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Synthesis and Characterization of a Binuclear Iron(III) Complex Bridged by 1-Aminocyclopropane-1-carboxylic Acid. Ethylene Production in the Presence of Hydrogen Peroxide

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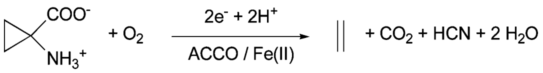
A μ-oxo-diiron(III) complex bridged by two molecules of 1-aminocyclopropane-1-carboxylic acid (ACCH) was prepared with the ligand 1,4,7-triazacyclononane (TACN): $[(TACN)Fe_2(\mu\text{-}O)(\mu\text{-}ACCH)_2](ClO_4)_4 \cdot 2H_2O$ (**1**). This complex was characterized, and its crystal structure was solved. The bridging amino acid moieties were found in their zwitterionic forms (noted as ACCH). Reactivity assays were performed in the presence of hydrogen peroxide, and **1** turned out to be the first example of a well-characterized iron−ACCH complex able to produce ethylene from the bound ACCH moiety. The reaction requires the presence of a few equivalents of base, probably involved in the deprotonation of the amine groups of the ACCH bridges.

Ethylene is a plant hormone involved in the control of a wide range of physiological processes. The last step of ethylene biosynthesis is catalyzed by ACC oxidase, which oxidizes 1-aminocyclopropane-1-carboxylic acid (ACCH) into ethylene in the presence of ferrous ions, dioxygen, and ascorbate. The products are ethylene, cyanhydric acid, and carbon dioxide (Scheme 1).[1]

The crystallographic structure of ACC oxidase from *Petunia hybrida* reveals an active site composed of a single ferrous ion coordinated to a 2-histidine-1-aspartate facial triad.[2] The catalytic mechanism of ACC oxidase remains the object of debates.[3] Spectroscopic studies have suggested that, during the first steps of the reaction, both ACC and dioxygen are coordinated to the iron(II) ion, generating an iron(III) superoxo intermediate. It has been proposed that the substrate, ACC, is ligated in a bidentate η^2-N,O-coordination mode.[4] There are only a few well-characterized metal−ACC(H) complexes and, to our knowledge, no iron−ACC(H) complex has been reported.[5,6] In the present paper, we describe the characterization and reactivity of a μ-oxo-diiron(III) complex bridged by two zwitterionic ACCH moieties that was obtained with the ligand 1,4,7-triazacyclononane (TACN).

Scheme 1. Reaction Catalyzed by ACC Oxidase



The complex $[Fe_2(TACN)_2(\mu\text{-}O)(\mu\text{-}ACCH)_2](ClO_4)_4 \cdot 2H_2O$ (**1**) was prepared by mixing the TACN ligand, $Fe(ClO_4)_3 \cdot xH_2O$, and ACCH in methanol (see the Supporting Information for experimental details). Suitable crystals for X-ray diffraction analysis were obtained, and the ORTEP drawing of the cation part of the complex is shown in Figure 1. The structure reveals two iron(III) ions in distorted octahedral geometries with facially coordinated tridentate TACN ligands and bridged by one oxo and by the carboxylate functions of two amino acid moieties. Moreover, the H atoms on the amino acid moieties were determined experimentally, and ACCH is found in its zwitterionic protonated

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10.1021/ic9001626 CCC: $40.75

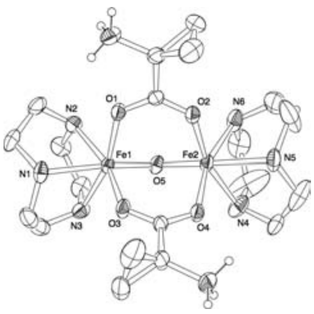

Figure 1. ORTEP drawing of the cation $[Fe_2(TACN)_2(\mu\text{-}O)(\mu\text{-}ACCH)_2]^{4+}$. Structural data: Fe1−O5 = 1.782(3) Å, Fe2−O5 = 1.787(3) Å, Fe1−Fe2 = 3.113 Å, and Fe1−O5−Fe2= 121.43(16)°. H atoms were omitted for clarity, except for those of the nitrogen from ACCH.

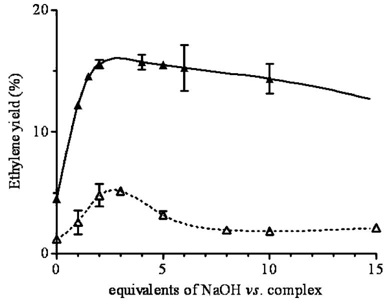

Figure 2. Ethylene production yields at 20 °C in methanol as a function of NaOH added (1 equiv = 5×10^{-4} M) by (−▲−) a 5×10^{-4} M solution of **1** and (--△--) a standard solution composed of 1 mM $\{Fe(NO_3)_3 \cdot 9H_2O$ + ACCH$\}$. Ethylene was measured 30 min after the addition of 5×10^{-2} M hydrogen peroxide.

form. Four perchlorate anions are visible in the structure (not shown). One of these perchlorates was found to be disordered and was refined in two different positions. The diiron core is symmetric with very similar Fe1−O5 and Fe2−O5 bonds (1.782 and 1.787 Å, respectively) and $Fe-O_{carboxylate}$ ranging from 2.048 to 2.058 Å. The Fe−N bonds located trans to the oxo bridge are slightly longer (Fe1−N1 and Fe2−N5 = 2.204 and 2.200 Å, respectively) than the other Fe−N bonds (2.139−2.150 Å) in the complex as a result of the trans effect of the oxo bridge. The Fe1−Fe2 distance is 3.113 Å, and the Fe1−O5−Fe2 angle is 121.4°. There are only a limited number of reported amino acid-bridged diiron(III) complexes. The few rare binuclear complexes were obtained in the group of Umakoshi, who described the synthesis and crystallographic structure of oxo-bridged binuclear complexes in which the iron ions were also bridged by the carboxylate function of various zwitterionic amino acids (AAH).[7,8] The structure of **1** is similar to the ones of TACN-based diiron(III) complexes obtained with L-valine and L-proline and to those of other (μ-oxo)bis(μ-carboxylato)diferric complexes.[7,9]

The magnetic behavior of **1** was recorded as a function of the temperature using a SQUID Quantum Design MPMS 5. Upon cooling, both χ_M and the product $\chi_M T$ continuously decrease, indicating an antiferromagnetically coupled high-spin iron(III) pair and thus a diamagnetic singlet ($S = 0$) ground state.[10] Experimental data were simulated by setting $g = 2$ and gave $J = -261$ cm^{-1} ($H = -JS_1 \cdot S_2$; $S_1 = S_2 = {}^5/_2$). This constant is in good agreement with the coupling constants determined for other μ-oxo(μ-amino acid)- or μ-oxo-μ-carboxylato-diiron(III) complexes and with the quantitative magnetostructural correlation reported by Gorun and Lippard.[11,12]

Ethylene formation was measured in 0.5 mM methanolic solutions of **1** in the presence of hydrogen peroxide. The solution was placed in a hermetically sealed tube at 20 °C, and then hydrogen peroxide was injected through the septum. Ethylene was measured by removing 1 mL of the headspace gas with a gastight syringe and injecting the sample into a gas chromatograph, as was previously described.[5] Ethylene was produced from complex **1** under these experimental conditions. The conversion of ACCH into ethylene strongly depends on the equivalents of base, and ethylene production was measured as a function of NaOH added (Figure 2). When no base is added to complex **1**, less than 5% of the bound ACCH is converted into ethylene. ACCH conversion reaches a maximum of ca. 16% after the addition of 2−3 equiv of NaOH and then slowly diminishes as base is further added. The addition of a large excess of base leads to discoloration of the initial solution as well as to a drastic drop of activity, probably due to the decomposition of **1** in a very basic medium. These results were compared to the ethylene production of a standard solution composed of an iron(III) nitrate salt at the same concentration (1 mM) in the presence of an equimolar amount of ACCH. With this standard solution, ACCH conversion into ethylene reaches ca. 4% in the presence of a few equivalents of base. Thus, ethylene production is more than 4-fold lower than that with complex **1**. Moreover, it was verified that free ACCH hardly produces ethylene in the same conditions.

There are only few reported functional models of ACC oxidase, and even less using iron. The group of Nishida described functional models of ACC oxidase using dinucleating ligands and several transition-metal ions.[13] These complexes require a large excess of hydrogen peroxide to convert ACCH into ethylene, and no metal−ACC(H) interaction was characterized. Furthermore, the best iron(III) complex gave a very low conversion yield (~0.01% vs iron complex) even in a basic medium with excess ACCH and hydrogen peroxide, which is 3200-fold lower than the one obtained with complex **1**.

A few equivalents of base are necessary to reach optimum activity. The addition of base on complex **1** was monitored by UV−visible spectroscopy. The UV−visible spectrum of

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1 in methanol consists of intense near-UV features (232, 335, and 376 nm), weaker bands around 400−550 nm (422, 472, 492, 512, and 550 nm), and a weak band found at 760 nm. This spectrum is very comparable to those obtained for similar μ-oxo(μ-amino acid)diiron(III) and μ-oxo-μ-carboxylato-diiron(III) complexes.[7,9] When a few equivalents of base is added, an increase of the spectral features around 400−500 nm is observed, as well as a small increase of the absorption band at 760 nm (see the Supporting Information). The electronic spectra of (μ-oxo)diiron(III) complexes have been shown to be quite sensitive to structural variations around the diiron core.[14] These results thus indicate that complex **1** is slightly modified by the addition of 2−3 equiv of base but that the diiron core is maintained. The addition of a further amount of base leads to the loss of the spectral characteristics of the diiron core and thus confirms that complex **1** is decomposed in a very basic medium.

The role of the base can be rationalized thanks to the work of Umakoshi et al., who have studied the redox properties of various μ-oxo-diiron(III) complexes bridged by one zwitterionic amino acid unit (noted AAH).[8] The authors have described a splitting of the reduction process to generate mixed-valent $Fe^{III}Fe^{II}$ species. This splitting is not observed for other μ-oxo-diiron(III) complexes that mostly display reversible one-electron reductions, and it rather seems to be typical for AAH-bridged complexes.[9,14] Umakoshi et al. have shown that it is probably due to the presence of two species in solution, (μ-O)$Fe_2^{III,III}$(μ-AAH) and (μ-O)$Fe_2^{III,III}$(μ−AA), that display different redox behavior. To verify whether **1** displays similar redox behavior or not, electrochemical experiments were conducted in the presence of Et_3N.[15] The observed cyclovoltammograms in the absence or in the presence of Et_3N are comparable to the ones described by the group of Umakoshi with a splitting of the reduction process ($E^{(I)} = -610$ mV and $E^{(II)} = -970$ mV vs SCE). The processes appear to be irreversible, and the interpretation was difficult because **1** contains an additional amino acid bridge. The reduction potentials are also strongly shifted toward the negative range as compared to the ones observed by Umakoshi et al., which is coherent with an additional negative charge (carboxylato bridge of the ACCH moiety) at the diiron core. The electrochemical studies thus suggest that the amines of the ACCH moieties are partially protonated in solution. It is admitted that the conversion of ACCH into ethylene by ACCO proceeds via the oxidation of the amine group to generate a radical cation.[16] Therefore, in the present case, the base is probably involved in the deprotonation of the amine groups prior to their oxidation. The base could also be partly involved in hydrogen peroxide deprotonation to facilitate its interaction with the complex.

The reaction of **1** with hydrogen peroxide in the presence of a few equivalents of base was monitored by UV−visible spectroscopy. At the end of the reaction, the spectral characteristics of **1** and of the diiron core are still present with lower intensity, indicating that part of the unreacted complex is intact. The rest is probably present as a mixture of iron species. A further addition of hydrogen peroxide leads to a further decrease in the intensity of the spectral features of **1**. This is followed by a small additional ethylene production, but it seems that supplementary hydrogen peroxide mostly leads to decomposition of **1** without ethylene evolution.

So far, many synthetic carboxylate-bridged complexes have been described as models for diiron-containing metalloproteins.[17] The reaction of diiron(III) complexes with hydrogen peroxide and the reaction of diiron(II) complexes with dioxygen are well documented and have led to the characterization of several intermediates.[18] In the present case, however, no intermediate could be detected even at low temperature. Although not fully representative of ACCO's active site, this complex is the first well-characterized example of an iron−ACCH complex displaying ethylene production activity. Efforts will be directed toward the identification of reaction intermediates and the understanding of the catalytic mechanism.

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Supporting Information Available: Experimental details for the synthesis of complex **1**, magnetic data, and a CIF file for complex **1** (CCDC 706176). This material is available free of charge via the Internet at http://pubs.acs.org.

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