

Fig. 2

Spectrophotometric titrations of DHL with increasing concentrations of different metal ions. To 50-100 ml of an anaerobic solution of DHL in 0.2 M Tris/sulfate pH 9.00, small volumes of concentrated aqueous solutions of the metal chlorides were added in the given molar ratios. Samples were anaerobically withdrawn for measuring the absorbance spectra 5 minutes after each addition

Work is in progress in order to crystallize and isolate all these compounds, to determine their structure and their electrochemical properties and to test their ability as substrates in the biosynthesis of naturally-occurring structures.

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PS4.7 — MO

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A UNIFIED CONCEPT OF ELECTRONIC PERTURBATIONS OF PORPHYRINS

This communication addresses the problem whether different observables on porphyrins, or hemes, are understandable within a unified model or whether each experimental technique expresses itself in its own language. Does a pattern exist such that an observed quantity of one parameter immediately predicts a quantity of another?

The effects of 2,4-substitutions on a number of observables are collected in Table 1. For most of them a working model based upon delocalized electron density suffices to give a unified view. Removing electron density from the annular structure weakens the N-H bond, increases its H-donor capacity (E_H), and lowers its pK_3 . It also diminishes its Soret band energy (increases λ_{Soret}) and reduces the electron availability at the iron atom. The latter effect, which is observed as a more positive FeIII/II reduction potential (ΔE_m), will also increase ν_{CO} and decrease ν_{FeC} of liganded CO. The trend of these observables persists in the succession $R=\text{CHO}$, COCH_3 , CHCH_2 , H , and C_2H_5 but for a 0,5 nm shift of the Soret band upon H for C_2H_5 substitution. The trend holds also for another two observables, the melting point T_m of the esters and the 1- CH_3 proton chemical shift δ_1 . In these cases, however, the mechanism is less obvious.

On the assumption that T_m reflects the strength of intermolecular attraction certain classes of tunable forces are plausible [1]. The basicity of the pyrrolic nitrogen atoms could modulate the strength of

an intermolecular hydrogen bond. Alternatively, the altered electron density could affect van der Waal's forces. In a first approximation this should correlate the dominant oscillator frequency, i.e. the Soret band energy, to T_m . Such a correlation does exist but opposite to that required to explain the T_m data. Dipole type interactions between the static charge displacements of the substituted porphyrins provide a third possible intermolecular force. Quantum chemical calculations on carbonyl hemes indicate a considerable change of the dipole moment from CH_2CH_3 to CHO even in the plane of the porphyrin.

The 1-CH_3 proton resonance obeys a contact shift as a result of spin density, ρ^{spin} , in the aromatic ring [2]. These shifts correspond to a net spin of 5×10^{-3} electrons at the nearest carbon atom whereas the total shift of electron density, ρ^{electron} , is in the order of some tenths of an electron. Hence it is crucial to reveal whether ρ^{spin} parallels ρ^{electron} or only reflects irregular ripples of ρ^{electron} . Unlike 2,4-substituents protein moieties induce C_2 symmetry changes such that 5- CH_3 resonances parallel 1- CH_3 resonances [3]. Also the protein-induced shifts follow a trend which relates larger chemical shifts to lower reduction potentials. This correlation becomes understandable if one postulates that an enhanced rhombic distortion gives a lower reduction potential. The enhanced x-y asymmetry will then also induce a larger pseudo-contact shift of the proton resonances.

The elucidation of correlations such as those in Table 1 requires a physical description of their

origin. Much information can, however, also be revealed by a statistical evaluation. A route to pattern recognition is offered by the SIMCA programmes [4].

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Table 1
Observables of 2,4-di(R)deuteroporphyrins

R	$T_m^a)$ (°C)	$E_H^b)$ (kJ/mol)	$pK_3^c)$ —	$\lambda_{\text{Soret}}^d)$ (nm)	$\Delta E_m^e)$ (mV)	$\Delta \nu_{\text{CO}}^f)$ (cm^{-1})	$\Delta \nu_{\text{FeC}}^g)$ (cm^{-1})	$\delta_1^h)$ (ppm)
C_2H_5	215	9.65	5.85	400	0	0	0	-12.63
H	219	9.69	5.50	399.5	+0.5	+3	—	-9.99
C_2H_3	228	10.28	4.80	407	+18	+3.5	-3	-9.31
COCH_3	235	10.36	3.35	421	+148	+10.5	-18	-0.72
CHO	289	10.69	~3.0	437	—	+20	—	(+0.10)

a) Melting points of porphyrin dimethyl esters, [1] plus refs. therein.

b) H-bond strengths of substituted pyrroles, $\text{RH}_3\text{C}_4\text{N}\cdots\text{H-F}$ [1].

c) Porphyrin dimethyl esters in detergents [5].

d) Porphyrin dimethyl esters in CHCl_3 [5,6].

e) Reconstituted Mb and HRPC_2 , relative C_2H_5 [7].

f) Reconstituted MbCO and HRPC_2CO and free carbonyl-hemes [7,8].

h) $S=1/2$ Fe(III) biscyano complexes of 2,4-substituted hemes (except for CHO), 1- CH_3 [2].



PS4.8 — TU

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STERIC HINDRANCE INFLUENCE OF IRON(II)-PORPHYRINS ON O₂ AND CO BINDINGS: INFRARED AND RESONANCE RAMAN STUDIES

In designing model compounds for the active site of oxygen carrier hemoproteins, it is important to know the influence of steric hindrance on the O₂ and CO bindings. Indeed if in natural compounds the CO moiety adopt an end-on bent or tilted geometry from the perpendicular to the porphyrin plane [1], in various model complexes CO has been found to adopt a linear geometry [2]. This difference is attributed to distal group steric effect in the formers. On the contrary in both systems the Fe-O-O is bent [3-4].

As part of our studies of stereochemical influence on the formation and reactivity of carbonylated and oxygenated model compounds, we have prepared a new series of porphyrin derivatives specifically designed to investigate such a distal steric effect. The first observations of their infrared (IR) and resonance Raman (RR) spectroscopic studies are reported here.

All the single-face hindered porphyrins are derived from 5,10,15,20 tetraphenylporphyrin. The efficient protection of CO and O₂ coordination site is provided by a polymethylene chain anchored through amide linkage in *ortho* position of two *meso* phenyl groups in a cross configuration. The two other *meso* phenyl groups are substituted in

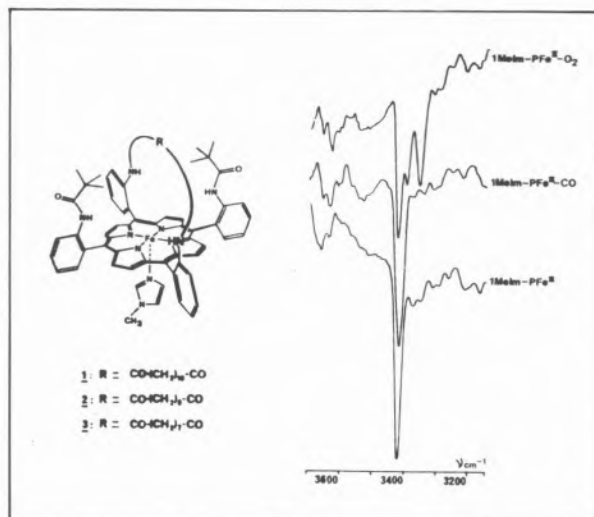


Fig. 1

IR NH stretching of compound 3

ortho position by a pivalamido residue. The presence of these "pickets" inhibits the sideways displacement of the anchoring "handle" in comparison with symmetrical "basket handle" porphyrins previously developed in our laboratory [5], as shown by the strong up-field shifts of methylene protons in ¹H NMR spectra of four-coordinated iron(II) derivatives which are affected by pseudo-contact interaction [6].

IR spectra of iron(II) CO and O₂ complexes were recorded on a Nicolet 5MX FT/IR spectrometer at 4 cm⁻¹ resolution. All RR spectra were obtained with excitation at 441,6 nm using a He/Cd laser (Liconix, model 4050). Samples were prepared by bubbling (1 atm) ¹²CO, ¹³CO (CEA, France, 99 atome %), ¹⁶O₂ and ¹⁸O₂ (CEA, France, 98 atome %) in a toluene or benzene solution of iron(II) derivatives (10⁻⁴ M) and *N*-methylimidazole (10⁻² M). IR spectra of carbonylated derivatives reveal a gradual shift of ¹²C-O stretching vibrations from 1960 cm⁻¹ (1) to 1948 cm⁻¹ (3) which are very different from the observed frequency at 1970 cm⁻¹ of unhindered ferrous-porphyrins CO [7]. It should be noted that compound 3 exhibits ν_{CO} not significantly different from those observed for carboxyhemoglobin and carboxymyoglobin [8]. RR spectra of ¹²CO-1 and ¹²CO-3 show a specific band at 488 cm⁻¹ and 506 cm⁻¹ respectively corresponding to the Fe-¹²CO stretching mode. These bands are sensitive to ¹³CO isotopic substitution and are shifted to 484 and 501 cm⁻¹. The decrease in the C=O stretching frequencies as well as the