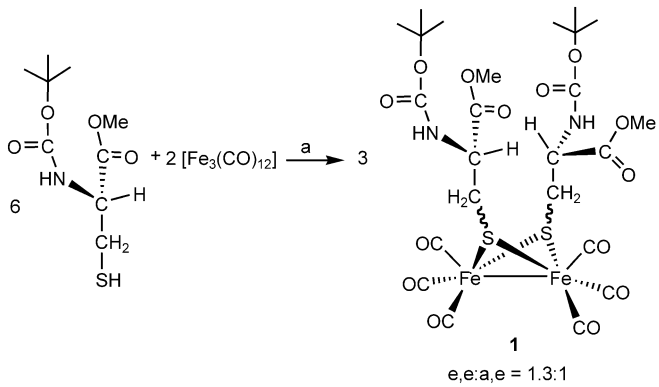
*Bioinorganic Chemistry*

# An Unusual Cyclization in a Bis(cysteinyl-*S*) Diiron Complex Related to the Active Site of Fe-Only Hydrogenases**

*Chengjiang He, Mei Wang,* Xiaofeng Zhang, Zhen Wang, Changneng Chen, Jianhui Liu, Björn Åkermark, and Licheng Sun**

The recent unveiling of high-quality crystal structures of Fe-only hydrogenases isolated from *Desulfovibrio desulfuricans* and *Clostridium pasteurianum* has renewed interest in classical organometallic diiron dithiolate complexes, $[(\mu\text{-}SR)_2Fe_2(CO)_6]$ (R = Me, Et, $CH_2CH{=}CH_2$, Ph). Crystallographic studies have revealed that cysteinyl ligands exist in the active sites of [NiFe] and Fe-only hydrogenases. The active site of the [NiFe] hydrogenase from *Desulfovibrio gigas* has been established as a pyramidal $[Fe(CN)_2(CO)]$ unit with the face opposite the CN and CO ligands coordinated to two bridging cysteinyl ligands that connect the Fe and Ni moieties.[1,2] These [NiFe] dinuclear units are connected to their respective proteins by multiple cysteine residues. The H cluster of the Fe-only hydrogenase contains a single cysteinyl ligand that connects a [4Fe4S] subcluster to a dinuclear iron subcluster through a sulfur atom.[3–6] These maps of the active sites of the two major families of hydrogenase hint that the cysteinyl ligand is indispensable for $H_2$ production and uptake by the metal-containing hydrogenases. Although the synthesis and reactions of diverse dithiolate $Fe^IFe^I$ hexacarbonyl compounds and their derivatives have attracted much attention,[7,8] no diiron carbonyl complex with cysteinyl ligands has been reported in the literature so far, despite the important function of the cysteinyl ligand in the H cluster. We are interested in the preparation of dinuclear iron complexes featuring one or two cysteinyl ligands with the aim of gaining an insight into the role of this amino acid in proton reduction by active-site model complexes.

The preparation of diiron dithiolate complexes by oxidative addition of thiols to iron(0) carbonyl compounds can be traced back more than half a century.[9] Many diiron complexes with the general formula $[(\mu\text{-}SR)_2Fe_2(CO)_6]$ can be obtained by using this traditional protocol.[10–13] Initial attempts to introduce two cysteinyl ligands to a dinuclear iron complex by treatment of $[Fe_3(CO)_{12}]$ with cysteine or its methyl ester were unsuccessful as the target product could not be separated from the resulting mixture by column chromatography. A protecting group (*tert*-butoxycarbonyl, Boc) was thus introduced at the amino group of the cysteine methyl ester. Treatment of the amino-protected L-cysteine ester with $[Fe_3(CO)_{12}]$ in refluxing MeOH for 1 h gave the predesigned diiron complex **1** as a dark-red crystalline product in about 30% yield (Scheme 1). The $^1H$ and $^{13}C$ NMR spectra of the product show that the bis(cysteinyl-*S*) diiron complex **1** obtained is a mixture of e,e (**1a**) and a,e (**1b**) isomers




***Scheme 1.*** Synthesis of complex **1**. a) MeOH, reflux, 1 h, 30%.

[*] C. He, Prof. M. Wang, Z. Wang, Dr. J. Liu, Prof. L. Sun
State Key Laboratory of Fine Chemicals
Dalian University of Technology
Zhongshan Road 158-46, Dalian 116012 (P.R. China)
Fax: (+86) 411-8370-2185
E-mail: symbueno@dlut.edu.cn
X. Zhang, Prof. C. Chen
State Key Laboratory of Structural Chemistry
Fujian Institute of Research on the Structure of Matter
Fuzhou 350002 (P.R. China)
Prof. B. Åkermark, Prof. L. Sun
Department of Organic Chemistry
Arrhenius Laboratory
Stockholm University
10691 Stockholm (Sweden)
E-mail: licheng.sun@organ.su.se

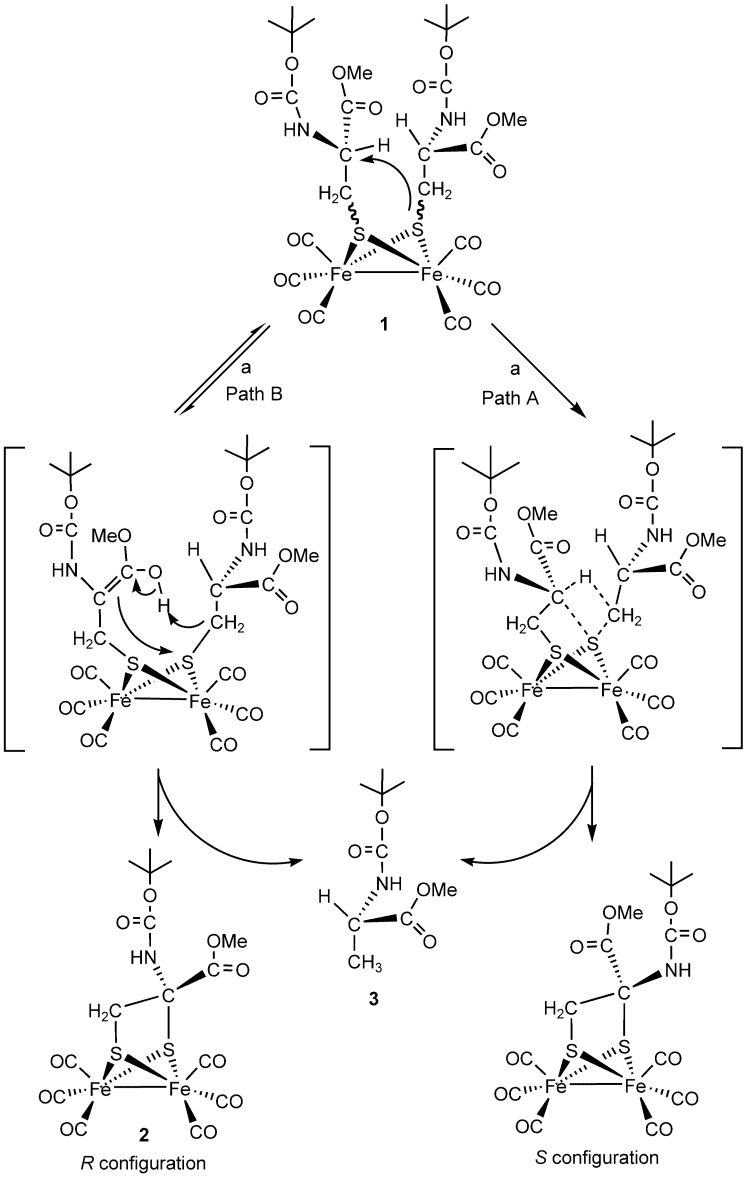
[**] Financial support for this work from the Ministry of Science and Technology of China and the Chinese National Natural Science Foundation (Grant nos. 20128005 and 20173006) is gratefully acknowledged.

 DOI: 10.1002/ange.200453961  

(e, equatorial; a, axial). The e,e to a,e ratio is 1.3:1, as determined by integration of the $^1$H NMR spectrum. This kind of geometrical isomerization, derived from the orientation of the α-C atom on the bridging thiolate, has been reported for $[(\mu\text{-}RS)_2Fe_2(CO)_6]$ complexes ($R = CH_3$, $CH_2CH_3$, $CH_2C_6H_5$) by King[9] and Seyferth.[14–18]

To the best of our knowledge, complex **1** is the first synthetic diiron carbonyl compound containing cysteinyl ligands. The complex is relatively stable in the solid state but is unstable in solution. When **1** was refluxed in MeOH for 30 h or in toluene and HOAc (2 equiv) for several hours, an unexpected intramolecular cyclization reaction occurred (Scheme 2). This reaction generated a two-carbon-bridged dithiolate diiron complex (**2**, yellow crystals) in about 5–12% yield by elimination of one equivalent of L-alanine methyl ester **3**, which has a Boc-protected amino group. The products were identified in solution by mass spectrometry. An uncharacterized precipitate and cysteinyl disulfide were also formed. The mechanism for this spontaneous intramolecular cyclization is not clear. One can imagine two paths for intramolecular cyclization of complex **1** in which one of the bridged sulfur atoms acts as a nucleophile (Scheme 2). The suggested nucleophilicity of the bridging sulfur atom is supported by the fact that S-alkylation of $[(\mu\text{-EDT})\{Fe(CO)_2(PMe_3)\}_2]$ (EDT = ethanedithiolato) with $EtOSO_2CF_3$ results in the formation of $[(\mu\text{-}SCH_2CH_2SEt)\{Fe(CO)_2(PMe_3)\}_2]^+[SO_3CF_3]^-$.[19] Both the paths shown in Scheme 2 lead to a chiral carbon-bridged diiron complex. In Path A, the sulfur atom attacks the stereogenic carbon atom from the side of the leaving hydrogen atom and the reaction proceeds through a transition state containing a four-membered ring. In Path B, an enolization takes place first, followed by an attack on the sulfur atom and displacement of an alanine derivative.

Although the details of the mechanism for this unprecedented intramolecular cyclization reaction are not clear, the molecular structure of the product **2** could be determined by X-ray crystallography.[20] The resulting structure is one of only a few well-characterized crystal structures of chiral carbon-bridged diiron complexes related to the active site of Fe-only hydrogenases.[21,22] Single-crystal X-ray diffraction studies of **2** show the typical butterfly framework of a 2Fe2S complex and the familiar pyramidal geometry around each iron atom (Figure 1). The Fe–Fe bond length (2.5101(8) Å) determined for **2** is comparable to the Fe–Fe bond lengths found in the structures of other $SCH_2CHRS$-bridged diiron hexacarbonyl complexes (Fe–Fe: R = H, 2.497(4); R = Me, 2.5196(7) Å).[22–24] The distance between the two bridging carbon atoms (C7–C8, 1.523(5) Å) is slightly longer in **2**




***Scheme 2.*** Proposed pathway for the formation of complex **2**. a) MeOH, reflux, 30 h or HOAc (2 equiv), $CH_3C_6H_5$, reflux, 2 h; 5–12%.

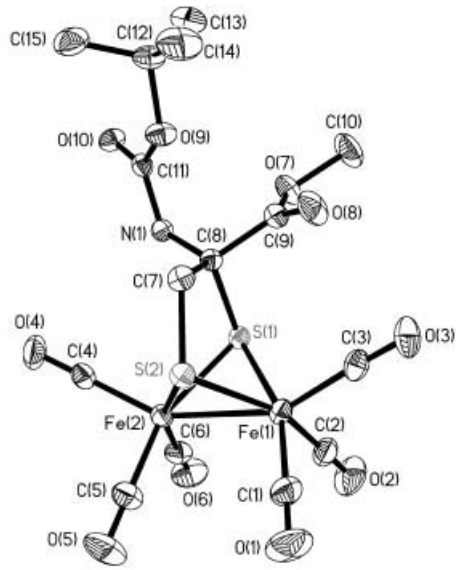



***Figure 1.*** Molecular structure of **2** with thermal ellipsoids set at 30% probability. Selected bond lengths [Å]: Fe1–Fe2, 2.5101(8); Fe1–S1, 2.2359(10); Fe1–S2, 2.2410(11); S1–C8, 1.863(4); S2–C7, 1.823(4); C7–C8, 1.523(5); N1–C8, 1.453(4); C8–C9, 1.535(5). Selected bond angels [°]: Fe1–S1–Fe2, 68.24(3); C11–N1–C8, 122.8(3); C8–C7–S2, 113.4(3); C7–C8–S1, 110.1(2); C7–C8–C9, 111.4(3); C9–C8–S1, 104.6(2); N1–C8–C7, 113.6(3); N1–C8–C9, 111.7(3); N1–C8–S1, 105.0(2).

than in the analogues complex $[(\mu\text{-}SCH_2CHRS)Fe_2(CO)_6]$ (bridging C–C: R = H, 1.487(6); R = Me, 1.496(6) Å). The reported mirror symmetry of $[(\mu\text{-}SCH_2CHRS)Fe_2(CO)_6]$ (R = H, Me)[22–24] in the plane defined by S1, S2, C7, and C8 is not present in **2**. The C8–C7–S2 bond angle in **2** is larger than the C7–C8–S1 angle by 3.3°. The most noteworthy characteristic of the crystal structure of **2** is that the stereogenic carbon atom in the bridge has an *R* configuration. In the light of the *R* configuration of the L-cysteine reactant, this structural result suggests that the bridging sulfur atom attacks the methine group while the C–H bond cleaves heterolytically, then the intramolecular cyclization reaction takes place with a turnover of the stereogenic carbon atom, that is, with an inversion of the configuration. An enolization mechanism (Scheme 2, Path B) would lead to a chiral carbon-bridged diiron complex mainly with the *R* configuration, while a concerted mechanism (Scheme 2, Path A) would be stereoselective for a chiral diiron product with an *S* configuration. The C11–N1 bond in the bridged-cysteinyl ligand is in an axial orientation, which avoids significant congestion of the ligands. A dimer of **2** in the solid state is formed by intermolecular H bonding of two amido groups (N(1)···O(10), 2.890(4) Å; symmetry: $-x+1, -y, -z+1$; Figure 2).

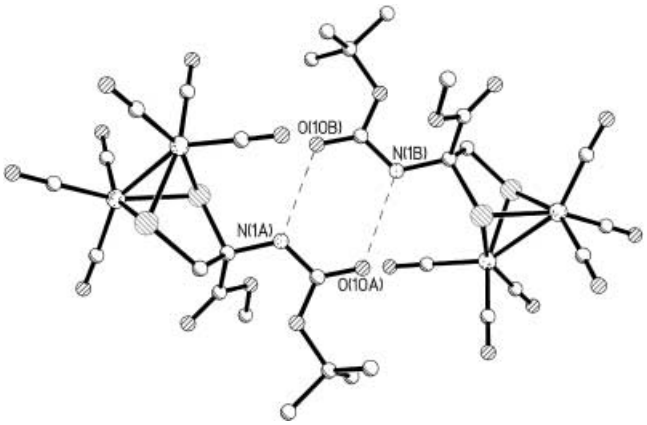



***Figure 2.*** Plot of the unit cell of **2**. The dotted lines represent H bonds between the N1 and O10 atoms of the two molecules.

In conclusion, a dinuclear iron complex (**1**) containing two protected cysteinyl ligands was prepared by the classical oxidative addition of thiols to $[Fe_3(CO)_{12}]$. An unusual intramolecular cyclization reaction of **1** gave the chiral carbon-bridged diiron complex **2**, whose structure is related to that of the active site of Fe-only hydrogenases. The molecular structure of **2** was determined by X-ray crystallography and suggests that the unprecedented nucleophilic cyclization reaction occurs with a inversion of configuration.

## *Experimental Section*

**1**: *N*-Boc-protected L-cysteine methyl ester (0.75 mL, 5.70 mmol, 7.5 M in $CHCl_3$) was added dropwise to a solution of $[Fe_3(CO)_{12}]$ (1.0 g, 1.98 mmol) in MeOH (20 mL). The mixture was refluxed for 1 h, which resulted in formation of a dark-red solution. The solvent was evaporated in vacuo and the residue was chromatographed on an $Al_2O_3$ column with hexane/$CH_2Cl_2$ (1:10) as the eluent. Recrystallization of the crude product in hexane/$CH_2Cl_2$ gave **1** as dark red crystals (640 mg). Yield: 30%; elemental analysis (%): calcd for $C_{24}H_{32}Fe_2N_2O_{14}S_2$: C, 38.52, H, 4.31, N, 3.74; found: C, 38.73, H, 4.48, N, 3.81; $^1$H NMR (400 MHz, $CDCl_3$): $\delta = 1.47$ (s, 18H; *t*Bu), 2.38, 2.50, 2.83 (3 × m, 4H; $SCH_2$), 3.78 (s; $OCH_3$, a,e), 3.83 (s; $OCH_3$, e,e), 3.85 (s, 6H; $OCH_3$, 57% e,e, 43% a,e), 4.39, 4.63 (2 × m, 2H; CH), 5.17, 5.42 ppm (d, 2H; NH); $^{13}$C NMR (400 MHz, $CDCl_3$): $\delta = 28.40$, 29.46 (2 × s; $CH_3$ of *t*Bu), 39.47, 40.88 (2 × s; $SCH_2$), 53.11, 54.17, 54.39 (3 × s; $OCH_3$), 80.56, 80.72 (2 × s; CH), 105.32 (s; C of *t*Bu), 154.92, 155.04 (2 × s; CONH), 170.15, 170.38, 170.69 (3 × s; COO), 207.86, 208.03, 208.83 ppm (3 × s; CO); IR ($CH_2Cl_2$): $\tilde{\nu} = 2073, 2036, 1994$ (CO), 1745, 1716 $cm^{-1}$ (C=O); ESI MS (*m*/*z*): 770.9 (100%) $[M+Na]^+$.

**2**: Either a mixture of $[Fe_3(CO)_{12}]$ and *N*-Boc-protected L-cysteine methyl ester or a solution of **1** in MeOH was refluxed for 30 h to give a dark-orange solution. Complex **2** was isolated by chromatography on silica gel with MeOH/$CH_2Cl_2$ (2:98 v/v) as the eluent. The first yellow band was collected and the solvent was removed in vacuo. The residue was crystallized by defusing hexane into a solution of the residue in $CH_2Cl_2$. Yield: 5–12%; $^1$H NMR (400 MHz, $CDCl_3$): $\delta = 1.46$ (s, 9H; *t*Bu), 2.65–3.08 (br s, 2H; $SCH_2$), 3.76 (s, 3H; $OCH_3$), 5.50 ppm (s, 1H; NH); IR ($CH_2Cl_2$): $\tilde{\nu} = 2085$ (w), 2075 (w), 2058 (m), 2042 (s), 1996 (s) (CO), 1743, 1718 $cm^{-1}$ (C=O); TOF-ESI MS (*m*/*z*): calcd for $[M+Na]^+$: 567.8734; found: 567.8735 (100%).

Received: February 6, 2004
Revised: April 13, 2004 [Z53961]

**Keywords:**
S ligands · cyclization · enzyme models · iron hydrogenases · structure elucidation

[1] M. W. W. Adams, E. I. Stiefel, *Curr. Opin. Chem. Biol.* **2001**, *5*, 214–220.
[2] M. Frey, *ChemBioChem* **2002**, *3*, 153–160.
[3] Y. Nicolet, C. Piras, P. Legrand, C. E. Hatchikian, J. C. Fontecilla-Camps, *Structure* **1999**, *7*, 13–23.
[4] J. W. Peters, W. N. Lanzilotta, B. J. Lemon, L. C. Seefeldt, *Science* **1998**, *282*, 1853–1858.
[5] R. Cammack, *Nature* **1999**, *397*, 214–215.
[6] Y. Nicolet, B. J. Lemon, J. C. Fontecilla-Camps, J. W. Peters, *Trends Biochem. Sci.* **2000**, *25*, 138–143.
[7] M. Y. Darensbourg, E. J. Lyon, J. J. Smee, *Coord. Chem. Rev.* **2000**, *206–207*, 533–561.
[8] D. J. Evans, C. J. Pickett, *Chem. Soc. Rev.* **2003**, *32*, 268–275.
[9] R. B. King, *J. Am. Chem. Soc.* **1962**, *84*, 2460.
[10] J. A. Beer, R. J. Haines, *J. Organomet. Chem.* **1970**, *24*, 757–767.
[11] N. S. Nametkin, V. D. Tyurin, M. A. Kukina, *J. Organomet. Chem.* **1978**, *149*, 355–370.
[12] X. Zhao, Y. M. Hsiao, C. H. Lai, J. H. Reibenspies, M. Y. Darensbourg, *Inorg. Chem.* **2002**, *41*, 699–708.
[13] A. Winter, L. Zsolnai, G. Huttner, *Z. Naturforsch. B* **1982**, *37*, 1430–1436.
[14] L. F. Dahl, C. H. Wei, *Inorg. Chem.* **1963**, *2*, 328–333.
[15] G. Bor, *J. Organomet. Chem.* **1968**, *11*, 195–197.
[16] G. L. Borgne, D. Grandjean, *J. Organomet. Chem.* **1977**, *131*, 429–438.
[17] D. Seyferth, R. S. Henderson, *J. Organomet. Chem.* **1981**, *218*, C34–C36.
[18] D. Seyferth, R. S. Henderson, L. C. Song, *Organometallics* **1982**, *1*, 125–133.
[19] X. Zhao, C. Y. Chiang, M. L. Miller, M. V. Rampersad, M. Y. Darensbourg, *J. Am. Chem. Soc.* **2003**, *125*, 518–524.
[20] Crystal data for **2**: $C_{15}H_{15}Fe_2NO_{10}S_2$, $M_r = 545.10$; triclinic; space group $P\bar{1}$; $a = 7.6905(4)$, $b = 12.4367(6)$, $c = 13.3307(7)$ Å, $\alpha =$

64.253(1), $\beta = 79.414(1)$, $\gamma = 89.292(1)°$, $V = 1125.6(1)$ Å$^3$; $\rho_{calcd} = 1.608$ g m$^{-3}$; $\mu = 1.523$ mm$^{-1}$; $T = 293(2)$ K; $Z = 2$; $R_1 = 0.0435$ and $wR_2 = 0.1037$ for 3373 reflections with $I > 2\sigma(I)$. CCDC-224401 (**2**) contains the supplementary crystallographic data for this paper. These data can be obtained free of charge via www.ccdc.cam.ac.uk/conts/retrieving.html (or from the Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB21EZ, UK; fax: (+44) 1223-336-033; or deposit@ccdc.cam.ac.uk).

[21] D. Seyferth, G. B. Womack, R. S. Henderson, *Organometallics* **1986**, *5*, 1568 – 1575.

[22] Q. Q. Zhang, R. S. Dickson, G. D. Fallon, R. Mayadunne, *J. Organomet. Chem.* **2001**, *627*, 201 – 205.

[23] D. L. Hughes, G. J. Leigh, D. R. Paulson, *Inorg. Chim. Acta* **1986**, *120*, 191 – 195.

[24] J. Messelhäuser, I. Lorenz, K. Huag, W. Hiller, *Z. Naturforsch. B* **1985**, *40*, 1064 – 1067.