

REVIEW ARTICLE

STRUCTURAL PROPOSALS FOR THE MANGANESE CENTERS OF THE OXYGEN EVOLVING COMPLEX: AN INORGANIC CHEMIST'S PERSPECTIVE*

VINCENT L. PECORARO

Department of Chemistry, University of Michigan, Ann Arbor, MI 48109, USA

(Received 26 February 1988; accepted 1 March 1988)

Abstract—This contribution provides an analysis of the basic coordination tendencies of manganese with specific emphasis on the biological chemistry of this element. The review is broken into four parts. First, a discussion of the basic coordination chemistry of manganese is mononuclear and multinuclear environments is presented. Second, the biophysical data essential to the development of models for the active center of the photosystem is examined. Third, recently reported mononuclear and cluster manganese compounds are profiled with an emphasis on relating the physical parameters of the models to the structure and function of the enzymatic system. Finally, a comparison is made between the OEC and other known manganoenzymes which metabolize the O_2^{2-} moiety.

INTRODUCTION

Direct biochemical and biophysical studies of the chloroplast associated oxygen evolving complex have provided significant insight into the workings and structure of the enzyme complex responsible for the photosynthetic water splitting reaction (Amesz, 1983; Dismukes, 1986). Among the important conclusions gleaned from these studies are (1) that each reaction center operates independently requiring four photochemically derived oxidizing equivalents per mole of oxygen formed, (2) that four manganese ions are required for catalysis, and (3) that some or all of the manganese ions are associated in cluster(s). Unfortunately, the interpretation of the accumulated biophysical data has been hindered due to the lack of structurally and chemically well defined manganese complexes that are biochemically relevant. Inorganic chemists have been enticed into photosynthetic research by sensing this deficiency. While not directly dealing with either chloroplasts or highly purified PSII core particles, these investigators have aimed at establishing benchmark complexes that should provide a significant contribution to the understanding of both structural and mechanistic properties of manganese in biological systems.

On the surface, it might not appear that model compounds could provide insight into the metallo active site structure or the mechanistic pathway operating in the water splitting process due to the complexity of the enzymic system which one cannot, at least in the near future, hope to mimic artificially. However, synthetic complexes can provide a wealth of information pertaining to reasonable structural types, appropriate ligands for high oxidation poten-

tials, reactivity mechanisms of clusters and magnetic and spectroscopic properties of manganese in variable oxidation states or nuclearity compounds. In the paragraphs below, I will discuss those areas where model studies are providing valuable information for the photobiologist.

BASIC COORDINATION CHEMISTRY OF MANGANESE

Mononuclear centers

It is important to establish the basic coordination (Cotton, A. and G. Wilkinson, 1980; Huheey, 1983) and chemical properties of manganese in different oxidation states before proceeding to complexes directly relating to water oxidation. Table 1 presents some of the basic properties of manganese in the +2, +3 and +4 oxidation states for *mononuclear* complexes. The degree to which manganese ions in *multinuclear* complexes comply to these parameters will depend on the extent of interaction between the metal centers. As discussed later, the magnitude of this interaction and the resulting modifications in the anticipated properties of the metal centers is under investigation in numerous laboratories.

With few exceptions [e.g., complexes containing very strong field ligands (Gross and Kaim, 1987a,b)] virtually all Mn(II), Mn(III) and Mn(IV) complexes are high spin. The *d* electron configuration of each oxidation state imparts certain preferences for coordination geometry. A simple ligand field model demonstrates that Mn(II), with 5 unpaired *d* electrons, has the same ligand field stabilization energy (LFSE) in all geometries. Thus, complexes of Mn(II) usually minimize ligand-ligand repulsions by forming tetrahedral or octahedral materials. In great contrast, Mn(IV) compounds (Kessissoglou *et al.*, 1986a; Kessissoglou *et al.*, 1987b; Pavacic *et al.*, 1986; Hartmann *et al.*, 1984) are strongly stabilized only as six-coordinate octahedral or pseudo-octahedral species since the 3 unpaired electrons con-

*Editor's note: This paper was presented at a symposium on Photosystem II and Oxygen Evolution in the Photosynthesis Apparatus, organized by G. T. Babcock, 15th Annual Meeting of the American Society for Photobiology.

Table 1. Properties of mononuclear manganese ions

Property	Mn(II)	Mn(III)	Mn(IV)
<i>d</i> Electron configuration	d^5	d^4	d^3
Coordination preference	6	6* or 5	6
Orbital configuration*	6A_1	5E	4A_2
<i>d-d</i> Transitions*‡	None	${}^5E \rightarrow {}^5T$ [${}^5B_2 \rightarrow {}^5B_1$]§ [${}^5E \rightarrow {}^5B_1$]§ [${}^5A_1 \rightarrow {}^5B_1$]§	${}^4A_2 \rightarrow {}^4T_2$ ${}^4A_2 \rightarrow {}^4T_1$
μ_{eff} (B.M.)	5.9	4.9	3.9
Lewis acidity strong	Strong	Strong	Very
Exchange kinetics	Very rapid	Rapid	Very slow
Oxidant	Very poor	Good	Good
Reductant	Poor	Good	Very poor

*Octahedral, high spin configuration.

‡Spin-allowed transitions.

§Tetragonally distorted octahedron (D_{4h} point group symmetry).

tribute $-12Dq$ LFSE in this geometry. The Mn(III) ion is least selective due to the one unpaired electron in the doubly degenerate e_g (antibonding) orbitals in octahedral symmetry. One expects a first order Jahn–Teller distortion away from octahedral symmetry, usually through a tetragonal elongation, which breaks the degeneracy of the e_g set and lowers the overall energy of the system (Stults *et al.*, 1979). Alternatively the symmetry can be broken by adopting 5 coordinate structures such as trigonal bipyramidal (Bellito *et al.*, 1971), square pyramidal (Pecoraro and Butler, 1986; Boucher and Day, 1977; VanAtta *et al.*, 1987) or polyhedra along a Berry type (Muerterties and Guggenberger, 1974) distortion pathway. One consequence of this lack of geometrical preference is that sequential oxidations of a manganese compound from Mn(II) to Mn(III) to Mn(IV) are often accompanied by major structural changes. One electron processes in the cyclic voltammetry of manganese species are often quasireversible or irreversible due to these structural changes which can occur on the time scale of the experiment (Coleman *et al.*, 1979; Coleman *et al.*, 1981; Yamaguchi and Sawyer, 1985; Kessissoglou *et al.*, 1987b).

In addition to the structural preferences of manganese compounds, the *d* electron configuration can be used to understand the other properties listed in Table 1. Both Mn(II) (6A_1) and Mn(IV) (4A_2) have orbitally singlet ground states while an octahedral Mn(III) ion (5E) is doubly degenerate. Ions with singlet ground states and small zero-field splittings have long paramagnetic relaxation times (Lewis and Morgan, 1968) while orbitally degenerate ions have shorter relaxation times. This observation has led Sharp (Srinivasan and Sharp, 1986a and 1986b) to study the flash induced enhancements in the NMR spin-lattice relaxation rate (R_1) of sol-

vent protons in PSII particles since both Mn(II) and Mn(IV) are predicted to be more effective relaxation traps than Mn(III).

The electronic spectra of transition elements are notoriously weak due to the symmetry forbidden nature of the transition. Two bands are predicted for Mn(IV), one for Mn(III) in octahedral symmetry and none for Mn(II). The latter is not only symmetry but also spin forbidden. Because the metal centered electronic transitions are predicted to have extinction coefficients $\cong 100 M^{-1} \text{ cm}^{-1}$ they are not expected to make a significant contribution to the visible spectrum of the reaction center. As discussed below, however, changes in the UV-vis spectra with flash events have been assigned to manganese oxidation state changes. These perturbations may arise from charge transfer excitations between the metal and its coordinated ligands.

In the +2, +3 and +4 oxidation states, manganese is classified as a hard Lewis acid. Therefore, it is expected to form the most stable complexes with hard Lewis bases such as oxyanions. Biologically relevant oxyanions include carboxylate, phenolate, alkoxide, hydroxide and oxide oxygen atoms. The nitrogen atoms of imidazole groups would also be expected to be good donors.

An interesting aspect of the coordination chemistry of manganese is the predicted exchange rates of ligands coordinated to the metal. Although many factors determine the actual exchange rate, two appear to dominate. The first is the change in LFSE on forming the intermediate species and the second is the energy required to break the electrostatic interaction between metal and donor. Most octahedral exchange reactions are primarily dissociative in nature, requiring a five coordinate activated complex. Little or no LFSE is lost when Mn(II) or Mn(III) form five coordinate complexes. In con-

trast, ligand dissociation subjects the Mn(IV) ion to a great loss of LFSE which drives up the activation energy for the process. Removing a neutral or anionic ligand from Mn(IV) is much more difficult than from Mn(II) due to electrostatic reasons as well. Thus, Mn(II) exhibits exchange rates (Basolo and Pearson, 1967) of 10^7 s^{-1} , Mn(III) $> 10^5 \text{ s}^{-1}$ and Mn(IV) is predicted to be $\ll 10^{-2} \text{ s}^{-1}$.

The water splitting reaction requires a strong oxidant. Clearly Mn(II) cannot fulfill the oxidizing capacity which is required. It is neither a good oxidant nor a good reductant, although in many reactions it can reduce dioxygen (Coleman and Taylor, 1977; Kessissoglou *et al.*, 1987b; Pecoraro and Butler, 1986; Sawyer *et al.*, 1977) to form Mn(III) or Mn(IV). The Mn(III) ion can function both as a good oxidant and as a good reductant. This dichotomous behavior results from the unpaired electron in the antibonding orbital set. The loss of this electron gives a very stable d^3 electronic configuration. Conversely, adding an electron generates a stable d^5 , half-filled shell configuration. The oxidizing nature of Mn(III) dominates in most cases. The Mn(IV) ion is a good oxidant and poor reductant. It can function both as a one or a two electron acceptor as the Mn(II) and Mn(III) oxidation states are stable. As will be discussed below, it is a common misconception that all Mn(IV) compounds are strongly oxidizing. This is not the case as the Mn(IV) coordination environment often leads to the thermodynamically most stable material.

Multinuclear complexes

The magnetic moments for single ions can be easily calculated*. Because spin-orbit coupling is extremely small for manganese, mononuclear compounds rarely deviate markedly from the spin only values. The magnetic properties of multinuclear manganese centers are more complex and, therefore, often provide insight into the mechanism and extent of interaction between metal centers. The magnitude of the coupling is usually reported in units of cm^{-1} as either J or $2J$. The standard convention is that complexes exhibiting antiferromagnetic coupling have negative J values, while ferromagnetic species have positive J . The J values can be extracted, while also accounting for zero-field splitting and g anisotropy, by fitting (usually to a modification of the Van Vleck equation) the variable temperature magnetic behaviour of the compound. Unfortunately, the tabulation of magnetic moments for multinuclear complexes in the inorganic literature has been inconsistent. Magnetic moments per

manganese ion are presented in some studies while others report moments per complex. These values can be related by dividing the moment per complex by the square root of the total number of manganese atoms.

As a general rule, biologically relevant multinuclear manganese complexes vary between very weak to moderate coupling between centers. Very weak coupling has been noted for diphenolate bridged Mn(II/II)¹ dimers (e.g., [Mn(SALPS)]₂ where $J = -1.6 \text{ cm}^{-1}$; Kessissoglou *et al.*, 1987a) and linear trinuclear manganese acetates (e.g. Mn(III/II/III) (SALADHP)₂(OAc)₄ (methanol)₂ where $J_{12} = -7 \text{ cm}^{-1}$; Li *et al.*, 1988). Stronger coupling ($J = -120 \text{ cm}^{-1}$) is observed for (bipy)₂-Mn(III)(O)₂Mn(IV)(bipy)₂ (Plaskin *et al.*, 1972). Wieghardt has recently reported the largest exchange coupling for a manganese dimer with a J for this complex equal to -414 cm^{-1} (Girerd *et al.*, 1987). Although this J value may appear large, it is actually small when compared to the very strong coupling that is often observed for binuclear copper complexes (Sorrell *et al.*, 1985) (e.g. the protein hemocyanin, which contains a binuclear copper center, is diamagnetic at room temperature with $J < -1200 \text{ cm}^{-1}$). The J coupling in tetranuclear manganese complexes (Vincent and Christou, 1987; Christmas *et al.*, 1987; Vincent *et al.*, 1987b) have ranged from -1.97 cm^{-1} to -22.5 cm^{-1} .

The magnitude of the exchange coupling is related to a number of factors. As expected, direct metal-metal bonding leads to extremely strong coupling. However, the moderate to high oxidation state manganese complexes usually do not exhibit direct metal-metal bonding. For these molecules, a superexchange pathway is an effective mechanism for magnetic interaction. The efficiency of superexchange depends on metal and ligand type, bonding distances and geometry. The metal and bridging ligand atomic orbitals must have sufficient overlap so that the resulting molecular orbital can efficiently delocalize spin over both metal centers. Therefore, deviations in Mn-L-Mn bond angles can significantly modify the exchange coupling by decreasing molecular orbital overlap. The oxidation and spin state of the participating ions is also important. Higher effective nuclear charge and smaller ionic radii lead to shorter Mn-ligand bond lengths. Shorter metal-metal distances occur and, more important, a shorter and more effective exchange pathway for electrons is produced. Within an oxidation state, a shorter Mn-L distance usually leads to stronger coupling. However, a comparison between oxidation states is more dangerous because the spatial orientation of magnetic orbitals (a magnetic orbital is a metal centered atomic orbital that has an unpaired electron) is essential to superexchange coupling. A high spin Mn(III) ion has four unpaired electrons with all the d orbitals but the $d_{x^2-y^2}$ half occupied. In contrast, an Mn(IV) ion

*Throughout the text the abbreviation Mn(W/X/Y/Z) has been used to designate the formal oxidation states of the manganese ions forming the cluster. Thus, Mn(II/II) indicates a dimer containing two Mn(II) ions. Similarly, Mn(III/II/III) will describe a trinuclear complex with two Mn(III) and one Mn(II) ions.

(d^5) also has two unfilled d orbitals ($d_{x^2-y^2}$ and d_{z^2}). If the magnetic exchange pathway for an Mn(III/III) dimer proceeds through the d_{z^2} orbitals of the metal ions, the oxidation of this compound to an Mn(IV/IV) species could result in dramatically different coupling since the optimal exchange path has been removed. The complexity of manganese magnetism is illustrated by a series of dimers which are antiferromagnetically coupled in the Mn(III/III) and Mn(III/IV) complexes and ferromagnetically coupled in the Mn(III/III) dimers (Girerd *et al.*, 1987). As discussed below, a model has been proposed for the magnetic properties of the OEC manganese cluster using a tetranuclear cluster that has both antiferromagnetic and ferromagnetic coupling (dePaula *et al.*, 1986).

Mixed valence complexes have special relevance to oxygenic photosynthesis as the manganese cluster in the S_2 state of the OEC is believed to fit this formulation (Dismukes and Siderer, 1981). A mixed valence complex is any multinuclear material in which an element is present in more than one oxidation state. Robin and Day (Robin and Day, 1967; Day, 1980) have presented a classification scheme for such complexes. Class I mixed valence compounds are valence localized systems. In this sense, each site acts independently of the other. This results in manganese centers with distinct chemical properties (e.g. wide variations in bond lengths between centers for identical ligand types). Rarely does a coordination complex completely fit this definition; however, compounds such as Mn(III/II/III) (SALADHP)₂(OAc)₄(CH₃OH)₂ described below (Li *et al.*, 1988) deviate only slightly from the limit of non-interaction. Class II complexes contain centers that are crystallographically dissimilar; however, many of the properties of the ions are similar. The classic example of such a material in manganese coordination chemistry is (bipy)₂Mn(III)(O)₂-Mn(IV)(bipy)₂ (Plaskin *et al.*, 1972). Class II complexes often exhibit Intervalence Charge Transfer Excitations (ICT) at low energy with large bandwidths. An ICT band for this complex is observed at 12 000 cm⁻¹ with a 4600 cm⁻¹ band width. Dismukes (Dismukes and Matis, 1984) has reported an ICT band at \approx 12 800 cm⁻¹ ($\epsilon = 500$) for the S_2 and S_3 states of the OEC suggesting that this center is a Class II complex. Theoretical models (Piepho *et al.*, 1978; Allen and Hush, 1967) have been presented for the ICT bands for those interested in the basis of this effect. Class III complexes are valence delocalized systems in which the ions cannot be distinguished crystallographically. Strong antiferromagnetic coupling in a system, such as observed for the Fe₄S₄ cubanes of oxidized ferredoxins, can lead to class III compounds. There are no examples of class III complexes within the biologically relevant manganese coordination chemistry literature.

When considering the interaction between ions in clusters, one must also consider the nature of the

bridging ligand. Bifunctional, aromatic ligands such as pyrazine or 4,4'-bipyridal have been employed to study electron transfer between ions in mixed valence complexes. Other groups, with more biological application, which are known to facilitate metal-metal bridges are Cl⁻, O²⁻, acetate and imidazole. EXAFS analysis of the OEC (Cole *et al.*, 1987; Kirby *et al.*, 1981a; Goodin *et al.*, 1984; Yachandra *et al.*, 1986) indicates a very short Mn-(C,N,O) distance of 1.75 Å that has been assigned to a bridging μ_2 -O²⁻ (or μ_3 -O²⁻) moiety. Manganese seems to have a high affinity towards forming acetate bridged species. Examples (Davies *et al.*, 1973; Bonadies *et al.*, 1988) of chain structures that use acetate as an intermolecular bridge between two adjacent manganese units and intramolecular acetate bridges that provide structural integrity to multinuclear (Li *et al.*, 1988; Bonadies *et al.*, 1988; Kessissoglou *et al.*, 1988a,b; Lis, 1980; Vincent and Christou, 1987; Christmas *et al.*, 1987; Vincent *et al.*, 1987a,b; Sheats *et al.*, 1987; Wieghardt *et al.*, 1985; Diril *et al.*, 1987; Lis, T. and Jezowska-Trzebiatowski, 1977; Bashkin *et al.*, 1987; Mathur *et al.*, 1987) complexes now exist. Exchange coupling between manganese is heightened with the 3-atom Mn-O-Mn oxo bridge relative to the 5-atom Mn-O-C-O-Mn acetate bridge for the reasons outlined above.

The role of chloride in oxygen evolution is intriguing. A chloride dependence for S -state advancement (Theg *et al.*, 1984; Itoh *et al.*, 1984) has been observed and explained (Sandusky and Yocum, 1983, 1984, 1986) through a structural requirement for the manganese cluster. In this model, chloride is needed to impart structural integrity and promote electron transfer between ions at the active site. This proposal is consistent with observed synthetically prepared halide bridged dimers (Mathur *et al.*, 1987) and a tetranuclear Mn(III/III/III/IV) cluster (Bashkin *et al.*, 1987).

STRUCTURAL PROPOSALS

The nuclearity of the active site manganese represents one of the most controversial topics in oxygen evolving chemistry. There is general agreement that some of the manganese ions are associated in a cluster; however, whether all of the manganese atoms form a single tetranuclear cluster or only a few are associated as trinuclear or binuclear units with mononuclear centers also being present is still being debated. Most of the structural proposals are based primarily on the behavior of the S_2 state epr spectra. The multiline (Dismukes and Siderer, 1981) and $g = 4.1$ signals (Casey and Sauer, 1984) have been examined under slightly altered conditions. Zimmerman and Rutherford (1986) have shown that the choice of cryoprotectant (e.g. using sucrose or ethylene glycol) can strongly influence the epr spectrum. PS II preparations containing 30% ethylene

glycol generates mostly the multiline signal when illuminated at 200 K. In contrast, illumination of membranes at 200 K using 0.4 M sucrose as cryoprotectant generates both the $g = 4.1$ and $g = 2$ multiline signals. Furthermore, the nuclear hyperfine interaction in the multiline signature is slightly different in samples containing 0.4 M sucrose versus 30% ethylene glycol (dePaula *et al.*, 1987). Because the epr active manganese center appears to be extremely sensitive to sample preparation and environment, conflicting epr data, especially regarding the temperature dependence of the multiline signal discussed below, have appeared (dePaula *et al.*, 1986, 1987; Hansson *et al.*, 1987). As a result, different structural proposals have been advanced which can be considered as single center [Brudvig] (dePaula *et al.*, 1985, 1986; Brudvig and Crabtree, 1986) or dual center [Vanngard] models (Hansson *et al.*, 1987). In addition to the stoichiometry of the centers, these models are distinguished by the explanations given for the origin of the $g = 4.1$ signal and the magnetic properties of the cluster that yields the multiline feature. The single center proposal requires a tetranuclear cluster that can undergo subtle conformational changes, while the second invokes two manganese centers in an electron transfer equilibrium.

Single center model

Brudvig has proposed that a tetrameric cubane type cluster undergoes sequential one electron oxidations until an adamantane type intermediate that liberates oxygen is formed (Brudvig and Crabtree, 1986). This view is supported by the temperature dependence of the S_2 state multiline signal which shows non-Curie law behavior. The data were fitted to a model in which the cluster has an $S = 3/2$ ground state configuration (dePaula *et al.*, 1986; dePaula *et al.*, 1986). The multiline signal then arises from a low lying $S = 1/2$ excited state which is thermally populated at 8 K. The requirement of a quartet ground state eliminates any model which suggests two non-interacting dimeric units but does not differentiate between tetrameric and trimeric stoichiometries. A signal at $g = 4.1$, which is also attributed to manganese, can be observed in certain S_2 preparations. This low field signal arises from an $S = 3/2$ component of the cluster in a perturbed conformation within the Brudvig framework.

Dual center models

The non-Curie law behavior of the multiline signal is observed for preparations using ethylene glycol as a cryoprotectant; however, a Curie law behavior for this signal is reported when sucrose is added to stabilize the system at low temperatures (Hansson *et al.*, 1987). This observation is fully consistent with a binuclear formulation. It appears

that the signal intensity at the $g = 2$ multiline decreases as the $g = 4.1$ component develops. These data prompted Vanngard to present an alternative structural proposal. In the Vanngard model, a mixed valence binuclear $S = 1/2$ center is the origin of the multiline feature. The $g = 4.1$ resonance, which also follows Curie's law, results from a mononuclear Mn(IV) center which is an $S = 3/2$ spin manifold. These two systems are thought to be in an electron transfer equilibrium such that when the binuclear center is oxidized, presumably an Mn(III/IV) center, the mononuclear center is reduced, forming an epr silent Mn(III), so that only a multiline signal is observed. In contrast, when conditions are altered, for example by the addition of ammonia, the equilibrium shifts so that the mononuclear center is oxidized to Mn(IV) and the cluster reduced to Mn(III/III) giving predominantly the $g = 4.1$ epr feature. Four manganese ions are necessary for oxygen evolution, therefore, this model would require an additional mononuclear manganese center which is not redox active in the lower S states.

The magnetic behavior reported by Brudvig and the electron transfer framework described by Vanngard can be mated to generate a third viable structural framework. In this model, a mixed valence trinuclear center which has an $S = 3/2$ ground state and a low lying $S = 1/2$ excited state could give rise to the non-Curie law dependence observed by Brudvig. The $g = 4.1$ signal could then arise from a mononuclear Mn(IV) center which is in electron transfer equilibrium with this trinuclear cluster. Thus, the $g = 2$ and $g = 4.1$ signals could be generated in a manner analogous to that proposed by Vanngard. Zimmermann and Rutherford (1984) had suggested that the species responsible for the $g = 4.1$ signal might function as an intermediate donor between the cluster and the reaction center; however, more recent studies (Zimmermann and Rutherford, 1986) indicate that the loss of the $g = 4.1$ signal was due to ethanol in the sample preparation. As discussed above, the S_2 state epr spectrum is extremely sensitive to small molecules such as ethanol that inhibit the formation of the $g = 4.1$ signal when samples are illuminated at 200 K. Only when ammonia is added can the $g = 4.1$ signal be restored. On the basis of these data, the authors concluded that the two epr spectral forms of manganese arise from the same center.

X-ray Absorption spectroscopy has been used to determine manganese–manganese and manganese heteroatom distances in the lower S states (Yachandra *et al.*, 1986a,b; Cole *et al.*, 1987; Yachandra *et al.*, 1987; George *et al.*, 1987). The EXAFS data indicate that there are at least two manganese atoms which are separated by 2.7 Å. The best fit to the data also suggests that there is a short manganese–(carbon, nitrogen, oxygen) atom of 1.78 Å. These observations are suggestive of μ -oxo-bridged

Mn(IV) ions. Manganese to heteroatom bond distance typical of Mn(III) and Mn(IV) complexes 1.9–2.0 Å were also reported. The EXAFS fit appears inconsistent with chloride being directly coordinated to the metal cluster. However, it is possible that one chloride bonded at a long distance might escape detection [(e.g. 2.7 Å as reported for a tetranuclear manganese cluster (*vide infra*))]. A long Mn–Mn separation (3.3 Å) has also been observed. Recent EXAFS (George *et al.*, 1987) show that this vector is anisotropic which rules out a symmetric metal cluster. However, it is possible that a highly distorted cubane structure could fit the available experimental data.

Over the past few years many interesting low molecular weight manganese complexes have appeared. A discussion of some of the materials that are relevant to the structural proposals presented above is undertaken in the following section.

SYNTHETIC MODELS

Tetranuclear centers

The characterization of an independent Mn₄ oxo-cubane, illustrated as Fig. 1A, originally proposed by Brudvig has not been reported; however, there is precedent for this structural type in a dodecanuclear mixed valence manganese acetate, Mn₁₂(CH₃CO₂)₁₆(H₂O)₄, reported by Lis, 1980. A central core of four Mn(IV) ions are bridged by four μ₃-oxo atoms in this structure. Two important features of this unit are the Mn–Mn vectors (dist_{avg} = 2.88 Å) and the Mn-(μ₃-O) distance (1.91 Å). These are important distances as EXAFS data (Yachandra *et al.*, 1986a,b; Cole *et al.*, 1987; George *et al.*, 1987) indicate that there are two Mn–Mn vectors (2.71 Å and 3.3 Å) and a very short Mn–O distance (1.79 Å). Clearly, this cubane cannot fit the EXAFS structural constraints. This is especially true of the long Mn–Mn vector. The dodecanuclear cluster contains another tetranuclear building block, shown as Fig. 1C, composed of a Mn(IV) and three Mn(III) ions. There is a short Mn(IV)–Mn(III) distance (2.77 Å) and longer Mn(III)–Mn(IV) (3.45 Å) and Mn(III)–Mn(III) (3.37 Å) distances. Furthermore, a short Mn–O distance of 1.85 is observed.

Christou (Vincent *et al.*, 1987a; Christmas *et al.*, 1987) has recently reported tetranuclear butterfly type clusters reminiscent of the structural unit seen in the dodecanuclear system described above. The tetranuclear Mn(III) complex has an [Mn₄O₂]⁸⁺ core with a short Mn–Mn distance of 2.85 Å and longer distances in the range 3.30–3.39 Å. Furthermore, this material possesses a very short Mn–O distance of 1.80 Å. Thus, it would appear that butterfly type units more closely mimic the EXAFS bond distances than does the cubane structure. Another very interesting tetranuclear complex [Mn(III)₃Mn(IV)₁(μ₃-O)₃(μ₃-Cl)] prepared by the

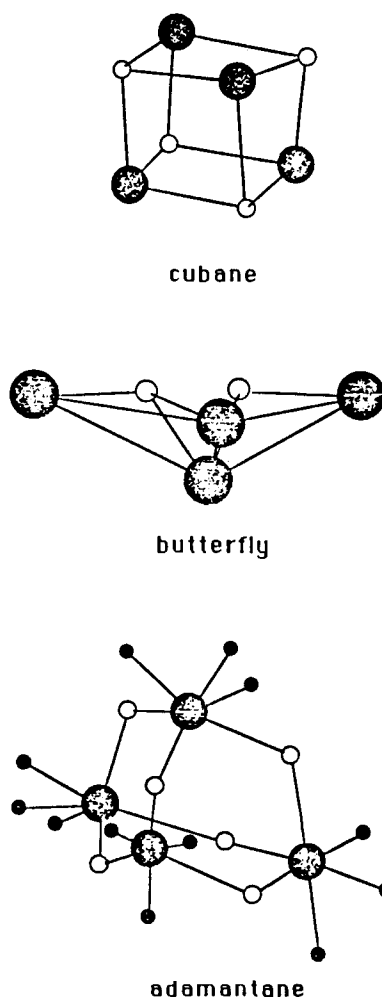


Figure 1. Three tetranuclear structural types which have been proposed for the OEC manganese cluster. (Top) cubane, Mn₄O₄; (Middle) adamantane, Mn₄O₆; (Bottom) butterfly, Mn₄O₂. The large black spheres are manganese atoms and the small white spheres are oxygen atoms. The solid lines between manganese atoms in the middle figure are meant to emphasize the butterfly structure and are not intended to represent Mn–Mn bonds.

same authors (Bashkin *et al.*, 1987) contains both chloride and imidazole coordination to manganese. The chloride ions act as terminal ligands to three Mn(III) ions with 2.2 Å Mn–Cl distance. Most interesting is a μ₃-Cl⁻ which bridges a triangular face of three Mn(III) ions. The Mn–Cl distance is quite long (2.7 Å) which suggests that a bridging chloride could have escaped detection in the original x-ray absorption studies of the OEC.

Wieghardt *et al.* (1983) have synthesized an adamantane type cluster, with an [Mn₄O₆]⁴⁺ core schematically illustrated as Fig. 1 (bottom), which is employed as the active oxidant in one mechanistic proposal (Brudvig and Crabtree, 1986). In this case, the Mn–Mn vectors are 3.21 Å, without a short Mn–Mn distance, and there is a short Mn–O distance of 1.79 Å. It is unlikely that this structure

would appear in the lower enzyme oxidation levels given these distances and the metal oxidation states. However, it is equally unlikely that an adamantane structure in this oxidation level can fulfill the reactivity pattern required of the S_4 state. This complex is stable even in boiling acetic acid, illustrating that often times Mn(IV) compounds are very stable species. The possibility of an adamantane cluster that contains an Mn(V) as the reactive species cannot be completely ruled out. To date there are no clusters containing Mn(V), although the existence of this oxidation state as an intermediate in olefin epoxidation reactions catalyzed by porphyrin or Schiff base ligands has been suggested (Srinivasan *et al.*, 1986).

Trinuclear centers

The best known trinuclear manganese complexes are the common basic acetates (Baikie *et al.*, 1978; Vincent *et al.*, 1987a) illustrated in Fig. 2. These materials contain manganese in a triangular array with two acetate bridging ligands between each metal ion and a μ_3 -oxo ligand occupying the central position of the cluster. The coordination sphere is completed with acetate or other weak bases such as pyridine. The basic acetates can be isolated as mixed valence Mn(III/III/II) or non-mixed valence Mn(III/III/III) compounds.

We have recently (Li *et al.*, 1988; Kessissoglou *et al.*, 1988b) isolated new structural classes of mixed valence trinuclear manganese acetates. The methanol adduct, Mn(III/II/III)(SALADHP)₂(OAc)₄(CH₃OH)₂, which is shown in Fig. 3(a) contains a central Mn(II) ion that is separated by 3.551 Å from the terminal Mn(III) ions which are related by a crystallographic inversion center. The central ion is bridged to each of the terminal ions by one alkoxide and two acetate oxygen moieties. The

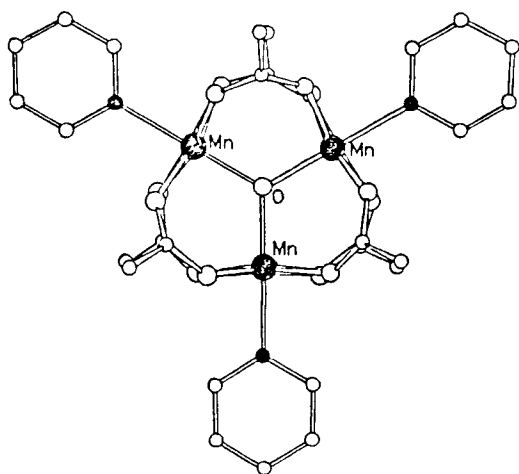


Figure 2. The standard structure type for the basic trinuclear acetates. The large black spheres are manganese atoms, the large white spheres are oxygen atoms, the small white spheres are carbon atoms and the small black spheres are nitrogens.

metals are very close to being valence trapped (Class I) as indicated by the magnetic properties, the lack of an intervalence charge transfer band and the distinctive Mn(II) and Mn(III) bond distances. We have studied in detail the magnetic and spectroscopic properties of the complex with methanol bound to the terminal manganese ions. The room temperature solid state moment is 8.1 B.M. per complex and exhibits non-Curie law behavior as the temperature is decreased. The Mn(III)/Mn(II) antiferromagnetic coupling is $J_{12} = -7 \text{ cm}^{-1}$. This material has an $S = 3/2$ ground state. Because the ground state electronic configuration of this trinuclear complex closely mimics the behavior suggested by Brudvig for the OEC, we investigated the low temperature x-band epr spectrum of this material in a methylene chloride/toluene glass at 4.1 K. A strong signal which exhibits ⁵⁵Mn hyperfine coupling at $g \approx 4$ is observed. A multiline component is observed in the $g = 2$ region at low temperature if DMF is used as the solvent rather than methylene chloride/toluene.

The sensitivity of the epr spectrum of the OEC to environment has been noted above. Small molecules and ions such as ammonia, ethanol and chloride cause shifts in the relative intensities of the 4.1 and multiline features (Beck and Brudvig, 1986; Beck *et al.*, 1986; Ono *et al.*, 1986; Hansson *et al.*, 1987; Zimmermann and Rutherford, 1986). We have recently synthesized (Kessissoglou *et al.*, 1988a) an analog to the methanol complex described above in which the only change in ligation is the substitution of water for the alcohol (Li *et al.*, 1988). This complex, illustrated as Fig. 3(b) has a Mn(III) to water oxygen distance of 2.29 Å. Most important, however, is that the Mn(III) to Mn(II) distance is 3.42 Å, a decrease of nearly 0.1 Å as compared to the methanol adduct. This system represents the first example of a structural change in a manganese cluster which is associated solely with the exchange of coordinated small molecules and illustrates structurally how the conformational change proposed by Brudvig could occur in trinuclear species. Even more recently (Kessissoglou *et al.*, 1988a), we have synthesized a structural isomer of Mn(III/II/III)(SALADHP)₂(OAc)₄(CH₃OH)₂ in which the linear motif is not conserved. This molecule, illustrated as Fig. 3c, contains a Mn2–Mn1–Mn2' angle which is 138.5°, however, the Mn1–Mn2 separation (3.50 Å) is very similar to the linear complex. As expected, the terminal Mn(III) ions are much closer (6.5 Å) in the bent structure than in the linear (7.1 Å). Another interesting difference between the two structural types is the relationship of the two solvent molecules. Both solvent molecules, which are related by a crystallographic O₂ axis, are disposed on the same side of the bent isomer. This contrasts with the linear complexes, which are related by an inversion center, where the solvent moieties lie on opposite sides of the molecular axis. Therefore, one

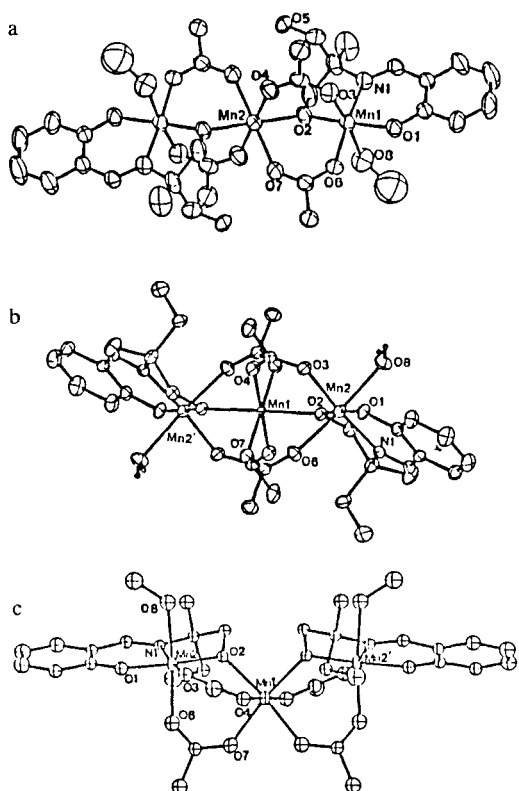


Figure 3. Three new structural types for trinuclear manganese complexes containing Schiff base ligands. (a) An ORTEP diagram of the linear mixed valence acetate Mn(III/II/III)(SALADHP)₂(OAc)₄(CH₃OH)₂. This complex has a Mn–Mn separation of 3.55 Å; (b) The water substituted analogue Mn(III/II/III)(SALADHP)₂(OAc)₄(CH₃OH)₂. The coordinated water is 2.29 Å from the Mn(III) ion and the Mn–Mn separation is 3.42 Å; (c) An ORTEP diagram of the bent form of Mn(III/II/III)(SALADHP)₂(OAc)₄(CH₃OH)₂. The Mn₂–Mn₁–Mn₂ angle is 138.5° and the Mn₁–Mn₂ separation is 3.50 Å.

can envisage that as the bent isomer is more highly oxidized, a compression of the cluster could occur which would bring the two solvent moieties close enough for oxidative chemistry to occur. A similar process could not occur with the linear molecules.

The linear trinuclear acetates described above mimic the magnetic properties of the OEC in the S₂ oxidation level, however, they are not realistic structural models for the center. Both the short Mn–O bond (1.79 Å) and the two Mn–Mn distances (2.7 Å and 3.3 Å) of the OEC are inconsistent with the methanol and water adducts of the trinuclear species.

Binuclear centers

Binuclear materials were the first complexes that were proposed as models for the oxygen evolving center. The epr spectral properties of mixed valence Mn(III/IV) species, especially L₂Mn(IV)(μ₂-O)₂Mn(III)L₂ where L is either bipyridal or phenanthroline, originally were used to explain the S₂ state multiline epr signal (Kirby *et al.*, 1981a,b). In

addition, the very short Mn–Mn and Mn–O distances were the first examples cited in support of bridging oxo groups in the manganese cluster. Binuclear mixed valence μ-oxo bridged species, containing tetradentate ligands, which are more easily oxidized than the bipyridal or phenanthroline complexes, have recently been synthesized (Collins *et al.*, 1987). Manganese analogues of iron dimers containing oxo and acetate bridges, which mimic the binuclear center in the iron protein hemerythrin, have appeared (Sheats *et al.*, 1987). All of the Mn(III/IV) binuclear complexes studied to date give S = 1/2 ground state configurations and usually give rise to g = 2 multiline spectra. Hendrickson (Mabad *et al.*, 1985) has shown that Mn(III/II) dimers can also show multiline features which have similar nuclear hyperfine coupling constants to the Mn(III/IV) dimers. From these data, he has concluded that oxidation state assignment based solely on the magnitude of the nuclear hyperfine coupling is risky since coordination number and environment can perturb these values. More recent reports (Diril *et al.*, 1987) of Mn(II/III) dimers prepared from binucleating ligands further substantiate this observation; however, controversy over this point still remains (Dismukes *et al.*, 1987).

Non-mixed valence dimers which show interesting bridging features have also appeared. Both Mn(II/II) (Kessissoglou *et al.*, 1987a; Mabad *et al.*, 1986) and Mn(III/III) diphenolate bridged dimers (Vincent *et al.*, 1986) have been prepared and structurally characterized. We isolated the Mn(II/II) dimer (Kessissoglou *et al.*, 1987a), [Mn(SALPS)]₂, shown in Fig. 4. The Mn(II) ions are six coordinate, but do not form an ideal octahedron due to steric constraints imposed by the ligand. The material is epr active at 77 K and exhibits very weak antiferromagnetic coupling ($J = -1.6 \text{ cm}^{-1}$). This material has two interesting structural features. First, the Mn–Mn distance is 3.3 Å, a value similar to the long distance in the OEC. Second, a disulfide sulfur

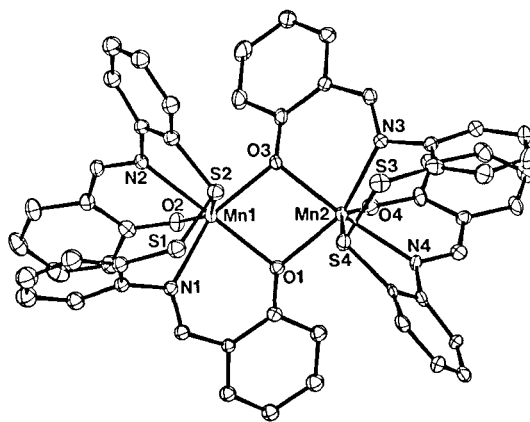


Figure 4. An ORTEP diagram of [Mn(II)SALPS]₂ showing the diphenolate bridge and the long bond between the Mn(II) and disulfide sulfur atom.

atom is coordinated to the metal at a very long 2.7 Å distance. This illustrates that heteroatoms other than manganese (e.g. chloride, bromide or sulfur) can occur at the short Mn–Mn distance seen in the EXAFS spectra. We have also synthesized (Bonadies *et al.*, 1988) [Mn(III)(2-hydroxy-1,3-bis-(salicylideneamino)propane)]₂(CH₃OH) which is an asymmetric dimer that has only one alkoxide oxygen atom bridging the two manganese atoms. The separation between the two metals (3.86 Å) is the longest so far established for two manganese ions that contain a bridging atom.

Mononuclear centers

Mononuclear centers play an important role in both of the electron transfer mechanisms described above. Structurally characterized Mn(IV) complexes are relatively rare (Kessissoglou *et al.*, 1986, 1987b; Pavacik *et al.*, 1986; Hartmann *et al.*, 1984; Camenzind *et al.*, 1982). In addition to Mn(IV) porphyrin and catecholate complexes, two interesting materials with an N₂O₄ coordination environment have been reported. The first of these (Kessissoglou *et al.*, 1986, 1987b), Mn(SALADHP)₂, is shown in Fig. 5. The complex exhibits a slightly distorted octahedral environment using two phenolate and two alkoxide oxygen atoms and two imine nitrogen atoms to encapsulate the metal as a neutral complex. The $E_{1/2}$ for the Mn(IV) to Mn(III) reduction in DMF vs the standard calomel electrode (SCE) is –350 mV. This demonstrates that Mn(IV) can be extremely stable to reduction. Another neutral Mn(IV) complex with an N₂O₄ coordination environment (Pavacik *et al.*, 1986), Mn(salicylate)₂-(bipyridine), has been prepared and the electro-

chemistry investigated. The complex exhibits a reductive wave at +440 mV vs SCE. The primary difference between the two materials is that Mn(SALADHP)₂ has strongly basic alkoxides ligands stabilizing the Mn(IV) ion while Mn(salicylate)₂(bipyridine) substitutes very weakly basic carboxylate moieties. One sees directly the trade off required for the enzyme active site. On one hand, utilization of strongly basic ligands such as oxo groups stabilize Mn(IV) intermediates; however, too many of these very basic moieties lead to a complex which is thermodynamically stable in the high oxidation states (for example the adamantane complex described above). Thus, incorporating weakly basic groups such as carboxylates will lead to more oxidizing sites.

The Mn(SALADHP)₂ complex has $\mu_{\text{eff}} = 3.9$ B.M. as expected for an $S = 3/2$ system. A signal at $g = 5.25$ is observed at 77 K in the x-band epr spectrum of a 5 mM solution in DMF:methanol (3:2). We have prepared other structurally similar materials (Kessissoglou, 1987b) which have low field components ranging between $g = 4$ and 5.3. This illustrates, using a structurally characterized mononuclear Mn(IV) complex, that the low field signal in the S_2 state of the OEC could arise from a mononuclear center as has been suggested by Vanngard.

OXIDATION STATE PROPOSALS

The application of x-ray absorption spectroscopy to photosystem II chemistry has not only provided useful information for defining structure but also oxidation states of manganese in many enzymic oxidation levels (Cole *et al.*, 1987; Yachandra *et al.*, 1986a,b; Kirby *et al.*, 1981a,b; George *et al.*, 1987). The near edge region (XANES) of the spectrum provides the most useful data for interpretation of metal oxidation state. The absolute x-ray absorption edge energies (≈ 6551.3 eV in the S_1 state and ≈ 6552.5 eV in the S_2 state) are in the range observed for model Mn(III) and Mn(IV) complexes described above. The edge shifts to higher energy by about 0.9–1.4 eV during the S_1 to S_2 transition suggesting that a metal centered oxidation has occurred. Little or no change is observed for the S_2 to S_3 transition which suggests that the manganese are not oxidized in this step. However, care must be taken in these interpretations as the edge energies reflect electron density on the manganese, not necessarily oxidation state. Therefore, a change in coordination number or type of ligand could mitigate the change in edge energy associated with an oxidation state change. It is also important to consider the internal consistency between the XANES and EXAFS analysis of the different OEC preparations. The fits of the EXAFS data clearly show a large feature at ≈ 1.75 Å that is normally assigned as an oxo-bridge between the manganese ions (Yachandra *et al.*, 1987). However, such short

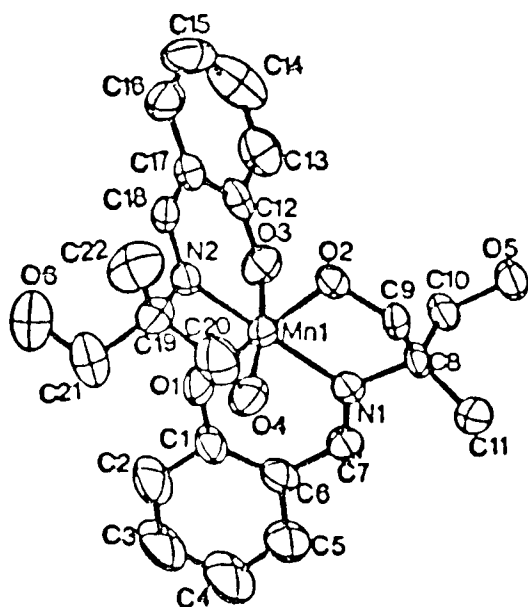


Figure 5. An ORTEP diagram of Mn(IV)(SALADHP)₂.

manganese oxygen distances have only been observed in clusters with manganese in the Mn(IV) oxidation state. In lower oxidation state materials, this bridging oxide is at least 0.1 Å longer (Vincent *et al.*, 1987). Therefore, the short Mn–O distance would suggest a cluster with one or more Mn(IV) ions while the edge energy of the S_1 state is much more consistent with an all Mn(III) formal metal oxidation state. Evidence from other techniques both support and contradict the interpretation of the S_2 to S_3 transition.

Sharp (Srinivasan and Sharp, 1986a,b) has monitored the change in relaxation enhancement (R_1) of water protons by the manganese centers after short saturating flashes of light. Efficient relaxation traps include Mn(II) and Mn(IV) ions, while the orbitally doublet Mn(III) ion is not. A strongly relaxing center disappeared on advancement from S_0 to S_1 while a relaxation enhancement was observed during the S_1 to S_2 transition. An additional flash caused neither enhanced nor detracted from the water relaxation rate. The model presented from these data invokes an S_0 level of Mn(II)₁Mn(III)₃ being oxidized to Mn(III)₄ at the S_1 state which would result in the decreased R_1 . The subsequent S_1 to S_2 transition also corresponds to a metal centered oxidation which generates a Mn(III)₃Mn(IV)₁ center(s) with enhanced water relaxation. As was the case with the x-ray absorption data, the S_2 to S_3 transition is not believed to occur with a metal centred oxidation. The interpretations of the relaxivity data relies on two important assumptions. The first is whether the mechanism for relaxation enhancement is through direct water interaction with the cluster or follows a spin diffusion mechanism through the membrane. If water coordination to the manganese center being oxidized is required in order to see a change in relaxation enhancement, it would be possible to see no change in the relaxivity due to the inaccessibility of the site. The second is whether the relaxivity of a cluster can be modeled by the theory for mononuclear ions or whether the relaxation rates will depend on the mechanism and magnitude of the exchange coupling in the cluster. Both of these points are currently under investigation.

Dismukes (Dismukes and Mathis, 1984) observed a low energy transition in the near IR which was assigned as an intervalence band for the S_2 state. One can not assign the oxidation states of the manganese on these data; however, an interesting feature of this electronic transition is that it arises from a mixed valence center. Advancement of the cluster to the S_3 state does not diminish the intensity of this absorption which suggests that the metal center has not changed valence. Once again, these data are consistent with the x-ray absorption edge spectra.

Although the water relaxation rate and x-ray edge and near IR spectra indicate that the metal cluster

is not oxidized on advancing to S_3 , it is clear that some change has occurred that affects the metals since the multiline epr signal disappears above S_2 . The loss of the multiline signature can be due to an oxidation or a change in spin relaxation rates. The experiments described above support the spin relaxation idea while studies of the UV absorption spectra have been interpreted in favor of sequential, metal centred oxidations. A UV absorption in the 300–350 nm range has been associated with the S state transitions by a number of groups. Dekker *et al.* (1984a,b) studied the flash induced changes in the UV spectrum and observed that the spectral energy and magnitude of the change was equivalent for the S_0 to S_1 , S_1 to S_2 , and S_2 to S_3 transitions. Each transition was then assigned as a Mn(III) to Mn(IV) oxidation by comparing the spectra to those obtained for Mn(III) and Mn(IV) gluconate complexes. This model requires that the S_0 oxidation level contains at least three Mn(III) ions and that the fourth ion is either Mn(III) or Mn(IV). The authors originally suggested that S_0 was Mn(III)₄. However, the Mn(III)₃Mn(IV)₁ or Mn(III)₃Mn(II)₁ oxidation levels are the most probable formulations because the S_2 level contains a non-integral spin system (either $S = 3/2$ or $1/2$). Brudvig and Crabtree have suggested a mechanism for the water splitting reaction using S_0 as Mn(III)₃Mn(IV)₁ while Christou has presented a scheme beginning with S_0 as Mn(III)₃Mn(II)₁ (*vide infra*).

The interpretation of the difference UV absorption studies is not universally accepted. The observed difference spectra were obtained after correcting for spectral changes occurring on the reducing side of PS II. This resulted in very weak signals after the subtraction. In addition, these are difference spectra, not simple absorption spectra, which can complicate the interpretation. One might have expected that the spectral energy of these transitions, if centered on a manganese cluster, would shift in energy as the cluster was sequentially oxidized. Only if the metals acted independently would the same energy and extinction coefficient be expected. These data are even more controversial since recent data suggest that the absorbance changes are not identical for each S state transition (Saygin and Witt, 1985) and can be fit to other models (Lavergne, 1986). Thus, one must question whether the observed signals are associated with manganese or some other PS II component. In addition, the assignment of the oxidations as Mn(III) to Mn(IV) has also been questioned since the Mn(II) to Mn(III) oxidation can exhibit difference spectral changes similar to those observed for the OEC (Vincent and Christou, 1986). Thus, it is unclear to what extent the difference UV spectral changes have provided firm information about the oxidation states of the metal ions. Given the available data, this author prefers the interpretation that the S_0 level corresponds to 1 Mn(II) and 3 Mn(III)

ions. This formulation was assumed in the discussions in the previous section.

MECHANISTIC PROPOSALS

The field of photosynthetic oxygen evolution is not at a loss for mechanistic proposals detailing the role of manganese in the water splitting reaction. Recent reviews that summarize some of these are given in the references. In this section, two recent chemical mechanisms (Brudvig and Crabtree, 1986; Vincent and Christou, 1986) that are based on tetranuclear formulations will be discussed.

Brudvig and Crabtree have presented a model, illustrated as Fig. 6, which incorporates an oxo-cubane structure in the lower S states which converts to an oxo-adamantane structure in the higher oxidation levels of the enzyme (S_3 and S_4). The S_0 state was assigned an oxidation state of $\text{Mn(III)}_3\text{Mn(IV)}_1$. Using the interpretations from the difference UV spectra of Dekker *et al.* (1984a,b), each S state transition was associated with a metal centered oxidation. This results in a non-integral spin system, $\text{Mn(III)}_1\text{Mn(IV)}_3$, for S_2 as required by the interpretation of the epr and magnetic data. On advancing to the S_3 state, two water molecules bind to the cluster with a simultaneous rearrangement of the metal atoms into an Mn_4O_6 adamantane type core. The final oxidation would then generate a catalytically competent $\text{Mn(IV)}_3\text{Mn(V)}_1$ center which facilitates the formation of the oxygen-oxygen bond by excising two bridging oxo groups from the core and regenerating the fully reduced oxo-cubane of S_0 .

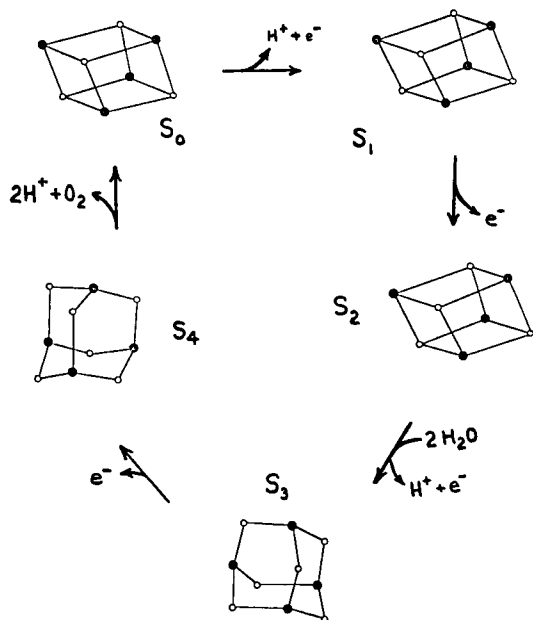


Figure 6. Mechanistic scheme for water oxidation presented by Brudvig and Crabtree and described in detail in text. Dark atoms are manganese ions and white atoms are oxo groups.

This mechanism accounts for many of the experimental observations for the OEC and is structurally consistent both with available model complexes and previously described clusters in biological systems. The cubane formulation is well established in iron-sulfur chemistry as an important structural motif. Manganese cubanes have not been isolated; however, a cubane core is known in the Mn_{12} cluster reported by Lis (1980). The most recent EXAFS analysis (George *et al.*, 1987) of S_1 shows that the long Mn-Mn distance (3.3 Å) is anisotropic. Therefore, a symmetric cubane is ruled out. However, this observation does not exclude either a highly distorted cubane or butterfly arrangement of manganese atoms, if the center is tetranuclear. Wieghardt *et al.* (1983) have characterized an adamantane manganese core which serves as justification for the higher S state structures. This Mn(IV)_4 core is chemically unreactive even under harsh conditions and shows no higher oxidation state electrochemistry. Whether ligand substitution on the non-oxo coordination sites of the manganese ions could allow oxidation to a formal Mn(V) oxidation state is an open question. Highly reactive *mononuclear* manganese(V) porphyrin and Schiff base complexes have been invoked as catalysts in olefin epoxidation chemistry. The existence of this oxidation state in manganese clusters has not, as yet, been detected. This 'missing link' of model chemistry is not necessarily a fatal flaw for this mechanism since one expects that the catalytically competent cluster should be unstable and may not be amenable to isolation. The requirement for a Mn(V) is partially dependent on the interpretation of the UV absorption data. If each S state transition does not proceed with metal oxidation or if the S_0 cluster is more highly reduced, as predicted by the NMR relaxivity data, a lower formal oxidation state for S_4 is expected. Oxygen bond formation occurs at a very late stage in this mechanism. This is consistent with the mass spectrometric data of Radmer and Ollinger (1986) which showed that isotopically labeled water is not incorporated into dioxygen until the final S state transition. Therefore, the water must be freely exchangeable over the time scale of the experiment to a very late stage in the catalytic cycle.

An alternative mechanistic proposal (Vincent and Christou, 1987), illustrated as Fig. 7, based on a series of tetranuclear butterfly type clusters has been advanced. In this, the double-pivot water oxidation scheme, the S_0 oxidation level is composed of three Mn(III) and an Mn(II) ion. These ions are arranged in a butterfly structure which contains two 'hinge' manganese ions at a short distance (2.85 Å) and two wing tip manganese atoms at approximately 3.3 Å from the hinge atoms. A short Mn-O distance is observed from two μ_3 -oxo atoms which bridge each triangular face defined by the two hinge manganese and one wing tip manganese atom. After two metal centered oxidations, the S_2 oxidation level is com-

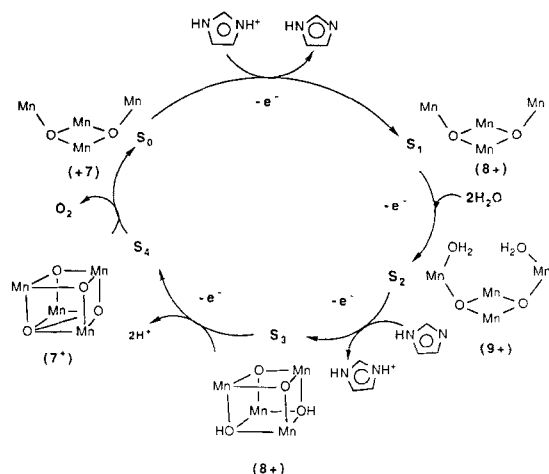


Figure 7. Mechanistic scheme for water oxidation presented by Christou and described in detail in text. Dark atoms are manganese ions and white atoms are oxo groups.

posed at an $\text{Mn(III)}_3\text{Mn(IV)}_1$ cluster. A butterfly with water molecules coordinated to the wing tip manganese was suggested. Additional oxidation of the cluster is thought to generate a cubane like structure which on advancement to S_4 , results in the liberation of dioxygen and the reformation of the S_0 state structure. An imidazole group has been suggested as an acid base catalyst during the S_0 to S_1 transition (releasing a proton) and during the S_2 to S_3 transition (accepting a proton) in order to facilitate cluster oxidation. Acid-base catalysis, whether using histidine or another moiety such as a carboxylate, is well established in other enzymatic reactions. Thus, precedent for an acid-base catalyst is known, although there is little or no experimental evidence addressing the question in the oxygen evolving complex.

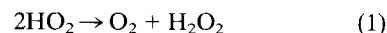
After proposing the above chemical mechanism, a tetranuclear structural type with a bridging chloride was isolated (Bashkin *et al.*, 1987). This complex may† give rise to a multiline type epr spectrum at 60 K. Although this S_2 state model was not part of the original scheme, it represents an interesting possible intermediate since it demonstrates the role of chloride as a cluster stabilizing ligand. This is consistent with the studies of Sandusky and Yocum (1983, 1984, 1986) which have demonstrated a chloride requirement for oxygen evolving activity.

RELATED MANGANESE ENZYMES

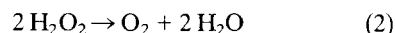
Redox functions for manganese are most often encountered in enzymes which are involved in the

†A multiline signal is observed when this complex is dissolved in DMF:toluene; however, as the authors have pointed out, the nuclearity of the cluster in this solution may not be the same as in the solid state. Therefore, there is a potential ambiguity as to whether this signal originates from a tetranuclear center or a binuclear Mn(III)Mn(IV) fragment of the cluster.

metabolism of the O_2^{n-} unit, where n may range between 0 and 2. A one electron redox process for the manganese superoxide dismutase (Ludwig *et al.*, 1986) has long been established. This enzyme shuttles between the Mn(II) and Mn(III) oxidation states while catalyzing reaction 1:



More recently, binuclear manganese enzymes with catalase activity (Beyer and Fridovich, 1985; Kono and Fridovich, 1983; Barynin *et al.*, 1986; Khangulov *et al.*, 1986), reaction 2, have



been purified from *Lactobacillus plantarum* and *Thermus thermophilus*. The *Thermus* enzyme has been crystallized and a low resolution x-ray structure is available (Khangulov *et al.*, 1986). In addition, the epr spectra of the native enzyme and a hydroxylamine-treated form have been reported (Khangulov *et al.*, 1986). It appears that the manganese ions are within 3.6 Å of one another in this preliminary structure. The native form gives a $g=2$ multiline epr spectrum reminiscent of the S_2 state signature of the OEC and the model complex $(\text{bipy})_2\text{Mn(III)(O)}_2\text{Mn(IV)(bipy)}_2$. In contrast, the epr spectrum of the hydroxylamine-treated sample resembles that of the binuclear $[\text{Mn(II)SALPS}]_2$. Indeed, the origin of the enzymic signal is believed to be from an $S = 1$ or $S = 2$ spin manifold of two weakly coupled Mn(II) ions. Apparently, an inactive enzyme results from the reduction of the core to the Mn(II/II) state by hydroxylamine (Kono and Fridovich, 1983). A model complex has been reported that will cause the decomposition of hydrogen peroxide (Mathur *et al.*, 1987). This compound functions between the Mn(II) and Mn(III) oxidation states. Thus, it would appear that this two electron chemistry is competent using either Mn(II/II) to Mn(III/III) or even higher oxidation state couples.

The OEC is also thought to be reduced by hydroxylamine treatment generating an S_{-1} oxidation level that is epr inactive. In addition, there have been reports that under appropriate conditions hydrogen peroxide can act as an electron donor to PS II (Schroder and Akerlund, H.-E., 1986) and that oxygen can be evolved in S_0 to S_2 cycles using hydrogen peroxide as a substrate (Frasch and Mei, 1987; Mano *et al.*, 1987). Further work on both the catalase and OEC manganese centers is required to elucidate the structural and/or electronic differences between these active sites.

An interesting relationship between all of these manganoenzymes is the conservation of the ratio of manganese atoms to oxidizing or reducing equivalents required for catalysis. Because manganese has multiple available intermediate oxidation states (+2, +3, +4), it is possible that a four electron oxidation of substrate could occur by a reduction of an Mn(IV/IV) dimer to the Mn(II/II) oxidation

level. However, it appears that in the manganese superoxide dismutase and manganese catalase that each metal participates in redox catalysis as a one electron donor or acceptor. By analogy, it is tempting to suggest that each manganese ion in biological systems acts as a single electron transfer agent. The requirement for four manganese ions in the OEC may result as much from a need to accumulate a sufficient number of one electron oxidants as from thermodynamic requirements for generating a highly oxidized center. A tetranuclear cluster would be most appealing within this framework.

SUMMARY

The previous discussion attempts to provide an underpinning for the basic properties of manganese, whether as mononuclear or multinuclear complexes, in biological systems. Complexes that have provided insight into both structural and oxidation state assignments have been emphasized. While the structural data base and physical properties of manganese clusters has increased greatly in the past few years, our understanding of the reactivity of these materials is in its infancy. For this reason, a thorough critique of the reactivity models for the OEC that have appeared recently was not undertaken. We have progressed to the awkward position of being able to state definitively that certain structures or mechanisms are ruled out, but are unable to assign unambiguously a structure or mechanism as correct. The reason for this uncomfortable situation is that the data base is incomplete. Therefore, inorganic chemists will continue to synthesize new compounds which will mimic more closely the properties of the biological centers. However, as the field matures, an increasing emphasis toward defining reactivity patterns of manganese catalyzed reactions will develop. The reader interested in this aspect of manganese chemistry may find references (Ashmawy *et al.*, 1984 and 1985; Ramaraj *et al.*, 1986) informative. The information gleaned from these studies, when combined with biophysical, biochemical and enzymatic data, will allow scientists across the many disciplines that have been studying oxygenic photosynthesis to ferret out the secrets of one of the most important and intriguing reactions in nature.

Acknowledgements‡—The author expresses his thanks to the following colleagues. First, are my research coworkers at The University of Michigan who carried out the original work from my laboratory discussed in this paper: Dr.

Dimitris P. Kessissoglou, Dr. Joseph Bonadies, Dr. William Butler, Ms. Xinhua Li and Mr. Myoung Soo Lah. Next, are our collaborators at Michigan and other universities: Professor Gerald T. Babcock and Dr. Chris Bender (Michigan State University), Professor William Hatfield and Mr. Martin Kirk (University of N. Carolina, Chapel Hill), Professor James Penner-Hahn and Dr. Richard Fronko (U. of Michigan) and Prof. Mike Maroney (U. of Mass., Amherst). I also have enjoyed stimulating conversations on the manganese centers in PS II with Professors Gary Brudvig, George Christou, G. Charles Dismukes, Dimitris Coucouvanis, Wayne Frasc, Bob Sharp, John Sheats, Karl Wieghardt and Charlie Yocum, and Drs. Julio de Paula and Jan Dekker. Finally, I wish to acknowledge financial support for our work over the past several years from the donors of the Petroleum Research fund which is administered by the American Chemical Society, The Horace H. Rackham Foundation and The Institute for Protein Structure and Design of the University of Michigan, and the US Department of Agriculture.

REFERENCES

- Allen, G. and N. S. Hush (1967) Intervalence-transfer absorption. Part 2. Theoretical considerations and spectroscopic data. *Prog. Inorg. Chem.* **8**, 391-443.
- Ashmawy, F. M., C. A. McAulliffe, R. V. Parish and J. Tames (1984) The photolysis of coordinated water in $Mn(\text{salm})(\text{H}_2\text{O})_2(\text{ClO}_4)_2$ [salm = N,N'-propylenebis(salicylideneaminato)] *J. Chem. Soc., Chem. Commun.* 14-16.
- Ashmawy, F. M., C. A. McAulliffe, R. V. Parish and J. Tames (1985) Water photolysis. Part 1. The photolysis of coordinated water in $[MnL(\text{H}_2\text{O})_2](\text{ClO}_4)_2$ (L = dianion of tetradentate O_2N_2 -donor Schiff bases). A Model for the manganese site in photosystem II of green plant photosynthesis. *J. Chem. Soc., Dalton Trans.* 1391-1397.
- Asmez, J. (1983) The role of manganese in photosynthetic oxygen evolution. *Biochim. Biophys. Acta* **726**, 1-12.
- Baikie, A. R. E., M. B. Hursthouse, D. B. New and P. Thornton (1978) Preparation, crystal structure, and magnetic properties of a trinuclear mixed-valence manganese carboxylate. *J. Chem. Soc., Chem. Comm.* 62-63.
- Barynin, V. V., A. A. Vagin, V. R. Melik-Adamyin, A. I. Grebenko, S. V. Khangulov, A. N. Popov, M. E. Anrianova and B. K. Vainshtein (1986) Three dimensional structure of T-catalase with 3 Å resolution. *Soviet Phys. Dokl.* **31**, 457-459.
- Bashkin, J. S., H.-R. Chang, W. E. Streib, J. C. Huffman, D. N. Hendrickson and G. Christou (1987) Modelling the photosynthetic water oxidation center: Preparation and physical properties of a tetranuclear oxide bridged Mn complex corresponding to the native S_2 state. *J. Am. Chem. Soc.* **109**, 6502-6504.
- Basolo, F. and R. G. Pearson (1967) *Mechanisms of Inorganic Reactions: A Study of Metal Complexes in Solution* (Second Edition). Wiley, New York.
- Beck, W. F. and G. W. Brudvig (1986) Binding of amines to the O_2 -evolving center of photosystem II. *Biochemistry* **25**, 6479-6486.
- Beck, W. F., J. C. de Paula and G. W. Brudvig (1986) Ammonia binds to the manganese site of the O_2 -evolving complex of photosystem II in the S_2 site. *J. Am. Chem. Soc.* **108**, 4018-4022.
- Bellito, C., A. A. G. Tomlinson and C. Furlani (1971) Crystal polarised spectra of pentachloromanganese(III) ion. *J. Chem. Soc. A*, 3267-3271.
- Beyer, Jr., W. F. and I. Fridovich (1985) Pseudocatalase from *Lactobacillus plantarum*: Evidence for a homopentameric structure containing two atoms of manganese per subunit. *Biochemistry* **24**, 6460-6467.

‡This manuscript was based on a presentation in a symposium on photosynthetic oxygen evolution held at the national meeting of the American Society for Photobiology in Bal Harbour, Florida. For this reason, special emphasis has been placed on new chemistry from the author's laboratory. Details of structures from other research groups can be obtained from the references provided.

- Bonadies, J. A., D. P. Kessissoglou, M. Maroney and V. L. Pecoraro (1988) Solution speciation of monomeric and dimeric manganese(III) Schiff base complexes using electrochemistry and proton NMR. *J. Am. Chem. Soc.* Submitted.
- Boucher, L. J. and V. W. Day (1977) Manganese Schiff base complexes. 5. Synthesis and spectroscopy of some anion complexes of N,N'-ethylene-bis-acetylacetonimine Mn(III). *Inorg. Chem.* **16**, 1360-1367.
- Brudvig, G. W. and R. H. Crabtree (1986) Mechanism for photosynthetic O₂ evolution. *Proc. Natl. Acad. Sci. USA* **83**, 4586-4588.
- Camenzind, M. J., F. J. Hollander and C. L. Hill (1982) Synthesis, ground electronic state, and crystal and molecular structure of the monomeric manganese(IV) porphyrin complex dimethoxy (5,10,15,20-tetraphenylporphyrinato)manganese(IV). *Inorg. Chem.* **21**, 4301-4308.
- Casey, J. L. and R. Sauer (1984) EPR detection of a cryogenically photogenerated intermediate in photosynthetic oxygen evolution. *Biochim. Biophys. Acta* **767**, 21-28.
- Christmas, C., J. B. Vincent, J. C. Huffman, G. Christou, H.-R. Chang and D. N. Hendrickson (1987) Support from model studies for the proposed existence of an S₋₁ oxidation level in the manganese assembly of the photosynthetic water oxidation centre. *J. Chem. Soc. Chem. Comm.* 1303-1304.
- Cole, J. L., V. K. Yachandra, A. E. McDermott, R. D. Guiles, R. D. Britt, S. L. Dehriheimer, K. Sauer and M. P. Klein (1987) Structure of the manganese complex of Photosystem II upon removal of the 33-Kilodalton extrinsic protein: An x-ray absorption spectroscopy study. *Biochemistry* **26**, 5967-5973.
- Coleman, W. M., R. K. Boggess and L. T. Taylor (1981) Electrochemical studies of manganese (II) complexes containing pentadentate ligands with O₂N₁, O₃N₂, and O₂SN₂ donor sets. *Inorg. Chem.* **20**, 700-706.
- Coleman, W. M., R. R. Goehring, L. T. Taylor, J. G. Mason and R. K. Boggess (1979) Electrochemical studies on a series of manganese (III) complexes containing symmetrical pentadentate ligands. *J. Am. Chem. Soc.* **101**, 2311-2315.
- Coleman, W. M. and L. T. Taylor (1977) Oxygen studies of manganese (II) complexes containing linear pentadentate ligands. *Inorg. Chem.* **16**, 1114-1119.
- Collins, M. A., D. J. Hodgson, K. Michelsen and D. K. Towle (1987) Synthesis and characterization of the binuclear complex Di-μ-oxo-bis[N,N'-bis(2-methylpyridyl)ethane-1,2-diamine]dimanganese(III,IV) Perchlorate trihydrate, [(bispicen)MnO]₂(ClO₄)₃·3H₂O. *J. Chem. Soc., Chem. Commun.* 1659-1660.
- Cotton, A. and G. Wilkinson (1980) *Advanced Inorganic Chemistry: A Comprehensive Text* (4th Edn). Wiley, New York.
- Davies, J. E., B. M. Gatehouse and K. S. Murray (1973) Crystal and molecular structure and magnetic properties of catena-μ-acetato-[NN'-ethylenebis(salicylaldiminato)]manganese(III). A linear-chain complex containing a single anti-anti acetate bridge. *J. Chem. Soc. Dalton*, 2523-2527.
- Day, P. (1980) An introduction to mixed-valence chemistry. In *Mixed-Valence Compounds* (Edited by D. B. Brown), NATO Advanced Study Institutes Series Vol. 58, pp. 3-24. D. Reidel, Boston.
- Dekker, J. P., H. J. V. Van Gorkom, M. Brok and L. Ouwehand (1984a) Optical characterization of photosystem II electron donors. *Biochim. Biophys. Acta* **764**, 301-309.
- Dekker, J. P., H. J. V. Van Gorkom, J. Wensink and L. Ouwehand (1984b) Absorption difference spectra of the successive redox states of the oxygen evolving apparatus of photosynthesis. *Biochim. Biophys. Acta* **767**, 1-9.
- Diril, H., H.-R. Chang, X. Zhang, S. K. Larsen, J. A. Potenza, C. G. Pierpont, H. J. Shugar, S. Isied and D. N. Hendrickson (1987) Binuclear mixed-valence MnII/MnIII complexes: Insight about the resolution of hyperfine structure in the EPR spectrum. *J. Am. Chem. Soc.* **109**, 6206-6208.
- Dismukes, G. C. (1986) The metal centers of the photosynthetic oxygen evolving complex. *Photochem. Photobiol.* **43**, 99-115.
- Dismukes, G. C. (1986) The metal centers of the photosynthetic oxygen evolving complex. *Photochem. Photobiol.* **43**, 99-115.
- Dismukes, G. C. and P. Mathis (1984) A near infrared electronic transition associated with the conversion between S-states of the photosynthetic O₂-evolving complex. *FEBS Lett.* **178**, 51-54.
- Dismukes, G. C., J. E. Sheats and J. A. Smegal (1987) Mn²⁺/Mn³⁺ and Mn³⁺/Mn⁴⁺ mixed valence binuclear manganese complexes of biological interest. *J. Am. Chem. Soc.* **109**, 7202-7203.
- Dismukes, G. C. and Y. Siderer (1981) Intermediates of a polynuclear manganese center involved in photosynthetic oxidation of water. *Proc. Natl. Acad. Sci. USA* **78**, 274-278.
- Frasch, W. D. and R. Mei (1987) Hydrogen peroxide as an alternate substrate for the oxygen-evolving complex. *Biochim. Biophys. Acta* **891**, 8-14.
- George, G., S. Cramer and R. Prince (1987) Presented at the 3rd International Conference on Bioinorganic Chemistry, Noordwijkerhout, Netherlands, June.
- Girerd, J. J., G. Blondin, J. Bonvoisin, M. F. Charlot, M. Corbella, S. Menage, S. E. Vitols, K. Wieghardt, M. Perree-Fauvet, J. Huet, A. Gaudemer and P. Boucly (1987) Magnetic properties of manganese clusters related to the active site of the oxygen evolving complex in photosynthesis. *Rec. Trav. Chim. Pays-Bas* **106**, 219.
- Gross, R. and W. Kaim (1987a) Deprotonated p-phenylenediamines as noninnocent ligands. Metal-to-ligand spin transfer in the ground state and ligand-to-metal charge transfer in the lowest excited state of low-spin manganese(II) complexes. *Inorg. Chem.* **26**, 3596-3600.
- Gross, R. and W. Kaim (1987b) Electron spin resonance and electronic spectroscopy of low-spin manganese(II) complexes (C₅R₄)(CO)₂Mn with I=hydrazido(1-), arylamido, anionic nitrile and purine type ligands. *J. Chem. Soc., Farad. Trans.* **83**, 3549-3564.
- Hansson, O., R. Aasa and T. Vanngard (1987) The origin of the multiline and g = 4.1 epr signals from the oxygen evolving system of photosystem II. *Biophys. J.* **51**, 825-832.
- Hartmann, J. R., B. M. Foxman and S. R. Cooper (1984) Higher valent manganese chemistry. Synthetic, structural, and solution studies on [Mn(catecholate)₃]ⁿ⁻ (n = 2,3) complexes. *Inorg. Chem.* **23**, 1381-1387.
- Huhey, J. E. (1983) *Inorganic Chemistry* (3rd Edn). Harper & Row, New York.
- Itoh, S., C. T. Yerkes, H. Koike, H. H. Robinson and A. R. Crofts (1984) Effects of chloride depletion on the electron donation from the water-oxidizing complex to the photosystem II reaction center as measured by the microsecond rise of chlorophyll fluorescence in isolated pea chloroplasts. *Biochim. Biophys. Acta* **766**, 612-622.
- Kessissoglou, D. P., C. Bender, M. Kirk and V. L. Pecoraro (1988a) Isolation of an aquated trinuclear mixed valence manganese acetate complex. *J. Chem. Soc., Chem. Commun.* Submitted.
- Kessissoglou, D. P., W. M. Butler and V. L. Pecoraro (1986a) Structural and spectroscopic characterization of the manganese(IV) Schiff base complex Mn(saladhp)₂(saladhp = 2-salicylideneiminato-1,3-dihydroxy-2-Methylpropane. *J. Chem. Soc. Chem. Commun.* 1253-1255.

- Kessissoglou, D. P., W. M. Butler and V. L. Pecoraro (1987a) Characterization of mono- and binuclear manganese (II) Schiff base complexes with metal-disulfide ligation. *Inorg. Chem.* **26**, 495–503.
- Kessissoglou, D. P., M. S. Lah and V. L. Pecoraro (1988b) A bent mixed-valence manganese (III/II/III) complex: A new class of trinuclear acetate bridged Schiff base compounds. *Angew. Chem.* Submitted.
- Kessissoglou, D. P., X. Li, W. M. Butler and V. L. Pecoraro (1987b) Mononuclear manganese(IV) complexes of hydroxyl-rich Schiff base Ligands. *Inorg. Chem.* **26**, 2487–2492.
- Khangulov, S. V., V. V. Barynin, V. R. Melik-Adamyin, A. I. Grebenko, N. V. Voevodskaya, L. A. Blumenfeld, S. N. Dobryakov and V. B. Il'Yasova (1986) EPR study of T-catalase from *Thermus thermophilus*. *Bioorg. Khim.* **12**, 741–748.
- Kirby, J. A., D. B. Goodin, T. Wydrzynski, A. S. Robertson and M. P. Klein (1981a) State of manganese in the photosynthetic apparatus. 1. Extended x-ray absorption fine structure studies of chloroplasts and di- μ -oxo bridged dimanganese model compounds. *J. Am. Chem. Soc.* **103**, 5529–5537.
- Kirby, J. A., A. S. Robertson, J. P. Smith, A. C. Thompson, S. R. Cooper and M. P. Klein (1981b) State of manganese in the photosynthetic apparatus. 2. X-ray absorption edge studies of manganese in photosynthetic membranes. *J. Am. Chem. Soc.* **103**, 5537–5542.
- Kono, Y. and I. Fridovich (1983) Inhibition and reactivation of Mn-catalase: implications for valence changes at the active site manganese. *J. Biol. Chem.* **258**, 13646–13648.
- Lavergne, J. (1986) Stoichiometry of the redox changes of manganese during the photosynthetic water oxidation cycle. *Photochem. Photobiol.* **43**, 311–317.
- Lewis, W. B. and L. O. Morgan (1968) *Paramagnetic Relaxation in Solutions in Transition Metal Chemistry* (Edited by R. L. Carlin), Vol. 4. Marcel-Dekker, New York.
- Li, X., D. P. Kessissoglou, M. L. Kirk, C. Bender and V. L. Pecoraro (1988) Isolation of a mixed valence trinuclear manganese complex potentially relevant to the photosynthetic oxygen evolving complex. *Inorg. Chem.* **27**, 1.
- Lis, T. (1980) Preparation, structure, and magnetic properties of a dodecanuclear mixed-valence manganese carboxylate. *Acta Cryst.* **B36**, 2042–2046.
- Lis, T. and B. Jezowska-Trzebiatowski (1977) The reaction between potassium permanganate and formic acid. Structural characterization of the polymeric compound $(K_2\{Mn(H_2O)_2[Mn_3O(HCOO)_9\}_2])_n$. *Acta Cryst.* **B33**, 2112–2116.
- Ludwig, M. L., K. A. Patridge and W. C. Stallings (1986) Manganese superoxide dismutase: Structure and properties. Manganese In *Metabolism and Enzyme Function*, pp. 405–430. Academic Press, New York.
- Mabad, B., P. Cassoux, J.-P. Tuchagues and D. N. Hendrickson (1986) Manganese(II) complexes of polydentate Schiff bases. 1. Synthesis, characterization, magnetic properties, and molecular structure. *Inorg. Chem.* **25**, 1420–1431.
- Mabad, B., J.-P. Tuchagues, Y. T. Hwang and D. N. Hendrickson (1985) Mixed-valence $Mn^{II}Mn^{III}$ complexes: Models for the manganese site in the photosynthetic electron-transfer chain. *J. Am. Chem. Soc.* **107**, 2801–2803.
- Mano, J., M.-A. Takahashi and K. Asada (1987) Oxygen evolution from hydrogen peroxide in photosystem II: Flash-induced catalytic activity of water-oxidizing photosystem II membranes. *Biochemistry* **26**, 2495–2501.
- Mathur, P., M. Crowder and G. C. Dismukes (1987) Dimanganese complexes of a septadentate ligand. Functional analogues of the manganese pseudocatalase. *J. Am. Chem. Soc.* **109**, 5227–5233.
- Muerteries, E. L. and L. J. Guggenberger (1984) Idealized polytopal forms. Description of real molecules referenced to idealized polygons or polyhedra in geometric reaction path form. *J. Am. Chem. Soc.* **96**, 1748–1756.
- Ono, T., J. L. Zimmerman, Y. Inoue and A. W. Rutherford (1986) EPR evidence for a modified S-state transition in chloride depleted Photosystem II. *Biochim. Biophys. Acta* **851**, 193–201.
- de Paula, J. C., W. F. Beck and G. W. Brudvig (1986) Magnetic properties of manganese in the photosynthetic O_2 -evolving complex. 2. Evidence for a manganese tetramer. *J. Am. Chem. Soc.* **108**, 4002–4009.
- de Paula, J. C., W. F. Beck, A.-F. Miller, R. B. Wilson and G. W. Brudvig (1987) Studies of the manganese site of photosystem II by electron spin resonance spectroscopy. *J. Chem. Soc. Faraday Trans I*, **83**, 3635–3651.
- de Paula, J. C., J. B. Innes and G. W. Brudvig (1985) Electron transfer in photosystem II at cryogenic temperatures. *Biochemistry* **24**, 8114–8120.
- Pavacik, P. S., J. C. Huffman and G. Christou (1986) A manganese(IV) complex with phenoxide- and carboxylate-like ligation: Preparation and structure of $[Mn(sal)_2(bipy)]$ (H_2sal = salicylic acid; $bipy$ = 2,2'-bipyridine). *J. Chem. Soc. Chem. Commun.* 43–44.
- Pecoraro, V. L. and W. M. Butler (1986) Structure of N,N' -ethylene-bis(salicylideneaminato) manganese (III) chloride acetonitrile solvate. *Acta Cryst.* **C42**, 1151–1153.
- Piepho, S. B., E. R. Krausz and P. N. Schatz (1978) Vibronic coupling model for calculation of mixed valence absorption profiles. *J. Am. Chem. Soc.* **100**, 2996–3005.
- Plaskin, P. M., R. C. Stoufer, M. Mathew and G. J. Palenik (1972) A novel antiferromagnetic oxo-bridged manganese complex. *J. Am. Chem. Soc.* **4**, 2121–2122.
- Ramaraj, R., A. Kira and M. Kaneko (1986) Oxygen evolution by water oxidation mediated by heterogeneous manganese complexes. *Angew. Chem. Int. Ed.* **25**, 825–827.
- Robin, M. B. and P. Day (1967) Mixed valence chemistry—A survey and classification. *Adv. Inorg. Chem. Radiochem.* **10**, 248–403.
- Sandusky, P. O. and C. F. Yocum (1983) The mechanism of amine inhibition of the photosynthetic oxygen evolving complex. *FEBS Lett.* **162**, 339.
- Sandusky, P. O. and C. F. Yocum (1984) The chloride requirement for photosynthetic oxygen evolution. Analysis of the effects of chloride and other anions on amine inhibition of the OEC. *Biochim. Biophys. Acta* **766**, 603–611.
- Sandusky, P. O. and C. F. Yocum (1986) The chloride requirement for photosynthetic oxygen evolution: factors affecting nucleophilic displacement of chloride from the oxygen-evolving complex. *Biochim. Biophys. Acta* **849**, 85–93.
- Sawyer, D. T., M. E. Bodini, L. A. Willis, T. L. Reichel and K. D. Magers (1977) Electrochemical and spectroscopic studies of manganese (II,III,IV) complexes as models for photosynthetic O_2 -evolution reactions (Edited by K. N. Raymond) Vol. 2, pp. 330–349. Am. Chem. Soc. Washington, DC.
- Saygin, O. and H. T. Witt (1985) Sequence of the redox changes of manganese and pattern of the changes of charges during water cleavage in photosynthesis: Optical events in the UV and red region in the presence and absence of hydroxylamine. *Photobiochem. Photobiophys.* **10**, 71–82.
- Schroder, W. P. and H.-E. Akerlund (1986) H_2O_2 Accessibility to the Photosystem II donor side in protein-depleted inside-out thylakoids measured as flash-induced oxygen production. *Biochim. Biophys. Acta*

- 848, 359–363.
- Sheats, J. E., R. S. Czernuszewicz, G. C. Dismukes, A. L. Rheingold, V. Petrouleas, J. Stubbe, W. H. Armstrong, R. H. Beer and S. J. Lippard (1987) Binuclear manganese(III) complexes of potential biological significance. *J. Am. Chem. Soc.* **109**, 1435–1444.
- Sorrell, T. N. (1985) Binuclear copper complexes: Synthetic models for the active site of type III copper proteins, biological and inorganic copper chemistry. Adenine Press, Guilderland, New York.
- Srinivasan, K., P. Michaud and J. K. Kochi (1986) Epoxidation of olefins with cationic (salen)Mn(III) complexes. The modulation of catalytic activity by substituents. *J. Am. Chem. Soc.* **108**, 2309–2320.
- Srinivasan, A. N. and R. R. Sharp (1986a) Flash-induced enhancements in the proton NMR relaxation rate of photosystem II particles. *Biochim. Biophys. Acta* **850**, 211–217.
- Srinivasan, A. N. and R. R. Sharp (1986b) Flash-induced enhancements in the proton NMR relaxation rate of photosystem II particles. *Biochim. Biophys. Acta* **850**, 211–217.
- Stults, B. R., B. S. Marianelli and V. W. Day (1979) Distortions of the coordination polyhedron in high-spin manganese (III) complexes. 3. Crystal and molecular structure of Tris(acetylacetonato)manganese(III): A tetragonally elongated octahedral form. *Inorg. Chem.* **18**, 1853–1858.
- Theg, S. M., P. Jurinsinic and P. H. Homann (1984) Studies on the mechanism of chloride action on photosynthetic water oxidation. *Biochim. Biophys. Acta* **766**, 636–646.
- VanAtta, R. B., C. E. Strouse, L. K. Hanson and J. S. Valentine (1987) [Peroxytetraphenylporphinato]manganese(II) anions. Synthesis, crystal structures, and electronic structures. *J. Am. Chem. Soc.* **109**, 1425–1434.
- Vincent, J. B., H.-R. Chang, K. Folting, J. C. Huffman, G. Christou and D. N. Hendrickson (1987a) Preparation and physical properties of trinuclear oxo-centered manganese complexes of the general formulation $[\text{Mn}_3\text{O}(\text{O}_2\text{CR})_6\text{L}_3]^{0,+}$ (R=Me or Ph; L=a neutral donor group) and the crystal structures of $[\text{Mn}_3\text{O}(\text{O}_2\text{CMe})_6(\text{pyr})_3](\text{pyr})$ and $[\text{Mn}_3\text{O}(\text{O}_2\text{CPh})_6(\text{pyr})_2(\text{H}_2\text{O})] \cdot 0.5 \text{ MeCN}$. *J. Am. Chem. Soc.* **109**, 5703–5711.
- Vincent, J. B., C. Christmas, J. C. Huffman, G. Christou, H.-R. Chang and D. N. Hendrickson (1987b) Modeling the photosynthetic water oxidation centre: Synthesis, structure, and magnetic properties of $[\text{Mn}_4\text{O}_2(\text{OAc})_7(\text{bipy})_2](\text{ClO}_4) \cdot 3\text{H}_2\text{O}$ (bipy = 2,2'-bipyridine). *J. Chem. Soc. Chem. Commun.* 236–238.
- Vincent, J. B. and G. Christou (1986) Model complexes suggest certain S-state changes of photosynthetic water-oxidation enzyme may involve an Mn(II)–Mn(III) transition. *FEBS Lett.* **207**, 250–252.
- Vincent, J. B. and G. Christou (1987) A molecular 'double pivot' mechanism for water oxidation. *Inorg. Chim. Acta* **136**, L41–L43.
- Vincent, J. B., K. Folting, J. C. Huffman and G. Christou (1986) Use of tetra-*n*-butylammonium permanganate for inorganic synthesis in non-aqueous solvents. Preparation and structure of a manganese(III) dimer containing bridging phenoxo oxygen atoms. *Inorg. Chem.* **25**, 996–999.
- Volker, M., G. Renger and A. W. Rutherford (1986) Effects of trypsin upon EPR signals arising from components of the donor side of Photosystem II. *Biochim. Biophys. Acta* **851**, 424–430.
- Wieghardt, K., V. Bossek and W. Gebert (1983) Synthesis of a tetranuclear manganese(IV) cluster with an adamantane skeleton: $[(\text{C}_6\text{H}_{15}\text{N}_3)_4\text{Mn}_4\text{O}_6]$. *Angew. Chemie, Int. Ed.* **22**, 328–329.
- Wieghardt, K., V. Bossek, D. Ventura and J. Weiss (1985) Assembly and structural characterization of binuclear μ -Oxo-di- μ -acetato bridged complexes of manganese(III). Analogues of the Di-iron(III) centre in hemerythrin. *J. Chem. Soc., Chem. Commun.* 347–349.
- Yachandra, V. K., R. D. Guiles, A. McDermott, R. D. Britt, S. L. Dexheimer, K. Sauer and M. P. Klein (1986a) The state of manganese in the photosynthetic apparatus. 4. Structure of the manganese complex in photosystem II studied using EXAFS spectroscopy. The S₁ state of the O₂-evolving photosystem II complex from spinach. *Biochim. Biophys. Acta* **850**, 324–332.
- Yachandra, V. K., R. D. Guiles, A. McDermott, J. L. Cole, R. D. Britt, S. L. Dexheimer, K. Sauer and M. P. Klein (1987) Comparison of the structure of the manganese complex in the S₁ and S₂ states of the photosynthetic oxygen evolving complex: an x-ray absorption spectroscopy study. *Biochemistry* **26**, 5974–5981.
- Yachandra, V. K., R. D. Guiles, K. Sauer and M. P. Klein (1986b) The state of manganese in the photosynthetic apparatus. 5. The chloride effect in photosynthetic oxygen evolution. Is halide coordinated to the EPR-active manganese in the O₂-evolving complex? Studies of the substructure of the low-temperature multiline EPR signal. *Biochim. Biophys. Acta* **850**, 333–342.
- Yamaguchi, K. and D. T. Sawyer (1985) Redox chemistry for the mononuclear tris(picolinato), tris(acetylacetonato), and tris(8-quinolinato) manganese(III) complexes: Reaction mimics for the water oxidation cofactor in photosystem II. *Inorg. Chem.* **24**, 971–976.
- Zimmerman, J.-L. and A. W. Rutherford (1984) EPR studies of the oxygen-evolving enzyme of photosystem II. *Biochim. Biophys. Acta* **767**, 160–167.
- Zimmerman, J.-L. and A. W. Rutherford (1986) Electron paramagnetic resonance properties of the S₂ state of the oxygen evolving complex of photosystem II. *Biochemistry* **25**, 4609–4613.