Models of High-Valent Heme Protein Intermediates: A Quantum Chemical Study of Iron(IV) Porphyrins with Two Univalent Axial π-Bonding Ligands

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This is a quantum chemical survey of a broad class of high-valent transition metal intermediates relevant to the catalytic cycles of the cytochromes P_{450} and related heme proteins, viz. iron(IV) and manganese(IV) porphyrins with two univalent π-bonding axial ligands. The axial ligands in these intermediates invariably exhibit a bent geometry, reflecting a competitive interplay of σ- and π-bonding effects. Compared to an oxo ligand, the univalent ligands are distinctly less able to delocalize electronic spin density away from the metal center, which might have significant implications for the chemical reactivity of these species.

Introduction

Over the last three decades, high-valent iron intermediates have been recognized as the key components of the catalytic cycles of a variety of heme and nonheme iron enzymes, including peroxidases, catalases, cytochromes P450, soluble methane monoxygenase (sMMO), and ribonucleotide reductase.[1,2] Perhaps the best known of these intermediates are the peroxidase compound I intermediates, the majority of which are best described as $S = {}^3/_2$ iron(IV)−oxo porphyrin π-cation radical species.[3] Iron(IV)−oxo intermediates have also been proposed for mononuclear nonheme iron enzymes such as the pterin-dependent enzyme phenylalanine hydroxylase and the α-ketoglutarate-dependent enzyme prolyl hydroxylase.[4,5] A key recent development in this field has been the isolation[6,7] and, in one case even X-ray crystallographic characterization,[8] of synthetic nonheme iron(IV)−oxo intermediates. Extensive spectroscopic,[1−8] structural,[1−8] and quantum chemical[9−12] studies aimed at obtaining a detailed understanding of the electronic structures and chemical reactivities of these reactive intermediates are underway in many laboratories around the world.

In this context, one class of high-valent iron porphyrin intermediates has remained relatively little explored, viz. iron(IV) porphyrins with two univalent π-donating axial ligands such as alkoxide, thiolate, or hydroxide. Indeed, a single study by Groves and co-workers[13] on Fe(IV) porphyrin intermediates with two axial methoxide ligands provides most of the spectroscopic characterization available for such Fe(IV) complexes, although a few more studies provide additional relevant information.[14−16] Iron(IV) intermediates of this type are actually also of interest from a biochemical perspective. As shown in Figure 1, they are implicated as key intermediates in the so-called "rebound" mechanism of heme-based oxygenases,[17−19] which has received significant support from both theory and experiment.[17,18,20,21] Second, the protonation state of the axial oxo ligand of peroxidase compound I and particularly of compound II intermediates has long remained a somewhat unresolved and controversial issue. Thus, whereas an EXAFS study indicated a short FeO distance of about 1.64 Å,[22] a recent high-resolution X-ray crystallographic[23] study indicate an FeO distance of about 1.9 Å for HRP-II (HRP = horseradish peroxidase), compared with a distance of about 1.7 Å for HRP-I. These crystallographic results appear to be consistent with a protonated ferryl unit in HRP-II. Moreover, the Fe−O stretching frequency of HRP-II,[24] 776 cm^{-1} at pH 6.0 and 787 cm^{-1} at pH 11.0, is significantly lower than those of synthetic model compounds such as Fe^{IV}-(OEP)(O)(1-MeIm) (828 cm^{-1}) and Fe^{IV}(TpivPP)(O)(1-MeIm) (807 cm^{-1}),[25] consistent with a very strongly hydrogen-bonded ferryl unit in HRP-II. In the same vein, chloroperoxidase compound II (CPO-II) does not even exhibit an ^{18}O-sensitive Fe−O stretching frequency in resonance Raman spectra for different excitation wavelengths,[26,27] which led one of us to suggest that CPO-II may be better described as an iron(IV)−hydroxo intermediate.[28] Our goal in this study is not to contribute to the ongoing debate on any of the above-mentioned biochemical results, but rather to view them against a chemical context, especially that provided by Groves' key contribution mentioned above.[13] We attempt to do so by presenting a quantum chemical survey of iron(IV) porphyrins with two univalent axial ligands such as alkoxide, thiolate, or hydroxide, examples of which are shown in Figure 2.

From the outset, DFT appeared to be the natural method of choice for such a study, given the major role it has already played in enhancing our understanding of the electronic structures of high-valent metalloporphyrins and related complexes.[9,12,29] Thus, extensive DFT studies have been reported for metalloporphyrin π-cation radicals,[30] peroxidase compound I and compound II intermediates,[31−38] high-spin iron(III) porphyrin π-cation radicals,[39] iron(V)−nitrido porphyrins,[40] manganese(IV) and manganese(V) porphyrin intermediates,[39,41] high-valent iron and manganese corroles,[42−45] Ni(III) porphyrins,[46,47] and Cu(III) corroles.[48] Of particular interest has been the question of noninnocent ligands, viz. whether a particular high-valent transition metal complex features a "truly" high-valent metal center or a noninnocent or partially oxidized porphyrin or other ligand.[9,42] DFT studies of some high-valent nonheme

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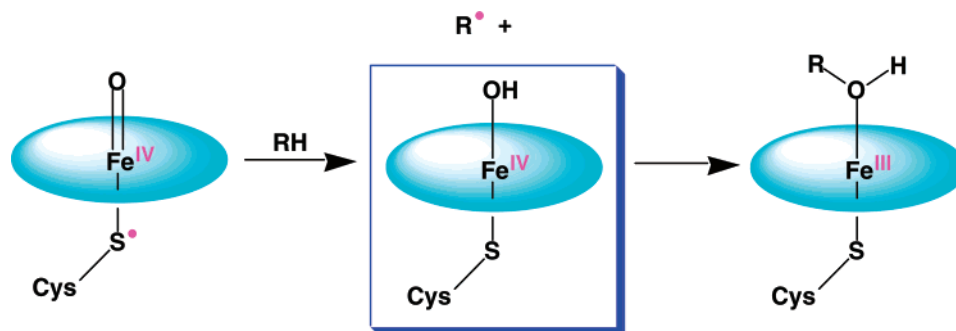High-Valent Heme Protein Intermediates

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Figure 1. High-valent iron porphyrin intermediates implicated as key intermediates in the so-called "rebound" mechanism of heme-based oxygenases.
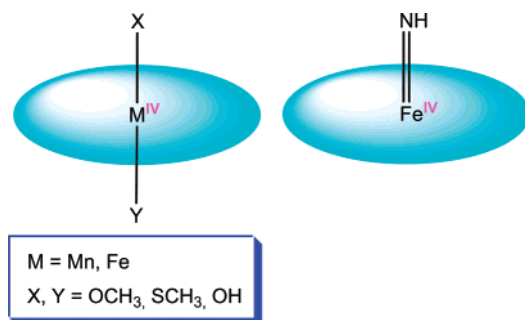

M = Mn, Fe

X, Y = OCH₃, SCH₃, OH

Figure 2. M(IV) porphyrin species studied. The complexes with X = Y = SCH₃ or X = Y = OH were not included in this study. For comparison, we have also studied the Fe(IV)–imido porphyrin shown.

iron–oxo intermediates may also be mentioned in this connection.[10–12,49]

In this study, we have sought to address two main aspects of iron(IV) porphyrins with two univalent π-donating axial ligands such as alkoxide, thiolate, or hydroxide. Not surprisingly, these highly reactive species have not yet been structurally characterized. Accordingly, a key point of interest in this study concerns the coordination geometry of the alkoxide, thiolate, and hydroxide ligands, specifically whether the Fe–O–C, Fe–S–C, and Fe–O–H angles are linear or quasi-linear, which would maximize metal–ligand π-bonding, or substantially bent. A second question of interest is related to whether the intermediates examined are "true" Fe(IV) species or whether they are better viewed as Fe(III) porphyrin π-cation radicals.[9] In addition, we have also compared the Fe(IV) intermediates of interest with analogous Mn(IV) complexes.[50]

DFT(PW91/TZP)[51] calculations with full geometry optimization were carried out on $M^{IV}(P)(X)(Y)$, for several combinations of M = Mn, Fe and X, Y = OCH₃, SCH₃, OH (Figure 2). For each molecule, the so-called "cis" and "trans" conformations (Figure 3) were studied. Figure 3 depicts key optimized geometry parameters and Mulliken atomic spin populations. In addition, the $Fe^{IV}(P)(NH)$ complex with the π-donating axial imido ligand was optimized with both C_{4v} and C_s symmetry constraints for comparison with our other results. The results of these calculations provide clear answers to the questions posed above, as mentioned below.

Structural Chemistry

A key structural result is that in each case examined, the M–O–C, M–O–H, and M–S–C angles are clearly bent, the M–O–C angles being about 130–135° and the M–O–H and M–S–C angles being close to the tetrahedral angle, 106–114°. In other words, the M–O or M–S π-bonding, though significant, is clearly not sufficient to result in linearization of these bond angles. To put these results in perspective, we have

examined whether the very strongly π-bonding imido ligand in $Fe^{IV}(P)(NH)$ favors a linear geometry, as we tacitly assumed in a previous[40] theoretical study. Indeed, a bent conformation is favored with an FeNH angle of 118.1° over an upright C_{4v} conformation by a rather large margin of 0.5 eV (13.6 kcal/mol). These strongly bent angles may be contrasted with the essentially linear Mn–N$_{imido}$–C$_{aryl}$ angle in an Mn(V)–arylimido corrole complex, which features a formal Mn–N$_{imido}$ triple bond.[52]

The optimized FeO distances are 1.81–1.85 Å, rather short for FeO distances in general, but consistent with the high-valent nature of the compounds in question. The optimized MnO distances are about 0.03 Å longer than the corresponding FeO distances in analogous Fe compounds. The same effect is also seen for M(IV)–oxo porphyrins, where an $Fe^{IV}O$ unit exhibits a shorter bond distance and a higher stretching frequency than an $Mn^{IV}O$ unit in an analogous complex.[41,53] We have suggested that this structural difference between Mn and Fe results from a spatially tighter 3d subshell for Mn, which somewhat inhibits its ability to form π-bonds, compared with Fe.[41] An alternative explanation involving a special stability of the half-filled t_{2g} subshell in the Mn(IV) case has also been offered.[53] In the same way, the optimized MnS distances (2.33–2.34 Å) are somewhat longer than the optimized FeS distances (2.27–2.29 Å). Some additional structural aspects of the species studied are as follows. The M–O vectors generally lie along the heme normal (indicated by a dotted line in Figure 3), whereas the M–S vectors are generally tilted by a few degrees. Finally, the cis and trans conformations of the species studied are essentially equienergetic, suggesting that the axial ligands can freely rotate about the heme normal.[54]

Spin Density Profiles

For all the compounds studied, the metal atoms and the axial ligands carry essentially all the unpaired electron density so that the metal centers may be regarded as "truly" high-valent and the porphyrin ligand as essentially "innocent" or "nonradicaloid",[9,55] although low-lying Fe(III) porphyrin cation radical states are a distinct possibility in certain cases, especially for the "rebound intermediate" of cytochrome P450 (Figure 1). Moreover, as shown in Figure 3, the Fe and Mn atoms in the species studied carry about 70–80% and 95%, respectively, of the total molecular spin density. By comparison, the metal atoms in $Fe^{IV}(P)(O)(pyridine)$ and $Mn^{IV}(P)(O)(pyridine)$ carry only about 55% and 75%, respectively, of the total molecular spin density.[41] In other words, an axial oxo ligand is significantly better at delocalizing spin density and presumably also positive charge away from the metal center than two univalent π-bonding axial ligands. We do not yet have a "feel" for the implications of this finding for the chemical reactivity of the molecules studied, but it is clearly an interesting issue, in our opinion.
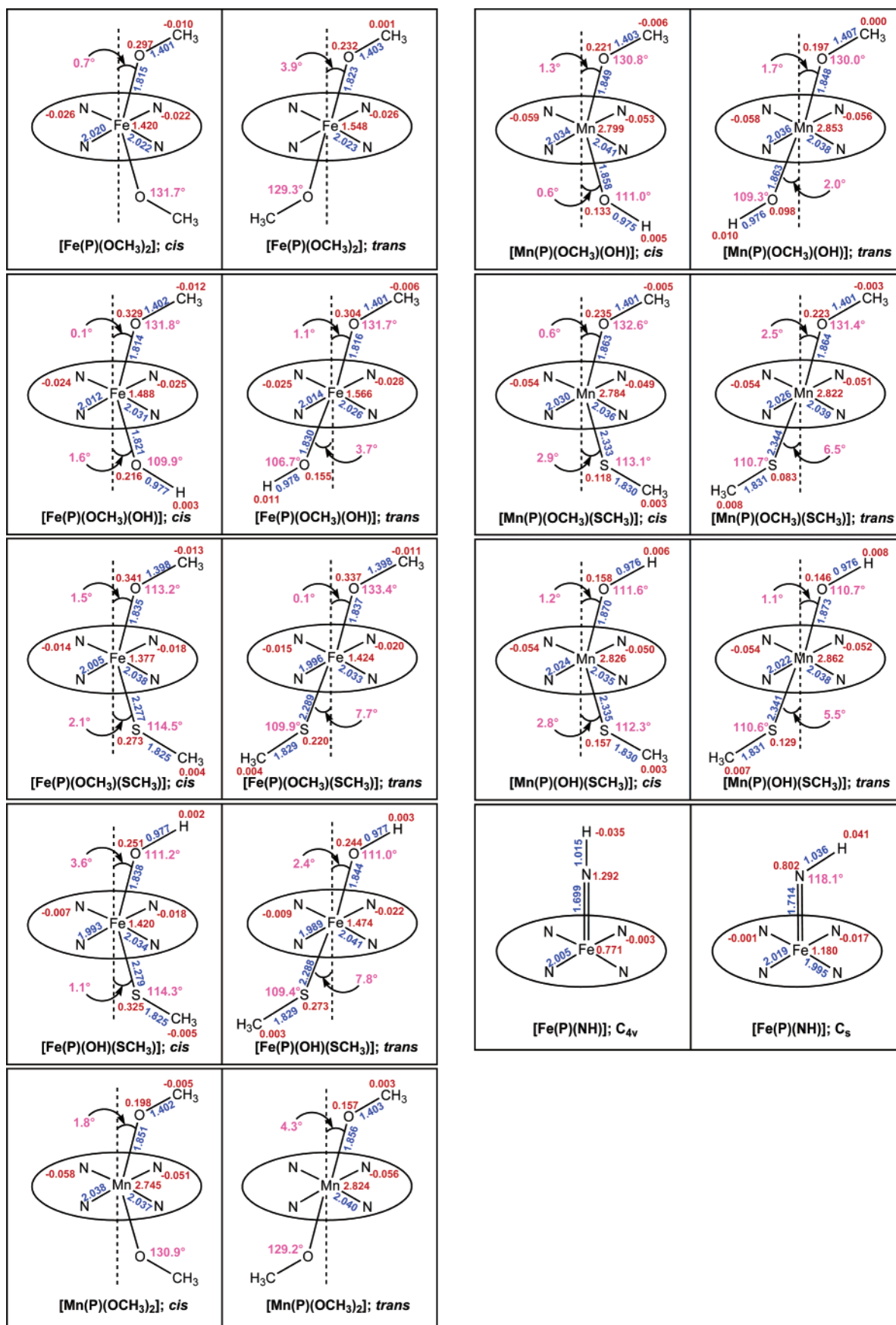
Figure 3. Selected optimized geometry parameters (Å, blue; deg, magenta) and gross atomic spin populations (red) for the different $M^{IV}(P)(X)(Y)$ (M = Fe, Mn, X, Y = OCH$_3$, OH, SCH$_3$) derivatives studied as well as for [Fe(P)(NH)].

A few additional observations about the spin density profiles are as follows. For the three different axial ligands examined, the ability to harbor unpaired electron density decreases along the series OCH$_3$ > SCH$_3$ > OH. Second, for each complex

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studied, the metal spin population is slightly higher in the trans conformation than in the cis conformation. Third, the higher spin populations on Mn compared with Fe, in other words, the lesser degree of metal-to-ligand spin delocalization in the Mn case, again might reflect the lower π-bonding ability of Mn. Fourth, the N_{imido} spin density is substantially lower in the more stable bent conformation of $Fe^{IV}(P)(NH)$ relative to the upright C_{4v} conformation, consistent with weaker $M-N_{imido}$ π-bonding and the slightly longer $Fe-N_{imido}$ bond distance in the C_s conformation. Finally, in contrast to genuine low-spin Fe(IV) intermediates, the $S = 1$ ground states of [Fe(porphyrinato)-(imidazole)$_2$]$^{2+}$ species have been described as reflecting strong ferromagnetic coupling between a low-spin Fe(III) center and a porphyrin radical.[56,57] Because strong metal–ligand ferromagnetic coupling is relatively unusual, we carried out a quantum chemical study of these species and concluded that the $S = 1$ ground states actually do reflect significant Fe(IV) character.[58]

Conclusion

In summary, we have presented a quantum chemical survey of iron(IV) and manganese(IV) porphyrin intermediates with two univalent π-bonding axial ligands. Besides providing a first quantum chemical description of Groves' key dimethoxyiron-(IV) intermediate,[13] the results, in our opinion, also add to the chemical context against which we view the "rebound" mechanism of cytochrome P$_{450}$, especially the "boxed" intermediate shown in Figure 1. The axial ligands in the species studied invariably exhibit a bent geometry, reflecting a competitive interplay of σ- and π-bonding effects. Compared to an oxo ligand, the univalent ligands are less able to delocalize electronic spin density away from the metal center; we do not know whether and how this electronic-structural feature contributes to the chemical reactivity of the complexes studied here.

Supporting Information Available: Optimized Cartesian coordinates. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) Dawson, J. H.; Sono, M. *Chem. Rev.* **1987**, *87*, 1255.

(2) Que, L., Jr.; Tolman, W. B. *Angew. Chem., Int. Ed.* **2002**, *41*, 1114.

(3) Terner, J.; Gold, V.; Weiss, R.; Mandon, D.; Trautwein, A. X. *J. Porphyrins Phthalocyanines* **2001**, *5*, 357.

(4) Que, L., Jr. In *Bioinorganic Catalysis*, 2nd ed.; Reedijk, J., Bouwman, E., Eds.; Marcel Dekker: New York, 1999, 269.

(5) Solomon, E. I.; Brunold, T. C.; Davis, M. I.; Kemsley, J. N.; Lee, S.-K.; Lehnert, N.; Neese, F.; Skulan, A. J.; Yang, Y.-S.; Zhou, J. *Chem. Rev.* **2000**, *100*, 235.

(6) Grapperhaus, C. A.; Mienert, B.; Bill, E.; Weyhermüller, T.; Wieghardt, K. *Inorg. Chem.* **2000**, *39*, 5306.

(7) Kim, M. H.; Rohde, J.-U.; Stubna, A.; Bukowski, M. R.; Costas, M.; Ho, R. Y. N.; Münck, E.; Nam, W.; Que, L., Jr. *Proc. Natl. Acad. Sci. U.S.A.* **2003**, *100*, 3665.

(8) Rohde, J.-U.; In, J. H.; Lim, M. H.; Brennessel, W. W.; Bukowski, M. R.; Stubna, A.; Münck, E.; Nam, W.; Que, L., Jr. *Science* **2003**, *299*, 1037.

(9) For a broad quantum chemical survey of high-valent metalloporphyrins, see: Ghosh, A.; Steene, E. *J. Biol. Inorg. Chem.* **2001**, *6*, 739.

(10) Siegbahn, P. E. M.; Blomberg, M. R. A. *Chem. Rev.* **2000**, *100*, 421.

(11) Guallar, V.; Gherman, B. F.; Lippard, S. J.; Friesner, R. A. *Curr. Opin. Chem. Biol.* **2002**, *6*, 236.

(12) For a critical discussion of the limitations of DFT vis-à-vis transition metal calculations, see: Ghosh, A.; Taylor, P. R. *Curr. Opin. Chem. Biol.* **2003**, *7*, 113.

(13) Groves, J. T.; Quinn, R.; McMurry, T. J.; Nakamura, M.; Lang, G.; Boso, B. *J. Am. Chem. Soc.* **1985**, *107*, 354.

(14) Calderwood, T. S.; Lee, W. A.; Bruice, T. C. *J. Am. Chem. Soc.* **1985**, *107*, 8272.

(15) Lee, W. A.; Calderwood, T. S.; Bruice, T. C. *Proc. Natl. Acad. Sci. U.S.A.* **1985**, *82*, 4301.

(16) Swistak, C.; X. H. Mu, Kadish, K. M. *Inorg. Chem.* **1987**, *26*, 4360.

(17) Filatov, M.; Harris, N.; Shaik, S. *Angew. Chem., Int. Ed.* **1999**, *38*, 3510.

(18) Kamachi, T.; Yoshizawa, K. *J. Am. Chem. Soc.* **2003**, *125*, 4652.

(19) For a recent review, see: Groves, J. T. *Proc. Natl. Acad. Sci. U.S.A.* **2003**, *100*, 3569.

(20) Shaik, S.; de Visser, S. P.; Ogliaro, F.; Schwarz, H.; Schröder, D. *Curr. Opin. Chem. Biol.* **2002**, *6*, 556.

(21) de Visser, S. P.; Ogliaro, F.; Harris, N.; Shaik, S. *J. Am. Chem. Soc.* **2001**, *123*, 3037.

(22) Penner-Hahn, J.; Eble, K. S.; McMurry, T. J.; Renner, M.; Balch, A. L.; Groves, J. T.; Dawson, J. H.; Hodgson, K. O. *J. Am. Chem. Soc.* **1986**, *108*, 7819.

(23) Berglund, G. I.; Carlsson, G. H.; Smith, A. T.; Szöke, H.; Henriksen, A.; Hajdu, J. *Nature* **2002**, *417*, 463.

(24) Sitter, A. J.; Reczek, C. M.; Terner, J. *J. Biol. Chem.* **1985**, *260*, 7515.

(25) Schappacher, M.; Chottard, G.; Weiss, R. *J. Chem. Soc., Chem. Commun.* **1986**, 93.

(26) Egawa, T.; Proshlyakov, H.; Miki, R.; Makino, T.; Ogura, T.; Kitagawa, T.; Ishimura, Y. *J. Biol. Inorg. Chem.* **2001**, *6*, 46.

(27) Hosten, C. M.; Sullivan, A. M.; Palaniappan, V.; Fitzgerald, M. M.; Terner, J. *J. Biol. Chem.* **1994**, *269*, 13966.

(28) Ghosh, A.; Steene, E. *J. Inorg. Biochem.* **2002**, *91*, 423.

(29) Ghosh, A.; Persson, B. J.; Taylor, P. R. *J. Biol. Inorg. Chem.* **2003**, *5*, 345.

(30) Vangberg, T.; Lie, R.; Ghosh, A. *J. Am. Chem. Soc.* **2002**, *124*, 8122−8130.

(31) For the first DFT calculations on Fe(IV)−oxo porphyrins, see: Ghosh, A.; Almlöf, J.; Que, L., Jr. *J. Phys. Chem.* **1994**, *98*, 5576.

(32) Deeth, R. J. *J. Am. Chem. Soc.* **1999**, *121*, 6074.

(33) Vangberg, T.; Ghosh, A. *J. Am. Chem. Soc.* **1999**, *121*, 12154.

(34) Harris, D. L. *Curr. Opin. Chem. Biol.* **2001**, *5*, 724.

(35) Kuramochi, H.; Noodleman, L.; Case, D. A *J. Am. Chem. Soc.* **1997**, *119*, 11442.

(36) Green, M. T. *J. Am. Chem. Soc.* **1998**, *120*, 10772.

(37) Green, M. T. *J. Am. Chem. Soc.* **1999**, *121*, 7939.

(38) Green, M. T. *J. Am. Chem. Soc.* **2001**, *123*, 9218.

(39) Ghosh, A.; Gonzalez, E.; Vangberg, T.; Taylor, *J. Porphyrin Phthalocyanines* **2001**, *5*, 345.

(40) Dey, A.; Ghosh, A. *J. Am. Chem. Soc.* **2002**, *124*, 3206.

(41) Ghosh, A.; Gonzalez, E. *Isr. J. Chem.* **2000**, *40*, 1.

(42) For our latest study on high-valent Fe and Cu corroles, see: Steene, E.; Day, A.; Ghosh, A. *J. Am. Chem. Soc.*, in press.

(43) Steene, E.; Wondimagegn, T.; Ghosh, A. *J. Phys. Chem. B* **2001**, *105*, 11406. Addition/Correction: *J. Phys. Chem. B.* **2002**, *106*, 4312.

(44) Tangen, E.; Ghosh, A. *J. Am. Chem. Soc.* **2002**, *124*, 8117.

(45) Zakharieva, O.; Schunemann, V.; Gerdan, M.; Licoccia, S.; Cai, S.; Walker, F. A.; Trautwein, A. X. *J. Am. Chem. Soc.* **2002**, *124*, 6636.

(46) Ghosh, A.; Wondimagegn, T.; Gonzalez, E.; Halvorsen, I. *J. Inorg. Biochem.* **2000**, *78*, 79.

(47) Wondimagegn, T.; Ghosh, A. *J. Am. Chem. Soc.* **2001**, *123*, 1543.

(48) Ghosh, A.; Wondimagegn, T.; Parusel, A. B. J. *J. Am. Chem. Soc.* **2000**, *122*, 5100.

(49) For the first DFT study of a high-valent iron diamond-core intermediate, see: Ghosh A.; Almlof J.; Que L. *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 770. For a follow-up study, see: Ghosh, A.; Tangen, E.; Gonzalez, E.; Que, L., Jr. *Angew. Chem. Int. Ed.*, in press.

(50) For a thorough experimental study of the electronic structure of Mn(IV) complexes, see: Kaustov, L.; Tal, M. E.; Shames, A. I.; Gross, Z. *Inorg. Chem.* **1997**, *36*, 3503.

(51) All calculations were carried out with the ADF program system, the VWN local exchange-correlation functional, PW91 gradient corrections, Slater-type triple-ζ plus polarization (TZP) basis sets, a fine mesh for numerical integration of matrix elements, and tight criteria for geometry optimizations. For additional technical details, the reader is referred to the program manual, available from: Scientific Computing and Modeling, Department of Theoretical Chemistry, Vrije Universiteit, 1081 HV Amsterdam, The Netherlands.

(52) Eikey, R. A.; Khan, S. I.; Abu-Omar, M. *Angew. Chem., Int. Ed.* **2002**, *41*, 3591.

(53) Czernuszewicz, R. S.; Su, Y. S.; Stern, M. K.; Macor, K. A.; Kim, D.; Groves, J. T.; Spiro, T. G. *J. Am. Chem. Soc.* **1988**, *110*, 4158.

(54) This is indeed what is expected. Dinitrosylheme intermediates are a surprising violation of this scenario; they strongly favor a cisoid orientation of the NO ligands: Conradie, J.; Wondimagegn, T.; Ghosh, A. *J. Am. Chem. Soc.* **2003**, *125*, 4968.

(55) As discussed in ref 9, the ^{1}H NMR spectrum of Fe^{IV}(TMP)(OCH$_3$)$_2$ exhibits significant paramagnetic shifts for the β-protons. However, as in the case Fe(porphyrinato)Cl complexes, we believe that these paramagnetic shifts result from very small unpaired spin populations at the β-carbons.

(56) This study presents a revised electronic-structural description of [Fe(porphyrinato)(imidazole)$_2$]$^{2+}$ intermediates relative to that given in the original study, ref 57: Gans, P.; Buisson, G.; Duée, E.; Marchon, J. C.; Erler, B. S.; Scholz, W. F.; Reed, C. A. *J. Am. Chem. Soc.* **1986**, *108*, 1223.

(57) Goff, H. M.; Phillippi, M. *J. Am. Chem. Soc.* **1983**, *105*, 7567.

(58) Conradie, J.; Ghosh, A. *J. Phys. Chem. B* **2003**, *107*, 6486.