

# A Nitrogenase Cluster Model $[\text{Fe}_8\text{S}_6\text{O}]$ with an Oxygen Unsymmetrically Bridging Two Proto- $\text{Fe}_4\text{S}_3$ Cubes: Relevancy to the Substrate Binding Mode of the FeMo Cofactor

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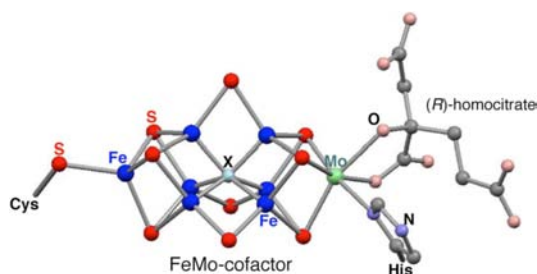
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## S Supporting Information

**ABSTRACT:** An oxygen-encapsulated iron sulfido cluster,  $[(\text{DmpS})\text{Fe}_4\text{S}_3\text{O}][(\text{DmpS})\text{Fe}_4\text{S}_3](\mu\text{-SDmp})_2(\mu\text{-OCPh}_3)$  (**2**; Dmp = 2,6-(mesityl)<sub>2</sub>C<sub>6</sub>H<sub>3</sub>), has been synthesized by the reaction of the preformed dinuclear iron thiolate/alkoxide  $[(\text{Ph}_3\text{CO})\text{Fe}]_2(\mu\text{-SDmp})_2$  (**1**) with  $1/8\text{S}_8$  and  $1/4\text{H}_2\text{O}$  in toluene. In the  $[\text{Fe}_8\text{S}_6\text{O}]$  core, the oxygen atom bridges unsymmetrically two incomplete  $\text{Fe}_4\text{S}_3$  cubes, and two coordinatively unsaturated iron atoms are weakly bound to mesityl rings. Relevancy of the cluster structure of **2** to the nitrogenase FeMo cofactor and its substrate binding mode is discussed.

Molybdenum-dependent nitrogenase,<sup>1</sup> which catalyzes the reduction of dinitrogen to ammonia, is composed of two component metalloproteins called the Fe protein and the MoFe protein. The active site of the enzyme is the MoFe protein, and an Fe/Mo sulfido cluster called the FeMo cofactor (Figure 1) is



**Figure 1.** Structure of the nitrogenase FeMo cofactor (X = C, N, or O).<sup>3</sup>

thought to serve as the binding and reduction site of dinitrogen. Until a decade ago, the cluster core of the FeMo cofactor had been considered to be  $[\text{MoFe}_7\text{S}_9]$  with a trigonal-prismatic  $\text{Fe}_6$  inner cage.<sup>2</sup> A high-resolution (1.16 Å) crystallographic analysis of the MoFe protein, however, revealed electron density from a single atom X at the center of the  $\text{Fe}_6$  inner frame.<sup>3</sup> The electron density associated with X and the Fe–X bond distances are consistent with light elements such as oxygen, nitrogen, and carbon. Recently, X has been strongly suggested to be carbon, based on data from several techniques.<sup>4</sup>

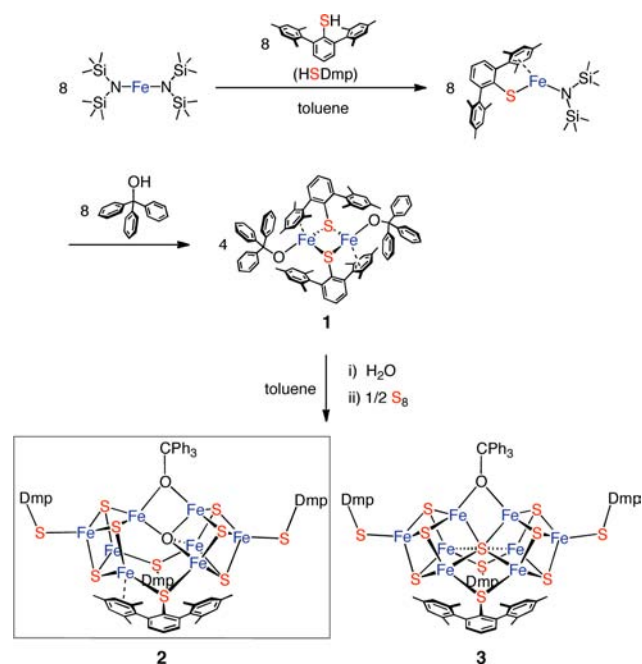
Because of the unparalleled structure and important function of the FeMo cofactor, the synthesis of model clusters has received considerable attention by inorganic chemists. Recently, Lee et al. reported cubane cluster  $[\text{Fe}_4\text{S}_3(\text{N}^t\text{Bu})\text{Cl}_4]^{2-}$ , which may represent a partial cluster structure of the FeMo cofactor.<sup>5</sup> A cuboidal  $[\text{Mo}_2\text{Fe}_2\text{S}_3\text{O}]$  cluster synthesized by Coucouvanis et al. is also relevant to part of the FeMo-cofactor cluster core.<sup>6</sup> Holm and co-workers have isolated  $[\text{M}_2\text{Fe}_6\text{S}_9]$  clusters (M = Mo, V, or W), which are topologically analogous to the P-cluster  $[\text{Fe}_8\text{S}_7]$  core of nitrogenase.<sup>7</sup> Previously, we have synthesized  $[\text{Fe}_8\text{S}_7]$  clusters reproducing the P-cluster core,<sup>8</sup> and recently we have discovered that the P-cluster core can be prepared via the reductive fusion of two all-ferric  $[\text{Fe}_4\text{S}_4]$  clusters.<sup>9</sup> We have also reported that the reaction of  $[(\text{TipS})\text{Fe}]_2(\mu\text{-SDmp})_2$  (Tip = 2,4,6-<sup>i</sup>Pr<sub>3</sub>C<sub>6</sub>H<sub>2</sub>, Dmp = 2,6-(Mes)<sub>2</sub>C<sub>6</sub>H<sub>3</sub>, and Mes = mesityl) or  $(\text{DME})\text{Fe}(\text{Mes})(\text{SDmp})$  (DME = 1,2-dimethoxyethane) with  $\text{S}_8$  in toluene generates the  $[\text{Fe}_8\text{S}_7]$  clusters  $[(\text{DmpS})\text{Fe}_4\text{S}_3]_2(\mu\text{-SDmp})_2(\mu\text{-SAr})(\mu_6\text{-S})$  (Ar = Tip or Mes).<sup>10</sup> These clusters consist of two incomplete cubane  $[\text{Fe}_4\text{S}_3]$  units connected by a six-coordinated sulfur atom and are similar to the FeMo cofactor in that the trigonal-prismatic cage of the six central iron atoms embraces a sulfur atom. In this study, we have attempted to synthesize  $[\text{Fe}_8\text{S}_x]$  clusters with a light atom relevant to X in the middle, modeling topologically the FeMo-cofactor core structure. Herein, we report the reaction of a new precursor,  $[(\text{Ph}_3\text{CO})\text{Fe}]_2(\mu\text{-SDmp})_2$  (**1**), with  $\text{S}_8$  and  $\text{H}_2\text{O}$  in toluene to produce the oxygen-incorporated  $[\text{Fe}_8\text{S}_6\text{O}]$  cluster  $[(\text{DmpS})\text{Fe}_4\text{S}_3\text{O}][(\text{DmpS})\text{Fe}_4\text{S}_3](\mu\text{-SDmp})_2(\mu\text{-OCPh}_3)$  (**2**; Scheme 1).

Precursor **1** was synthesized using a modification of the procedure for iron(II) thiolate complexes of the type  $\text{Fe}(\text{SAr})(\text{SAr}')$  or  $[(\text{ArS})\text{Fe}]_2(\mu\text{-SAr}')$  (Ar, Ar' = bulky aryl groups).<sup>10a,11</sup> Treatment of **1** with  $\text{S}_8$  and  $\text{H}_2\text{O}$  (24 mM in toluene) in the ratio 4:1/2:1 ( $1/\text{S}_8/\text{H}_2\text{O}$ ) in toluene resulted in the formation of a dark-yellowish-brown solution, which gradually turned dark brown over a few days. The reaction mixture was subjected to size-exclusion high-performance liquid chromatography (HPLC), eluting with toluene under dinitrogen, and the first fraction was collected. The black solid obtained therefrom was extracted with  $\text{C}_6\text{H}_6/\text{Et}_2\text{O}$ , from which black crystals were obtained. According to X-ray crystallo-

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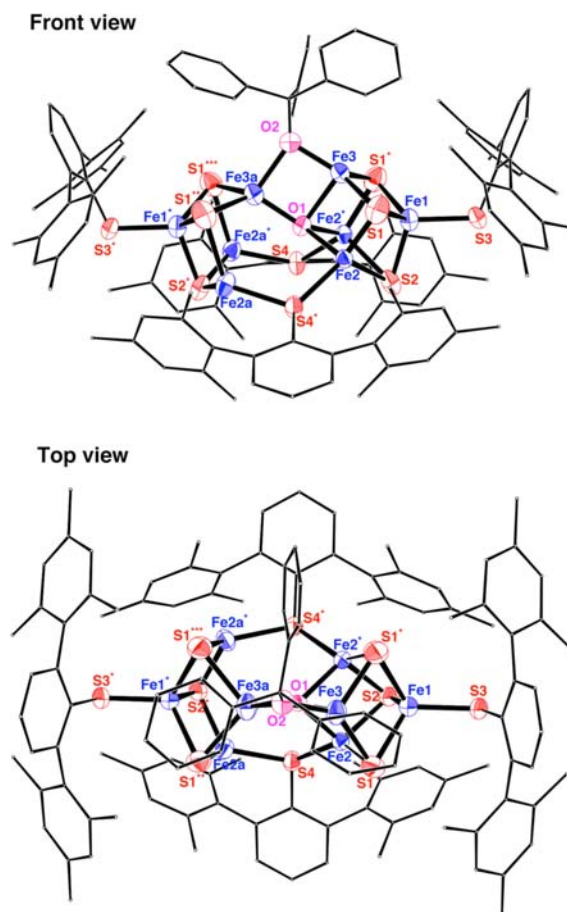
Scheme 1



graphic analysis, the crystals often show compositional disorder of two clusters: one is the  $[\text{Fe}_8\text{S}_6\text{O}]$  cluster **2**, and the other is the sulfur-centered  $[\text{Fe}_8\text{S}_7]$  cluster  $[(\text{DmpS})\text{Fe}_4\text{S}_3]_2(\mu\text{-SDmp})_2(\mu\text{-OCPh}_3)(\mu_6\text{-S})$  (**3**). The latter cluster **3** is an analogue of the  $[\text{Fe}_8\text{S}_7]$  clusters, which we reported previously.<sup>10</sup> The occupancy ratio of **2** and **3** in the crystals varies from 75:25 to 100:0 (**2/3**). Thus, the black crystals may contain both clusters **2** (dominant) and **3** (trace). The yield of black crystals remains low (7%) because of the high solubility of **2** in  $\text{C}_6\text{H}_6/\text{Et}_2\text{O}$ .

Here we describe the structural characterization of **2** obtained from a crystallographically pure sample. This cluster is located at a Wyckoff site *g* of space group  $P4_2/mnm$  (No. 136) at the intersection of two mirror planes. As a result, the cluster is divided into four symmetry-equivalent sites. The structure of **2** revealed two half-occupied oxygen positions disordered across a mirror plane located between two incomplete  $\text{Fe}_4\text{S}_3$  cubes. In addition, the iron atoms at the inner positions, Fe2 and Fe3, are also found to be split into two positions, each in a 1:1 ratio. An ORTEP drawing of **2** is depicted in Figure 2, where only one of the two components is shown for clarity. The  $[\text{Fe}_8\text{S}_6\text{O}]$  core of **2** is comprised of an incomplete-cuboidal  $[\text{Fe}_4\text{S}_3]$  and a cuboidal  $[\text{Fe}_4\text{S}_3\text{O}]$  unit. A  $\mu\text{-OCPh}_3$  and two  $\mu\text{-SDmp}$  ligands bridge the two units, which are also linked by an  $\text{Fe3a-O1}$  bond. Although there are six iron atoms around O1, only four are at bonding distances [1.910(6)–2.190(5) Å], and the two remaining distances [ $\text{Fe2a-O1}/\text{Fe2a}^*\text{-O1}$ , 3.361(5) Å] indicate that O1 does not form chemical bonds with these iron centers. Therefore, the central oxygen atom (O1) is  $\mu_4$ -ligated.

The coordination geometries of all iron centers except for Fe3 are close to tetrahedral. At first glance, Fe2a and Fe2a\* seem to be three-coordinate. However, upon closer inspection, we see that these iron atoms interact with the mesityl rings of the  $\mu\text{-SDmp}$  ligands with a shortest Fe–C distance of 2.505(2) Å, to complete their tetrahedral coordination. This interaction allows Fe2a and Fe2a\* to stay far from the central oxygen, and the Fe2a–Fe2a\* distance is very long [3.609(2) Å]. The



**Figure 2.** Molecular views of **2** with thermal ellipsoids of iron, sulfur, and oxygen atoms at the 50% probability level. Only one of the two orientations is shown for clarity.

geometry of Fe3 is trigonal-bipyramidal, and the iron atom lies in the plane defined by two sulfur atoms and the oxygen atom of the  $\mu\text{-OCPh}_3$  ligand (S1, S1\*, and O2) with a deviation of only 0.026(2) Å. The central oxygen O1 is coordinated to Fe3 from below the S1–S1\*–O2 plane, and a phenyl group of  $\mu\text{-OCPh}_3$  is located above this plane with a short Fe3–C distance of 2.6747(4) Å. An agostic C–H...Fe interaction may be present because the phenyl group does not face Fe3 but rather orients one of the C–H bonds toward Fe3.

Although both cluster **2** and the FeMo cofactor have a light atom in the middle, the bridging modes of the central atoms are different. The oxygen O1 of **2** is  $\mu_4$ -ligated, while X of the FeMo cofactor is bound to six iron atoms. In the case of **2**, two inner iron atoms move away from the central oxygen O1 and interact with the mesityl rings. This coordination mode models the interaction of the core iron atoms in the FeMo cofactor, binding to substrates such as dinitrogen, hydrazines, and alkynes. Interestingly, one of the inner iron atoms of the FeMo cofactor has been suggested to accommodate the substrates, based on spectroscopic studies of mutants of the MoFe protein<sup>12</sup> and on theoretical studies.<sup>13</sup> On the other hand, there is no obvious substrate binding site in the reported structure of the FeMo cofactor, because all metal atoms appear to be coordinatively saturated, in that all iron atoms are tetrahedral and the molybdenum is octahedral. The observed structure has been suggested to represent a resting state of the FeMo cofactor, and according to kinetic studies, the cluster may be

reduced by three or four electrons before it binds dinitrogen.<sup>14</sup> This increase of the electron density within the cluster in the reduced form may facilitate Fe–X bond cleavage to generate a substrate binding site. In this regard, an important role of the light atom X may be to allow opening of the FeMo-cofactor structure without breaking apart. This flexibility of the cluster is illustrated by comparing the structures of **2** and **3** and analogous Fe<sub>8</sub>S<sub>7</sub> clusters, which have a central sulfur atom. In the tight structure of **3**, all six of the central iron atoms are four-coordinate tetrahedral. This contrasts with the more open structure of **2**, where two of the iron atoms move away from the central oxygen atom, as well as the iron atoms in the other cube, and instead initiate interaction with the mesityl rings.

In summary, we have synthesized a novel [Fe<sub>8</sub>S<sub>6</sub>O] cluster, **2**, from the reaction of **1**, S<sub>8</sub>, and H<sub>2</sub>O. X-ray analysis of **2** reveals an unusual Fe–S cluster structure with an oxygen atom unsymmetrically bridging two incomplete Fe<sub>4</sub>S<sub>3</sub> cubes. Two iron atoms interact with the mesityl rings, supporting the concept<sup>15</sup> that an inner iron atom of the FeMo cofactor may dissociate from the central atom X, allowing an external substrate to bind to the iron. This result also suggests a possible, but subtle, role for the molybdenum atom in the FeMo-cofactor structure. Without molybdenum in **2**, the central oxygen atom can equally move to either proto-Fe<sub>4</sub>S<sub>3</sub> cube, while the presence of molybdenum may favor association of the oxygen atom with one cube, rendering the other cube coordinatively unsaturated and suitable as the substrate binding site. Further, if cluster **2** represents the active-site structure of the all-iron nitrogenase,<sup>16,17</sup> the above argument implies a decrease of at least 50% in the activity of the all-iron nitrogenase compared to the more common molybdenum form.

## ■ ASSOCIATED CONTENT

### ■ Supporting Information

Synthesis and crystal structure determinations for **1** and **2** and a mixture of **2** and **3** and a CIF file of the X-ray crystallographic data for **1** and **2** and a mixed crystal of **2** and **3**. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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### Notes

The authors declare no competing financial interest.

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