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SYNTHESIS OF 8-DEMETHYL-8-FORMYL PROTOPORPHYRIN IX AND OF 8-DEMETHYL PROTOPORPHYRIN IX

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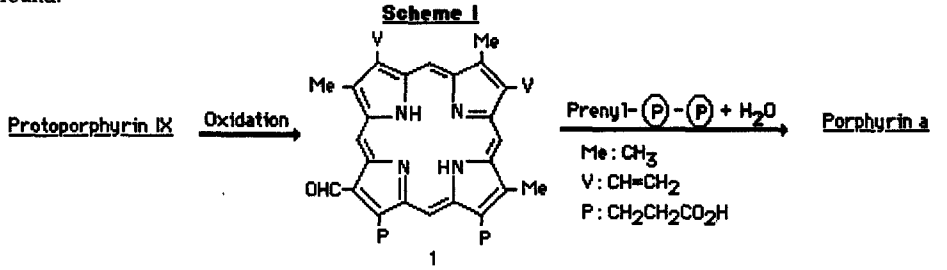
ABSTRACT: A 8-demethyl-8-formyl-2,4-bis(β-chloroethyl) porphyrin and its 8-demethyl derivative were obtained from the oxidative cyclization of 2,4-bis (β-chloroethyl)-6-(β-ethoxycarbonylethyl)-7-(β-methoxycarbonylethyl)-1',1,3,5,8-pentamethyl a,c-biladiene dihydrobromide with copper (II) chloride in dimethyl formamide (DMF) in the presence of iodine and air. Under these reaction conditions the cyclization took place at 25°C and the formylporphyrin was obtained in 25% yield together with the C-8 unsubstituted porphyrin which was obtained in 50% yield. The latter could also be obtained in 65% yield by the oxidative cyclization with copper (II) chloride in DMF at 25°C of the β-unsubstituted 1,7-bis (β-chloroethyl)-4-(β-methoxycarbonylethyl)-5-(β-ethoxycarbonylethyl)-1',2,6,8,8'-pentamethyl-a,c-biladiene dihydrobromide. The formylation attemps at the C-8 unsubstituted position of this porphyrin were however unsuccesful, when either N,N-diisobutyl formamide (Vilsmeier-Haak reaction) or dichloromethyl methyl ether (Friedel Crafts reaction) were used. The title porphyrins were obtained from the aforementioned 2,4-bis(β-chloroethyl) porphyrins by vinylation of the latter with base. 8-Demethyl-8-formyl protoporphyrin IX is a valuable intermediate to probe into the biosynthesis of heme a.

Heme a (iron-porphyrin a) is the prosthetic group of cytochrome oxidase, the terminal enzyme of the respiratory chain (Warburg's atmungsferment). Removal of iron from heme a affords porphyrin a, a porphyrin where the 2-vinyl residue of the ubiquitous protoporphyrin IX [1] is replaced by an isoprene side chain and where its 8-methyl residue has been oxidized to a formyl group. Very little is known about the biosynthesis of porphyrin a. It is conceivable that it is originated in protoporphyrin IX either by a prior oxidation to 8-demethyl-8-formylprotoporphyrin IX **1** followed by alkylation of the 2-vinyl residue by an isoprene pyrophosphate (Scheme I), or the other way around.

Scheme I

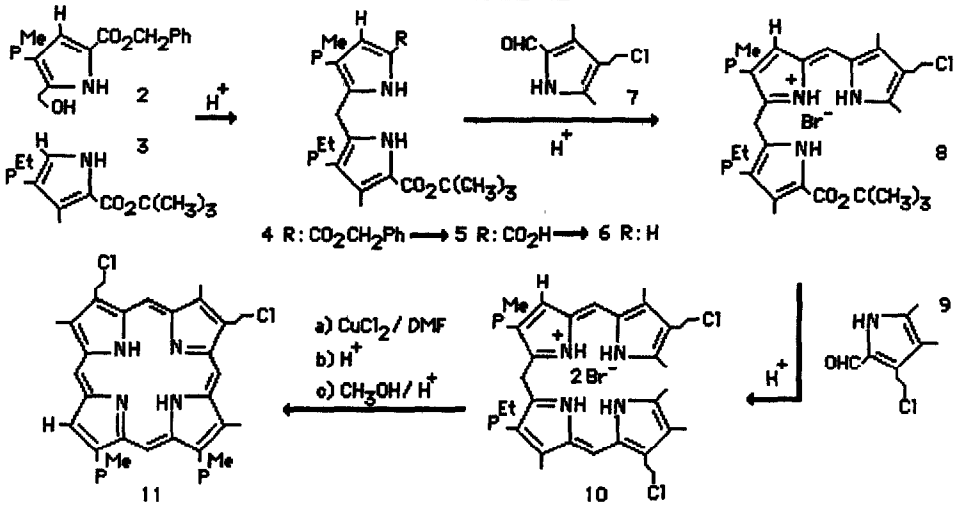


1

Recent findings showed that radical type oxidations of heme b, the prosthetic group of widely distributed hemoproteins such as horseradish and cytochrome c peroxidases, result in the formation of iron-porphyrin **1** due to the oxidation of the exposed C-8 edge in the protein pocket.[2] This fact could explain the unusual presence of a formyl residue at C-8 in heme a and could also be the starting step of its biosynthesis. An efficient synthesis of **1** will therefore be of help to probe into the biosynthetic pathway of heme a by making use of specifically labeled precursors. Porphyrin **1** was obtained by Clezy and Fookes [3] through the synthesis of a porphyrin precursor substituted with an 8-acetyl residue which was then degraded to give the 8-formyl group. The same approach was used for the synthesis of porphyrin a.[4] We looked for a synthetic procedure where the formyl group could be introduced in a one-step reaction.

Of the many porphyrin syntheses which can be found in the literature,[5] it is increasingly clear that symmetrically substituted porphyrins can best be prepared by condensation of two dipyrrylmethane halves,[6] while asymmetrically substituted porphyrins can be obtained by the cyclization of 1',8'-dimethyl-a,c-biladiene salts in the presence of oxidizing copper (II) salts.[7] The latter procedure was also used for the cyclization of b-bilene salts.[3,4] It was decided to prepare an 8-unsubstituted porphyrin **11** conveniently substituted at C-2 and C-4 with 2-chloroethy side-chains as precursors of the vinyl residues (Scheme II). Since regioselective formylations of β-unsubstituted porphyrins have been reported,[8,9] the synthesis of **11** as a precursor of **1** seemed fully justified.

Scheme II

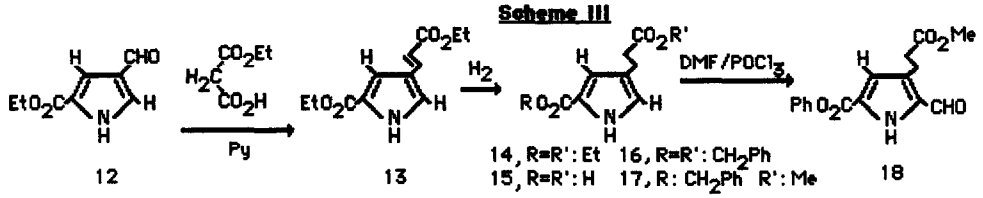


The direct synthesis of **11** was approached through β-unsubstituted a,c-biladiene in view of the good results reported for the synthesis of asymmetric porphyrins starting with a,c-biladienes[7].The former was first constructed in such a way as to avoid that the 1',8'-dimethyl groups should be flanked by the β-unsubstituted carbon, since it has been reported that when this is the case the cyclization with copper salts in hot dimethyl formamide leads to the formation of secondary porphyrins.[7b] By condensation of the 2-hydroxymethylpyrrole **2** with the α-unsubstituted pyrrole **3**, the β-unsubstituted dipyrrylmethane **4** was obtained. Hydrogenolysis over Pd/C then afforded the acid **5**. The attempted condensation of **5** with the formyl pyrrole **7**[7c] in the presence of p-toluensulfonic acid failed since **5** was not decarboxylated under these reaction conditions. Decarboxylation of **5** was achieved by heating in vacuo, and the resulting dipyrrylmethane **6** was condensed with **7** in the presence of hydrobromic acid to give the tripyrrene hydrobromide **8**. This β-unsubstituted tripyrrene **8** was unstable and could not be stored but had to be converted as soon as possible into the biladiene. By treatment of **8** with trifluoroacetic acid (to cleave and decarboxylate the t-butyloxycarbonyl residue) in the presence of the 2-formyl pyrrole **9** and hydrobromic acid, it was posible to obtain the a,c-biladiene dihydrobromide **10**. It has been reported[10] that β-unsubstituted biladienes are cleaved to by-products when the cyclizations are attempted in hot (150°C) dimethyl formamide in the presence of the copper salts (the usual cyclization conditions[7]). Fortunately, the oxidative cyclization of **10** with copper chloride in dimethyl formamide could be achieved at 20°C. It was followed by the demetalation of the copper chelate of the porphyrin, and by a transesterification step with methanol. It was thus posible to obtain the dimethyl ester of

porphyrin **11** in 65% yield from **10**.

The synthesis of the new pyrroles employed in this synthetic sequence are summarized in Scheme III. The 4-formylpyrrole **12** was condensed with ethyl malonate to give the acrylate **13** which was reduced to the propionate **14**. Saponification gave the diacid **15**, which by treatment with benzyl chloride in dimethyl formamide [44] gave the dibenzyl ester **16**.The latter was transesterified with methanol in acid medium to give **17**. A Vilsmeier-Haack reaction on **17** gave **18**, which was reduced with sodium borohydride in methanol-methylene chloride to the hydroxymethyl pyrrole **2**. The formyl pyrrole **9** was obtained from 2-benzyloxycarbonyl-3-(β-chloroethyl)-4,5-dimethylpyrrole[45] by prior hydrogenolysis to the acid followed by treatment of the latter with triethyl orthoformate in acid medium.

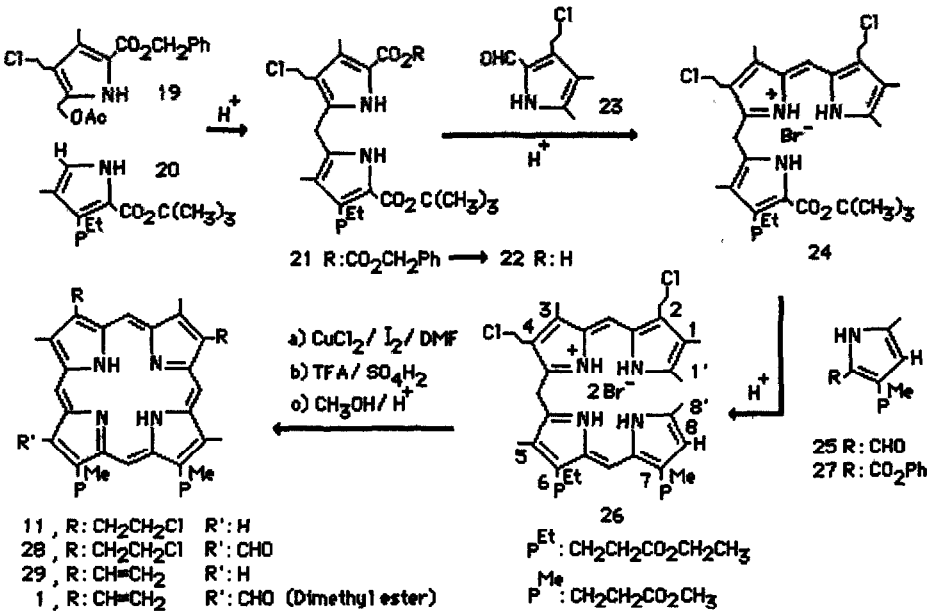
Scheme III



All attempts to formylate the C-8 position of **11** were unsuccesful. The hindered N,N-diisobutyl formamide was used in a Vilsmeier-Haak reaction to avoid substitution at the meso-carbons [8], but the yields of the obtained 8-formyl porphyrin were very low. The use of a Friedel-Crafts reaction with dichloromethyl methyl ether[9] on the iron-porphyrin also failed to give the desired porphyrin. It was therefore decided to attempt the cyclization of an a,c-biladiene β-unsubstituted at a position flanking the 1',8'-dimethyl residues, and to take advantage of the observation[10] that if an oxidative cyclization with copper salts is carried out in the presence of iodine, a formyl residue is introduced at the unsubstituted carbon. The formyl group is very likely originated in one of the methyl residues[7b].The sequence leading to the needed a,c-biladiene **26** is summarized in Scheme IV. Condensation of the 2-acetoxymethyl pyrrole **19** with the α-unsubstituted pyrrole **20** in acetic acid in the presence of p-toluensulfonic acid gave the dipyrrylmethane **21** in 70% yield. Catalytic hydrogenation of **21** over Pd gave the dipyrrylmethane acid **22**. Treatment of **22** with p-toluensulfonic acid in methylene chloride (to achieve decarboxylation), followed by *in situ* condensation with the formyl pyrrol **2 3** afforded the tripyrrene **24**. Treatment of **24** with trifluoroacetic acid to achieve the cleavage of the t-butyloxycarbonyl residue, followed by an *in situ* condensation with the 2-formyl pyrrole **25** in the presence of hydrobromic acid gave the a,c-biladiene dihydrobromide **26** in 80% yield. The formyl pyrrole **25** was prepared from the benzyloxycarbonyl pyrrole **27** [14] as described above.

The a,c-biladiene **26** was cyclized following the procedure described for the synthesis of **11** and gave a mixture of the copper chelates of **11** and **28**. They were easily separated by chromatography. The chelates were then demetalated by acid, and the porphyrins were transesterified with methanol-acid. The C-8 unsustituted porphyrin **11** was formed in 50% yield and the C-8 formyl pophyrin **28** was formed in 25% yield. The overall yield of the reaction can therefore be considered as very satisfactory. Vinylation of the 2-chloroethyl residues of **11** to give **29** was achieved with base, while in **28** the formyl residue had to be protected by an *in situ* acetal formation with ethylene glycol during the basic treatment. The dimethyl ester of **1** was thus obtained in 90 % yield from **28**. This synthetic approach made posible the obtention of regioselective labelled **1** by a relatively simple sequence.

Scheme IV



EXPERIMENTAL SECTION

General procedures: Melting points were determined on a Kofler melting point apparatus and are uncorrected.1H-NMR spectra were routinely recorded on a Varian FT-80 A spectrometer, except for the spectra of tripyrrenes, biladienes, and porphyrins which were recorded at 270 MHz on a Bruker WH-270. Mass spectra were obtained with a Varian CH-7 spectrometer. The silica gel used in column chromatography was TLC Kieselgel (Merck). TLC was performed on precoated silica gel F-254 plates (Merck,0.25 mm layer thickness). The substances were spotted by spraying the plates with Ehrlich's reagent (2% p-(dimethylamino) benzaldehyde in 6N HCl), or by treatment with bromine vapor which gave orange or red colors with the dipyrrylmethanes.

Ethyl 2-ethoxycarbonyl-4-pyrroleacrylate **13**:

A solution of 16.7 g (0.1mol) of the 4-formyl pyrrole **12** [11] in 100 mL of dry pyridine to which 2 mL of piperidine and 75 mL of acid ethyl malonate were added, was heated at 90 °C during 6 h and then at 130 °C for additional 3 h. The solution was then poured over 500 mL of ice-water, the precipitate of **13** was filtered and crystallized from ethanol-water: 22.1 g (93%); mp 104 °C; ^{1}H-NMR ($CDCl_3$) δ 7.47(d,1H, CH=CHCO), 7.05 (m, 2H, H-3 and H-5), 6.05 (d, 1H, =CHCO), 4.20 (m, 4H, CH_2CH_3), 1.28(m,6H,CH_3). Anal.Calcd. for $C_{12}H_{15}NO_4$: C,60.76; H,6.33; N, 5.91. Found: C, 60.83; H, 6.39; N,5.99.

Ethyl 2-ethoxycarbonyl-4-pyrrolepropionate **14**:

The acrylate **13** (20 g) was dissolved in 200 mL of ethanol and was reduced with hydrogen over 2 g of 10% *Pd on charcoal at 50 psi during 2 h. The catalyst was filtered off and after evaporation of the solvent* 19.8 g (98%) of **14** were obtained as a colorless oil; mass spectrum, m/e (relative intensity) 239 (M^+,80), 152 (M^+ - $CH_2CH_2CO_2C_2H_5$,100); ^{1}H-NMR ($CDCl_3$) δ 6.77, 6.74(s, s, 1H, 1H, H-5 and H-3), 4.20 (m, 4H, CH_2CH_3), 3.22 (m,4H, CH_2CH_2), 1.30 (m, 6H, CH_3).

2-Carboxy-4- pyrrolepropionic acid **15**.

The diethyl ester **14** (18.9 g) was dissolved in a mixture of 100 mL of ethanol and 100 mL of 10 % sodium hydroxide. The solution was evaporated to dryness in an open vessel by heating at 100 °C. The residue was redissolved in 20 mL of water, the solution was adjusted to pH 2 with hydrochloric acid, the precipitate of **15** was filtered, dried, and crystallized from ethanol-water: 15 g (98%); mp 173-174 °C. Anal.Calcd. for $C_8H_9NO_4$: C, 52.46; H, 4.92; N, 7.65. Found: C, 52.50; H, 4.98; N,7.70.

Benzyl 2-benzyloxycarbonyl-4-pyrrolepropionate 16:

A solution of 15 g of the diacid 15 in a mixture of 500 mL of dimethyl formamide, 200 mL of triethylamine and 200 mL of benzyl chloride were kept at 20 °C during 48 h. The solution was then evaporated in vacuo at 90°C the residue was partitioned between chloroform (200 mL) and water (100 mL), the organic layer was separated, washed again with water (50 mL) and evaporated to dryness. The residue was dissolved in a small volume of benzene containing 0.5 % of methanol, and was chromatographed through a silica column (3x40 cm) packed, prewashed, and eluted with the same solvent. The fractions containing 16 were pooled and evaporated to dryness. The residue of 16 was recrystallized from benzene-heptane: 28.3 g (95%); mp 51-52 °C; ^{1}H-NMR ($CDCl_3$) δ 7.41,7.33 (b, b, 5H, 5H, Ph), 6.78, 6.70(m, m, 1H, 1H, H-5 and H-3), 5.33, 5.14 (s, s, 2H, 2H, $\underline{CH_2}$Ph), 2.73 (m, 4H, $\underline{CH_2CH_2}$). Anal. Calcd. for $C_{22}H_{21}NO_4$: C,72.73; H, 5.78; N, 3.86. Found: C,72.70; H, 5.81; N,3.90.

Methyl 2-benzyloxycarbonyl-4-pyrrolepropionate 17:

A solution of 28 g of 16 in 1 L of dry methanol containing 5% of sulfuric acid was kept during 18 h at 20°C. It was then poured over 2 L of ice-water and 500 mL of chloroform, the organic layer was separated, the aqueous layer was reextracted with chloroform (250 mL), the organic layers were pooled, washed first with a saturated sodium bicarbonate solution and then with water, and finally evaporated to dryness. The residue was dissolved in a small volume of 1% methanol in benzene and chromatographed on a silica gel column as described above using the same solvent.The fractions containing 17 were pooled and evaporated to dryness. The residue was crystallized from benzene-heptane: 17.2g (78%); mp 55-56 °C; ^{1}H-NMR ($CDCl_3$) δ 7.40 (b, 5H, Ph), 6.82(m, 2H, H-5 and H-3), 5.30 (b, 2H, $\underline{CH_2}$Ph), 3.70 (s,3H, OCH_3), 2.70 (m,4H,$\underline{CH_2CH_2}$CO). Anal. Calcd. for $C_{16}H_{17}NO_4$: C, 66.90; H,5.92; N,4.88. Found: C, 66.85; H, 5.90; N,4.97.

Benzyl 2-formyl-3-(β-methoxycarbonylethyl)-5-pyrrolecarboxylate 18:

A mixture of 25 mL of phosphorous oxychloride and 38 mL of dry dimethyl formamide was kept at 20 °C during 1h and was then diluted with 75 mL of 1,2-dichloroethane. A solution of 15 g of 17 in 200 mL of 1,2-dichloroethane was then slowly added to the former mixture with constant stirring. The resulting solution was heated at 85 °C during 4 h; it was then diluted by addition of crushed ice and adjusted to pH 8 with a concentrated sodium hydroxide solution. The mixture was further heated at 70 °C during 1h, cooled, extracted with chloroform (200 mL), the organic layer was separated, washed with water to neutral, and evaporated to dryness. The residue of 18 was crystallized from benzene-heptane: 14.6 g (89 %); mp, 67-68 °C; ^{1}H-NMR ($CDCl_3$) δ 9.75 (s, 1H, CHO), 7.33 (b, 5H, Ph), 6.75(d, 1H,H-4), 5.30 (s,2H, $\underline{CH_2}$Ph), 3.65 (s, 3H, OCH_3), 3.05(t, 2H, $\underline{CH_2}CH_2CO$), 2.60 (t, 2H, $\underline{CH_2}CO$).Anal. Calcd. for $C_{17}H_{17}NO_5$: C, 64.76; H, 5.40; N,4.44. Found: C, 64.72; H, 5.36; N, 4.35.

Benzyl 2-hydroxymethyl-3-(β-methoxycarbonylethyl)-5-pyrrolecarboxylate. 2

Sodium borohydride (10 g) was added to a solution of 10 g of benzyl 2-formyl-3-(β-methoxycarbonyl ethyl)-5-pyrrolecarboxylate in a mixture of 250 mL of dry methanol and 250 mL of dry methylene chloride. The mixture was stirred at 20°C during 20 min, it was then poured over water (500 mL), the solution was adjusted to pH 6 with dilute hydrochloric acid, the organic layer was separated, and the aqueous solution was extracted with chloroform (100 mL). The pooled organic layers were evaporated to dryness, and the residue was dissolved in a small volume of 5% methanol in chloroform and chromatographed on a silica gel column as described above using the same solvent. The fractions containing 2 were pooled, evaporated to dryness, and the residue was crystallized from benzene-hexane: 8 g (86%); mp 63-64°C; ^{1}H-NMR ($CDCl_3$) δ 7.38 (b, 5H, Ph), 6.78 (d, 1H, H-4), 5.31 (s, 2H, $\underline{CH_2}$Ph), 4.90(b, 2H, $\underline{CH_2}OH$), 3.67(s, 3H, OCH_3), 3.28 (b, 1H, OH), 2.68(m, 4H, CH_2CH_2). Anal. Calcd. for $C_{17}H_{19}NO_5$: C, 64.35; H, 5.99; N,4.42. Found: C, 64.30; H, 6.02; N, 4.60.

tert-Butyl-3-[β-(ethoxycarbonylethyl)]-3'-[β-(methoxycarbonylethyl)]-4-methyl-5'-[benzyloxycarbonyl]dipyrrylmethane-5 carboxylate 4.

The dipyrrylmethane was obtained by condensation of 1.58 g (5 mmol) of 2 and 1.40 g (5 mmol) of 3 [64] dissolved in 150 mL of dry methylene chloride in the presence of 250 mg of p-toluenesulfonic acid. The solution was heated under a stream of nitrogen at 40°C for 4 h after which it was poured over 300 mL of water, the organic layer was separated, the aqueous layer was extracted with chloroform (3 x100mL), the organic layers were pooled, washed with an 8% sodium bicarbonate solution, then with water, and finally evaporated to dryness in vacuo. The residue was dissolved in a small volume of 2% methanol in benzene and was chromatographed through a silica gel column which was packed and eluted with the same solvent. The elution fractions which contained 4 were pooled, evaporated to dryness, and the residue crystallized from ethanol-water; 2.2 g (70 %); mp 112-114°C; ^{1} H-NMR ($CDCl_3$) δ 7.35(b, 5H, Ph), 6.75 (d, 1H, H-4'), 5.26 (s,2H, $\underline{CH_2}$Ph), 4.07(q, 2H, $\underline{CH_2}CH_3$), 3.96 (s, 2H, -CH_2), 3.70 (s, 3H, OCH_3), 2.70 (m, 8H, $\underline{CH_2CH_2}$CO), 2.30 (s, 3H, CH_3-4), 1.55(s, 9H, $(CH_3)_3$), 1.35(t, 3H, $CH_2\underline{CH_3}$). Anal. Calcd. for $C_{32}H_{40}N_2O_8$: C, 66.21; H, 6.89; N, 4.83. Found: C, 66.18; H, 6.78; N,5.00.

tert-Butyl-3-(β-(ethoxycarbonylethyl)]-3'-[β-(methoxycarbonylethyl)]-4-methyl-5'-carboxydipyrrylmethane-5-carboxylate 5:

A solution of 4 (2 g) in 200 mL of ethanol was reduced with hydrogen over 400 mg of 10 % Pd on charcoal at 50 psi during 4 h. The catalyst was filtered off, the solution was evaporated to dryness at 40°C in vacuo and the residue was dissolved in a small volume of 5% methanol in chloroform and chromatographed through a silica gel column (3 x40 cm) packed and eluted with the same solvent. The fractions containing 5 were pooled, evaporated to dryness, and the residue was crystallized from methanol-water; 1.57 g (92%); mp 148-149°C; ^{1}H-NMR (DMSO-d_6) δ

11.0 (b,1H, CO_2H), 6.60 (d,1H, H-4'), 4.15 (q, 2H, $\underline{CH_2}CH_3$), 3.94 (s, 2H,-CH_2), 3.75 (s, 3H, OCH_3), 2.65(m, 8H, $\underline{CH_2}\underline{CH_2}$), 2.25 (s, 3H, CH_3-4), 1.75 (s, 9H, $(CH_3)_3$), 1.27 (t, 3H, $CH_2\underline{CH_3}$). Anal.Calcd.for $C_{25}H_{34}N_2O_6$: C,61.22; H,6.94; N, 5.71. Found: C, 61.35; H,7.00; N, 5.98.

tert-Butyl 3-[ß-(ethoxycarbonylethyl)]-3'-[ß-(methoxycarbonyl)]-4-methyl-dipyrrylmethane-5-carboxylate 6.

Dipyrrylmethane acid 5 (1.5 g) was decarboxylated in 250 mg fractions. Each fraction was dissolved in 5 mL of methanol containig 0.1 mL of triethylamine, the solution was evaporated to dryness in a round bottomed flask, the latter was repeatedly evacuated and flushed with nitrogen, and the residue was heated at 185°C/ 0.1 torr during 6 min. The decarboxylated fractions were pooled, dissolved in a small volume of 2% methanol in benzene and chromatographed on a silica-gel column (2 x15 cm) using the same solvent. Dipyrrylmethane 6 was recovered from the eluate as a colorless oil: 990 mg (73%); mass spectrum, m/e (relative intensity) 446 (M^+,43); ^{1}H-NMR ($CDCl_3$) δ 6.60(t, 1H, H-5'), 6.00 (t, 1H, H-4'), 4.12(q,2H, $\underline{CH_2}CH_3$), 3.94 (s, 2H, CH_2), 3.74 (s, 3H, OCH_3), 2.78(m, 8H, $CH_2\underline{CH_2}$), 2.32(s, 3H, CH_3-4), 1.60 (s, 9H, $C(CH_3)_3$), 1.30(t, 3H, $CH_2\underline{CH_3}$).

tert-Butyl-1,5,6'-trimethyl-2-[ß(ethoxycarbonylethyl)]-3-[ß-(methoxycarbonylethyl)]-6-[ß-chloroethyl]tripyrrene-b-1'carboxylate hydrobromide 8.

Dipyrrylmethane 6 (446 mg, 1mmol) and the aldehyde 7 [7c] (186 mg,1mmol) were dissolved in 25 mL of dry methanol, 0.2 mL of 33% hydrobromic acid in acetic acid was added, and the mixture was kept in the dark at 20°C during 1 h under moisture exclusion. The solution was then poured into water, the aqueous solution was adjusted to pH 8 with sodium bicarbonate, and was extracted with chloroform (3 x 25 mL). The chloroform extracts were dried (Na_2SO_4), concentrated in vacuo at 40°C to 0.5 mL, a drop of the hydrobromic-acetic acid solution was added, and the solution was finally evaporated to dryness in vacuo. The residue was dissolved in methylene chloride and the hydrobromide 8 was precipitated by dry ether: 420 mg (60%); mp 142-143°C; ^{1}H-NMR ($CDCl_3$) δ 7.00 (s, 1H, -CH=), 6.82(b,1H, H-4), 4.38(s, 2H, -CH_2-), 4.13 (q, 2H, $\underline{CH_2}CH_3$), 3.68(s, 3H, OCH_3), 3.52(t, 2H, $\underline{CH_2}CH_2Cl$), 2.88-2.67 (m, 6H, $CH_2\underline{CH_2}Cl$, $\underline{CH_2}CH_2CO$), 2.60 (s, 3H, CH_3), 2.48(t, 2H, CH_2CO), 2.32(t, 2H, CH_2CO), 2.23(s, 3H, CH_3), 2.18(s, 3H, CH_3), 1.56 (s, 9H, $C(CH_3)_3$), 1.18(t, 3H, $CH_2\underline{CH_3}$); vis max ($CHCl_3$), 490 nm (ε: 92,000). Anal. Calcd. for $C_{33}H_{43}N_3O_6BrCl$: C,57.02; H,6.48; N,6.05. Found: C,57.18; H,6.60; N,6.10.

1,7-Bis(ß-chloroethyl)-4-(ß-methoxycarbonylethyl)-5-(ß-ethoxycarbonylethyl)-1',2,6,8,8'-pentamethyl-a,c-biladiene dihydrobromide 10:

Tripyrrene hydrobromide 8 (417 mg, 0.6 mmol) was added to 15 mL of trifluoroacetic acid which had been previously degassed by bubbling nitrogen during 20 min. The solution was stirred with nitrogen at 20°C during 10 min (until the tripyrrene dissolved). A solution of formyl pyrrole 9 (111 mg, 0.6mmol) in 15 mL of dry methanol was then added followed by 1 mL of 33% hydrobromic acid in acetic acid, and the mixture was kept at 20°C during 2.5 h. Dry ether was then added to the solution cooled to 0°C, the precipitated a,c-biladiene dihydrobromide was filtered, dried, and crystallized from methanol-ether: 354 mg (70%); mp > 300°C; ^{1}H-NMR ($CDCl_3$) δ 7.11, 7.08(s, s,1H 1H, =CH-), 6.81(s,1H, H-3), 5.20(s, 2H, CH_2), 3.89 (q,2H, $\underline{CH_2}CH_3$), 3.62(t, 2H, $\underline{CH_2}CH_2Cl$), 3.56 (t, 2H, $\underline{CH_2}CH_2Cl$), 3.62 (s, 3H, OCH_3), 3.12 (t, 2H, $CH_2\underline{CH_2}Cl$), 2.90 (t, 2H, $CH_2\underline{CH_2}Cl$), 2.87-2.70 (m, 8H, $\underline{CH_2}CH_2$), 2.66 (s, 6H, 2 CH_3), 2.30 (s, 3H, CH_3), 2.24(s, 3H, CH_3), 2.02 (s, 3H, CH_3),1.08 (t, 3H, $CH_2\underline{CH_3}$); vis max, ($CHCl_3$) 447nm (ε:104,000), 522 mn (ε:103,000). Anal.Calcd. for $C_{37}H_{48}N_4O_4Br_2Cl_2$: C, 52.67; H, 5.69; N, 6.64. Found: C, 52.71; H,5.73; N, 6.70.

2,4-Bis(ß-chloroethyl)-6,7-bis [ß-methoxycarbonylethyll-1,3,5- trimethylporphyrin 11.

Anhydrous copper chloride (300 mg) was added to a solution of 300 mg of a,c-biladiene 10 in 200 mL of dry dimethyl formamide. The mixture was stirred at 20°C during 2 h, chloroform (400 mL) was then added, the solution was washed with water (2 x 200 mL) and evaporated to dryness in vacuo. The residue was dissolved in a small volume of 0.5 % of methanol in chloroform and was filtered through a silica gel column. The red eluates which contained the copper chelate of 11 were pooled and evaporated to dryness. The residue (186 mg, 70%) was dissolved in a cooled mixture of 1.5 mL trifluoroacetic acid and 7.5 mL of concentrated sulfuric acid, and the solution was kept at 20°C during 30 min. It was then poured over water (100 mL) and the porphyrin was extracted with chloroform, the organic layer was washed with a saturated solution of sodium bicarbonate, then with water, dried (Na_2SO_4), and evaporated to dryness in vacuo.The residue was dissolved in 10 mL of anhydrous methanol which contained 5% sulfuric acid , and the solution was kept at 20°C during 15 h. It was then diluted with 50 mL of chloroform, the organic layer was washed with water, then with a saturated sodium bicarbonate solution, again with water, and evaporated to dryness. Porphyrin 11 dimethyl ester was crystallized from chloroform-methanol: 158 mg (93%); mp 171-173°C; ^{1}H-NMR(CD_2Cl_2) δ 10.14, 10.05, 9.99, 9.97 (s, s, s, s, 1H,1H, 1H, 1H, meso-H), 9.09 (s,1H, H-8), 4.52 (t, 4H, $\underline{CH_2}CH_2Cl$), 4.49, 4.37 (t, t, 2H, 2H, $\underline{CH_2}CH_2CO$), 4.34 (t, 4H, $CH_2\underline{CH_2}Cl$), 3.79, 3.66, 3.63, 3.61 (s, s, s, s, 3H, 3H, 3H, 6H, CH_3, OCH_3), 3.48, 3.27 (t, t, 2H, 2H, $CH_2\underline{CH_2}CO$), -4.08 (s, 2H, NH); mass spectrum, m/e (relative intensity) 649 (M^+,43), 648 (M^+-1, 93). Anal. Calcd. for $C_{35}H_{38}N_4O_4Cl_2$: C,64.71; H, 5.85; N, 8.63. Found: C, 64.82; H, 5.90; N, 8.75.

tert-Butyl-3,4'-dimethyl-4[ß-(ethoxycarbonyl)ethyll-3'-[ß-chloroethyl]-5'-[(benzyloxy)carbonyll]dipyrrylmethane-5-carboxylate 21

A solution of 3.5 g (0.01 mol) of the acetate 19 [12] in 30 mL of dry methylene chloride was added dropwise to a stirred solution of 2.8 g (0.01mol) of the α-unsubstituted pyrrole 20 [13] and 160 mg of p-toluensulfonic acid in 30 mL of dry methylene chloride. The solution was then heated at 40°C during 1h. It was then poured into 100 mL of

water, the organic layer was separated, the aqueous layer was extracted with methylene chloride (3 x 50 mL), the pooled organic solvents were washed with a saturated sodium bicarbonate solution, then with water, dried (Na_2SO_4), and evaporated to dryness in vacuo. The dipyrrylmethane **21** was purified by filtration through a TLC silica gel column (4 x 30 cm) using 2% methanol in benzene as eluant. The fractions containing the dipyrrylmethane **21** (monitored by TLC) were pooled and evaporated to dryness. The oily residue (4g, 70%) had the following: mass spectrum, m/e (relative intensity) 571 (M^+,39), 570 (37), 515 (M^+ -isobutylene, 57), 470 (514- CO_2, 100);^{1}H-NMR ($CDCl_3$) δ 9.23, 9.28 (s, s, 1H,1H, NH), 7.15 (b, 5H, Ph), 5.14 (s, 2H, CH_2Ph), 4.01(q,2H, CH_2CH_3), 3.68(s, 2H, pyrr-CH_2-pyrr), 3.22 (t, 2H, CH_2CH_2Cl), 2.96-2.62 (m, 4H, CH_2CH_2CO, CH_2Cl), 2.37 (t, 2H, CH_2CO), 2.17, 1.90 (s, s, 3H,3H,CH_3), 1.43 (s, 9H, $C(CH_3)_3$), 1.15 (t, 3H, CH_2CH_3)

tert-Butyl 1-[ß(ethoxycarbonyl)ethyll-2,4,6,6'-tetramethyl-3,5-bis[ß-chloroethyll tripyrrene-b-1'-carboxylate hydrobromide **24**

Dipyrrylmethane **21** (1.14 g, 2mmol) dissolved in 150 mL of ethanol was reduced with hydrogen over 0.25 g of 10 % Pd on charcoal during 2 h at 50 psi. The catalyst was filtered, the solvent evaporated to dryness in vacuo, and the residue was dissolved in ethanol and precipitated by addition of water. The acid **22** (895 mg, 93%, 1.86 mmol) thus obtained was dissolved in 40 mL of dry methylene chloride, formyl pyrrole **23** [7a] (353 mg, 1.90 mmol) and 900 mg of p-toluensulfonic acid hydrate dissolved in 10 mL of methanol were added, and the solution was kept during 1h at 20°C while flushed with nitrogen. The solution was then poured over 100 mL of water, the organic layer was washed with a saturated sodium bicarbonate solution, then again with water, dried (Na_2SO_4), and evaporated to dryness in vacuo at 25°C.The residue was dissolved in dry methylene chloride and hydrogen bromide was passed through the solution during 10 sec while the latter was cooled in ice-water.The hydrobromide was precipitated by addition of dry ether, filtered off, and washed with dry ether to give 1.02 g (80%): mp 132-133°C; mass spectrum m/e (relative intensity) 605(M^+- Br,42), 603 (87), 568 (605-HCl, 100); ^{1}H-NMR ($CDCl_3$) δ 13.57, 13.58 (b, b, 1H, 1H, NH), 10.25 (b, 1H, NH), 7.08 (s,1H,=CH-), 4.35(s, 2H, -CH_2-), 4.08 (q, 2H, OCH_2CH_3) ,3.62, 3.31 (t, t, 2H, 2H, CH_2CH_2Cl), 3.11 (t, 2H, CH_2CH_2CO), 2.95 (t, 2H, CH_2CH_2Cl), 2.85 (t, 2H, CH_2CH_2Cl), 2.46 (t, 2H, CH_2CH_2CO), 2.67,2.26,2.06,2.03 (s, s, s, s ,3H each, CH_3), 1.57 (s, 9H, $C(CH_3)_3$), 1.21(t, 3H, CH_2CH_3); vis max ($CHCl_3$) 495 nm (ε: 79,200). Anal.Calcd. for $C_{32}H_{44}N_3O_4BrCl_2$: C, 56.06; H,6.42; N, 6.13. Found: C, 56.00; H, 6.38; N,6.16.

2,4-Bis(ß-chloroethyl)-6(ß-ethoxycarbonylethyl)-7(ß-methoxycarbonylethyl)-1',1,3,5,8'-pentamethyl a,c-biladiene dihydrobromide **26:**

The tripyrrene **24** (685 mg, 1 mmol) was hydrolyzed and decarboxylated with trifluoroacetic acid as described for the synthesis of **10** and then condensed with 195 mg (1 mmol) of **25** following the described procedure. The disappearence of the vis absortion of **24** (494 nm) and its replacement by the vis max of **26** (446 nm and 521 nm) was complete in 1h, after which the dihydrobromide **26** was precipitated by addition of dry ether: 672 mg (80%); mp > 300°C; vis ($CHCl_3$) max: 446 nm (ε:104,000), 521(ε:103,000); ^{1}H-NMR (CD_2Cl_2) δ 7.50, 7.13 (s, s, 1H, 1H, =CH-), 6.22 (s,1H, H-8), 5.21(s, 2H, CH_2), 4.04 (q, 2H, OCH_2CH_3), 3.64 (s, 3H, OCH_3), 3.62 (t, 2H, CH_2CH_2Cl), 3.16-2.91(m, 10 H, CH_2CH_2CO, CH_2CH_2Cl), 2.66,2.48 (t, t, 2H, 2H, CH_2CH_2Cl), 2.70, 2.28, 2.03, 1.98 (s, s, s, s, 3H, 3H, 3H, 6H, CH_3), 1.16 (t, 3H, OCH_2CH_3). Anal. Calcd. for $C_{37}H_{48}N_4O_4Br_2Cl_2$: C, 52.67; H,5.69; N,6.64. Found: C,52.63;H,5.65; N, 6.70.

2,4-Bis(ß-chloroethyl)-6,7-bis[ß-methoxycarbonylethyll-8-formyl-1,3,5-trimethylporphyrin **28.**

The a,c-biladiene **26** (600 mg) was cyclized using the procedure described for the synthesis of **11** with 2.8 g of dry copper chloride and 5.8 g of iodine in 260 mL of dimethyl formamide. The copper chelates of **11** and of **28** were separated by the chromatography step on silica gel using chloroform as the elution solvent. The red chelate of **11** was eluted first, while the green chelate **28** followed. The chelates were recovered by pooling and evaporating the eluted fractions, and each chelate was dissolved in a mixture of 6 mL of trifluoroacetic acid and 30 mL of sulfuric acid. The work-up followed the procedure described for the obtention of **11** dimethyl ester: 236 mg (50%) of **11** were obtained, mp 171-173°C; as well as 123 mg (25%) of **28**; mp 237-239°C; ^{1}H-NMR (CD_2Cl_2) δ 11.48 (s, 1H, CHO), 10.81(s,1H, H-d), 10.25, 9.92, 9.89 (s, s,. s, 1H, 1H, 1H, meso-H), 4.63 (t, 2H, CH_2CH_2CO), 4.52 (t, 4H, CH_2CH_2Cl), 4.38 (t, 2H, CH_2CH_2CO), 4.35(t, 4H, CH_2Cl), 3.67, 3.65, 3.61, 3.56 (s, s, s, s, 3H, 3H, 3H, 6H, CH_3, OCH_3), 3.35, 3.27 (t, t, 2H, 2H, CH_2CO), -3.84 (s, 2H, NH); mass spectrum, m/e (relative intensity); 677 (M^+, 37), 676 (M^+-1,100). Anal. Calcd. for $C_{36}H_{38}N_4O_5Cl_2$: C, 63.81; H,5.61, N,8.27.Found: C, 63.75; H, 5.68; N,8.30.

6,7-Bis [ß-methoxycarbonylethyll-1,3,5-trimethyl- 2,4-divinylporphyrin **29.**

The bis(ß-chloroethyl) porphyrin **11** (200 mg) was dissolved in a mixture of 120 mL of pyridine, 20 mL of water, and 25 mL of a 3% sodium bicarbonate solution. The mixture was heated to reflux during 2 h under a strem of nitrogen. It was then cooled, 25 mL of a 25% acetic acid solution was added, and the solution was concentrated in vacuo to a small volume. The precipitate of the diacid porphyrin was filtered, washed with water, dried, dissolved in 300 mL of 5% sulfuric acid in anhydrous methanol and the solution was kept at 20°C during 18 h. It was then diluted with an equal volume of chloroform, washed with water, then with a saturated sodium bicarbonate solution, again with water, and it was finally evaporated to dryness in vacuo. The residue of **29** was crystallized from chloroform-methanol: 174 mg (90%); mp 210-211°C; ^{1}H-NMR ($CDCl_3$) δ 10.06, 10.05, 10.00, 9.97 (s, s, s, s, 1H, 1H, 1H, 1H, meso-H), 9.03 (s, 1H, H-8), 8.30-8.18 (m, 2H, CH=CH_2), 6.43-6.15 (m, 4H, CH=CH_2), 4.49,4.46 (t, t, 2H, 2H, CH_2CH_2CO), 3.81, 3.68, 3.66, 3.61, 3.58 (s, s, s, s, s, 3H, 3H, 3H, 3H, 3H, CH_3, OCH_3), 3.47, 3.25 (t, t, 2H, 2H, CH_2CO), -4.17 (s, 2H, NH); mass spectrum, m/e (relative intensity) 576 (M^+,80). Anal. Calcd. for $C_{35}H_{36}N_4O_4$: C,

72.92; H, 6.25; N, 9.72. Found: C, 72.98; H,6.20; N, 9.79.

6,7-Bis(β-methoxycarbonylethyl)-8-formyl-1,3,5-trimethyl-2,4-divinylporphyrin (8-demethyl-8-formyl protoporphyrin IX dimethyl ester) 1:

Formyl bis (β-chloroethyl) porphyrin **28** (200 mg) was dissolved in 140 mL of dry benzene, 4 mL of ethylene glycol and 25 mg of p-toluenesulfonic acid were added, and the mixture was heated under reflux during 1 h. Cloroform (200 mL) was added to the cooled solution, the organic mixture was washed with water, dried (Na_2SO_4), and evaporated in vacuo. The residue was dissolved in 100 mL of pyridine, and the solution was flushed with nitrogen during 15 min while heated to reflux. It was then cooled , 50 mL of a 1.5% sodium hydroxide solution was added, and the mixture was heated under reflux during 2 h. It was then cooled, acidified with 25 mL of 25 % acetic acid, reduced to a small volume in vacuo, the porphyrin precipitate was filtered, dried, and dissolved in 300 mL of 5% sulfuric acid in methanol. After 18 h at 20°C, the solution was diluted with an equal volume of chloroform, the organic solution was washed with water, then with a saturated sodium bicarbonate solution, again with water, dried (Na_2SO_4) evaporated to dryness in vacuo, and the residue of **1** was crystallized from methylene chloride-methanol:157mg (90%); mp 223-225°C (lit.[3] 228-229°C); ^{1}H-NMR (CD_2Cl_2) δ 11.26 (s,1H, CHO), 10.14 (s, 1H, H-d), 9.64, 9.52, 9.46 (s, s, s, 1H, 1H, 1H, meso-H), 8.18-7.93 (m, 2H, $\underline{CH}=CH_2$), 6.35-6.14 (m, 4H, $CH=\underline{CH_2}$), 4.42, 4.25 (t, t, 2H, 2H, $\underline{CH_2}CH_2CO$); 3.60, 3.58, 3.52, 3.49, 3.40 (s, s, s, s, s, 3H, 3H, 3H, 3H, 3H, OCH_3, CH_3), 3.21-3.10 (m, 4H, CH_2CO), -5.27 (s, 2H, NH); uv-vis max ($CHCl_3$): 648 nm (ε: 2,100), 587 (ε:12,800), 565 (ε: 19,500), 522 (ε: 9,800), 421(ε:166,000). Anal. Calcd. for $C_{36}H_{36}N_4O_5$: C,71.52; H, 5.96; N, 9.27.Found: C, 71.50; H, 6.00; N, 9.31.

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