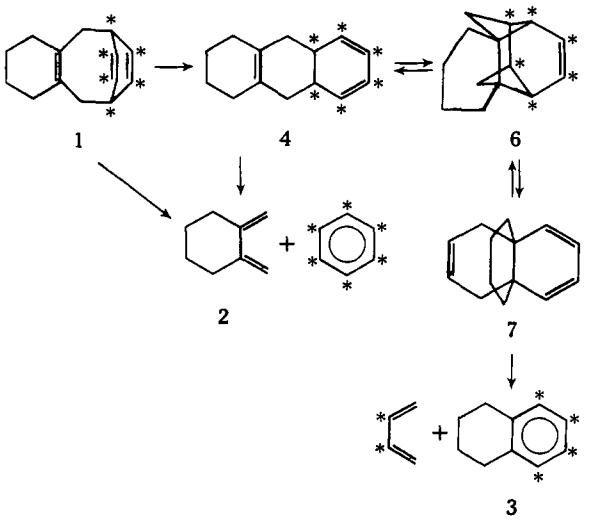
Scheme III



tions implicated in the rearrangements of annulated bicyclo[4.2.0]octatrienes[6] and the degenerate reactions of cyclooctatetraene[7] studied by Paquette and co-workers and also in the thermal behavior of several unsaturated bicyclo[4.2.2] and [4.2.1] species.[8]

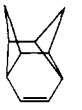
In order to add weight to these proposals, the specifically labeled 3,4,5,6,13,14-hexadeuterio analogue of **1**, prepared by photoaddition of **2** to hexadeuteriobenzene, was also pyrolyzed, and the products were analyzed for deuterium content. The mass spectra of **2** and of benzene were in accord with complete retention of all deuterium in the benzene, while those of **3** and butadiene showed that these molecules were d_4 and d_2, respectively.[12] Additionally samples of tetralin (d_4) and butadiene (d_2) were obtained by preparative GLC and their NMR spectra recorded.

The absence of absorptions (<2% of those of tetralin-H_{12}) in the aromatic region for the tetralin (d_4) indicated no aromatic protons and characterized the tetralin as the 2,3,4,5-tetradeuterio derivative. The NMR spectrum of the butadiene (d_2) consisted of two simple absorptions ($\tau(CCl_4)$ 4.90 (s, 1 H) and 4.96 (s, 1 H)) with negligible absorption in the τ 3.0–4.5 region (<4% of that for butadiene H_6). The observed signals coincide with those of the geminal protons on C_1 and C_4 in fully protonated butadiene and the absence of other absorptions characterizes the butadiene as the 2,3-dideutero derivative. Scheme III traces, by means of asterisks, the fate of the deuterium label through the various isomers in the proposed mechanism and it can be seen that the product isotopic pattern observed is just that expected.

Further investigations of this mechanism and of the thermal stability of the proposed intermediates are under way in our laboratory.

References and Notes

(1) J. C. Berridge, D. Bryce-Smith, and A. Gilbert, *Tetrahedron Lett.*, 2325 (1975).
(2) Samples were prepared by evaporating solvent from 0.50 cm^3 of a solution of **1** in *n*-pentane. The technique gave samples reproducible within ±3% but analysis suggested possible mass losses during transfer up to 14%.
(3) R. Srinivasan and A. A. Levi, *J. Am. Chem. Soc.*, **86**, 3756 (1964).
(4) Thermochemical estimates[5] place **5** about 33 (±5) kcal mol^{-1} higher in enthalpy than **1**. A possible Arrhenius equation consistent with the measured rate constant is $\log (k/s^{-1}) = 14.5 - 37.3$ kcal $mol^{-1}/RT \ln 10$.
(5) Based on methods described in S. W. Benson, "Thermochemical Kinetics", Wiley, New York, N.Y., 1968.
(6) L. A. Paquette, R. E. Wingard, Jr., and J. M. Photis, *J. Am. Chem. Soc.*, **96**, 5801 (1974), and references cited therein.
(7) L. A. Paquette, M. Oku, W. E. Heyd, and R. H. Meisinger, *J. Am. Chem. Soc.*, **96**, 5815 (1974).
(8) (a) W. von Philipsborn, J. Altman, E. Babad, J. J. Bloomfield, D. Ginsburg, and M. B. Rubin, *Helv. Chim. Acta*, **53**, 725 (1970); (b) L. A. Paquette, R. H. Meisinger, and R. E. Wingard, Jr., *J. Am. Chem. Soc.*, **95**, 2230 (1973); (c) M. J. Goldstein and S. A. Kline, *Tetrahedron Lett.*, 1089 (1973), and references cited in these papers.
(9) Thermochemical estimates[5] place **6** about 21 (±6) kcal mol^{-1} above **4**. This means that **6** itself must be very kinetically labile. The prototype, nonannulated molecule, tetracyclo[4.4.0.$0^{2,9}$.$0^{5,8}$]dec-3-ene, shown below, and hereafter named felicene,[10] may well be even less stable



than the *cis*^{2}-bishomobenzene intermediate implicated in analogous rearrangements.[6,7] Another polycyclic olefin which rearranges by a retro-Diels–Alder reaction is basketene[11] which has on the other hand been isolated.
(10) The winning entry in an ad hoc competition involving the authors, A. C. Clements, G. Kennedy, D. A. Penny, and D. J. Smith.
(11) H. H. Westberg, E. N. Cain, and S. Masamune, *J. Am. Chem. Soc.*, **91**, 7512 (1969); correction *ibid.*, **92**, 5291 (1970).
(12) The 70-eV spectra showed parent ions (M) at 136 amu (tetralin) and 56 amu (butadiene). Additionally the M + 1 peaks were in accord with ^{13}C natural abundance in both cases within experimental error indicating the assigned deuterium distribution to apply to at least 95% of these products.

A. Gilbert, R. Walsh*
Department of Chemistry, University of Reading
Whiteknights, Reading, RG6 2 AD, United Kingdom

Received October 24, 1975

Oxygen Binding to Mercaptide–Heme Complexes. Models for Reduced Cytochrome P-450

Sir:

The sustained interest in studying oxygen binding to hemoproteins had led, in the past 2 years, to the preparation of model systems, for both oxymyoglobin and oxyhemoglobin.[1-7] In the majority of these systems, the presence of a nitrogen base at the fifth coordination site of the iron(II) porphyrin was found to be important for heme oxygenation. In this communication we wish to report the binding of oxygen to a heme that contains a mercaptide ion as the axial ligand.

We recently described the preparation of a model compound for reduced cytochrome P-450 as well as its CO complex, in which *n*-butyl mercaptide ion, whose reactivity was enhanced by using a crown ether cation scavenger, served as axial ligand for the protoheme.[8,9] This compound bound CO reversibly and exhibited striking spectral resemblances to those of the P-450 enzyme. However, when this compound was exposed to O_2 at ambient temperature, a drastic spectral change was observed presumably due to the following reactions: oxygenation of heme, autoxidation of heme, and oxidation of the mercaptide ion which would not only decrease the effective concentration of mercaptide available for heme coordination but could aslo result in the production of oxidation products capable of binding to the heme. Since all of these reactions could take place simultaneously spectral evidence for the oxygen binding to heme was ambiguous when the observations were made at room temperature.

It was found, nevertheless, that at lower temperatures the latter two reactions were sufficiently inhibited to allow us to observe clean, and reversible, oxygen binding. Thus at −45° addition of O_2 to the heme-mercaptide complex in dimethylacetamide (DMA) resulted in the spectral change a → b in Figure 1.[10] The spectrum of the oxygen adduct showed no deterioration after 1 h under these conditions. Addition of excess cold pyridine to this solution displaced O_2 and re-

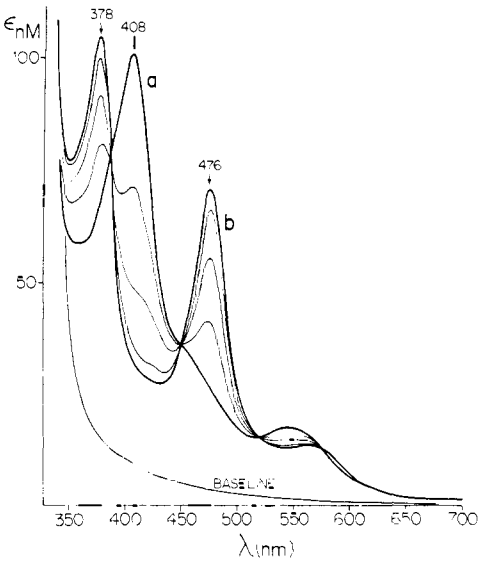


Figure 1. Changes in absorption spectra upon oxygenation of mercaptide-protoheme dimethyl ester complex in DMA at −45°. (a) in vacuo; (b) under 100 Torr of O_2, $[O_2]$ = 2.3, 4.7, and 7.0 Torr, for the intermediate curves, respectively. The baseline was recorded with a DMA solution of BuSK–dibenzo-18-crown-6 complex of the same concentration but without heme.

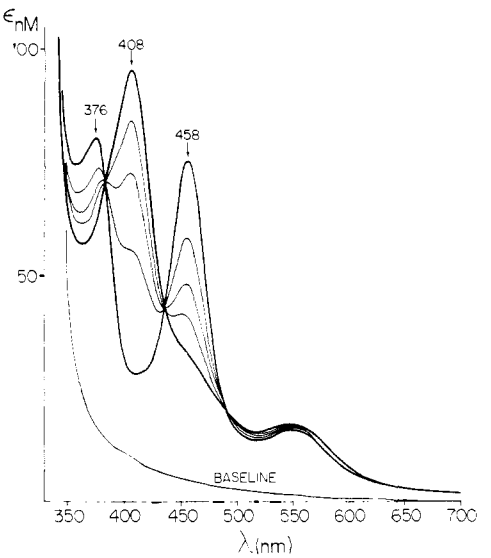


Figure 2. Formation of the mercaptide-protoheme-CO complex in DMA at 24°. [CO] = 0, 4.7, 11, 30, and 760 Torr, respectively.

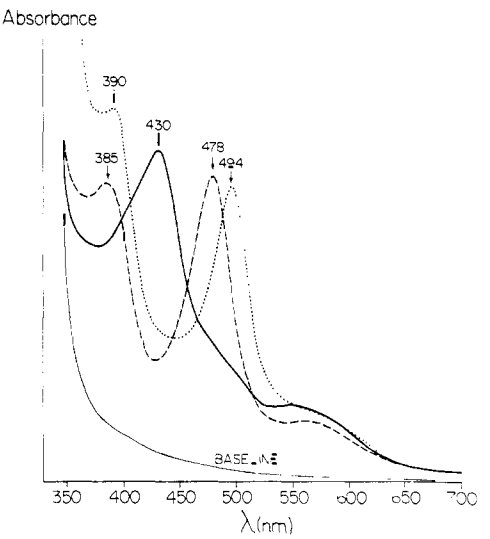


Figure 3. Absorption spectra of O_2 and CO adducts of mercaptide-2,4-diacetyldeuteroheme dimethyl ester complex in DMA: (—) deoxy complex, under argon at 24°; (- - -), CO complex at 24°; (• • •) O_2 complex at −45°.

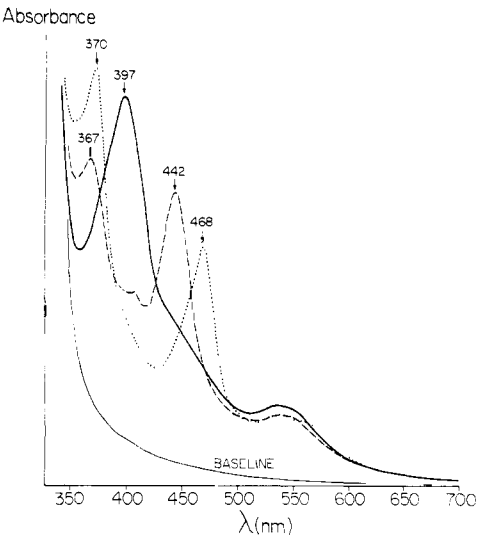


Figure 4. Absorption spectra of O_2 and CO adducts of mercaptide-mesoheme complex in DMA: (—) deoxy complex under argon at 24°, (- - -) CO complex at 24°, (• • •) O_2 complex at −45°.

sulted in the typical pyridine hemochrome spectrum. The stoichiometry of oxygen binding to generate the species with spectrum a has been quantitatively determined by this displacement reaction to give a value of Fe:O_2 of 1:1 at −45°.[11]

It is significant that the Soret band of this $[RS\text{-heme-}O_2]^-$ complex has split into two peaks: one at 476 nm and a second one at 378 nm. In contrast, oxygenated P-450 of both bacterial[12-14] and microsomal[15] origins was found to have a single Soret peak at 418 nm and visible bands around 550 and 580 nm. Careful examination of the region 370–390 nm of the carbon monoxide complex $[RS\text{-heme-CO}]^-$ also revealed a second Soret peak at 376 nm (Figure 2). This intense near-uv band had been observed in many previously published P-450–CO spectra,[13,16] but its significance was not recognized, presumably because this part of the spectrum could easily be mistaken for the strong absorption of dithionite commonly employed as the reducing agent for the enzyme. This type of two-Soret-peak spectra can further be demonstrated to be a general feature of all CO- or O_2-ferrous porphyrin complexes that have mercaptide ion as the axial ligand (Figures 3 and 4).[17]

The wavelength of the Soret peaks is apparently very sensitive to the electron density at the iron atom. With electron-withdrawing groups on the porphyrin ring, decreased electron density at the iron shifts the long wavelength Soret peak to 494 nm in 2,4-diacetyldeuteroheme, while electron rich mesoheme has the peak at 468 nm. Another interesting property of these mercaptide-heme complexes is that, unlike ordinary heme coordination chemistry, their affinity towards O_2 binding is just as strong as CO binding. ($P_{1/2}$ was about 2 Torr for protoheme at −45° in our system.) We believe that the p orbital of sulfur in the mercaptide must interact strongly with iron and result in strong π-electron donation from the sulfur to iron and subsequently to the π^* orbitals of O_2.[18-20] It is this unusually large basicity of the mercaptide compared to other nitrogen π bases which serves to narrow the gap between O_2 and CO binding, since it has been demonstrated that O_2 binding to heme iron demands considerably more π back-bonding than CO.[18]

The splitting of the Soret band of [RS-heme-CO]$^-$ and [RS-heme-O_2]$^-$ has been correlated with other metalloporphyrins such as Sb(III) and Bi(III) porphyrins that bear a close spectral resemblance and a theorectical interpretation for this splitting as well as the red shifting of the Soret band has been proposed.[21] The notable spectral discrepancies between the [RS-heme-O_2]$^-$ complex reported here and the oxygenated P-450 enzymic system is, however, open to speculation at this stage. Nevertheless the present results suggest that while in the ferric deoxy and ferrous CO forms of P-450 a deprotonated cysteine residue may serve as the axial ligand, the oxygenated form of P-450 *cannot* be bound to a mercaptide ion. Whether this change in axial ligation, upon oxygen binding, results from the change of mercaptide to mercaptan or replacement of mercaptide for a non-sulfur-containing ligand must await further experimentation. However, this intriguing change in axial ligation at the critical step of the enzymic reaction may be ultimately related to the activation of molecular oxygen for the hydroxylation reactions catalyzed by these systems.

Acknowledgments. This work is a contribution from the Bioinorganic Chemistry Group and was supported by operating and negotiated development grants from the National Research Council of Canada and the United States National Institutes of Health (AM 17989).

References and Notes

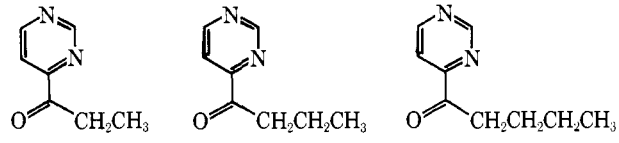
(1) (a) C. K. Chang and T. G. Traylor, *Proc. Natl. Acad. Sci. U.S.A.*, **70**, 2647 (1973); (b) *J. Am. Chem. Soc.*, **95**, 5810 (1973).
(2) W. S. Brinigar, C. K. Chang, J. Geibel, and T. G. Traylor, *J. Am. Chem. Soc.*, **96**, 5597 (1974).
(3) W. S. Brinigar and C. K. Chang, *J. Am. Chem. Soc.*, **96**, 5595 (1974).
(4) J. P. Collman, R. G. Gagne, C. A. Reed, T. R. Halbert, G. Lang, and W. T. Robinson, *J. Am. Chem. Soc.*, **97**, 1427 (1975).
(5) (a) J. E. Baldwin and J. Huff, *J. Am. Chem. Soc.*, **95**, 5757 (1973); (b) J. Almog, J. E. Baldwin, and J. Huff, *ibid.*, **97**, 227 (1975).
(6) G. C. Wagner and R. J. Kassner, *J. Am. Chem. Soc.*, **96**, 5593 (1974).
(7) D. L. Anderson, C. J. Weschler, and F. Basolo, *J. Am. Chem. Soc.*, **96**, 5599 (1974).
(8) C. K. Chang and D. Dolphin, *J. Am. Chem. Soc.*, **97**, 5948 (1975). See also ref 9.
(9) J. P. Collman and T. N. Sorrell, *J. Am. Chem. Soc.*, **97**, 4133 (1975).
(10) The spectrum a was obtained by mixing a DMA solution of heme and a DMA solution of the crown ether complex of potassium butyl mercaptide under argon. The concentration of heme after mixing was about 10^{-5} M; BuSK, 0.6 M; and dibenzo-18-crown-6, 0.3 M. The system also contained 5% H_2O in order to depress the freezing point of DMA to below −50°.
(11) Oxygen bound to mercaptide-heme cannot be removed completely by evacuation at −45° but can be replaced by CO under 1 atm of CO.
(12) Y. Ishimura, V. Ullrich, and J. A. Peterson, *Biochem. Biophys. Res. Commun.*, **42**, 140 (1971).
(13) J. A. Peterson, Y. Ishimura, and B. W. Griffin, *Arch. Biochem. Biophy.*, **149**, 197 (1972).
(14) I. C. Gunsalus, J. R. Meeks, J. D. Lipscomb, P. Debrunner, and E. Munck in "Molecular Mechanisms of Oxygen Activation", O. Hayashi, Ed., Academic Press, New York, N.Y., 1974, p 559.
(15) H. Schleyer, D. Y. Cooper, and O. Rosenthal in "Oxidases and Related Redox Systems", T. E. King, H. S. Mason, and M. Morrison, Ed., University Park Press, Baltimore, Md., 1973, p 469.
(16) F. P. Guengerich, D. P. Ballou, and M. J. Coon, *J. Biol. Chem.*, **250**, 7405 (1975).
(17) In the absence of mercaptide, protheme in DMA can be oxygenated at −45° to give a "normal" oxyheme spectrum: 407, 535, and 571 nm, similar to those published in ref 3. Replacement of mercaptide by mercaptan failed to bring about either type of oxygenated spectrum; but rapid oxidation of the heme, even at low temperature, took place.
(18) C. K. Chang and T. G. Traylor, *J. Am. Chem. Soc.*, **95**, 8477 (1973).
(19) C. K. Chang and T. G. Traylor, *Proc. Natl. Acad. Sci. U.S.A.*, **72**, 1166 (1975).
(20) J. Peisach, J. A. Berzofsky, and W. E. Blumberg in "Oxidases and Related Redox Systems", T. E. King, H. S. Mason, and M. Morrison, Ed., Vol. 1, University Park Press, Baltimore, Md., 1973, p 265.
(21) L. K. Hanson, W. A. Eaton, S. G. Sligar, I. C. Gunsalus, M. Gouterman and C. R. Connell, manuscript in preparation.

C. K. Chang, D. Dolphin*
Department of Chemistry
The University of British Columbia
Vancouver, British Columbia, Canada V6T 1W5
Received August 26, 1975

Photochemistry as a Probe of the Excited State Properties of Molecules. IV.[1] A Determination of the Relative Triplet Reactivities of the Carbon–Nitrogen and Carbon–Oxygen Chromophores of 4-Acylpyrimidines. Photochemical Cyclopropanol Formation

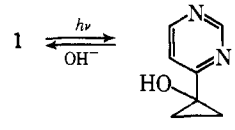
Sir:

In recent years the photochemistry arising from the C=N portion of azaaromatic molecules has received considerable attention.[4] A number of studies have compared the excited-state behavior of the C=N chromophore of aza aromatics to that of the C=O chromophore of ketones.[5] Although the studies have clearly demonstrated qualitative analogies in photochemical behavior, no quantitative comparisons of the excited-state reactivities of the two chromophores have been made. We recently reported[2] that 4-acylpyrimidines could uniquely undergo intramolecular hydrogen abstraction by way of both C=N and C=O triplets. We have since examined the relative triplet reactivities of the C=N and C=O chromophores of 4-propionylpyrimidine (**1**), 4-butyrylpyrimidine (**2**), and 4-valerylpyrimidine (**3**). In this communication we wish to report findings which indicate that the C=N and C=O triplets of 4-acylpyrimidines possess similar reactivity, but surprisingly dissimilar selectivity towards intramolecular hydrogen atom abstraction.



1 **2** **3**

Irradiations of the 4-acylpyrimidines (**1**), (**2**), and (**3**) were carried out at 313 nm in benzene as previously described.[2b] Under the photolysis conditions, ketone (**1**) was found to rearrange exclusively to 1-(4-pyrimidyl)-1-cyclopropanol (**4**). The structure of the cyclopropanol (**4**) is based on its spectral characteristics (ir (CCl_4) 2.80 μ, NMR ($CDCl_3$) methylene δ 1.42 (4 H) multiplet, hydroxyl 2.96 (1 H) broad singlet, aromatic 7.45 (1 H) doublet, 8.60 (1 H) doublet, 9.06 (1 H) singlet; mass spectrum (*m/e*) 136 M^+, 135, 108, 107, 80 (base peak), 79, 57, 55, 53, 52) and on the fact that it reverts back to ketone (**1**) upon treatment with 0.1 N NaOH. Under the same irradiation conditions, ketone (**2**) afforded a mixture of 4-acetylpyrimidine (**5**) and 1-(4-pyrimidyl)-2-methyl-1-cyclopropanol (**6**),[2b] while ketone (**3**) underwent virtually exclusive type II cleavage to 4-acetylpyrimidine (**5**).



4

Since cyclopropanol formation can occur only via γ-hydrogen abstraction by C=N triplets and type II elimination can result only from triplet C=O hydrogen abstraction,[2] each reaction can serve as a probe of the relative triplet reactivities of the two chromophores. The side chain of the pyrimidyl ketone (**1**) permits only C=N abstraction of primary hydrogen atoms. The side chain of the pyrimidyl ketone (**2**), on the other hand, allows for primary C=O hydrogen abstraction and secondary C=N hydrogen abstraction, while that of the pyrimidyl ketone (**3**) permits only secondary C=N and C=O hydrogen abstraction. The nature of the products obtained from the photolysis of ketones **1**, **2**, and **3** clearly indicates that exclusive C=N hydrogen