

A μ -Oxo Mixed-metal Complex of Copper(II) and an Iron(III)-porphyrin as a Resting State Model for the Cytochrome c Oxidase Active Site¹

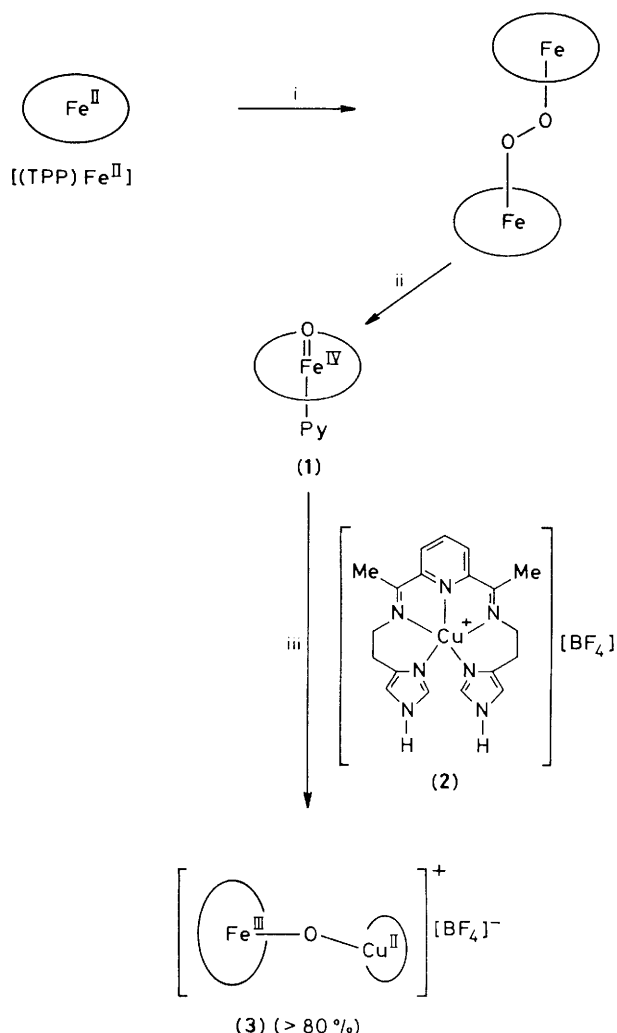
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The complex $(\text{TPP})\text{Fe}^{\text{IV}} : \text{O}$ (TPP = tetraphenylporphyrin) reacts in solution with an imidazole-bearing Cu^{I} complex to form a stable mixed-metal μ -oxo-species, $[\text{Fe}^{\text{3+}}-\text{O}-\text{Cu}^{\text{2+}}]^+$, as a resting state model compound for the binuclear active site of cytochrome c oxidase.

Aerobic organisms utilize dioxygen (O_2) for respiration and metabolism in various ways. Of the metalloenzymes that

activate the O-O bond, cytochrome c oxidase is the most complex, containing two heme (Cyt a and Cyt a₃) and two



Scheme 1. Reagents and conditions: i, CH_2Cl_2 , (a) -78°C , (b) O_2 ; ii, pyridine (py); iii, MeCN, argon purge.

copper (Cu_{T} and Cu_{I}) centres per functioning unit.² Spectroscopic measurements have indicated that the active site (where oxidase catalyses the reaction $\text{O}_2 + 4\text{H}^+ + 4\text{e}^- \rightarrow 2\text{H}_2\text{O}$) is binuclear in Cyt a_3 and Cu_{I} .³ Furthermore, for the fully-oxidized or resting state of the enzyme, full-temperature magnetochemical measurements have indicated that the $(\text{Cyt } a_3)^{3+}$ ($S = 5/2$) and $\text{Cu}_{\text{I}}^{2+}$ ($S = 1/2$) sites are strongly coupled *antiferromagnetically* ($-J \geq 200 \text{ cm}^{-1}$) to give a resultant $S = 2$ ground state.^{4,5} Such strong coupling implies a $[(\text{Cyt } a_3)^{3+}(\text{B})-\text{Cu}_{\text{I}}^{2+}]$ structure where (B) is a biochemically feasible bridge such as an imidazolato-group from histidine,^{4,6} an oxo-bridge from O_2 ,⁷⁻⁹ or perhaps a mercapto-bridge from cysteine.¹⁰ In biomimicry studies to elucidate further the mechanism and active site structure of oxidase, we^{7,11} and others¹²⁻¹⁴ have initially focused only on the synthesis and study of $[\text{Fe}(\text{B})-\text{Cu}]$ species where (B) is imidazolato and related ligands. In this work, we report the synthesis, isolation, and characterization of the first such case where (B) is an oxo-bridge originating from O_2 .

LaMar and Balch¹⁵ first reported the strong oxidizing power of $(\text{TPP})\text{Fe}^{\text{IV}}:\text{O}$ (TPP = tetraphenylporphyrin), an iron-

porphyrin complex derived from *meso*-tetraphenylporphyrin and O_2 [(1) in Scheme 1]. When the copper(I) complex, (2), which has been previously shown to bind O_2 reversibly under ambient conditions in solution,^{16,17} is treated with $(\text{TPP})\text{Fe}^{\text{IV}}:\text{O}$, a new compound of empirical formula $\text{FeCuC}_{63}\text{H}_{51}\text{N}_{11}\text{OBF}_4$ is isolated in $>80\%$ yield. From its elemental analysis, univalent electrolyte behaviour in MeCN ($\Lambda = 154.4 \mu\text{S l cm}^{-1} \text{ mol}^{-1}$), and its spectroscopic and magnetic properties (*vide infra*), this new species has been characterized as being the $[\text{Fe}^{3+}-\text{O}-\text{Cu}^{2+}] [\text{BF}_4]$ compound (3) shown in Scheme 1 [which also gives the required synthetic conditions for (3)].

Lacking direct structural evidence, one of the most definitive methods for establishing the presence of the oxo-bridge in (3) is by ^{18}O replacement of the mainly ^{16}O bridge by using naturally-occurring O_2 , and observing the consequential shift of the $[\text{Fe}^{3+}-\text{O}-\text{Cu}^{2+}]^+$ asymmetric stretching frequency in the i.r. spectrum. For $[\text{Fe}^{3+-16}\text{O}-\text{Fe}^{3+}]$ in $[(\text{TPP})\text{Fe}^{\text{III}}]_2$ ^{16}O this asymmetric stretching mode reportedly occurs as a doublet at 890.2 and 873.8 cm^{-1} ,¹⁸ while for $[\text{Fe}^{3+-18}\text{O}-\text{Fe}^{3+}]$ in $[(\text{TPP})\text{Fe}^{\text{III}}]_2$ ^{18}O we found that the main spectral change is the appearance of a new absorption band at 831.3 cm^{-1} [Figure 1(a)] (the other half of the doublet is probably masked by strong neighbouring absorptions due to the porphyrin ring). Figure 1(b) details the i.r. spectra for $[\text{Fe}^{3+-16}\text{O}-\text{Cu}^{2+}] [\text{BF}_4]$ and $[\text{Fe}^{3+-18}\text{O}-\text{Cu}^{2+}] [\text{BF}_4]$ (from $>90\%$ $^{18}\text{O}_2$) as Nujol mulls in the region of interest. By analogy with $[(\text{TPP})\text{Fe}^{\text{III}}]_2\text{O}$, the absorptions at 890.2 and 872.8 cm^{-1} in the $[\text{Fe}^{3+-16}\text{O}-\text{Cu}^{2+}]^+$ spectrum can be assigned to the asymmetric stretching mode, although they are $10 \times$ less intense than those of $[(\text{TPP})\text{Fe}^{\text{III}}]_2\text{O}$. However, in the $[\text{Fe}^{3+-18}\text{O}-\text{Cu}^{2+}]^+$ spectrum, these peaks are absent and there is instead a new absorption at 880.5 cm^{-1} ; the complementary peak to that at 880.5 cm^{-1} is most probably obscured by the intense band at 847.7 cm^{-1} , which also accounts for the increased absorbance in that area of the spectrum. This interpretation gives the expected shift to lower energy, paralleling that of the $[\text{Fe}^{3+}-\text{O}-\text{Fe}^{3+}]$ system. In any case, from comparison of the $[\text{Fe}^{3+-16}\text{O}-\text{Cu}^{2+}]^+$ and $[\text{Fe}^{3+-18}\text{O}-\text{Cu}^{2+}]^+$ spectra it is readily apparent that there are differences that can be attributed to isotopic substitution in an oxo-bridging structure.

Physical measurements of (3) have shown it to have a Curie-like temperature-dependent magnetic susceptibility, with μ_{eff} ranging from $4.23 \mu_{\text{B}}$ at 298 K to $3.95 \mu_{\text{B}}$ at 10 K . Furthermore, the complex is e.s.r. silent at 10 K , with an ^{57}Fe Mössbauer spectrum consisting of a doublet with $\delta(\text{Fe}^0) = 0.23 \text{ mm s}^{-1}$ and $\Delta E_{\text{Q}} = 2.06 \text{ mm s}^{-1}$ at 136 K . Taken together, these results indicate strong *antiferromagnetic* coupling through the oxo-bridge between iron(III) and copper(II) to give a ground state with $S = 1$ characteristics.† Thus, the synthetic $[\text{Fe}^{3+}-\text{O}-\text{Cu}^{2+}]^+$ centre in (3) correctly mimics resting oxidase in that strong *antiferromagnetic* coupling is apparently present, but differs from the enzyme in that the coupled resultant ground state appears as $S = 1$ in nature rather than $S = 2$.‡ Of course, the present $S = 1$ spin state is also e.s.r. silent, like the enzyme situation where $S = 2$.

Finally, the u.v.-visible spectrum of (3) resembles that of the $[(\text{TPP})\text{Fe}^{\text{III}}]_2\text{O}$ dimer in that it has a strong absorption in

† These magnetic and spectroscopic results compare favourably with a well characterized $S = 1$ tetraphenylporphyrin complex, $[\text{TPP}\text{Fe}^{\text{II}}]$, as reported by Collman, *et al.*, (J. P. Collman, J. L. Hoard, N. Kim, G. Lang, and C. A. Reed, *J. Am. Chem. Soc.*, 1975, 97, 4344), for which the Mössbauer parameters are $\delta(\text{Fe}^0) = 0.51 \text{ mm s}^{-1}$ and $\Delta E_{\text{Q}} = 1.52 \text{ mm s}^{-1}$ at 77 K and $\mu_{\text{eff}}(\text{RT}) = 4.3 \mu_{\text{B}}$.

‡ In the present $[\text{Fe}^{3+}-\text{O}-\text{Cu}^{2+}]$ case, the $S = 1$ ground state is taken to arise from strong *antiferromagnetic* coupling between Fe^{3+} ($S = 3/2$) and Cu^{2+} ($S = 1/2$).

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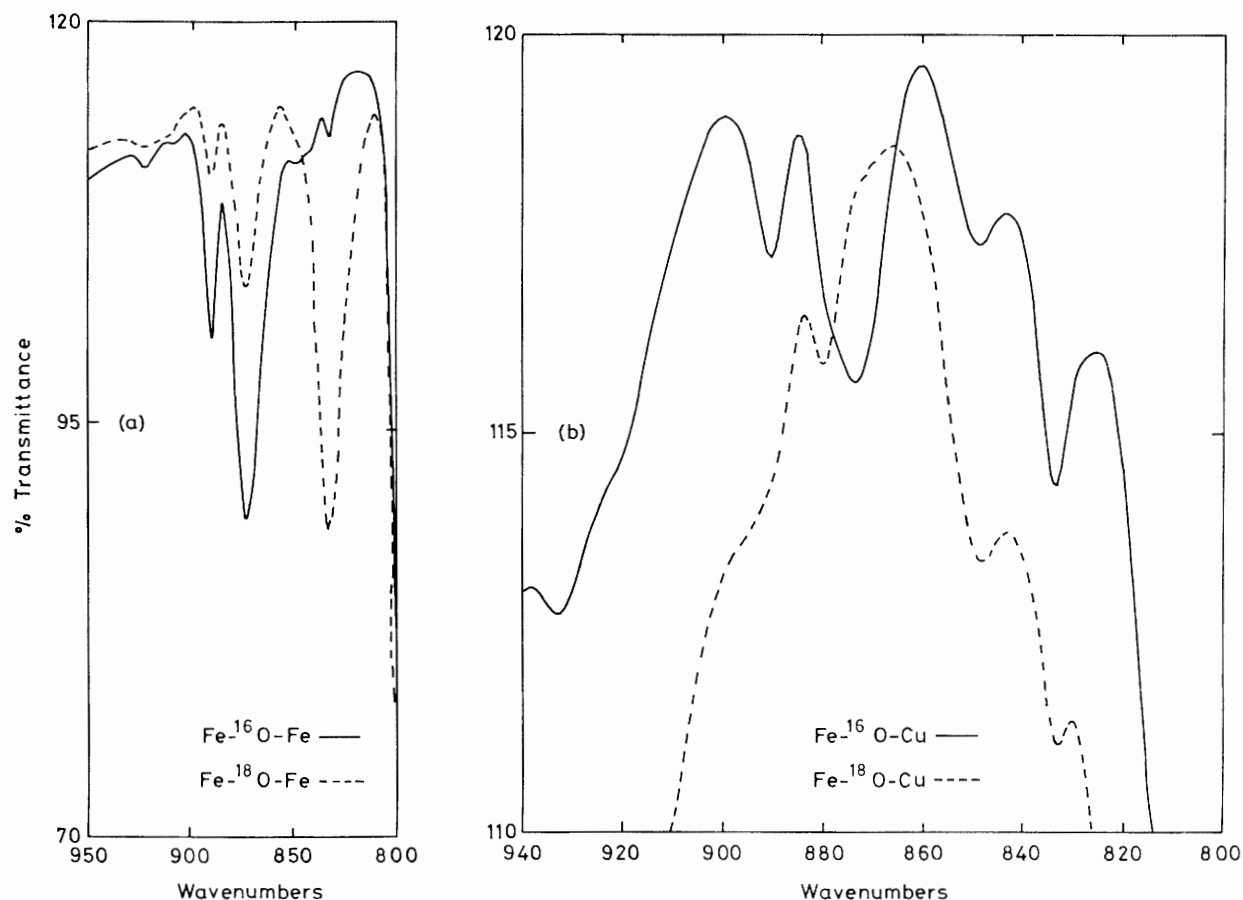


Figure 1 F.t.-i.r. absorption spectra for (a) $[\text{Fe}^{3+}\text{-}^{16}\text{O-Fe}^{3+}]$ and $[\text{Fe}^{3+}\text{-}^{18}\text{O-Fe}^{3+}]$ in $[(\text{TPP})\text{Fe}]_2\text{O}$ and (b) $[\text{Fe}^{3+}\text{-}^{16}\text{O-Cu}^{2+}]^+$ and $[\text{Fe}^{3+}\text{-}^{18}\text{O-Cu}^{2+}]^+$ in (3) as Nujol mulls ($800\text{--}940\text{ cm}^{-1}$ region).

the Soret region at 413 nm ($\epsilon = 1.22 \times 10^5$) with two other absorbances in the visible region at 552 nm ($\epsilon = 1.30 \times 10^4$) and 583 nm ($\epsilon = 6.71 \times 10^3$).[†] Even though $[(\text{TPP})\text{Fe}^{111}]_2\text{O}$ would probably be a major impurity in (3), absorbances in the visible spectrum at 571 and 610 nm corresponding to $[(\text{TPP})\text{Fe}^{111}]_2\text{O}$ are totally absent.

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[†] The electronic spectrum of $[(\text{TPP})\text{Fe}^{111}]_2\text{O}$ is well known, with λ_{max} at 408 nm ($\epsilon = 8.81 \times 10^4$), 571 nm ($\epsilon = 7.64 \times 10^3$), and 610 nm ($\epsilon = 3.47 \times 10^3$).