Oxygen diffusion near the heme binding site of horseradish peroxidase

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The quenching by molecular oxygen of the fluorescence of several probes complexed to apohorseradish peroxidase has been studied by intensity and time-resolved fluorescence methods. The probes utilized include 1-anilino-8-naphthalene sulfonic acid,4,4?-bis(1-anilino-8-naphthalene sulfonic acid), and 2-p-toluidinylnaphthalene-6-sulfonic acid. These results are contrasted to those obtained using apohorseradish peroxidase complexed with protoporphyrin IX. The resistance of these complexes to denaturation by guanidine hydrochloride was also determined. The results demonstrate a dramatic increase in oxygen accessibility to the naphthalene probes compared to protoporphyrin IX, which can be correlated to the increased stability of the protein-protoporphyrin IX complex.