Synthetic and Biosynthetic Studies of Porphyrins. Part 8.[1] Syntheses of Hepta-, Hexa-, and Penta-carboxylic Porphyrins Related to Uroporphyrin-I

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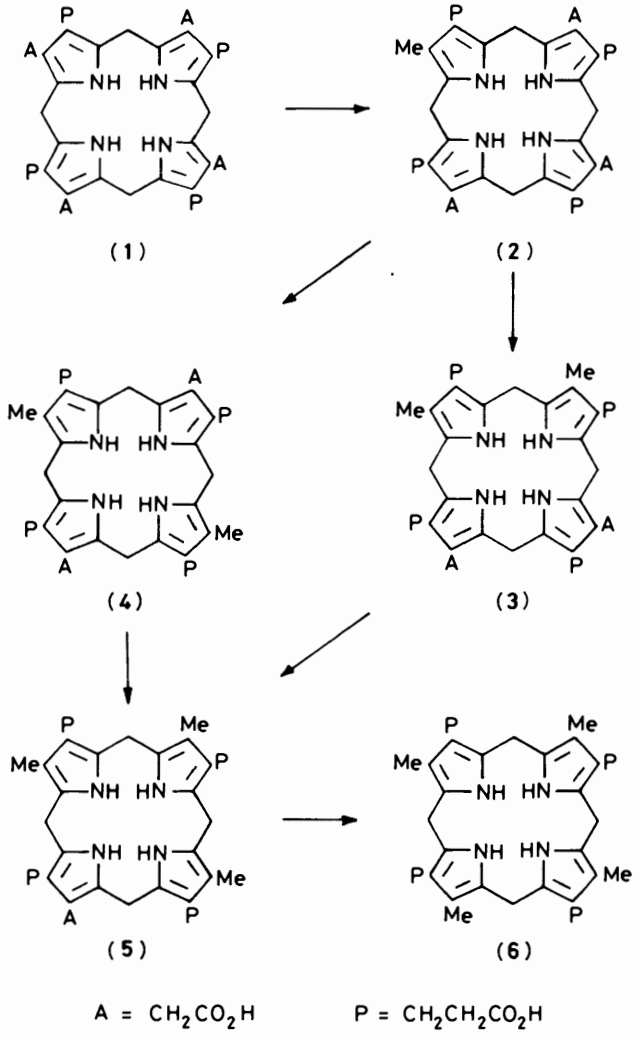
The title porphyrins, of interest as abnormal metabolites in porphyrin biosynthesis, have been synthesized by the Fischer, and *b*-oxobilane routes, and compared with the naturally derived materials. Enzymic experiments have shown that the conversion of uroporphyrinogen-I into coproporphyrinogen-I both *in vivo* and *in vitro* is non-specific and occurs by both possible pathways *via* the two intermediate hexacarboxylic porphyrinogens.

The biochemical conversion of uroporphyrinogen-III into coproporphyrinogen-III involves stepwise decarboxylation of the four acetic acid side-chains in a preferential clockwise manner starting with the D-ring acetic acid and then successively the A, B, and C ring acetic acid residues.[2] Small amounts of other isomers have also been observed both in normal and abnormal metabolism, but the enzyme uroporphyrinogendecarboxylase is sufficiently non-specific to decarboxylate all these as well. Indeed this enzyme can also degrade uroporphyrinogens-II, -III, and -IV to the corresponding coproporphyrinogens[3] but, whereas, the type -I isomer and its decarboxylation products are found in abnormal metabolism (probably because of a defect in the uroporphyrinogen cosynthetase enzyme), the type -II and -IV isomers do not occur in nature.[4] For these reasons we thought that it would be interesting to study the enzymic conversion of uroporphyrinogen-I into coproporphyrinogen-I in detail in order to discover whether or not the process involved both possible routes shown in Scheme 1 or only one of them. We have, therefore, synthesized[5] the heptacarboxylic porphyrin methyl ester, the two hexacarboxylic porphyrin methyl esters, and the pentacarboxylic porphyrin methyl ester related to the type -I porphyrinogens shown in Scheme 1.

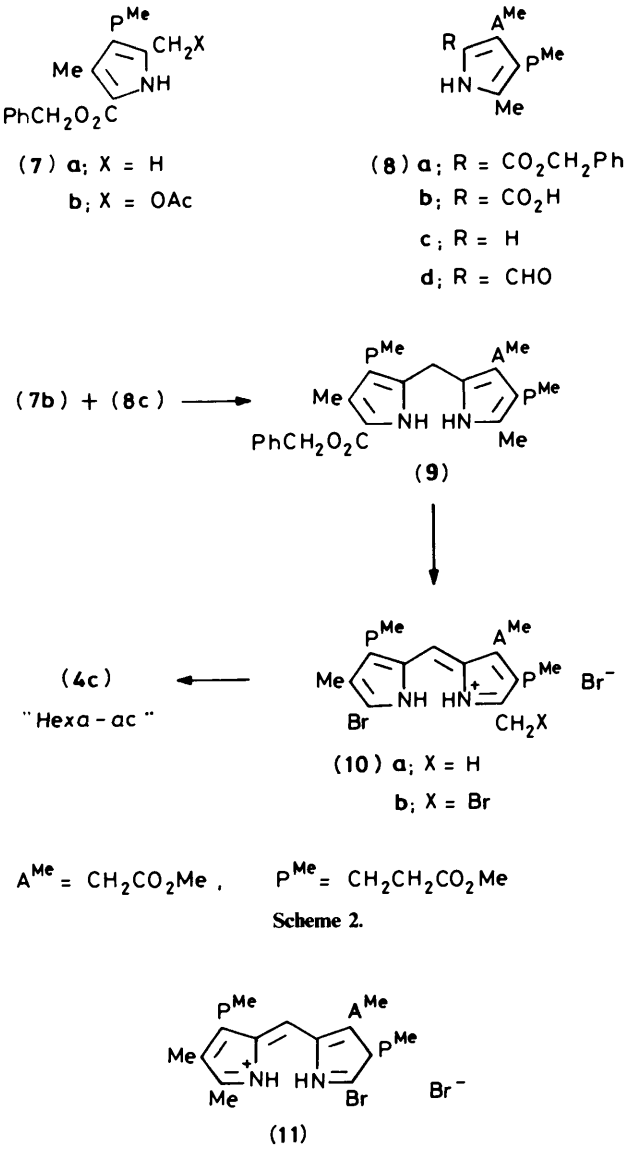


Scheme 1. (a) Porphyrinogens, (b) corresponding porphyrin acids, and (c) porphyrin methyl esters

The centrosymmetrical hexacarboxylic porphyrin was the simplest to synthesize, and this was accomplished by the classical Fischer method[6] as shown in Scheme 2. The pyrromethane (**9**) was first synthesized by coupling the acetoxymethyl pyrrole (**7b**) [prepared by lead tetra-acetate acetoxylation of the corresponding methyl pyrrole (**7a**)] with the α-free pyrrole (**8c**) in boiling methanol containing a catalytic amount of toluene-*p*-sulphonic acid. [The α-free pyrrole was prepared by hydrogenolysis of the benzyl ester (**8a**) followed by decarboxylation of the intermediate acid (**8b**).] Hydrogenolysis of the pyrromethane (**9**) over palladium–charcoal then afforded the corresponding pyrromethanecarboxylic acid, which was immediately treated with bromine in hot acetic acid to afford the required pyrromethene (**10a**); if reaction was prolonged, and an excess bromine was used further bromination occurred and a mixture of the pyrromethene (**10a**) and the bromethyl analogue (**10b**) was formed (as shown by n.m.r. spectroscopy). The pyrromethene (**10a**) [or the mixture with (**10b**)] when heated in molten methylsuccinic acid gave the desired porphyrin hexamethyl ester (**4c**) in 11% yield after work-up and chromatographic purification.

This particular method of synthesizing the porphyrin was chosen rather than *via* the alternative pyrromethene (**11**), because it seemed that steric effects in the self-condensation of pyrromethenes (**10a**), or (**10b**), would be lower than with the pyrromethene (**11**) in which substitution would be required α to a propionate side-chain rather than to a methyl group.

The other three porphyrins were all synthesized by the *b*-oxobilane route.[7] In this method there are, in general, four different positions for siting the oxo function, and since each pyrrole used to synthesize the intermediate pyrromethane can be constructed, with or without the functionalised methylene carbon or amide residue which will form the *meso*-bridge carbon atoms, there are eight possible ways of synthesizing the porphyrin. We chose, in fact, to place the oxo group at the potential δ-position of the pentacarboxylic porphyrin (**5c**),



Scheme 2.

firstly because the required intermediate pyrroles for the synthesis of the two pyrromethanes (**12b**) and (**13a**) were readily accessible, and secondly because the unsubstituted α-position in pyrromethane (**12b**) was adjacent to a methyl, rather than an acetate or propionate, group and would, therefore, be expected to offer least steric hindrance during formation[8] of the imine salt intermediate (**14a**) en route to the desired *b*-oxobilane (**15a**). The pyrromethane (**12b**) could, moreover, also be utilised as a common intermediate for the synthesis of the hexacarboxylic porphyrin (**3c**) and the heptacarboxylic porphyrin (**2c**) as shown in Scheme 3.

The pyrromethane (**12a**) was first synthesized from the pyrrole intermediates (**18a**) and (**19a**), which had been prepared by established methods. Pyrrole (**18a**) was converted into the α-free pyrrole (**18d**) by a standard series of transformations[9] *via* the acid (**18b**) and the iodopyrrole (**18c**). Activation of the pyrrole (**19a**) by lead tetra-acetate afforded the acetoxymethylpyrrole (**19b**), which was coupled with the α-free pyrrole (**18d**) in acetic acid containing toluene-*p*-sulphonic acid to give the pyrromethane (**12a**) in excellent yield (80%).

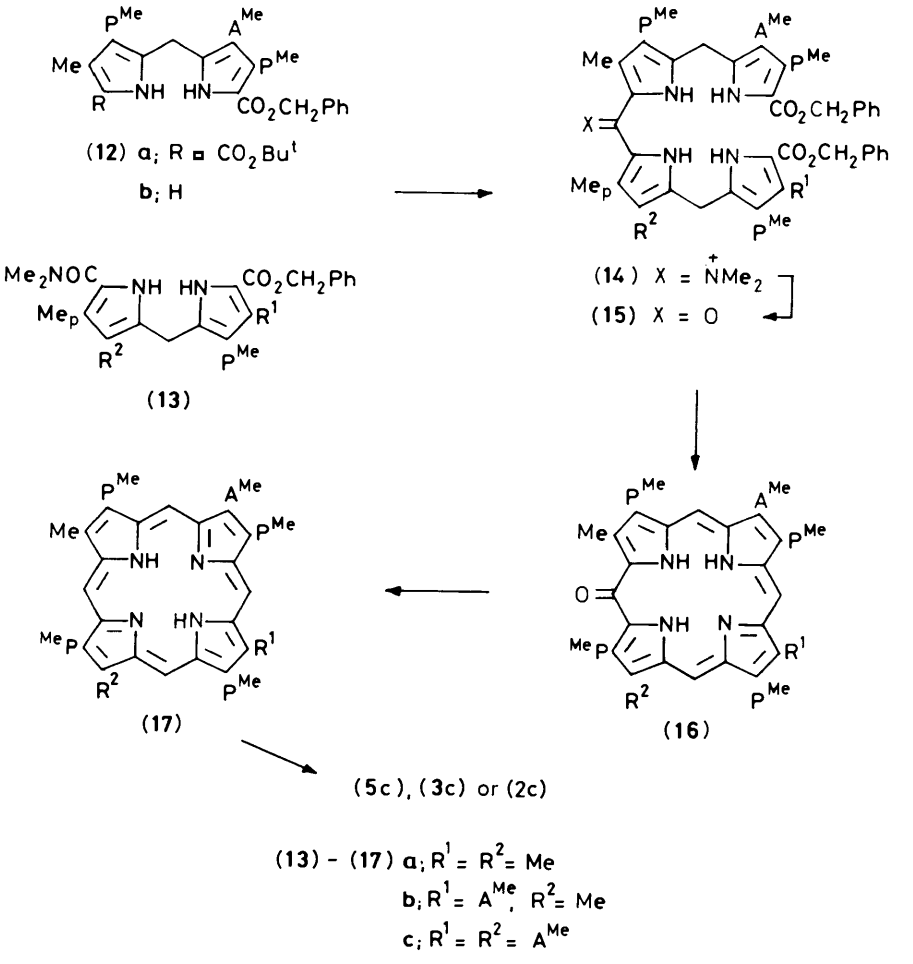
For the synthesis of the pyrromethane amide (**13a**) the two pyrroles (**7b**) and (**20c**) were required. The former was readily available from the pyrrole (**7a**) by treatment with lead tetra-acetate but a somewhat lengthier synthesis was required for the latter. The pyrrole (**7a**) was trichlorinated with sulphuryl chloride and hydrolysed to the acid (**20a**); treatment of the acid chloride (**20b**) with dimethylamine gas afforded the amide (**20c**). The latter was hydrogenolysed to the acid (**21a**), iodinatively decarboxylated to the iodopyrrole (**21b**) and catalytically hydrogenated over platinum to the α-free pyrrole (**21c**). The two pyrroles (**7b**) and (**21c**) were coupled in acetic acid containing toluene-*p*-sulphonic acid at 95 °C to give the desired pyrromethane amide (**13a**); a small amount of the symmetrical pyrromethane (**22**) was also formed by self-condensation of the acetoxymethylpyrrole (**7b**), and separated by chromatography.

The pyrromethane amide (**13a**) was then activated as its phosphoryl chloride complex, and coupled with the α-free pyrromethane (**12b**) in methylene chloride. The resulting imine salt (**14a**) was purified by chromatography on alumina, less polar by-products being eluted first. Hydrolysis of the imine salt with boiling aqueous sodium carbonate in a two-phase system with benzene afforded the desired *b*-oxobilane dibenzyl ester (**15a**) in 30% overall yield from the pyrromethanes. Hydrogenolysis of the benzyl groups followed by treatment with trimethyl orthoformate–trichloroacetic acid in methylene chloride then effected cyclisation to the oxophlorin (**16a**). The latter was purified chromatographically, and was converted directly into the *meso*-acetoxyporphyrin (**17a**) (30%) with acetic anhydride and pyridine. Hydrogenation of the latter over palladium–charcoal, followed by careful re-oxidation of the intermediate *meso*-unsubstituted porphyrinogen then led to the desired pentacarboxylic porphyrin (**5c**).

The hexacarboxylic porphyrin (**3c**) and the heptacarboxylic porphyrin (**2c**) were subsequently synthesized in an exactly analogous manner (see Scheme 3). The pyrromethane (**13b**) was prepared from the acetoxymethyl pyrrole (**23b**) and the α-free pyrrole (**21c**), and the pyrromethane (**13c**) from the same acetoxymethylpyrrole (**23b**) and the α-free pyrrole amide (**25c**). The latter was synthesized from the pyrrole (**23a**) by an analogous method to that used for pyrrole (**21c**) *via* pyrroles (**24a**), (**24b**), (**24c**), (**25a**), and (**25b**).

During the course of synthesizing the various pyrroles required for this work we also briefly investigated variations of the methods which have become established in recent years. One alternative was to adapt a route developed by Plieninger.[10] Thus the diketone (**26a**), prepared from acetylacetone and ethyl bromoacetate in 60% yield, was converted into the magnesium complex and treated with 3-ethoxycarbonylpropionyl chloride in dry ether. The intermediate (**26b**) was not isolated but deacetylated directly with aqueous ammonia to give the dioxoheptanoate (**27**) (in 35% overall yield from acetylacetone). Knorr condensation with diethyl oxoiminomalonate then afforded the pyrrole (**28c**), but in relatively low yield (16%). When dibenzyl oximinomalonate was used instead of the diethyl ester, an intractable oil was obtained which could not be crystallised, and which did not give the expected n.m.r. spectrum for the pyrrole benzyl ester (**28b**). Further work on this route was therefore abandoned in view of the problems in distilling the oxoheptanoate (**27**) under very low pressure (preferably 0.01 mmHg to avoid decomposition) and the low yields obtained with the diethyl oximinomalonate Knorr reaction. The pyrrole methyl diethyl ester (**28a**) was, however, converted by transesterification with benzyl alcohol followed by methanol[9] into the well known pyrrole α-benzyl ester (**28c**).

We also investigated an approach to the synthesis of the pentacarboxylic porphyrin by the tripyrrene-biladiene route[11] (Scheme 4). Thus the unsymmetrical pyrromethane (**30a**) was prepared in 74% yield by acid-catalysed coupling of the acetoxymethyl pyrrole (**7b**) with the α-free pyrrole (**29**) in methylene chloride–methanol. For the synthesis of the desired tripyrrene (**32**) we required the formyl pyrrole (**31c**) and this was



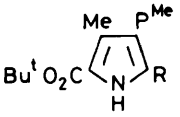
Scheme 3.

synthesized from the benzyl ester (**31a**) by hydrogenolysis over palladium–charcoal, followed by acid-catalysed decarboxylation and formylation of the intermediate carboxylic acid (**31b**) with trimethyl orthoformate and trifluoroacetic acid. The pyrromethane (**30a**) was then hydrogenolysed to the carboxylic acid (**30b**) and treated directly with the formylpyrrole (**31c**) in methylene chloride containing toluene-*p*-sulphonic acid. However, although the visible spectra indicated formation of a pyrromethene-like species no crystalline product could be obtained, even though a variety of conditions were investigated. The n.m.r. spectrum of the crude product was also very ill-resolved and at this stage this approach was abandoned as the alternative *b*-oxobilane route being studied simultaneously had proved more fruitful.

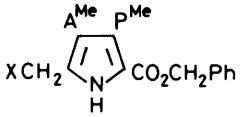
Comparisons with the Natural Porphyrins.—The four synthetic porphyrins were then compared with samples of naturally derived Type-I porphyrins obtained from human patients, and from a porphyric bull, suffering from the rare autosomal recessive disorder congenital erythropoietic porphyria[4] (Gunter's disease). In this condition substantial amounts of type-I porphyrins are excreted, and also found in the blood stream (giving rise to acute photosensitivity, which causes skin lesions on exposure to sunlight). In this type of porphyria the greatly increased production of type-I uro- and coproporphyrinogens in the developing erythroid cells of the bone marrow suggests an imbalance in the co-operative activity of hydroxymethylbilane synthase (EC 4.3.1.8) and uroporphyrinogen-III cosynthetase. This red cell haemolysates from patients with congenital erythropoietic porphyria convert porphobilinogen into a mixture of type-I and type-III isomers, and these studies led to the proposal that the genetic error is a deficiency of the cosynthetase enzyme.[12] However, the haemoglobin concentration in red cells of these patients is normal, despite the increased erythropoiesis, and the large overall increase in total porphyrin production has also been attributed to an increase in the activity of the enzyme ALA synthetase (or of PBG deaminase) leading to an overproduction of PBG and of uroporphyrinogens.[13] However, the currently accepted view[14] is that the main genetic defect is a primary deficiency of uroporphyrinogen-III cosynthetase; the overall increase in porphyrin production can be attributed to feedback control of haem upon ALA synthase activity (to make up for the ALA and PBG diverted to the type-I porphyrinogens).

Three samples of type-I porphyrins from a porphyric bull were sent to us by Dr. T. K. With, (*a*) crude pentacarboxylic-porphyrin as its methyl ester, (*b*) heptacarboxylic porphyrin methyl ester, and (*c*) a mixture of porphyrins from the urine adsorbed on talc. The crude pentacarboxylic porphyrin methyl ester (1.5 mg) had m.p. 222—224 °C, *i.e.* much lower than the pure synthetic porphyrin (**5c**), m.p. 289—290 °C. H.p.l.c. analysis showed that it was contaminated with a substantial amount of coproporphyrin methyl esters as well as traces of other porphyrins, and we were unable to purify it fully by preparative t.l.c. However, the major component had the same retention time on both t.l.c. and h.p.l.c. as the synthetic pentacarboxylic porphyrin (**5c**).

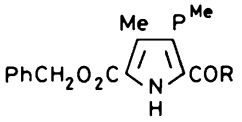
The natural heptacarboxylic porphyrin methyl ester (1.5 mg)



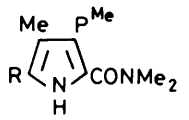
(18) a; R = Me
b; R = CO_2H
c; R = I
d; R = H



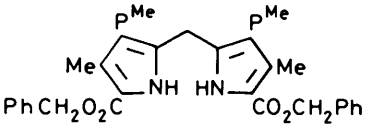
(19) a; X = H
b; X = OAc



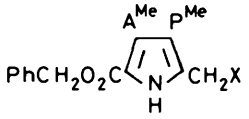
(20) a; R = OH
b; R = Cl
c; R = NMe_2



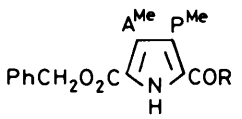
(21) a; R = CO_2H
b; R = I
c; R = H



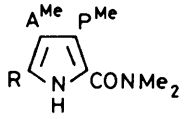
(22)



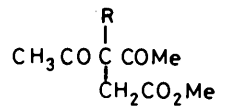
(23) a; X = H
b; X = OAc



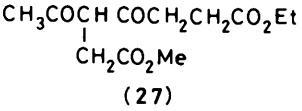
(24) a, R = OH
b, R = Cl
c, R = NMe_2



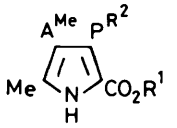
(25) a; R = CO_2H
b; R = I
c; R = H



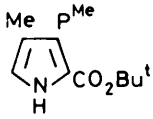
(26) a; R = H
b; R = $COCH_2CH_2CO_2Et$



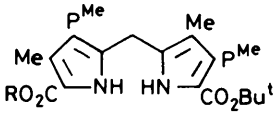
(27)



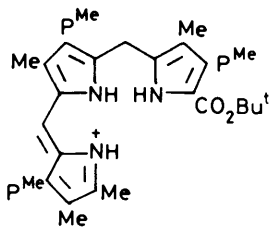
(28) a; $R^1 = R^2 = Et$
b; $R^1 = CH_2Ph$, $R^2 = Et$
c; $R^1 = CH_2Ph$, $R^2 = Me$

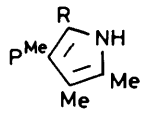


(29)



(30) a; R = CH_2Ph
b; R = H





(31) a; R = CO_2CH_2Ph
b; R = CO_2H
c; R = CHO

(5c)

Scheme 4.

had m.p. 240—252 °C, undepressed by admixture with the synthetic porphyrin (m.p. 254—255 °C). The n.m.r. spectrum was also closely similar to that of the synthetic material.

The mixture of free porphyrins on talc was extracted and converted into the corresponding methyl esters by treatment with methanolic sulphuric acid. After chromatography on alumina, the resulting mixture of porphyrins was shown by t.l.c. and h.p.l.c. to consist mainly of uro- and copro-porphyrin esters. The hexacarboxylic ester fraction only amounted to *ca.* 1—2% of the total porphyrin present, and since it was insufficient for isolation, it was analysed by h.p.l.c. (see below).

Samples of hexacarboxylic porphyrin methyl esters were also obtained from two patients with congenital erythropoietic porphyria; one was kindly provided by Professor G. H. Elder and Dr. D. C. Nicholson, and the other from Barcelona by Dr. S. G. Smith. Both were purified by chromatography, firstly on an open column of alumina and then by h.p.l.c. before more detailed studies of their isomeric composition by h.p.l.c. (see below).

The h.p.l.c. method, developed by our colleague Dr. K. R. N. Rao,[5] afforded separation of both the synthetic hexacarboxylic porphyrin hexamethyl ester isomers (**3c**) and (**4c**) on 5 μm Partisil using chloroform containing 1% acetone (by volume) as eluant. This system also enabled the simultaneous separation of the normal type-III hexacarboxylic porphyrin ester (**33**) from both the type-I isomers. Comparisons with the naturally derived material showed that the hexacarboxylic porphyrin fraction from the porphyric bull was a mixture of all three hexacarboxylic porphyrins (**3c**), (**4c**), and (**33**). Samples from the patient showed mainly the type-I porphyrins (**3c**) and (**4c**), with a small amount of type-III porphyrin (**26**), whereas the Barcelona patient's sample showed that the unsymmetrical hexacarboxylic porphyrin (**3c**) was present in about twice the amount of either the symmetrical type-I porphyrin (**4c**) or the type-III porphyrin (**33**).

In addition to these studies we also carried out incubations of the heptacarboxylic porphyrinogen (**2a**) with chicken red cell haemolysates.[15,16] Within *ca.* 4—5 min the amount of hexacarboxylic porphyrinogen formed had reached a maximum (*ca.* 5—10%), most of it being further transformed to the pentacarboxylic porphyrinogen (**5c**) and to coproporphyrinogen-I (**6c**). The incubation was, therefore, stopped after 5 min and the porphyrinogens allowed to oxidise in air to the corresponding porphyrins before being converted into the methyl esters. The hexacarboxylic porphyrin fraction was then separated and analysis by h.p.l.c. showed that it contained almost equal amounts of the two isomeric type-I porphyrins (**3c**) and (**4c**).

Typical h.p.l.c. traces of the various naturally derived type-I hexacarboxylic porphyrin esters, and comparisons with the synthetic compounds, have previously been published.[5] We conclude, as a result of the studies described in this paper that, the metabolism of uroporphyrinogen-I is nonspecific both *in vivo* and *in vitro* and occurs by both possible pathways (Scheme 1), whereas the pathway from uroporphyrinogen-III to coproporphyrinogen-III is relatively specific[2] (even though there are many more possible alternative routes).

Experimental

M.p.'s were determined on a Kofler block and are uncorrected. Reactions were monitored by t.l.c., h.p.l.c. and by u.v. and visible spectra as appropriate. In a number of instances, especially with pyrromethanes and oxobilanes, the intermediates in porphyrin synthesis were only obtained as gums or foams; however, the purity of all these intermediates was confirmed in each case by t.l.c. or h.p.l.c. and by spectroscopic methods (usually n.m.r.) before proceeding to the next stages in the syntheses. ^{1}H N.m.r. spectra were determined with a Perkin-Elmer R32 90 MHz instrument, and mass spectra with a Varian-CHSD instrument either by F.D. (wire current 10—20 μA) or by E.I. (70 eV at 200—250 °C).

Ethyl 5-Methoxycarbonylmethyl-4,6-dioxoheptanoate (**27**).—Ethanol (11.5 ml) and carbon tetrachloride (1.9 ml) were added to magnesium turnings (1.83 g), and acetyl acetone (28.3 g) was subsequently added at such a rate that the mixture boiled gently. After the addition was complete, ether (75 ml) was added and the mixture was heated under reflux for 6 h to ensure that reaction was complete. Ethoxycarbonylpropionyl chloride (26.3 g) was then added slowly at a rate sufficient to maintain reflux. The solution was finally boiled for a further 3 h and after cooling to 5 °C was poured into ammonium hydroxide solution (10%; 75 ml) and stirred for 1 h. The product was extracted with ether (2 × 50 ml) and the extracts washed with water (50 ml) and dried ($MgSO_4$). After removal of the ether the residual oil was distilled *in vacuo* to yield the *dioxo ester* (**27**) (28.5 g, 35%) as an oil, b.p. 140 °C/0.035 mmHg (Found: C, 55.0; H, 6.6. $C_{12}H_{18}O_8$ requires C, 55.8; H, 6.7%); τ($CDCl_3$) 8.8 (3 H, t, OCH_2CH_3), 7.75 (3 H, s, OCH_3), 7.35 and 7.1 (4 H, m, CH_2CH_2), 7.15 (2 H, d, $CHCH_2$), 6.36 (3 H, s, OCH_3), 6.36 (1 H, t, $CHCH_2$), and 5.8 (2 H, m, CH_2CH_3).

Pyrroles

Ethyl 3-(2-Ethoxycarbonylethyl)-4-methoxycarbonylmethyl-5-methylpyrrole-2-carboxylate (**28a**).—A solution of ethyl 5-methoxycarbonylmethyl-4,6-dioxoheptanoate (3.0 g) in acetic acid was warmed to 70 °C, and treated slowly, and concurrently, with a solution of diethyl oximinomalonate (2.2 g) in acetic acid (3.5 ml) and with a mixture of zinc dust (2.6 g) and anhydrous sodium acetate (2.2 g), at such a rate as to keep the temperature of the well stirred mixture at 70—80 °C. After completion of the addition, the mixture was heated at 90—95 °C for 1 h and then under reflux for 2 h before being poured onto crushed ice (500 ml). After all the ice had melted the crude product was filtered off, dried, and crystallised from methylene chloride–light petroleum (b.p. 60—80 °C) to give the desired *pyrrole* (**28a**) (0.6 g, 16%) as needles, m.p. 89—91 °C. (Found: C, 59.0; H, 4.3. $C_{16}H_{23}NO_6$ requires C, 59.1, H, 7.1; N, 4.3%); τ($CDCl_3$) 8.70 (6 H, m, 2 × OCH_2CH_3), 7.80 (3 H, s, pyr-CH_3), *ca.* 7.5 and 7.0 (each 2 H, m, $-CH_2CH_2-$), 6.60 (2 H, s, CH_2CO_2Et), 6.36 (3 H, s, OCH_3), *ca.* 5.70 (4 H, m, 2 × OCH_2CH_3), and 0.56 (1 H, br, NH).

Benzyl 3-(2-Methoxycarbonylethyl)-4,5-dimethylpyrrole-2-carboxylate (**31a**).—(*a*) Methyl 5-methyl-4, 6-dioxoheptanoate[10] (10 g) in acetic acid (50 ml) was heated to *ca.* 90 °C, and then a solution of dibenzyl oximinomalonate (17 g) in acetic acid (20 ml) and an intimate mixture of zinc dust (12 g) with anhydrous sodium acetate (12 g) were added simultaneously and at such a rate that the well stirred mixture remained at 90—95 °C. The mixture was stirred for a further 1 h at 90—95 °C, and was then poured onto crushed ice (500 ml). After the ice had melted the crude product was filtered off, dissolved in methylene chloride (500 ml), and the solution washed with aqueous sodium hydrogen carbonate (5%; 100 ml) and water (100 ml), and then dried ($MgSO_4$). After removal of solvent the residual oil was taken up in hot, light petroleum (b.p. 60—80 °C) and on cooling it gave the required pyrrole (2.2 g, 13%), m.p. 90—93 °C (lit.,[10] m.p. 95—96 °C) τ($CDCl_3$) 8.10 (3 H, s, pyr-CH_3), 7.80 (3 H, s, pyr-CH_3), 7.5 and 7.0 (each 2 H, m, CH_2CH_2), 6.4 (3 H, s, OCH_3), 4.72 (2 H, s, $CH_2C_6H_5$), 2.63 (5 H, s, C_6H_5), and 0.9 (1 H, br, NH).

(*b*) The pyrrole was also prepared from methyl 5-methyl-4,6-dioxoheptanoate (4.4 g) benzyl oximinoacetoacetate (5.2 g), zinc dust (5 g), and anhydrous sodium acetate (5 g) in acetic acid following a similar procedure to that described in (*a*) above. A mixture of two products was, however, obtained which was separated by chromatography on alumina (Grade III) eluting with increasingly polar mixtures of light petroleum (b.p. 60—80°) and ethyl acetate. The first fraction afforded benzyl 3, 4, 5-trimethylpyrrole-2-carboxylate (1.5 g, 30%), m.p. 113—115 °C from light petroleum (b.p. 60—80 °C) (lit.,[17] m.p. 119—120 °C); τ($CDCl_3$) 8.12, 7.88, and 7.86 (each 3 H, s, pyr-CH_3), 4.72 (2 H, s, $CH_2C_6H_5$), 2.62 (5 H, s, C_6H_5), and 1.2 (1 H, br, NH). The second fraction was the required pyrrole (2.2 g, 30%), m.p. 91—93 °C, identical (mixed m.p. and n.m.r. spectrum) with the product prepared in (*a*). (H.p.l.c. analysis of the crude product on silica using ethyl acetate–cyclohexane (20:80, v:v) confirmed that the two pyrroles were present in almost equal yields).

(*c*) This pyrrole was also prepared from the corresponding ethyl pyrrole-2-carboxylate (**31d**) (see below) by transesterification to the corresponding dibenzyl ester (**31c**) (60%), m.p. 128—129 °C (lit.,[18] m.p. 131—132 °C), followed by partial transesterification with methanolic sodium methoxide. The desired benzyl methoxycarbonylethylpyrrole-2-carboxylate, m.p. 94—95 °C, was obtained in 70% yield, and proved to be identical in all respects with the pyrrole prepared as in (*a*) and (*b*) above.

Ethyl 3-(2-Methoxycarbonylethyl)-4,5-dimethylpyrrole-2-carboxylate (**31d**).—This compound was prepared from ethyl 5-methyl-4,6-dioxoheptanoate and diethyl oximinomalonate in an exactly analogous fashion to the corresponding benzyl ester (**31a**). The required *pyrrole* (**31d**) was obtained in 27% yield, m.p. 79—82 °C from methylene chloride–light petroleum, (b.p. 60—80 °C) (Found: C, 61.5; H, 7.3; N, 5.2. $C_{13}H_{19}NO_4$ requires C, 61.7; H, 7.5; N, 5.5%); τ($CDCl_3$) 8.7 (3 H, t, CH_2CH_2), 8.1 and 7.85 (each 3 H, s, pyr-CH_3), *ca.* 7.5 and 7.0 (each 2 H, m, CH_2CH_2), 6.35 (3 H, s, OCH_3), 5.7 (2 H, q, OCH_2CH_3), and −1.0 (1 H, br, NH).

2-Formyl-3-(2-methoxycarbonylethyl)-4,5-dimethylpyrrole (**31c**).—Benzyl 3-(2-methoxycarbonylethyl)-4,5-dimethylpyrrole-2-carboxylate (**31a**) (6.2 g) in dry methanol (70 ml) was hydrogenated over palladium–charcoal (10%; 0.25 g) at 20 °C and 760 mmHg. After uptake (441 ml) was complete, the catalyst was filtered off through Celite, and the solvent removed under reduced pressure. The residue was crystallised from methylene chloride–light petroleum (b.p. 40–60 °C) to yield the intermediate *pyrrolecarboxylic acid* (**31b**) (4.2 g, 95%), m.p. 130—131 °C. This was treated directly with trifluoroacetic acid (17 ml) and stirred at 25 °C for 30 min before being cooled to

5 °C; trimethyl orthoformate (15 ml) was then added. After 15 min the brownish solution was poured into water (250 ml), and the brown solid which had precipitated was extracted into methylene chloride (2 × 150 ml). The extracts were washed with aqueous sodium hydrogen carbonate (5%; 100 ml) and water (100 ml), dried ($MgSO_4$), and decolourised with a little charcoal. After removal of the solvent under reduced pressure the residue was crystallised from methylene chloride–light petroleum (b.p. 60—80 °C) to afford the *formylpyrrole* (**31c**) (0.6 g, 15%), m.p. 92—92.5 °C (Found: C, 63.1; H, 7.1; N, 6.3. $C_{11}H_{15}NO_3$ requires C, 63.2; H, 7.2; N, 6.7%); τ($CDCl_3$) 8.10 and 7.80 (each 3 H, s, pyr-CH_3), 7.50 and 7.00 (each 2 H, m, CH_2CH_2), 6.40 (3 H, s, OCH_3), 0.52 (1 H, s, CHO), and −0.6 (1 H, br, NH).

t-Butyl 3-(2-*methoxycarbonylethyl*)-4-*methylpyrrole*-2-*carboxylate* (**29**).—The corresponding 5-iodopyrrole[19] (2.0 g), was hydrogenated in ethanol solution over platinum oxide (0.2 g) at 20 °C and 760 mmHg. After uptake was complete, catalyst and solvent were removed, and the residue crystallised from light petroleum (b.p. 60—80 °C) to afford the α-*free pyrrole* (1.28 g, 95%), m.p. 65—66 °C from light petroleum (b.p. 60—80 °C) (Found: C, 62.8; H, 7.8; N, 4.85. $C_{14}H_{21}NO_4$ requires C, 62.9; H, 7.9; N, 5.2%); τ($CDCl_3$) 8.46 [9 H, s, $C(CH_3)_3$], 8.01 (3 H, s, pyr-CH_3), 7.5 and 7.0 (each 2 H, m, CH_2CH_2), 6.35 (3 H, s, OCH_3), 3.40 (1 H, d, pyr-H), and 0.9 (1 H, br, NH).

5-*Benzyloxycarbonyl*-4-*methoxycarbonylmethyl*-3-(2-*methoxycarbonylethyl*)*pyrrole*-2-*carboxylic Acid* (**24a**).—Freshly distilled sulphuryl chloride (6.9 ml) was added at 10 °C over 1 h to the stirred solution of the 2-methylpyrrole (**23a**) (10.2 g) in dry ether (150 ml) and the mixture was kept at 20 °C for 5 days. The reaction was shown to be complete by evaporating a small portion of the mixture (10 ml) to dryness at 20 °C. The n.m.r. spectrum of the residual oil illustrated the disappearance of the 2-methyl protons (τ 7.88). After removal of ether and sulphuryl chloride under reduced pressure, hydrolysis was accomplished by stirring the mixture of the residual oil in dioxane (100 ml) and anhydrous sodium acetate (15 g) in water (80 ml) at 60—65 °C for 3 h. The cooled solution was extracted with ether (2 × 100 ml) and the organic layer was separated, and extracted with an excess of aqueous sodium carbonate (5%). The combined aqueous extracts were acidified slowly with dilute acetic acid at room temperature to yield colourless plates of the required *pyrrolecarboxylic acid* (5.2 g, 47%), m.p. 150—152 °C [from CH_2Cl_2–Me_2SO–light petroleum (b.p. 60—80 °C)] (Found: C, 59.8; H, 5.25; N, 3.4, $C_{20}H_{21}NO_8$ requires C, 59.55; H, 5.2; N, 3.5); τ($CDCl_3$) 7.0 and 7.5 (each 2 H, m, CH_2CH_2), 6.5 (6 H, s, 2 × OCH_3), 6.20 (2 H, s, CH_2CO_2Me), 4.75 (2 H, s, $PhCH_2$), 2.67 (5 H, s, Ph), −0.1 (1 H, m br, NH), and −5.4 (1 H, br, OH).

t-Butyl 4-(2-*Methoxycarbonylethyl*)-3-*methylpyrrole*-2-*carboxylate* (**17d**).—A mixture of t-butyl 5-iodo-4-(2-methoxycarbonylethyl)-3-methylpyrrole-2-carboxylate (**17c**)[20] (25 g) anhydrous sodium acetate (10 g), and platinum oxide (0.1 g) in absolute ethanol (40 ml) was shaken under hydrogen at 20 °C and 760 mmHg. After uptake of hydrogen (1.5 l) was complete (4 h), the solution was filtered and evaporated under reduced pressure to give a colourless oil, which crystallised from light petroleum (b.p. 60—80 °C) to yield the α-free pyrrole (15.4 g, 91%), m.p. 46—47 °C (lit.,[21] m.p. 51—52 °C), τ($CDCl_3$) 8.45 (9 H, s, CMe_3), 7.75 (3 H, s, pyr-CH_3), 7.45 and 7.3 (each 2 H, m, CH_2CH_2), 6.35 (3 H, s, OCH_3), 3.4 (1 H, d, pyr-H), and 0.6 (1 H, br, NH); *m/z* 267 (M^+, 30%), 211 ($M^+ - C_4H_8$) (68%), 174 (30%), 138 (10%), and 120 (55%).

4-(2-*Methoxycarbonylethyl*)-3-*methoxycarbonylmethyl*-5-*methylpyrrole* (**8c**).—A mixture of benzyl 4-(2-methoxycarbonylethyl)-3-methoxycarbonylmethyl-5-methylpyrrole-2-carboxylate (**8a**)[9] (3.53 g), absolute ethanol (100 ml), and palladised charcoal (10%; 0.3 g) was shaken under hydrogen at 20 °C and 760 mmHg. After 1.5 h hydrogen ceased to be taken up. The solution was filtered through Celite and evaporated to dryness. The residual solid, m.p. 132—134 °C, was mixed with water (300 ml) and the mixture refluxed under nitrogen (oil-bath at 120—125 °C) for 1.5 h. The cooled, red solution was extracted with methylene chloride. The solution was washed with dilute aqueous sodium carbonate and water, dried ($MgSO_4$), and evaporated to dryness. The residual α-free pyrrole (2.03 g, 90%) was used immediately without further purification for pyrromethane preparation.

Benzyl 5-*Acetoxymethyl*-3-(2-*methoxycarbonylethyl*)-4-*methoxycarbonylmethylpyrrole*-2-*carboxylate* (**18b**).—To a stirred solution of the corresponding 5-methylpyrrole[9] (**18a**) (3.15 g) in glacial acetic acid (30 ml) lead tetra-acetate (4.4 g) (dried *in vacuo* at 45 °C for 1 h) was added over a period of 20 min. Stirring was continued for a further 3 h, after which the mixture was poured onto crushed ice (500 ml). The solid was filtered off and dissolved in methylene chloride (250 ml). After being washed with dilute aqueous sodium carbonate and water the solution was dried ($MgSO_4$) and evaporated under reduced pressure. The residual oil crystallised from methylene chloride–light petroleum (b.p. 60—80 °C) to afford the 5-*acetoxymethylpyrrole* (1.9 g, 50%), m.p. 104—105 °C (Found: C, 61.1; H, 5.9; N, 3.15. $C_{22}H_{25}NO_8$ requires C, 61.25; H, 5.8; N, 3.25); τ($CDCl_3$) 7.98 (3 H, s, $COCH_3$), 7.5 and 7.0 (each 2 H, t, CH_2CH_2), 6.48 (2 H, s, CH_2CO_2Me), 4.96 (2 H, pyr-CH_2C), 4.72 (2 H, s, CH_2Ph), 2.66 (5 H, s, Ph), and 0.55 (1 H, br, NH).

5-*Dimethylamido*-4-(2-*methoxycarbonylethyl*)-3-*methoxycarbonylmethyl*-2-*carboxylic Acid* (**25a**).—Powdered 5-benzyloxycarbonyl-3-(2-methoxycarbonylethyl)-4-methoxycarbonylmethylpyrrole-2-carboxylic acid (**24a**) (10.0 g) was added to stirred thionyl chloride (80 ml) over 20 min. The mixture was heated at 40—45 °C for 30—40 min and then evaporated under reduced pressure to give the acid chloride (**24b**) as a yellow oil which was dissolved in dry benzene (200 ml). Dimethylamine gas [generated from the 30% aqueous solution (150 ml) by distillation and drying by passage over sodium hydroxide pellets] was passed through the benzene solution for 30 min; the mixture was then kept for 2 h before adding to water (150 ml). The benzene was washed with water (2 × 50 ml), dilute aqueous acetic acid (100 ml) and then water again (50 ml) before being dried ($MgSO_4$). After removal of the benzene under reduced pressure, the desired intermediate dimethylamidopyrrole (**24c**) (8.5 g, 80%) was obtained as a yellow oil which could not be crystallised; τ($CDCl_3$) 7.50 and 7.3 (4 H, m, CH_2CH_2), 7.02 (6 H, s, NMe_2), 6.48 and 6.40 (6 H, s, OCH_3), 6.19 (2 H, s, CH_2CO_2Me), 4.77 (2 H, s, $PhCH_2$), 2.70 (5 H, s, Ph), and −0.1 (1 H, br, NH); *m/z* (f.d.) 860 ($2M^+$, 100%) and 430 (M^+, 60%).

The foregoing pyrrole amide (6 g) in absolute methanol (20 ml) was hydrogenated over 10% palladised charcoal (0.2 g) at 20 °C and 760 mmHg. Hydrogen (250 ml) was taken up during 19 h. The solution was then filtered through Celite and evaporated under reduced pressure. The residual oil was triturated with light petroleum (b.p. 60—80 °C) to give the *pyrrole acid* (2.93 g, 62%) as crystals m.p. 185—189 °C after crystallisation from methylene chloride and dimethyl sulphoxide–light petroleum (b.p. 60—80 °C) (Found: C, 50.9; H, 5.2; N, 7.4. $C_{15}H_{20}N_2O_7$ requires C, 52.9; N, 8.2%); τ($CDCl_3$–Me_2SO) *ca.* 7.45 (4 H, m, CH_2CH_2), 7.01 (6 H, s, NMe_2), 6.37 and 6.40 (6 H, s, 2 × OMe), 6.18 (2 H, s, CH_2CO_2Me), and −1.2 (1 H, br, NH).

5-Dimethylamido-2-iodo-4-(2-methoxycarbonylethyl)-3-methoxycarbonylmethylpyrrole (**25b**).—A solution of iodine (2.75 g) and potassium iodide (4 g) in water (75 ml) and methanol (25 ml) was added over 2 h at 60—65 °C to a stirred solution of the foregoing pyrrolecarboxylic acid (**25a**) (3.4 g) in aqueous sodium hydrogen carbonate [2.5 g in water (25 ml)] and methanol (25 ml). After completion of the addition the mixture was heated at 60 °C for 3 h, cooled, and poured into water (50 ml). The yellowish solid product was filtered off, taken up in methylene chloride (50 ml), and the solution washed with dilute aqueous sodium carbonate (20 ml), aqueous sodium hydrogen sulphite (10 ml), and water (20 ml), and then dried ($MgSO_4$). After removal of the methylene chloride under reduced pressure, the residue was recrystallised from methylene chloride–light petroleum (b.p. 60–80 °C) to give the desired *iodopyrrole* (2.6 g, 64%), m.p. 169—170 °C (Found: C, 39.5; H, 4.5; N, 6.9. $C_{14}H_{19}IN_2O_5$ requires C, 39.8; H, 4.5; N, 6.6%); τ($CDCl_3$) *ca.* 7.45 and 7.1 (4 H, m, CH_2CH_2), 6.94 (6 H, s, NMe_2), 6.57 (2 H, s, CH_2CO_2Me), 6.36 and 6.33 (6 H, s, 2 × OMe), and −0.25 (1 H, br, NH); *m/z* (f.d.) 4.22 (M^+) (100%).

5-Dimethylamido-4-(2-methoxycarbonylethyl)-3-methoxycarbonylmethylpyrrole (**25c**).—The foregoing iodopyrrole (2.2 g) and anhydrous sodium acetate (1 g) in absolute ethanol (40 ml) was hydrogenated over platinum oxide (0.1 g) at 20 °C and 760 mmHg. The catalyst was removed by filtration through Celite, and the solvent removed under reduced pressure to give the desired α-free dimethylamidopyrrole (1.4 g, 91%) as a pale yellow oil which could not be crystallised, and was used directly for pyrromethane synthesis; τ($CDCl_3$) 7.50 and 7.18 (4 H, m, CH_2CH_2), 6.97 (6 H, s, NMe_2), 6.55 (2 H, s, CH_2CO_2Me), 6.40 and 6.32 (6 H, s, OMe), 3.35 (1 H, d, pyr-H), and 0.07 (1 H, br, NH); *m/z* (f.d.) 296 (M^+, 100%).

Pyrromethanes and Pyrromethenes

Benzyl 3′,4-Bis(2-methoxycarbonylethyl)-3-methoxycarbonylmethyl-4′,5-dimethylpyrromethane-5′-carboxylate (**9**).—A mixture of crude α-free pyrrole prepared from benzyl 4-(2-methoxycarbonylethyl)-3-methoxycarbonylmethyl-5-methylpyrrole-2-carboxylate (**8c**) (300 mg), toluene-*p*-sulphonic acid monohydrate (8.3 mg), benzyl 5-acetoxymethyl-4-(2-methoxycarbonylethyl)-3-methylpyrrole-2-carboxylate (**7b**) (278 mg) in absolute methanol, (3 ml) was boiled under nitrogen for 3 h. The mixture was then diluted with water until it became cloudy after which it was stored in a refrigerator overnight to give pink crystals. These were recrystallised from methylene chloride–light petroleum (b.p. 60—80 °C) to yield the required *pyrromethane* (320 mg, 78%), m.p. 96—98 °C (Found: C, 65.65; H, 6.65; N, 5.2. $C_{30}H_{36}N_2O_8$ requires C, 65.2; H, 6.6; N, 5.1%); τ($CDCl_3$) 7.90 and 7.70 (each 3 H, s, pyr-Me), 7.3 and 7.4 (each 2 H, m, CH_2CH_2), 6.55 (2 H, s, pyr-CH_2), 6.35 (6 H, s, OMe), 6.18 (2 H, s, pyr-CH_2-pyr), 4.71 (2 H, s, CH_2Ph), 2.6 (5 H, m, Ph), and 1.4 and 0.2 (each 1 H, br, NH); *m/z* (f.d.) 552 (M^+, 100%).

t-Butyl 5′-Benzyloxycarbonyl-3,4′-bis(2-methoxycarbonylethyl)-3′-methoxycarbonylmethyl-4-methylpyrromethane-5-carboxylate (**12a**).—A mixture of t-butyl 4-(2′-methoxycarbonylethyl)-3-methylpyrrole-2-carboxylate (**17d**) (500 mg), benzyl 5-acetoxymethyl-3-(2-methoxycarbonylethyl)-4-methoxycarbonylmethylpyrrole-2-carboxylate (**18b**) (807 mg), toluene-*p*-sulphonic acid monohydrate (25 mg, 0.07 mmol) in acetic acid (9 ml) was heated at 45 °C for 3 h. The acetic acid was removed under reduced pressure at 40 °C and the residual oil was chromatographed on alumina (grade III) eluting with 5% ethyl acetate in light petroleum (b.p. 60—80 °C). The *product* was obtained as a colourless oil (917 mg, 76%) which could not be crystallised; τ($CDCl_3$) 8.52 (9 H, s, CMe_3), 7.77 (3 H, s, pyr-CH_3), 7.4 and 7.0 (each 2 H, m, CH_2CH_2), 6.3, 6.43, and 6.5 (each 3 H, s, OCH_3), 4.78 (2 H, s, CH_2Ph), 2.7 (5 H, s, Ph), 0.25 (1 H, br, NH), and 0.03 (1 H, br, NH); *m/z* (f.d.) 638 (M^+, 100%) and 548 ($M^+ - CH_2C_6H_5$, 10%).

Benzyl 3,4′-Bis(2-methoxycarbonylethyl)-3′-methoxycarbonylmethyl-4-methylpyrromethane-5′-carboxylate (**12b**).—To t-butyl 5′-benzyloxycarbonyl-3,4′-bis(2-methoxycarbonylethyl)-3′-methoxycarbonylmethyl-4-methylpyrromethane-5-carboxylate (**12a**) (4.17 g) was added at room temperature trifluoroacetic acid (30 ml) and the mixture was stirred under nitrogen for 1 h at 20 °C. The trifluoroacetic acid was removed under reduced pressure at 20 °C, and the residual red oil was taken up in methylene chloride (50 ml) and the solution washed with dilute aqueous sodium (25 ml). The resulting yellow solution was washed with water, dried ($MgSO_4$), and evaporated under reduced pressure to yield a pink oil which could not be crystallised (3.25 g, 93%). The oil was dried *in vacuo* (0.1 mmHg) at 20 °C overnight and used without further purification; τ($CDCl_3$) 8.00 (3 H, s, pyr-CH_3), 7.45, 7.3, and 7.05 (8 H, m, CH_2CH_2), 6.05, 6.39, and 6.3 (11 H, s, OCH_3 and CH_2CO_2Me), 6.15 (2 H, s, pyr-CH_2-pyr), 4.8 (2 H, s, CH_2Ph), 3.62 (1 H, s, pyr-H), 2.70 (5 H, s, Ph), and 1.05 (1 H, br, NH) and 0.25 (1 H, br, NH); *m/z* (f.d.) 538 (100%, M^+).

Benzyl 5-Dimethylamido-3,4′-dimethyl-4,3′-bis(2′-methoxycarbonylethyl)pyrromethane-5′-carboxylate (**13a**).—A mixture of 5-dimethylamido-4-(2′-methoxycarbonylethyl)-3-methylpyrrole (**21c**) (0.96 g) and benzyl 5-acetoxymethyl-4-(2-methoxycarbonylethyl)-3-methylpyrrole-2-carboxylate (**19b**) (1.49 g) in acetic acid (20 ml) was stirred and heated at 100 °C for 2.5 h. The acetic acid was then removed under reduced pressure and the residual oil taken up in methylene chloride (50 ml) and washed with dilute aqueous sodium carbonate (25 ml) and water (25 ml) and dried ($MgSO_4$). Evaporation under reduced pressure gave a red oil which was chromatographed on alumina (grade III) in methylene chloride–light petroleum (b.p. 60—80 °C). The *product* was obtained as colourless needles (1.55 g, 70%) from methylene chloride–light petroleum (b.p. 60–80 °C), m.p. 137—138 °C (Found: C, 65.9; H, 6.9; N, 7.8. $C_{30}H_{37}N_3O_7$ requires C, 65.4; H, 6.7; N, 7.6%); τ($CDCl_3$) 8.05 and 7.81 (6 H, s, pyr-CH_3), 7.6 and 7.3 (8 H, t, CH_2CH_2), 7.3 (6 H, s, NMe_2), 6.38 (6 H, s, OCH_3), 6.22 (2 H, s, pyr-CH_2-pyr), 4.77 (2 H, s, CH_2Ph), 2.68 (5 H, s, Ph), and 0.6 and 0.44 (each 1 H, br, NH).

Dibenzyl 3,3′-bis(2-methoxycarbonylethyl)-4,4′-dimethylpyrromethane-5,5′-dicarboxylate (**22**) (40 mg) was also obtained from a first fraction of the eluate, m.p. 100—101 °C (from methylene chloride–light petroleum) (lit.,[20] m.p. 99.5—100 °C) τ($CDCl_3$) 7.75 (6 H, s, pyr-CH_3), 7.6 and 7.3 (8 H, m, CH_2CH_2), 6.45 (6 H, s, OCH_3), 6.05 (2 H, s, pyr-CH_2-pyr), 4.77 (4 H, s, CH_2Ph), 2.70 (10 H, s, Ph), and 0.80 (2 H, br, 2 NH).

Benzyl 5-Dimethylamido-4,3′-bis(2-methoxycarbonylethyl)-4-methoxycarbonylmethyl-3-methylpyrromethane-5′-carboxylate (**13b**).—A mixture of benzyl 5-acetoxymethyl-4-(2-methoxycarbonylethyl)-3-methoxycarbonylmethylpyrrole-?-carboxylate (**23b**) (1.25 g) and 5-dimethylamido-4-(2′-methoxycarbonylethyl)-3-methylpyrrole (**21c**) (0.80 g) in acetic acid (15 ml) was heated under nitrogen at 90—95 °C for 3 h. The acetic acid was evaporated under reduced pressure to give a red oil which was chromatographed on alumina (grade III) eluting with ethyl acetate–light petroleum (b.p. 60–80 °C) to yield the required *amidopyrromethane* (770 mg, 44%), m.p. 108—109 °C (Found: C, 62.2; H, 6.0; N, 6.3. $C_{32}H_{39}N_3O_9$ requires C, 62.05; H, 6.4; N, 6.9%); *m/z* (f.d.) 609 (M^+, 100%).

Benzyl 5'-Dimethylamido-3,4'-bis(2-methoxycarbonylethyl)-4,3'-bismethoxycarbonylmethylpyrromethane-5-carboxylate (**13c**).—This compound was prepared by the procedure described above [for the amidopyrromethane (**13a**) and (**13b**)] from the condensation of 5-dimethylamido-4-(2-methoxycarbonylethyl)-3-methoxycarbonylmethylpyrrole (**25c**) (1.5 g) and benzyl 5-acetoxymethyl-4-(2-methoxycarbonylethyl)-3-methoxycarbonylmethylpyrrole-2-carboxylate (**23b**) (2.1 g). The product was an oil (2.3 g, 69%) which could not be crystallised; τ($CDCl_3$) *ca.* 7.5 and 7.3 (8 H, br, CH_2CH_2), 7.05 (6 H, s, NMe_2), *ca.* 6.41 (12 H, m, OCH_3), 6.56 (2 H, s, CH_2CO_2Me), 6.22 (2 H, s, CH_2CO_2Me), 6.16 (2 H, s, pyr-CH_2-pyr), 4.8 (2 H, s, $PhCH_2$), 2.68 (5 H, m, Ph), 0.12 (1 H, br, NH), and −0.22 (1 H, br, NH); *m/z* (f.d.) 667 (M^+) (100%).

Benzyl 3,4'-Bis(2-methoxycarbonylethyl)-3',4-dimethyl-5'-t-butyloxycarbonylpyrromethane-5-carboxylate (**30a**).—A mixture of t-butyl 3-(2-methoxycarbonylethyl)-4-methylpyrrole-2-carboxylate (**29**) (133 mg) and benzyl 5-acetoxymethyl-4-(2-methoxycarbonylethyl)-3-methylpyrrole-2-carboxylate (**7b**) (187 mg) in methylene chloride (3 ml), absolute methanol (0.5 ml), and toluene-*p*-sulphonic acid monohydrate (6.7 mg) was boiled for 1 h under reflux. Removal of the solvent under reduced pressure afforded a red oil, which crystallised from aqueous methanol to yield the *pyrromethane* (204 mg, 70%), m.p. 139—142 °C (Found: C, 65.5; H, 7.3; N, 6.1. $C_{32}H_{40}N_2O_8$ requires C, 66.2; H, 6.9; N, 4.8%); τ($CDCl_3$) 8.52 (9 H, s, $OCMe_3$), 8.05 (3 H, s, pyr-CH_3), 7.76 (3 H, s, pyr-CH_3), *ca.* 6.9—7.7 (8 H, m, CH_2CH_2), 6.37 (6 H, s, OMe), 6.12 (2 H, s, pyr-CH_2-pyr), 4.76 (2 H, s, $PhCH_2$), 2.7 (5 H, s, Ph), and 0.8 and 0.58 (2 H, br, NH); *m/z* (f.d.) 580 (M^+, 100%).

5-Bromo-3,4'-(2-methoxycarbonylethyl)-3'-methoxycarbonylmethyl-5',4-dimethylpyrromethene Hydrobromide (**10a**).—Benzyl 4,5'-dimethyl-3'-methoxycarbonylmethyl-3,4'-bis(2-methoxycarbonylethyl)pyrromethane-5-carboxylate (**9**) (124 mg) in absolute ethanol (2 ml) was hydrogenated over palladium–charcoal (10%; 0.2 g) at 20 °C and 760 mmHg. After hydrogen uptake was complete the solution was filtered through Celite, the solvent removed under reduced pressure and the residue was dissolved in acetic acid (2 ml). Bromine (170 mg) in acetic acid (2 ml) was added and the mixture heated at 100 °C for 30 min. Removal of the acetic acid and excess of bromine under reduced pressure afforded a black oil which crystallised from methylene chloride–light petroleum (b.p. 60–80 °C) to yield the *pyrromethane hydrobromide* as deep red needles (65 mg, 50%), m.p. 210—211 °C; $\lambda_{max.}$($CHCl_3$); 497 nm (log ε 5.02); τ($CDCl_3$) 8 (3 H, s, 5'-Me), 7.31 (3 H, s, 4-Me), 6.45, 6.40, and 6.35 (each 3 H, s, OMe), 6.95—7.45 (8 H, m, CH_2CH_2), 6.81 (4 H, s, CH_2CO_2Me), 2.61 (1 H, s, CH-pyr), and −3.75 (2 H, br, NH).

Oxobilanes

Dibenzyl 1,3,5,7-Tetrakis(2-methoxycarbonylethyl)-2-methoxycarbonylmethyl-4,6,8-trimethyl-b-oxobilane-1',8'-dicarboxylate (**15a**).—To benzyl 3,4'-bis(2-methoxycarbonylethyl)-4,3'-dimethylpyrromethane-5-carboxylate (1.42 g) was added phosphoryl chloride (12 ml). The mixture was heated at 50—60 °C under nitrogen for 35 min after which it was evaporated under reduced pressure. To the residual oil was added 1,2-dibromoethane (10 ml) and the mixture was again evaporated under reduced pressure; this was repeated three times to remove excess of phosphoryl chloride. The complex was dissolved in freshly distilled methylene chloride (5 ml) and mixed with the solution of benzyl 3-methoxycarbonylmethyl-3,4-bis(2-methoxycarbonylethyl)-4'-methylpyrromethane-5-carboxylate (1.54 g) in freshly distilled methylene chloride (5 ml). The mixture was boiled under N_2 and in the dark (oil-bath at 50—55 °C) for 43 h until the new absorption at $\lambda_{max.}$ 412 nm had reached a maximum. Methylene chloride (50 ml) was added and the mixture was washed with water (50 ml) and dried ($MgSO_4$). Evaporation of the solvent under reduced pressure yielded a brown oil which was chromatographed on alumina (grade III) in ethyl acetate–light petroleum (b.p. 60—80 °C). The proportion of ethyl acetate was increased stepwise from 10 to 100% (until the eluate was colourless) and the desired imine salt (**14a**) was then stripped off from the column with methanol. After evaporation of the solvent the imine salt (1.69, 59%) was obtained as an oil.

This was taken up in benzene (100 ml) and vigorously stirred with 10% aqueous sodium carbonate (100 ml) and heated (oil-bath temp. 75—80 °C) under nitrogen and in the dark for 4 h until the peak at λ 345 nm had reached a maximum. The mixture was extracted with more benzene (100 ml) and the organic layer was separated, washed with water (100 ml), and dried ($MgSO_4$). The benzene was removed under reduced pressure to give a brown oil which was chromatographed on alumina (grade III) in ethyl acetate–benzene. The required *b*-oxobilane (1.02 g, 65%) was obtained as a thick yellow oil which could not be crystallised, but its n.m.r. spectrum and t.l.c. (one spot) showed that it was essentially a single compound pure enough for conversion to porphyrin: τ($CDCl_3$) 8.07 and 7.79 (9 H, s, pyr-CH_3), 6.9—7.7 (16 H, m, CH_2CH_2), 6.35—6.62 (17 H, m, OCH_3 and CH_2CO_2Me), 6.1 and 6.18 (each 2 H, s, pyr-CH_2-pyr), 4.76 (4 H, s, CH_2Ph), 2.68 (10 H, s, Ph), 1.1, 0.82, 0.45, and 0.15 (each 1 H, br, NH).

Dibenzyl 2,4,6,8-Tetrakis(2-methoxycarbonylethyl)-1,3,7-trismethoxycarbonylmethyl-5-methyl-b-oxobilane-1',8-dicarboxylate (**15b**).—This compound (357 mg, 36%) was similarly obtained, by the procedure described for the preceding b-oxobilane, from benzyl 5'-dimethylamido-3,4'-bis(2-methoxycarbonylethyl)-4,3'-bismethoxycarbonylmethyl-pyrromethane-5-carboxylate (576 mg) and t-butyl benzyl 3,4'-bis(2'-methoxycarbonylethyl)-3'-methoxycarbonylmethyl-4-methylpyrromethane-5,5'-dicarboxylate (1.10 g). It was an oil which could not be crystallised, $\lambda_{max.}$ 350 nm ($\varepsilon_{max.}$ *ca.* 27 600); τ($CDCl_3$) 8.25 (3 H, s, pyr-CH_3), *ca.* 7.55 and 7.00 (16 H, br, CH_2CH_2), 6.45 (21 H, m, OCH_3), 6.22 and 6.10 (8 H, br, pyr-CH_2-pyr and CH_2CO_2Me), 4.80 (4 H, s, CH_2Ph), 2.7 (10 H, m, Ph), and 0.21, 0.09, 0.10, and 0.28 (4 H, br, NH).

Dibenzyl 2,4,8,7-Tetrakis(2'-methoxycarbonylethyl)-1,7-bismethoxycarbonylmethyl-3,5-dimethyl-b-oxobilane-1',8'-dicarboxylate (**15c**).—This compound was prepared by the same method as that described for two preceding *b*-oxobilanes in 23% yield (based on the amidopyrromethane) from benzyl 3',4-bis(2'-methoxycarbonylethyl)-3-methoxycarbonylmethyl-4-methylpyrromethane-5-carboxylate and benzyl 5'-dimethylamido-3,4'-bis(2-methoxycarbonylethyl)-4-methoxycarbonylmethyl-3'-methylpyrromethane-5-carboxylate. It could not be crystallised but was shown to be pure by n.m.r. spectroscopy: τ($CDCl_3$) 8.07 (6 H, s, pyr-CH_3), *ca.* 7.05—*ca.* 7.90 (16 H, m, CH_2CH_2), *ca.* 6.20—6.65 (22 H, m, OCH_3 and CH_2CO_2Me), 6.17 and 6.11 (each 2 H, s, pyr-CH_2-pyr), 6.80 and 6.81 (4 H, s, CH_2Ph), 2.7 (10 H, m, Ph), and 0.65, 0.57, 0.35, and 0.10 (4 H, br, NH).

Porphyrins

β-Acetoxy-2-methoxycarbonylmethyl-1,3,5,7-tetrakis(2'-methoxycarbonylethyl)-4,6,8-trimethylporphin (**17a**).—Dibenzyl 1,3,5,7-tetrakis(2'-methoxycarbonylmethyl)-4,6,8-trimethyl-*b*-oxobilane-1',8'-dicarboxylate (1.02 g) in tetrahydrofuran (20 ml) containing triethylamine (5 drops) was hydrogenated over

palladium–charcoal (10%; 150 mg) at 20 °C and 760 mmHg. The catalyst was filtered off through Celite and the solution evaporated to dryness at 20 °C under reduced pressure. The residual oil was dissolved in dry methylene chloride (190 ml) and the solution treated with trimethyl orthoformate (3 ml) and M-trichloroacetic acid (61 ml) (10 g in 61 ml). The mixture was stirred in the dark at 20 °C in the air for 3 h, and then treated with pyridine (7.6 ml). The resulting green solution was stirred overnight (20 h) and evaporated under reduced pressure. The residual solid was stirred with pyridine (15 ml) and acetic anhydride (50 ml) for 40 min. The resulting red solution was evaporated under reduced pressure first at 60 °C (15 mmHg) then at 50 °C (0.1 mmHg) to yield a dark solid, which was chromatographed alumina (grade III) eluting with 10% tetrahydrofuran in methylene chloride. The red band afforded the β-*acetoxyporphyrin* (225 mg, 28% from *b*-oxobilane), m.p. 201—202 °C (Found: C, 64.1; H, 5.85; N, 6.8. $C_{44}H_{50}N_4O_{12}$ requires C, 63.9; H, 6.05; N, 6.8); $\lambda_{max.}$($CHCl_3$) 403 (log ε 5.12), 499 (4.11), 535 (3.95), 570 (3.60), and 624 nm (3.38); τ ($CDCl_3$; 0.06M) 7.3 (3 H, s, $COCH_3$), 6.61, 6.50, 6.47, 6.41, 6.35, 6.31, and 6.22 (24 H, s, OCH_3 and pyr-CH_3); *ca.* 6.83 and *ca.* 5.84 (16 H, m, CH_2CH_2), 5.2 (2 H, s, CH_2CO_2Me) 0.32 (1 H, s, *meso*-H), 0.10 (1 H, s, *meso*-H), and 0.04 (1 H, s, *meso*-H), and 13.85 (2 H, br, NH); *m/z* (f.d.) 826 (M^+, 100%).

α-*Acetoxy*-1,3,5,7-*tetrakis*(2-*methoxycarbonylethyl*)-4,6-*bismethoxycarbonylmethyl*-2,8-*dimethylporphin* (**17b**).—This compound was synthesized by the procedure described for the preceding acetoxyporphyrin in 12% yield (based on the corresponding *b*-oxobilane), m.p. 203—204 °C (Found: C, 62.3; H, 5.9; N, 6.2. $C_{46}H_{52}N_4O_{14}$ requires C, 62.4; H, 5.9; N, 6.3%); $\lambda_{max.}$($CHCl_3$) 403 (log ε 5.12), 496 (4.01), 531 (3.82), 569 (3.91), and 624 (3.37); τ($CDCl_3$) 7.0 (3 H, s, $COCH_3$), *ca.* 6.65—*ca.* 6.95 and *ca.* 5.60—*ca.* 6.0 (16 H, m, CH_2CH_2), *ca.* 6.10—*ca.* 6.60 (24 H, m, OCH_3 and pyr-CH_3), 5.20 (4 H, s, CH_2CO_2Me), 0.28 and 0.08 (3 H, s, *meso*-H), and 13.85 (2 H, br, NH); *m/z* (f.d.) 884 (M^+, 100%).

α-*Acetoxy*-1,3,5,7-*tetrakis*(2-*methoxycarbonylethyl*)-4,6,8-*trismethoxycarbonylmethyl*-2-*methylporphin* (**17c**) was obtained in the same way as the preceding *meso*-acetoxyporphyrins in 24% yield (67 mg), m.p. 208–209 °C from the appropriate *b*-oxobilane (357 mg) from methylene chloride–light petroleum (b.p. 60—80 °C) (Found: C, 61.0; H, 5.7; N, 6.0. $C_{48}H_{54}N_4O_{16}$ requires C, 61.15; H, 5.7; N, 5.9%); $\lambda_{max.}$($CHCl_3$) 403 (log ε 5.12), 499 (4.11), 535 (3.95), 570 (3.60), and 624 (3.39); τ ($CDCl_3$; 0.02M) 7.00 (3 H, s, $COCH_3$), *ca.* 6.72 and *ca.* 5.7 (16 H, m, CH_2CH_2), 6.48, 6.35, 6.26, 6.3, and 6.18 (24 H, s, OCH_3 and pyr-CH_3), 5.19, 5.16, and 4.92 (6 H, s, CH_2CO_2Me), 0.18, 0.02, and 0.13 (3 H, s, *meso*-H), and 14.70 (2 H, br, NH); *m/z* (f.d.) 942 (M^+) (100%).

1,3,5,7-*Tetrakis*(2-*methoxycarbonylethyl*)-2-*methoxycarbonylmethyl*-4,6,8-*trimethylporphin* (**5c**).—β-Acetoxy-2-methoxycarbonylmethyl-1,3,5,7-tetrakis(2-methoxycarbonylethyl)-4,6,8-trimethylporphin (51 mg) in tetrahydrofuran (20 ml) containing triethylamine (5 drops) was hydrogenated at 20 °C and 760 mmHg over palladised charcoal (10%; 50 mg). Hydrogen (15 ml) was taken up (3 h) and the solution was filtered through Celite, which was subsequently washed well by tetrahydrofuran. The resulting colourless filtrate was treated immediately with 2,3-dichloro-5,6-dicyano-*p*-benzoquinone (43 mg) in dry benzene (1 ml). The red solution was evaporated under reduced pressure and the residual oil was chromatographed on alumina (grade III) in methylene chloride–benzene (1:1). The required *porphyrin* (17.0 mg, 36%) crystallised from methylene chloride–methanol, m.p. 289—290 °C (Found: C, 65.7; H, 6.4; N, 7.4. $C_{42}H_{48}N_4O_{10}$ requires C, 65.6; H, 6.25; N, 7.3%); $\lambda_{max.}$($CHCl_3$) 402 (log ε 5.12), 497 (4.11), 532 (3.93), 569 (3.61), and 624 nm (3.38); τ ($CDCl_3$; 0.5M) *ca.* 6.8 and *ca.* 5.7 (16 H, m, CH_2CH_2), 6.5, 6.48, 6.35, 6.30, and 6.25 (24 H, s, OCH_3 and por-CH_3), 5.05 (2 H, s, CH_2CO_2Me), 0.12, 0.07, and 0.01 (4 H, s, *meso*-H), and 14.5 (2 H, s, NH); *m/z* (f.d.) 768 (M^+, 100%).

1,7-*Bismethoxycarbonylmethyl*-2,4,6,8-*tetrakismethoxycarbonylethyl*-3,5-*dimethylporphin* (**3c**).—This was prepared by reduction of the corresponding acetoxyporphyrin and re-oxidation of the resulting porphyrinogen employing the procedure described above in 47% yield; it had m.p. 195—200 °C from methylene chloride–methanol (Found: C, 63.7; H, 6.0; N, 6.2. $C_{44}H_{50}N_4O_{12}$ requires C, 64.0; H, 5.9; N, 6.8%); $\lambda_{max.}$($CHCl_3$) 403 (log ε 5.12), 496 (4.01), 531 (3.82), 568 (3.90), and 624 (3.36); τ ($CDCl_3$; 0.04M) 6.55—*ca.* 6.9 and *ca.* 5.5—*ca.* 5.9 (16 H, br, CH_2CH_2), *ca.* 6.2—*ca.* 6.5 (24 H, m, OCH_3 and pyr-CH_3), 4.95 and 5.00 (4 H, s, CH_2CO_2Me), and *ca.* 0.9—*ca.* 1.10 (4 H, q, *meso*-H); *m/z* (f.d.) 826 (M^+, 100%).

2,4,6,8-*Tetrakis*(2′-*methoxycarbonylethyl*)-1,3,7-*trismethoxycarbonylmethyl*-5-*methylporphin* (**2c**).—This was prepared in 69% yield from the corresponding α-acetoxyporphyrin by the procedures described above for the other porphyrins. It had m.p. 254—255 °C (Found: C, 62.3; H, 5.8; N, 6.3. $C_{46}H_{52}N_4O_{14}$ requires C, 62.4; H, 5.9; N, 6.3%). Mixed m.p. with naturally derived material (m.p. 249—252 °C) was 249—253 °C; $\lambda_{max.}$($CHCl_3$) 403 (log ε 5.12), 499 (4.11), 535 (3.95), 570 (3.60), and 642 (3.38) nm; τ ($CDCl_3$; 0.02M) 6.70 and 5.65 (16 H, br, CH_2CH_2), 6.41, 6.34, 6.3, and 6.22 (24 H, m, OCH_3 and pyr-CH_3), 5.02, 4.95, and 4.9 (6 H, s, CH_2CO_2Me), 0.00 −0.08, and 1.00 (4 H, s, *meso*-H), and 13.98 (2 H, br, NH); *m/z* (f.d.) 884 (M^+, 100%).

2,4,6,8-*Tetrakis*(2′-*methoxycarbonylethyl*)-3,7-*bismethoxycarbonylmethyl*-1,5-*dimethylporphin* (**4c**).—A mixture of the pyrromethane (**10a**) (65 mg) and methylsuccinic acid (380 mg) was heated under nitrogen at 118 °C (b.p. of acetic acid) for 4—5 h. Then water (50 ml) was added and the mixture centrifuged. The black solid was dried *in vacuo* overnight and esterified at 20 °C and in the dark with methanolic sulphuric acid (5%; 100 ml). The mixture was neutralized by 5% ammonium hydroxide (50 ml) and extracted with methylene chloride (2 × 50 ml). The extract was washed with water, dried ($MgSO_4$), and evaporated under reduced pressure. The dark residue recrystallised from methylene chloride–methanol to give the *porphyrin* (**4c**) (99 mg, 10%) as deep red needles, m.p. 294—295 °C (Found: C, 63.6; H, 5.8; N, 6.2. $C_{44}H_{50}N_4O_{12}$ requires C, 63.9; H, 6.1; N, 6.8%); $\lambda_{max.}$($CHCl_3$) 403 (log ε 5.12), 499 (4.11), 5.35 (3.95), 570 (3.60), and 624 nm (3.38); τ ($CDCl_3$; 0.02M) *ca.* 6.50—*ca.* 6.85 and 5.45—5.75 (8 H, m, CH_2CH_2), 6.25, 6.35, and 6.4 (18 H, s, OCH_3), 4.95 (4 H, s, CH_2CO_2Me), 0.09 and −0.15 (4 H, s, *meso*-H), and 13.78 (2 H, br, NH); *m/z* (f.d.) 826 (M^+, 100%).

Incubation of the Heptacarboxylic Porphyrinogen (**2a**) *with Chicken Red Cell Haemolysates.*—The corresponding heptacarboxylic porphyrin heptamethyl ester (**2c**) (1 mg) was hydrolysed to the free acid in 25% hydrochloric acid (2 ml) at 20 °C overnight. After removal of the acid under reduced pressure the residue was dissolved in dilute ammonium hydroxide (0.05M; 4 ml) and potassium phosphate (1M; 1 ml), and reduced to porphyrinogen by shaking with sodium amalgam (3%; 5 g) at 0—5 °C under nitrogen. The solution was filtered rapidly through glass wool, and the pH adjusted to 7.3 by dropwise addition of acetic acid, before dilution to 10 ml with deionised water (which had previously been saturated with nitrogen). This solution was then added directly to a freshly prepared haemolysate (100 ml) of chicken erthrocytes (prepared as described elsewhere),[10] and the mixture kept at 37 °C

for 5 min. Ethyl acetate–acetic acid (100 ml; 70/30, v/v) was then added, and after being shaken briefly the mixture was centrifuged and filtered. The residue was re-extracted with further portions of ethyl acetate–acetic acid (75 ml) until no further orange fluorescence was observed under u.v. light. The organic layer was separated, washed with sodium acetate (100 ml; 3%), and then with water (100 ml). The porphyrin was extracted into hydrochloric acid (10%; several 15 ml portions), and any haem present was removed by extraction with ether (50 ml; peroxide-free). The aqueous solution was adjusted to pH 7.3 (Congo Red) with saturated aqueous sodium acetate, and the porphyrin extracted into ether (3 × 50 ml). The ether extracts were washed with deionised water (50 ml), and the ether evaporated to dryness. The residue was esterified by treatment with methanolic sulphuric acid (5%) (10 ml) for 16 h and the product isolated by pouring onto crushed ice (5 g) followed by extraction with chloroform (2 × 10 ml). The extracts were washed with dilute ammonia (5%; 20 ml) and cold water (2 × 10 ml). The solvent was removed under reduced pressure and the residual esters dried *in vacuo* (0.01 mmHg), before analysis by h.p.l.c. The hexacarboxylic porphyrin was shown[5] to be a mixture of the two possible type I isomers (**3**) and (**4**).

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