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Spin State/Stereochemistry Relationships in Metalloporphyrins: the Vibronic Approach

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Consider the pseudo Jahn–Teller instability [1] of the planar nuclear configuration of the MeP system with respect to the out-of-porphyrin plane displacement and to the change of hole diameter depending on the electronic state and the nature of the metal. The planar MeP has the D_{4h} symmetry, the out-of-plane metal displacement is of A_{2u} symmetry, and the change in hole diameter corresponds to the totally symmetric displacement. It may be concluded from selection rules and 3d MeP electronic structure calculations [2], that in $Fe^{II}P$ and $Mn^{II}P$ there are intermediate-spin ground, ${}^{i}\psi_0$, and excited, ${}^{i}\psi_1$, electronic states, which are mixed by A_{2u} displacements. The ${}^{i}\psi_1$ state can be constructed from the ${}^{i}\psi_0$ one by a one-electron excitation $a_{2u}(\pi) \to a_{1g}(d_{z^2})$.

The lowest high-spin states ${}^{h}\psi_0$ can be obtained from the ${}^{i}\psi_0$ one by electron $e_g(d\pi) \to b_{1g}(d_{x^2-y^2})$ excitations. These states are mixed by A_{2u} displacements with two near lying excited states. The first, ${}^{h}\psi_1$, can be obtained from the ${}^{h}\psi_0$ state by electron $a_{2u}(\pi) \to a_{1g}(d_{z^2})$ excitation, the mixing ${}^{h}\psi_0 - {}^{h}\psi_1$ being characterized by the same vibronic parameters as the ${}^{i}\psi_0 - {}^{i}\psi_1$ mixing. The second state ${}^{h}\psi_2$ is formed by the $b_{1g}(d_{x^2-y^2}) \to b_{2u}(\pi^*)$ excitation.

We have shown recently [3, 4] that in both cases the $\psi_0 - \psi_1$ mixing gives the main contribution to the instability of the planar MeP configuration, whereas the contribution of the $\psi_0 - \psi_2$ mixing per se is insufficient for the formation of such an instability (herefrom it may be concluded that in MeP with completely occupied $a_{1g}(d_{z^2})$ MO, *e.g.* in $Cu^{II}P$, $Zn^{II}P$, and diamagnetic $Ni^{II}P$, the in-plane position of the metal is stable due to the lack of low lying ψ_1 states).

On the other hand the occupation of the antibonding $b_{1g}(d_{x^2-y^2})$ MO in the ${}^{h}\psi_0$ state results, first, in the increase of the hole diameter due to the stronger interaction with the totally symmetric displacements and, second, in an additional reduction of A_{2u} force constant. Therefore, under the out-of-plane metal displacement the energy lowering in the ${}^{h}\psi_0$ state occurs more readily than in the ${}^{i}\psi_0$ one. As a result the intersection of the two terms takes place, provided the $\psi_0 - \psi_1$ vibronic mixing is strong enough (for the corresponding quantitative criterion see Ref. [4]). Thus for large enough out-of-plane displacements of the metal atom the ${}^{h}\psi_0$ state becomes the ground one, in contrast with the planar configuration case, where ${}^{i}\psi_0$ is the ground state and *vice versa*, if the MeP system is found in the high-spin state, it means that the size of the hole is greater and the out-of-plane metal displacement is larger, compared with ${}^{i}\psi_0$. Note, that as far as the out-of-plane displacement of the metal atom is determined by the $a_{2u}(\pi) - a_{1g}(d_{z^2})$ MO mixing, the essential influence of the environment of the MeP (both peripheral and axial) on the magnitude of this displacement and spin equilibrium has to be expected.

The results obtained are in qualitative agreement with the experimental data available [5].

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Catalytic Mechanisms and Reactivity of Peroxidases, Catalases and Iron–Porphyrin Model Systems

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Water-soluble iron(III)–porphyrins display both peroxidase and catalase-like properties [1, 2]. In both enzymic and model systems the basic catalytic mechanism is:

Catalyst + Oxidising Substrate →
Oxidised Catalyst Intermediate

Oxidised Catalyst + Reducing Substrate →
Catalyst + Oxidised Product

The kinetics of both phases of the catalytic cycles have been studied with the objective of elucidating the factors (steric, electronic, electrostatic) which influence reactivity, the natures of the intermediates and the mechanistic origins of enzymic reactivity and selectivity. The reactions of peroxidases (different proteins with the same iron-porphyrin prosthetic group) have been studied with a range of oxidising substrates (hydroperoxides) [3–5] and reducing substrates [6, 7]. Parallel studies have been made with protein-free, water-soluble iron(III)–porphyrins

[3, 8–10] where, in addition, the influence of variation in the porphyrin ligand on reactivity has been examined [11]. The stoichiometry of formation, spectra and reactivity of intermediates formed with a variety of oxidising substrates (hydroperoxides, chlorite) have also been studied [12, 13].

The results reveal some remarkably simple relationships between the reactivities of enzymic and model species and these afford an approach to assessment of the co-catalytic roles played by protein in the expression of enzymic function.

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Q14

A Novel Mechanism of Hemoglobin Cooperativity

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Perutz has proposed that the cooperative effect of oxygen binding in tetrameric hemoglobin arises from an equilibration between two quaternary structures: the liganded or relaxed (R) structure with oxygen affinity comparable to the isolated subunit affinity and the unliganded, tensed (T) structure with oxygen affinity lowered by constraining salt bridges [1–3]. Perutz argues that the equilibrium between these two structures is primarily governed by displacement of the iron and proximal histidine from the mean porphyrin plane and that much of the free energy of heme–heme interaction is stored in salt bridges which break upon oxygenation. This theory has received support from other workers [4–6].

On the other hand EXAFS studies have shown that iron–pyrrol nitrogen bond distances do not differ between deoxyhemoglobin A and deoxyhemoglobin Kempsey (β99 asp → asn) which is a high affinity mutant essentially devoid of cooperativity [7]. Thermodynamic studies of the temperature dependence of $\alpha\beta$dimer–tetramer equilibrium point out that the free energy of heme–heme interaction is stored in hydrogen bonds between dimers and not salt bridges [8]. Replacement of iron by cobalt, which remains low spin even in the unliganded tetramer, only slightly diminishes n, the empirical measure of cooperativity [9–11]. Finally, the displacement of iron from the porphyrin plane in deoxyhemoglobin A seems to differ little from iron displacement in the noncooperative monomer deoxymyoglobin [12].

We wish to propose a truly alternative mechanism for cooperative ligand binding by hemoglobin which does not necessitate metal movement or salt bridge energetics. The important molecular movement for an increase in oxygen affinity is porphyrin sliding from the hydrophobic protein interior to a position with increased porphyrin exposure to solvent. In hemoglobin A, this movement is rigidly coupled to breaking the hydrogen bond between β99 asp and α42 tyr as the porphyrin moves towards the protein exterior, and upon oxygenation formation of a new hydrogen bond between β102 asn and α94 asp. Protein crystallographic studies report that the porphyrins of both α and β subunits are more exposed to water in the met-form than for deoxyhemoglobin [2]. The porphyrin is more exposed to solvent in the β chain of the high affinity mutant deoxyhemoglobin Yakima (β99 asp → his) than A [13]. Studies on model cobaltoporphyrins report that oxygen affinity is more dependent upon porphyrin–solvent interactions than upon 2,4-substituents or ligand *trans* to oxygen [14, 15]. Stellwagen has pointed out that the redox potential of hemoproteins is dependent upon the degree of porphyrin exposure to solvent, with the redox potential decreasing as porphyrin exposure increases [16]. That oxygen affinity increases with increasing porphyrin exposure to solvent is consistent with all the above facts and is tied together by the experiments of Basolo and coworkers who have shown that an inverse linear correlation exists between the logK of oxygenation and cobalt(II/III) redox potential for a series of equatorially substituted cobalt complexes [17, 18].

Recent work upon energetics across the $\alpha_1\beta_2$ interface of hemoglobin shows that amino acid mutations in this region drastically alter cooperative energetics while substitutions at other parts of the molecule have little effect upon cooperativity [19]. Substitution of β99 asp by his, gln or gly destroys the lone hydrogen bond which connects the deoxy-dimers and abolishes all cooperative energy,