CHEMISTRY
A EUROPEAN JOURNAL

COMMUNICATION

DOI: 10.1002/chem.201300495

Reversible Dimerization of Mononuclear Models of [Fe]-Hydrogenase

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[Fe]-Hydrogenase, which can activate H_2 in the presence of methenyltetrahydromethanopterin (methenyl-H_4MPT^+), is the only mononuclear hydrogenase known.[1–3] In its active site, there are two *cis*-CO, a cysteine sulfur atom (Cys 176), and a bidentate pyridinol acyl ligand (Figure 1, A).[4,5] The coordination position *trans* to the acyl group is regarded as the H_2-binding position.[6]

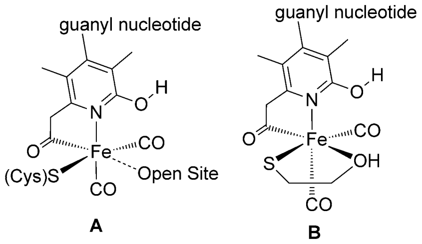

Figure 1. The active site structure of [Fe]-hydrogenase (**A**) and the proposed structure of the protein-free FeGP cofactor extracted with 2-mercaptoethanol (**B**).

[Fe]-Hydrogenase also contains an iron guanylylpyridinol (FeGP) cofactor, which can be extracted by denaturation of the enzyme in the presence of 2-mercaptoethanol.[7,8] The protein-free cofactor is too unstable to be identified; however, its structure was proposed based on the active site structure of [Fe]-hydrogenase (Figure 1, B).[9–11]

Following the elucidation of the active site of [Fe]-hydrogenase, a number of synthetic models have been synthesized.[12–28] Recently, we prepared a five-coordinate model complex $[Fe(2\text{-}CH_2CO\text{-}6\text{-}MeOC_5H_3N)(CO)_2\{S\text{-}(2,6\text{-}Me_2C_6H_3)\}]$ (**1**) with an open site.[25] However, **1** did not react with H_2 under ambient conditions. In continuation of this work, similar mononuclear models with alkyl thiolate ligands were targeted. However, only dimeric complexes were obtained, both in the solid state and in solution.[27] Herein, we describe the equilibrium between the dimeric complexes and their monomeric components in solution.

Reaction of $[Fe(2\text{-}CH_2CO\text{-}6\text{-}MeOC_5H_3N)(CO)_3I]$ (**2**)[25] with $(4\text{-}NO_2\text{-}C_6H_4)SNa$ gave a dinuclear complex that has a formula of $[\{Fe(2\text{-}CH_2CO\text{-}6\text{-}MeOC_5H_3N)(CO)_2\{S\text{-}(4\text{-}NO_2\text{-}C_6H_4)\}\}_2]$ (**3a**) in the solid state, as revealed by X-ray crystallography (Scheme 1). Complex **3a** has a C_2 symmetry (Figure 2). The C_2 axis passes through the midpoint of Fe1–Fe2 and is perpendicular to the plane of Fe1-S1-Fe2-S2. Each iron ion is coordinated in an octahedral geometry by two *cis*-CO, two thiolate ligands, and the acylmethylpyridinyl moiety. The IR spectrum of **3a** in the solid state shows four intense ν(CO) absorption bands.

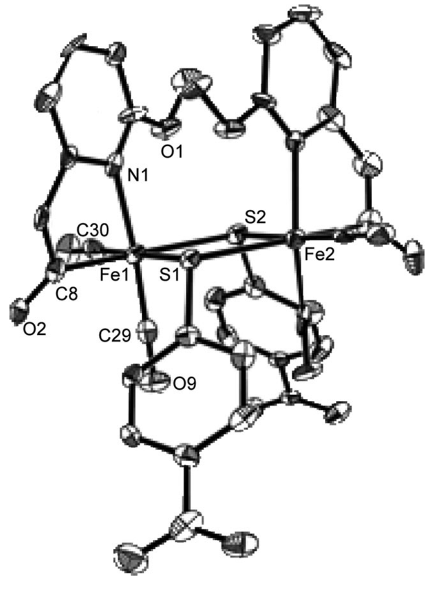

Figure 2. Solid-state structure of **3a**. The thermal ellipsoids are displayed at a 50% probability. Selected bond lengths [Å] and angles [°]: Fe1–N1 2.069(11), Fe1–C8 1.947(13), Fe1–C29 1.754(14), Fe1–C30 1.794(15), Fe1–S1 2.367(4), Fe1–S2, 2.469(4); C29-Fe1-C30 90.4(6), C8-Fe1-N1, 82.9(5).

The ^1H NMR spectrum of **3a** in CD_3CN exhibits three signals at 7.83, 7.06, and 6.82 ppm for the pyridyl rings, two signals at 7.79 and 7.29 ppm for the phenyl rings, and one doublet at 4.52 and one multiplet at 3.88 ppm for the diastereotopic methylene hydrogen atoms and the methoxy groups.[29] In the previously reported dinuclear complex $[\{Fe(6\text{-}MeO\text{-}C_5H_3N\text{-}2\text{-}CH_2CO)(CO)_2(SCH_2CH_2OH)\}_2]$ (**4**),[27] four multiplets were found for $\text{-}CH_2CH_2OH$ groups because the

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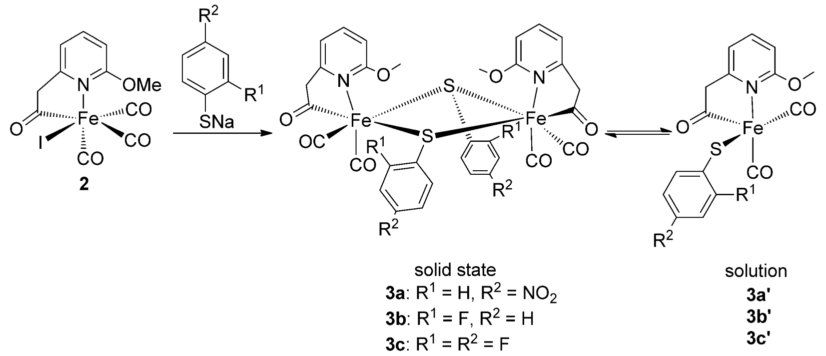
Supporting information for this article is available on the WWW under http://dx.doi.org/10.1002/chem.201300495.



Scheme 1. Reactions of **2** with sodium aryl thiolate.

bulky ligand environment around the Fe centers restricts the rotation of the S-CH_2 bonds. In principle, the Fe centers in **3a** should be more sterically shielded than in **4**, so it was surprising that only two instead of four signals for the phenyl rings were observed in the solution NMR spectrum. This abnormality prompted us to record the IR spectrum of **3a** in solution as well. Interestingly, only two intense ν(CO) absorption bands were observed in CH_3CN and $CHCl_3$ (Table 1). The reaction of **3a** with CH_3CN was further monitored in $CDCl_3$ by ^1H NMR spectroscopy, and no reactivity was found, confirming that CH_3CN was not coordinated with Fe. These observations indicate that a five-coordinate monomer (**3a'**) rather than the dimeric **3a** is the main species in solution (Scheme 1). It is worth noting that the ν(CO) absorptions in CH_3CN of **3a'** are almost identical to that of the FeGP cofactor (Table 1).

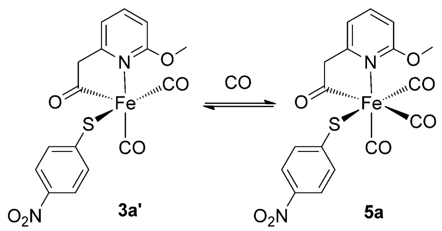
Table 1. Selected infrared data.

Complex	$\tilde{\nu}_{CO}$ [cm^{-1}]
3a[a]	2024, 2007, 1977, 1962
3a'[b]	2032, 1973
3a'[c]	2038, 1977
3b[a]	2031, 2011, 1980, 1965
3b'[c]	2036, 1975
3c[a]	2028, 2004, 1986, 1961
3c'[b]	2030, 1967
3d'[a]	2025, 1962
3d'[b]	2030, 1967
3e[a]	2025, 1994, 1955, 1934
3e'[b]	2015, 1951
5a[b]	2083, 2034, 2008
5d[b]	2085, 2035, 2008
[Fe]-hydrogenase[d]	2011, 1944
FeGP cofactor[d]	2031, 1972
CO-inhibited cofactor[e]	2080 (the other two bands were not identified)

[a] Spectrum of a solid sample on KBr disk. [b] Spectrum of a sample dissolved in CH_3CN. [c] Spectrum of a sample dissolved in $CHCl_3$. [d] Spectrum of a sample dissolved in water; data from reference [30]. [e] Spectrum of a sample dissolved in water; data from reference [31].

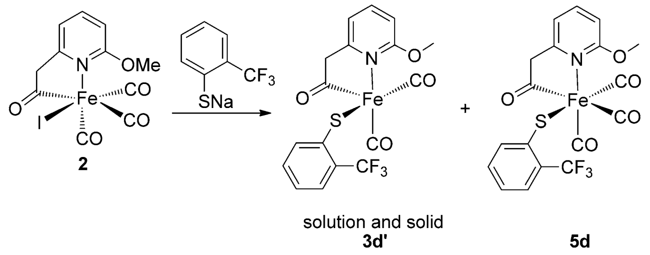
Complex **3a'** is unstable in solution at room temperature; its half-life was about 1 h in the dark. One of the decomposition products, interestingly, was the tris(carbonyl) complex [Fe(2-CH_2CO-6-$MeOC_5H_3N$)$(CO)_3${S-(4-NO_2-C_6H_4)}] (**5a**), which was the product of the reaction of **3a'** with CO (Scheme 2).[29] This kind of carbonylation was demonstrated by Rauchfuss et al. before.[16]

Reaction of **2** with (2-F-C_6H_4)SNa and (2,4-F_2-C_6H_3)SNa gave [{Fe(2-CH_2CO-6-$MeOC_5H_3N$)$(CO)_2${S-(2-F-C_6H_4)}}$_2$] (**3b**) and [{Fe(2-CH_2CO-6-$MeOC_5H_3N$)$(CO)_2${S-(2,4-F_2-C_6H_3)}}$_2$] (**3c**), respectively (Scheme 1). Like **3a**, these compounds are dimeric in the solid state. The structure of **3b** was also confirmed by X-ray crystallography.[29] Both **3b** and **3c** convert to monomers (**3b'** and **3c'**) in solution (Scheme 1), which was confirmed by IR spectroscopy (Table 1). The half-lives of **3b'** and **3c'** were both about 2 h in the dark.



Scheme 2. Reaction of **3a'** with CO.

Reaction of **2** with (2-CF_3-C_6H_4)SNa gave a mixture of [Fe(2-CH_2CO-6-$MeOC_5H_3N$)$(CO)_2${S-(2-CF_3-C_6H_4)}] (**3d'**) and [Fe(2-CH_2CO-6-$MeOC_5H_3N$)$(CO)_3${S-(2-CF_3-C_6H_4)}] (**5d**) (Scheme 3). At −30 °C, two species could be observed with a ratio of 5:4 in the NMR spectrum of the product mixture.[29] At room temperature, the signals from the two species coalesced.[29] Thus, **3d'** and **5d** were involved in a fast self-exchange reaction, concomitant with the transfer of a CO ligand. Unfortunately, pure **3d'** could not be isolated from the mixture. When CO (1 atm) was added into this mixture, **3d'** was converted into **5d**.[29]

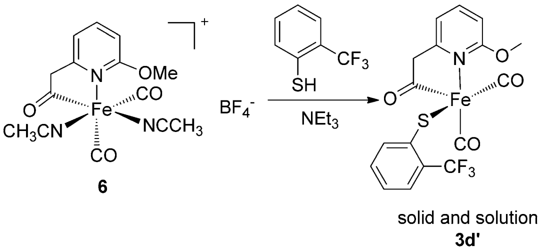


Scheme 3. Reaction of **2** with (2-CF_3-C_6H_4)SNa.

Analogous reactions of **2** with sodium aryl thiolates without an electron-withdrawing group, for example, PhSNa, (2-

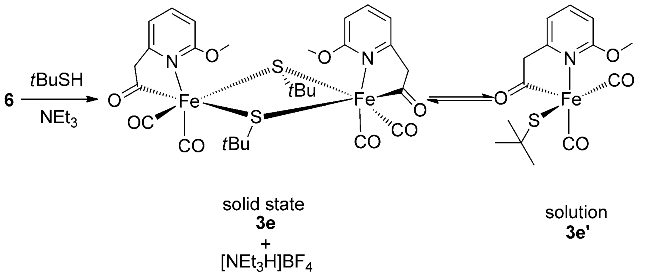
MeC_6H_4)SNa, (2-EtC_6H_4)SNa, (2-*i*PrC_6H_4)SNa, (2-OMe-C_6H_4)SNa, and (4-MeC_6H_4)SNa, did not yield isolable complexes, because of the instability of the expected products.

To obtain pure **3d′**, an alternative route was attempted: [Fe(2-CH_2CO-6-MeOC_5H_3N)$(CO)_2(CH_3CN)_2$]BF_4 (**6**)[26] was treated with (2-CF_3-C_6H_4)SH in the presence of NEt_3 at −30°C (Scheme 4). The reaction was fast, indicated by an instantaneous change of color of the reaction medium from yellow to red. The IR spectra of **3d**, collected in the solid state and in solution, both show two intense *ν*(CO) absorption bands (Table 1). This result indicates that **3d′** exists in a monomeric form even in the solid state. The half-life of **3d′** in solution was about 0.5 h.



Scheme 4. Reaction of **6** with (2-CF_3-C_6H_4)SH and NEt_3.

A similar method was applied for the synthesis of [{Fe(2-CH_2CO-6-MeOC_5H_3N)$(CO)_2${S-(*t*Bu)}}$_2$] (**3e**) (Scheme 5). Although the formation of **3e** could be confirmed by ^{1}H NMR and IR spectroscopy, this compound could not be isolated due to its high instability in solution (**3e′**) even at −30°C. Interestingly, different from **4**, complex **3e** converts to a monomeric form (**3e′**) in solution, evidenced by IR spectroscopy (Table 1). This is the first five-coordinate mononuclear Fe complex with an alkyl thiolate ligand that models the main structural features of [Fe]-hydrogenase.



Scheme 5. Reactions of **6** with *t*BuSH and NEt_3.

Like **1**, **3a′**–**3e′** do not react with H_2 under ambient conditions. This is not surprising, because [Fe]-hydrogenase only activates H_2 in the presence of methenyl-H_4MPT^+.[1–3] It is proposed that the 2-hydroxyl group in the pyridyl ring of the active site of [Fe]-hydrogenase may play a very important role.[32] Research on developing models with acylmethylpyridinol ligand is ongoing in our labs.

In summary, a series of new iron acyl thiolate complexes have been synthesized and characterized. These complexes can be used as structural and reactivity models for the active site of [Fe]-hydrogenase. With electron-withdrawing aryl thiolate ligands, five-coordinate mononuclear iron complexes are the dominate species in solution. However, their dimeric forms are more stable in the solid state. Substitution at the *ortho*-position of the aryl thiolate is required to favor the monomeric form in the solid-state. The first five-coordinate iron acyl model complex with an alkyl thiolate ligand (**3e′**) has also been prepared, albeit not isolated. The insights gained from this study are useful for the development of the next generation of model compounds.

Acknowledgements

This work is supported by the National Natural Science Foundation of China (Nos. 21242012 and 21201049), the Fundamental Research Funds for the Central Universities (Grant No. HIT. NSRIF. 2013042), and the Swiss National Science Foundation (project no. 200020_134473/1).

Keywords: alkyl · enzyme models · hydrogenase · iron · thiolate

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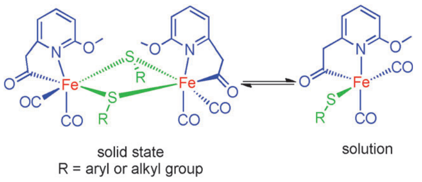
Received: February 7, 2013

Enzyme models on the catwalk! A series of new model complexes of the active site of [Fe]-hydrogenase have been synthesized and characterized. These complexes are monomeric in solution, but dimeric in the solid state.



Enzyme Models

B. Hu, D. Chen, X. Hu** . . . ▮▮▮▮–▮▮▮▮

Reversible Dimerization of Mononuclear Models of [Fe]-Hydrogenase

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A unique bidentate pyridinol cofactor…

… is to be found in [Fe]-hydrogenase. In their Communication on page ■■ ff., D. Chen, X. Hu and B. Hu demonstrate the synthesis and reactivities of a series of mononuclear models, including the first five-coordinate complex with an alkyl thiolate ligand, that mimic the main structure of [Fe]-hydrogenase active site. The equilibrium between these models and their dimeric components is also described.