5,10,15,20-tetraphenylporphyrinate(-2)) as a halide ligand-to-metal charge-transfer (LMCT) transition. Irradiation into this band leads to rapid photoreduction of the iron atom and dissociation of X•. In the presence of O_2, photoinitiation of hydrocarbon oxidation occurs.

Most of the features in the optical absorption spectrum of Fe(porph)(X) complexes are due to porphyrin-localized $\pi-\pi^*$ transitions.[5] A strong band to higher energy than the Soret band has not been analyzed in the literature. The influence of axial ligation on the energy of this transition permits the unambiguous assignment of this band as a halogen-to-metal charge transfer. In comparison to the Soret or Q-bands, which are $\pi-\pi^*$ transitions and therefore virtually unaffected by the choice of axial anion, this LMCT band shifts strongly to lower energy as the ligand's electronegativity decreases, as shown in Figure 1.

Irradiation into this CT band, and *only* into this band, causes the clean photoreduction of the iron atom in the absence of O_2 (as shown in Figure 2). The rate of $Fe^{II}(TPP)$ production has a strong solvent dependence: cumene > ethylbenzene > toluene > cyclohexane. As shown in Figure 3, an excellent linear free energy relationship exists between the photoreduction rates and the solvents' bond dissociation energies. This is consistent with the abstraction of hydrogen atoms from the solvent as a key step in the overall photoreduction of the iron porphyrin. For Fe(TPP)(Cl) in a 1 M solution of cumene in benzene, the photoreduction quantum yield over 362 ± 11 nm is 5.1×10^{-4}. Laser flash photolysis (at 355 nm) of Fe(TPP)(I) indicates that $Fe^{II}(TPP)$ is formed within the lifetime of the laser pulse (10-ns fwhm). In benzene and in the absence of O_2, this transient does not decay on the nanosecond timescale and only partially returns after tens of microseconds. Our data are consistent with the following mechanism:

$$\text{Fe(TPP)(X)} \xrightarrow[\text{LMCT}]{h\nu} \text{Fe(TPP)} + \text{Cl}^\bullet \quad (1)$$

$$\text{Fe(TPP)} + \text{Cl}^\bullet \rightarrow \text{Fe(TPP)(Cl)} \quad (2)$$

$$\text{Cl}^\bullet + \text{RH} \rightarrow \text{HCl} + \text{R}^\bullet \quad (3)$$

$$\text{R}^\bullet \rightarrow \text{products} \quad (4)$$

Radicals are known to react with Fe(II) porphyrins under acidic, aqueous conditions to give Fe(III) and alkanes.[6] Under our very different conditions, however, we find no evidence for such reoxidation, presumably due to the effectiveness of the secondary reactions of R• (e.g., R-R formation).

This photoreduction mechanism suggests that photocatalytic hydrocarbon hydroxylation with O_2 should be possible via a peroxyl radical chain autooxidation.[7] In fact, upon photolysis of Fe(TPP)(X) in the presence of both substrate and O_2, photoinitiation of hydrocarbon oxidation is observed. In a typical experiment, a benzene solution which was 1 M in substrate and 1 mM in Fe(TPP)(Cl) was irradiated with a Xe arc lamp filtered at 362 ± 6 nm; aliquots were removed periodically and analyzed by capillary GC and GC/MS. For cyclohexene, the expected allylic oxidation products are found (cyclohexen-3-ol (23%) and cyclohexen-3-one (77%)) with a quantum yield of 0.26 and with 150 equiv of products produced per equiv of porphyrin consumed. For cumene, the usual autoxidation products are formed (cumyl alcohol (76%) and acetophenone (24%)), with a quantum yield of 0.30 and with 160 equiv of products produced per equiv of porphyrin consumed. Other substrates with lower oxidizability ratios[7] (e.g., toluene) are not hydroxylated to a significant degree. The eventual fate of the iron porphyrin is observed to be principally $(FeTPP)_2O$, derived from the autoxidation of Fe(TPP), which may be reconverted to Fe(TPP)(Cl) upon addition of HCl.[8]

These results demonstrate that the photochemistry of even simple Fe(III) porphyrins is quite rich and allows a new entry into the oxidation of hydrocarbons with O_2. Further work on the photochemistry of metalloporphyrin complexes with oxoanions is under way.[9]

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Interconversion of Conformation of Apomyoglobin Adsorbed on Hydrophobic Silica Gel

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We wish to report what is, to our knowledge, the first direct observation of multiple conformations and of interconversion between them, for a protein adsorbed to a solid surface. The behavior of proteins adsorbed at solid-liquid interfaces is an area of considerable interest.[1,2] A key concern is the degree of reorganization in the three-dimensional protein structure, if any, that accompanies adsorption. The results from the few studies to date show that changes in protein conformation upon adsorption may range from undetectable to substantial,[3,4] depending on the protein and substrate. A promising technique is intrinsic fluorescence spectroscopy,[5] recently employed to probe changes in fibronectin conformation upon adsorption to hydrophobic silica.[6] We report here on the direct observation of the intrinsic fluorescence characteristics of apomyoglobin in the adsorbed state on a hydrophobic, microparticulate silica gel—Zorbax 100.[7] Our data suggest that the degree of reorganization of the protein in the adsorbed state is dependent on the pH of the contact buffer. At acid pH, the protein is more unfolded and interacts to a greater extent with the surface than at neutral pH and partial reversibility between the two surface conformers is observed.

Sperm whale apomyoglobin was prepared by the methyl ethyl ketone method.[8] Protein solutions were buffered in 20 mM phosphate. The quartz column flow cell[9] was packed with fresh

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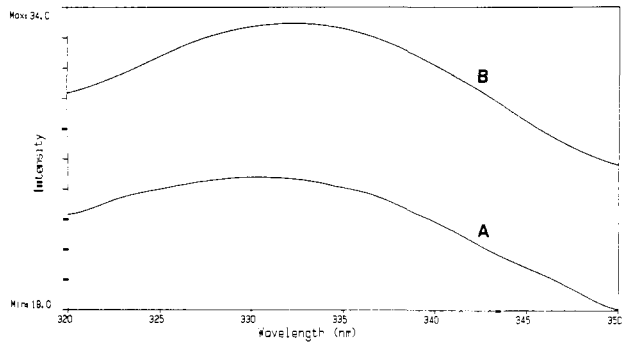

Figure 1. Fluorescence spectra of apomyoglobin adsorbed to Zorbax silica from pH 4.0 buffer: (A) after clearing the flowcell void volume with 1.0 mL of pH 4.0 buffer; (B) after pumping 2.0 mL of pH 7.5 buffer through the cell. The emission maxima of (A) and (B) occur at 330.3 and 332.3 nm, respectively; the intensity units are relative.

Zorbax prior to each experiment. Fluorescence spectra were acquired at stopped flow on a Perkin-Elmer MPF-66 spectrofluorimeter (Ex = 295; slits = 3 nm) at 22 °C. Scattering and silica background luminescence were treated as previously described.[9] After preequilibration with buffer and acquisition of the background spectrum for correction of spectra, a solution of 4.8 μM protein (pH 4.0) was pumped through the cell. When the luminescence from the adsorbed protein became constant, the column was flushed free of solution-phase protein with fresh pH 4.0 buffer. The background-corrected spectrum was obtained, the cell flushed with 2.0 mL of pH 7.5 buffer, and the spectrum obtained again. In another experiment, after apomyoglobin adsorption, the cell was flushed 8 times alternately with fresh pH 4.0 and 7.5 buffer and spectra were recorded.

The emission maxima of apomyoglobin in solution in pH 7.5 and pH 4.0 buffer are 330 and 335 nm.[10] The 5-nm red shift observed upon acidification of the solution is associated with destruction of the heme-binding pocket and loss of approximately half of the α-helical content of the protein.[11] Fluorescence spectra of apomyoglobin adsorbed to Zorbax at pH 4.0 and 7.5 are shown in Figure 1. The emission maximum of apomyoglobin at pH 4.0 is 330 nm, a 5-nm blue shift from the solution value. This suggests that the two tryptophans (positions 7 and 14 in the A α-helix) are in a more hydrophobic environment compared to the solution conformation. This could occur if the protein is either folded into a more compact configuration upon adsorption or is unfolded to allow interaction of the exposed tryptophans with the octadecyl chains bound to the silica gel. We believe the latter case is more likely in light of the red shift in the emission maximum to 332 nm and the increase in intensity observed after pumping pH 7.5 buffer through the cell. It is highly unlikely that apomyoglobin in the adsorbed state is more unfolded at pH 7.5 than at pH 4.0. The 2-nm red shift must represent partial refolding of the adsorbed protein, resulting in less interaction between the tryptophan residues and the hydrophobic surface. The 332-nm emission maximum is 2 nm to the red from that in solution at pH 7.5, indicating partial unfolding of the adsorbed protein at neutral pH. The emission maximum of apomyoglobin adsorbed from pH 7.5 buffer is 334 nm (a 4-nm red shift from the solution maximum of 330 nm). This suggests that the pH 4.0 surface form can be only partially refolded to a pH 7.5 surface form when adsorption has taken place at the more acidic pH.

The correlated fluctuation of the emission maximum and relative intensity (at the maximum) of adsorbed apomyoglobin with the pH of the contact buffer suggests that interconversion between the folded and unfolded states is at least partially reversible (Figure 2). Repeated interconversion is accompanied by a gradual red shift and an overall decrease in fluorescence intensity. The latter may be due to UV photodegradation (observed for albumin adsorbed on silica[1]). A gradual decline in luminescence which cannot be attributed to UV degradation but is associated with adsorption for long periods of time was also observed.

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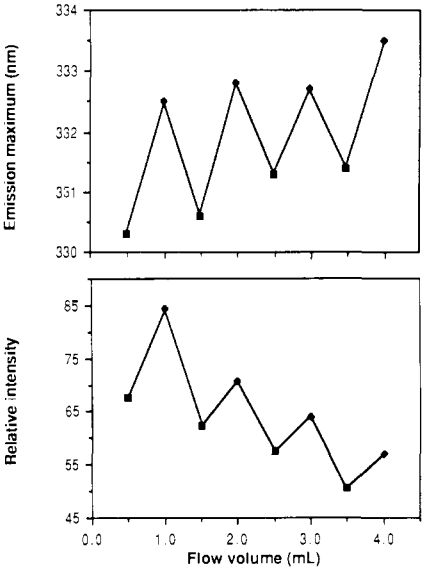

Figure 2. Fluorescence emission maxima and relative intensity (at the maximum) of apomyoglobin adsorbed to Zorbax silica from pH 4.0 buffer: (■) after pumping 0.5 mL of pH 4.0 buffer through the flowcell; (♦) after pumping 0.5 mL of pH 7.5 buffer through the cell.

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A Novel Hypervalent Antimony Ate Complex (12-Sb-6)[1]

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The chemistry of hypervalent ate complexes has recently attracted increasing interest.[2] Concerning that of antimony, there is no example of stable organic 12-Sb-6 compounds other than $Ph_6Sb^-Li^+$,[3a] although several 12-Sb-6 species have been proposed as intermediates in the reactions of Sb(V) with nucleophiles.[4]

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