

Synthetic Porphyrins II

Preparation and Spectra of Some Metal Chelates of *para*-Substituted-*meso*-Tetraphenylporphines

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Various new metalloporphyrins, including both Fe(II) and Fe(III), Co(II), and Co(III) complexes of *p*-substituted-tetraphenylporphines, were synthesized for use as possible model compounds in the study of the mechanisms of the oxidation-reduction of the heme-enzymes and cobalamines. Characteristic differences were observed in the near-ultraviolet, visible, and infrared spectra of the complexes of different metal ions. These compounds may also be of interest as specific radiation sensitizing or protecting agents.

THE IMPORTANT biological roles of metalloporphyrins (heme, chlorophyll) and of the structurally related metalocorrins (vitamin B₁₂) suggested that the appropriate metal chelates of certain synthetic *para*-substituted-*meso*-tetraphenylporphine derivatives (1) may serve as antimetabolites, or as model compounds in the study of the modes of action of these interesting natural substances. In addition, such compounds would be expected to be effective in absorbing radiation energy at certain wavelengths and may serve either as radiation sensitizing or as radiation protecting agents (2). It is of particular interest that some porphyrin derivatives apparently show selective distribution properties in the various tissues. Hematoporphyrin was reported to accumulate selectively in tumors and other rapidly growing tissues (3-5) and was used as a fluorescent indicator for the delineation of neoplastic tissue in cancer patients (6). It would seem, therefore, that certain metalloporphyrins may be used as tumor-selective radiation sensitizers or "carriers" of various cytotoxic groups and/or metals [e.g., Hg (7) or ⁵⁷Co (8)] in the chemotherapy of neoplastic diseases.

Recently, Winkelman and co-workers reported (8, 9) that a sulfonated derivative of *meso*-tetraphenylporphine was even more selectively localized (attaining a 50-100 times higher concentration in the tumor tissues) than hematoporphyrin, but that the ⁵⁷Co chelate of this sulfonated compound lost its metal and only the free ligand was absorbed. This is in contrast with the reported stabilities of various injected Zn (3), Hg (7), and ⁶⁴Cu (10) porphyrins through-

out their organ-uptake and/or excretion and is, of course, also in contradiction with the known stability of the Co-corrin derivative, vitamin B₁₂. Thus, the fate of the chelated metal during the absorption and localization of the metalloporphyrins in the tissues requires further investigation.

In the present paper, the synthesis and physicochemical properties of various metal chelates of the recently reported (1) *para*-substituted-*meso*-tetraphenylporphines are described. Due to the interest in obtaining model compounds for the study of the mechanisms of oxidation-reduction of the various heme-enzymes as well as of the cobalt in the various cobalamincoenzyme mediated reactions, particular efforts were centered on the synthesis of metalloporphyrins containing Fe and Co, respectively, at various oxidation states. The structures and chemical names of the new metalloporphyrins described in this paper are shown in Table I.

RESULTS AND DISCUSSION

The metalloporphyrins (I to VII) were synthesized by refluxing the ligand (1) and the metal salt in a suitable solvent (see under *Experimental*). Progress of the reactions was followed by observing the changes in the visible spectra of the reaction mixtures. The reactions were stopped when complete disappearance of the characteristic visible absorption maxima of the ligands occurred. The choice of solvent, due to solubility problems, was limited to pyridine, glacial acetic acid, and 88% formic acid. The suitable solvent for each preparation could be found only by carrying out small-scale reactions. The choice of metal salts was limited to the readily available chlorides or acetates. The ferrous complex (compound IV) was prepared and purified under nitrogen atmosphere, since the Fe(II) rapidly oxidized to Fe(III).

Compound VIII was prepared by two alternative methods, that is, either by direct chelation of the corresponding ligand, *meso*-tetra-(*p*-carboxyphenyl)porphine, or by alkaline hydrolysis of *meso*-tetra-(*p*-carbomethoxyphenyl)porphinocobalt(II) (compound VII). The latter compound (VII) was solu-

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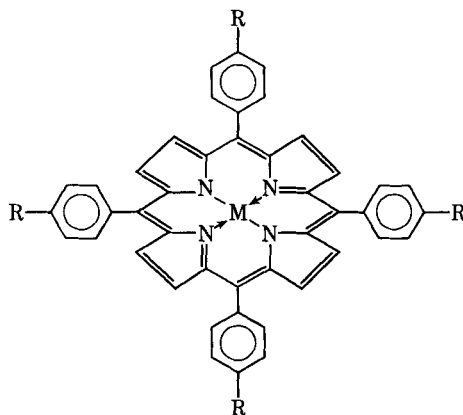
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TABLE I—STRUCTURES AND CHEMICAL NAMES OF NEW SYNTHETIC METALLOPORPHYRINS



	R	M	Name
I	CH ₃	Co(II)	<i>meso</i> -Tetra- <i>p</i> -tolylporphinocobalt(II)
II	CH ₃	Cu(II)	<i>meso</i> -Tetra- <i>p</i> -tolylporphincopper(II)
III	CH ₃	Zn(II)	<i>meso</i> -Tetra- <i>p</i> -tolylporphinozinc(II)
IV	CH ₃	Fe(II)	<i>meso</i> -Tetra- <i>p</i> -tolylporphinoiron(II)
V	CH ₃	Cl-Fe(III)	<i>meso</i> -Tetra- <i>p</i> -tolylporphinoiron(III) chloride
VI	CN	Cl-Fe(III)	<i>meso</i> -Tetra-(<i>p</i> -cyanophenyl)porphinoiron(III) chloride
VII	CO ₂ CH ₃	Co(II)	<i>meso</i> -Tetra-(<i>p</i> -carbomethoxyphenyl)porphinocobalt(II)
VIII	COOH	Co(II)	<i>meso</i> -Tetra-(<i>p</i> -carboxyphenyl)porphinocobalt(II)
IX	CH ₂ OH	Co(II)	<i>meso</i> -Tetra-(<i>p</i> -hydroxymethylphenyl)porphinocobalt(II)
X	CH ₃	Br-Co(III)	<i>meso</i> -Tetra- <i>p</i> -tolylporphinocobalt(III) bromide

ble in dry tetrahydrofuran and was also used as the starting material for the synthesis of *meso*-tetra-(*p*-hydroxymethylphenyl)porphinocobalt(II) (compound IX). Before attempting to reduce VII with lithium aluminum hydride, *meso*-tetra-*p*-tolylporphinocobalt(II) (compound I), as a model compound, was treated with lithium aluminum hydride in dry tetrahydrofuran solution in order to determine whether the solubility of the cobalt complex and the chelation or the oxidation state of cobalt(II) would be affected by a reaction with the reagent. Only the unchanged starting material (I) was isolated; thus lithium aluminum hydride did not seem to have any effect on the chelated Co(II) ion. However, it is quite possible that the divalent cobalt in I was reduced by lithium aluminum hydride to monovalent cobalt but the latter was rapidly reoxidized to divalent cobalt during work-up (11). Thus, reduction of VII with lithium aluminum hydride gave the Co(II) chelate of the hydroxymethyl derivative (IX) in 79.3% yield.

Meso-tetra-*p*-tolylporphinoiron(III) (compound X) was prepared from I in a manner analogous to the synthesis of the corresponding derivative of aetioporphyrin (I) by Johnson and Kay (12). Attempts were also made according to the procedure of the same authors (12) to synthesize *meso*-tetra-*p*-tolylporphindicyanocobaltate. The product obtained contained theoretical amount of cyanide radicals, nitrogen and hydrogen, but the analytic results for C, Co, and K were unsatisfactory. The material was stable in alcohol but slowly decomposed in chlorinated solvents. Further work on the nature of the *meso*-tetra-*p*-tolylporphindicyanocobaltate is in progress.

The near-ultraviolet and visible spectra of the metal complexes of tetraphenylporphines are characterized by a strong "Soret band," in the 400–440 m μ region, and by another strong absorption band ("K-band") in the visible spectral region; the latter is usually accompanied by much weaker vibrational bands ("R-bands") (13). Thus, these metalloporphyrins differ from the corresponding "free base" *meso*-tetraphenylporphines by the absence of the characteristic five-banded patterns in the visible spectra of the latter (1), and also from other metalloporphyrins (e.g., etio-type) which generally show a different, two-banded (α and β -bands) pattern in their visible spectral region (14).

The absorption maxima of the new metalloporphyrins are shown in Table II. Molar extinction coefficients of all, except III, VI, and VIII, have been calculated in two different solvents. Compounds III and VIII could be quantitatively dissolved only in pyridine and 5% aqueous sodium carbonate, respectively, whereas compound VI could not be quantitatively dissolved in any solvent. It is apparent from Table II that a change of the solvent from benzene (or chloroform) to pyridine, results in certain changes in the positions or intensities (or both) of the absorption bands; these changes vary in magnitude from one metal chelate to another. In either solvent, the spectra show characteristically different patterns dependent on the chelated metal ion; different *p*-phenyl substituents in the ligands have relatively small influence on the spectral patterns of the metal chelates. The Soret bands of all metal chelates, with the exception of Cu^{II} and Zn^{II}, are significantly less intense than those of the corresponding free base porphyrins

TABLE II—NEAR ULTRAVIOLET AND VISIBLE ABSORPTION SPECTRA OF NEW METALLOPORPHYRINS

Compd. (Solvent)	"Soret Band"	$\lambda_{\max.}$ (m μ) ($\epsilon \times 10^{-3}$)				
		"K and R Bands"				
I (Benzene)	415		530	612		
	(279)		(17.0)	(3.96)		
(Pyridine)	414		538			
	(147)		(9.6)			
II (Benzene)	424	523	552	592	650	
	(472)	(4.83)	(16.7)	(4.83)	(1.36)	
(Pyridine)	431	520	564	605		
	(672)	(6.5)	(20.8)	(13.6)		
III (Benzene)	424	514	549	590		
	(650)		(14.2)	(3.4)		
(Pyridine)	422		545	570		
	(121)		(9.64)	(5.66)		
IV (Benzene)	416		571	614		
	(112)		(8.3)	(4.8)		
V (Benzene)	420		512	547	587	650
	(136)		(5.62)	(2.25)	(1.5)	(1.5)
(Pyridine)	424	485	530	590	649	694
	(218)	(10.0)	(16.2)	(2.9)	(2.1)	(1.6)
VI (Chloroform)	419		527	560	598	651
	(Pyridine)	426	508	548	572	600
VII (Tetrahydrofuran)	411		526			
	(253)		(14.8)			
(Pyridine)	416		534			
	(166)		(6.8)			
VIII (5% aq. Na ₂ CO ₃)	430		547	586		
	(247)		(13.5)	(6.3)		
IX (Tetrahydrofuran)	410		528			
	(226)		(13.5)			
(Pyridine)	414		534			
	(196)		(7.6)			
X (Benzene)	410		545			
	(122)		(13.3)			
(Pyridine)	439		556	594		
	(170)		(8.2)	(5.0)		

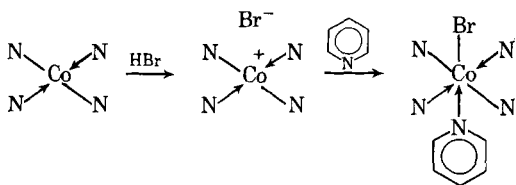
(1). The number of the visible absorption bands of the metal chelates vary between one (Co^{II}) and five (Fe^{III}).

The spectrum of the Co(III) complex (X) very markedly differs from those of the Co(II) chelates in pyridine solution, but the difference is less pronounced when the spectra are compared in benzene. The more significant difference in the spectrum of X in pyridine is probably due to the extra ligand action of a molecule of pyridine with the trivalent cobalt. The spectrum of X in pyridine may correspond to that of an octahedral pyrimidinobromocobalt(III) complex, as proposed by Johnson and Kay (12) (Scheme I).

The spectra of the Fe(II) complex (IV) in benzene and pyridine are similar to each other, except for the position of the Soret band. The intensity of this band is remarkably low in both solvents. No other Fe(II) complex of a *meso*-tetraphenylporphine has been reported previously. The spectra of the corresponding Fe(III) complex(V) in either benzene or pyridine, are quite different from those of the Fe(II) complex. Although the spectra of V in the two solvents considerably differ from each other, their

composite pattern shows remarkable similarity to the spectrum of the free base porphyrin (1) in the positions of the absorption bands. However, the intensity of the Soret band of V is extremely low (particularly, in the benzene spectrum), just as in the case of the Fe(II) complex (IV).

The infrared spectra of the new metalloporphyrins, obtained in potassium bromide phase, are shown in Table III. The weak N-H stretching vibrations above 3300 cm.⁻¹ observable in the spectra of all the ligands (1) having no hydroxyl group, are missing from the spectra of their chelates, due to the replacement of N-H protons by metal ions. Probably the most characteristic difference between the infrared spectra of a tetraphenylporphine ligand and its metal chelates is that the three bands appearing between 960–1000 cm.⁻¹ in the spectra of the former (1) collapse to one strong band near 1000 cm.⁻¹ in the spectra of the latter. This absorption band has been associated with a C-H vibration of the pyrrole rings. In a limited series of metalloporphyrins containing certain divalent cations, Thomas and Martell (15) were able to correlate the relative frequencies of this band with the relative stabilities of chelation. In the present series, the metal complexes of *meso*-tetra-*p*-tolylporphine, (I–V, and X) show the following relative order of frequencies for the "1000 cm.⁻¹ band": Co(II) > Fe(II), Fe(III) > Cu(II) > Zn(II) > Co(III). Except for the relative positions of the iron chelates, this series seems to agree with the stability order of metalloporphyrins as determined by various methods (16, 17).



Scheme I

TABLE III—CHARACTERISTIC INFRARED ABSORPTION SPECTRAL BANDS OF METALLOPORPHYRINS I TO X

I	II	III	IV	V	VI	VII	VIII	IX	X	Assignments
								3420 (m)		O—H (stretch)
					2175 (m)					—C≡N
						1725 (vs)	1690 (vs)			—C=O
1602 (vw)	1595 (w)				1602 (w)	1600 (s)	1600 (s)	1600 (w)		—C=C— (phenyl)
1535 (m)	1520 (vw)	1525 (vw)	1524 (wsh)		1535 (m)	1565 (w)	1565 (w)	1540 (vw)		—C=C— (pyrrole)
1500 (m)	1480 (m)	1500 (m)	1480 (m)	1480 (m)		1480 (wsh)		1500 (vw)	1475 (m)	—C=C— (phenyl)
1442 (m)	1435 (w)	1439 (w)	1440 (w)	1440 (wsh)	1440 (w)	1435 (m)			1440 (m)	—C—H bend (pyrrole)
1347 (s)	1330 (s)	1340 (s)	1325 (s)	1327 (m)	1320 (s)	1360 (m)	1350 (m)	1343 (s)	1340 (s)	=C—N— stretch
1202 (m)	1202 (ms)	1205 (m)	1200 (ms)	1200 (w-m)		1207 (m)	1240–1260 (s; broad)	1200 (m)		
1178 (ms)	1176 (ms)	1180 (ms)	1178 (ms)	1178 (s)		1175 (m)	1204 (m)	1175 (m)		
1105 (m)	1104 (m)	1104 (m)	1105 (m)	1107 (w)	1113 (m)	1105 (s)	1175 (m)	1103 (w)	1102 (m)	<i>p</i> -Substituted phenyl
1070 (ms)	1064 (ms)	1070 (ms)	1065 (m)	1067 (w)			1100 (m)	1070 (m)		
1000 (vs)	995 (vs)	992 (vs)	997 (vs)	997 (vs)	996 (vs)	1002 (vs)	1002 (vs)	998 (vs)	992 (vs)	—C—H rock
			871 (s)			867 (m)	866 (m)			
847 (w)	846 (m)	845 (m)	846 (w)	844 (w)		827 (m)	879 (m)	850 (w)		
797 (vs)	794 (vs)	798 (vs)	796 (vs)	800 (vs)	800 (vs)	795 (s)	795 (vs)	796 (vs)	794 (s)	<i>p</i> -Substituted phenyl
			750 (m)			767 (s)	767 (m)			Pyrrole ring
720 (ms)	720 (ms)	719 (ms)	719 (s)	722 (s)		718 (ms)	715 (m)	713 (ms)	713 (m)	
	693 (m)									

TABLE IV—SYNTHESIS OF METALLOPORPHYRINS I TO IX BY INSERTION OF METAL-ION INTO APPROPRIATE LIGAND (FREE BASE PORPHYRIN)

Compd.	Ligand	Metal Salt	Time of Reflux	Solvent		Anal., %	
				Reaction	Recrystallization	Calcd.	Found
I	<i>p</i> -CH ₃ -TPP ^a	CoCl ₂ ·6H ₂ O	22 hr.	Pyridine	Chloroform	C, 79.23; H, 4.95; N, 7.64	Found C, 79.25; H, 4.83; N, 7.71
II	<i>p</i> -CH ₃ -TPP	Zn(OCOCH ₃) ₂	30 min.	Pyridine	Chloroform-ethanol	C, 78.74; H, 4.92; N, 7.66	Found C, 77.91; H, 4.80; N, 7.84
III	<i>p</i> -CH ₃ -TPP	Cu(OCOCH ₃) ₂	30 min.	Pyridine	Chloroform	C, 78.54; H, 4.91; N, 7.64	Found C, 78.89; H, 4.90; N, 7.51
IV	<i>p</i> -CH ₃ -TPP	FeCl ₂	12 hr.	Pyridine	Chloroform-ethanol	C, 79.57; H, 4.97; N, 7.74	Found C, 79.00; H, 4.82; N, 7.78
V	<i>p</i> -CH ₃ -TPP	FeCl ₃	5 hr.	Pyridine	Chloroform-ethanol	C, 75.85; H, 4.74; N, 7.37	Found C, 76.27; H, 4.72; N, 7.54
VI	<i>p</i> -CN-TPP	FeCl ₃	5 hr.	Pyridine	Pyridine	C, 71.69; H, 2.99; N, 13.94	Found C, 72.10; H, 3.33; N, 13.52
VII	<i>p</i> -CO ₂ Me-TPP	CoCl ₂ ·6H ₂ O	3 hr.	Acetic acid	Chloroform	C, 69.10; H, 3.99; N, 6.20	Found C, 69.49; H, 4.60; N, 6.14
VIII	<i>p</i> -COOH-TPP	CoCl ₂ ·6H ₂ O	3.5 hr.	90% HCOOH	AcOH- HCOOH	Co, 6.97	Found Co, 6.96
IX	<i>p</i> -CH ₂ OH-TPP	CoCl ₂ ·6H ₂ O	1 hr.	Acetic acid	Tetrahydrofuran- chloroform	C, 72.82; H, 4.55; N, 7.08	Found C, 73.61; H, 4.93; N, 7.14

^a TPP = *meso*-tetraphenylporphine.EXPERIMENTAL¹

General Method of Synthesis by Insertion of Metal Ion—In general, the metalloporphyrins were prepared by refluxing the appropriate ligand (I) with 4–5 times excess of a metal salt and a solvent, for a certain length of time (until disappearance of the

free base spectral bands) as given in Table IV. After evaporation of the solvent, the excess inorganic salt was removed by washing with water, and the reaction product, obtained in the residue in essentially quantitative yield, was recrystallized from a suitable solvent or solvent mixture (Table IV).

Alternative Method for the Preparation of VIII—The *meso*-tetra-(*p*-carboxyphenyl)porphino-cobalt-

¹ Microanalyses were performed by Galbraith Laboratories, Inc., Knoxville, Tenn.

(II) was also synthesized by basic hydrolysis of *meso*-tetra-(*p*-carbomethoxyphenyl)porphino-cobalt(II) (VII). A mixture of VII, 3 Gm., 4% aqueous potassium hydroxide solution, 100 ml., and tetrahydrofuran, 500 ml., was refluxed for 24 hr. The solvents were removed *in vacuo*. The residue was dissolved in water and neutralized with 6 *N* sulfuric acid. The purple precipitate was centrifuged and repeatedly washed with water. The material was dried over anhydrous calcium chloride. Yield: 2.66 Gm. (94.5%). This product had identical spectra with that obtained by the insertion method.

Alternative Method for the Preparation of IX—A mixture of *meso*-tetra-(*p*-carbomethoxyphenyl)-porphino-cobalt(II) (VII), 1.7 Gm., lithium aluminum hydride, 1.7 Gm., and tetrahydrofuran (freshly distilled over lithium aluminum hydride), 50 ml., was refluxed for 2 hr., then 1 additional Gm. of lithium aluminum hydride and 50 ml. of dry tetrahydrofuran were added to the reaction mixture. Refluxing was continued for 2 more hr., followed by decomposition of the unreacted lithium aluminum hydride by moist ether at 0°. The slurry thus obtained was filtered and washed with 150 ml. of tetrahydrofuran. On evaporation of the solvent, 1 Gm. of purple crystalline solid was obtained. An additional 0.16 Gm. of material was obtained by further washing of the aluminum hydroxide slurry with 100 ml. of tetrahydrofuran, followed by evaporation of the solvent. Total yield: 1.16 Gm. (79.3%).

Synthesis of Meso-tetra-*p*-tolylporphino-cobalt(III)Bromide(X)—The *meso*-tetra-*p*-tolylporphino-cobalt(II) (I), 300 mg., was dissolved in 50 ml. of chloroform, and to this was added 50 ml. of methanol and 1 ml. of 47.5% hydrobromic acid. The mixture was stirred at room temperature for 2 hr., and then 50 ml. of chloroform was added. The reaction mixture was washed with water, dried over anhydrous sodium sulfate, and evaporated to dryness. The residue was crystallized from a mixture of chloroform and petroleum ether (b.p. 40–60°). Yield: 300 mg. (90%). An analytical sample was obtained by recrystallization from the same solvent mixture and drying over phosphorus pentoxide at 100° for 8 hr.

Anal.—Calcd. for C₄₈H₃₆BrCoN₄: C, 71.38; H, 4.46; N, 6.94. Found: C, 71.29; H, 4.62; N, 6.73.

Spectral Measurements—The near ultraviolet and visible absorption spectra (Table II) were determined on Beckman DU (attached to a Gilford absorbance indicator, model 2000) and Beckman DB spectrophotometers. The infrared spectra (Table III) were determined on Perkin-Elmer model 137 and Beckman model IR-8 spectrophotometers.

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Keyphrases

Porphyrins—synthetic
 Metalloporphyrins—synthesis
meso-Tetraphenylporphyrins, *para*-substituted metal chelates
 UV spectrophotometry—structure
 Visible spectrophotometry—structure
 IR spectrophotometry—structure