TABLE I.

Ionizable ligands	Content hemin ($mg/g_{polym.}$)	Kinetic $W \times 10^4$ M RSH / $L.s.g_{pol.}$
NHCH:NCH:CH[a] (imidazole)	5.0	12.1
=NH; ≡N; ≡N–	7.5	9.2
=NH; $-NH_2$	2.4	7.5
$-NH_2$; =NH; ≡N	2.0	7.3
$C_6H_5NCH_3$	2.3	4.5
COOH[b]	<0.1	4.6
$Po(OH)_2$	<0.1	0
$SO_2(OH)$	0	0
C_6H_5NCOOH[c]	<0.1	3.7
$C_6H_5NCH_3$; $PO(OH)_2$	<0.1	3.0

[a]Anion. [b]Cation. [c]Ampholyte type ligands.

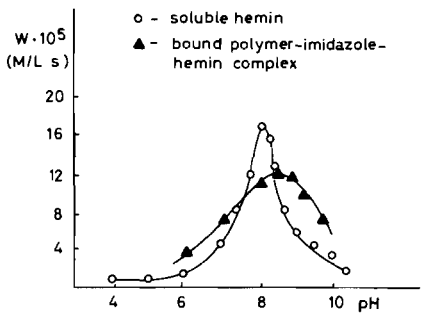

Fig. 1. Catalytic activity.

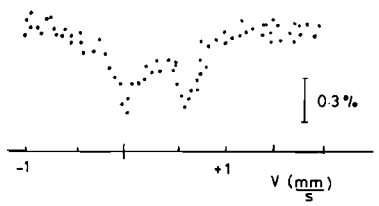

Fig. 2. Mössbauer spectrum of polymer–imidazole–hemin complex at 77 K.

It was found that the adsorption of hemin was higher for polymers with anion type ligands compared with polymers with cation type ones. The ESR spectra of bound species were similar to free hemin ones ($g \simeq 6$, $g \simeq 2$) and were used with optical spectra for controlling the binding process.

The oxidation reaction was obtained at 40 °C in 10 mL water solution containing 2×10^{-3} *M* RSH and 0.1 g polymer-bound hemin species. The highest catalytic activity was observed on polymer–imidazole–hemin complex (1st line of Table I, Fig. 1.)

The reaction for free hemin is first order in RSH, hem, O_2: $W = kC_{RSH}C_{hem.}C_{O_2}$ with concentration of RSH ($5 \times 10^{-2} \div 3 \times 10^{-1}$ *M*); hemin ($3.2 \times 10^{-6} \div 4 \times 10^{-4}$ *M*); O_2($0.9 \times 10^{-3} \div 8.1 \times 10^{-3}$ *M*). The pH dependence of W of bound and free hemin is shown in Fig. 1.

The shift of the peak of the Soret band up to 388 nm of soluble hemin shows some aggregation of hemin in solution. The Mössbauer spectra of polymer-bound hemin species at 300 K and 77 K yield resolved quadrupole doublets with nearly the same values for ΔE_Q (0.7–0.8 mm/s) and δ_{Fe}(0.4–0.5 mm/s) that correspond to high spin ferric states. The differences between these spectra and the unresolved doublet of free hemin arise from Fe–ligands binding and changes in electronic spin relaxation rate due to the distribution of hemin through the polymer matrix. A more detailed Mössbauer study will be carried out with ^{57}Fe enriched samples and measurements at helium temperatures.

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Energetic Correlation of Oxygen Affinities and Soret Absorption Maxima of Deoxyhemeproteins

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The oxygen affinities of hemeproteins have been shown to exhibit a wide range of values [1]. The structural perturbations which influence the oxygen affinity and other properties of these proteins have not been fully elucidated. It has previously been observed that a shift in the positions of the Soret absorption maxima of the α and β chains of hemoglobin is associated with a change in oxygen affinity [2]. A comparison of the physical chemical properties of various monohemeproteins has revealed that a correlation exists between the oxygen affinities and the Soret absorption maxima of the deoxyhemeproteins. Table I shows the $P_{1/2}$ values, corresponding

TABLE I. O_2 Affinities and Soret Absorption Maxima of Deoxyhemeproteins.

Hemeprotein	$P_{1/2}$[a] Torr	λ nm	Ref.
Aplysia Mb	2.7	438	1, 3
Horse Mb	0.70	435	4, 5
Sperm Whale Mb	0.51	434	6, 7
Human Hb α chain	0.46	430	8, 2
Human Hb β chain	0.40	428.5	8, 2
Soybean Lb	0.05	427	9, 10

[a]Pressure of oxygen for one-half oxygenation at pH 7.0 and 20 °C.

to the partial pressures of oxygen at one half oxygenation, together with the Soret absorption maxima of the deoxyhemeproteins. Oxygen affinity appears to decrease as the absorption maximum increases. Furthermore, differences in the free energies of binding of the hemeproteins appear to be related to the differences in energies between the Soret absorption maxima. The standard free energy of binding may be given by

$$\Delta G^\circ = RT \ln P_{1/2}$$

The difference between the free energies of oxygen binding to two hemeproteins is then

$$\Delta(\Delta G^\circ) = RT \ln {}^2P_{1/2}/{}^1P_{1/2}$$

The energy associated with the Soret absorption is

$$E = hc/\lambda$$

The molar equivalent of the difference in energies between the transitions of two hemeproteins is then

$$\Delta E = Nhc\left(\frac{1}{\lambda_2} - \frac{1}{\lambda_1}\right)$$

Table II compares these energy differences for the proteins in Table I. The results show that in general the energy differences are strikingly close to one another. The correlation suggests that for these proteins the difference in oxygen affinities and the difference in wavelengths may be dependent on a common structural perturbation. Banerjee *et al.* [2] have proposed that the O_2 affinity and Soret absorption band are associated with the Fe(II)–histidine axial bond length. Changes in the bond length alter the Fe(II) spin-state equilibrium. It was suggested that alterations in the Fe(II)–N bond length change the spin-pairing energy associated with conversion of the high-spin deoxyhemeprotein to the low-spin oxyhemeprotein. Changes in the Fe(II)–histidine bond length may result from conformational restraints of the protein structure [11] as evidenced by model studies [12, 13]. Other structural factors have also been suggested to influence the oxygen affinity of hemeproteins [14]. It has been shown that the oxygen affinity of model complexes increases with the polarity of the heme environment [14, 16]. This observation is consistent with spectroscopic studies which have demonstrated that the heme in soybean Lb is more exposed to solvent than the heme in sperm whale Mb [10, 17]. Likewise, the relative positions of absorption maxima may be indicative of different heme environments as indicated by the effect of solvent absorption maxima of model heme complexes and chromophores [10, 18–20]. Differences between the O_2 affinities of the monohemeproteins in Table I may, therefore, also be associated with differences in the polarities of the heme environments. These seemingly different explanations may be rationalized by concluding that the polarity of the heme environment effects the Fe(II)–histidine bond length and may thus represent an underlying structural parameter which determines the oxygen affinity as well as other physical chemical properties of the heme group.

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TABLE II. Free Energy of Oxygen Binding and Soret Absorption Energy Differences between Hemeproteins.[a,b]

Hemeprotein	*Aplysia* Mb	HMb	SWMb	α-chain	β-chain	Lb
Aplysia Mb		799	986	1050	1130	2360
		450	602	1210	1450	1680
Horse Mb	799		187	248	331	1560
	450		151	764	997	1230
Sperm Whale Mb	986	187		61	143	1370
	602	151		613	846	1080
Human Hb α-chain	1050	248	61		83	1310
	1210	764	613		233	467
Human Hb β-chain	1130	331	143	83		1230
	1450	997	846	233		234
Soybean Lb	2360	1560	1370	1310	1230	
	1680	1230	1080	467	234	

[a]Energy differences are in calories. [b]The upper number is the energy difference from $P_{1/2}$ data and the lower number is the energy difference from absorption maxima.

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Q25

A Comparison of CO Bound to Metalloporphyrins and Metal Surfaces

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A thorough understanding of the metal–ligand bond is of great importance in biochemistry as well as in catalysis. Much insight can be gained by an analysis of the various factors that govern the properties of this bond for a model system such as carbon monoxide bound to iron in heme or iron embedded in a metal surface [1]. The delocalized sp-states in a metal show a large dispersion and form an electron gas whereas the Fe 4s electrons in heme reside in the surrounding porphyrin. The porphyrin, basically a poly-pyrrol and thus 'metallic', can transfer long-range effects connecting substituents in the periphery with the Fe–C=O or Fe–O=O complex. Such properties can be simulated by a model in which iron is embedded in an electron gas but with different density for metalloporphyrins compared to metals. To what extent the response of such an electron gas to an external field will influence the matrix elements of photoexcitations is not known. The 3d states of a typical transition metal such as iron are well localized within the Wigner–Seitz cell. It is thus plausible that *e.g.* a CO molecule is bound to a particular surface atom since the bond is believed to be a combined $CO5\sigma$–$Fe3d\sigma$ and $Fe3d\pi$–$CO2\pi$ bond. The different crystal field separations of 3d states in the reactive pentacoordinated heme compared to the hexacoordinated heme–CO complex is parallelled by the band-narrowing of 3d-states in the surface layer of a metal. Any change of the 3d states either in metalloporphyrins or in metal surfaces will influence the chemical properties since the changes occur close to the Fermi level, *i.e.* the mean energy of the highest occupied and lowest unoccupied molecular orbitals. The stretch frequencies of the metal–carbon (ν_{MC}) and carbon–oxygen (ν_{CO}) bonds are sensitive probes of these changes. The analysis of such vibrational shifts in terms of electronic structure is however not always straightforward. A decreased redox potential (work-function) is followed by a decreased ν_{CO} [2, 3]. This can be interpreted in terms of an enhanced backbonding to the antibonding $CO2\pi$ level close to the Fermi level. The work-function decrease is provided by preadsorbed potassium on an iron catalyst which means that we cannot completely rule out the possibility of direct CO–K interaction.

In heme models, where the decrease is provided by substitutions in the periphery of the porphyrin, the pyrrols separate the substituents from the CO ligand and direct overlap is negligible. Moreover at high CO coverages of the metal surface we have the possibility of direct CO–CO overlap and formation of $CO2\pi$ electron bands which will influence the occupation numbers and thus the vibrational frequencies [4, 5]. At intermediate coverages where ordered overlayers occur the ν_{MC} frequency can be influenced by phonon bands. The isolation of the direct Fe–CO electronic effects is however possible for a few model systems. One such system is CO bound to iron adsorbed at dilute coverages onto aluminium, a metal which is inert to CO. Another system is heme solvation as well as steric effects or H-bonds to a protein can be ruled out. A comparison of iron and copper porphyrins is valuable. In a Fe–C=O complex the π states will be heavily mixed whereas in Cu–C=O the effect will be merely a broadened 2π level. This is plausible since a copper surface or a copper porphyrin has a low density of states at the Fermi level compared to iron due to an almost completely filled 3d shell.