Excited state potential energy surfaces and their interactions in Fe$^{IV}$=O active sites

Martin Srnec,$^{a,b}$ Shaun D. Wong$^a$ and Edward I. Solomon$^a$

The non-heme ferryl active sites are of significant interest for their application in biomedical and green catalysis. These sites have been shown to have an $S = 1$ or $S = 2$ ground spin state; the latter is functional in biology. Low-temperature magnetic circular dichroism (LT MCD) spectroscopy probes the nature of the excited states in these species including ligand-field (LF) states that are otherwise difficult to study by other spectroscopies. In particular, the temperature dependences of MCD features enable their unambiguous assignment and thus determination of the low-lying excited states in two prototypical $S = 1$ and $S = 2$ NHFe$^{IV}$=O complexes. Furthermore, some MCD bands exhibit vibronic structures that allow mapping of excited-state interactions and their effects on the potential energy surfaces (PESs). For the $S = 2$ species, there is also an unusual spectral feature in both near-infrared absorption and MCD spectra – Fano antiresonance (dip in Abs) and Fano resonance (sharp peak in MCD) that indicates the weak spin–orbit coupling of an $S = 1$ state with the $S = 2$ LF state. These experimental data are correlated with quantum-chemical calculations that are further extended to analyze the low-lying electronic states and the evolution of their multiconfigurational characters along the Fe–O PESs. These investigations show that the lowest-energy states develop oxyl Fe$^{III}$ character at distances that are relevant to the transition state (TS) for H-atom abstraction and define the frontier molecular orbitals that participate in the reactivity of $S = 1$ vs. $S = 2$ non-heme Fe$^{IV}$=O active sites. The $S = 1$ species has only one available channel that requires the C–H bond of a substrate to approach perpendicular to the Fe–oxo bond (the $\pi$ channel). In contrast, there are three channels (one $\sigma$ and two $\pi$) available for the $S = 2$ non-heme Fe$^{IV}$=O system allowing C–H substrate approach both along and perpendicular to the Fe–oxo bond that have important implications for enzymatic selectivity.

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Shaun Wong received his B.S. in chemistry from UC Berkeley and his Ph.D. from Stanford University. His work centered on elucidating the electronic structures and reaction mechanisms of low- and high-spin Fe(IV)-oxo species in enzymes and model complexes.
Introduction

Mononuclear non-heme iron (NHFe) enzymes, ubiquitous in living organisms, are involved in many vital biological processes including the regulation of hypoxia, demethylation of DNA, antibiotic, and natural product biosynthesis and bioremediation, and are related to disease states.1,2 Most of these enzymes use an S = 2 Fe3+ center to activate O2 to perform various ‘difficult’ and formally spin-forbidden reactions including hydroxylation, halogenation, desaturation, and electrophilic aromatic substitution on unreactive singlet organic substrates that require the cleavage of strong aliphatic or aromatic C–H bonds. The mononuclear NHFe enzymes can be divided into six classes3 based on their O2 activation or aromatic C electrophilic aromatic substitution on unreactive singlet Fe2S2 Rieske center that gives rise to a reactive high-spin mechanism (Table 1; here we omit classes where substrates are active substrate that include isopenicillin-8,9). Provided the S = 1/2 FeII center is the only source of electrons as is in the case for \( \text{Fe}^{III} \) OOH intermediate.5

Similarly, in bleomycin, a glycopeptide antibiotic used in anticancer therapy,6 dioxygen is activated to form a low-spin (S = 1/2) FeIII–OOH intermediate.7 The FeIII center is the only source of electrons as in the case for the class of enzymes that have no cofactor and a non-redox active substrate that include isopenicillin-N-synthase (IPNS), the 1e– reduction of O2 can give the thermodynamically unfavored high-spin (\( S_{\text{total}} = 2 \)) FeIII-O2–2 intermediate.8 In the two most prevalent NHFe classes that use either a pterin or an \( \alpha \)-keto glutarate as a cofactor, the formation of high-spin \( S = 2 \) FeIV=O (ferryl) intermediates proceed through the four-electron reduction of O2 (2e– from Fe3+ and 2e– from the cofactor).9 Note these NHFe oxygen intermediates, in spite of their elusive nature, have attracted much attention for their reactivities and selectivities as exemplified by studies on the high-spin (S = 2) FeIV=O intermediate in syringomycin halogenase (SyrB2).10–13

Efforts to understand the general physico-chemical factors that control reaction mechanisms of NHFe enzymes have also catalyzed interest in the syntheses and crystallographic characterizations of model NHFe complexes that include low-spin (S = 1) and high-spin (S = 2) FeIV=O complexes.14–19 These enable an important step toward the elucidation of geometric and electronic properties contributing to NHFe function: the spectroscopic and quantum-chemical elucidation of the frontier molecular orbitals (FMOs) and their contributions to reactivity in model S = 1 and S = 2 FeIV=O systems.20–28 For the S = 1 FeIV=O species, the pair of singly-occupied \( \sigma^* \) FMOs, resulting from the strong antibonding interaction between oxo \( p_x,p_y \) and Fe \( d_{x^2,y^2,z^2} \) orbitals, were shown to define the \( \pi \) channel for H-atom abstraction (HAA) which requires for reactivity the perpendicular orientation of the substrate C–H bond with respect to the Fe–oxo bond.21 For the S = 2 FeIV=O species, two types of FMOs were defined, i.e., \( \sigma^* \) and \( \sigma^* \). The \( \sigma^* \) defines a \( \pi \) channel similar to the S = 1 FeIV=O species, while \( \sigma^* \) arises from the strong \( \sigma \) antibonding interaction of the \( p_z \) with the Fe \( d_y \) orbital and defines the \( \pi \) channel for HAA that requires collinear orientation of the substrate C–H bond relative to FeIV=O bond.22 This additional \( \sigma \) channel in S = 2 complexes was elucidated on the basis of spin-unrestricted MO theory as given in Fig. 1. In going from an S = 1 to an S = 2 FeIV=O system, the excitation of a \( \beta^* \) from the non-bonding \( d_{xy} \) orbital into the \( \alpha,d_{y^2} \) orbital leads to a considerable electron–electron exchange stabilization (referred in the literature to as spin polarization20 or exchange enhancement21) that shifts the \( \alpha \)-MO manifold down in energy; thus the unoccupied \( \alpha,d^\pi \) becomes energetically comparable to the \( \sigma^* \) FMOs and accessible for reactivity.21 The flexibility of S = 2 species in using \( \sigma^* \) and/or \( \sigma^* \) FMOs were shown to be key to the reaction selectivity in SyrB212 where \( \sigma^* \) vs. \( \sigma^* \) control halogenation vs. hydroxylation of different substrates, and in HmaS vs. HPPD,29 where \( \sigma^* \) vs. \( \sigma^* \) participate in HAA vs. electrophilic aromatic substitution (EAS) on the same substrate.

From the experimental perspective, these FMOs can be studied through electronic spectroscopy particularly of the ligand-field (LF) transitions. These correspond to excitations into low-lying unoccupied d orbitals that get significant oxo character (hence their electrophilicity) through covalent bonding. However, these absorption (Abs) spectra are generally quite featureless due to band overlaps and thus not very informative. By contrast, the variable-temperature magnetic circular dichroism (VT MCD) spectra are much more resolved and thus give unique insight into these excited states and thus the FMOs that are key to electrophilic reactivity.

In this perspective, we focus on our spectroscopic results on the first structurally defined \( -C_2 \), symmetric S = 1 FeIV=O...
Table 1  Classes of O₂-activating NHFe species with examples

<table>
<thead>
<tr>
<th>Class</th>
<th>Reaction</th>
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<tbody>
<tr>
<td>Extradiol Dioxygenases</td>
<td>2,3-dihydroxybiphenyl 1,2-hydroxylase</td>
</tr>
<tr>
<td>Pterin-dependent Hydroxylases</td>
<td>Phenylalanine hydroxylase</td>
</tr>
<tr>
<td>α-KG-dependent Dioxygenases</td>
<td>Clavaminate synthase II</td>
</tr>
<tr>
<td>Rieske Dioxygenases</td>
<td>Phthalate dioxygenase</td>
</tr>
<tr>
<td>Non-redox Substrate Enzymes</td>
<td>Isopenicillin N synthase</td>
</tr>
<tr>
<td>Bleomycin</td>
<td>DNA → Bleomycin → base propenalis</td>
</tr>
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</table>

Fig. 1  MO diagrams of [(CH₃CN)(TMC)Fe⁴⁺=O] in S = 1 (ground) and S = 2 spin states, showing that excitation of β-dₓᵧ e⁻ into α-dₓ²₋ᵧ² orbital leads to spin polarization of the α-manifold and an additional low-energy α-dᵧ² FMO available for reactivity.

Fig. 2  Two crystallographically characterized NHFe⁴⁺=O complexes (crystal structures from ref. 30 and 31). Atoms are color-coded as follows: C (green), N (blue), O (red) and Fe (orange). The d-manifold splittings in both C₄ᵥ- and C₃ᵥ-like symmetric structures, along with electron occupations, are qualitatively depicted.

complex [chelated by the tetradequate 1,4,8,11-tetramethyl-1,4,8,11-tetraazaacyclotetradecane (TMC) ligand with an axially bound acetonitrile and denoted as (CH₃CN)(TMC)Fe⁴⁺=O; see Fig. 2]¹⁰ and the first structurally defined C₃ᵥ-like symmetric S = 2 Fe⁴⁺=O complex [chelated by the tetradequate 1,1,1-tris-(2-[N₂(1,1,3,3-tetramethylguanidino)]ethyl)amine (TMG₃tren)
ligand; \{TMG₄tren\}Fe⁴⁺–O₂; see Fig. 2). Both complexes were synthesized and structurally defined by Prof. L. Que and colleagues. In this perspective, we will show how VT MCD spectroscopy (combined with Abs spectroscopy and quantum-chemical calculations) allowed the description of low-lying excited states, their interactions that shape excited-state potential-energy surfaces and how these allow the experimental definition of FMOs that ultimately provide insight into reactivity of \(S = 1\) and \(S = 2\) Fe⁴⁺–O species of biological and catalytic significance.

\textbf{\(S = 1\) NHFe⁴⁺–O species}

The Abs and MCD spectra of the \(S = 1\) (CH₃CN)[TMC]Fe⁴⁺–O model complex are shown in Fig. 3A and B, respectively. While the Abs spectrum clearly shows three spectral features (at \(\sim 12,250, \sim 17,500\) (weak) and \(24,900\) cm\(^{-1}\) (strong)), the VT MCD spectra are much more resolved such that the near-infrared (NIR) feature centered at \(12,250\) cm\(^{-1}\) is composed of three overlapping bands (one having vibronic structure), two of which have positive MCD intensity (bands labelled I and III at \(\sim 10,400\) and \(\sim 12,900\) cm\(^{-1}\) in Fig. 3B) while one is negative (band II at \(\sim 10,600\) cm\(^{-1}\) with sharp features). Additionally, the temperature dependence of these bands gives valuable information on the polarizations of the electronic transitions in frozen solution, where all orientation of the molecules are present. These polarizations allow the ground state to be coupled to the excited states leading to (along with the analysis of \(C_D/D_0\) ratios) the assignment of the MCD/Abs bands. This is given by the selection rules for MCD transitions from a zero-field-split (ZFS) \(S = 1\) ground spin state that is further split due to interaction with the external magnetic field (Fig. 4C and ref. 20, 21).

Using the phenomenological spin Hamiltonian that includes the ZFS term \(D(S^2 – S(S + 1)/3) + E(S_x^2 – S_y^2)\) with axial \(D\) and rhombic \(E\) ZFS parameters, the analysis of magnetic Mössbauer spectra of the (CH₃CN)[TMC]Fe⁴⁺–O complex showed \(E\) to be negligible and \(D = 29\) cm\(^{-1}\). As depicted in Fig. 3C, for this positive \(D\) value, the ground triplet state splits into two sublevels with two-fold degenerate \(M_S = \pm 1\) lying above the \(M_S = 0\) sublevel. From the selection rules for MCD intensity, the magnetic field \((H)\) must be perpendicular to the polarization direction of the transition. Thus a \(z\)-polarized transition requires \(H\) to be perpendicular to the molecular \(z\) axis (coinciding with the Fe–oxo bond). In this case, the Zeeman-induced mixing between \(M_S = -1\) and \(M_S = 0\) results in significant \(M_S = -1\) character in the ground sublevel at \(H = 7\) T. From this MCD-active sublevel, the \(z\)-polarized transition exhibits the highest MCD intensity at lowest temperature, \(T \sim 2\) K that decreases with increasing \(T\) because of increased Boltzmann populations of higher-energy sublevels: the MCD-inactive \(M_S = 0\) and opposite-signed \(M_S = +1\). For an \((x,y)\)-polarized transition, \(H\) is required to be parallel to the molecular \(z\) axis. This leaves the MCD-inactive \(M_S = 0\) as the ground sublevel at \(H = 7\) T. As a consequence, the intensity of a \((x,y)\)-polarized MCD band is low at low temperature and increases with increasing \(T\) because of an increased Boltzmann population of the first excited MCD-active \(M_S = -1\) sublevel. Further increasing the \(T\) decreases the MCD intensity because the third sublevel, \(M_S = +1\), becomes populated, producing MCD intensity of opposite sign.

From these selection rules, band I in Fig. 3B was shown to be \(z\)-polarized (decreasing in intensity with increasing \(T\)), while bands II and III are \((x,y)\)-polarized (both first increase and then decrease with increasing \(T\)). In particular, the different polarizations (and thus temperature behavior) of the two overlapping bands I and II allowed these to be decoupled and band II to be unambiguously associated with the MCD vibronic structure (plotted as a positive band in Fig. 3D). From group theory and DFT calculations, band II was assigned as the lowest-energy \((x,y)\)-polarized \(d_{xy} \rightarrow \pi^*_{xz/yz}\) ligand-field (LF) transition (MOs shown in Fig. 3E; band I and III, IV and V are assigned as indicated in Fig. 3B). Importantly, this transition,
in Fig. 3D gives $\Delta r_{\text{Fe-O}} (= 0.14 \text{ Å})$, which was determined from eqn (1):

$$\Delta r_{\text{Fe-O}} = \sqrt{\hbar S_{\text{Fe-O}}/4\pi \mu_{\text{Fe-O}}}$$  \hspace{1cm} (1)

where $S_{\text{Fe-O}} (= 3.0)$ is the Huang-Rhys parameter given by eqn (2):

$$S = \sqrt{\frac{\Delta I_{0-n}}{\Delta I_{0-0}}}$$  \hspace{1cm} (2)

with $\Delta I_{0-n}$ and $\Delta I_{0-0}$ being the intensity of the first and $(n + 1)$-th peak in the progression, and $\mu$ ($\approx 15$ amu) is the reduced mass of the (ligand+Fe)-oxo stretching mode. The frequency of the Fe-oxo stretching mode in the ground state is known from the IR and NRVS spectra ($\sim$830 cm$^{-1}$).$^{30,33}$ From these data, PESs of the ground and excited state along Fe-oxo stretching mode can be defined experimentally (Fig. 3F). Clearly, the excited state is associated with a distorted PES having its minimum shifted to longer Fe-oxo bond length as compared to the PES of the ground state, which reflects the weaker Fe-oxo bond due to excitation of a nonbonding d $e^-$ into the antibonding Fe-oxo d*$\pi$ FMO. This strong $\pi^h/\pi^*$ interaction results in significant oxo $\sigma$ character in the dx* orbital that is key to activating it for reactivity. Note that the low-energy two-fold degenerate dx* orbital set is much lower in energy than Fe-oxo dx* (the latter is associated with the band V at $\sim$24 900 cm$^{-1}$ in Fig. 3B and displayed in Fig. 3E). These results have important implications for the H-atom abstraction (HAA) reaction coordinate for $S = 1$ Fe$^{IV}=$O systems. HAA must involve this low-energy Fe dx* orbital that accepts the $e^-$ from the substrate requiring good overlap with the C-H $\sigma$ orbital. Since the dx* is oriented perpendicular to the Fe-oxo bond (thus having significant oxo $p_{\sigma y}$ $\pi$ character), a perpendicular orientation of the C-H relative to the Fe-oxo bond is required for an accessible HAA trajectory (Scheme 1). Note, in the model complexes, there are likely steric effects of the equatorial chelate that hinder this $\pi$-channel in $S = 1$ Fe$^{IV}=$O sites (see ref. 34).

Alternatively to the $S = 1$ $\pi$ channel, several computational works suggested a possible spin crossover from $S = 1$ to $S = 2$ in the HAA process.$^{35,36}$ The feasibility of such a two-state mechanism depends on the accessibility of a $S = 2$ state that is obtained by the excitation of a $\beta-$dx$^y$ $e^-$ into the $\alpha-$dx$^y$ $\pi$-orbital. This would lead to a large stabilization of the $\alpha-$dx$^y$ $\sigma^*$ orbital (Fig. 1). This stabilization would provide the $\sigma^*$ FMO for HAA, which requires the collinear orientation of the substrate C-H relative to Fe-oxo bond and thus could overcome the chelate steric hindrance in some $S = 1$ systems.

Scheme 1. The dx* FMO interacting with the electron-donating C-H $\sigma$ orbital (left) and thus defining H-atom abstraction reaction coordinate for the $S = 1$ NHFe$^{IV}=$O species (right).
S = 2 NHFe⁴⁺ – O species

The Abs spectrum of the prototypical $S = 2$ ferryl complex, $(\text{TMG}_3\text{tren})\text{Fe}^{IV} = \text{O}^{33,37}$ exhibits three noticeable spectral features (Fig. 4A). Within the 6000–16 000 cm⁻¹ region, there is a weak Abs band centered at 12 500 cm⁻¹ (labelled as band I) having a dip in its intensity at ∼12 000 cm⁻¹, while the 16 000–30 000 cm⁻¹ (UV-vis) region is characterized by an intense broad band at 25 000 cm⁻¹ (band II) with a weak shoulder at 19 000 cm⁻¹ (band III). These bands become much more revealing in the LT MCD spectrum. Thus, as shown in Fig. 4B, the NIR feature I in fact consists of three overlapping bands: a lower-energy positive and higher-energy negative vibronic progression with a spacing of 710 and 880 cm⁻¹, respectively, and one positive sharp peak that is at the same energy as the dip in Abs (dashed vertical line in Fig. 4A and B). Additionally, the MCD reveals that four bands contribute to the Abs envelope in the UV-vis region (bands II, III, IV and V in Fig. 4A and B), where the negatively signed band II has well-resolved vibronic structure with a spacing of 490 cm⁻¹.

These MCD features (I–III) clearly exhibit two different VT behaviors that reflect different band polarizations. Feature I first increases and then decreases in intensity with increasing temperature, while bands II and III decrease in intensity with increasing $T$ (Fig. 4D) and following the theory described in section 2, the temperature dependences show that feature I is ($x_3y$)-polarized, and bands II and III are $z$-polarized.

Features I–V were also characterized in terms of their $C_δ/D_δ$ ratios $^{33}$ (included in Fig. 4B), with a large value for I (0.250) and significantly smaller values for II–V (<0.040). These indicate that feature I corresponds to a LF transition, while bands II–V are charge-transfer (CT) transitions. These experimental data, further correlated to TD-DFT calculations (Fig. 4E), led to the unambiguous assignment of the three lowest-energy features: Band I is the two-fold degenerate ($x_3y$)-polarized $\alpha\,\text{d}^{\pi}\pi^* \rightarrow \text{d}^{\pi}\pi^*$ LF transition (i.e., $^3\text{A}_1 \rightarrow \text{LF}^5\text{E}$), and bands II–III are the $z$-polarized $\beta\,\text{oxo}\,x_3y \rightarrow \text{d}^{\pi}\pi^*$ CT transitions. The molecular orbitals involved along with the transitions to the lowest-energy excited state LF $^5\text{E}$ are schematically depicted in Fig. 5A and B, respectively.

MCD feature I associated with the lowest excited state LF $^5\text{E}$ has a derivative band shape (Fig. 4B) that is primarily due to spin-orbit coupling (SOC) within the LF $^5\text{E}$ state. This gives rise to two oppositely signed C-term MCD features forming a pseudo-A term with the right circularly polarized (RCP; i.e., negative) component at higher energies (Fig. 5C). The in-state SOC between the spatially two-fold degenerate components of the LF $^5\text{E}$ state (depicted in Fig. 5B) is effectively controlled by the $z$-component of the SOC operator $\lambda S_L z$ (where $\lambda$ is a many-electron state-specific SOC constant) by acting on wavefunctions $|\text{M}^{\text{eff}}_L\rangle = \pm 1, M_S = 0, \pm 1, \pm 2$, where the effective magnetic quantum number $M^{\text{eff}}_L$ reflects the one-electron configuration subspace $d^{2\pi}_{xy}d^{\sigma}_{yz}, d^{2\pi}_{xz}d^{\sigma}_{yx}$ of the LF $^5\text{E}$. This SOC produces two fold-degenerate sublevels of the LF $^5\text{E}$ state with an energy separation between the lower-energy LCP and higher-energy RCP state of $2\lambda = 2(\zeta_{3d}/4) = 260$ cm⁻¹ ($\zeta_{3d}$ is the one-electron SOC constant for Fe$^{IV}$: 520 cm⁻¹)$^{38}$ as shown in Fig. 5C. The relation $\lambda = \zeta/4$ for this case is given by eqn (3):

$$
\begin{align*}
|\text{M}^{\text{eff}}_L\rangle = & \pm 1, \text{M}_S = |2|\pm 1, \text{S}|M^{\text{eff}}_L\rangle = \pm 1, \text{M}_S = 2 \\
& = \left\langle \frac{1}{\sqrt{2}} (d_{yz} + id_{xz}) \alpha \mid \zeta_{3d} l_s z \pm \frac{1}{\sqrt{2}} (d_{yz} - id_{xz}) \alpha \right\rangle 
\end{align*}
$$

Note these in-state SOC-perturbed MCD-active (LCP and RCP) states would have potential energy surfaces that do not differ in their shape. However, this is different from what is
observed from the NIR MCD pseudo-A term spectra that exhibit two very distinct vibronic progressions (Fig. 4B, right). These reflect a large difference in the frequency of the Fe-oxo stretching mode in the LCP vs. RCP component of the LF $^5E$ excited state ($\nu_{Fe-O} = 710 \text{ vs. } 880 \text{ cm}^{-1}$); hence there must be a difference in the shapes of the PESs associated with these states. Since the latter value of $\nu_{Fe-O}$ is even higher than the Fe-oxo frequency in the ground state (820–840 cm$^{-1}$), the LF $^5E$-based RCP state must undergo a strong SOC interaction with a nearby and spectroscopically spin-forbidden state. This would produce two RCP-active PESs, where the higher-energy PES is associated with the higher $\nu_{Fe-O}$ relative to its lower-energy cognate.

Indeed, the NIR MCD pseudo-A term is best fit by three (one positive and two negative) FC progressions and a sharp peak (Fig. 6), where the higher-energy RCP FC progression (green) has a larger $\nu_{Fe-O}$ than its lower-energy RCP FC cognate (dark blue) that is largely masked by the more intense LCP FC progression (dotted red). As was the case for band II of the $S = 1$ Fe$^{V}=O$ species in MCD (section 2), all of the FC fits contributing to this pseudo-A term (as well as the FC fit of the negative MCD band II associated with the oxo-to-Fe CT state in Fig. 4C, third panel) are characterized in terms of the zero-vibronic 0–0 transition ($E_{00}$), the $\nu_{Fe-O}$ and the Huang-Rhys factor ($S$), which is defined by eqn (2). These are given in Fig. 6.

There is an energy equivalence between the sharp positive peak in the $^5E$ MCD spectrum (solid red line in Fig. 6) and the dip in the Abs spectrum (Fig. 7A and B). This peak, due to its sharpness (the full width at half maximum is <400 cm$^{-1}$) and its positive MCD intensity, indicates a weak SOC interaction of the spin-allowed (broad) LCP component of the LF$^5E$ with a sharpness (the full width at half maximum is <400 cm$^{-1}$) and its positive MCD intensity, indicates a weak SOC interaction of the spin-allowed (broad) LCP component of the LF$^5E$ with a sharp peak (Fig. 6), where the higher-energy RCP FC progression (green) has a larger $\nu_{Fe-O}$ than its lower-energy RCP FC cognate (dark blue) that is largely masked by the more intense LCP FC progression (dotted red). As was the case for band II of the $S = 1$ Fe$^{V}=O$ species in MCD (section 2), all of the FC fits contributing to this pseudo-A term (as well as the FC fit of the negative MCD band II associated with the oxo-to-Fe CT state in Fig. 4C, third panel) are characterized in terms of the zero-vibronic 0–0 transition ($E_{00}$), the $\nu_{Fe-O}$ and the Huang-Rhys factor ($S$), which is defined by eqn (2). These are given in Fig. 6.

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$$R(\nu) = \frac{\alpha(\nu)}{\alpha_0(\nu)} = 1 + p\frac{q^2 + 2q(\nu - \nu_0)\gamma^{-1} - 1}{1 + (\nu - \nu_0)^2\gamma^{-2}}$$

where

$$p = \frac{\alpha(\nu_0)}{\alpha_0(\nu_0)} = \frac{\langle \psi_S | H_{\text{int}} | \mu \rangle \langle \mu | \psi_{GS} \rangle^2}{\langle \psi_S | H_{\text{int}} | \mu \rangle | \psi_{GS} \rangle^2}$$  \hfill (5, 6 and 7)

$$q = \frac{\langle \psi_S | H_{\text{int}} | \mu \rangle | \psi_{GS} \rangle}{\langle \psi_S | H_{\text{int}} | \psi_S \rangle^2}$$

$$\gamma = \frac{\langle \psi_S | H_{\text{int}} | \psi_S \rangle^2}{\langle \psi_S | H_{\text{int}} | \psi_S \rangle^2}.$$

$\alpha_0$ is line shape of the broad “continuum” (background) band ($\alpha_0 = \varepsilon_0$ and $\Delta \varepsilon_0$ in Abs and MCD, respectively), $\alpha(\nu_0)$ is the absorption at the center of the interference, $\psi_{GS}$ is the ground state, $\psi_S$ and $\psi_{\nu}$ are broad (spectroscopically allowed) and sharp (spectroscopically forbidden) excited states, $\psi_{\nu}$ is the modified sharp state that results from the interaction of $\psi_0$ with $\psi_S$ (here the interaction Hamiltonian $H_{\text{int}}$ is SOC), $\psi_{\nu}$ is a certain fraction of the background $\psi_S$ state connected to $\psi_{\nu}$ and $\mu$ is the electric dipole transition moment.

The Fano feature, $R(\nu)$, is experimentally defined for Abs by the ratio $\varepsilon_{\text{observed}}/\varepsilon_{\text{background}}$ and for MCD by $\varepsilon_{\text{observed}}/\varepsilon_{\text{background}}$ (Fig. 7). Theoretically, this is determined by the parameters $\nu_0$, $p$, $\lambda$ and $q$ (eqn (4)) that control its energy, height, width...
and shape (e.g., \(q = \infty\), 1 and 0 correspond to a Lorentzian peak, dispersion-like curve and Lorentzian dip, respectively). The parameter \(p\) reflects a fraction of the broad state (LF \(\Delta E\)) that weakly interacts via SOC with the sharp state. The parameter \(q\) is related to the ratio of the transition-dipole matrix elements that contribute excitations into the SOC-modified sharp state and the fraction of the broad state, while the parameter \(\gamma\) is a measure of the SOC between the sharp and the broad state (scaled down by the overlap of the sharp peak with the broad LF \(\Delta E\)-based continuum).

Fig. 7A and B give the analyses of the Abs and MCD Fano features. The \(\gamma\) and \(p\) parameters remain essentially unchanged in going from Abs to MCD. This is in line with the expectation that both the SOC strength (related to \(\gamma\)) and the parameter \(p\) (in eqn (5)) are independent of the Abs/MCD selection rules. Note the inter-state SOC off-diagonal matrix element is estimated from \(\gamma\) to be \(\sim 100 \text{ cm}^{-1}\), indicating a weak SOC interaction. However, there is a significant difference between Fano \(q_{\text{Abs}}\) and \(q_{\text{MCD}}\) parameters (0.15 and 5.0) that reflects the different selection rules in Abs vs. MCD. For Abs, all of the populated sublevels of the \(S = 2\) ground state, \(M_s = 0, \pm 1\) and \(\pm 2\), contribute to the spin-allowed transition to the LF \(\Delta E\) excited state. Among these only the \(M_s = -1\) sublevel contributes to the spin-forbidden Fano transition. For MCD, from all the ground-state \(M_s = 0, \pm 1\) and \(\pm 2\) sublevels, only the \(M_s = -1\) sublevel contributes to both the Fano spin-forbidden and spin-allowed positive MCD intensity. Therefore, the fraction of the ground-state sublevels contributing to the Fano transition is larger in MCD than in Abs, which results in \(q_{\text{MCD}} > q_{\text{Abs}}\) (eqn (6)).

The above FC (and Fano) analyses enable the experimental characterization of the low-lying, spectroscopically spin-allowed and spin-forbidden, excited states in the \(S = 2\) Fe\(^{3+}\)=O complex along with their in-state and inter-state SOC interactions. As a result, the experimental PESs of these excited states can be plotted along the Fe-oxo coordinate (Fig. 8A).

Correlating \textit{ab initio} multireference (CASPT2) calculations with these experimental data, the lowest-energy \(S = 2\) (LF \(\Delta E\)) excited state along with two close-lying \(S = 1\) excited states (LF \(\Delta E\) and \(\Delta A_1\)) were identified. In fact, the LF \(\Delta E\) is quasi-degenerate with LF \(\Delta E\) at the CASPT2 equilibrium geometry (left column in Fig. 8B). In going from non-relativistic to relativistic (CASPT2-based) calculations (from left to right column in Fig. 8B), a strong inter-state SOC interaction of the \(\Delta E\) state with the RCP component of the in-state SOC-perturbed LF \(\Delta E\) is revealed, while there is a weak inter-state SOC of the \(\Delta A_1\) with the LF \(\Delta E\)-based LCP component (i.e., the origin of the Fano interference). These produce four MCD-active states (labelled as 1–4 in Fig. 8B according to their relative energies). States 1 and 3 contribute positive MCD intensity (LCP transitions), while states 2 and 4 contribute negative MCD intensity (RCP transitions). These calculations nicely correlate with the experimental relative energies of states 1–4 (PES energies taken at the vertical line in Fig. 8A). Furthermore, the calculated PESs associated with these states (Fig. 8C) reproduce the characteristics of the experimental PESs from Fig. 8A: (i) The strong SOC interaction between \(\Delta E\) and LF \(\Delta E\) in the vicinity of

<table>
<thead>
<tr>
<th>State(^a)</th>
<th>(\Delta E_{\text{vertical}}) (^b) (\text{cm}^{-1})</th>
<th>(\Delta \gamma_{\text{Fe-O}}) (^d) (\text{cm}^{-1})</th>
<th>(\gamma_{\text{Fe-O}}) (\text{Å}^{-1})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ground state</td>
<td>0.0</td>
<td>820 (^a)</td>
<td>0.0</td>
</tr>
<tr>
<td>1</td>
<td>10 160</td>
<td>620</td>
<td>0.16</td>
</tr>
<tr>
<td>2</td>
<td>10 700</td>
<td>710</td>
<td>0.13</td>
</tr>
<tr>
<td>3</td>
<td>12 355</td>
<td>880</td>
<td>0.08</td>
</tr>
<tr>
<td>4</td>
<td>19 500</td>
<td>490</td>
<td>0.21</td>
</tr>
</tbody>
</table>

\(^a\) States are labeled as in Fig. 8. \(^b\) Electronic vertical transition from \(\Delta A_1\) ground state to highest-intensity component of Frank-Condon progression. \(^c\) Frequency of excited-state Fe-oxo stretching mode. \(^d\) Distortion of excited-state geometry along Fe-oxo stretching mode calculated from eqn (1) in the text. \(^e\) Electronic vertical transition from \(\Delta A_1\) ground state at CASPT2 equilibrium geometry (with Fe-oxo bond length of 1.6 Å). \(^f\) Estimated from 1/2 \(\sqrt{\kappa / \mu}\) where \(\kappa\) obtained from fit of CASPT2 potential energy surfaces with third-order Taylor expansion surfaces (fitted SOC-perturbed PESs from Fig. 8). \(^g\) The \(\Delta \gamma_{\text{Fe-O}}\) taken as difference between equilibrium geometries of \(\Delta A_1\) state and excited state. \(^h\) Taken from ref. 34. \(^i\) Taken from Table S2 in ref. 22.
the ground-state equilibrium geometry produces PESs 1 and 3 (green and blue lines in Fig. 8C), the latter of which is much less shifted and distorted as compared to the first; (ii) PES 2 (red line), whose shape is unaffected by weak SOC with state 4 (red bar), is less distorted than PES 1 and more than PES 3; PES 5 (magenta line), which corresponds to the oxo-to-Fe CT state, is the most distorted and shifted among all of the PESs investigated. More quantitative evaluations of the calculated PESs and their comparisons with experiment are provided in Table 2.

These spectroscopically-calibrated calculations can be further extended to analyses of the characters in the ground- and excited-state wavefunctions, and their evolutions along the Fe–O bond coordinate. This inspection reveals important electronic configuration interactions that contribute to the characters of the ground $^3A_1$ and the lowest excited $^5E$ state. For the equilibrium Fe–O bond length, the $^3A_1$ is characterized by the dominant Fe$^{IV}(S=2)$O$^{2−}\pi_z\pi^2_x\pi^2_yd_1^1d_0^0d_{1z}^1d_{xz}^1d_{yz}^1$ configuration (depicted in Fig. 5A) with a minor contribution from the Fe$^{III}(S=5/2)$O$^{−}\pi^2_x\pi^2_yd_1^1d_0^0d_{1z}^1d_{xz}^1d_{yz}^1$ configuration that results from spin polarization of the $\alpha e^−$ from the oxo $\sigma_z$ into the $d_z^2$ orbital (Fig. 8D, top panel left). In elongating the Fe–O bond, the weight of the first and latter configuration decreases and increases, respectively, such that the latter becomes dominant at Fe–O distances longer than 1.88 Å (Fig. 8D, top panel right). Thus for this elongation (relevant for TS in HAA), the $^3A_1$ state gains strong oxyl character with the unpaired $\alpha e^−$ in the oxo $\sigma_z$ orbital (directed along the z axis of the Fe–O bond). For the $^5E$ state at the Fe–O bond length of the ground state, the character is mostly the Fe$^{IV}(S=2)$O$^{2−}\pi_z\pi^2_x\pi^2_yd_1^1d_0^0d_{1z}^1d_{xz}^1d_{yz}^1$ configuration (Fig. 5B), blended with a minor component of the Fe$^{III}(S=5/2)$O$^{−}\pi^2_x\pi^2_yd_1^1d_0^0d_{1z}^1d_{xz}^1d_{yz}^1$ configuration (Fig. 8D, lower panel left shown only for the LCP component). The weights of these two configurations evolve such that the latter becomes dominant for distances larger than 1.72 Å (Fig. 8D, lower panel right). Thus, this $^5E$ state becomes strongly oxyl in character at the TS for HAA with the unpaired $\alpha e^−$ in the oxo $\pi_x/\pi_y$ orbital (perpendicular to the axis of the Fe–O bond) with five $\alpha e^−$ on the Fe$^{III}$ center. Finally, the second excited state, the oxo-to-Fe CT state, retains its character over entire Fe–O PES, i.e., this CT

Scheme 2 The $d\pi^*$ and $d\sigma^*$ FMOs interacting with the electron-donating C–H $\sigma$ orbital defining possible H-atom abstraction reaction coordinates for an $S = 2$ NHFe$^{IV}$=O species (right).

Scheme 3 Vibronically resolved VT MCD spectra calibrate the theoretical PESs allowing the evaluation of their wavefunction evolution and FMOs along the Fe–O bond stretch that govern mechanism for H-atom abstraction in $S = 1$ vs. $S = 2$ NHFe$^{IV}$=O species.
state has oxyl character with the unpaired α e− in the oxo πcxy orbital with three α e− on the FeIII center. This corresponds to an oxyl FeIII of intermediate spin (SFe = 3/2) at the TS. Thus these PESs define FMOs that have directionality (π vs. σ; see Scheme 2) and reactivity in being holes on the oxo at the TS as required for efficient overlap for HAA.

Concluding comments

LT MCD spectroscopy is a powerful method for investigation of transitions into low-lying excited states of both the S = 1 and S = 2 NHFeIV=O active sites, particularly of LF (d-d) transitions. The great advantage of MCD lies in decoupling of the overlapping bands due to their different selection rules that gives different signs of bands as well as their temperature behaviors that further provide information on the polarization of transitions and thus their unambiguous assignments. In addition, the MCD spectra exhibit vibronic structures that provide information on the excited-state PESs along the Fe–O coordinate and their (SOC) interactions. These spectroscopic results, in combination with ab initio multireference calculations, reveal the nature of the FMOs of different electronic states (and associated FMOs) evolve along the Fe–O PESs giving rise to oxyl FeIII character at Fe–O distances relevant to the TS for H-atom abstraction. For these TS-active site in controlling reaction selectivity as in the halogenation chemistry.

Acknowledgements

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References


32 Ratio of the MCD transition dipole moment ($C_0$) and dipole strength ($D_0$). Experimentally, this ratio is estimated using $C_0/D_0 = k_BT/\mu_B H (\Delta \varepsilon/\varepsilon)_{\text{max}}$, where $k_B$ and $\mu_B$ are Boltzmann constant and Bohr magneton, $T$ is the temperature, $H$ is the applied magnetic field, $\varepsilon$ is molar extinction coefficient obtained from absorption and $\Delta \varepsilon$ is the difference between the molar extinction coefficients for left- and right-circularly polarized light. In general, $C_0/D_0 > 0.1$ for ligand-field transitions, while charge-transfer transitions are characterized by $C_0/D_0 < 0.1$.


42 To estimate of the inter-state SOC matrix element between the broad and sharp state, we used eqn (7) considering that only 7% of the broad positive band overlaps with the sharp peak (Fig. 6): $\langle \Psi_b | H_{\text{int}} | \Psi_s \rangle = (0.07)^{-1} \sqrt{\gamma/\pi}$. **