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Energetics of Proton-Coupled Electron Transfer in High-Valent $Mn_2(\mu\text{-}O)_2$ Systems: Models for Water Oxidation by the Oxygen-Evolving Complex of Photosystem II

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The reducing equivalents generated during oxygenic photosynthesis are ultimately supplied by the four-electron oxidation of water to dioxygen.[1,2] The photochemical charge separation of the chromophore P_{680} initiates transfer of these equivalents to the reducing side of photosystem II (PSII) one electron at a time. On the oxidizing side, the hole produced in P_{680} is filled by an electron transferred from the oxygen evolving complex (OEC), a tetranuclear manganese cluster at which water oxidation occurs. A redox-active tyrosine (Y_{161} of the D1 polypeptide in PSII, also known as Y_Z) has been implicated as an electron transfer intermediate between the OEC and P_{680}.[1] The oxidizing equivalents required for water oxidation are stored by the OEC, resulting in an enzyme that traverses five oxidation states during catalysis. Each of the series of oxidation states produced in the OEC is referred to as an S-state in the Kok formalism,[3] with the most reduced state being S_0 and the most oxidized state S_4. The rate-limiting step for water oxidation in the $S_3 \rightarrow S_4 \rightarrow S_0$ transition is reduction of $Y_Z{}^{\bullet}$.[4] Recently, Babcock[5,6] and Britt[7,8] have each proposed that $Y_Z{}^{\bullet}$ may participate directly in water oxidation. Furthermore, Babcock has proposed[6] a detailed chemical mechanism that invokes H-atom abstraction from water or hydroxide bound to the Mn_4 cluster. It has been argued that this is not thermodynamically feasible since the O−H bond dissociation energy [*D*(O−H)] of water is 119 kcal/mol, about 35 kcal/mol higher than in tyrosine. However, no *D*(O−H) values have been available for water-derived ligands bound to manganese in an appropriate model. Herein we present thermodynamic data that can test this generalized proposal for the first time.

Structural studies of the intermediate S-states of the OEC by X-ray absorption spectroscopy have suggested a structural motif for the tetranuclear manganese cluster consisting of a pair of bis oxo-bridged, high-valent Mn dimers $[Mn_2(\mu\text{-}O)_2]$.[9,10] In order to understand the possible roles for the $Mn_2(\mu\text{-}O)_2$ structural unit in the chemistry of the OEC, we have studied a series of model complexes,[11−14] $[Mn^{IV}(X\text{-salpn})(\mu\text{-}O)]_2$ (salpn = *N*,*N*′-bis(salicylidene)-1,3-propanediamine, X = electron donating or accepting groups on the phenolate ring) with 2.7 Å Mn−Mn distances,[15] as observed in the OEC. These complexes are also effective catalysts of H_2O_2 disproportionation,[16,17] reminiscent of the catalase activity in the $S_2 \rightarrow S_0$ cycle.[18] Thus, the $[Mn(X\text{-salpn})(\mu\text{-}O)]_2$ complexes are both structural and partial functional models for the OEC. Here, we explore the energetics of proton-coupled electron transfer from protonated $Mn_2(\mu\text{-}O)_2$ units by studying the individual energy contributions of the oxidation and deprotonation steps for $[Mn(X\text{-salpn})(\mu\text{-}O)]_2$ with X = H, 3,5-di(Cl), and 3,5-di(NO_2). Providing a range of 8 pK_a units and 800 mV for each oxidation or protonation state, these complexes allow quantitation of the critical role protons may play in the highly efficient oxidation of the OEC.

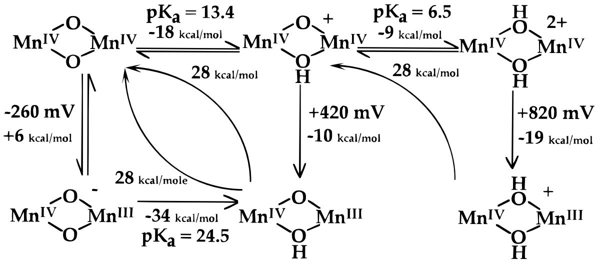

Figure 1. Thermodynamic cycle of protonations and reductions of $[Mn(salpn)(\mu\text{-}O)]_2$ in acetonitrile, showing energies for reaction 1 (eq 1) in the text across the diagonals. Each Mn shown is bound to the tetradentate salpn ligand.

For each of the three derivatives, the pK_a was determined in acetonitrile for both the protonated $Mn_2{}^{IV}$ and $Mn^{III}Mn^{IV}$ dimers. Titrations of various acids for which the pK_a values are known in acetonitrile were monitored spectrophotometrically for the $Mn_2{}^{IV}$ dimers[19] and electrochemically for the $Mn^{III}Mn^{IV}$ dimers.[20] Potentials were determined by cyclic voltammetry (CV) for the $Mn_2{}^{IV} \rightarrow Mn^{III}Mn^{IV}$ reduction of both the bis(μ-O) [reversible] and (μ-O,μ-OH) [electrochemically irreversible] protonation states for each derivative.[21] In addition, the pK_a and the reduction potential for the doubly protonated complex [Mn^{IV}-

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(19) As reported previously for the X = H and 3,5-di(Cl) derivatives.[11] The pK_a for X = 3,5-di(NO_2) was determined by titration of triflic acid into CH_3CN solutions buffered by 10 mM water (pK_a = 2.3) and by 10 mM DMF (pK_a = 6.0). pK_a values in CH_3CN for the reference acids are from the following: Izutsu, K. *Acid-Base Dissociation Constants in Dipolar Aprotic Solvents*, IUPAC Chemical Data Series 35; Blackwell Scientific Publications: Brookline Village, MA, 1990.

(20) Titrations were performed with 20 mM tetrabutylammonium hexafluorophosphate ($TBAPF_6$) electrolyte in CH_3CN for the 3,5-di(NO_2) derivative and in CH_2Cl_2 for the others. Protonation of the $Mn^{III}Mn^{IV}$ dimer was monitored by loss of the anodic current relative to the cathodic current in the $Mn_2{}^{IV} \rightarrow Mn^{III}Mn^{IV}$ couple upon titration of acid into the unprotonated $Mn_2{}^{IV}$. Values determined in CH_2Cl_2 are corrected by 0.9 units on the basis of comparison of values determined for $[Mn^{IV}(salpn)]_2(\mu\text{-}O,OH)^+$ in both solvents using the CH_3CN reference values. Reference acids: for X = 3,5-di(NO_2), lutidinium (pK_a = 14.05); for X = 3,5-di(Cl), acetic acid (pK_a = 22.3) and a 1,1,3,3-tetramethylguanidine (tmg) buffer (pK_a = 23.3); for X = H, tmg buffer and phenol (pK_a = 27).

(21) Conditions: solvents as indicated in Table 1 with 100 mM $TBAPF_6$ electrolyte; scan rate = 50 or 100 mV/s; glassy carbon working electrode, Pt wire auxiliary electrode, saturated calomel (SCE) reference electrode. Values reported vs the normal hydrogen electrode (NHE) [$E_{1/2}(NHE) \approx E_{1/2}(SCE) + 0.244$ V]. The CV was taken for $[Mn^{IV}(3,5\text{-di}(NO_2)salpn)(\mu\text{-}O)]_2$ in both CH_2Cl_2 and CH_3CN, showing reversible $Mn_2{}^{IV} \rightarrow Mn^{III}Mn^{IV}$ couples within 30 mV for the two solvents.

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Table 1. Energies of Oxidation Plus Deprotonation and O−H Bond Dissociation for $Mn_2(\mu\text{-O},\mu\text{-OH})$ Complexes

ligand	redox couple	E_c $(\mu\text{-O})_2$[a]	E_c $(\mu\text{-O},\mu\text{-OH})$[a]	pK_a (ox)	pK_a (red)	ΔG, D(O−H)[b]	ref
salpn	(IV,IV) → (III,IV)	-0.26^c	$+0.42^d$	13.4 ± 0.2^d	24.5 ± 0.7^e	$28,^d$ 76	this work, 11
3,5-di(Cl)salpn	(IV,IV) → (III,IV)	0.0^c	$+0.64^d$	10.8 ± 0.3^d	20.5 ± 1.0^e	$29,^d$ 77	this work, 11
3,5-di(NO_2)salpn	(IV,IV) → (III,IV)	$+0.51^d$	$+1.1^d$	5.0 ± 0.8^d	13.3 ± 0.7^d	$31,^d$ 79	this work
bpy	(III,IV) → (III,III)		$+1.1^f$	2.3^f		$29,^f$ 84	23, 24

[a] Although the CVs for the unprotonated complexes show reversible electrochemistry, cathodic potentials (E_c) are given for consistency with the irreversible data for the protonated complexes. All potentials are given in volts vs NHE. [b] ΔG is for the half reaction, as in eq 1 in the text, and is specific for the indicated solvent. D(O−H) is the bond dissociation energy as described in the text. Both values are given in kilocalories per mole, with estimated error bars of 2−3 kcal/mol. [c] Measured in CH_2Cl_2. [d] Measured in CH_3CN. [e] Measured in CH_2Cl_2 and corrected to reflect pK_a values in CH_3CN.[18] [f] Measured in water.

$(salpn)(\mu\text{-OH})]_2^{2+}$ have been determined.[22] The thermodynamic cycle of proton and redox reactions for the unsubstituted salpn system is shown in Figure 1. Although the pK_a values and reduction potentials for each derivative vary greatly, the total ΔG for reaction 1 (eq 1) in acetonitrile varies only slightly, averaging 29 ± 3 kcal/mol relative to the normal hydrogen electrode (NHE). Comparison to the $[Mn(bpy)_2(\mu\text{-O})]_2^{n+}$ system[23,24] shows that a similar ΔG value may be obtained in water with a $Mn_2(\mu\text{-O})_2$ complex with different auxiliary ligands, charge, and oxidation state couple ($Mn^{III}Mn^{IV} \rightarrow Mn_2^{III}$). These data are presented in Table 1.

$$Mn_2^{III/IV}H \rightarrow Mn_2^{IV/IV} + H^+ + e^- \qquad (1)$$

We may now use the data from model complexes to evaluate the thermodynamic constraints of the OEC. The energetics of photosystem II leave little margin for inefficiency in the required electron transfers. At pH 5.0, the operative acidity for lumenal membranes, the four-electron oxidation of water requires ~3.6 V.[2] The potential of the $S_0 \rightarrow S_1$ transition is estimated to be about 0.3−0.6 V, based on the ability of Y_D^+ ($E° \approx 0.75$ V), the radical of a second redox-active tyrosine in PSII, to oxidize S_0 to S_1[25] and the ability of certain reductants examined by Yocum and co-workers to reduce S_1 to S_0.[26] Thus, about 3.1 V must be acquired in the remaining three S-state advancements. After charge separation of P_{680}, 1.2 V of oxidizing power are available from P_{680}^+ for oxidation of Y_Z.[2] Babcock et al. have estimated that $Y_Z^\bullet$ is capable of providing 1.0 V of oxidizing power to the manganese cluster.[2] Even if the $S_3 \rightarrow S_4$ transition does not require a significant driving force for electron transfer, so that 1.1 V is available to the OEC in this final oxidation, a minimum of close to 1.0 V, or all of the oxidizing power available from $Y_Z^\bullet$, must be accommodated by the OEC in each of the intermediate S-state transitions. These successive electron transfers must be accomplished without the potential of the OEC becoming so high that early, incomplete oxidation of water or another substrate occurs or that subsequent oxidation by $Y_Z^\bullet$ is energetically disfavored. One way by which these electrochemical requirements may be satisfied is by the coupling of proton transfer to the electron transfer. The average energy for proton-coupled electron transfer in the X-salpn systems in acetonitrile is 29 ± 3 kcal/mol, which is equal to 1.26 ± 0.13 V, while the bpy system shows a similar energy in water. Thus, changing the Mn ligands allows tuning of this dielectric-dependent ΔG around a value of about 1.2 V. Within experimental error, this is precisely the energy which must be accommodated by the OEC upon each of the intermediate S-state advancements.

While the above analysis addresses the general case of energy transfer from Y_Z to the OEC, these data may alternatively be considered in terms of relative bond energies to test the specific case in which the energy transfer from $Y_Z^\bullet$ to the OEC occurs as abstraction of $H^\bullet$ from a water-derived ligand bound to manganese by $Y_Z^\bullet$, as proposed by Babcock.[6] While [D(O−H)] for water is much too high at 119 kcal/mol for hydrogen atom extraction by tyrosyl radical, with a D(O−H) for tyrosine calculated by Lind et al.[27] to be 86.5 kcal/mol, values for D(O−H) of water-derived ligands bound to manganese clusters like those in the OEC have not been reported. The value of D(O−H) for the hydroxo bridge in a protonated $Mn_2(\mu\text{-O})_2$ system may be calculated from pK_a values and reduction potentials in a manner adapted from that used by Mayer and co-workers[28,29] in studying hydrogen abstraction from toluene by permanganate. We determine D(O−H) of the hydroxo bridge in the manganese dimers to be 77 ± 3 kcal/mol. This value is supported by the ability of $[Mn^{IV}(salpn)(\mu\text{-O})]_2$ to abstract a hydrogen atom from 2,4-di(*tert*-butyl)phenol (D(O−H) ≈ 82 kcal/mol) but not from phenol (D(C−H) = 90 kcal/mol).[30,31] The value of D(O−H) for the hydroxo bridge is lower than that for tyrosine, so that the reaction in eq 2 should proceed easily. Thus, the energetics of hydrogen atom transfer from a protonated, high-valent $Mn_2(\mu\text{-O})_2$ center is consistent with the mechanism proposed by Babcock[6] for oxidation of the OEC.

$$\text{tyrosyl}^\bullet + Mn_2(\mu\text{-O},\mu\text{-OH})^{n+} \rightarrow \text{tyrosine} + Mn_2(\mu\text{-O})_2^{n+} \qquad (2)$$

In summary, the data presented here show that, in the general case, high-valent $Mn_2(\mu\text{-O})_2$ centers in various protonation states provide a nearly perfect energetic match for efficient energy transfer from $Y_Z^\bullet$. This may explain why photosystem II has evolved to use this unusual type of metal center which appears to be unique to PSII among biological systems. When applied to the specific mechanism proposed by Babcock and co-workers, the analysis of D(O−H) energies indicates that the likely structural motif of the OEC is thermodynamically consistent with hydrogen atom transfer from the OEC to $Y_Z^\bullet$.

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(22) For solubility reasons, the potential actually determined was for $[Mn^{IV}(salpn)]_2(\mu\text{-OH},\mu\text{-OCH}_3)^{2+}$ from the difference in potential from $[Mn^{IV}(salpn)]_2(\mu\text{-O},\mu\text{-OCH}_3)^+$ at −35 °C. The cathodic potential for $[Mn(salpn)]_2(\mu\text{-O},\mu\text{-OH})^+$ is less than 20 mV higher than that for $[Mn(salpn)]_2(\mu\text{-O},\mu\text{-OCH}_3)^+$ in either CH_3CN or CH_2Cl_2. The pK_a in acetonitrile reported here (6.5 ± 0.7) has been redetermined more accurately than our previously reported estimate[12] by titration of H_2SO_4 (pK_a = 7.25) monitored by stopped flow measurements.

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