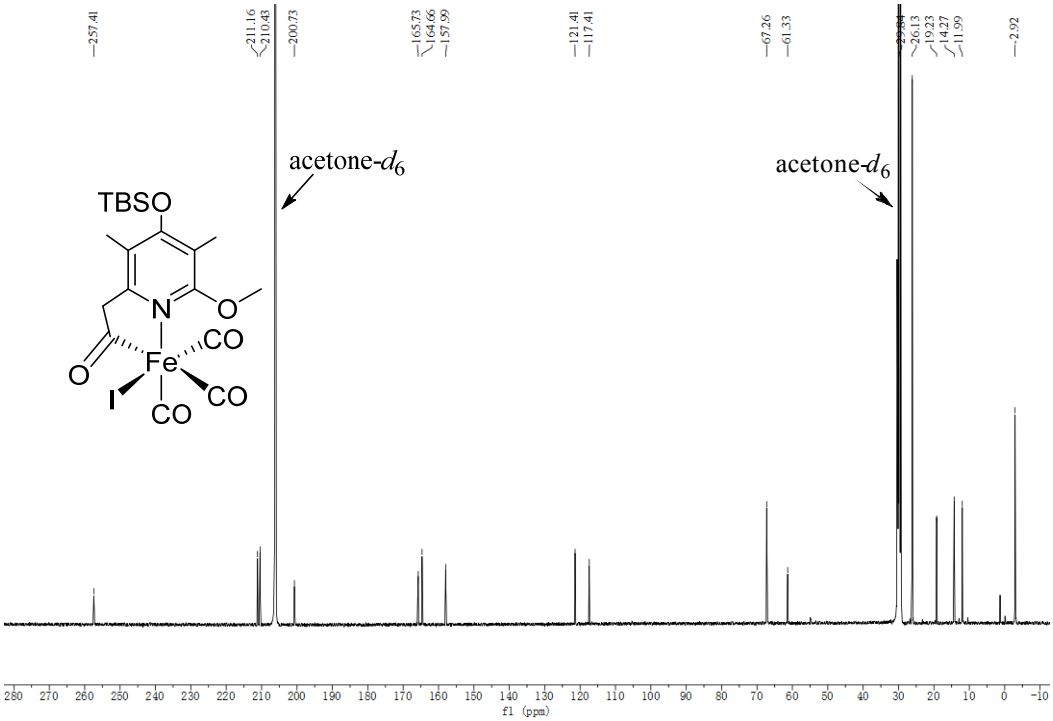


Figure S12. ^{13}C NMR spectrum of **4**

5. IR and ^{1}H (^{13}C) NMR spectra of **5**

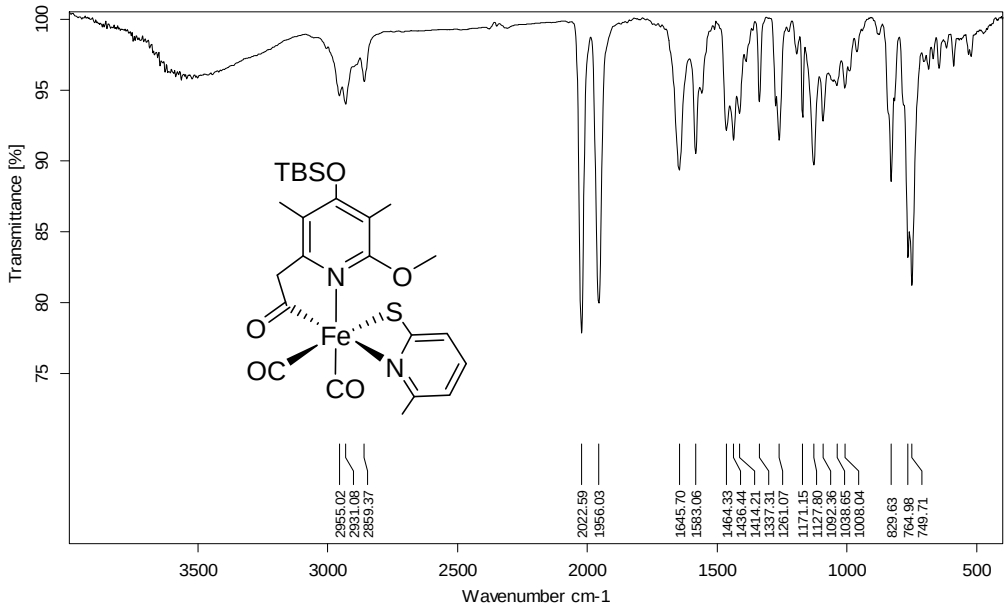


Figure S13. IR spectrum of **5**

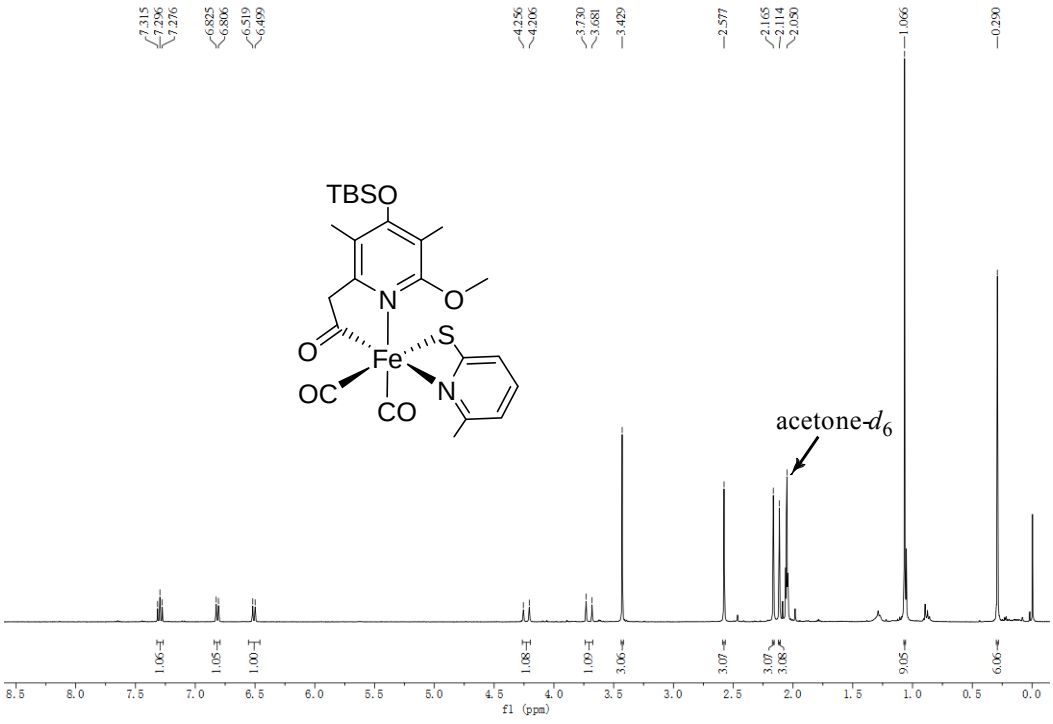


Figure S14. ^{1}H NMR spectrum of **5**

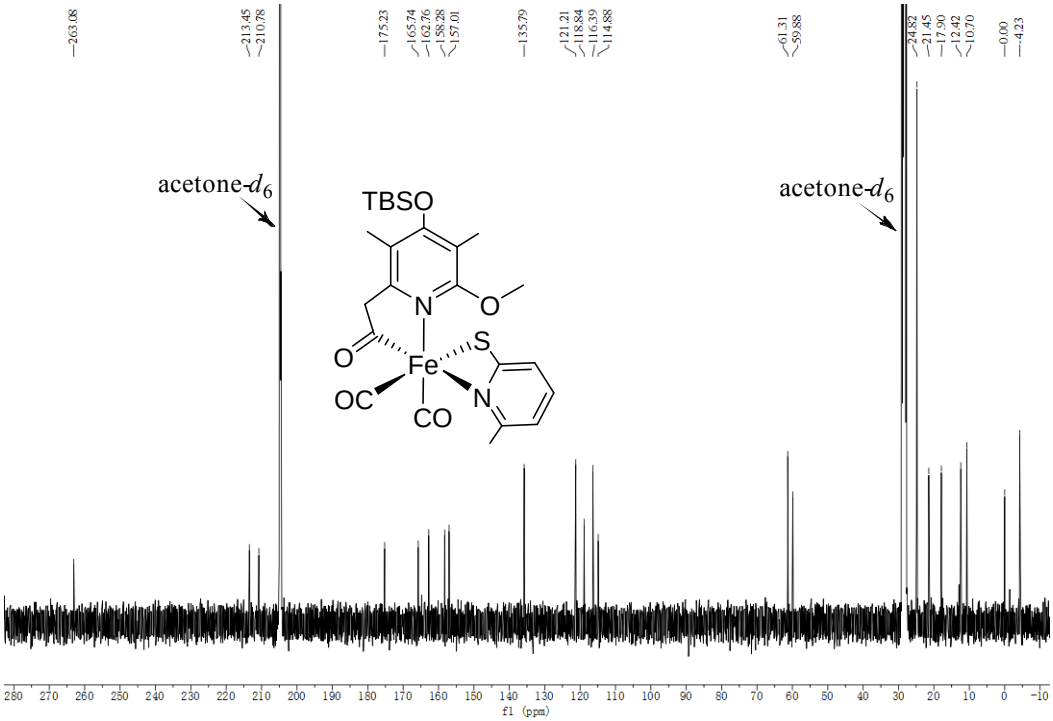


Figure S15. ^{13}C NMR spectrum of **5**

6. IR and ^{1}H (^{13}C) NMR spectra of **6**

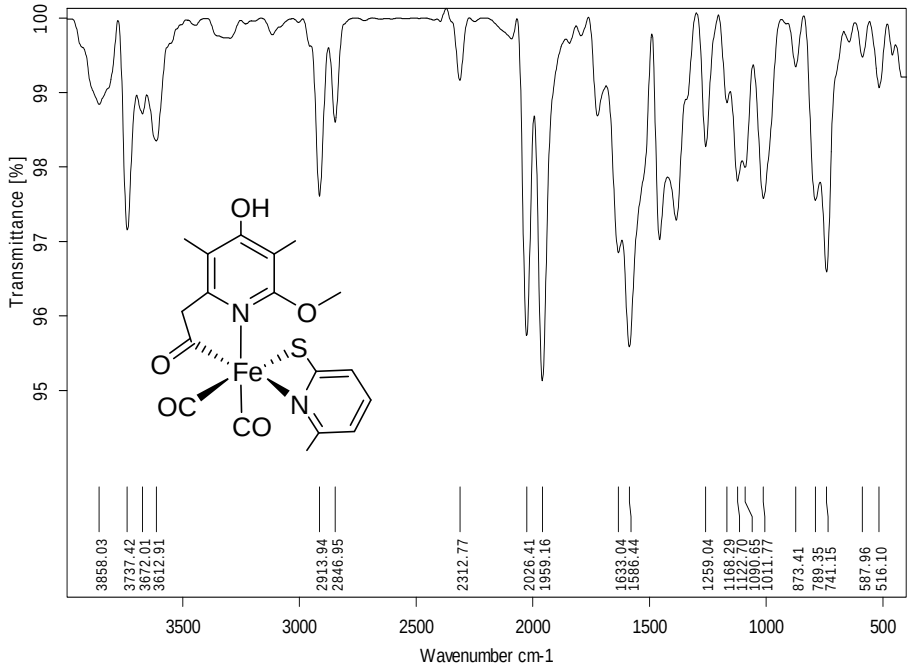


Figure S16. IR spectrum of **6**

Figure S17. ^{1}H NMR spectrum of **6**

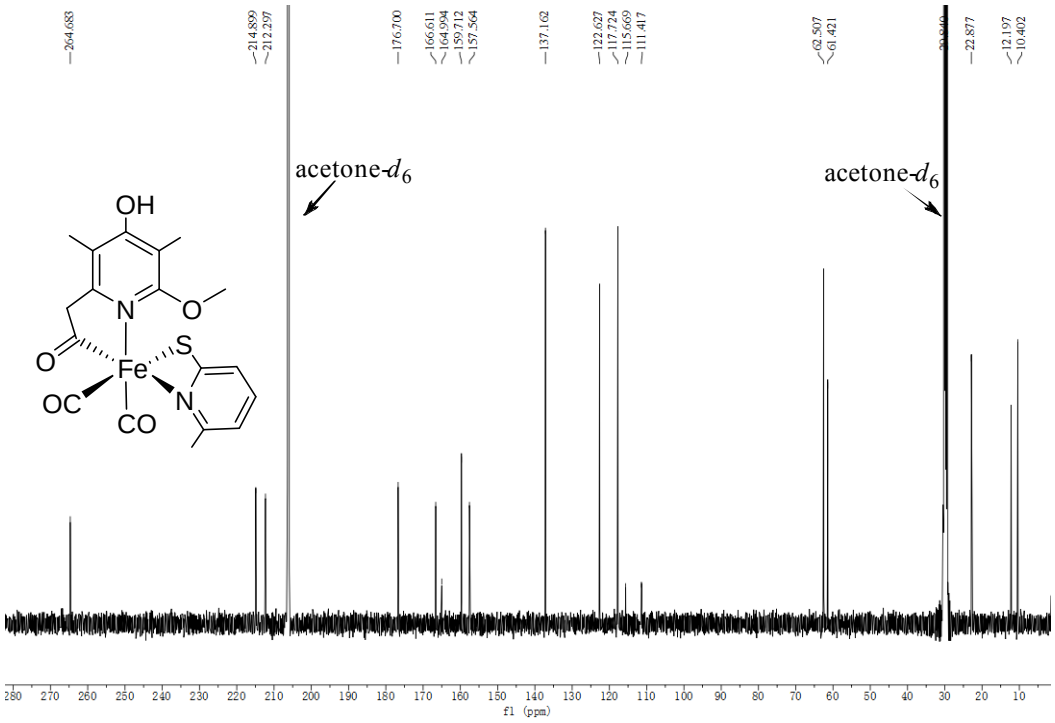


Figure S18. ^{13}C NMR spectrum of **6**

7. IR and ^{1}H (^{13}C, ^{31}P) NMR spectra of 7

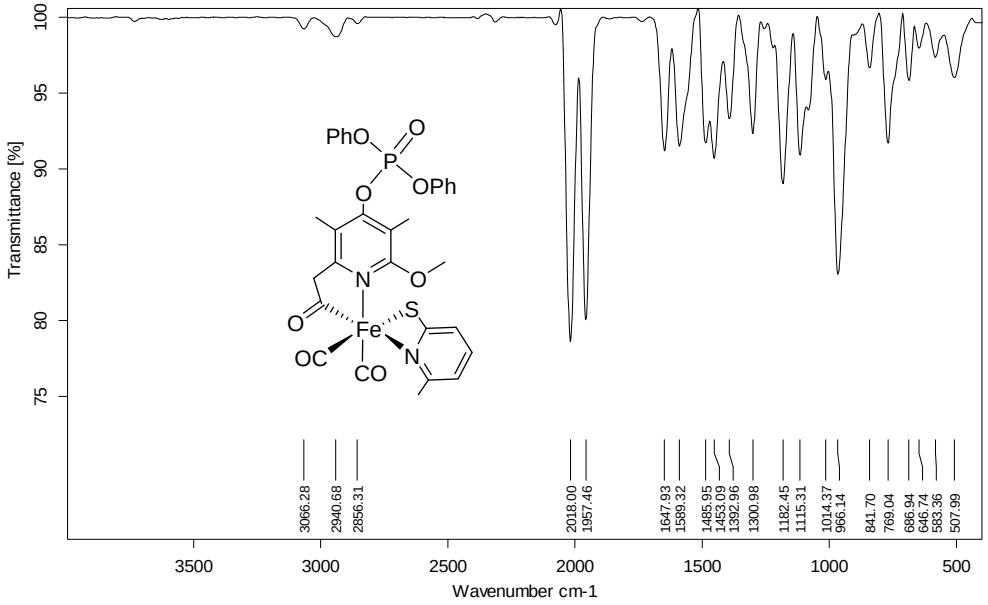


Figure S19. IR spectrum of **7**

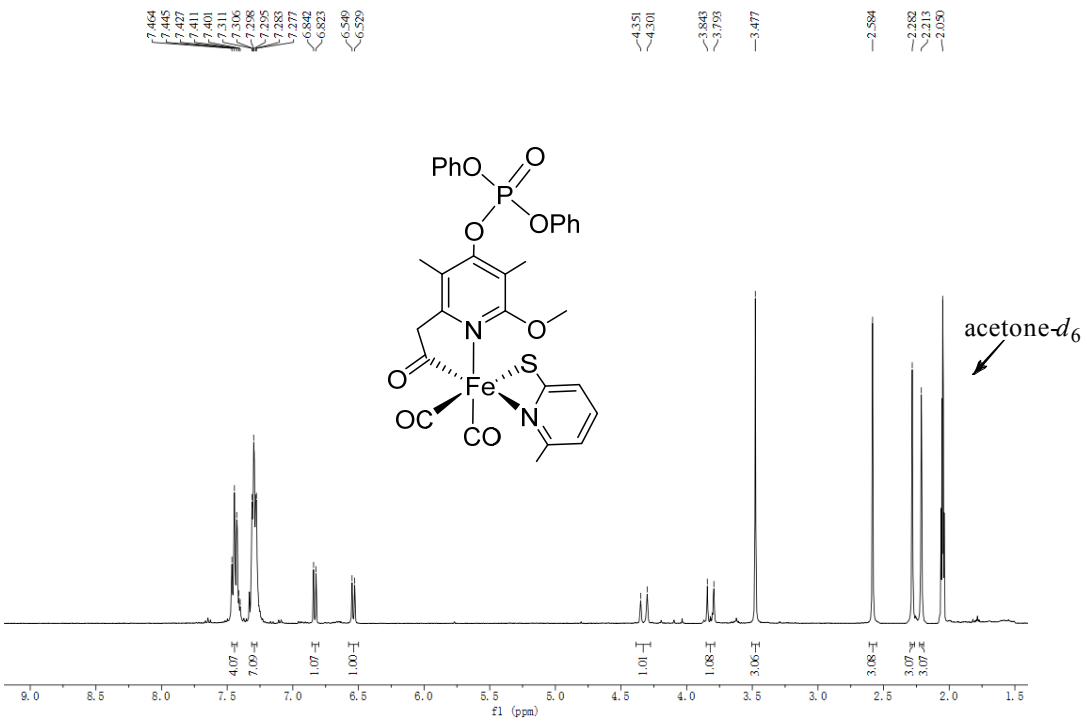


Figure S20. ^{1}H NMR spectrum of **7**

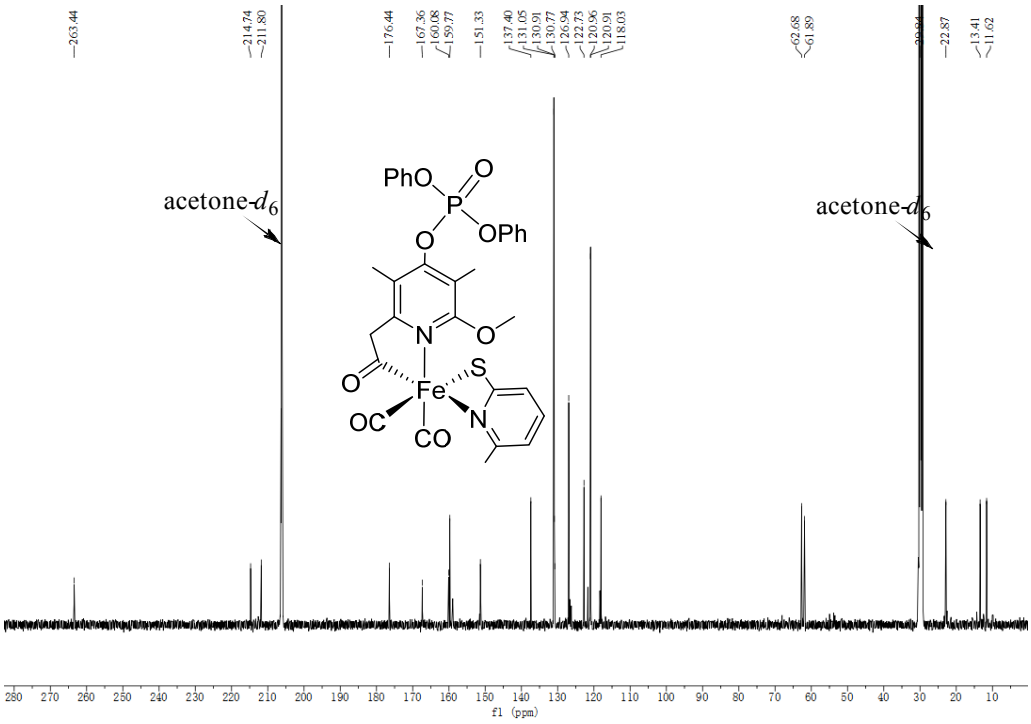


Figure S21. ^{13}C NMR spectrum of **7**

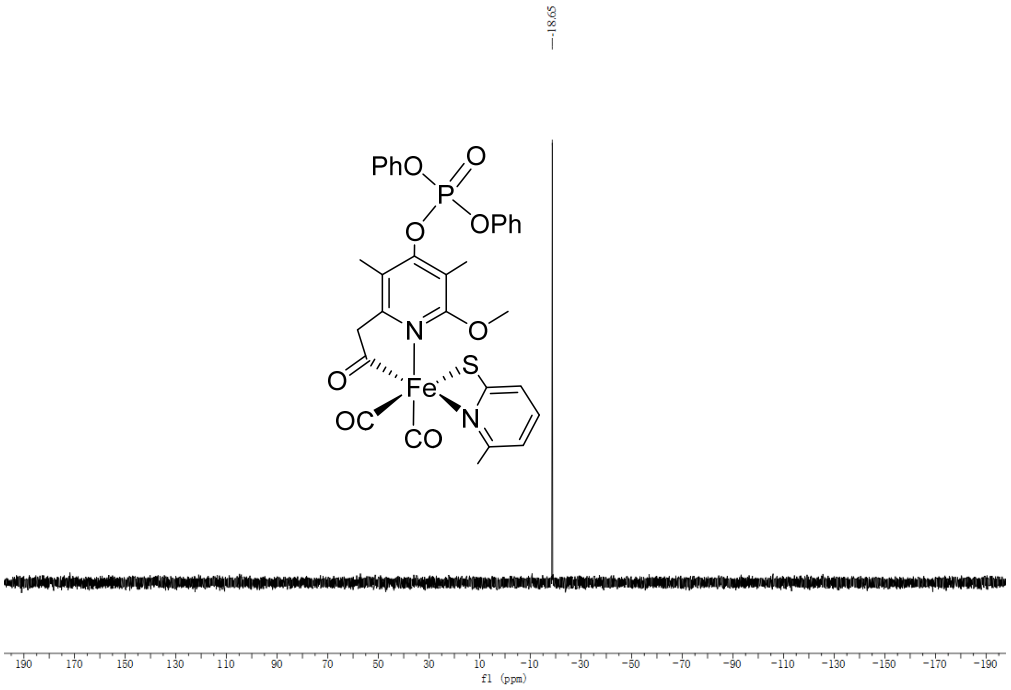


Figure S22. ^{31}P NMR spectrum of **7**

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