The Iron Centre of the Cluster-free Hydrogenase (Hmd): Low Spin Fe(II) or Low Spin Fe(0)?

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1. Preparations of the ligand 2,2'-(Pyridin-2-ylmethylazanediyl)diethanethiol, H_2L [1]

A mixture of 2-(aminomethyl)pyridine (0.3mL, 3mmol) and ethylene sulfide (0.5mL, 8.4mmol) in toluene (3ml) sealed in a Schlenk tube under N_2 was heated at 100°C on stirring for 40 hours. The solution was concentrated to about 0.5mL under vacuum and purified by flash chromatography (ethyl acetate) to produce a stinky colorless oily liquid (0.62g, 90%). IR (neat): SH 2541cm^{-1}; MS(ES+): [M+1]/z = 229, [M-C_2H_4SH+2H]/z = 169, [M-2(C_2H_4SH)+3H]/z =109; ^{1}HNMR($CDCl_3$)：1.695(2H, s, 2S*H*), 2.675(4H, q, 2CH_2SH, J = 10.62Hz,), 2.779(4H, t, 2NCH_2, J = 6.8Hz), 3.796(2H, s, PyCH_2), 7.183(1H, t, J = 6.13 Hz, Py), 7.530(1H, d, J = 7.75 Hz, Py), 7.685(1H, t, J = 7.6 Hz, Py), 8.529(1H, d, J=4.55 Hz, Py); ^{13}C NMR ($CDCl_3$): 23.122, 57.600, 60.530, 122.611, 123.551, 136.946, 149.428, 159.581.

2 Reaction of the ligand with $FeSO_4 \cdot 7H_2O$ [2]

To a suspension of $FeSO_4 \cdot 7H_2O$ (139.1mg) in MeOH (1mL) at ice temperature was dropwise added a solution (1.5mL) of 2,2'-(pyridin-2-ylmethylazanediyl)-diethanethiol (114mg, 0.5mmol) and Et_3N (0.14mL, 1mmol) in MeOH on stirring under CO atmosphere. In 10minutes, the reaction turned to dark red and the reaction was filtered. The filtrate was stored in a freezer at -25°C and dark red crystal blocks were collected in two days. Infrared absorption bands of the complex in different organic solvents are as follows: MeOH, 2025.0, 1971.0cm^{-1}; Ethanol, 2024.9, 1971.5; DCM, 2021.7, 1966.7cm^{-1}; $CDCl_3$, 2024.7, 1974.8cm^{-1}. ^{1}H NMR ($CDCl_3$): 8.943 (d, *H*-3-Py, J = 4.48Hz), 7.711 (t, *H*-4-Py, J = 6.92Hz), 7.327 (t, ill-resolved, *H*-5-Py),

7.211 (d, *H*-6-Py, J = 7.28Hz), 4.398 (s, C*H*$_2$-2-Py), 3.522 (d, 2C*H*$_2$, J = 5.60Hz), 2.765 (d, C*H*$_2$, J = 11.88Hz), 2.572 (t, ill-resolved, C*H*$_2$). Microanalysis for $C_{12}H_{14}N_2O_2S_2Fe$ (338.32), cal. (found): C%, 42.61 (42.52), H%, 4.17 (4.20), N%, 8.28 (8.03).

3. Mössbauer spectroscopic diagrams

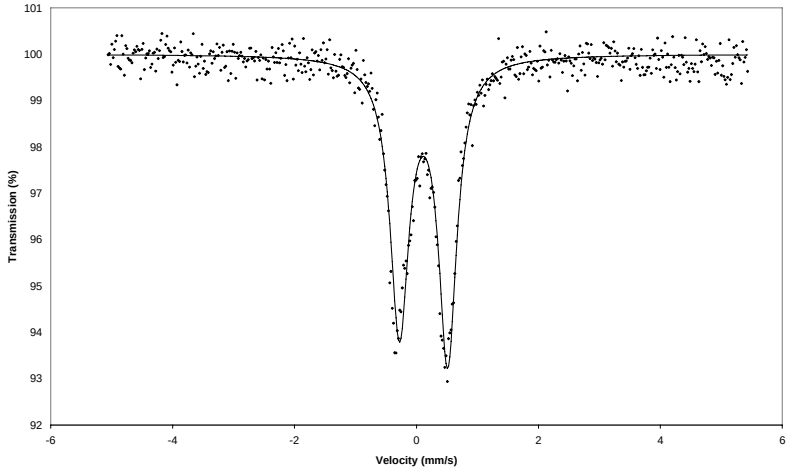

Fig. S1a At 80 K (anaerobic mounting, small holder)

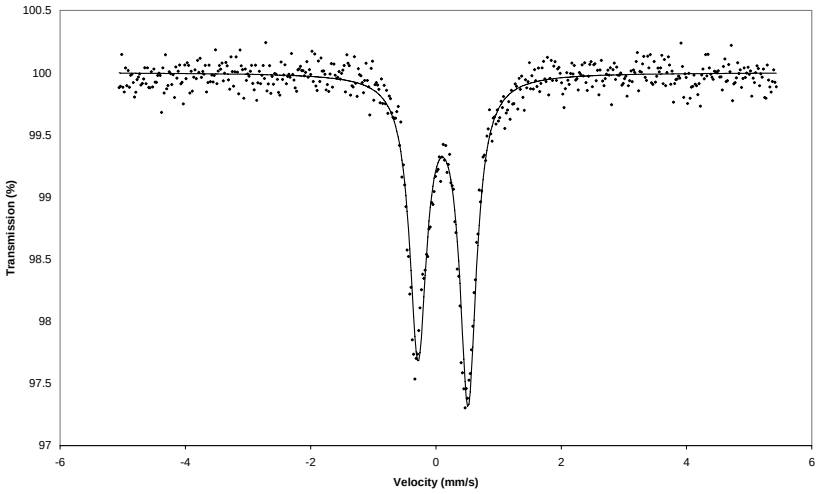

Fig. S1 3b At 80 K (aerobic mounting, ground with boron nitride, large holder, stored overnight before data collection)

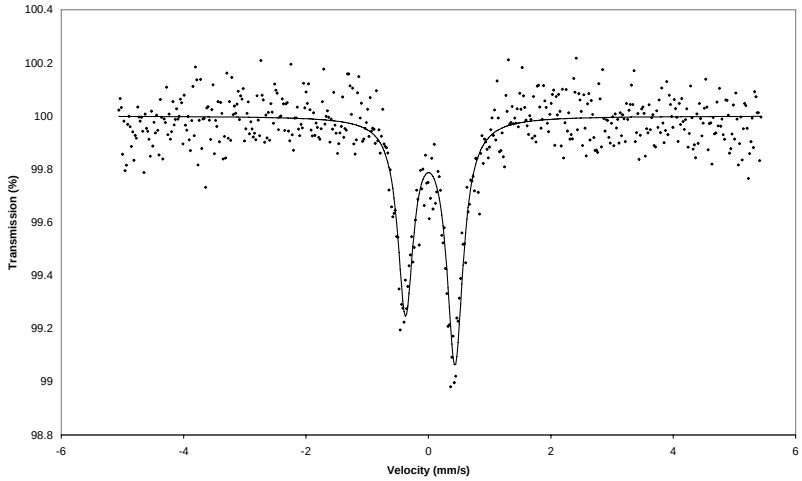

Fig. S1c At 293 K (aerobic mounting, ground with boron nitride).

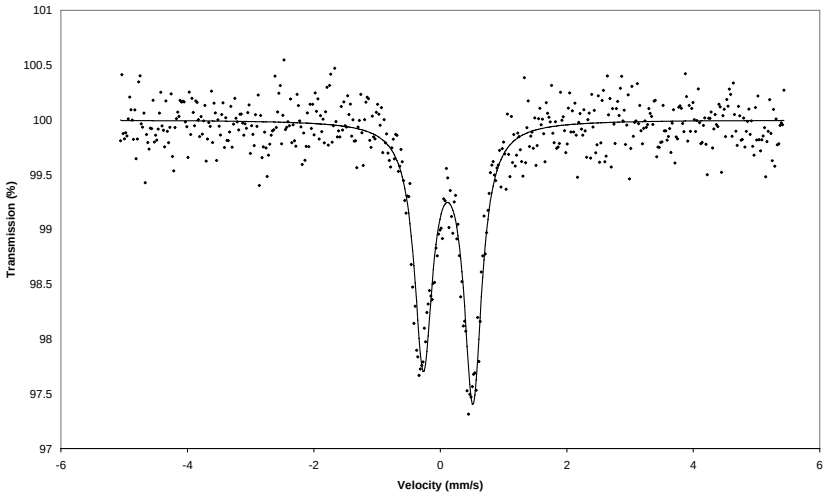

Fig. S1d At 80 K repeated after recording spectrum at 293 K (aerobic mounting, ground with boron nitride).

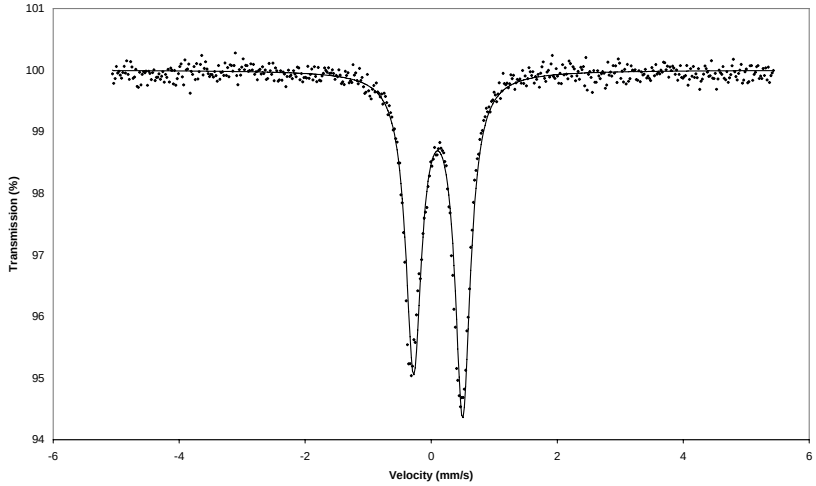

Fig. S1e At 80 K (anaerobic mounting, ground with boron nitride, large holder)

PARAMETERS (mm s^{-1}) All at 80 K except DAV956 at 293 K

	i.s.	**q.s.**	**h.w.h.m.**
Fig. S1a	0.10	0.79	0.19
Fig. S1b	0.10	0.80	0.17
Fig. S1c	0.01	0.81	0.17
Fig. S1d	0.10	0.79	0.18
Fig. S1e	0.10	0.78	0.15

Errors ≤ ± 0.01 mm s^{-1}.
Source ^{57}Co, 89 MBq.

4. Electrochemistry of the complex, [Fe(*cis*-CO)$_2$L]

The electrochemistry was carried out in 0.5M [NBu_4][BF_4]/methanol under CO atmosphere at ice temperature in the dark. We use vitreous carbon disk as working electrode, Pt rod as counter electrode, Ag/AgCl in (0.45M [NBu_4][BF_4] + 0.05M [NBu_4]Cl)/dichloromethane as reference electrode.

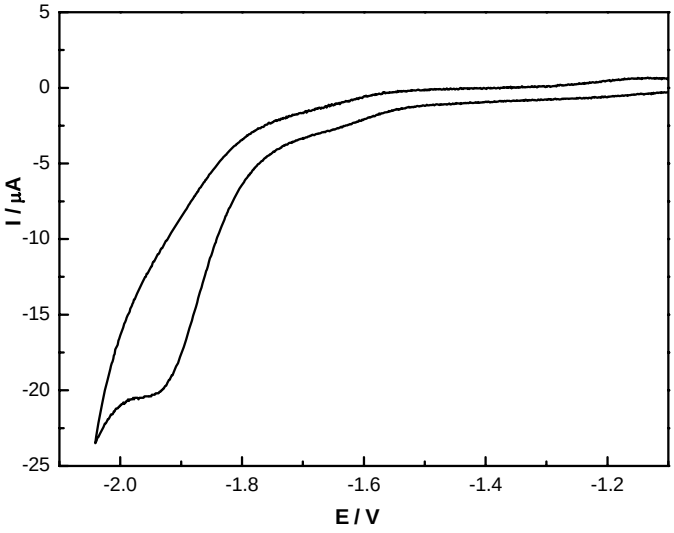

Figure S2 Cyclic voltammetry of the model complex in methanol (The potential is quoted against FC^{+}/FC couple).

5. UV/Vis spectrum in methanol

Spectrum was recorded in a 1.0 cm quartz cell in methanol. The concentration was 5.32×10^{-5}mmol/mL

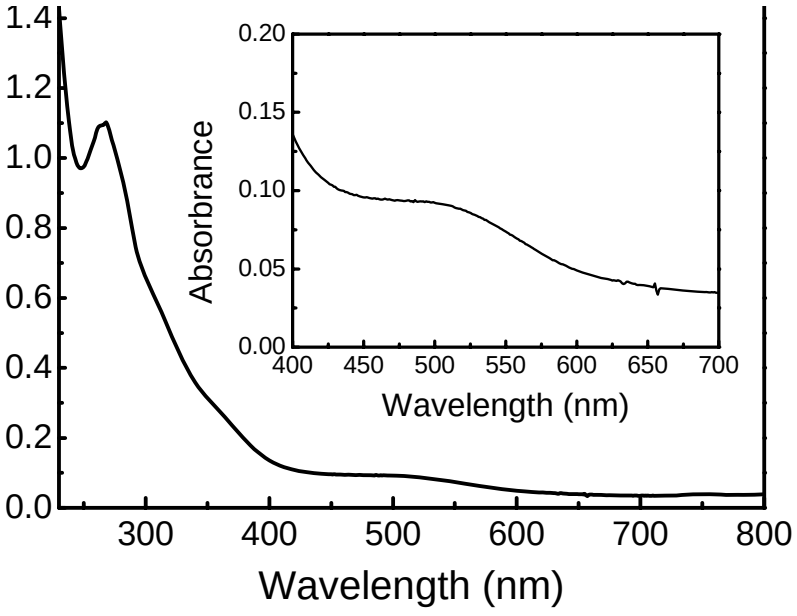

Figure S3 UV/Vis spectrum of the complex, [Fe(*cis*-$CO)_2$**L**] in methanol.

6. Bond lengths, angles, crystal data and structure refinement for $C_{12}H_{14}FeN_2O_2S_2$

Table S1. Crystal data and structure refinement for 070912C

Identification code	070912C
Empirical formula	$C_{12}H_{14}FeN_2O_2S_2$
Formula weight	338.22
Temperature	293(2) K
Wavelength	0.71073 Å
Crystal system, space group	Triclinic, P-1
Unit cell dimensions	a = 7.1525(11) Å α = 91.659(2)°
	b = 7.6915(12) Å β = 95.127(2)°
	c = 13.464(2) Å γ = 109.7330(10)°
Volume	693.05(18) $Å^3$
Z, Calculated density	2, 1.621 Mg/m^3
Absorption coefficient	1.387 mm^{-1}
F(000)	348
Crystal size	0.30 x 0.24 x 0.10 mm
Theta range for data collection	2.82 to 24.99°
Limiting indices	-8<=h<=8, -8<=k<=9, -15<=l<=15
Reflections collected / unique	4118 / 2205 [R(int) = 0.0232]
Completeness to theta = 24.99	90.4 %
Max. and min. transmission	0.8738 and 0.6811
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	2205 / 0 / 172
Goodness-of-fit on F^2	1.010
Final R indices [I>2sigma(I)]	R1 = 0.0446, wR2 = 0.1207
R indices (all data)	R1 = 0.0588, wR2 = 0.1353
Largest diff. peak and hole	0.422 and -0.537 e. $Å^{-3}$

Table S2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters ($Å^2$ x 10^3) for 070912c. U(eq) is defined as one third of the trace of the orthogonalized Uij tensor.

	x	y	z	U(eq)
Fe(1)	1086(1)	7337(1)	2589(1)	45(1)
S(2)	262(2)	9401(2)	1579(1)	60(1)
S(3)	2028(2)	5113(1)	3398(1)	59(1)
C(1)	3311(5)	10703(5)	3979(3)	50(1)
C(5)	5156(5)	9732(5)	2915(3)	47(1)
C(2)	4943(6)	12133(5)	4394(3)	59(1)
C(4)	6863(6)	11162(5)	3295(3)	58(1)
C(3)	6765(6)	12361(5)	4051(3)	62(1)
N(1)	3376(4)	9461(4)	3264(2)	44(1)
C(7)	-895(6)	5560(6)	1920(3)	60(1)
C(6)	-503(5)	7414(5)	3493(3)	51(1)
O(1)	-1533(4)	7437(5)	4090(2)	78(1)
O(2)	-2225(5)	4422(5)	1502(2)	89(1)
C(8)	5184(5)	8328(5)	2139(3)	56(1)
N(2)	3147(4)	7259(4)	1652(2)	49(1)
C(10)	2383(6)	9832(7)	863(3)	67(1)
C(9)	2770(6)	8041(7)	684(3)	67(1)
C(12)	3204(7)	4349(6)	2414(4)	78(1)
C(11)	3037(6)	5294(6)	1455(3)	66(1)

Table S3. Bond lengths [Å] and angles [°] for 070912c.

Fe(1)-C(6)	1.751(4)
Fe(1)-C(7)	1.755(4)
Fe(1)-N(1)	2.005(3)
Fe(1)-N(2)	2.039(3)
Fe(1)-S(3)	2.3067(11)
Fe(1)-S(2)	2.3089(12)
S(2)-C(10)	1.811(4)
S(3)-C(12)	1.816(5)
C(1)-N(1)	1.352(4)
C(1)-C(2)	1.366(5)
C(1)-H(1)	0.9300
C(5)-N(1)	1.348(4)
C(5)-C(4)	1.382(5)
C(5)-C(8)	1.488(5)
C(2)-C(3)	1.378(6)
C(2)-H(2)	0.9300
C(4)-C(3)	1.377(6)
C(4)-H(4)	0.9300
C(3)-H(3)	0.9300
C(7)-O(2)	1.141(4)
C(6)-O(1)	1.141(4)
C(8)-N(2)	1.489(4)
C(8)-H(8A)	0.9700
C(8)-H(8B)	0.9700
N(2)-C(9)	1.491(5)
N(2)-C(11)	1.501(5)
C(10)-C(9)	1.511(6)
C(10)-H(10A)	0.9700
C(10)-H(10B)	0.9700
C(9)-H(9A)	0.9700
C(9)-H(9B)	0.9700
C(12)-C(11)	1.515(7)
C(12)-H(12A)	0.9700
C(12)-H(12B)	0.9700
C(11)-H(11A)	0.9700
C(11)-H(11B)	0.9700
C(6)-Fe(1)-C(7)	89.07(17)
C(6)-Fe(1)-N(1)	93.43(14)
C(7)-Fe(1)-N(1)	175.94(16)
C(6)-Fe(1)-N(2)	174.27(14)
C(7)-Fe(1)-N(2)	95.00(15)

N(1)-Fe(1)-N(2)	82.74(11)
C(6)-Fe(1)-S(3)	91.64(13)
C(7)-Fe(1)-S(3)	88.81(15)
N(1)-Fe(1)-S(3)	94.32(9)
N(2)-Fe(1)-S(3)	84.43(10)
C(6)-Fe(1)-S(2)	95.65(13)
C(7)-Fe(1)-S(2)	87.92(15)
N(1)-Fe(1)-S(2)	88.64(9)
N(2)-Fe(1)-S(2)	88.55(10)
S(3)-Fe(1)-S(2)	171.96(4)
C(10)-S(2)-Fe(1)	95.29(15)
C(12)-S(3)-Fe(1)	99.60(17)
N(1)-C(1)-C(2)	123.9(3)
N(1)-C(1)-H(1)	118.0
C(2)-C(1)-H(1)	118.0
N(1)-C(5)-C(4)	122.1(3)
N(1)-C(5)-C(8)	115.6(3)
C(4)-C(5)-C(8)	122.2(3)
C(1)-C(2)-C(3)	118.5(4)
C(1)-C(2)-H(2)	120.7
C(3)-C(2)-H(2)	120.7
C(3)-C(4)-C(5)	119.6(4)
C(3)-C(4)-H(4)	120.2
C(5)-C(4)-H(4)	120.2
C(4)-C(3)-C(2)	118.9(4)
C(4)-C(3)-H(3)	120.6
C(2)-C(3)-H(3)	120.6
C(5)-N(1)-C(1)	116.9(3)
C(5)-N(1)-Fe(1)	115.5(2)
C(1)-N(1)-Fe(1)	127.5(2)
O(2)-C(7)-Fe(1)	177.8(4)
O(1)-C(6)-Fe(1)	178.7(4)
C(5)-C(8)-N(2)	112.2(3)
C(5)-C(8)-H(8A)	109.2
N(2)-C(8)-H(8A)	109.2
C(5)-C(8)-H(8B)	109.2
N(2)-C(8)-H(8B)	109.2
H(8A)-C(8)-H(8B)	107.9
C(8)-N(2)-C(9)	110.3(3)
C(8)-N(2)-C(11)	108.0(3)
C(9)-N(2)-C(11)	109.1(3)
C(8)-N(2)-Fe(1)	109.2(2)
C(9)-N(2)-Fe(1)	110.5(2)
C(11)-N(2)-Fe(1)	109.7(2)

C(9)-C(10)-S(2)	109.0(3)
C(9)-C(10)-H(10A)	109.9
S(2)-C(10)-H(10A)	109.9
C(9)-C(10)-H(10B)	109.9
S(2)-C(10)-H(10B)	109.9
H(10A)-C(10)-H(10B)	108.3
N(2)-C(9)-C(10)	110.6(3)
N(2)-C(9)-H(9A)	109.5
C(10)-C(9)-H(9A)	109.5
N(2)-C(9)-H(9B)	109.5
C(10)-C(9)-H(9B)	109.5
H(9A)-C(9)-H(9B)	108.1
C(11)-C(12)-S(3)	112.3(3)
C(11)-C(12)-H(12A)	109.1
S(3)-C(12)-H(12A)	109.1
C(11)-C(12)-H(12B)	109.1
S(3)-C(12)-H(12B)	109.1
H(12A)-C(12)-H(12B)	107.9
N(2)-C(11)-C(12)	112.0(3)
N(2)-C(11)-H(11A)	109.2
C(12)-C(11)-H(11A)	109.2
N(2)-C(11)-H(11B)	109.2
C(12)-C(11)-H(11B)	109.2
H(11A)-C(11)-H(11B)	107.9

Table S4. Anisotropic displacement parameters ($Å^2$ x 10^3) for 070912c. The anisotropic displacement factor exponent takes the form: exp {-2π2(h2a*2U11 + ... + 2hka*b*U12)}

	U11	U22	U33	U23	U13	U12
Fe(1)	38(1)	60(1)	37(1)	3(1)	7(1)	17(1)
S(2)	51(1)	87(1)	51(1)	19(1)	11(1)	33(1)
S(3)	61(1)	62(1)	56(1)	9(1)	8(1)	22(1)
C(1)	53(2)	56(2)	46(2)	5(2)	10(2)	22(2)
C(5)	46(2)	54(2)	42(2)	10(2)	7(2)	16(2)
C(2)	66(3)	53(2)	56(3)	-1(2)	6(2)	20(2)
C(4)	45(2)	62(2)	65(3)	13(2)	11(2)	13(2)
C(3)	58(2)	51(2)	65(3)	1(2)	-6(2)	7(2)
N(1)	45(2)	53(2)	38(2)	8(1)	10(1)	19(1)
C(7)	49(2)	76(3)	52(2)	-1(2)	9(2)	19(2)
C(6)	48(2)	60(2)	47(2)	4(2)	9(2)	20(2)
O(1)	68(2)	108(2)	64(2)	12(2)	36(2)	31(2)
O(2)	59(2)	111(3)	75(2)	-25(2)	5(2)	5(2)
C(8)	40(2)	73(3)	59(2)	2(2)	11(2)	22(2)
N(2)	42(2)	65(2)	40(2)	-1(2)	9(1)	20(1)
C(10)	65(3)	101(3)	46(2)	28(2)	21(2)	37(2)
C(9)	62(3)	103(3)	41(2)	8(2)	16(2)	32(2)
C(12)	81(3)	72(3)	93(4)	8(3)	29(3)	36(2)
C(11)	61(2)	75(3)	67(3)	-12(2)	15(2)	30(2)

Table S5. Hydrogen coordinates (x 10^4) and isotropic displacement parameters ($Å^2$ x 10^3) for 070912c.

	x	y	z	U(eq)
H(1)	2081	10576	4203	60
H(2)	4827	12935	4896	70
H(4)	8071	11313	3040	70
H(3)	7908	13309	4326	74
H(8A)	5772	7480	2444	67
H(8B)	6017	8945	1635	67
H(10A)	2109	10313	230	81
H(10B)	3552	10743	1227	81
H(9A)	1625	7157	290	80
H(9B)	3919	8264	311	80
H(12A)	4603	4606	2638	94
H(12B)	2580	3021	2284	94
H(11A)	4108	5278	1064	79
H(11B)	1779	4610	1065	79

Crystal structure analysis of $C_{12}\ H_{14}\ Fe\ N_2\ O_2\ S_2$ [3, 4]

Crystal data: $C_{12}H_{14}FeN_2O_2S_2$, M = 338.22. Triclinic, space group P-1, a = 7.1525(11) Å, b = 7.6915(12) Å, c = 13.464(2) Å, α= 91.659(2)°, β = 95.127(2)°, γ = 109.7330(10)°, V = 693.15(18) $Å^3$. Z = 2, Dc = 1.621 g cm^{-3}, F(000) = 348, T = 293(2) K, μ(Mo-Kα) = 1.367 cm^{-1}, λ(Mo-Kα) = 0.71073 Å.

Crystals are dark-red blocks. From a sample under oil, one, *ca* 0.30 x 0.20 x 0.10 mm, was mounted on a glass fibre and fixed on an APEX CCD II diffractometer equipped with Mo-Kα radiation and graphite monochromator. Intensity data were measured by ω- and φ-scans.

Total no. of reflections recorded, to θ_{max} = 25.55°, was 4118 of which 2205 were unique (Rint = 0.023); 1722 were 'observed' with $I > 2\sigma_I$. The structure was solved by direct method using SHELXL-97 programs, and refined by full-matrix least-squares on F^2. All non-hydrogen atoms were refined anisotropically. Hydrogen atoms were added theoretically and refined riding on their mother atoms. The weighting scheme used was $w = 1/\{s^2(F_0^2)+(0.0650P)^2+0.11P\}$ where $P = (F_o^2+2F_c^2)/3$. The refinement was converged to final R_1 = 0.045, wR_2 = 0.135 [I>2*sigma(I)*], S = 1.010. the largest difference peak and hole were 0.42 and -0.54 *e*. $Å^{-3}$

References

1. M. Mikuruya and T. Kotera, *Chem. Lett.*, 1998, **10**, 971-972.
2. J. Takács, E. Soós, Z. Nagy-Magos and L. Markó, *Inorg. Chim. Acta*, 1989, **166**, 39-46.
3. G. M. Sheldrick, SHELX-97 – Programs for crystal structure determination (SHELXS) and refinement (SHELXL), University of Göttingen, Germany (1997).
4. L. J. Farrugia, *J. Appl. Cryst.*, (1999) **32**, 837-838.