

The bioinorganic chemistry of iron in oxygenases and supramolecular assemblies

John T. Groves*

Department of Chemistry, Princeton University, Princeton, NJ 08544

The bioinorganic chemistry of iron is central to life processes. Organisms must recruit iron from their environment, control iron storage and trafficking within cells, assemble the complex, iron-containing redox cofactors of metalloproteins, and manage a myriad of biochemical transformations by those enzymes. The coordination chemistry and the variable oxidation states of iron provide the essential mechanistic machinery of this metabolism. Our current understanding of several aspects of the chemistry of iron in biology are discussed with an emphasis on the oxygen activation and transfer reactions mediated by heme and nonheme iron proteins and the interactions of amphiphilic iron siderophores with lipid membranes.

That an iron-containing enzyme mediated the activation and transfer of molecular oxygen into its substrate was first demonstrated by Hayaishi *et al.* (1) in the 1950s. It was shown, in some of the first mechanistically informative oxygen isotopic measurements, that both of the inserted oxygen atoms in the conversion of catechol to cis-muconic acid derived from O₂ and not water. These findings challenged the then-firmly held view that oxygen in biomolecules was derived exclusively from water via hydration processes. The biosynthesis of cholesterol and its precursor, lanosterol, from the hydrocarbon squalene were also shown to derive their oxygen functionality from molecular oxygen (2). Here, a single oxygen atom derived from molecular oxygen while the other was transformed to water. Later, the prostaglandins were shown to derive from the incorporation of two molecules of oxygen to form, initially, an alkyl hydroperoxide-endoperoxide. Thus, what appeared at first to be an obscure process of bacteria and fungi became recognized as a major theme of aerobic metabolism in higher plants and animals. The subsequent search for “active oxygen species” and efforts to elucidate and understand the molecular mechanisms of oxygen activation and transfer have been richly rewarding. The roles of iron in these wonderfully varied processes have been a major force in the development of both bioinorganic chemistry and chemical catalysis over the past three decades. Novel and unusual iron redox chemistry has appeared as our understanding of biological iron acquisition, transport and storage, and enzymatic oxidation strategies has developed. The goal of this perspective is to discuss several aspects of the current state of bioinorganic chemistry relating to iron and our understanding of the chemical pathways and mechanisms by which iron–oxygen systems function.

Oxygen Activation by Heme Proteins

The heme-containing metalloenzymes cytochrome P450 (3), chloroperoxidase

(CPO, refs. 4 and 5), NO synthase (NOS, ref. 6), and their relatives catalyze a host of crucial biological oxidation reactions. Highly specific P450s are involved in the selective oxygenations of steroid and prostaglandin biosynthesis. Myeloperoxidase, which is a CPO, is an integral part of the immune response, and NOS is the source of the highly regulated signal transducer NO. Certain fungal CPOs and bacterial P450s have been genetically engineered for large-scale biotransformations (7–10). The active sites of these three protein families, known in detail from a number of x-ray crystal structures (4, 11–13), are remarkably similar. All three have an iron-protoporphyrin IX center coordinated to a cysteine thiolate. All of them are oxidoreductases that activate molecular oxygen (O₂), in the cases of P450 and NOS, or hydrogen peroxide in the case of CPO, at the iron center and incorporate one of the oxygen atoms into a wide variety of biological substrates, with concurrent transformation of the other oxygen atom to H₂O. All three are proposed to initiate their chemistry through the oxidation of a resting iron(III) state (1) to a reactive oxoiron(IV)–porphyrin cation radical intermediate (2) (Fig. 1). A depiction of the CPO active site derived from the crystal structure of this protein from *Calderionomyces fumago* is shown in Fig. 5, which is published as supporting information on the PNAS web site, www.pnas.org. The structure, biochemistry, molecular biology, and the chemistry of cytochrome P450 and related model systems have been extensively reviewed (14–18).

Our understanding of the mechanism of action of these heme proteins comes from the direct observation of intermediates in the catalytic cycle through a variety of spectroscopic techniques, the use of diagnostic substrates with mechanistically revealing rearrangements during oxidation, and the parallel development of the chemistry of synthetic metalloporphyrins. The principal features of the consensus mechanism of cytochrome P450 (19) are as outlined in Scheme 1: binding of substrate to the enzyme, sometimes accompanied by a

spin-state change of the iron, to afford an enzyme-substrate adduct 3; reduction of the ferric cytochrome P450 by an associated reductase with an NADPH-derived electron to the ferrous cytochrome P450 4; binding of molecular oxygen to the ferrous heme to produce a ferrous cytochrome P450-dioxygen complex 5, similar to the situation in oxymyoglobin; a second one-electron reduction and protonation to arrive at the Fe(III)-hydroperoxy complex 6; protonation and heterolytic cleavage of the O—O bond in 6 with concurrent production of a water molecule to form a reactive iron-oxo intermediate 7; and, finally, oxygen atom transfer from this iron-oxo complex 7 to the bound substrate to form the oxygenated product complex 8. Product dissociation completes the cycle.

There were a number of important realizations in the course of elucidating this mechanism. That hydrogen peroxide, alkyl hydroperoxides, periodate, and iodosylbenzene were also functional with cytochrome P450 suggested that the chemistry of “oxygen activation” was the two-electron reduction of molecular oxygen to hydrogen peroxide and that, in analogy to the peroxidases, the active oxygen species was a ferryl (or oxene) complex Fe=O, formally iron(V). It was shown that a synthetic oxoiron(IV)porphyrin cation radical species could be formed at low temperature by the oxidation of an iron(III) precursor with peroxyacids (9 → 10) (20). Intermediate 10 did have the requisite reactivity to transfer an oxygen atom to hydrocarbon substrates. It is this oxygen atom transfer from the oxygen donor to form the Fe=O intermediate 7 and the subsequent oxygen transfer to form the substrate complex 8 that has been termed oxygen rebound. Such an iron-oxo species (compound I) has been observed for the CPO of *C. fumago* (21), but the active species of cytochrome P450 has remained elusive. Very recently, it has been shown that an intermediate with the spectral properties similar to those of CPO com-

*E-mail: jtgroves@princeton.edu.

PNAS

[HOME](#) [HELP](#) [FEEDBACK](#)

[SUBSCRIPTIONS](#) [ARCHIVE](#) [SEARCH](#)

[TABLE OF CONTENTS](#)

This Article

PDF version of:
Groves 100 (7): 3569. (2003)

- ▶ [Abstract](#) **FREE**
- ▶ [Full Text \(HTML\)](#)
- ▶ [Supporting Figure](#)
- ▶ [Alert me when this article is cited](#)
- ▶ [Alert me if a correction is posted](#)
- ▶ [Citation Map](#)

Services

- ▶ [Email this article to a colleague](#)
- ▶ [Similar articles in this journal](#)
- ▶ [Similar articles in ISI Web of Science](#)
- ▶ [Similar articles in PubMed](#)
- ▶ [Alert me to new issues of the journal](#)
- ▶ [Add to My File Cabinet](#)
- ▶ [Download to citation manager](#)
- ▶ [Request Copyright Permission](#)

Citing Articles

- ▶ [Citing Articles via CrossRef](#)
- ▶ [Citing Articles via ISI Web of Science \(75\)](#)
- ▶ [Citing Articles via Google Scholar](#)

Google Scholar

- ▶ [Articles by Groves, J. T.](#)
- ▶ [Search for Related Content](#)

PubMed

- ▶ [PubMed Citation](#)
- ▶ [Articles by Groves, J. T.](#)
- ▶ **Pubmed/NCBI databases**
 - [Compound via MeSH](#)
 - [Substance via MeSH](#)
- ▶ **Hazardous Substances DB**
 - [IRON](#)
 - [OXYGEN](#)

Related Content

- ▶ [Bioinorganic Chemistry Special Feature](#)

Social Bookmarking



[What's this?](#)

Help

Adobe Acrobat plugin users:

[View article in full window](#)

Printing problems?

To print a PDF using the Acrobat plugin, use the printer button in the plugin's toolbar, located immediately above the document.

[Download Adobe Reader](#)

Institution: Princeton University Library | [Sign In via User Name/Password](#)