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The Nitrogen Oxide Complex of the Iron(II) Protoporphyrin IX Dimethyl Ester

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Synopsis. A nitrogen oxide complex of the iron(II) protoporphyrin IX dimethyl ester was synthesized and characterized by means of its infrared spectrum and electron paramagnetic resonance (EPR) spectra at the X- and Q-band frequencies. The EPR spectra in the glass state and in the polycrystalline powder state distinctly indicated the existence of three *g*-values, showing a rhombic symmetry.

Since the nitrogen oxide molecule has an unpaired electron and coordinates to iron(II) hemoproteins, it has been widely used as a probe for investigating the symmetry around the heme iron and the electronic structure of the heme group.[1] The nitrogen oxide complex of the iron(II) protoporphyrin IX dimethyl ester, ONFe(PPDME), is an interesting model compound in relation to such nitrosylhemoproteins.

TABLE 1. IR DATA OF ONFe(PPDME) (in cm^{-1})

Method or solvent	Dielectric constant	Strteching frequency CO (ester)	NO
Nujol mull		1739	1656
KBr disk		1735	1655
CCl_4	2.24	1743	1684
Benzene	2.28	1741	1672
Toluene	2.38	1739	1673
$CHCl_3$	4.81	1732	1675
CH_2Cl_2	7.77	1735	1669
$ClCH_2CH_2Cl$	10.7	1737	1665
N-Methyl imidazole		1735	1616

Table 1 shows the infrared stretching frequencies of the NO (ν_{NO}) and carbonyl groups (ν_{CO}) at the porphyrin periphery for ONFe(PPDME). The ν_{NO} values, measured by the KBr disk technique, are smaller than those for nitrosyltetraphenylporphinatoiron(II), ONFe(TPP),[2] and nitrosyloctaethylporphinatoiron(II)[3] (1670 and 1671 cm^{-1} respectively). In addition, the ν_{NO} value in *N*-methylimidazole (NMIm), which occupies an axial position *trans* to the nitrosyl group, is smaller than that (1625 cm^{-1}) for ONFe(TPP)(NMIm),[4] while it is nearly identical with that (1615 cm^{-1}) for nitrosylhemoglobin (NOHb),[5] in which the imidazole group of proximal histidine is a *trans* axial ligand. Thus, differences in substituents on the porphyrin periphery affect the NO stretching mode for the nitrosyl porphyrin Fe(II) complex. As is shown in Table 1, the ν_{CO} and ν_{NO} values are sensitive to the solvent. Contrary to the case of the ν_{CO} value, the ν_{NO} value shifts essentially to a lower frequency with an increase in the polarity of the solvent, in agreement with the results of Maxwell and Caughey.[5] Such a solvent effect may reflect a considerable degree of polarization in ONFe(PPDME) resulting from the delocalization of the unpaired electron of the nitrosyl group and the d_π-p_π back bonding.[6]

The X-band EPR spectra in the acetone, dichloromethane, and toluene glass states distinctly indicate the existence of three *g*-values, suggesting a bending of the Fe–N–O unit and rhombic symmetry (Fig. 1 and Table 2). In chloroform glass and polycrystalline powder states, the three *g*-values can not be estimated from the X-band spectra, but they can from the Q-band spectra (Fig. 2). As is shown in Table 2, the *g*-values in the glass state for ONFe(PPDME) are comparable to those for ONFe(TPP),[7] while the coupling constants of the ^{14}N hyperfine splitting (hfs) for the former are slightly smaller than those for the latter. From the facts that the observed g_3-value is essentially equal to the free spin value and that the hfs of NO nitrogen is clearly

TABLE 2. EPR PARAMETERS OF ONFe(PPDME)

	State	Temp[a]	g_{iso}	g_1	g_2	g_3	A_{iso}	A_1	A_2	A_3 (G)
ONFe(PPDME)	powder	RT(Q)		2.093	2.04	2.009				
		LNT(Q)		2.099	2.02	2.006				
	in toluene	RT	2.052				15.8			
		LNT		2.102	2.058	2.010		13	16	16.7
	in $CHCl_3$	RT	2.052				15.8			
		LNT		2.08		2.010				16.6
		LNT(Q)		2.09	2.06	2.010				
	in CH_2Cl_2	RT	2.052				16.1			
		LNT		2.098	2.059	2.010		14	16	16.5
	in acetone	RT	2.051				16.1			
		LNT		2.111	2.061	2.010		12.8	16.4	16.5
ONFe(TPP)[7]	in toluene	RT	2.051				16.3			
		−140 °C		2.103	2.064	2.010		12.6	17.2	17.3

a) RT, room temp; LNT, liquid nitrogen temp; Q, measured at the Q-band freq.

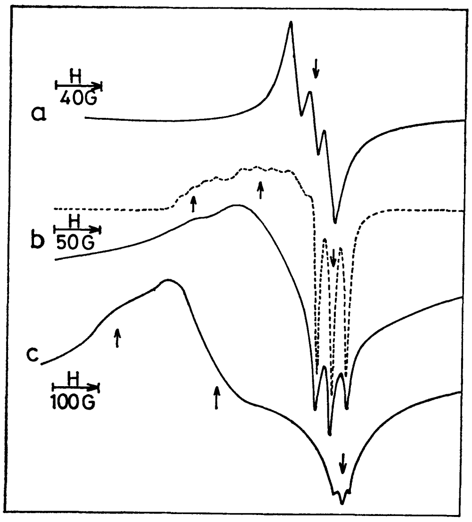

Fig. 1. EPR spectra for ONFe(PPDME) in $CHCl_3$ (——) and in acetone (-----): (a) X-band spectra at room temp (soln); (b) X- and (c) Q-band spectra at liq N_2 temp (frozen glass).

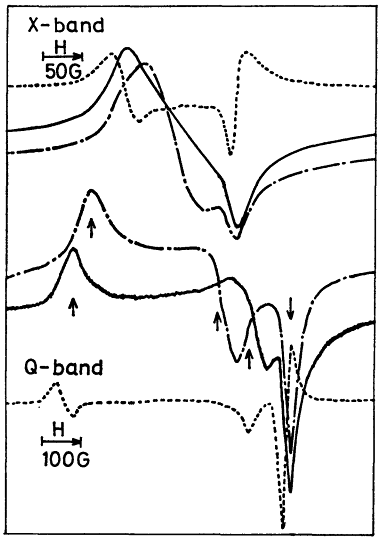

Fig. 2. X- and Q-band EPR spectra for ONFe(PPDME) in polycrystalline powder state at room temp (-·-·-, 1st derivative) and at liq N_2 temp (——, 1st derv.;----, 2nd derv.).

resolved at g_3 absorption, it may be inferred that the g_3 absorption is to be assigned to the g_z absorption and that the unpaired electron of the nitrosyl group is considerably delocalized to the iron d_{z^2} orbital. The g-values for ONFe(PPDME) are found to vary with the experimental conditions, such as the temperature and the powder or solution states, and, among the rest, the g_2-value is especially highly sensitive to such conditions. The g_y-value for NO-protoheme complexes with different nitrogenous bases has been reported to be particularly sensitive to the kind of base.⁶⁾ The g_y-value for the $(d_{z^2})^1$ configuration is dependent on the energy separation between d_{z^2} and d_{xz} orbitals and the electron population in each orbital.⁸⁾ Thus, the energy and the electron population of the d_π orbital can be expected to be sensitive to environments around Fe–N–O unit in the nitrosylprotoheme complexes.

It has been found that, upon the addition of inositol hexaphosphate (IHP) to NOHb, the hfs from imidazole ^{14}N of proximal histidine (N_ε) is weakened and that from ^{14}NO is intensified.⁹⁾ The coupling constant (16.5 G) and the g-value (about 2.0) of the central peak for the ^{14}NO triplet in IHP-treated NOHb are nearly identical with those of ONFe(PPDME) in the glass state, showing that the structure of the nitrosylprotoheme in IHP-treated NOHb is similar to that of ONFe(PPDME). These EPR results suggest that the Fe–N_ε bond in IHP-treated NOHb is stretched or cleaved.⁵⁾

Experimental

Synthesis. ONFe(PPDME) was synthesized by a procedure analogous to that used for ONFe(TPP).²⁾ One gram of ClFe(PPDME) was added to a chloroform(10 ml)–dry pyridine(1 ml) mixture. The solution thus obtained was deoxidized by bubbling pure N_2 for 10 min. Nitrogen oxide, passed through a KOH column to free it of higher nitrogen oxides, was bubbled into the solution for an hour, and then the excess nitrogen oxide was removed by bubbling N_2 through the solution. Methanol, distilled from Mg, was added to the solution until crystals began to precipitate. The products were filtered off, washed with methanol, and recrystallized from chloroform-methanol, to obtain dark purple crystals of ONFe(PPDME). Calcd for $FeC_{36}H_{36}N_5O_5$: C, 64.10; H, 5.38; N, 10.38; Fe, 8.28%. Found: C, 63.85; H, 5.47; N, 10.35; Fe, 8.22%. The analytical results and the IR spectrum confirmed the absence of pyridine in the product. The procdut was handled in a dry box filled with nitrogen.

Measurements. The IR absorption spectra were recorded on Jasco DS-402G and IR-G IR grating spectrophotometers. The EPR measurements were carried out using a JES-ME-3X (X-band) spectrometer and a JES-PE-3Q (Q-band) spectrometer, both with 100 KHz field modulation. The second derivative display was obtained by the use of the 80 Hz field modulation. As a standard, DPPH powder and MgO powder doped with Mn^{2+} were used. All the solvents were spectrograde reagents and were deoxidized by bubbling pure N_2 prior to use.

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