Synthetic and Biosynthetic Studies of Porphyrins. Part 2.[1] Synthesis of Isopempto- and Isochlorocruoro-porphyrins

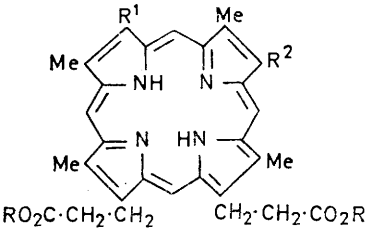
By **David E. Games, Anthony H. Jackson,*** and **Peter J. O'Hanlon,** Department of Chemistry, University College, Cardiff CF1 1XL

Isopempto- and isochlorocruoro-porphyrin dimethyl esters have been prepared by application of the MacDonald porphyrin synthesis. The vinyl group was introduced by elimination of hydrogen chloride from chloroethyl side-chains carried through from an intermediate pyrrole, and the formyl group (of isochlorocruoroporphyrin) was introduced at the porphyrin stage with butyl dichloromethyl ether in the presence of tin(IV) chloride.

THE iron(II) chelate of chlorocruoroporphyrin (1a) (or spirographis porphyrin) is the prosthetic group of the oxygen carrying haemoprotein of the polychaete worm *Spirographis spallanzanii*.[2] Its structure [3] was originally assigned by Fischer, but there was a slight element of ambiguity and the isomeric structure (1b) could not be completely excluded. Partial syntheses of chlorocruoroporphyrin dimethyl ester (2a) from protoporphyrin-IX [4-6] (2c) and from deuteroporphyrin-IX [7] (2d) did not make possible an unequivocal distinction between the two structures; this was only achieved recently by ring synthesis and subsequent elaboration of the vinyl and formyl side-chains.[8]

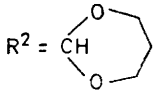
In the course of earlier work in Liverpool,[8] chlorocruoroporphyrin dimethyl ester (2a) was synthesised *via* the *b*-oxobilane procedure, as well as the related pempto- and isopempto-porphyrin esters (2e and f). However the ring synthesis of isochlorocruoroporphyrin (1b) remained to be completed, and we decided to investigate its preparation *via* the MacDonald route.[9] As in the earlier work the synthesis of a porphyrin with a free β-position was envisaged so that the formyl group could be introduced at a later stage; it was hoped to elaborate the vinyl group from a chloroethyl side-chain introduced originally at the pyrrole stage.

The pyrrole (4b) required as a precursor of ring B of



(1) R = H
(2) R = Me

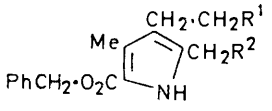
a; R^1 = CHO, R^2 = CH:CH_2
b; R^1 = CH:CH_2 R^2 = CHO
c; R^1 = R^2 = CH:CH_2
d; R^1 = R^2 = H
e; R^1 = H, R^2 = CH:CH_2
f; R^1 = CH:CH_2, R^2 = H
g; R^1 = CH_2·CH_2Cl, R^2 = H
h R^1 = CH_2·CH_2Cl, R^2 = CHO
j R^1 = CH_2·CH_2Cl,



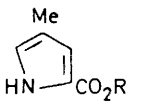
chlorocruoroporphyrin was prepared by transesterification of the related methyl ester (4a), which is now

[1] Part 1, P. W. Couch, D. E. Games, and A. H. Jackson, preceding paper.
[2] O. Warburg, *Biochem. Z.*, 1930, **227**, 171.
[3] H. Fischer and C. von Seeman, *Z. physiol. Chem.*, 1936, **242**, 133.
[4] H. Fischer and K. O. Dielmann, *Z. physiol. Chem.*, 1944, **280**, 186.
[5] R. Lemberg and A. Parker, *Austral. J. Exp. Biol.*, 1952, **30**, 163.
[6] H. H. Inhoffen, G. Bliesener, and H. Brockmann, *Annalen*, 1969, **730**, 173.
[7] H. Fischer and G. Wecker, *Z. physiol. Chem.*, 1942, **272**, 1.
[8] A. H. Jackson, G. W. Kenner, and J. Wass, *J.C.S. Perkin I*, 1974, 480.
[9] *Cf.* G. P. Arsenault, E. Bullock, and S. F. MacDonald, *J. Amer. Chem. Soc.*, 1960, **82**, 4384.

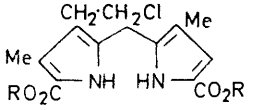
readily available by a five-stage synthesis from pyrrole.[10] (Earlier we had attempted to methylate pyrrole-2-carboxylic esters or 2-formylpyrrole under Friedel–Crafts type conditions but without success.). The precursor of ring A was the chloroethylpyrrole (3b) prepared by treatment of the hydroxyethylpyrrole [11] (3a) with thionyl chloride. Acetoxylation with lead tetra-acetate then afforded the corresponding acetoxymethyl derivative (3c) which was coupled directly with the pyrrole benzyl ester (4b) in methanol at 30 °C in the presence of a catalytic amount of toluene-*p*-sulphonic acid [12] to give the pyrromethane (5a) in 80% yield; condensation in hot acetic acid as in the original MacDonald procedure [13] gave lower yields. Hydrogenolysis of this dibenzyl ester (5a) afforded the pyrromethanedicarboxylic acid (5b), which was condensed with the diformylpyrromethane (6b) in a mixture of methanol and methylene chloride



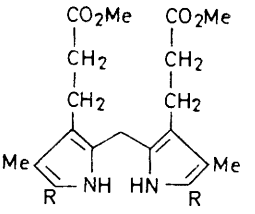
(3) a; R^1 = OH, R^2 = H
b; R^1 = Cl, R^2 = H
c; R^1 = Cl, R^2 = OAc



(4) a; R = Me
b; R = CH_2Ph



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



(6) a; R = CO_2H
b; R = CHO

containing toluene-*p*-sulphonic acid.[14] Subsequently an excess of zinc acetate was added and the mixture left in air until porphyrin production had reached a maximum (estimated spectroscopically from the Soret band at 400 nm). The zinc was removed and the side-chain re-esterified with methanolic sulphuric acid prior to column chromatography. The required porphyrin (2g) was obtained in 12% yield; coproporphyrin-II tetramethyl ester was also obtained in 3% yield, presumably from self-condensation of the diformylpyrromethane (6b). The relatively low yield of porphyrin was not improved by varying the reaction conditions (or by using the original MacDonald method [9]); this was attributed to the effects of the free β-position in the pyrromethane (5b).

The formyl group was introduced in 48% yield by treatment of the iron(III) complex of the porphyrin (2g) with butyl dichloromethyl ether and tin(IV) chloride, followed by removal of the iron with iron(II) sulphate and acidic work-up. Protection of the formyl porphyrin (2h) as its 2,2-dimethyltrimethylene acetal (2j) (as in the synthesis of chlorocruoroporphyrin itself [8]) and treatment with zinc acetate gave the zinc complex. The latter underwent elimination of hydrogen chloride on dissolution in t-butyl alcohol containing potassium t-butoxide, the reaction being followed by the small bathochromic shift in the visible spectrum. Isochlorocruoroporphyrin dimethyl ester (2b) was finally obtained, after removal of the zinc and re-esterification with methanolic sulphuric acid, in 36% overall yield from the β-free porphyrin (2g). The m.p. (226—228°) corresponded closely with that obtained [6] by the Braunschweig group (225°) for the isomer of chlorocruoroporphyrin prepared by photo-oxidation of protoporphyrin dimethyl ester; furthermore, mixed m.p. comparisons confirmed its identity with a synthetic specimen prepared by a different route.[15] The n.m.r. spectra of the two isomers showed small but significant differences, but the mass spectra, as expected, were essentially identical.

Isopemptoporphyrin dimethyl ester (2f) was also synthesised in good yield from the 2-chloroethylporphyrin (2g), by elimination of hydrogen chloride from the zinc complex with potassium t-butoxide in t-butyl alcohol. The product proved to be identical (mixed m.p.) with the earlier sample prepared in Liverpool,[8] and with another synthetic specimen kindly provided by Professor Clexy;[15] its physical properties were also identical with material prepared by a different synthesis in Nottingham.[16]

The work described in this paper thus completes the earlier studies in Liverpool [6] aimed at a full and unambiguous proof of structure for both pempto- and chlorocruoro-porphyrins. It provides a new route to the isomers which were required for biochemical studies. More recently new syntheses of pempto-, isopempto-, chlorocruoro-, and isochlorocruoro-porphyrins have been achieved from protoporphyrin-IX by degradation and manipulation of the vinyl side-chains.[17]

EXPERIMENTAL

M.p.s were determined on a hot-stage apparatus. N.m.r. spectra were determined with a Perkin-Elmer R14 100 MHz spectrometer, and mass spectra with a Varian CH5D instrument. Reactions were monitored whenever possible by t.l.c. on silica gel, and by u.v.–visible spectroscopy; column chromatography was carried out on alumina (Brockmann grade III).

Benzyl 4-Ethoxycarbonylmethyl-3,5-dimethylpyrrole-2-carboxylate.—Sodium nitrite (36 g) in water (125 ml) was slowly

[10] P. E. Sonnet, *J. Medicin Chem.*, 1971, **15**, 97.
[11] R. P. Carr, A. H. Jackson, G. W. Kenner, and G. S. Sach, *J. Chem. Soc.* (*C*), 1971, 487.
[12] *Cf.* A. M. d'A.R. Gonsalves, G. W. Kenner, and K. M. Smith, *Tetrahedron Letters*, 1972, 2203.
[13] E. J. Tarlton, S. F. MacDonald, and E. Baltazzi, *J. Amer. Chem. Soc.*, 1960, **82**, 4389.
[14] *Cf.* A. M. d'A.R. Gonsalves, G. W. Kenner, and K. M. Smith, *Chem. Comm.*, 1971, 1304.
[15] P. S. Clezy and V. Diakiw, *Austral. J. Chem.*, 1975, **28**, 1589, 2703.
[16] R. Grigg, A. W. Johnson, and M. Roche, *J. Chem. Soc.* (*C*), 1970, 1928.
[17] G. W. Kenner, J. M. E. Quirke, and K. M. Smith, personal communication.

added to a stirred solution of benzyl acetoacetate (96 g) in glacial acetic acid (150 ml), with the temperature kept below 10 °C. After 2 h in the refrigerator the solution was slowly added to a solution of ethyl 3-acetyl-4-oxopentanoate (86 g) in glacial acetic acid (100 ml), and a mixture of zinc dust (92.5 g) and anhydrous sodium acetate (92.5 g) was added simultaneously, with the temperature kept below 80 °C. When addition was complete the mixture was further heated under reflux for 1 h and then poured into water (4 l). The solid product was filtered off and recrystallised twice from methanol to yield crystals of the *pyrrole* (68 g, 44%), m.p. 79—80° (Found: C, 68.8; H, 6.6; N, 4.5. $C_{18}H_{21}NO_4$ requires C, 68.6; H, 6.7; N, 4.4%), τ ($CDCl_3$) 5.89 (q) and 8.78 (t) (OCH_2·CH_3), 6.66 (CH_2), 7.80 (5-CH_3), 7.72 (3-CH_3), 2.63 and 4.72 ($PhCH_2$), and 1.20 (NH).

Benzyl 4-(2-*Hydroxyethyl*)-3,5-*dimethylpyrrole*-2-*carboxylate* (3a).—Boron trifluoride–ether complex (64 ml) was added dropwise to sodium borohydride (12.5 g) in bis-(2-methoxyethyl) ether (25 ml), and the diborane generated was passed in a slow stream of nitrogen through a solution of benzyl 4-ethoxycarbonylmethyl-3,5-dimethylpyrrole-2-carboxylate (10 g) in tetrahydrofuran (50 ml) for 45 min. Methanol was then carefully added until effervescence ceased. The solvents were removed under reduced pressure and the *hydroxyethylpyrrole* (7.4 g, 84%) crystallised from benzene–light petroleum (b.p. 60—80 °C) as needles, m.p. 120—121° (lit.,[11] 120—121.5°), $\nu_{max.}$ (Nujol) 3 435 (OH), 3 317 (NH), and 1 670 cm^{-1} (ester C=O), τ ($CDCl_3$) 7.82 (5-CH_3), 7.72 (3-CH_3), 6.37 (t) and 7.38 (t) (CH_2CH_2), 7.92 (OH), 2.64 and 4.71 ($PhCH_2$), and 1.0br (NH).

Benzyl 4-(2-*Chloroethyl*)-3,5-*dimethylpyrrole*-2-*carboxylate* (3b).—Benzyl 4-(2-hydroxyethyl)-3,5-dimethylpyrrole-2-carboxylate (10.0 g) was dissolved in benzene (200 ml) and anhydrous potassium carbonate (20 g) was added, followed by thionyl chloride (6 ml). The mixture was heated under reflux for 3 h. The solution was filtered and evaporated to dryness under reduced pressure, and the residue recrystallised from benzene to yield the *pyrrole* (9.7 g, 91%), m.p. 121—121° (Found: C, 66.0; H, 6.2; N, 4.8. $C_{16}H_{18}ClNO_2$ requires C, 65.9; H, 6.2; N, 4.8%), τ ($CDCl_3$) 7.8 (5-CH_3), 7.72 (3-CH_3), 6.51 (t) and 6.18 (t) (CH_2·CH_2Cl), 2.66 and 4.73 ($PhCH_2$), and 1.03br (NH), *m/e* 293 (2%), 291 (6), and 91 (100).

Benzyl 5-*Acetoxymethyl*-4-(2-*chloroethyl*)-3-*methylpyrrole*-2-*carboxylate* (3c).—Benzyl 4-(2-chloroethyl)-3,5-dimethylpyrrole-2-carboxylate (7.5 g) was dissolved in glacial acetic acid (300 ml), lead tetra-acetate (126 g) was added, and the mixture was stirred for 3 h at 20 °C. It was then poured into water (300 ml) and the precipitate filtered off, washed with water, and dried *in vacuo*. Recrystallisation from methylene chloride yielded the 5-*acetoxymethylpyrrole* (8.4 g, 93%), m.p. 164—166° (Found: C, 61.8; H, 5.7; N, 4.0. $C_{18}H_{20}ClNO_4$ requires C, 61.8; H, 5.6; N, 4.3%), τ ($CDCl_3$) 7.72 (3-CH_3), 7.97 (OAc), 6.48 (t) and 7.08 (t) (CH_2·CH_2Cl), 4.98 (CH_2), 2.65 and 4.72 ($PhCH_2$), and 0.77br (NH), *m/e* 349 (3%), 351 (2), and 91 (100).

Benzyl 4-*Methylpyrrole*-2-*carboxylate* (4b).—The corresponding methyl ester (4.7 g) was dissolved in benzyl alcohol (75 ml), and sodium (0.15 g) was added in small portions. The mixture was stirred at 30 °C and 13 mmHg for 6 h and then the excess of benzyl alcohol was distilled off (60 °C and 0.6 mmHg) and the residue dissolved in ether. The solution was washed with water until the washings were neutral and then dried ($MgSO_4$). Removal of ether under reduced pressure left the crude product, which crystallised from methylene chloride–light petroleum (b.p. 40—60 °C) to yield *benzyl* 4-*methylpyrrole*-2-*carboxylate* (6.9 g, 95%), m.p. 40—42° (Found: C, 72.4; H, 6.3; N, 6.5. $C_{13}H_{13}NO_2$ requires C, 72.6; H, 6.1; N, 6.5%), τ ($CDCl_3$) 7.96 (4-CH_3), 2.70 and 4.77 ($PhCH_2$), 3.42 and 3.26 (3- and 5-H), and 0.57br (NH), *m/e* 215 (25%), 108 (4), 91 (100), and 65 (14).

3,3′-*Bis*-(2-*methoxycarbonylethyl*)-4,4′-*dimethylpyrromethane*-5,5′-*dicarbaldehyde* (6b).—(i) A suspension of pyrromethane-5,5′-dicarboxylic acid (6a) (5 g) in *NN*-dimethylformamide (20 ml) was refluxed under nitrogen for 25 min. The solution was cooled to 0 °C and maintained below 5 °C while benzoyl chloride (7.5 ml) was added carefully and as rapidly as possible. After 15 min benzene (15 ml) was added and the mixture allowed to return to room temperature. After 1 h the imine salt was filtered off and washed with benzene, and the moist solid was added to aqueous ethanol (50%; 65 ml) containing sodium carbonate (4 g). The stirred mixture was heated at 100 °C for 15 min; a benzene azeotrope distilled off. Water (65 ml) was added to the hot solution and stirring continued until the residual sodium carbonate had dissolved. Next day the precipitate was filtered off, dried *in vacuo*, and crystallised from methylene chloride to yield the pyrromethane (2.8 g, 61%), m.p. 180—181° (lit.,[18] 180—181°; lit.[19] 184—185°), τ ($CDCl_3$) 7.72 (4- and 4′-CH_3), 7.22 (t), 7.53 (t), and 6.31 (s) (CH_2·CH_2·CO_2·CH_3), 5.91 (CH_2), 0.57 (CHO), and −0.53 (NH).

(ii) Finely ground pyrromethane-5,5′-dicarboxylic acid (6a) (1.1 g) was added in small portions to trifluoroacetic acid (6 ml) and the mixture was stirred for 5 min. The solution was cooled to 0 °C and triethyl orthoformate (1.4 ml) was added. The mixture was kept at 0 °C for 5 min and then poured into water (80 ml). The precipitate was filtered off and added to a mixture of ethanol (10 ml) and aqueous M-ammonium hydroxide (20 ml). After 10 min the product was filtered off and crystallised from ethanol to yield the pyrromethane (0.65 g, 63%), identical with the product prepared as in (i).

Dibenzyl 3-(2-*Chloroethyl*)-3′,4-*dimethylpyrromethane*-5,5′-*dicarboxylate* (5a).—To a suspension of benzyl 5-acetoxymethyl-4-(2-chloroethyl)-3-methylpyrrole-2-carboxylate (2.10 g) and benzyl 4-methylpyrrole-2-carboxylate (1.29 g) in methanol (60 ml) was added toluene-*p*-sulphonic acid hydrate (60 mg), and the mixture was heated under nitrogen for 4 h at 35 °C. The crude product was filtered off, washed with ice-cold methanol, and recrystallised from methylene chloride to give the *pyrromethane* (2.40 g, 80%), m.p. 152—153° (Found: C, 68.9; H, 5.6; N, 5.5. $C_{29}H_{29}ClN_2O_4$ requires C, 68.9; H, 5.6; N, 5.5%), τ ($CDCl_3$) 7.96 (3′-CH_3), 7.73 (4-CH_3), 6.61 (t) and 7.17 (t) (CH_2·CH_2·Cl), 6.21 (CH_2), 2.76, 4.81, and 4.83 (5- and 5′-$PhCH_2$), 3.30 (4′-H), and 0.60 (NH), *m/e* 504 (3%), 506 (1%), and 91 (100).

3-(2-*Chloroethyl*)-3′,4-*dimethylpyrromethane*-5,5′-*dicarboxylic Acid* (5b).—The foregoing pyrromethane dibenzyl ester (870 mg) was dissolved in tetrahydrofuran (30 ml) and hydrogenated over 10% palladium–charcoal (800 mg) with one drop of triethylamine. The mixture was filtered through Celite and evaporated to dryness under reduced pressure to yield the *pyrromethane*-5,5′-*dicarboxylic acid* (550 mg, 98%), m.p. 175° (decomp.) (Found: C, 55.5; H, 5.6; N, 8.2. $C_{15}H_{17}ClN_2O_4$ requires C, 55.5; H, 5.3; N, 8.1%), τ (C_5D_5N) 7.90 (3′-CH_3), 7.44 (4-CH_3), 6.49 (t) and 7.06 (t) (CH_2·CH_2·Cl),

[18] R. Chong, P. S. Clezy, A. J. Liepa, and A. W. Nichol, *Austral. J. Chem.*, 1969, **22**, 229.

[19] A. H. Jackson, G. W. Kenner, and J. Wass, *J.C.S. Perkin I*, 1972, 1475.

5.77 (CH_2), 2.89 (4′-H), and −2.21 and −1.88 (NH), *m/e* 324 (0.5%), 282 (2), 280 (4), and 86 (100).

2-(2-*Chloroethyl*)-6,7-*bis*-(2-*methoxycarbonylethyl*)-1,3,5,-8-*tetramethylporphin* (2g).—3,3′-Bis-(2-methoxycarbonylethyl)-4,4′-dimethylpyrromethane-5,5′-dicarbaldehyde (0.78 g) was suspended in a solution of 3-(2-chloroethyl)-3′,4-dimethylpyrromethane-5,5′-dicarboxylic acid (0.69 g) in methylene chloride (750 ml) and, with strict exclusion of light, toluene-*p*-sulphonic acid hydrate (2.0 g) in methanol (35 ml) was added. The mixture was stirred for 24 h in the dark at 36 °C and then treated with a saturated solution of zinc acetate in methanol (35 ml). Stirring in the dark at 36 °C was continued until the Soret band had reached a maximum; the mixture was then washed with water, aqueous sodium hydrogen carbonate solution, and water again. The organic layer was dried ($MgSO_4$) and the residue obtained after removal of solvent was stirred overnight in the dark in 5% (v/v) sulphuric acid in methanol (250 ml) and then poured into methylene chloride and water. The organic phase was washed with aqueous sodium hydrogen carbonate and water, dried ($MgSO_4$), and evaporated to dryness under reduced pressure. The residue was chromatographed twice on alumina, first with methylene chloride to obtain the porphyrin fraction and secondly with methylene chloride–benzene (1 : 1). The major porphyrin fraction was evaporated to dryness and the residue crystallised from methylene chloride–methanol to yield the porphyrin (140 mg, 12%), m.p. 199—201° (lit.,[8] 199—210°), $\lambda_{max.}$ (CH_2Cl_2) 399 (ε 166 200), 496 (13 300), 529 (6 020), 568 (5 620), and 623 (3 361) nm, τ (0.1M in $CDCl_3$) 0.24, 0.24, 0.35, and 0.43 (4 *meso*-H), 1.12 (4-H), 5.79 (t), 6.83 (t), and 6.36 (s) (6- and 7-CH_2·CH_2·CO_2·CH_3), 5.8—6.0 (m) (CH_2·CH_2Cl), and 6.44, 6.56, 6.60, and 6.74 (4 ring CH_3), *m/e* 600 (63%), 602 (25), 566 (28), 527 (23), 52 (30), and 50 (100) (Found: M^+, 600.353 and 602.350. $C_{34}H_{37}ClN_4O_4$ requires *M*, 600.350 and 602.347).

A second porphyrin eluted crystallised from methylene chloride–methanol to give coproporphyrin-II tetramethyl ester (46 mg, 3.5%), m.p. 288—290° (lit., 287°), $\lambda_{max.}$ (CH_2Cl_2) 398, 498, 531, 570, and 624 nm, τ (0.1M in $CDCl_3$) 0.06 (4 *meso*-H), 5.68 (t), 6.77 (t), and 6.38 (s) (2-, 3-, 6-, and 7-CH_2·CH_2·CO_2·CH_3), 6.47 (4 ring CH_3), and 13.9 (NH), *m/e* 710 (100%) (Found: M^+, 710.329. Calc. for $C_{40}H_{46}N_4O_8$: *M*, 710.331).

6,7-*Bis*-(2-*methoxycarbonylethyl*)-1,3,5,8-*tetramethyl*-2-*vinylporphin* (*Isopemptoporphyrin Dimethyl Ester*) (2f).—2-(2-Chloroethyl)-6,7-bis-(2-methoxycarbonylethyl)-1,3,5,8-tetramethylporphin (15 mg) was dissolved in methylene chloride (6 ml) and treated with a saturated solution of zinc acetate in methanol (6 ml). After the mixture had been warmed on a water bath the colour changed to a deep red, indicating metal insertion. The solution was washed with water, dried ($MgSO_4$), and evaporated to dryness, and the residue dried under reduced pressure at 60 °C. The zinc salt was dissolved in tetrahydrofuran (0.5 ml) and 1M-potassium t-butoxide in t-butyl alcohol (8 ml) was added; the mixture was stirred in nitrogen for 5 h. The solution was then poured into ethyl acetate (57 ml) and water (75 ml) and the aqueous phase was adjusted to pH 4 with dilute sulphuric acid. After separating the organic layer, the aqueous layer was extracted with ethyl acetate and the combined extracts were washed with water, dried ($MgSO_4$), and evaporated to dryness. The zinc salt was dissolved in 5% (v/v) sulphuric acid in methanol (20 ml); the solution was kept at room temperature in the dark overnight then poured into methylene chloride (50 ml) and water (50 ml), and the aqueous phase was adjusted to pH 6 with dilute ammonium hydroxide. The organic phase was separated and the aqueous phase extracted with methylene chloride, and the combined extracts were washed with water and dried ($MgSO_4$) before evaporation to dryness under reduced pressure followed by chromatography of the residue on alumina (20 g), first with methylene chloride, and then with methylene chloride–benzene (4 : 1). The porphyrin fraction was evaporated to dryness and the residue crystallised from methylene chloride–methanol to give the porphyrin (6 mg, 43%), m.p. 218—220° (lit.,[8] 221—222°), not depressed on admixture with the sample prepared previously,[8] nor by admixture with a sample kindly provided by Professor P. S. Clezy; $\lambda_{max.}$ (CH_2Cl_2) 401 (ε 178 400), 501 (17 100), 536 (12 800), 573 (8 570), and 626 nm (4 230), τ ($CDCl_3$) 0.03, 0.09, 0.14, and 0.14 (4 *meso*-H), 1.03 (4-H), 2.7—2.9 (m) and 3.6—4.1 (m) (CH·CH_2), 5.69 (m), 6.79 (m), and 6.36 (s) (6- and 7-CH_2·CH_2·CO_2·CH_3), and 6.36, 6.44, 6.47, and 6.50 (4 ring CH_3), *m/e* 564 (100%) and 491 (25) (Found: M^+, 564.374. Calc. for $C_{34}H_{36}N_4O_4$: *M*, 564.275).

2-(2-*Chloroethyl*)-4-*formyl*-6,7-*bis*-(2-*methoxycarbonylethyl*)-1,3,5,8-*tetramethylporphin* (2h).—2-(2-Chloroethyl)-6,7-bis-(2-methoxycarbonylethyl)-1,3,5,8-tetramethylporphin (40 mg) in refluxing glacial acetic acid (20 ml) was treated with a freshly prepared solution of iron(II) acetate in acetic acid [5 ml of a solution of iron (0.5 g) in glacial acetic acid (30 ml)]. Refluxing was continued for a further 5 min and the acetic acid removed under reduced pressure. The residue was dissolved in methylene chloride (80 ml) and water (80 ml) and the organic phase washed with aqueous 10% sodium carbonate and water until the washings were neutral. The methylene chloride solution was dried ($MgSO_4$) and briefly treated with hydrogen chloride before evaporation to dryness under reduced pressure. The haem was dissolved in methylene chloride (20 ml) and treated with butyl dichloromethyl ether (2 ml) and tin(IV) chloride (0.5 ml). Stirring was continued for 30 min at 20 °C during which time the colour changed from brown to green. The mixture was poured into methylene chloride (150 ml) and water (150 ml), the organic phase was separated, and the aqueous phase was extracted with methylene chloride. The combined extracts were washed with water until the washings were neutral, dried ($MgSO_4$), and evaporated to dryness under reduced pressure. The haem obtained was dissolved in the minimum quantity of pyridine and diluted with glacial acetic acid (290 ml). A freshly prepared solution of iron(II) sulphate (1.16 g) in concentrated hydrochloric acid (11.6 ml) was added to this stirred solution at 20 °C under nitrogen. The mixture was kept under nitrogen for 15 min, during which time the colour changed from brown to violet. Then the mixture was poured into a mixture of methylene chloride (140 ml) and saturated sodium acetate solution (280 ml). The organic extracts were washed with aqueous 10% sodium carbonate followed by water, dried ($MgSO_4$), and evaporated to dryness under reduced pressure. The residue was chromatographed twice on alumina (50 g) first in methylene chloride and secondly in methylene chloride–benzene (1 : 1), and the porphyrin fraction was evaporated to dryness and crystallised from methylene chloride–methanol to give the *porphyrin* (20 mg, 48%), m.p. 233—235° (Found M^+, 628.248 and 630.240. $C_{35}H_{37}ClN_4O_5$ requires *M* 628.345 and 630.242), $\lambda_{max.}$ (CH_2Cl_2) 412 (ε 165 900), 515 (8 220), 556 (13 230), 582 (8 020), and 643 nm (1 600), τ ($CDCl_3$) −1.0, −0.27, 0.47, 0.52, and 1.27 (4 *meso*-H and CHO), 6.0—6.4

(m) (CH_2·CH_2Cl), 5.83 (m) and 6.75 (m) (CH_2·CH_2·CO), 6.31 and 6.34 (6- and 7-OCH_3), and 6.59, 6.59, 6.74, and 8.62 (4 ring CH_3), *m/e* 628 (15%), 630 (6), 594 (12), 52 (31), and 50 (100).

2-*Vinyl*-4-*formyl*-6,7-*bis*-(2-*methoxycarbonylethyl*)-1,3,5,8-*tetramethylporphin* (*Isochlorocruoroporphyrin Dimethyl Ester*) (2b).— 2-(2-Chloroethyl)-4-formyl-6,7-bis-(2-methoxycarbonylethyl)-1,3,5,8-tetramethylporphin (15 mg) was dissolved in a minimal quantity of dry methylene chloride and treated with 2,2-dimethyltrimethylene glycol (26 mg) in benzene (15 ml) and a crystal of toluene-*p*-sulphonic acid. The methylene chloride and a portion of the benzene (5 ml) were distilled from the mixture and the remaining solution was boiled under reflux for 2 h. The benzene solution was poured into methylene chloride and saturated aqueous sodium acetate and the aqueous layer extracted with methylene chloride. The combined extracts were washed with 10% sodium carbonate solution and dried ($MgSO_4$). The solvents were removed under reduced pressure and the residue dissolved in methylene chloride (3 ml). A saturated solution of zinc acetate in methanol (6 ml) was added to the solution and the mixture warmed for 4 min before pouring into ethyl acetate (25 ml) and saturated aqueous sodium acetate (25 ml). The organic layer was separated, washed with water, dried ($MgSO_4$), and evaporated to dryness. The zinc salt was dried at 60 °C and 1 mmHg for 1 h before dissolving it in tetrahydrofuran (3 ml) and adding 1M-potassium t-butoxide in t-butyl alcohol (10 ml). The mixture was stirred under nitrogen in the dark at 20 °C for 5 h, and then poured into ethyl acetate (75 ml) and water (75 ml). The aqueous phase was adjusted to pH 4 with dilute sulphuric acid and extracted with ethyl acetate. The combined extracts were washed with water and evaporated to dryness; benzene was added and evaporated off to remove traces of water. The zinc salt was dissolved in 5% (v/v) sulphuric acid in methanol (15 ml) and left overnight in the dark. The methanolic solution was poured into methylene chloride (25 ml) and water (25 ml) and the aqueous phase adjusted to pH 6 with dilute ammonium hydroxide. The organic phase was separated and the aqueous phase extracted with methylene chloride. The combined extracts were washed thoroughly with water before drying ($MgSO_4$) and evaporation to dryness. The residue was chromatographed twice on alumina (20 g) with methylene chloride as eluant. The porphyrin fraction was evaporated to dryness and crystallised from methylene chloride–methanol to give isochlorocruoroporphyrin dimethyl ester (5 mg, 36%), m.p. 228—230° (corrected) (lit.,[6] 225°), mixed m.p. with a sample (m.p. 233—255°) kindly provided by Professor P. S. Clezy 229—231° (Found: M^+, 592.266. Calc. for $C_{35}H_{36}N_4O_5$: *M*, 592.268), $\lambda_{max.}$ (CH_2Cl_2) 417 (ε 153 600), 518 (10 100), 559 (13 500), 586 (8 100), and 645 nm (2 160), *m/e* 592 (100%), 593 (38), 594 (26), 521 (12), 520 (16), and 519 (47).

We thank the S.R.C. for a maintenance grant (to P. J. O'H.) and Professor P. S. Clezy (University of New South Wales) for exchange of samples.

[6/851 *Received*, 4*th May*, 1976]