Synthesis of Cationic Metalloporphyrin Precursors Related to the Design of DNA Cleavers

Christiane Casas, Bernadette Saint-Jalmes, Christophe Loup, C. Jeffrey Lacey, and Bernard Meunier*

Laboratoire de Chimie de Coordination du CNRS, 205 route de Narbonne, 31077 Toulouse cedex, France

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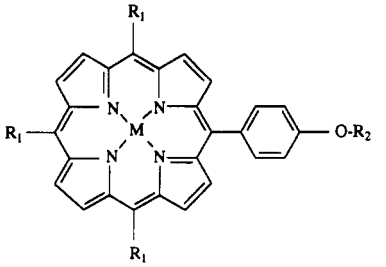
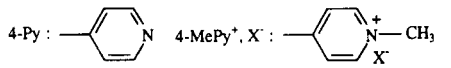
The design of specific DNA cleavers is a growing field of research for several reasons: (i) DNA is the biological target of a large number of antitumor agents which are able to cleave double-stranded nucleic acids, bleomycin,[1] neocarzinostatin,[2] and enediyne molecules,[3] (ii) transition metal complexes linked to a DNA recognizing molecule (intercalating agents,[4] peptides,[5] or oligonucleotides[6]) are also able to cleave selectively single- or double-stranded nucleic acids, and (iii) some of these hybrid molecules, "DNA cleaver-vector", might have a future as antiviral or antitumoral agents.[7] Cationic manganese–porphyrin complexes are among the efficient DNA cleavers because of their affinity for nucleic acids and their capacity to oxidize the sugar carbon–hydrogen bonds of deoxyribose units.[8] Mechanistic studies on DNA breaks mediated by cationic manganese–porphyrins strongly suggest that high-valent manganese–oxo species might be responsible for DNA cleavage instead of diffusible oxygen species like hydroxyl radicals.[8,9] Such metal–oxo species were also proposed for "activated bleomycin", an antitumoral antibiotic able to cleave DNA.[1a,c,10] DNA cleavage selectivity can be modulated by linking the tris(methylpyridiniumyl)porphyrinatomanganese motif to different vectors: an intercalating agent like 9-methoxyellipticine[11] or an oligonucleotide.[12] This DNA cleaver exhibits also an anti-HIV activity alone or linked to 9-methoxyellipticine.[13] For these different reasons, we decided to undergo the preparation of a series of water-soluble, cationic porphyrin ligands based on the tris(methylpyridiniumyl)porphyrin motif (Table I). All these unsymmetric porphyrins contain a *meso*-phenyl substituent with a tether terminating with an amine, alcohol, or acid function to allow the linkage of the corresponding metalloporphyrins to various vectors (intercalators, peptides, or oligonucleotides).

Table I. Structures of Porphyrin Derivatives 1 to 14-Mn



porphyrins	R_1[a]	R_2	M
1	4-Py	CO-Et	H_2
2	4-Py	H	H_2
3-H_2	4-Py	$(CH_2)_4COOEt$	H_2
3-Ni	4-Py	$(CH_2)_4COOEt$	Ni^{II}
3-Mn	4-Py	$(CH_2)_4COOEt$	Mn^{III}
4-H_2	4-MePy$^+$, I^-	$(CH_2)_4COOEt$	H_2
4-Ni	4-MePy$^+$, I^-	$(CH_2)_4COOEt$	Ni^{II}
4-Mn	4-MePy$^+$, I^-	$(CH_2)_4COOEt$	Mn^{III}
5-H_2	4-Py	$(CH_2)_4COOH$	H_2
5-Mn	4-Py	$(CH_2)_4COOH$	Mn^{III}
6-H_2	4-MePy$^+$, I^-	$(CH_2)_4COOH$	H_4^{2+}, 2 Cl^-
6-Ni	4-MePy$^+$, I^-	$(CH_2)_4COOH$	Ni^{II}
6-Mn	4-MePy$^+$, I^-	$(CH_2)_4COOH$	Mn^{III}
7-H_2	4-Py	$(CH_2)_3OCPh_3$	H_2
7-Ni	4-Py	$(CH_2)_3OCPh_3$	Ni^{II}
7-Mn	4-Py	$(CH_2)_3OCPh_3$	Mn^{III}
8-Ni	4-MePy$^+$, I^-	$(CH_2)_3OCPh_3$	Ni^{II}
8-Mn	4-MePy$^+$, I^-	$(CH_2)_3OCPh_3$	Mn^{III}
9-H_2	4-Py	$(CH_2)_3OH$	H_2
10-Ni	4-MePy$^+$, AcO^-	$(CH_2)_3OH$	Ni^{II}
10-Mn	4-MePy$^+$, AcO^-	$(CH_2)_3OH$	Mn^{III}
11-H_2	4-Py	$(CH_2)_3NHCPh_3$	H_2
11-Ni	4-Py	$(CH_2)_3NHCPh_3$	Ni^{II}
11-Mn	4-Py	$(CH_2)_3NHCPh_3$	Mn^{III}
12-Ni	4-MePy$^+$, I^-	$(CH_2)_3NHCPh_3$	Ni^{II}
12-Mn	4-MePy$^+$, I^-	$(CH_2)_3NHCPh_3$	Mn^{III}
13-H_2	4-Py	$(CH_2)_3NH_2$	H_2
14-Ni	4-MePy$^+$, AcO^-	$(CH_2)_3NH_2$	Ni^{II}
14-Mn	4-MePy$^+$, AcO^-	$(CH_2)_3NH_2$	Mn^{III}

[a] Key:



Results and Discussion

Preparation of the Unsymmetric Trispyridylporphyrin Precursor 1. The synthesis of the unsymmetric porphyrin 1 having three pyridyl groups and a phenyl substituent with a *p*-hydroxyl function protected by a propionyl moiety was carried out in propionic acid in the presence of acetic anhydride according to a slightly modified version of the classical Adler–Longo method.[14] The presence of a small quantity of acetic anhydride allowed the in situ esterification of the *p*-phenol function by propionic acid, facilitating then the recovery of 1 from the complex mixture of the different porphyrin isomers. Porphyrins with free phenol functions do not migrate as

* Author to whom correspondence should be addressed.

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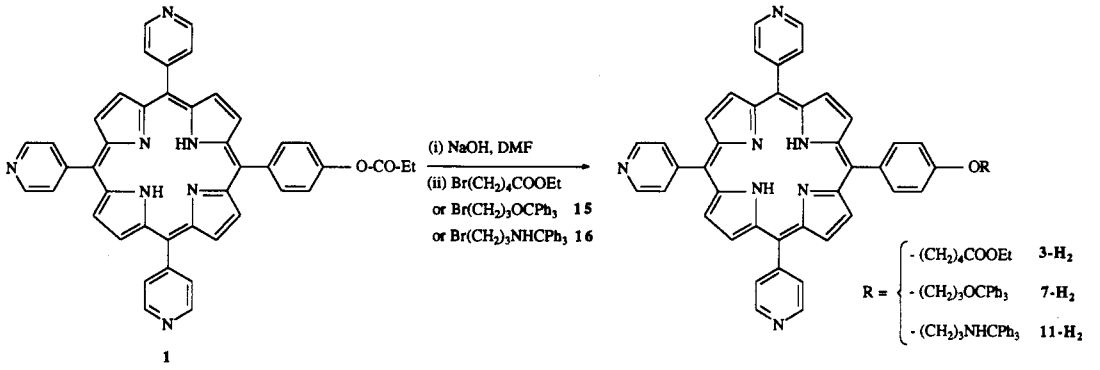

Figure 1. Preparation of porphyrins 3-, 5- and 11-H_2 from the unsymmetrical porphyrin precursor 1.

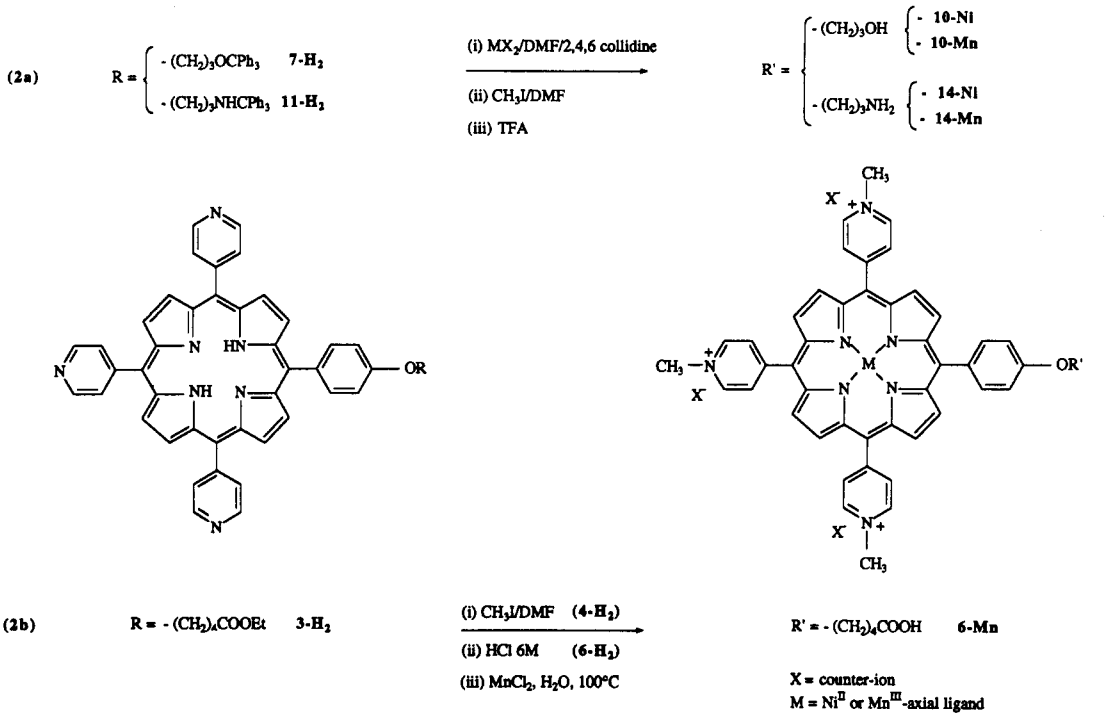

Figure 2. (a) Preparation of manganese(III)- and nickel(II)-porphyrin complexes 10-Ni, 10-Mn, 14-Ni, and 14-Mn. (b) Preparation of the manganese(III)-porphyrin complex 6-Mn with a tether bearing a terminal acid function.

well as the corresponding macrocycle with esterified phenols. The final yield (6–7%) is similar to that observed in other preparations of unsymmetrically substituted tetraarylporphyrins with tolyl groups.[15] The corresponding porphyrin with the free phenol **2** was easily obtained by saponification with a satisfactory yield of 95%. Starting directly from the trispyridylporphyrin **1** several unsymmetrical porphyrins having a protected acid, alcohol, or amine function (3-H_2, 7-H_2, and 11-H_2, respectively) have been prepared by alkylation of the esterified *p*-phenol in strongly basic conditions (Figure 1). Purification of these compounds with protected arms was easily performed by column chromatography on silica and precipitation from a dichloromethane solution by addition of hexane (yields are ranging from 71–90%). In wet DMF, the porphyrin derivative 5-H_2 with a terminal free acid function is the final reaction product. The two other compounds with terminal alcohol and amine functions, 7-H_2 and 11-H_2, were obtained by hydrolysis of the corresponding protected precursors with trifluoroacetic acid.

Metalation and methylation of porphyrin precursors 7-H_2 and 11-H_2 were usually performed on compounds having a protected arm to avoid any possible interactions of the terminal function during the metal insertion step and also to allow a selective methylation of the three pyridine moieties without risk of chemical modification of the terminal function (Figure 2a). We prepared the manganese derivatives of these porphyrin derivatives because of the high nuclease activity of cationic porphyrinatomanganese complexes observed in previous studies.[8,11b] The corresponding nickel porphyrins were prepared (i) to be used as inactive metalloporphyrins for control experiments in DNA cleavage or biological studies at cellular level and (ii) in order to analyze by NMR spectroscopy reaction products subsequent to metalation. Both activities of hybrid molecules containing a metallotris(methylpyridiniumyl)prophyrin motif were found to be metal-dependent and are only observed with metals endowed with redox properties similar to manganese or iron.[11] Yields of metalation reactions ranged from 60–70% after purification by column chromatography.

Metalloporphyrins 3-Ni, 3-Mn, 7-Ni, 7-Mn, 11-Ni, and 11-Mn as well as porphyrin 3-H_2 were methylated in DMF

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with a large excess of methyl iodide for 3 h at room temperature. In these reaction conditions, methylation is quantitative, and products are easily purified by addition of ether to a concentrated methanolic solution of (methylpyridiniumyl)porphyrin derivatives.

For the preparation of the cationic manganese porphyrin with a terminal acid function 6-Mn, the metalation as the final step might be preferred over the methylation procedure (Figure 2b). Such a possibility is illustrated by the metalation of the tris(methylpyridiniumyl)porphyrin 6-H_2. Because of its poor solubility in organic solvent, this cationic porphyrin precursor was metalated by manganese dichloride in water. This latter manganese salt is also preferred to manganese acetate for solubility reasons. Final purification of the water-soluble manganese–porphyrin complex was achieved by addition of THF to an aqueous solution of 6-Mn.

The described preparations allow access to a large variety of different cationic metalloporphyrins tethered to different vectors, not only intercalating agents[11] but also oligonucleotides, polypeptides, and proteins.

Experimental Section

General Methods. Proton NMR spectra were recorded at 200 or 250 MHz in the indicated solvent, and shifts are reported in ppm. Mass spectra (DCI, NH_3) were performed at the Chemistry Department of the University Paul Sabatier (Toulouse).

Chemicals. All reagents or compounds not explicitly referenced were obtained from commercial sources and used as received. DMF was dried overnight on barium oxide and distilled under reduced pressure. CH_2Cl_2 was dried on alumina. TLC was performed using Merck silica gel $60F_{254}$ or neutral alumina $150F_{254}$ precoated plates, 0.2-mm thickness. Column chromatography was carried out using silica gel (70–230 mesh, Merck) or basic alumina (70–230 mesh, Merck).

Synthesis of Tritylated Bromopropanol (15). A mixture of 3-bromopropanol (1 mL, 11 mmol), trityl bromide (2.84 g, 8.8 mmol), triethylamine (1.5 mL, 11 mmol), and 4-(dimethylamino)-pyridine (54 mg, 0.44 mmol) was stirred in dry dichloromethane (20 mL). After 4 h at room temperature, the organic layer was washed three times with water and dried with Na_2SO_4. Evaporation of dichloromethane was carried out to the limit of solubility, and **15** crystallized as a white powder (4.2 g, 95%): R_f 0.63 (SiO_2, CH_2Cl_2–hexane (50/50)); ^1H NMR (CD_2Cl_2) δ 7.43 (m, 6 H, 2,6-trityl), 7.30 (m, 9 H, 3,4,5-trityl), 3.60 (t, 2 H, J = 6.7 Hz, $BrCH_2$), 3.21 (t, 2 H, J = 5.9 Hz, CH_2O), 2.14 (m, 2 H, CH_2); MS m/e 382 ($M^+ + 1$). Anal. Calcd for $C_{22}H_{21}OBr$: C, 69.46; H, 5.57. Found: C, 70.35; H, 5.86.

Synthesis of Tritylated Bromopropylamine (16). To a solution of 3-bromopropylamine hydrobromide (1 g, 4.56 mmol) and trityl bromide (1.47 g, 4.56 mmol) in dry dichloromethane (17 mL) under nitrogen was added triethylamine (1.3 mL, 9.5 mmol), and the mixture was stirred for 5 h at room temperature. The organic layer was then washed three times with water and the product obtained as for **15** (1.05 g, 61%): R_f 0.64 (SiO_2, CH_2Cl_2–hexane (50/50)); ^1H NMR ($CDCl_3$) δ 7.42 (m, 6 H, 2,6-trityl), 7.25 (m, 9 H, 3,4,5-trityl), 3.53 (t, 2 H, J = 6.7 Hz, $BrCH_2$), 2.24 (t, 2 H, J = 6.2 Hz, CH_2N), 1.99 (m, 2 H, CH_2), 1.48 (s, 1 H, NH); MS m/e 380 ($M^+ + 1$). Anal. Calcd for $C_{22}H_{22}NBr$: C, 69.64; H, 5.85; N, 3.69. Found: C, 70.17; H, 5.98; N, 3.57.

Preparation of Porphyrin Precursors 1 and 2. 5-[4-[(Ethylcarbonyl)oxy]phenyl]-10,15,20-tris(4-pyridyl)porphyrin (1). A mixture of 4-hydroxybenzotriazole (3.64 g, 30 mmol), propionic acid (234 mL), and acetic anhydride (12 mL) was heated at 110 °C with stirring. To this solution were successively and slowly added 4-pyridinecarboxaldehyde (8.54 mL, 97 mmol) and pyrrole (8.55 mL, 116 mmol). The resulting mixture was refluxed for 1.5 h. The volume of solvent was reduced to 50 mL under reduced pressure. The mixture was then neutralized with a $KHCO_3$ solution, filtered through a glass frit, and washed several times with water. The crude material was extracted by a mixture of CH_2Cl_2–EtOH (95/5, v/v) and purified by chromatography (alumina layered on silica, ratio = 1/2, v/v; CH_2Cl_2–EtOH (97/3)). Evaporation of solvent afforded 200 mg of **1** as a purple powder (6–7%): R_f 0.5 (SiO_2, CH_2Cl_2–EtOH (97/3)); ^1H NMR ($CDCl_3$) δ 9.04 (d, 6 H, J = 5.3 Hz, 2,6-pyridine), 8.95 (d, 2 H, J = 4.7 Hz, β-pyrrole), 8.84 (s, 4 H, β-pyrrole), 8.79 (d, 2 H, J = 4.8 Hz, β-pyrrole), 8.20 (d, 2 H, J = 8.4 Hz, 2,6-phenyl), 8.15 (d, 6 H, J = 5.5 Hz, 3,5-pyridine), 7.52 (d, 2 H, J = 8.3 Hz, 3,5-phenyl), 2.80 (q, 2 H, J = 7.5 Hz, CH_2), 1.47 (t, 3 H, J = 7.5 Hz, CH_3), −2.59 (s, 2 H, NH pyrrole); UV–vis (4.85 μM in $CHCl_3$) λ_{max} (ϵ, $M^{-1} \times cm^{-1}$) 640 (2×10^3), 584 (6.2×10^3), 544 (6.2×10^3), 510 (1.4×10^4), and 414 nm (38×10^4, Soret band); MS m/e 690 ($M^+ + 1$); IR (KBr pellet) carbonyl at 1757 cm^{-1}.

5-(4-Hydroxyphenyl)-10,15,20-tris(4-pyridyl)porphyrin (2). To a solution of **1** in DMF was added powdered sodium hydroxide (50 equiv), and the mixture was stirred at room temperature for approximately 15 min. The pH of the reaction mixture was then adjusted to 7 by addition of 1 M HCl and the product extracted from the aqueous layer with dichloromethane. The organic layer was washed three times with saturated salt solution and dried over Na_2SO_4. The product was precipitated by addition of hexane to dichloromethane–ethanol (90/10) and dried under vacuum (yield = 80%): R_f 0.34 (SiO_2, CH_2Cl_2–EtOH (95/5)); ^1H NMR (CD_3COOD) δ 9.41 (d, 6 H, J = 5.7 Hz, 2,6-pyridinium), 9.14 (d, 2 H, J = 4.9 Hz, β-pyrrole), 9.09 (s, 4 H, β-pyrrole), 9.01 (d, 2 H, J = 4.9 Hz, β-pyrrole), 8.79 (d, 6 H, J = 5.5 Hz, 3,5-pyridinium), 8.22 (d, 2 H, J = 8.3 Hz, 2,6-phenyl), 7.43 (d, 2 H, J = 8.3 Hz, 3,5-phenyl); UV–vis (4.78 μM in CH_2Cl_2–EtOH (95/5)) λ_{max} (ϵ, $M^{-1} \times cm^{-1}$) 640 (6.2×10^3), 58 (8.4×10^3), 546 (8.4×10^3), 510 (2.1×10^4), and 416 nm (28×10^4, Soret band); MS m/e 634 ($M^+ + 1$).

Preparation of Porphyrins 3-H_2, 7-H_2, and 11-H_2. Compounds 3-H_2, 7-H_2, and 11-H_2 were synthesized according to the same following procedure. To a solution of **1** (100 mg, 0.14 mmol) in dry DMF (6 mL) under nitrogen was added powdered sodium hydroxide (170 mg, 4.25 mmol), and the mixture was stirred for 30 min (the color of the solution changed from purple to green). Formation of the phenolate form of **2** was checked by TLC before addition of the alkyl bromide: ethyl 5-bromovalerate, **15** or **16** (0.23 mmol). The mixture was stirred for 3 h at room temperature and the solution adjusted to pH 6 with 1 M HCl. Water and dichloromethane were added to the solution to extract the desired product. The aqueous layer was washed several times with dichloromethane, until the combinated organic extracts were colorless, dried over sodium sulfate, and evaporated to dryness. Products were purified on silica with a CH_2Cl_2–EtOH (95/5 v/v) eluant and precipitated from a mixture of dichloromethane–hexane. Yields ranged from 71% (3-H_2) to 90% (7-H_2 and 11-H_2).

5-[4-[5-(Ethoxycarbonyl)-1-butyloxy]phenyl]-10,15,20-tris(4-pyridyl)porphyrin (3-H_2): ^1H NMR (CD_2Cl_2) δ 9.02 (d, 6 H, J = 5.6 Hz, 2,6-pyridine), 8.97 (d, 2 H, J = 4.9 Hz, β-pyrrole), 8.88 (s, 4 H, β-pyrrole), 8.85 (d, 2 H, J = 5.0 Hz, β-pyrrole), 8.16 (d, 6 H, J = 5.8 Hz, 3,5-pyridine), 8.10 (d, 2 H, J = 8.6 Hz, 2,6-phenyl), 7.30 (d, 2 H, J = 8.5 Hz, 3,5-phenyl), 4.27 (t, 2 H, J = 5.6 Hz, OCH_2), 4.17 (q, 2 H, J = 7.1 Hz, $COOCH_2$), 2.50 (t, 2 H, J = 6.8 Hz, CH_2COO), 2.00 (m, 4 H, 2 CH_2), 1.29 (t, 3 H, J = 7.1 Hz, CH_3), −2.92 (s, 2 H, NH pyrroles); UV–vis (2.60 μM in CH_2Cl_2) λ_{max} (ϵ, $M^{-1} \times cm^{-1}$) 644 (3.0×10^3), 588 (5.0×10^3), 548 (7.0×10^3), 512 (1.8×10^4), and 417 nm (36×10^4, Soret band); MS m/e 762 ($M^+ + 1$). Anal. Calcd for $C_{48}H_{39}H_7O_3$: C, 75.67; H, 5.16; N, 12.87. Found: C, 74.91; H, 5.32; N, 12.49.

5-[4-[[3-(Trityloxy)-1-propyl]oxy]phenyl]-10,15,20-tris(4-pyridyl)prophyrin (7-H_2): R_f 0.63 (SiO_2, CH_2Cl_2–ethanol (95/5)); ^1H NMR (CD_2Cl_2) δ 9.04 (d, 6 H, J = 5.8 Hz, 2,6-pyridine), 9.01 (d, 2 H, β-pyrrole), 8.91 (s, 4 H, β-pyrrole), 8.87 (d, 2 H, J = 4.9 Hz, β-pyrrole), 8.20 (d, 6 H, J = 5.7 Hz, 3,5-pyridine), 8.13 (d, 2 H, J = 8.6 Hz, 2,6-phenyl), 7.56 (m, 6 H, 2,6-trityl), 7.35 (m, 9 H, 3,4,5-trityl + 2 H, 3,5-phenyl), 4.46 (t, 2 H, J = 6.2 Hz, OCH_2), 3.45 (t, 2 H, J = 6.0 Hz, CH_2O), 2.28 (m, 2 H, CH_2), −2.88 (s, 2 H, NH pyrrole); UV–vis (1.61 μM in CH_2Cl_2) λ_{max} (ϵ, $M^{-1} \times cm^{-1}$) 650 (3.1×10^3), 590 (6.2×10^3), 550 (7.7×10^3), 515 (1.5×10^4), and 420 nm (27×10^4, Soret band); MS m/e 934 ($M^+ + 1$).

Anal. Calcd for $C_{63}H_{47}N_7O_2$: C, 81.00; H, 5.07; N, 10.50. Found: C, 81.14; H, 5.82; N, 9.84.

5-[4-[[3-(Tritylamino)-1-propyl]oxy]phenyl]-10,15,20-tris-(4-pyridyl)porphyrin (11-H_2): ^{1}H NMR (CD_2Cl_2) δ 9.02 (d, 6 H, J = 5.8 Hz, 2,6-pyridine), 8.99 (d, 2 H, β-pyrrole), 8.89 (s, 4 H, β-pyrrole), 8.87 (d, 2 H, β-pyrrole), 8.17 (d, 6 H, J = 5.9 Hz, 3,5-pyridine), 8.11 (d, 2 H, 2,6-phenyl), 7.57 (m, 6 H, 2,6-trityl), 7.34 (m, 9 H, 3,4,5-trityl + 2H, 3,5-phenyl), 4.44 (t, 2 H, OCH_2), 2.49 (t, 2 H, CH_2N), 2.15 (m, 2 H, CH_2), -2.90 (s, 2 H, NH pyrrole); UV-vis (2.45 μM in CH_2Cl_2) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 646 (4.0×10^3), 590 (6.0×10^3), 550 (8.0×10^3), 514 (2.2×10^4), and 420 nm (40×10^4, Soret band); MS m/e 933 ($M^+ + 1$). Anal. Calcd for $C_{63}H_{48}N_8O$: C, 81.09; H, 5.18; N, 12.01. Found: C, 80.25; H, 5.65; H, 12.03.

Hydrolysis of 3-H_2 to 5-H_2. The procedure is similar to that employed for the preparation of 3-H_2. But in this case, use of reagent-grade DMF allowed the direct formation of the porphyrin ligand with the free terminal acid function. The pH of the reaction mixture was then adjusted to 4 with 1 M HCl and the product extracted from the aqueous layer with dichloromethane. The organic layer was washed three times with water and dried with Na_2SO_4. The product was precipitated by addition of hexane to a dichloromethane solution (yield = 92%): ^{1}H NMR (CD_3COOD) δ 9.36 (d, 6 H, J = 6.3 Hz, 2,6-pyridinium), 9.15 (d, 2 H, J = 4.9 Hz, β-pyrrole), 9.08 (s, 4 H, β-pyrrole), 9.01 (d, 2 H, J = 49 Hz, β-pyrrole), 8.70 (d, 6 H, J = 6.3 Hz, 3,5-pyridinium), 8.25 (d, 2 H, J = 8.6 Hz, 2,6-phenyl), 7.46 (d, 2 H, J = 8.6 Hz, 3,5-phenyl), 4.39 (t, 2 H, OCH_2), 2.67 (t, 2 H, CH_2COO); UV-vis (3.6 μM in CH_2Cl_2–EtOH (80/20)) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 644 (3.8×10^3), 586 (5.5×10^3), 543 (6.3×10^3), 516 (1.8×10^4), and 420 nm (37×10^4, Soret band); MS m/e 734 ($M^+ + 1$).

Hydrolysis of 7-H_2 to 9-H_2. To a solution of 10% trifluoroacetic acid in dichloromethane (10 mL) was added 33 mg (35 μmol) of 7-H_2 with stirring at room temperature. The reaction was monitored by TLC as usual (SiO_2, dichloromethane–ethanol (95/5)) until deprotection was complete (1 h). In these conditions, the R_f value of the starting material is 0.63 and that of 9-H_2 is 0.26. The pH of the reaction mixture was then adjusted to 7 with 10 M sodium hydroxide and the product extracted with dichloromethane. The organic layer was washed three times with water and dried with Na_2SO_4. The product was precipitated from the dichloromethane solution by addition of hexane (yield = 60%); ^{1}H NMR (CD_3COOD) δ 9.36 (d, 6 H, J = 5.5 Hz, 2,6-pyridinium), 9.14 (d, 2 H, J = 4.9 Hz, β-pyrrole), 9.08 (s, 4 H, β-pyrrole), 9.01 (d, 2 H, J = 4.9 Hz, β-pyrrole), 8.71 (d, 6 H, J = 5.0 Hz, 3,5-pyridinium), 8.22 (d, 2 H, J = 8.5 Hz, 2,6-phenyl), 7.45 (d, 2 H, J = 8.6 Hz, 3,5-phenyl), 4.47 (t, 2 H, J = 6.0 Hz, OCH_2), 4.11 (q, 2 H, J = 6.2 Hz, CH_2O), 2.32 (m, 2 H, CH_2); UV-vis (16.2 μM in CH_2Cl_2) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 644 (1.2×10^3), 588 (2×10^3), 548 (2.6×10^3), 513 (6.7×10^4), and 417 nm (15×10^4, Soret band). Anal. Calcd for $C_{44}H_{33}N_7O_2 \cdot 2H_2O$: C, 72.61; H, 5.12; N, 13.47. Found: C, 72.60; H, 5.13; N, 13.48.

Hydrolysis of 11-H_2 to 13-H_2. The deprotection of 11-H_2 was performed in a mixture of 50/50 trifluoroacetic acid/water for 1 h at room temperature. The pH of the reaction mixture was then adjusted to 7 with citric acid and the product extracted with dichloromethane. The organic layer was washed three times with water and dried with Na_2SO_4. The product was precipitated by addition of ether to a methanol solution of the product (yield = 60%): ^{1}H NMR (CD_3COOD) δ 9.37 (d, 6 H, J = 5.9 Hz, 2,6-pyridinium), 9.14 (d, 2 H, J = 4.9 Hz, β-pyrrole), 9.08 (s, 4 H, β-pyrrole), 9.02 (d, 2 H, J = 4.6 Hz, β-pyrrole), 8.71 (d, 6 H, J = 5.7 Hz, 3,5-pyridinium), 8.26 (d, 2 H, J = 8.0 Hz, 2,6-phenyl), 7.48 (d, 2 H, J = 8.6 Hz, 3,5-phenyl), 4.51 (t, 2 H, OCH_2), 3.56 (t, 2 H, CH_2N), 2.48 (m, 2 H, CH_2); UV-vis (5.5 μM in CH_2Cl_2–EtOH (98/2)) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 646 (3.2×10^3), 590 (5.5×10^3), 550 (6.8×10^3), 515 (1.7×10^4), and 420 nm (38×10^4, Soret band).

General Procedure for Metalation of Porphyrins with a Protected Terminal Function. A solution of porphyrin 3-H_2 (50 mg, 0.066 mmol) and 2,4,6-collidine (87 μL, 0.66 mmol) in DMF was stirred at 140 °C for 15 min. Then $Ni^{II}Cl_2 \cdot 6H_2O$ (157 mg, 0.66 mmol) was added and the mixture stirred for 3 h. A UV-visible spectrum after this time showed the reduction of the Q-band number from four to two. After the mixture was cooled, 200 mL of dichloromethane–ethanol (95/5) solution was added. The organic layer was washed several times with water (until it was colorless), dried with Na_2SO_4, and evaporated. The product was dried under vacuum at 140 °C to remove all collidine (yield for 3-Ni = 70%). 7-Ni and 11-Ni were prepared according to the same procedure.

Metalation by manganese was done with $Mn^{II}(OAc)_2 \cdot 4H_2O$ (10 equiv with respect to the porphyrin) in a similar fashion. In this case, at the end of the reaction, the DMF was evaporated and the residue dissolved in CH_2Cl_2–EtOH (90/10). The solution was filtered and then purified by column chromatography on alumina gel (CH_2Cl_2–EtOH (90/10)). The solvent was removed under reduced pressure and the final product precipitated in a mixture (CH_2Cl_2–EtOH–hexane, yield = 60%). Porphyrins 3-Mn, 7-Mn, and 11-Mn were prepared according this procedure.

3-Ni: UV-vis (8.4 μM in DMF) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 558 (8.2×10^3) and 416 nm (12×10^4, Soret band). 7-Ni: UV-vis (2.6 μM in DMF) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 526 (1.1×10^3) and 414 nm (18×10^4, Soret band). 11-Ni: UV-vis (6.1 μM in DMF) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 528 (8.1×10^3) and 416 nm (10×10^4, Soret band).

3-Mn: UV-vis (5.3 μM in CH_2Cl_2–EtOH (90/10)) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 600 (5.7×10^3), 566 (8.6×10^3), and 470 nm (7.6×10^4, Soret band). 7-Mn: UV-vis (5.3 μM in CH_2Cl_2–EtOH (90/10)) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 600 (5.7×10^3), 566 (8.6×10^3), and 470 nm (7.6×10^4, Soret band). 11-Mn: UV-vis (6.1 μM in CH_2Cl_2–EtOH (90/10)) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 564 (9.6×10^3) and 468 nm (9.7×10^4, Soret band).

General Procedure for Methylation of Porphyrins. Metalloporphyrins 3-Ni, 3-Mn, 7-Ni, 7-Mn, 11-Ni, and 11-Mn and porphyrin 3-H_2 were methylated in DMF with a large excess of methyl iodide (120 equiv) for 3 h at room temperature. The reaction was monitored by TLC with a mixture of CH_2Cl_2–EtOH (95/5) as eluant (the final product does not migrate on TLC). The solvent and the excess of methyl iodide were removed under vacuum, at room temperature for methyl iodide and at 140 °C for DMF. The residue was taken up in methanol and precipitated with diethyl ether. This reaction is quantitative. Cationic metalloporphyrins 4-H_2, 4-Ni, 4-Mn, 8-Ni, 8-Mn, 12-Ni, and 12-Mn were obtained according to this procedure.

4-H_2: ^{1}H NMR (DMSO) δ 9.59 (d, 6 H, J = 6.6 Hz, 2,6-pyridinium), 9.26 (s, 4 H, β-pyrrole), 9.17 (s, 4 H, β-pyrrole), 9.12 (d, 6 H, J = 6.5 Hz, 3,5-pyridinium), 8.25 (d, 2 H, J = 8.5 Hz, 2,6-phenyl), 7.55 (d, 2 H, J = 8.6 Hz, 3,5-phenyl), 4.83 (s, 9 H, N^+-Me), 4.42 (t, 2 H, J = 5.3 Hz, OCH_2), 4.24 (q, 2 H, J = 7.1 Hz, OCH_2), 2.00 (m, 4 H, 2 CH_2), 1.36 (t, 3 H, J = 7.1 Hz, CH_3), -2.88 (s, 2 H, NH pyrroles).

4-Ni: ^{1}H NMR (DMSO) δ 10.61 (ls, 4 H, β-pyrrole), 10.52 (ls, 4 H, β-pyrrole), 9.49 (d, 6 H, J = 5.6 Hz, 2,6-pyridinium), 8.94 (d, 6 H, J = 5.3 Hz, 3,5-pyridinium), 8.05 (d, 2 H, J = 8.2 Hz, 2,6-phenyl), 7.44 (d, 2 H, J = 7.9 Hz, 3,5-phenyl), 4.75 (s, 9 H, N^+-Me), 4.33 (t, 2 H, OCH_2), 4.18 (q, 2 H, J = 6.9 Hz, $COOCH_2$), 1.95 (m, 4 H, 2 CH_2), 1.29 (t, 3 H, J = 6.7 Hz, CH_3).

8-Ni: ^{1}H NMR (DMSO) δ 10.65 (m, 8 H, β-pyrrole), 9.55 (d, 6 H, J = 5.8 Hz, 2,6-pyridinium), 8.11 (d, 2 H, 2,6-phenyl), 7.60 (m, 2 H, 3,5-phenyl + 15 H, trityl), 4.80 (s, 9 H, N^+-Me), 4.55 (t, 2 H, OCH_2), 3.51 (t, 2 H, CH_2O), 2.30 (m, 2 H, CH_2), 1.95 (m, 4 H, 2 CH_2), 1.29 (t, 3 H, J = 6.7 Hz, CH_3).

Synthesis of 6-Mn (Figure 2). The cationic porphyrin 4-H_2 (110 mg, 93 μmol) was dissolved in 6 M HCl (16.5 mL) and stirred at room temperature for 3.5 h. The solution was filtered and the solvent removed under reduced pressure. The product was redissolved in water, the solution filtered, and the solvent evaporated. The product was dried under vacuum at 80 °C (yield = 93%). 6-H_2: ^{1}H NMR (DMSO) δ 9.61 (d, 6 H, J = 6.3 Hz, 2,6-pyridinium), 9.26 (s, 4 H, β-pyrroles), 9.17 (s, 4 H, β-pyrroles), 9.12 (d, 6 H, J = 6.7 Hz, 3,5-pyridinium), 8.26 (d, 2 H, J = 8.6 Hz, 2,6-phenyl), 7.56 (d, 2 H, J = 8.6 Hz, 3,5-phenyl), 4.84 (s, 9 H, N^+-Me), 4.41 (m, 2 H, OCH_2), 2.02 (m, 4 H, 2 CH_2), -2.87 (s, 2 H, NH pyrroles); UV-vis (8.3 μM in MeOH) λ_{max} (ε, $M^{-1} \times cm^{-1}$) 518 (15.2×10^3) and 428 nm (20×10^4, Soret band). Anal. Calcd for $C_{49}H_{44}I_3N_7O_3 \cdot 3H_2O$: C, 45.74; H, 4.07; N, 7.62. Found: C, 45.57; H, 4.24; N, 7.99.

This cationic porphyrin 6-H_2 with a free terminal acid function (30 mg, 24 μmol) was dissolved in 2.8 mL of $MnCl_2 \cdot 4H_2O$ solution (1 mg/60 μL of water). The mixture was stirred at 100 °C for 4 h. After cooling, DMF was added (9.8 mL) and the product precipitated by addition of THF. The product was redissolved

in water and the precipitation procedure repeated. 6-Mn was obtained with a 73% yield: UV–vis (30 μM in H_2O) λ_{max} (ϵ, $M^{-1} \times cm^{-1}$) 466 nm (9.8×10^4, Soret band).

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