*Tetrahedron* Vol. 46, No. 22, pp. 7599–7659, 1990
Printed in Great Britain.

0040-4020/90 $5.00+.00


# THE TOTAL SYNTHESIS OF CHLOROPHYLL *a*

ROBERT BURNS WOODWARD†, WILLIAM A. AYER, JOHN M. BEATON, FRIEDRICH BICKELHAUPT, RAYMOND BONNETT*, PAUL BUCHSCHACHER, GERHARD L. CLOSS, HANS DUTLER, JOHN HANNAH, FRED P. HAUCK, SHÔ ITÔ, ALBERT LANGEMANN, EUGENE LE GOFF, WILLY LEIMGRUBER†, WALTER LWOWSKI, JÜRGEN SAUER, ZDENEK VALENTA and HEINRICH VOLZ
Converse Memorial Laboratory, Harvard University, Cambridge, Massachusetts 02138, U.S.A.

(*Received* 5 *June* 1990)

## CONTENTS



*Correspondence to this author at Department of Chemistry, Queen Mary and Westfield College, Mile End Road, London E1 4NS, U.K.
† Deceased.

**Abstract**—The total synthesis of chlorophyll *a* starting from Knorr's pyrrole (**1**) is described with full experimental detail. Forty six stages are involved to reach the target molecule, chlorin $e_6$ trimethyl ester (**46**), from which the preparation of chlorophyll *a* has already been described.

The four pyrroles which are required for rings A, B, C and D are elaborated largely by known reactions, although with considerable improvements. These pyrroles are manipulated to give two dipyrrin derivatives: a left-hand component (**26**, comprising rings A and D) and a right-hand component (the thioaldehyde **31**, comprising rings B and C). These are brought together in a carefully controlled, stepwise, condensation to give a single porphyrin product (**35**) in 50% yield. This synthesis of an unsymmetrically-substituted porphyrin bearing an electron-withdrawing substituent and a *meso*-substituent is seen as a very considerable advance, both in conceptual and practical terms, over earlier approaches. During the course of the closure of the macrocycle, intermediates which exemplify a new group of dihydroporphyrins, the phlorins (e.g. **34**), are recognised.

Eleven steps remain. The porphyrin (**35**) is shown to undergo dehydrogenation (again via a phlorin intermediate) on brief treatment with acetic acid in air to give the *meso*-acrylic acid derivative (**36**), which in acetic acid under nitrogen at 110° slowly reaches equilibrium with the purpurin (**37**). The introduction of the reactive vinyl group at C-3 has been delayed until this point in the synthesis. Photo-oxygenation of the product, the vinylpurpurin (**38**), cleaves the cyclopenteno-ring giving the methoxalylpurpurin (**39**). A reverse Claisen reaction now generates the methoxylactone, *rac*-isopurpurin 5 methyl ester (**40**), the first substance in this synthetic series which can be compared with a sample (albeit optically active) derived from natural chlorophyll *a*. *rac*-Isopurpurin 5 methyl ester (**40**) is hydrolysed to the lactol, chlorin 5 (**41**), which is resolved (diastereoisomeric salts with quinine). The less soluble salt gives synthetic *act*-chlorin 5, identical with a sample of natural provenance. Diazomethane treatment of the free acid (**42**) yields purpurin 5 dimethyl ester (**43**), again identical with the naturally-derived compound. Treatment with hydrogen cyanide in triethylamine leads to the cyanolactone (**44**), reductive cleavage and methylation of which give the chlorin $e_6$ nitrile (**45**). Methanolysis of this furnishes synthetic crystalline chlorin $e_6$ trimethyl ester (**46**), identical (mp, mixed mp, electronic spectrum, infra red spectrum) with a naturally-derived sample, so completing the total synthesis.

## 1. INTRODUCTION

This paper describes, with full experimental detail, the total synthesis of chlorophyll *a*. A preliminary communication,[1] and review articles,[2,3,4,5] have already appeared.

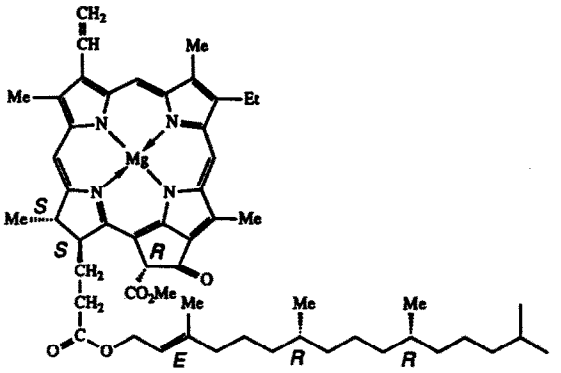
Chlorophyll has a complex and fascinating chemistry,[6] but differs from many of the complex natural substances (such as strychnine, for example) which organic chemists may choose to study in that it constantly impinges on everyday life. Although it is so common that it may often go unnoticed, it adds an important aesthetic dimension to the world we see. But it has the singular merit that, besides being beautiful, it is useful—indeed, essential. As the photosynthetic pigment, chlorophyll is directly responsible for the production of much of the food that we eat, and for all of the oxygen that we breath. Some scientists take the view that chlorophyll, or a close relative of it, had to be involved in the genesis of living things, and in the generation of an oxygen-containing atmosphere on our planet in past geological ages. What is without doubt is that, at the present time, chlorophyll photochemistry occurring in the chloroplast is the basis leading to essentially all the stored energy in the biosphere. Given a fair chance, to which the Amazonian rain forest should clearly be allowed to contribute, it helps to control atmospheric levels of carbon dioxide and so limits the 'greenhouse effect' on earth.

In historical terms the chemical study of chlorophyll falls into three main phases. At the beginning of this century Willstätter and his coworkers, in remarkable studies in an experimentally difficult area, isolated chlorophyll *a* as a pure substance, established the molecular formula as $C_{55}H_{72}N_4O_5Mg$, and carried out various transformations.[7] In a second phase the work was continued in the thirties by Stoll, by Conant, and by Hans Fischer, the last with particular vigour. Fischer charted a large number of chemical reactions in the chlorophyll series, and, by a classical iterative process,[8] arrived at the chemical structure[9,10] for chlorophyll which, while it once attracted some scepticism, is now seen to have stood the test of time.

In the forties, progress was again interrupted by war, and by Fischer's death. In the third phase the problems of stereochemistry, which had earlier very reasonably been put to one side, were solved. The *trans*-stereochemistry of ring D was established by Linstead,[11] and the absolute stereo-

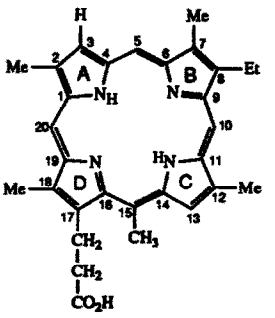
chemistry of the phytyl side chain [2′-*trans* (7′*R*, 11′*R*)] was determined by Weedon[12] at much the same time that the work here described was in progress.

The absolute stereochemistry at ring D and the isocyclic ring was not then known. From the point of view of synthesis this was not an impediment, and, indeed, in the preliminary communication[1] the unnatural enantiomer of (for example) chlorin $e_6$ trimethyl ester was drawn. However the absolute stereochemistries at C-13$^2$, C-17 and C-18 have subsequently been determined as 13$^2$-*R*, 17-*S*, 18-*S* by proton NMR studies on pheophorbides with chiral 13$^2$-alkoxy groups,[13]* by Horeau analysis of 13$^1$-hydroxypheophorbides,[14] by chemical correlation[15] of ring D oxidation products with degradation products from (−)α-santonin (of known absolute stereochemistry), and by X-ray structure analysis of ethyl chlorophyllide *a*.[16] The upshot of all this effort is that the structure of chlorophyll *a*, written now with correct absolute stereochemical detail, is as shown in (**101**). This was the overall synthetic objective.



(**101**) Chlorophyll *a*

During the 1930s Fischer devised a plan for the synthesis of chlorophyll *a*, which was continued after World War II by Strell and his colleagues. It suffered from various unsatisfactory steps (e.g. the synthesis of 3-deethylphylloporphyrin (**102**) in 0.6% yield is accompanied by many other porphyrins[17]) and ambiguities (e.g. the degree of regiospecificity of the reduction of 3-deethylphylloporphyrin with sodium in boiling amyl alcohol to give the corresponding chlorin[18]). Often yields are not stated, and a resolution of a racemic synthetic compound to reach the natural series has not been carried out. The approach has been reviewed,[4,6] and does not merit further attention here.



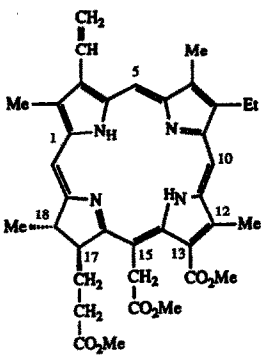
(**102**) 3-De-ethylphylloporphyrin

* For meaning of superscripts see structure (**103**).

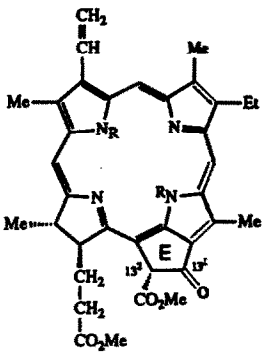
The nomenclature used in this paper follows IUPAC–IUB recommendations.[19] Fischer trivial names are used where appropriate, but no new trivial names for specific compounds are introduced. The numbering scheme and ring designations used in the paper follow the semi-systematic pattern with the structure orientated in the conventional way, as illustrated for (**102**). In the experimental section each compound is named once using the full systematic name (see p. 7635). Synthetic intermediates on the direct synthetic route are numbered (**1**) to (**46**); all other compounds are numbered starting at (**101**).

## 2. SYNTHETIC TARGET AND ITS APPROACH

During the extensive earlier work on chlorophyll *a* referred to above, a number of partial syntheses had been carried out. A consideration of these now allowed the overall synthetic objective (**101**) to be simplified to give the operational synthetic target. This emerged as chlorin $e_6$ trimethyl ester (**46**).



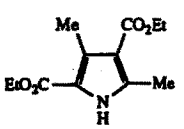
(**46**) Chlorin $e_6$ trimethyl ester



(**103**) R = H, methyl pheophorbide *a*
(**104**) 2R = Mg, methyl chlorophyllide *a*

Thus Fischer had shown that Dieckmann cyclisation of chlorin $e_6$ trimethyl ester with methanolic potassium hydroxide in pyridine[20] or with sodium methoxide in methanol–acetone[21] gives methyl pheophorbide *a* (**103**). Two long established pathways of partial synthesis are then available. One, outlined by Stoll and Wiedemann,[6] proceeds by transesterification to pheophytin *a*, followed by metallation[22] to chlorophyll *a*. Since the present work was completed, Smith and Lewis,[22a] in developing an alternative partial synthesis, have improved this sequence of reactions. The other pathway reverses the steps: magnesium is introduced into methyl pheophorbide *a* (**103**) with the ethyl magnesium bromide–ethanol reagent[23] in pyridine–ether under nitrogen to give the magnesium complex, methyl chlorophyllide *a* (**104**), and this is followed by the enzymatic introduction[7,24] of the phytyl residue to give chlorophyll *a* (**101**). Finally, phytol of the correct absolute stereochemistry has been synthesised by Weedon and his colleagues.[12]

With the target molecule defined in this way as (**46**) it was now necessary to develop strategy. Chlorin $e_6$ trimethyl ester (**46**) is evidently made up of units of the pyrrole type, and there was a large body of established pyrrole chemistry[10] with which to construct the individual building blocks for rings A, B, C and D. In the event (see Fig. 1) they were all elaborated from a single pyrrole, Knorr's pyrrole, (**1**), which was made in large quantity (several kilograms). What was not available



(**1**)

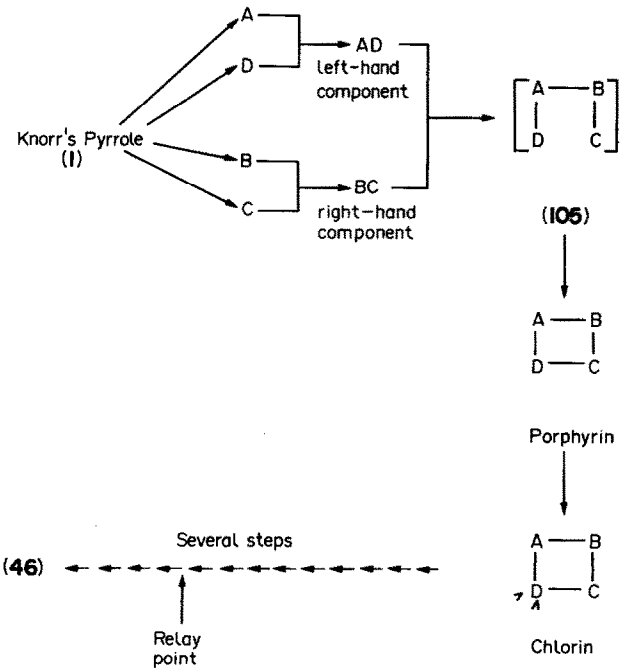



Fig. 1. General strategy for the synthesis of chlorin $e_6$ trimethyl ester (**46**).

was a porphyrin (or chlorin) synthesis which gave good yields with unsymmetrically-substituted systems when electron-withdrawing $\beta$-substituents were present. Since methods for regioselective hydrogen transfer had to be discovered, and several delicate steps would be necessary before the target molecule was reached, it was essential to have an adequate amount of porphyrin. Consequently, a new type of synthesis would have to be devised which was regioselective at the ring closure stage to ensure high yields and avoid porphyrin mixtures which would be difficult to separate and which, when separated, had to be identified one by one. This regioselective closure was to involve a linear tetrapyrrole intermediate (**105**). This was the first example of an approach the general principle of which has subsequently been widely developed and applied.[25]

The exact way in which the chlorin was to be formed could not be known at the outset. This had to await the development of a successful porphyrin synthesis which was thus central to the entire effort. However, certain considerations relevant to the generation and properties of the chlorin system, as found in chlorophyll derivatives, were developed at an early stage, and it is appropriate to consider these now.

2.1. *Peripheral overcrowding effect*

A feature of a precursor for chlorin $e_6$ trimethyl ester (**46**) is likely to be a porphyrin which (like **46**) has substituents at all the available positions in the lower part of the molecule (i.e. 12, 13, 15, 17, 18). Since the porphyrin system is aromatic and, though relatively easily deformed, tends to be approximately planar, this means that this part of the molecule is under severe steric constraint, as illustrated diagramatically in Fig. 2.

This steric compression might be relieved in various ways. Firstly, by the simple extrusion of a substituent, illustrated by many examples in the chemistry of the chlorophyll-related porphyrins.[10] For example,[26] treatment of chloroporphyrin $e_6$ trimethyl ester (**106**) with hydrogen iodide in acetic

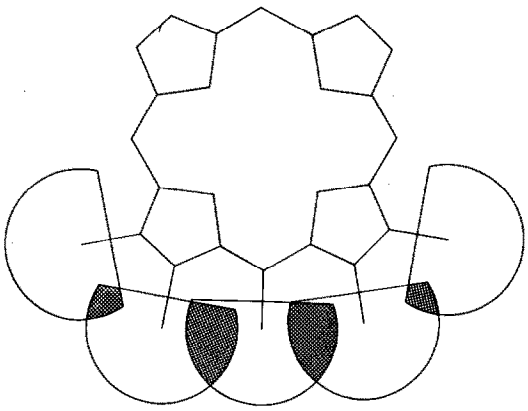
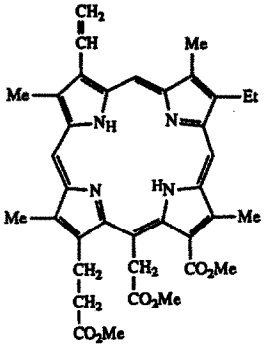
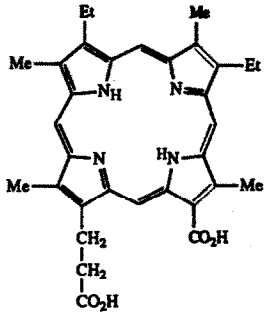


Fig. 2. Diagram to illustrate steric interaction in the 12,13,15,17,18-pentasubstituted porphyrin system. (From the original drawing by RBW).

acid gives rhodoporphyrin (**107**, as a monomethyl ester), with the remarkable (but see later) loss of the 15-acetic acid residue.




(106) Chloroporphyrin $e_6$ trimethyl ester




(107) Rhodoporphyrin

Secondly the non-bonded interaction could be reduced by forging a bond between two of the peripheral substituents, as illustrated in Fig. 3. Again, there are many examples of cyclisation to be

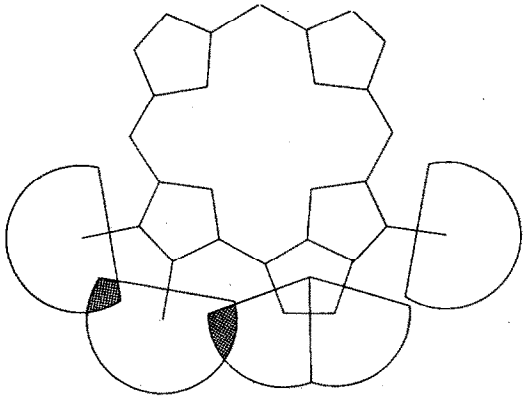
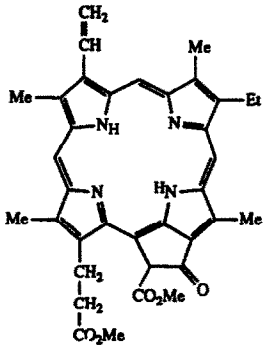


Fig. 3. Diagram to illustrate relief of steric interaction in 12,13,15,17,18-pentasubstituted porphyrin by cyclisation. (From the original drawing by RBW).

found in the chlorophyll-related porphyrins, especially where the *meso*-substituent is involved. Thus, heating chloroporphyrin $e_6$ trimethyl ester (**106**) in acetic acid at *ca.* 100° for two hours gives a quantitative yield of the cyclised product (**108**, pheoporphyrin $a_5$ dimethyl ester).[27]



(108) Pheoporphyrin $a_5$ dimethyl ester

The third way in which the peripheral steric interaction might be diminished would be to generate a chlorin, with ring D (or ring C) reduced, thus displacing two of the five substituents out of the plane, as illustrated in Fig. 4. The stability conferred by this change is perhaps best illustrated by

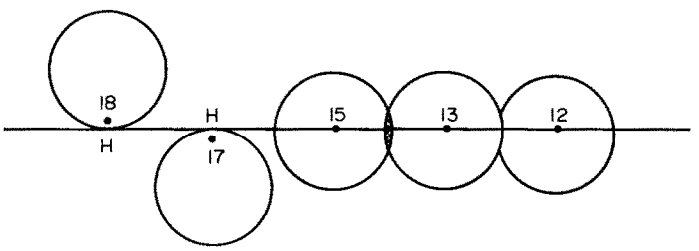
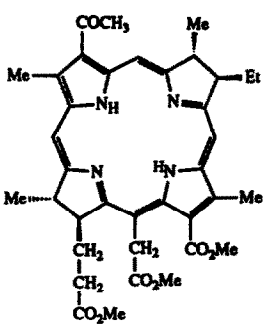
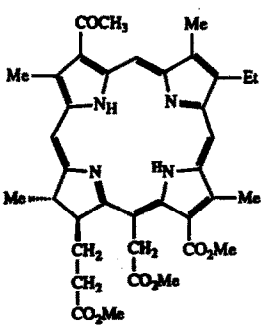



Fig. 4. Diagram to illustrate relief of steric compression in 12,13,15,17,18-pentasubstituted porphyrin by reduction of β-positions of one of the five membered rings (ring D illustrated).

the reverse process, since *meso*-substitution adjacent to the reduced ring is known to make the dehydrogenation of chlorin to porphyrin more difficult. For example, the dehydrogenation of bacteriochlorin $e_6$ trimethyl ester (**109**) with quinones occurs at room temperature to dehydrogenate



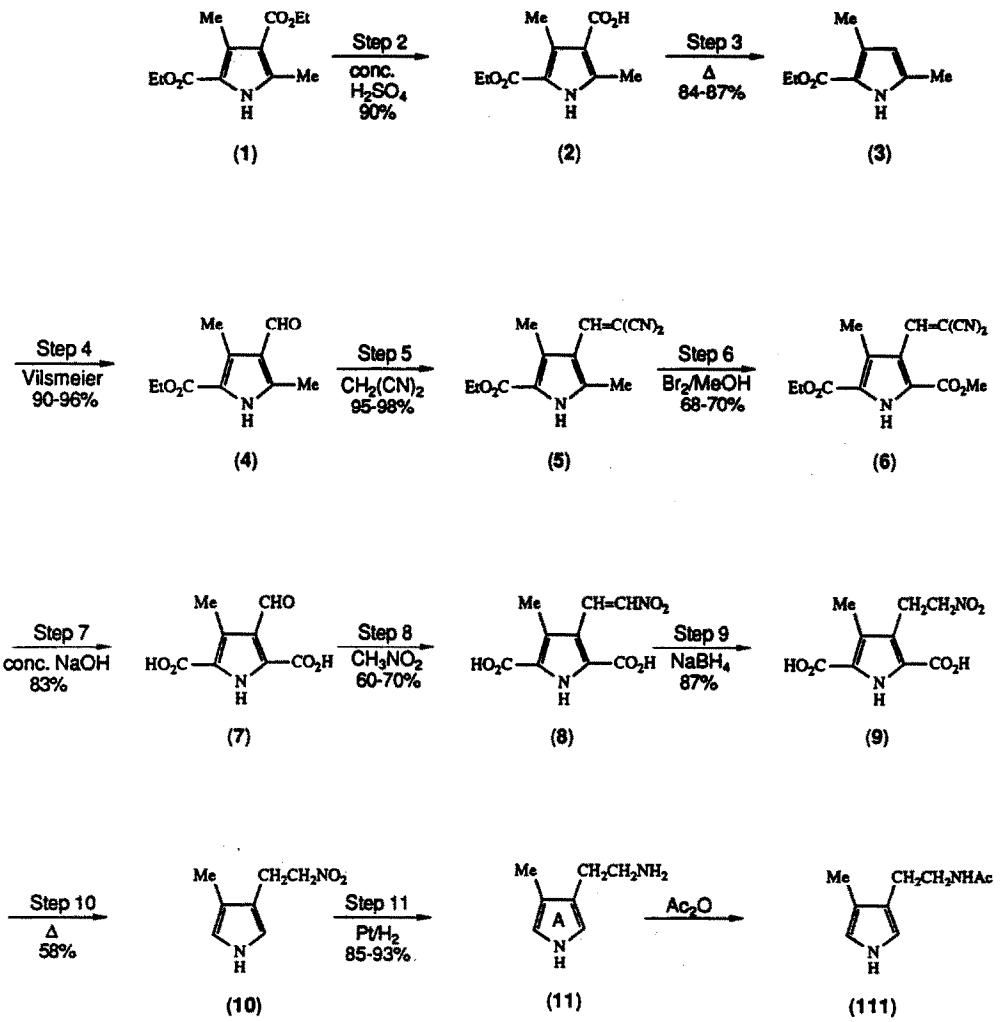
(109) Bacteriochlorin $e_6$ trimethyl ester



(110)

ring B, that is, the ring without flanking *meso*-substituents, to give the chlorin (**110**): dehydrogenation of this to give the corresponding porphyrin requires more vigorous conditions,[28] the difference being attributed to the extra steric compression caused by the 15-acetic acid residue in the porphyrin. Again, 5-chlorooctaethylchlorin is dehydrogenated to the porphyrin with DDQ (chloroform at room temperature) but the corresponding 5,20-dichlorochlorin is unaffected under these conditions.[29]

This third factor led us to anticipate that, with a properly constructed porphyrin in hand, we could find a suitable route to transfer hydrogens to the desired site. It was evident that the naturally-occurring chlorophyll *a* (**101**) had a stereochemistry ($13^2$-17-transoid, 17-18-transoid) which minimised the peripheral overcrowding effect of substituents associated with the chiral centres. We needed to generate only those at C-17 and C-18, and, provided that these could be set up under equilibrating conditions, we could reasonably expect to be able to arrive at the thermodynamically more stable transoid geometry.




Scheme 1. Synthesis of ring A of chlorophyll *a*.

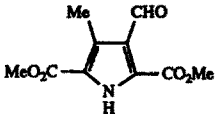
### 3. RINGS A, B, C, D

The synthesis was based on one starting point, Knorr's pyrrole (Fig. 1), and proceeded through twenty five steps to construct the four rings A, B, C, D, needed to make the dipyrrin derivatives (**26**) and (**27**). Of these twenty five steps, only five were entirely new; the rest relied heavily on earlier work, especially that of Fischer,[10] although considerable improvements were made to many of the steps, and these are therefore recorded.

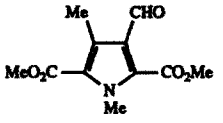
Knorr's pyrrole (**1**) was prepared from ethyl acetoacetate by a variant of MacDonald's modification[30] of Knorr's original synthesis.[31] It was necessary to make a large quantity of this substance, and the procedure used proved adaptable to the pilot plant scale so that several kilograms of (**1**) were available for our subsequent work.[32]

#### 3.1. *Ring A*

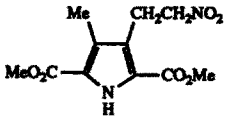
Extensive preliminary experimentation had led to the conclusion that the regioselectivity in the B–C linkage would require a pyrrole with a β-aminoethyl substituent, as in (**11**), which would fulfil a two-fold function since it was also destined to emerge as the vinyl group at C-3. Pyrrole (**11**) was prepared as shown in Scheme 1. Selective hydrolysis of Knorr's pyrrole (**1**) in concentrated sulphuric acid[33] gave the β-monocarboxylic acid (**2**), which was decarboxylated thermally in ethanolamine[34] giving (**3**). Vilsmeier formylation[34] now produced the aldehyde (**4**) in excellent yield on a kilogram scale. Fischer and Weiss[35] had carried out the reaction (**4**) → (**5**) with malononitrile in acetic anhydride; we found that this condensation was more conveniently effected in warm absolute ethanol with ethylamine as catalyst. The conversion of the α-methyl group to α-methoxycarbonyl (**5** → **6**) in a one pot reaction with bromine and methanol had been described by Fischer and Zeile,[36] and it was found possible, with care, to scale up this reaction to give consistent yields of 68–70% on a 100–500 g scale. Fischer and Zeile had also reported the hydrolysis and cleavage of the dicyanovinyl diester (**6**) on a small scale with a large volume of 0.1 M-sodium hydroxide. This procedure was impractical for larger quantities, and in any case the reaction under these conditions was slow. However, hydrolysis and cleavage using concentrated aqueous sodium hydroxide gave (**7**), obtained as a fawn powder which was characterised as the colourless dimethyl ester (**112**) formed with a slight excess of diazomethane (excess diazomethane gave the N-methyl derivative **113**).




(**112**)



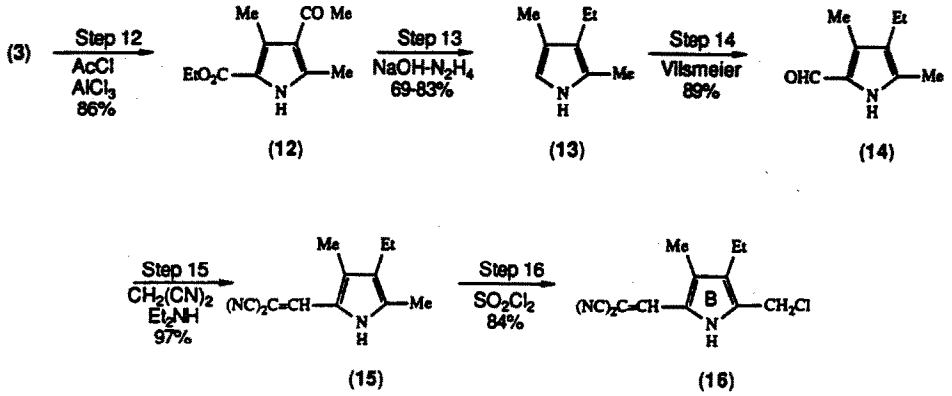

(**113**)




(**114**)

Condensation of the aldehyde (**7**) with nitromethane in the presence of diethylamine gave (*ca.* 65%) the β-nitrovinyl pyrrole (**8**), which was directly reduced by sodium borohydride (87% yield) to the corresponding β-nitroethyl compound (**9**). The latter was characterised as its dimethyl ester (**114**), colourless needles, mp 125.5–126°, the electronic spectrum of which was very similar to that of 3-ethyl-2,5-di(ethoxycarbonyl)-4-methylpyrrole [$\lambda_{max}$ (EtOH) 222 (17 700), 282 nm (21 900)].

It was now necessary to remove the carboxylic acid functions, generating β,β′-dialkylpyrrole systems which needed careful manipulation and storage. The decarboxylation was achieved in a sodium acetate–potassium acetate melt under nitrogen to give the α-free pyrrole (**10**) as a colourless oil in 58% yield, and this was catalytically reduced to the ring A intermediate (**11**) in excellent yield. The β-aminoethyl compound (**11**) was characterised as its N-acetyl derivative (**111**). The sequence (**1**) → (**11**) was the longest of the preliminary pathways to the corner pyrrole units, and had been achieved in an overall yield of 14%.



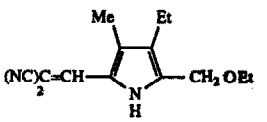

Scheme 2. Synthesis of ring B.

3.2. *Ring B*

Ring B has simple alkyl $\beta$-substituents, but required a protected $\alpha$-aldehyde function which would later feature, it was hoped, in the regiospecific linking of rings A and B. The system sought is shown at (**16**).

The route, shown in Scheme 2, included kryptopyrrole (**13**), and normally this would be approached by a classical Knorr synthesis (ethyl acetoacetate and pentan-2,4-dione)[37] or by the Kleinspehn–Johnson modification (benzyl acetoacetate and 3-ethyl-pentan-2,4-dione).[38] In our circumstances the availability of large quantities of Knorr's pyrrole (**1**) and thence of (**3**), led to us using a different route. The pyrrole (**3**) was acetylated[39] with acetyl chloride in dichloromethane in the presence of aluminium (III) chloride giving the acetyl derivative (**12**) in 86% yield. Wolff-Kishner reduction with concomitant hydrolysis and decarboxylation gave kryptopyrrole (**13**).[40] Vilsmeier formylation gave the $\alpha$-formylpyrrole (**14**). The formyl group was protected as the dicyanovinyl derivative, using the preparative method of Fischer and Neber.[41] Finally, the product (**15**) of this step was prepared for linkage with ring C (to give the dipyrrylmethane derivative **27**) by converting the $\alpha$-methyl function to $\alpha$-chloromethyl using sulphuryl chloride as halogenating agent to give (**16**) in good yield. Interestingly, we found that the chloromethyl derivative (**16**) was particularly light sensitive, but the corresponding ethoxymethyl derivative (**115**) was not.

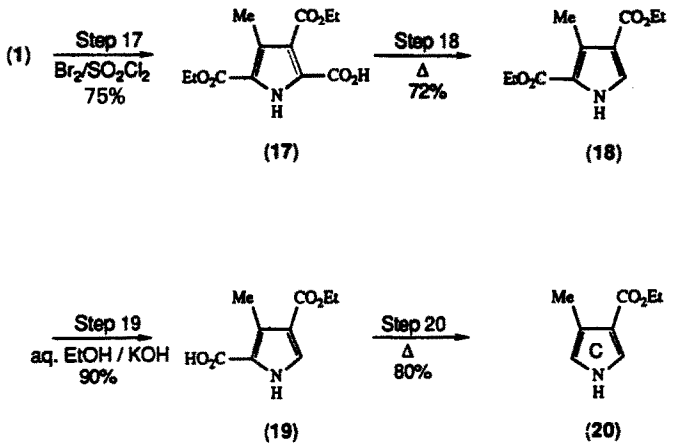
The overall yield (**3**) → (**16**) was 52%.




(**115**)

3.3. *Ring C*

The ring C unit, 3-ethoxycarbonyl-4-methylpyrrole (**20**) was first made by Fischer and Wiedemann,[42] who condensed aminoacetone and ethyl oxaloacetate to obtain 3-ethoxycarbonyl-4-methylpyrrole-2-carboxylic acid, which was then decarboxylated. Later Corwin[43] and Treibs[44] prepared the ester (**20**) by hydrolysis of (**17**) to the 2,5-dicarboxylic acid, which was subjected to iodinative decarboxylation, followed by removal of the 2,5-iodo substituents by catalytic hydrogenation. In our hands, Corwin's method gave excellent results, but was considerably less convenient, particularly




Scheme 3. Synthesis of ring C.

on a large scale, than that outlined in Scheme 3. More recently Le Goff[45] has shown that cycloaddition of *p*-tosylmethylisocyanide to ethyl crotonate offers a direct route to (**20**) which would be particularly convenient for small scale preparations.

Regioselective oxidation of the α-methyl group of Knorr's pyrrole (**1**) occurred, as described by Corwin,[46] with bromine–sulphuryl chloride to give the monocarboxylic acid (**17**) which (contrary to Corwin's reports[46,47]) can be smoothly decarboxylated (copper bronze, 230–240°) giving (**18**). Hydrolysis with strong base[48] again caused a regioselective reaction to give (**19**) which was decarboxylated as before to give the α-free ring C unit (**20**). The decarboxylation with copper bronze is much more effective and convenient than that using glycerol[48] as the thermal support.

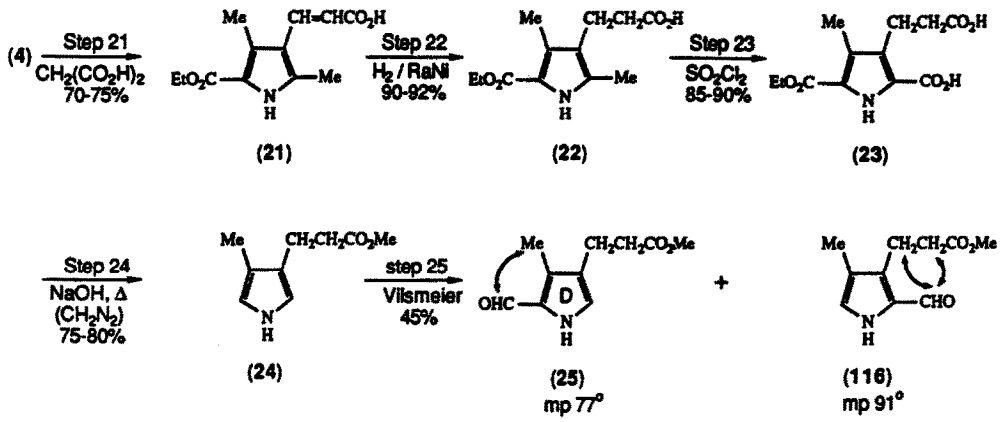
The overall yield (**1**) → (**20**) was 39%.

### 3.4. *Ring D*

It was proposed that rings A and D should be linked together (Fig. 1) in the form of a pyrromethene salt (**26**) and hence the ring D unit required was the formyl pyrrole (**25**) which, even with two β-alkyl substituents, was expected to be a relatively stable compound.

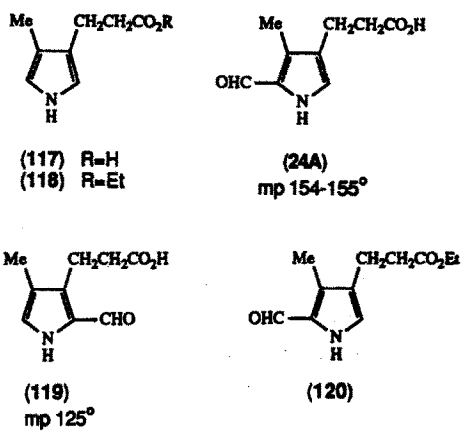
The route is shown in Scheme 4. The β-acrylic acid (**21**) was prepared from the β-formylpyrrole (**4**) using the Knoevenagel condensation (malonic acid, aniline) following Fischer and Andersag.[49] Fischer had reduced this to (**22**) using sodium amalgam,[49] but we found that the catalytic reduction described by MacDonald[50] was more convenient. Various esters of the acid (**22**) are available by direct ring synthesis from acetoacetates and 4-acetyl-5-oxo-hexanoates[38,51] but for present purposes we needed to take maximum benefit from the large quantities of (**1**), and hence (**4**), which were available.

The β-propionic acid (**22**) had earlier[44,52] been oxidised to the α-carboxylic acid (**23**) using a two-step procedure (conversion to the α-bromomethylpyrrole, followed by treatment with sulphuryl chloride, and hydrolysis). However, we experienced no difficulty in carrying out the conversion directly with sulphuryl chloride to give the α-carboxylic acid (**23**) in yields approaching 90%. In the removal of the α-substituents of (**23**) under alkaline conditions we again followed Fischer.[52,53] The reaction requires an autoclave at 180–185°, and the temperature is critical: a reaction at 170° resulted in the hydrolysis of the α-ethoxycarbonyl group, but decarboxylation was not effected. Treatment with diazomethane was needed to give the methyl ester (**24**). Alternatively the free acid (**117**, opsopyrrole carboxylic acid) could be isolated, or, with diazoethane, the corresponding ethyl ester (**118**) was obtained.



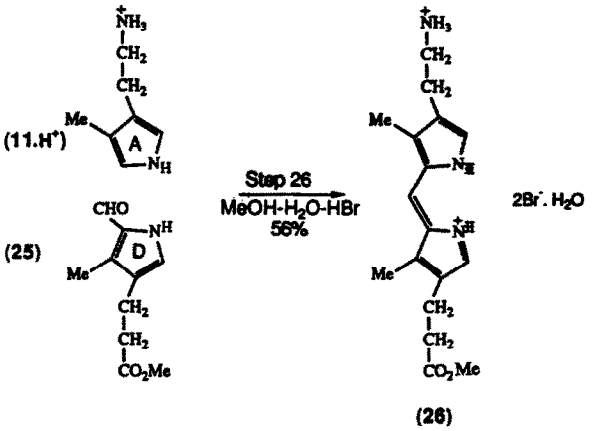

Scheme 4. Synthesis of ring D. The arrows on structures (**25**) and (**116**) refer to observed nuclear Overhauser enhancement effects.

The final step, a Vilsmeier formylation, was regioselective but not regiospecific. Two products were obtained—the required 5-formyl compound (**25**) and its 2-formyl isomer (**116**) in the ratio of about 4 : 1. The formation of the second component (**116**) had not been previously recognised:[54] the regioselectivity is attributed to the extra steric bulk of the β-methoxycarbonylethyl function (compared with methyl) causing C-2 to be relatively more hindered towards the approach of the electrophile, but evidently the effect is not overwhelming.




The separation of the isomeric substances was achieved by taking advantage of the lower solubility in water of the required acid (**24A**) compared with that of the isomeric acid (**119**), followed by methylation with diazomethane. The corresponding ethyl ester (**120**) is best made by esterification of the major acid (**24A**) with diazoethane. The assignment of structure to the individual isomers has been confirmed by nOe observations. For the 5-formyl ester (**25**), irradiation at the formyl signal (δ 9.60) gives a positive difference signal at δ 2.30 (the C-4 methyl group), whereas for the 2-formyl isomer (**116**) irradiation at δ 9.62 shows no effect in the region of the C-4 methyl singlet (δ 2.06) but a strong positive difference signal at δ 3.05 and a weak one at δ 2.58, assigned to the 3′ and 3″ methylenes, respectively, of the propionic acid ester side chain.

This separation of isomers depressed the overall yield of this sequence (**4** → **25**) to 22%, but was, of course, essential for the stages which were to follow.



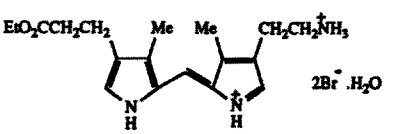

Scheme 5. Synthesis of left-hand component.

## 4. LEFT HAND, RIGHT HAND

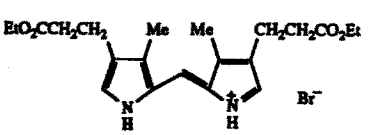
The left-hand component was to be the pyrromethene salt (**26**) shown in Scheme 5. The preparation of pyrromethenes by the acid-catalysed condensation of an α-formylpyrrole with an α-free pyrrole is in general a rapid and smooth reaction. However, in our experience, which includes many examples not described here, success depends on getting the methene out of the reaction medium in a crystalline form as rapidly as possible. After one or two recrystallisations (e.g. from methanol–HBr) the methene salts are pure, and when pure are stable. Crude materials, by contrast, deteriorate rapidly, and where the methene salt does not come rapidly out of solution it is difficult to isolate the desired compound by subsequent manipulative or separation procedures, even where spectroscopic examination reveals that a considerable amount of methene has been formed. These difficulties are perhaps to be attributed to oligomerisation processes, especially when, as here, free α-positions are present.

Continuing in a practical vein, two other points may be noted. Since the acidity of freshly-prepared hydrogen bromide in methanol decreases rapidly, the solution should be titrated directly before use. Secondly, the methene salts decompose rather than melt and the decomposition point depends on the rate of heating and the temperature at which gradual heating is commenced. Hence, in this series, melting behaviour is not a particularly useful parameter for characterisation purposes.

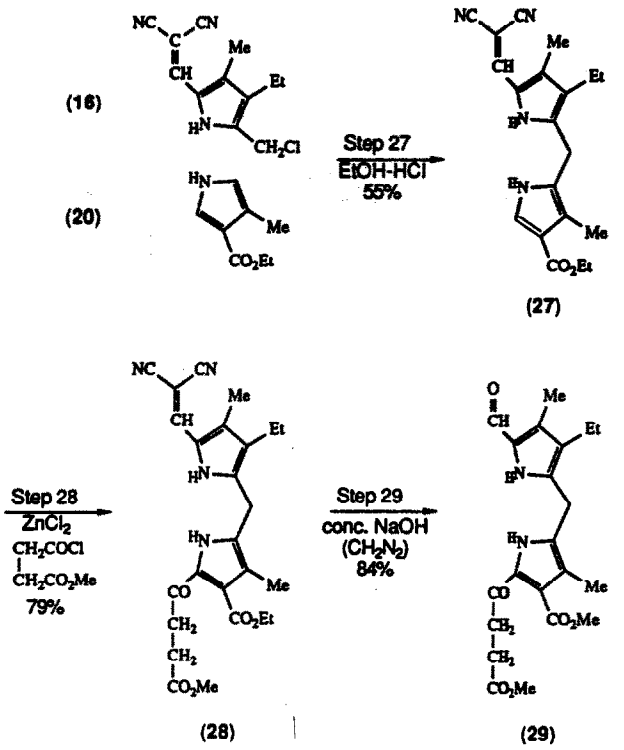
The ring A component (**11**) and the ring D component (**25**) were reacted in methanolic hydrogen bromide containing a small proportion of water at −25°. The orange pyrromethene dihydrobromide (**26**) crystallised out and was recrystallised (56% yield) from aqueous methanolic hydrogen bromide. The corresponding ethyl ester (**121**) was prepared analogously from (**11**) and the aldehyde (**120**). Similarly reaction of (**118**) with aldehyde (**120**) gave the symmetrically substituted pyrromethene salt (**122**).




(**121**)




(**122**)

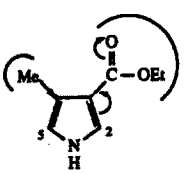



Scheme 6. Generation of the initial version of the right-hand component.

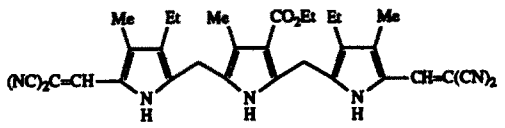
As far as we can ascertain these reactions are all regiospecific as drawn. We suppose that not only is a steric factor directing the reaction (as in the case above, **24** → **25**) but that, for (**26**) and (**121**), the amine function will be protonated under the reaction conditions and will exert a strongly inhibiting electrostatic effect on electrophilic approach towards the adjacent α-position.

Synthesis of the right-hand component went through a series of later modifications to perfect the regiospecificity of the A–B closure (Fig. 1), but the initial objective was the dipyrrylmethane (dipyrrane, **29**) which had carbonyl groups of different reactivity at C-1 and C-9 (Scheme 6).

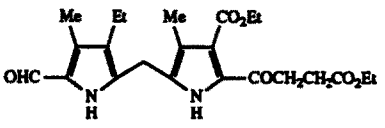
Condensation of the α-chloromethylpyrrole (**16**) with the ring C unit (**20**) in acidic ethanol gave the dipyrrylmethane (**27**) in 55% yield. In the two earlier cases where alternative α-positions were available the reaction was regioselective when controlled by a single factor (a steric effect, **24** → **25** + **116**) but apparently regiospecific when controlled by a steric effect and an electrostatic one (**11** → **26**). In the present step we suppose that both steric and electronic effects are operating: the ethoxycarbonyl group has a greater bulk than methyl; at the same time it is deactivating C-2, while the methyl group is activating C-5 (**123**, arrows). A second isomer was not detected and, if




(**123**)



(124)



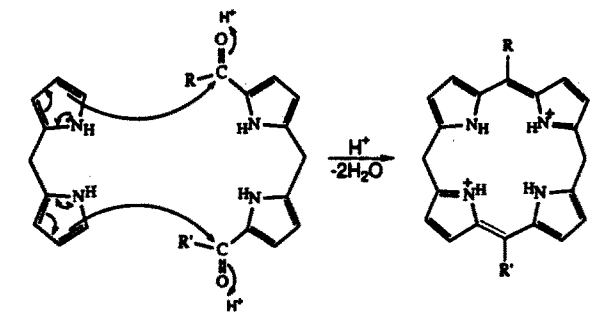
(125)

formed, it must have been a very minor product. However, that electrophilic attack can subsequently occur at C-2 of 3-ethoxycarbonyl-4-methylpyrrole was shown by the isolation of the tripyrrane (**124**) as a by-product (10%). This substance was light sensitive and exhibited a pronounced greenish fluorescence which permitted its detection even in small amount. The structure of the tripyrrane was confirmed by its deliberate synthesis from the dipyrrylmethane (**27**) and excess of the chloromethyl pyrrole (**16**).

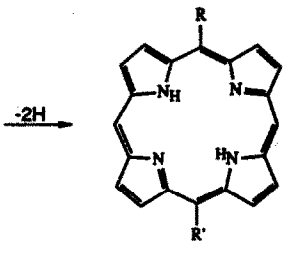
The dipyrrylmethane (**27**) was reacted in Friedel–Crafts fashion ($ZnCl_2$) with $\beta$-methoxycarbonylpropionyl chloride giving the ketone (**28**) in 79% yield. The structure of this substance was confirmed by its NMR spectrum: irradiation at the *meso*-methylene signal ($\delta$ 4.00) caused nuclear Overhauser enhancement of signals due to the imino, methyl, and ethyl protons nearby. Cleavage of the protecting dicyanovinyl function with *ca.* 30% aqueous sodium hydroxide at 95° under nitrogen gave, after treatment with diazomethane in methanolic ether, 84% of the colourless ketoaldehyde (**29**). The corresponding diethyl ester (**125**) was obtained in an analogous fashion, but using diazoethane in ethanolic ether.

## 5. MACROCYCLISATION

The critical porphyrin synthesis was now at hand. The concept behind our approach was to employ a two-fold pyrromethene condensation between an $\alpha,\alpha'$-free dipyrrylmethane (**126**) and an $\alpha,\alpha'$-dicarbonyldipyrrylmethane (**127**) as illustrated in Scheme 7. This was expected to lead to



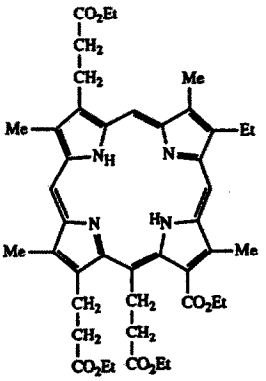
(126) (127) (128)



(129)

Scheme 7. Scheme for proposed macrocyclic synthesis.

the 5,15-dihydroporphyrin salt (**128**) which on dehydrogenation would give the porphyrin (**129**). Preliminary experiments showed that this scheme of porphyrin synthesis worked reasonably well even when an electron-withdrawing substituent was present on the ring. Thus catalytic reduction of the symmetrically-substituted pyrromethene salt (**122**) gave the colourless dipyrrylmethane which was immediately condensed under hydrogen with the dipyrrylmethane diethyl ester (**125**) in hydrogen bromide-acetic acid. The intermediate dihydro compound (corresponding to **128**, but see later) was then oxidised with iodine giving the porphyrin tetraethyl ester (**130**) in 40% yield. This is clearly a very respectable yield, especially when compared with those obtained using previous routes to compounds of this type.[10]



**(130)**

This general synthesis had been independently discovered by MacDonald and his colleagues[55] for the case where R = R[1] = H (Scheme 7) and is now conveniently referred to by his name.

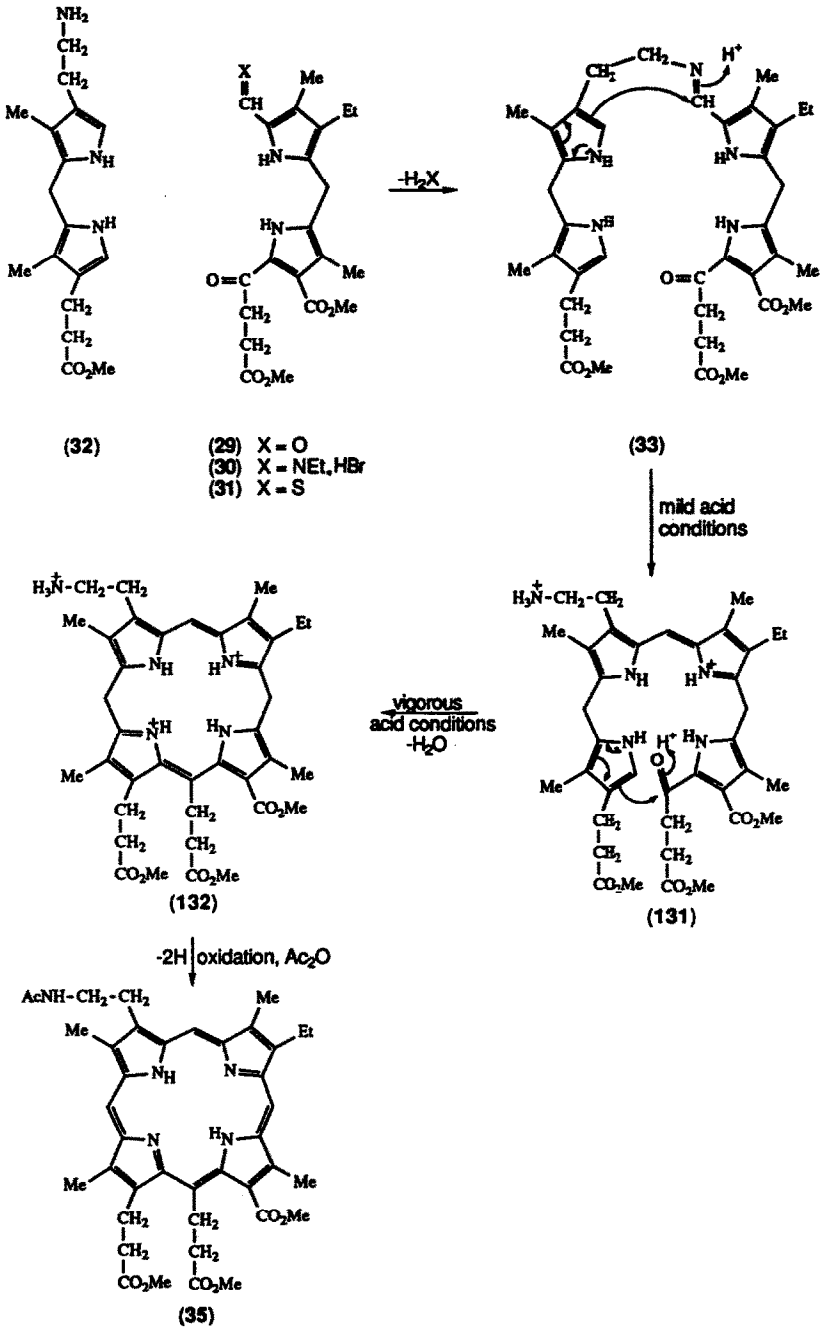
However, the synthetic approach in Scheme 7, although it works well for the case where either the left-hand component (as in **130**) or the right-hand component is symmetrically $\beta$-substituted, is not satisfactory for a porphyrin which, on retrosynthetic analysis, does not lead to a dipyrrane with this property. A porphyrin designed as a precursor for chlorin $e_6$ trimethyl ester necessarily lacks symmetry in this sense, and a new approach is needed.

Our plan was to modify the chemistry shown in Scheme 7 so that ring A had a $\beta$-aminoethyl substituent (as in **32**), while the reactivities of ring B and ring C were differentiated by making ring B an aldehyde (R = H) and ring C a ketone (R′ = $CH_2CH_2CO_2Me$) as in (**29**) (Scheme 8).

It was envisaged that acid-catalysed *aldo*-azomethine formation would occur regiospecifically to give (**33**), which is set up to undergo a second regiospecific condensation (**33**, arrows) thus generating a single bilene-*b* (**131**) with the correct substitution pattern. (This bilene-*b* corresponds to the linear tetrapyrrole represented in an abstract way as (**105**) in Fig. 1). More forcing acid conditions were then expected to cause a second condensation between rings C and D (**131**, arrows) thus closing the macrocycle in the desired way and leading to (**132**), which could be dehydrogenated to the desired porphyrin (**35**).

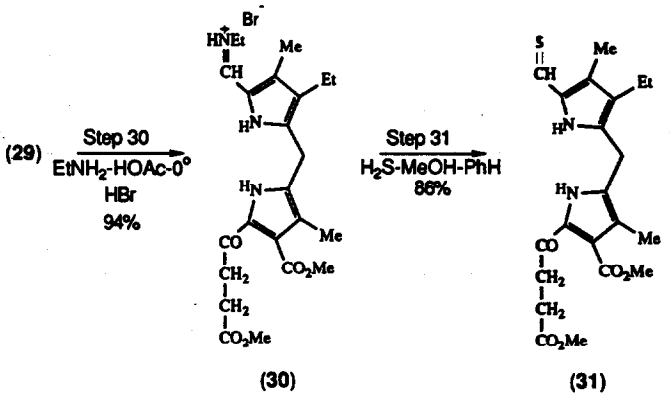
After much experimentation it emerged that this differentiation (aldehyde versus ketone) in reactivity was insufficiently great. It was found that the mild acidic conditions required to generate the azomethine (**33**) from the aldehyde (**29**) and the amine (**32**) caused the latter to be destroyed. Such dipyrrylmethanes without electron-withdrawing substituents are known to be prone to dismutation and pyrrole-exchange reactions, and are particularly fragile when both $\alpha$-positions are unsubstituted.[56]

The reactivity of the aldehyde function of (**29**) was therefore modified by making the Schiff's base (**30**) and carrying out the amine exchange reaction (**32** + **30** → **33**). This reaction was shown to



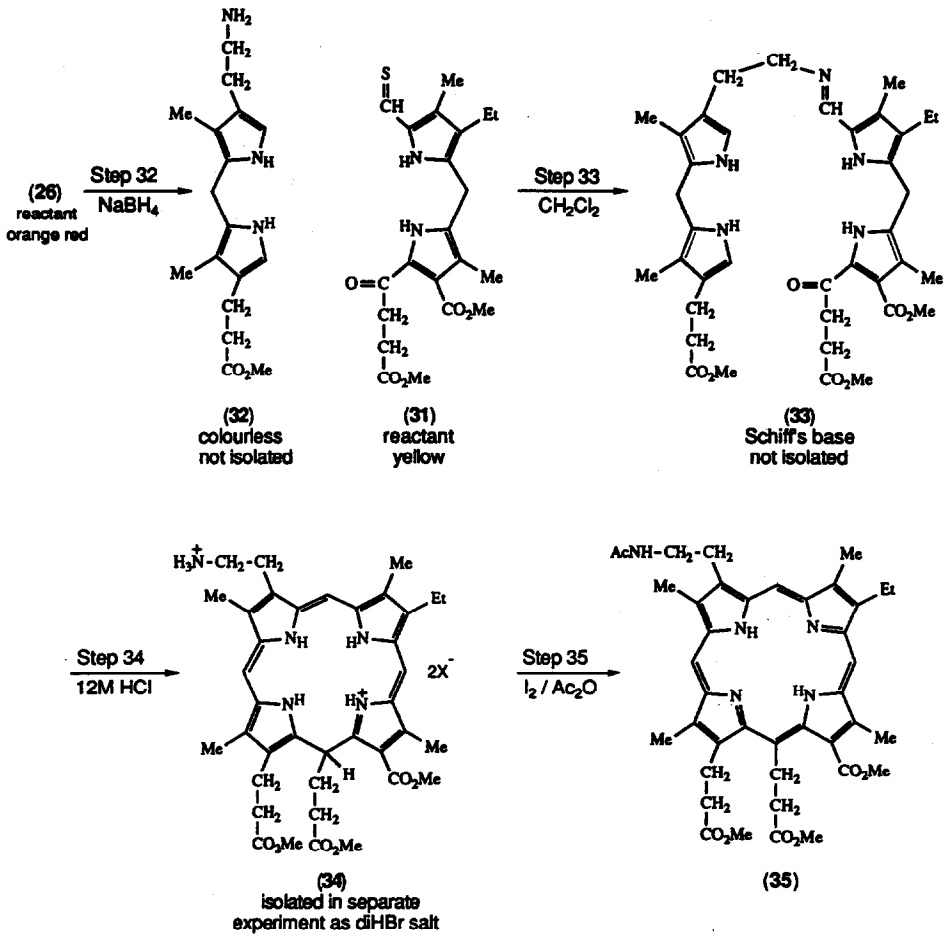

Scheme 8. Scheme for regiospecific cyclisation to generate the porphyrin system via a linear tetrapyrrole (bilene *b*) intermediate (**131** cf. **105**).

work reasonably well, and might have been developed into a workable procedure. However, this approach was overtaken by the discovery that (**30**) could be smoothly converted, by hydrogen sulphide in benzene/methanol into the thioaldehyde (**31**). The latter compound reacted with primary amines to give azomethines in neutral organic solvents and hence the thioaldehyde (**31**) became the right-hand component of choice (Scheme 9).




Scheme 9. Synthesis of the final version of the right-hand component.

A further period of careful experimentation ensued to discover the best conditions for making the macrocycle from the two halves. This is described in detail in the Experimental Section, but it is appropriate to mention certain features here (Scheme 10).




Scheme 10. Observed and isolated stages in the regiospecific synthesis of porphyrin (35).

The experiment was carried out as a continuous sequence using a specially constructed but simple reactor (Figs 19 and 20, Experimental Section). The orange pyrromethene (**26**) was reduced with sodium borohydride to the sensitive colourless dipyrrylmethane (**32**), which was not isolated but extracted at once into dichloromethane, dried, and treated without delay with an equivalent amount of the thioaldehyde (**31**).

Solvent was removed at once, and the resultant Schiff's base (**33**) was assayed by treating a tiny sample with dilute ethanolic hydrogen chloride to give the bilene-*b* salt (**131**), $\lambda_{max}$ 311, 358, 499 nm (Fig. 21, experimental) while the bulk of the product was treated with saturated methanolic hydrogen chloride at room temperature. During this treatment the solution changed in colour from pale yellow to orange to red to a darkish red brown, the intermediate colours being attributed to the pyrromethene salt chromophores present in stages (**131**) and (**132**). The intermediate dihydroporphyrin salt(s) had $\lambda_{max}$ 425, 456, and 713 nm in methanolic hydrogen chloride (Fig. 22, experimental). Oxidation with iodine and acetylation with acetic anhydride then gave porphyrin (**35**) which was chromatographed on Florisil to remove any minor impurities and which crystallised from methanol-dichlormethane as violet needles, mp 233.5–234°. The sequence of steps 32 → 33 → 34 → 35 required careful experimentation, but proceeded in 50% yield overall. More than 50 g of porphyrin (**35**) was prepared in this way.

Porphyrin (**35**) behaved as a single pure substance in all respects, including thin layer chromatography. It had a satisfactory elemental analysis, and the acurate mass of the molecular ion measured by the FAB technique agreed with that calculated for $C_{40}H_{47}N_5O_7+H$ (M+H = 710.355).

In the visible spectrum of porphyrin (**35**) the effect of the *meso*-substituent (→ phyllo-type spectrum) and the *β*-electron withdrawing substituent (→ rhodo-type spectrum) oppose one another and the result is of the general form of an etio-type spectrum, and is similar to that shown by chloroporphyrin $e_6$ trimethyl ester (**106**) (Fig. 5). The $^1$H-NMR spectrum showed three singlets at *δ* 10.12, 10.09 and 9.91 representing *meso*-protons; and three aryl methyl groups at *δ* 3.73, 3.75 and

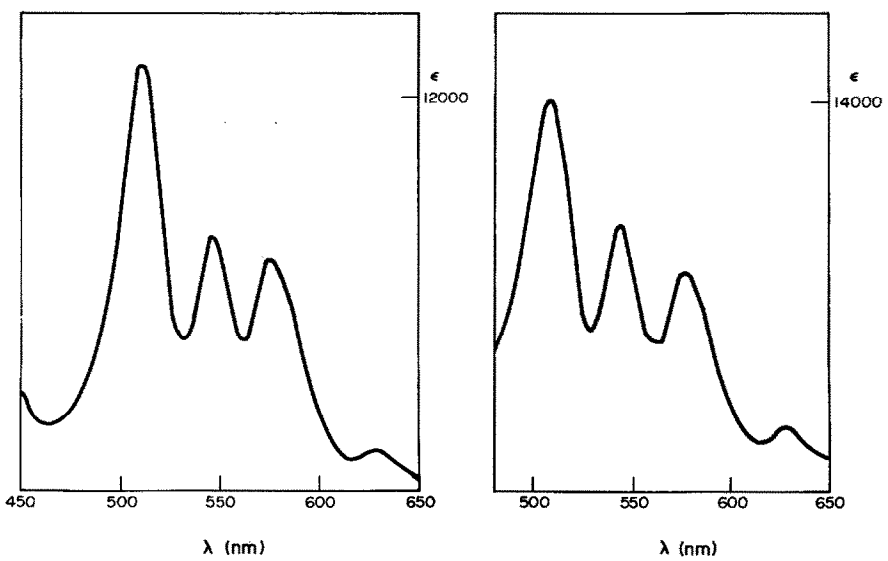



Fig. 5. Visible spectra of porphyrin (**35**) (left, in $CH_2Cl_2$) and chloroporphyrin $e_6$ trimethyl ester (**106**) derived from natural sources (right, in dioxan).

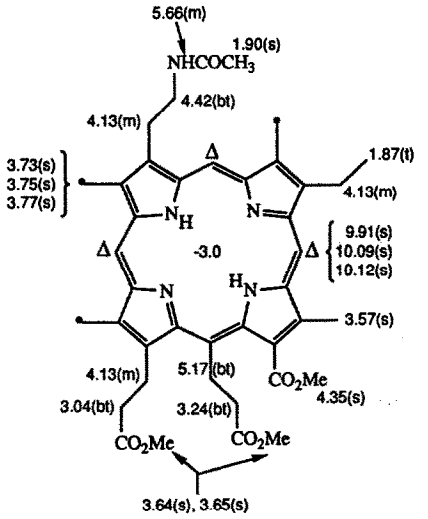
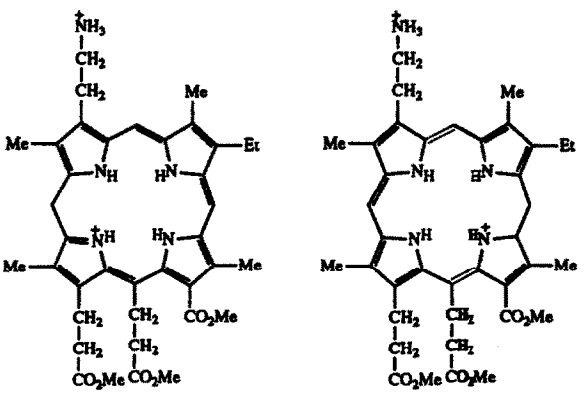



Fig. 6. $^1$H-NMR spectroscopic assignments for porphyrin (**35**).

3.77 corresponding to the methyl groups on rings A, B, and D, but not necessarily respectively. The other signals are assigned as shown in Fig. 6.

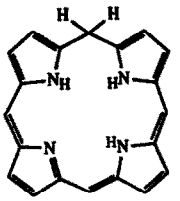
It may be observed that the dihydroporphyrin intermediate (**34**) detected in the experimental work is not the same as that (**132**) which we had conjectured at the planning stage. We suppose that the system (**132**) is initially formed, but that it is deprotonated to give either or both of the cations (**133**) and (**134**). Tautomerisation then leads to the stable isolable cation (**34**). We see here the operation in the synthetic work of the peripheral overcrowding effect discussed earlier: in (**34**)



(**133**) (**134**)

peripheral overcrowding of substituents is reduced by displacing one of them—the *meso*-substituent—from the plane by attaching it to an $sp^3$ centre, and this is the favoured tautomer.

The dihydroporphyrin salt (**34**) is an example of a new class of dihydroporphyrins, the 5,22-dihydroporphyrins (**135**), for which we introduce the trivial name phlorins. These compounds are



(135)

isomeric with the chlorins. They are monoacidic bases, the free bases being a striking imperial blue, while the salts are olive green: these colours reflect a bathochromic shift in going from the free base form ($\lambda_{max}$ 387, 620 nm) to the salt ($\lambda_{max}$ 431, 723 nm) (Fig. 7). This is reminiscent of the change which occurs on monoprotonation of the pyrromethene system, and, although the phlorins (unlike the chlorins) do not have macrocyclic aromaticity, they may be regarded as extended pyrromethenes, and, like them, as the mono-salts, are quite stable. An exception in this regard occurs with oxidising agents such as iodine, oxygen, or quinones such as chloranil, which (as in **34** → **35**) cause rapid dehydrogenation to give the porphyrin system.

Because of our interest in these new compounds, we returned to the chemical sequence illustrated in Scheme 10 in an attempt to isolate and characterise the phlorin stage. This was obtained in low yield as the dihydrobromide (**34**, X = Br) which formed dark bluish green rhombs from acetone. The visible spectrum of the salt in dichloromethane is noteworthy in showing a sharp band in the Soret region; this might lead to confusion with the porphyrin system were it not that the molar extinction is much lower in the present case [$\lambda_{max}$ ($CH_2Cl_2$) 431 nm, $\varepsilon$ 57 800].

## 6. PORPHYRIN REACTIONS: A PORPHYRIN–PHLORIN INTERCONVERSION

In studying the chemistry of the new porphyrin (**35**) with an eye on the ultimate objective we encountered a remarkable reaction. When (**35**) was heated for a brief period (1 hour) in acetic acid in the absence of oxygen there was a pronounced change in the visible spectrum: a new band appeared (Fig. 8) at 721 nm, recalling that of the phlorin salt system already encountered (Fig. 7). It became clear that the porphyrin (**35**) had suffered a clean acid-catalysed equilibration with the

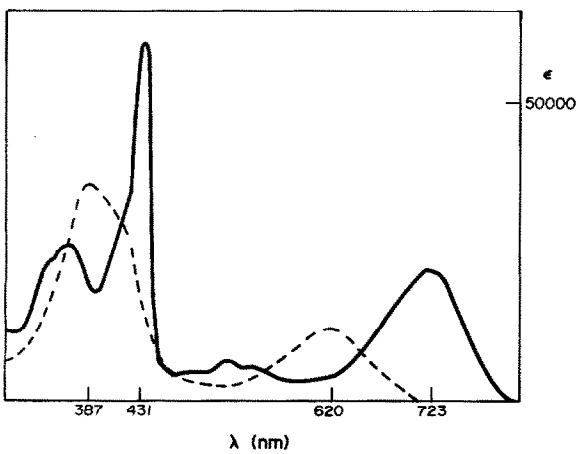
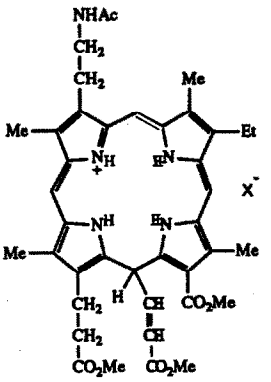
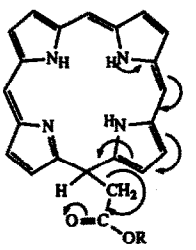



Fig. 7. Visible spectra of the phlorin salt (**34**) (solid line) and the corresponding neutral phlorin (dashed line) (both in $CH_2Cl_2$, X = Br).




(136)




(137)

phlorin acrylic ester (**136**). The two hydrogen atoms from the 15-propionic acid function had not been induced to migrate to the adjacent $\beta$-positions and so generate a chlorin. (Indeed this type of isomerisation was not observed under any conditions examined.) But the discovery of the porphyrin–phlorin equilibration nevertheless offered a way forward (Scheme 11).

The equilibration slightly favoured the porphyrin ($K_{\text{phlorin/porphyrin}} = 0.60$). It was possible to isolate the two components by diluting with ether and extracting the porphyrin with 0.7 M-HCl, the neutral spectrum of the porphyrin being shown in Fig. 9. The spectrum of the phlorin salt remaining in the organic layer is shown in Fig. 10. The equilibrium constant illustrates again the considerable thermodynamic stability of the phlorin salt system, no doubt assisted in this example by relief of the peripheral overcrowding in the direction (**35**) → (**136**). The reaction may be rationalised in terms of a series of protonations and deprotonations as shown in Scheme 12.

When the above equilibration was carried out in acetic acid for a short period in the presence of air, the phlorin was oxidised to the corresponding porphyrin acrylic acid ester (**36**) which was obtained in 91% yield (Scheme 11). Quantitative determination of the oxygen taken up in this process showed that one gram-atom of oxygen was consumed for each mole of porphyrin (**35**) dehydrogenated.

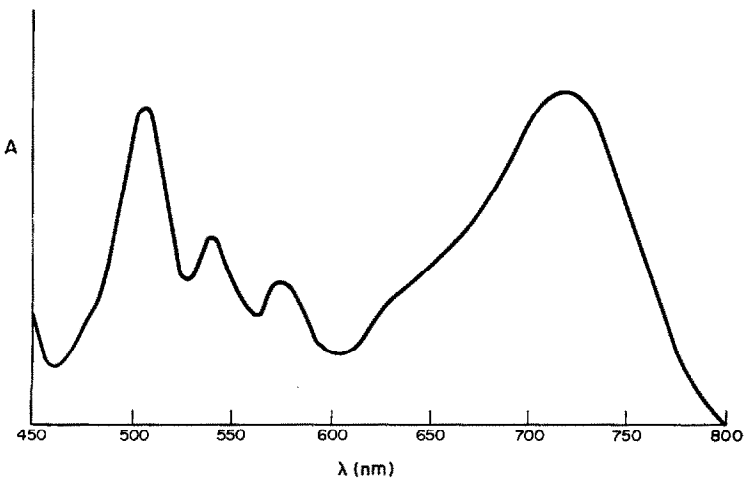
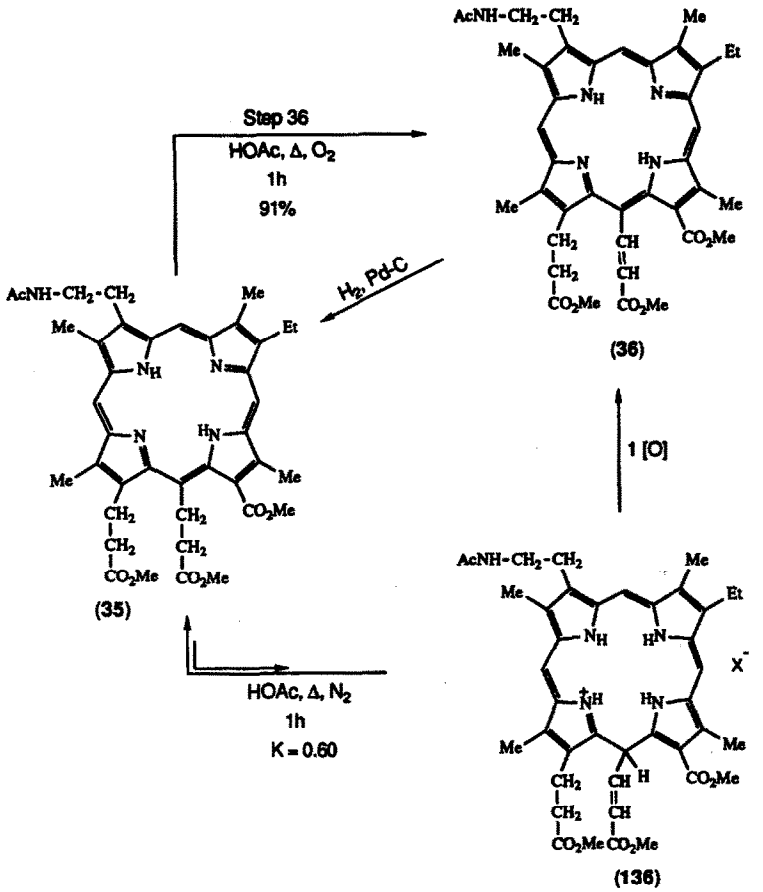



Fig. 8. Visible spectrum of the equilibrium mixture of porphyrin (**35**) and phlorin (**136**) (HOAc, 100°, 1 h, no oxygen).




Scheme 11. Chemical modification of the first porphyrin (35) in the synthetic sequence.

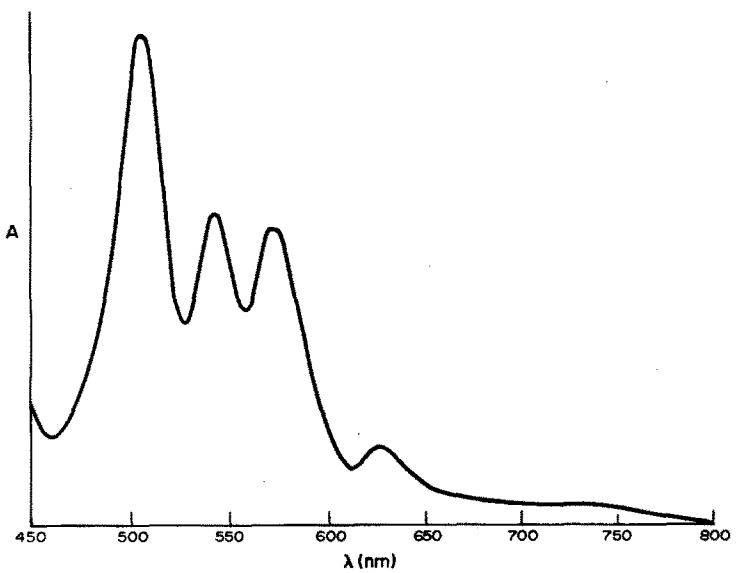



Fig. 9. Porphyrin (35) extracted from equilibrium mixture by fractionation between ether and 0.7 M-HCl.

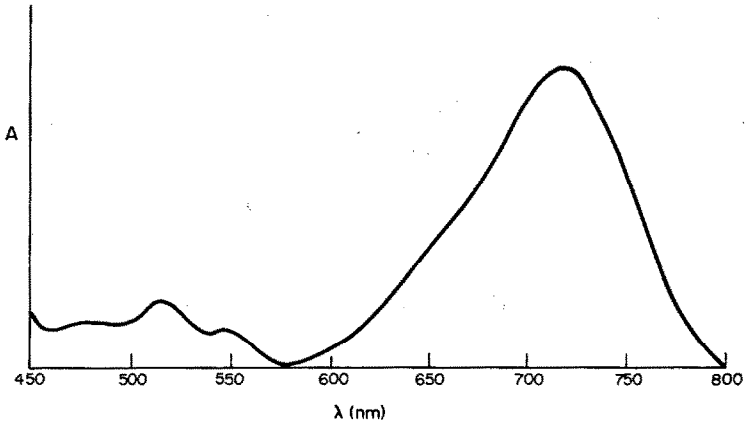
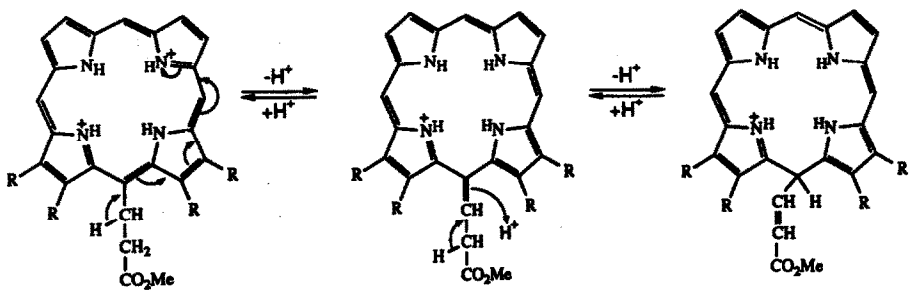



Fig. 10. Phlorin salt (**136**) remaining in supernatant after extracting porphyrin (**35**) from the equilibrium mixture.




Scheme 12. Acid-catalysed equilibration of porphyrin and phlorin.

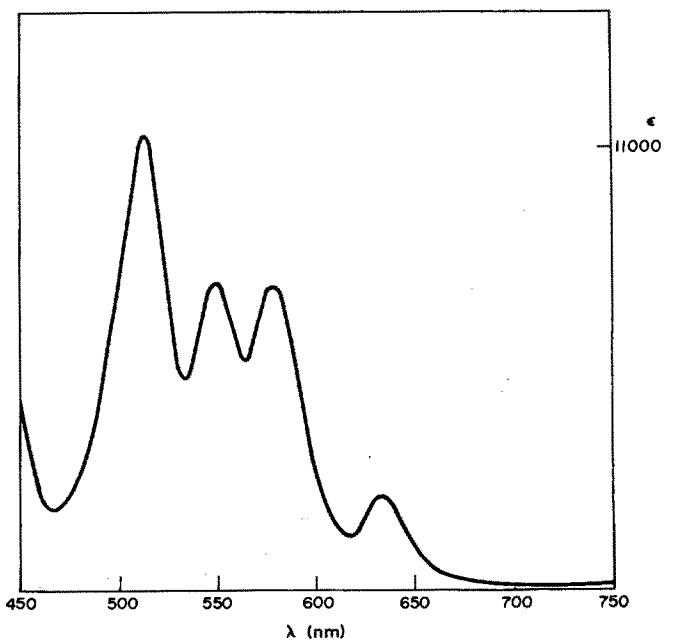



Fig. 11. Visible spectrum of the porphyrin *meso*-acrylic ester (**36**) in $CH_2Cl_2$—MeOH.

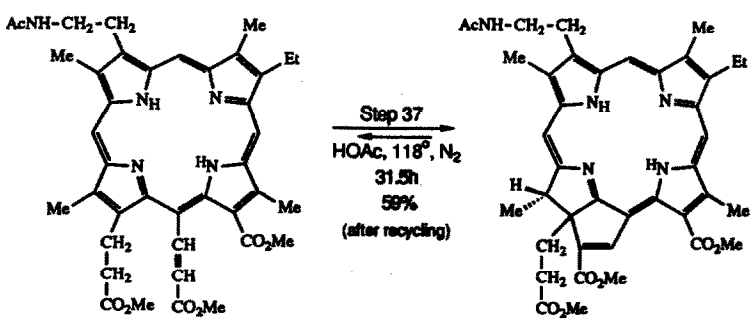
The new porphyrin (**36**) formed large violet spears, mp 251–252°, from methanol–dichloromethane. Peaks in the visible spectrum of (**36**) (Fig. 11) showed a small bathochromic shift (*ca.* 3 nm) with respect to porphyrin (**35**); this relatively small effect would accord with the double bond of the *meso*-acrylic ester function being turned out of the plane, and out of conjugation with the porphyrin ring. The infrared spectrum did not provide clear-cut evidence for an additional conjugated ester function but the NMR spectrum did so (>—CH=CH—CO, doublets at $\delta$ 10.23 and $\delta$ 6.25 respectively, with J = 15.5 Hz, according with *trans* geometry). On silica plates porphyrins (**35**) and (**36**) were virtually indistinguishable but on silver nitrate impregnated silica plates the more unsaturated porphyrin (**36**) had the lower mobility. As expected, catalytic reduction of the porphyrin 15-acrylic acid ester (**36**) regenerated the 15-propionic acid ester (**35**). The reduction proceeded through the sensitive *leuco*-compound and the isolated yield was modest (25%).

At this stage it is of interest to mention the reasons for having a propionic acid residue at C-15 in porphyrin (**35**) rather than an acetic acid residue which, after all, is present at that position in the target molecule (**46**). Firstly, preliminary experiments with the readily prepared 15-propionic acid ester (**130**) had shown a porphyrin–phlorin equilibration analogous to that discussed above. Secondly, it was suspected that when a dihydroporphyrin (i.e. phlorin) was formed, during the macrocyclisation or subsequently, a *meso*-acetic acid residue would tend to be extruded, with the relief of peripheral overcrowding, as indicated in partial structure (**137**, arrows), with considerable diminution in the yield of the desired product. The formation[26] of rhodoporphyrin methyl ester (**107**) on reduction of chloroporphyrin $e_6$ trimethyl ester (**106**) illustrates this effect; such considerations were sufficient to dissuade us from going for the obvious C-15 acetic acid substituent.

## 7. ENTRY INTO THE CHLORIN SERIES

To arrive at porphyrin (**36**) it had been necessary to heat the precursor (**35**) in acetic acid in air for 1 h, when essentially a dehydrogenation had occurred, but longer heating caused problems. It was eventually discovered that if the *meso*-acrylic ester derivative (**36**) was now refluxed in acetic acid (in the absence of air and for much longer), an equilibrium was established with the purpurin (**37**), that is, a chlorin with a *meso*-electron-withdrawing group (Scheme 13). This is believed to be the first occasion on which a reversible porphyrin–chlorin interconversion has been observed. Under the conditions described the equilibrium constant, $K_{\text{purpurin/porphyrin}}$, was *ca.* 1.7 and by recycling the recovered unchanged porphyrin it was possible to isolate the purpurin (**37**) in a yield of 59%.

Purpurins do not have the pure green colour of ordinary chlorins, but rather have a puce or




(**36**) (**37**)

Scheme 13. The porphyrin–purpurin interconversion.

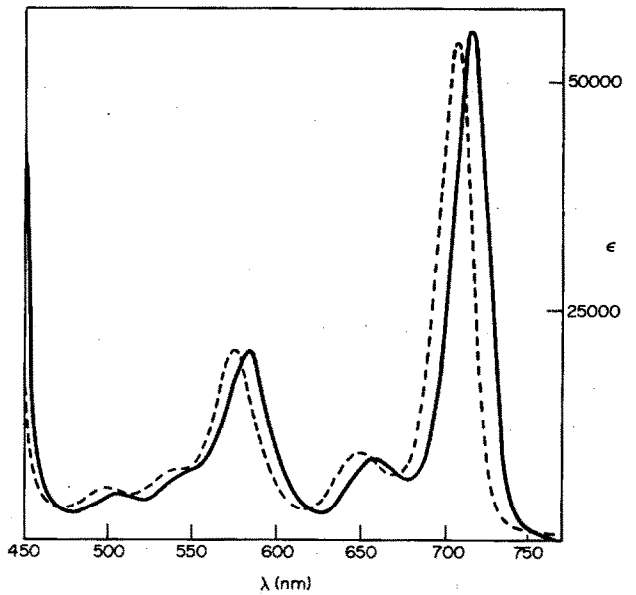
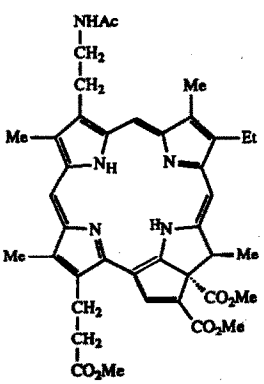



Fig. 12. Visible spectra of the purpurin (**37**, dashed line) and the vinylpurpurin (**38**, solid line), in $CH_2Cl_2$.

khaki tinge to their greenness. Purpurin (**37**) was no exception: from brownish purple solutions in methanol–dichloromethane it formed blue rhombic plates which melted at 310° with decomposition. The visible spectrum (Fig. 12) had a strong band in the 700 nm region, characteristic of chlorins, with additional moderate absorption in the *ca.* 550 nm region usually found with purpurins.

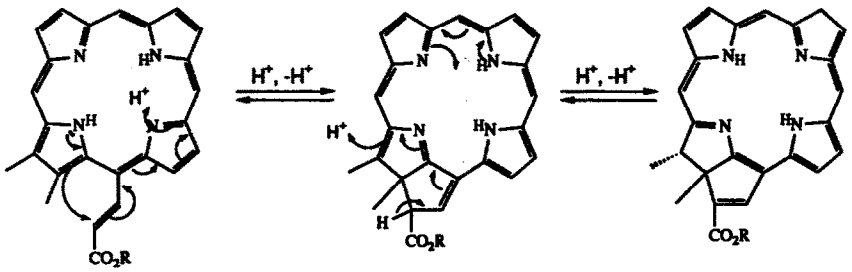
The reaction is regarded as an acid-catalysed cyclisation, since we have observed it to occur under other acidic conditions (e.g. D-camphorsulphonic acid in diphenyl ether). Various pathways can be written[5] and one is presented in Scheme 14. Cyclisation, with the relief of steric strain is believed to be an important factor again, and since the $sp^3$ centres at C-17 and C-18 are established under equilibrium conditions, the favoured transoid geometry is expected as shown.

The cyclisation could equally well be written as occurring at C-13, and there is some evidence that a minor by-product, formed under certain acid conditions, has structure (**138**). However, this




(138)

cyclisation interrupts the conjugation between the C-13 ester function and the aromatic ring, and it is this factor which keeps the equilibrium concentration of this component at a low or (in acetic acid) negligible level.



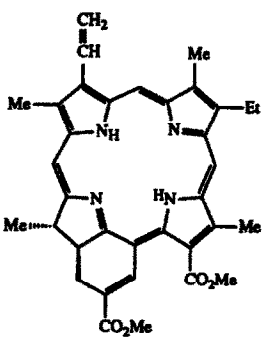
Scheme 14. Mechanistic rationalisation for the interconversion (**36**) → (**37**) (partial structures).

## 8. CHLORIN ELABORATION: THE COMPLETION OF THE SYNTHESIS

### 8.1. *Chlorin elaboration*

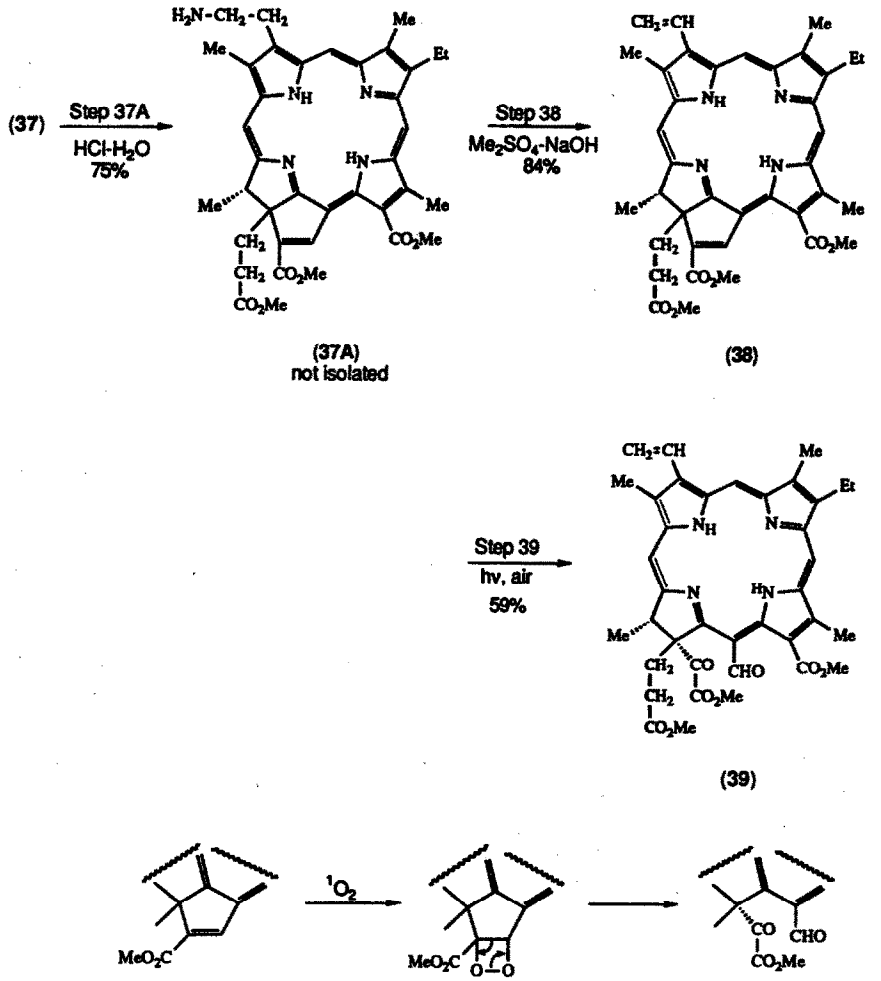
Up to this point the vinyl group at C-3 had been protected as an acetylaminoethyl group, and it was decided to remove this protection at this stage. There were, as it turned out, still nine stages to go, but some of them were related to known aspects of chlorophyll chemistry, where the vinyl group was known to survive. A decision had to be made to carry out the deprotection at a point where large amounts (several grams) of material were available in order to be ready for difficulties still to come: it was here.

The deprotection involved acid hydrolysis to form the *β*-aminoethyl purpurin (**37A**) which was obtained in 75% yield (Scheme 15). It was a sensitive compound and was immediately subjected to Hofmann elimination by treatment with dimethyl sulphate in alkali to give the vinylpurpurin (**38**) isolated as narrow purple prisms which decomposed at 285–290° without melting. The visible spectrum was similar to that of the precursor purpurin (**37**) but showed a small bathochromic shift consistent with the extra conjugation (Fig. 12). With the strong absorption in the 570 nm region, and an even stronger band at about 700 nm, these spectra call to mind the spectrum of neopurpurin 4 dimethyl ester[57] which has a rather similar chromophore (**139**).



**(139) Neopurpurin 4 dimethyl ester**
**$\lambda_{max}$ (dioxan) (ε) 492 (4900), 526 (5000), 561 (17500), 631 (6000), 693 nm (50 000), data read from curve.[57]**

For the above reactions (and, of course, for some of the earlier ones: see experimental) an inert atmosphere was used in the absence of light, and this regime was strictly adhered to henceforward, except in the next step! It was thought to be only fitting that a photochemical reaction should be employed in the synthetic pathway to chlorophyll: in this case one might be forgiven for thinking that both the sun and good fortune shone upon the venture.




Scheme 15. Deprotection of the vinyl group of purpurin (**37**) and photo-oxygenation of the C-15[1]–C-15[2] double bond of the resulting vinylpurpurin (**38**) to give the methoxalylpurpurin (**39**). A possible mechanism for the photo-oxygenation is shown.

In the vinylpurpurin (**38**) one hydrogen was correctly placed at a $\beta$-position of ring D (C-18), but the C-17 centre remained to be modified. This modification was effected following the discovery that the substance was susceptible to photoreaction in air. This was not in itself surprising and would normally, as we have already implied, be avoided because it would be likely to lead to complex decomposition. In this case, however, under controlled conditions a single product was formed in a spectroscopic yield of about 75%. Thus, as shown in Scheme 15, irradiation of the vinylpurpurin (**38**) in dichloromethane at 0° with a tungsten lamp for 35 minutes gave, in the presence of air, the methoxalylpurpurin (**39**) in which oxidative cleavage of the C-15[1]–C-15[2] double bond had occurred. The product was isolated in 59% yield as dark crystals decomposing at *ca.* 250–255°. With a formyl group at C-15, the colour and visible spectrum remained that of a purpurin. In the $^1$H-NMR spectrum the formyl proton appeared as a singlet at $\delta$ 11.22, the C-5 and C-10 *meso*-protons at $\delta$ 9.45 and 9.00, with the C-20 *meso*-proton flanking the reduced ring at higher field ($\delta$ 8.28) as expected. These magnetic resonance features accord with those found in the known purpurin 5 dimethyl ester (**43**, see later). The photoreaction is regarded as a photo-oxygenation in which singlet

oxygen is generated from ground state oxygen by the triplet purpurin, thus:

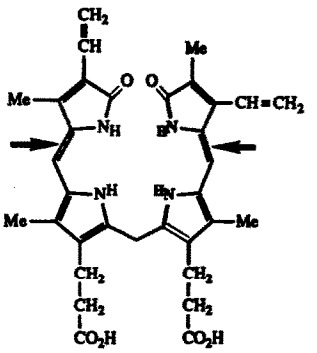
$$\text{Purpurin (38) } S_0 \xrightarrow{h\nu} \text{Purpurin (38) } S_1$$

$$\text{Purpurin (38) } S_1 \xrightarrow{\text{isc}} \text{Purpurin (38) } T_1$$

$$\text{Purpurin (38) } T_1 + {}^3O_2 \longrightarrow \text{Purpurin (38) } S_0 + {}^1O_2$$

$$\text{Purpurin (38) } S_0 + {}^1O_2 \longrightarrow \text{Purpurin (39).}$$

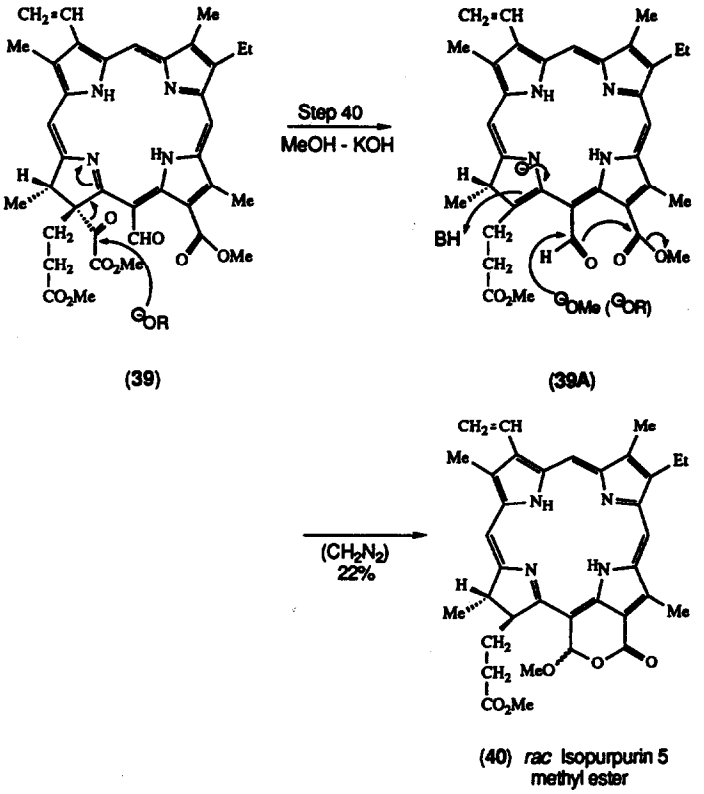
The detailed mechanism is not known, but a dioxetane intermediate as shown in Scheme 15 is regarded as plausible. That the reaction should have left the vinyl group untouched is remarkable but not exceptional. Thus, although bilirubin IXα has two vinyl substituents, cleavage with singlet oxygen occurs, in part, at the bridge double bonds (**140**, arrows) to give methylvinylmaleimide and a dipyrrylmethane dialdehyde.[58]



**(140) Bilirubin IXα**

Since the methoxalyl function is, in general, vulnerable to chemical modifications including removal, we were now close to systems which were derivable from chlorophyll *a*. Since the earlier exploration of chlorophyll chemistry[6,7,10] had, as we have seen, been very extensive, we hoped soon to penetrate into its foothills and find a recognisable path. This was accomplished on treating the methoxalylpurpurin (**39**) with alkali under mild conditions, followed by diazomethane to esterify any carboxylic acid that had formed. This gave the methoxylactone (**40**), racemic isopurpurin 5 methyl ester, albeit in only modest yield (22% isolated). The proposed manner of formation of this substance is outlined in Scheme 16. A reverse Claisen reaction removes the methoxalyl residue, and the protonation of the anion under alkaline equilibrating conditions ensures that the transoid stereochemistry at the β,β-positions of ring D is retained as shown (**39A**, arrows). Base-promoted cyclisation to give the lactol ring, a common feature of chlorophyll chemistry, then ensues to give (**40**). The stereochemistry of the C-15[1] substituent (OMe) is not known, but it is expected to be most stable in the α-configuration for the ring D stereochemistry as drawn.[59]

Isopurpurin 5 dimethyl ester had been obtained by Fischer and Strell[60] from purpurin 5 dimethyl ester by treatment with alkali followed by diazomethane and acid fractionation. The earlier workers had not formulated the substance correctly: the record is not completely clear on this point, but structure (**141**) appears to have been preferred. However, we did not observe a band which could be assigned to OH stretching in the infrared spectrum. Both isopurpurin 5 dimethyl ester (**40**) and chlorin 5 (**41**, see later) had similar chlorin-type spectra (Fig. 13), indicating that the electron-withdrawing effect of the group at C-15 has been removed. These observations, and a consideration of the chemistry involved, including the operation of the peripheral overcrowding effect, led us to the




Scheme 16. Synthesis of *racemic*-isopurpurin 5 methyl ester (**40**).

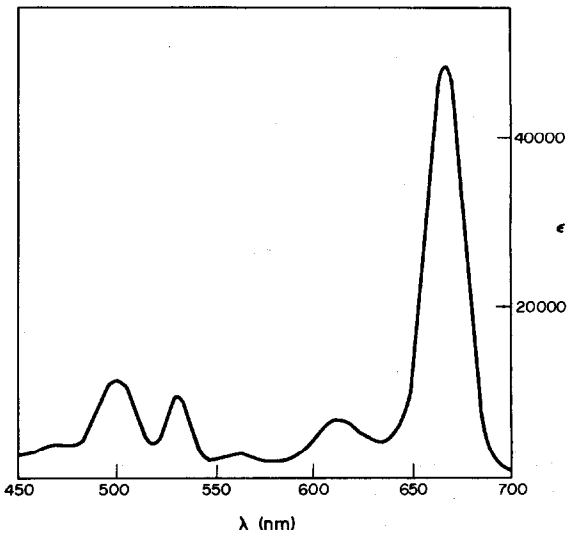
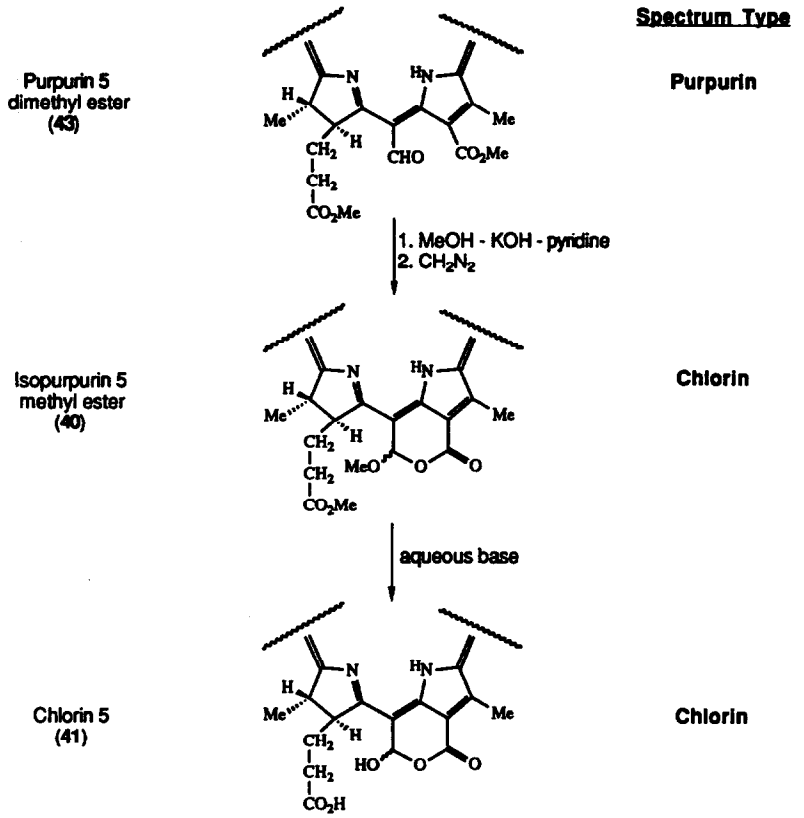
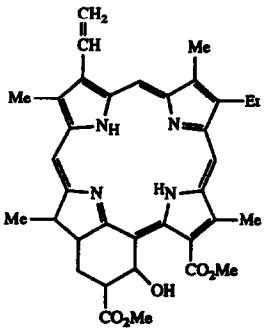



Fig. 13. Visible spectrum of racemic isopurpurin 5 methyl ester in $CH_2Cl_2$.




Scheme 17. Structural relationship between purpurin 5 dimethyl ester, isopurpurin 5 methyl ester and chlorin 5.

methoxylactone formulation shown in Scheme 17 as an interpretation of these changes. This was supported by a detailed interpretation of the proton NMR spectrum of isopurpurin 5 methyl ester. Thus the C-15[1] proton appeared as a singlet at $\delta$ 7.73: the methoxy group also located at C-15[1], appeared at $\delta$ 3.88, while no methoxy singlet was found at *ca.* $\delta$ 4.3, expected for a 13-$CO_2Me$ function. This evidence supports structure (**40**) and rules out such structures as (**141**).



(**141**)

It was now possible for the first time in this sequence to compare a synthetic (albeit racemic) compound i.e. isopurpurin 5 methyl ester with a natural (17*S*, 18*S*) sample.

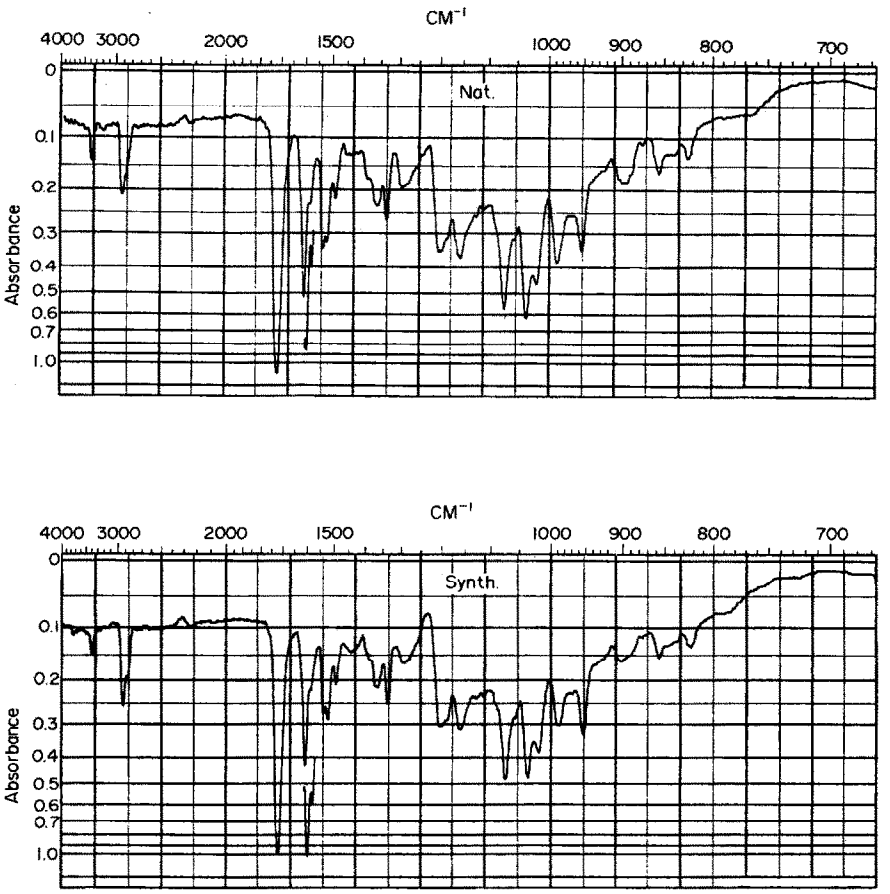


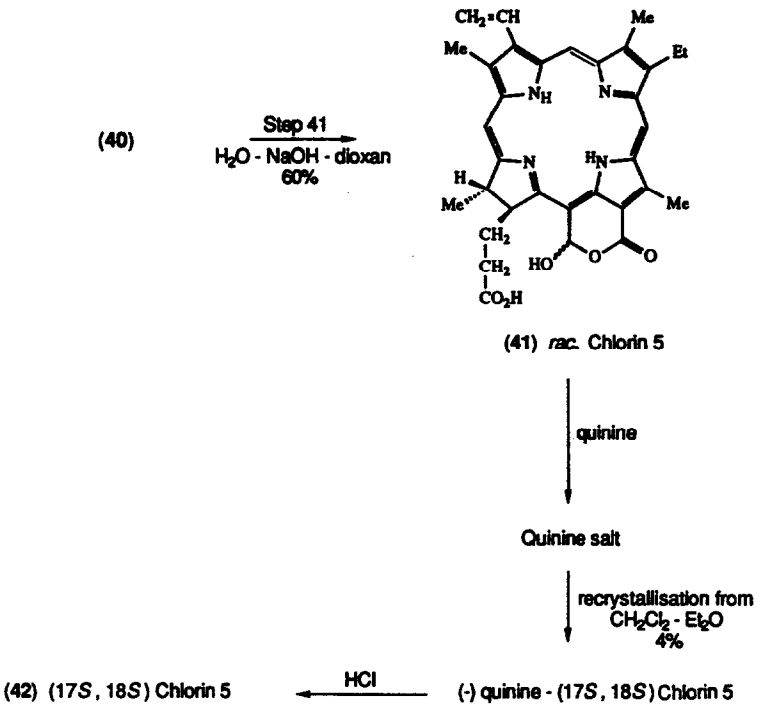

Fig. 14. Infrared spectrum of isopurpurin 5 methyl ester in $CH_2Cl_2$. Top: natural, 2.5 mg in 0.28 ml. Bottom: synthetic, 2.3 mg in 0.27 ml.

The visible spectra of the two samples were the same (see Fig. 13): more persuasively, the infrared spectra in dichloromethane solution also indicated identity (Fig. 14).

8.2. *Resolution*

When the racemic isopurpurin 5 methyl ester (**40**) was hydrolysed with sodium hydroxide in aqueous dioxan, it was converted into racemic chlorin 5 (**41**, Scheme 18) in 60% yield. The visible spectrum was identical with that of a sample of optically active chlorin 5 derived from natural sources.[61] Chlorin 5 (**41**) possesses a carboxylic acid function, and it was at this point that resolution was effected.

Various bases (cinchonidine, cinchonine, quinidine, morphine, thebaine, brucine and strychnine) were examined for this purpose: eventually, however, resolution was achieved using quinine, which was purified for the purpose by the method of Thron and Dirscherl.[62] In fact, two molar equivalents of quinine were needed to produce the conditions for the diastereoisomeric salt to crystallise. Crystallisation and recrystallisation of the quinine–chlorin 5 salt were wasteful but gave a sample having a specific rotation $[\alpha]^{23}_{546}$ of $+1236°$; for comparison, the specific rotation of the quinine salt made from naturally-derived chlorin 5 was $+1215°$ in the same solvent (acetone). The visible spectra of the synthetic salt and the natural salt were identical.



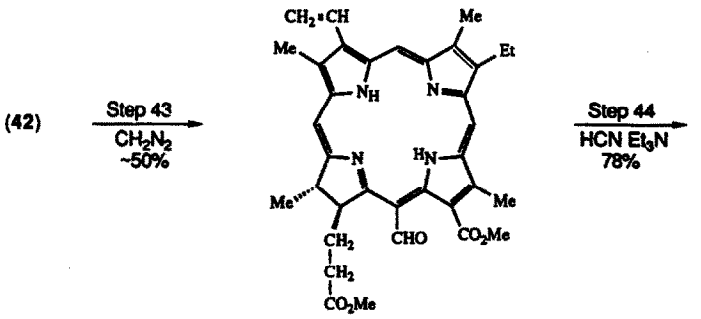

Scheme 18. Chlorin 5 and its resolution.

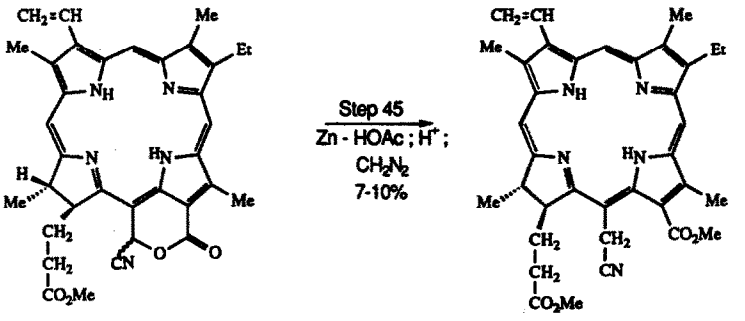
8.3. *Relay position, and final elaboration of peripheral substituents*

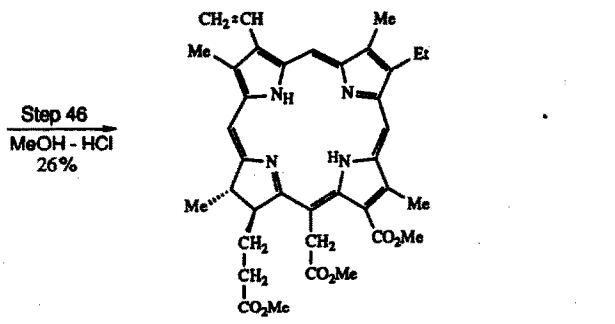
Synthetic optically active chlorin 5 (**42**) was recovered from the quinine salt after acidification (Scheme 18). The visible spectrum in acetone was essentially identical with that of the naturally-derived material. The specific rotations were also identical within the experimental error ($[\alpha]_{546}^{23}$ for synthetic chlorin 5 = +1810°; for natural chlorin 5 = +1823°). The chlorin 5 was not crystallised but was converted, by treatment with diazomethane in methanol-ether, into purpurin 5 dimethyl ester (**43**) as shown in Scheme 19. This was obtained in crystalline form in very low yield overall (2%) from racemic chlorin 5 (**41**). This low yield reflects the considerable difficulty of the resolution step; the yield of the quinine salt of chlorin 5 recrystallised to enantiomeric purity was only 4%. The optically-active purpurin 5 dimethyl ester so obtained was identified with the naturally-derived compound by visible and infrared spectroscopy (Figs 15 and 16, respectively). The melting point was 191.5–195°; the naturally-derived sample had mp 192–195.5°, and the mixture showed no depression. The melting point of racemic purpurin 5 dimethyl ester synthesised along the same route (but omitting the resolution step) was 221–222.5°.

The repeated establishment of identity with material of natural provenance at various points, [(17*S*, 18*S*)-chlorin 5 and its quinine salt, (17*S*, 18*S*)-purpurin 5 dimethyl ester], allowed us, at this stage, to replenish our stocks of material from natural sources. Compound (**43**), purpurin 5 dimethyl ester, was chosen as the relay point, since the identity of natural and synthetic optically active samples was secure and since (**43**) is conveniently available from methyl pheophorbide *a* (**103**).[63]

Treatment of purpurin 5 dimethyl ester with hydrogen cyanide in dichloromethane in the presence of triethylamine gave the cyanolactone (**44**) in 78% yield. This substance has been described before,[64] but since the description referred to an infusible compound, whereas our sample had a well-defined (if elevated) melting point, we have repeated the characterisation including elemental analyses. Disconcertingly the infrared spectrum of the cyanolactone (**44**) did not show a clearly




Scheme 19. The final steps (42) → (46).

identifiable peak in the triple bond region. However, the intensity of the C≡N stretching mode is known to be diminished when oxygenated functions are included in the molecule, especially when, as here, the oxygen-containing group is attached to the same carbon as is the nitrile.[65] Hydrogenolysis of the cyanolactone (**44**) with zinc dust in acetic acid, followed by acid treatment (to remove any zinc inevitably complexed by the macrocycle) and esterification with diazomethane gave the chlorin $e_6$ nitrile (**45**). The complexity of these three steps resulted in a disappointingly low yield, but at least it was consistent in the range 7–10% on a scale of 20 to 115 mg. The product (**45**) had a visible spectrum characteristic of a chlorin, and in the infrared the nitrile function was revealed as a weak sharp band at 2240 $cm^{-1}$.

The final step involved the methanolysis of the nitrile function of (**45**). This was achieved with saturated anhydrous methanolic hydrogen chloride at 0° for 16 h. Chromatographic purification, followed by three crystallisations from moist dichloromethane–acetone–methanol gave chlorin $e_6$

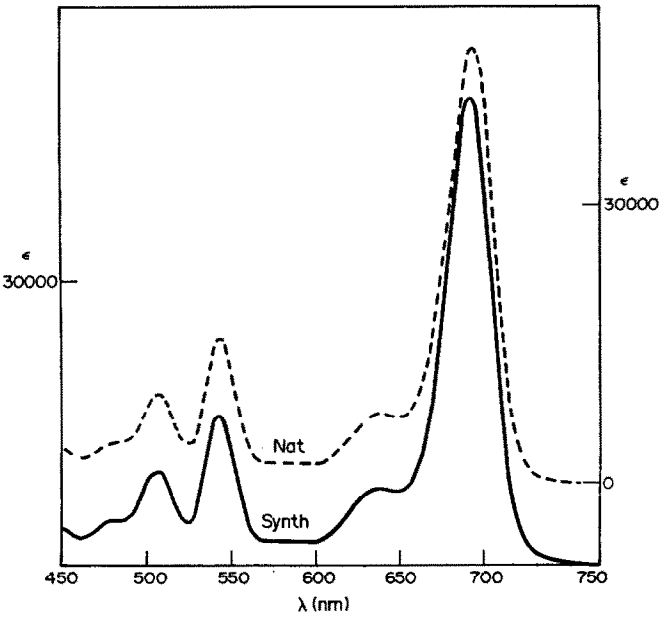



Fig. 15. Visible spectrum of purpurin 5 dimethyl ester in $CH_2Cl_2$. Dashed line—natural, right-hand ordinate; solid line—synthetic, left-hand ordinate.

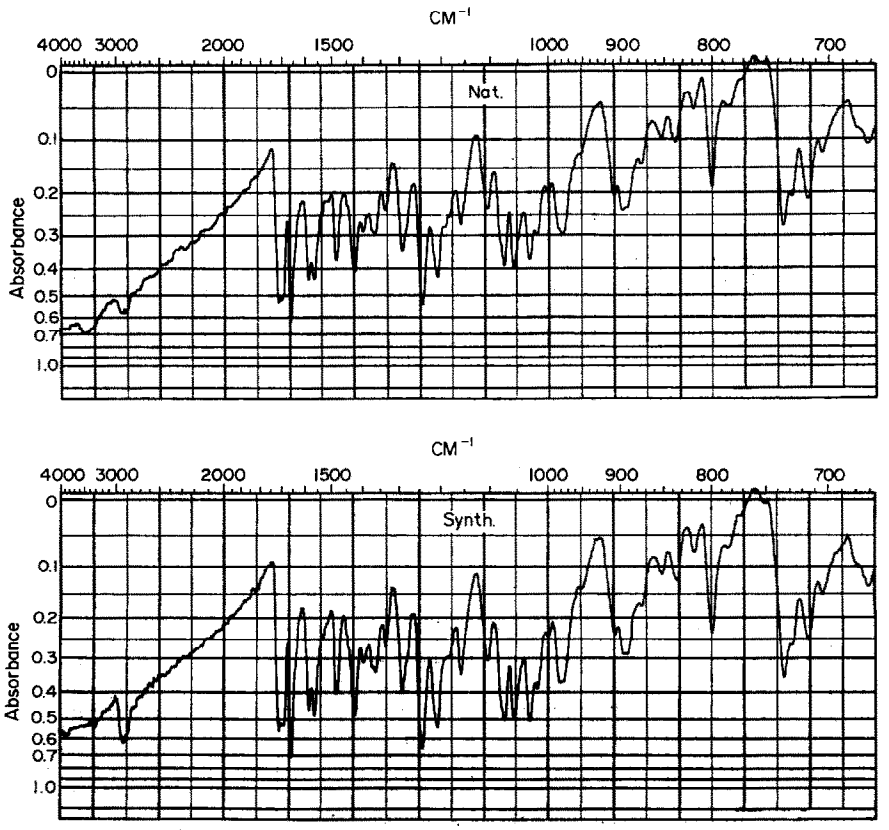



Fig. 16. Infrared spectrum of purpurin 5 dimethyl ester in KBr. Top: natural. Bottom: synthetic.

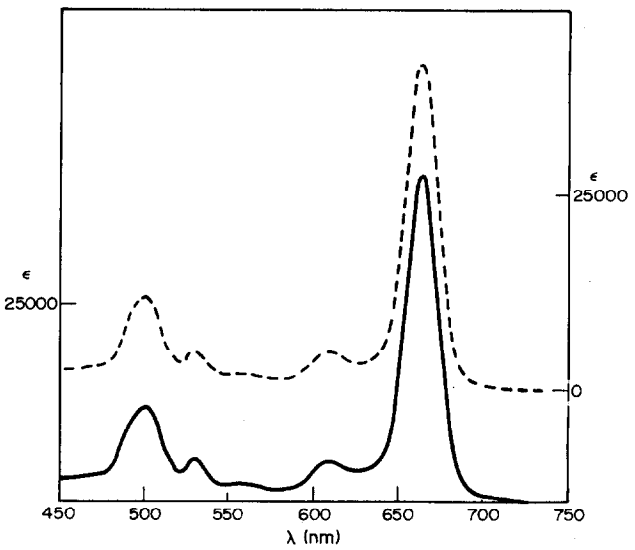



Fig. 17. Visible spectrum of chlorin $e_6$ trimethyl ester in $CH_2Cl_2$. Dashed line—natural, right-hand ordinate; solid line—synthetic, left-hand ordinate.

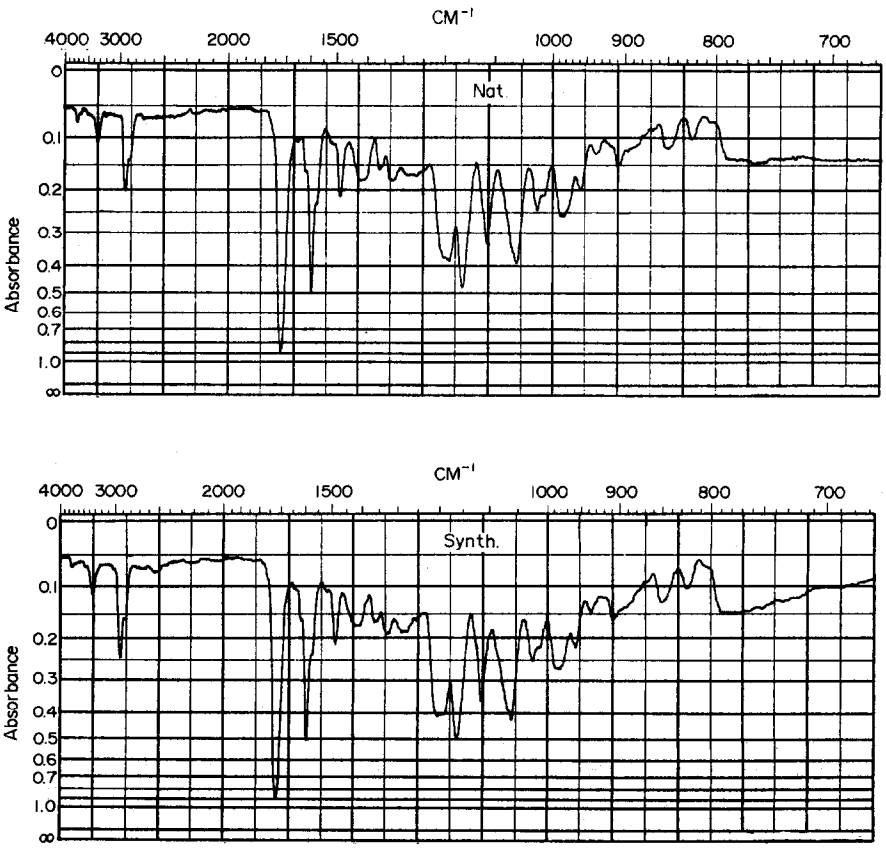



Fig. 18. Infrared spectrum of chlorin $e_6$ trimethyl ester in $CH_2Cl_2$. Top: natural, 3.8 mg in 0.3 ml. Bottom: synthetic, 3.8 mg in 0.3 ml.

trimethyl ester (**46**) in 26% yield (Scheme 19). The melting point of (**46**) so prepared was 207.5–208.5°: the naturally-derived sample had mp 208–209.5°, and the mixed melting point was 207.5–209°. The visible spectra and infrared spectra of the synthetic and natural materials were essentially identical, in each case, as shown in Fig. 17 and Fig. 18, respectively.

As we outlined at the outset of this paper, this step completes the first total chemical synthesis of chlorophyll *a*. The route chosen has been a long and difficult one, but it has the merit of being firmly based in known pyrrole chemistry, and in proceeding by well-defined steps, which are described in detail in the following section. It is interesting to note that Inhoffen and his colleagues[66] have, subsequent to our original report,[1] extended the synthetic scheme to include chlorophyll *b*. Thus the two major chlorophylls of the plant kingdom have now been synthesised in the laboratory, and a great deal of fascinating chemistry has been uncovered on the way.

## 9. EXPERIMENTAL

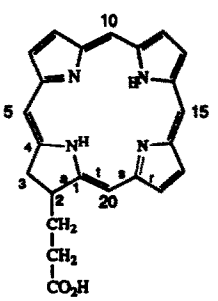
### 9.1. *General*

Column chromatography was carried out on Florisil, a synthetic activated magnesium silicate made according to U.S. Patent 2,593,625 (Jan. 29 1946) by the Floridin Co, Tallahassee, Florida. Solvent ratios refer to volumes. Melting points are not corrected; for the macrocyclic substances they were taken on a Kofler block.

Spectroscopic instrumentation was as follows: electronic spectra—Cary 11 and Cary 14; infrared spectra—Perkin Elmer 21 and Infracord: NMR spectra at 60 MHz, Varian HR60. Optical rotations refer to the mercury line at 546.07 nm using the Rudolph photoelectric optical rotation spectrometer. In those cases where the spectroscopic or other data reported here do not exactly coincide with those reported in the preliminary communication,[1] the present values are preferred (for example, because they are later determinations).

[Aug. 1989. Some additional experiments on the original samples were carried out at Queen Mary College. They are denoted by square brackets. The following equipment was used. Infrared spectra: Perkin Elmer—1600 FTIR; NMR spectra—Bruker AM 250 (Mr G. Coumbarides) and WH 400 (ULIRS service, Mr P. Haycock); mass spectra VG ZAB-E (SERC Mass Spectrometry Service, Swansea). TLC observations were made on Merck DC plastic sheets covered with Kieselgel 60 or Kieselgel 60 $F_{254}$ to ascertain the homogeneity of the samples examined. This support did not distinguish porphyrin (**35**) from porphyrin (**36**): these were distinguished on $AgNO_3$-impregnated plates (aqueous $AgNO_3$ spray, air-dried overnight in dark)].

The names of the tetrapyrrole macrocycles given first in the titles are systematic, following the IUPAC/IUB rules.[19] In the systems considered here the principal function is the propionic acid in ring D, which becomes C-2, as shown in (**142**).




(142)

By an extension of the linear tetrapyrrole nomenclature,[19] dipyrrylmethanes are systematically called dipyrranes (rather than 5,10-dihydrodipyrrins) and 5,10,15,17-tetrahydrotripyrrins are called tripyrranes.

9.2. *Synthesis of ring A*

*Step* 1. *Diethyl* 3,5-*dimethylpyrrole*-2,4-*dicarboxylate* (**1**, *Knorr's pyrrole*). Sodium nitrite (134 g) in water (185 ml) was added dropwise over about 2 h to a stirred solution of ethyl acetoacetate (250 g) in glacial acetic acid (500 ml), previously cooled to 4° in an ice–salt bath, at such a rate that the temperature did not exceed 7°. The clear solution was stirred for a further hour at room temperature, and a second portion of ethyl acetoacetate (250 g) was added. The resulting solution was added rapidly to a vigorously stirred ice-cooled mixture of acetic acid (900 ml) and zinc dust (375 g) in a 3 l round bottom (r-b) flask. The addition was so conducted that the temperature was allowed to rise rapidly to 70°, and then gradually to 95° over about 45 min. The mixture was stirred and heated at 100° for a further 1.5 h before being decanted hot from zinc and other solids into 10 l of stirred water. The residual zinc was washed with acetic acid (2 × 60 ml) and the washings were added to the aqueous liquor. The diester had precipitated: it was collected by filtration and crystallised from 95% ethanol (1.5 l) with hot filtration giving the product (**1**) as very pale yellow needles (262 g, 57%) mp 136–137° (lit.[31] mp 136–137°).

Yields of 57–61% were consistently obtained in laboratory operations on a 200–1000 g scale. The procedure also proved suitable for scaling up in the pilot plant to the multikilogram level.[32]

*Step* 2. 5-*Ethoxycarbonyl*-2,4-*dimethylpyrrole*-3-*carboxylic acid* (**2**). This acid was prepared consistently in 90% yield by treating the diester (**1**, 100 g–1000 g) with conc. sulphuric acid.[33] The reaction was also satisfactorily scaled up to the multikilogram level.[32] Generally *ca.* 8% of unhydrolysed ester (**1**) was recovered. The crude precipitated acid (**2**) was very finely divided and filtering and drying were tedious. The material had mp 269–270° (lit.[33] mp 273°) and was sufficiently pure to be used directly for the next step.

*Step* 3. *Ethyl* 3,5-*Dimethylpyrrole*-2-*carboxylate* (**3**). This was prepared by the method of Chu and Chu.[34] Mp 124–125° (lit.[34] mp 124.5–125°); yields 84–87% on a 200–300 g scale.

*Step* 4. *Ethyl* 4-*formyl*-3,5-*dimethylpyrrole*-2-*carboxylate* (**4**). The formylation method of Chu and Chu[34] was modified for large-scale use. To a stirred, ice-cooled mixture of (**3**) (211 g) and freshly distilled DMF (300 g), phosphorus oxychloride (235 g) was added over 30 min. If the dark brown reaction mixture became too viscous for stirring before the addition was complete, the ice bath was removed, and the temperature was allowed to rise spontaneously; if necessary, the stirred mixture was warmed to 60° during the final stages of addition. After the addition and any ensuing exothermic reaction were complete, the mixture was further stirred for 2 h at 95°. It was then poured into ice water (2 l) with stirring to give a clear solution of the immonium salt. For hydrolysis, the solution was brought to pH4 by the addition of sodium acetate, and heated on the steam bath for 2 h. The precipitated aldehyde was collected from the cold solution by filtration and crystallised from ethanol–water (1 : 1 *ca.* 1.5 l) giving colourless needles (234 g, 95%), of the aldehyde (**4**), mp 144–145° (lit.[34] mp 145–145.5°). The reaction was carried out consistently on a scale of 100 g to 2500 g to give yields of 90–96%.

*Step* 5. *Ethyl* 4-(*β*,*β*-*dicyanovinyl*)-3,5-*dimethylpyrrole*-2-*carboxylate* (**5**). Ethyl 4-formyl-3,5-dimethylpyrrole-2-carboxylate (**4**) (100 g) and freshly distilled malononitrile (50 g) dissolved in absolute ethanol (700 ml) were warmed to 50° and treated with diethylamine (30 ml) in one lot with stirring. The temperature rose to *ca.* 60°, and within 1 min separation of the crystalline product commenced. After stirring for a further 5 min, the mixture was heated on the steam bath for 5 min, and then cooled to 0°. The product was collected by filtration, washed with small portions of absolute ethanol, and dried overnight in air at 60° giving 119 g (95%) of the product (**5**) as pale yellow matted needles, mp 223–224° (lit.[35] mp 214°). On a scale of 100 g–1000 g the reaction consistently gave yields in the range 95–98%.

*Step* 6. 2 *Methyl* 5 *ethyl* 3-(*β*,*β*-*dicyanovinyl*)-4-*methylpyrrole*-2,5-*dicarboxylate* (**6**). The ester (**5**) (100 g) was suspended in boiling anhydrous methanol [2000 ml, to which water (7.4 ml) had been added] in a 5 l 3-necked r-b flask fitted with two very efficient large bore condensers. Bromine (200 ml) was added during the course of 2 min, or as rapidly as possible. The addition of the bromine

brought about a very violent reaction (vigorous reflux, large amounts of HBr and $Br_2$ vapour: FUME CUPBOARD). During the course of the additions, the suspended (**5**) dissolved to give a clear dark red solution, from which the product shortly began to separate. After the addition, the refluxing was continued for 2 min. The reaction mixture was then rapidly cooled to 5°. The product was removed by filtration and washed thoroughly with small portions of cold methanol (400 ml in total) until it was colourless to give 80.6 g (68%) of the product (**6**) as colourless matted needles, mp 192–193° (lit.[36] mp 187°).

The preparation was carried out consistently on the 100–500 g scale giving yields of 68–70%. On the larger scale it is desirable to introduce seed crystals at the appropriate time if crystals do not form spontaneously.

*Step* 7. 3-*Formyl*-4-*methylpyrrole*-2,5-*dicarboxylic acid* (**7**). The β,β-dicyanovinyl compound (**6**) (200 g) was suspended in aqueous sodium hydroxide (250 g NaOH, 2.5 l water, previously flushed $N_2$) and heated on a steam bath under $N_2$. The suspended material gradually dissolved giving a clear bright yellow solution, and ammonia was evolved. After 3.5 h the mixture was chilled to 10° and carefully acidified to pH2 by dropwise addition of cold conc. HCl with constant vigorous stirring and cooling (⩾20°). The mixture was cooled to 0°. The bulky precipitate was removed by filtration, washed thoroughly with water until it was free of mineral acid, and dried over $CaCl_2$ *in vacuo* at room temperature giving 114 g (83%) of the diacid (**7**) as a pale fawn microcystalline powder, mp decomp. >300°. $\lambda_{max}$ (95% EtOH) (ε) 242 (29 500), 262 nm (13 000). Treatment of the diacid (**7**) with a slight excess of ethereal diazomethane gave the corresponding dimethyl ester (**112**), colourless short needles, mp 182° (lit.[36] mp 180°) from aqueous acetic acid. $\nu_{max}$ ($CHCl_3$) 3380, 1710, 1675 $cm^{-1}$. Excess of ethereal diazomethane gave dimethyl 3-formyl-1,4-dimethylpyrrole-2,5-dicarboxylate, (**113**) colourless long needles, mp 108–109°, from aqueous acetic acid. $\nu_{max}$ ($CHCl_3$) no pyrrolic NH stretch, 1720, 1670 $cm^{-1}$.

*Step* 8. 4-*Methyl*-3-(β-*nitrovinyl*)*pyrrole*-2,5-*dicarboxylic acid* (**8**). 3-Formyl-4-methylpyrrole-2,5-dicarboxylic acid (**7**) (100 g) suspended in absolute alcohol (1 l) was treated with diethylamine (110 ml) and the mixture was warmed for a short while until a clear orange solution was obtained. Freshly distilled nitromethane (32.5 g) was added, and the mixture was refluxed for 2.5 h. The ethanol was evaporated, and the residual yellow, partly crystalline, material was taken up in water (1 l). The solution was cooled to 15° and carefully treated with conc. HCl (1 l) as rapidly as possible consistent with the temperature not rising above 25°. During the course of the acidification a precipitate (crystalline or oily/crystalline) usually formed which redissolved on further addition of acid, and was then replaced by new precipitated material. When the acidification and separation of the second precipitate were complete, the crude product was collected by filtration, washed with water thoroughly, and dried *in vacuo* over calcium chloride. The dry product (95 g) was crystallised from boiling acetic acid (*ca.* 2.5 l) giving the nitrovinylpyrrole diacid as a very pale yellow to yellow–brown microcrystalline powder (77 g, 68%), mp >300°, slow decomp. $\lambda_{max}$ (95% EtOH) (ε) 219 (10 400), 267 sh (21 900), 270 sh (22 200), 272 (22 600), and 335 nm (13 200). The reaction was carried out consistently on the 20–400 g scale with yields of 60–70%. [August 1989. ν(KBr) 3300, *ca.* 2800(b), 1685(s), 1635, 1560, 1515, 1415, 1330, 1265, 1220 $cm^{-1}$].

*Step* 9. 4-*Methyl*-3-(β-*nitroethyl*)*pyrrole*-2,5-*dicarboxylic acid* (**9**). The nitrovinylpyrrole diacid (**8**) (307 g) was dissolved in water (9 l) containing sodium bicarbonate (221 g). Solid sodium borohydride (51.0 g) was added in portions with shaking over 10 min (colour change: yellow–orange → light brown). After a further 30 min at room temperature the mixture was cooled to 10° and carefully acidified to Congo Red with conc. HCl (temp. ⩾20°). The resulting voluminous precipitate was removed by filtration, washed well with water, giving the product (**9**) as a brown powder (270.1 g, 87%). mp: darkened from *ca.* 216°, charred *ca.* 222–226°. Recrystallisation from aqueous methanol gave very pale pink fluffy crystals, mp: darkened *ca.* 215°, charred 226–230°. $\lambda_{max}$ (95% EtOH) (ε) 217 (19 500), 278 nm (20 100).

The acid was characterised as the dimethyl ester (**114**) formed with excess ethereal diazomethane,

which crystallised from methanol as needles, mp 125.5–126°. (Found: C, 48.9; H, 5.4; N, 10.5. $C_{11}H_{14}N_2O_6$ requires C, 48.9; H, 5.2; N, 10.4%). $\lambda_{max}$ (95% EtOH) ($\varepsilon$) 218 (17 100), 280 nm (19 400). $\nu_{max}$ ($CHCl_3$) 3380, 1700, 1545, 1375 $cm^{-1}$.

*Step* 10. 4-*Methyl*-3-(*β-nitroethyl*)*pyrrole* (**10**). 4-Methyl-3-(β-nitroethyl)pyrrole-2,5-dicarboxylic acid (**9**) (24.2 g), sodium acetate trihydrate (36 g), and potassium acetate (36 g) were ground together in a mortar and the intimate mixture was transferred to a 500 ml flask equipped with a short reflux condenser, stirrer, and an inlet through which a nitrogen atmosphere was maintained throughout. The reaction mixture was immersed in an oil bath at 100°, and the temperature was increased. At 115° (bath) liquefaction occurred and decarboxylation commenced. Stirring was started and the bath was maintained at 125–135° until evolution of $CO_2$ had almost ceased (*ca.* 25 min). The reaction mixture was then partitioned between dichloromethane and water. The water layer was washed with two further portions of dichloromethane, and the combined organic extracts were washed (aq. $NaHCO_3$) and dried ($Na_2SO_4$). The solvent was evaporated and the residual greenish-brown oil (10 g) was distilled giving 8.9 g (58%) of the product (**10**) as a colourless oil, bp 127° at 1.5 mm, $\eta_D^{25}$ 1.5200. $\lambda_{max}$ (95% EtOH): end absorption only. $\nu_{max}$ ($CHCl_3$) 3520, 1555, 1385, no band at *ca.* 1700 $cm^{-1}$.

The reaction was carried out reproducibly on a scale of 10–30 g to give yields of 50–58%. The product is air sensitive, and is best used for the next stage as soon as possible.

*Step* 11. 3-(*β-Aminoethyl*)-4-*methylpyrrole* (**11**). The nitro compound (**10**) (9.00 g), methanol (100 ml), and platinum oxide (308 mg) were shaken together under hydrogen in a medium-pressure Parr hydrogenation apparatus, initially at 48.5 lb $in^{-2}$ until no further pressure drop occurred. The theoretical amount of hydrogen was absorbed in 20 min to 3 h, depending on the batch of catalyst. The catalyst was removed, the solvent was evaporated, and the residual oil was distilled (bp 71–68° at 0.04 mm) giving the colourless amino compound (6.77 g) (93%) with $\eta_D^{27.7}$ 1.5316. $\lambda_{max}$ (95% EtOH): end absorption only. $\nu_{max}$ ($CHCl_3$) 3600(s), 3460, 3255, 1590, 1445 $cm^{-1}$. On a scale of 3 g–12 g the reduction gave yields of 85–93%. The amine (**11**) could also be prepared (50–60% yield) by reduction of the nitro compound with lithium aluminium hydride in boiling ether.

The amine (**11**) rapidly takes up carbon dioxide in air: it is stored cold under nitrogen. It was characterised as the N-acetyl derivative prepared by reaction with acetic anhydride in benzene/triethylamine.

3-(*β-Acetylaminoethyl*)-4-*methylpyrrole* (**111**), colourless prisms, mp 81–81.5° from benzene. (Found: N, 16.75. $C_9H_{14}N_2O$ requires N, 16.85%). $\lambda_{max}$ (95% EtOH)—end absorption only. $\nu_{max}$ ($CHCl_3$) 3510(s), 3355, 1670 $cm^{-1}$.

9.3. *Synthesis of ring B*

*Step* 12. *Ethyl* 4-*acetyl*-3,5-*dimethylpyrrole*-2-*carboxylate* (**12**). Aluminium chloride (370 g) was carefully added to ethyl 3,5-dimethylpyrrole-2-carboxylate (**3**) (370 g) in dichloromethane (3.5 l). Acetyl chloride (337 g) was then added rapidly, and the reaction mixture was stirred and heated under reflux for 2 h. Water (750 ml) was added to the cooled mixture, and the bulk of the solvent was removed *in vacuo*. The residue was taken up in the minimum of hot ethanol. From the cooled solution a first crop of pure product (**12**) (288 g, mp 144–147°, lit.[37] mp 143–144°) crystallised. The mother liquor gave a further 109 g of material of the same quality after two crystallisations with clarification with Florisil and charcoal. Total yield 86%.

*Step* 13. 3-*Ethyl*-2,4-*dimethylpyrrole* (*Kryptopyrrole*) (**13**). This was made by the reaction of sodium hydroxide and hydrazine hydrate with ethyl 4-acetyl-3,5-dimethylpyrrole-2-carboxylate (**12**) following the procedure of Treibs and Schmidt.[40]

*Step* 14. 4-*Ethyl*-2-*formyl*-3,5-*dimethylpyrrole* (**14**). A cooled mixture of phosphorus oxychloride (242 g) and freshly distilled dimethylformamide (450 ml) was added during 20 min to a solution of kryptopyrrole (**13**) (176 g) in freshly distilled dimethylformamide (450 ml) previously cooled to −30° in an acetone/solid $CO_2$ bath (temp $\not>$ −25°). When the addition was complete the yellow

solution was poured into ice-cold 5% aqueous sodium hydroxide (8 l) and stirred for 1 h. The aldehyde crystallised as yellow plates, which were removed by filtration, washed with water, and dissolved in boiling ether (*ca.* 2 l). The solution was concentrated to about half volume, diluted with an equal volume of hexane, and again concentrated to about half volume. The product (**14**) crystallised in two crops of colourless prisms, 180 g, mp 100–102° (lit.[69] mp 105–106°). A further 12 g of pure material was obtained from the mother liquors after treatment with silica gel and concentration. Total yield 89%. $\lambda_{max}$ (85% EtOH) ($\varepsilon$) 270–280 sh (6 600), 312 nm (21 000).

*Step* 15. 2-(*β,β-Dicyanovinyl*)-4-*ethyl*-3,5-*dimethylpyrrole* (**15**). The method was that of Fischer and Neber.[41] The foregoing aldehyde (**14**) (305 g) and freshly distilled malononitrile (147 g) in 95% ethanol (2 l) were treated with diethylamine (2 ml) with swirling. The mixture warmed spontaneously, and separation of the product began at once: the mixture was warmed gently on the steam bath for 1 h, and then allowed to stand overnight. The product was collected and washed with ethanol (*ca.* 1 l) giving beautiful yellow needles (391 g, 97%) of the dicyanovinyl derivative (**15**), mp 191.5° (lit.[41] mp 191.5°). $\lambda_{max}$ (EtOH) ($\varepsilon$) 312 (4 300), 403 nm (43 200). $\nu_{max}$ ($CHCl_3$) 3410, 2210(s), 1595, 1530 $cm^{-1}$.

*Step* 16. 5-*Chloromethyl*-2-(*β,β-dicyanovinyl*)-4-*ethyl*-3-*methylpyrrole* (**16**). A stirred suspension of the pyrrole (**15**) (70 g) in glacial acetic acid (2 l) was heated to 55°, and freshly distilled colourless sulphuryl chloride (50 g) was added dropwise during 15 min. Stirring was continued at 55° for 1 h more. At no time did the solid dissolve completely, but as the reaction proceeded prisms of the starting material were replaced by needles of the product.

The reaction mixture was cooled to 20°, the product was removed by filtration, washed with acetic acid (2 × 100 ml), and light petroleum (bp 30–60°, 2 × 250 ml) and then dried in air giving the chloromethyl compound (**16**) (69 g, 84%) as well-formed needles, mp 189–192°, decomp. (Found: C, 62.55; H, 5.25; N, 18.55; Cl, 14.7. Calcd. for $C_{12}H_{12}ClN_3$: C, 61.65; H, 5.2; N, 18.0; Cl 15.15%). $\nu_{max}$ (KI) 3310, 2200(s), 1580, 1540 $cm^{-1}$.

On warming (**16**) in ethanol for a short time it was converted to 2-(*β,β-dicyanovinyl*)-5-*ethoxymethyl*-4-*ethyl*-3-*methylpyrrole* (**115**), yellow needles, mp 109.5–110.5°, from ethanol. (Found: C, 69.4; H, 7.05; N, 17.1. $C_{14}H_{17}N_3O$ requires C, 69.1; H, 7.05; N, 17.25%). $\nu_{max}$ (KI) 3400, 2205(s), 1600, 1535 $cm^{-1}$.

9.4. *Synthesis of ring C*

*Step* 17. 3,5-*Diethoxycarbonyl*-4-*methylpyrrole*-2-*carboxylic acid* (**17**). This was prepared from Knorr's pyrrole (**1**) using Corwin's method[46] (successive treatment with bromine and sulphuryl chloride, followed by hydrolysis) giving the desired acid (**17**), mp 150–151° (lit.[46] mp 150°) in yields of 70–75% using 1 kg of starting material. Addition of 5% by weight of acetic anhydride to the glacial acetic acid solvent was advantageous in that it resulted in lower working temperatures. In addition to the desired acid, 13% of 3,5-diethoxycarbonyl-2-formyl-4-methylpyrrole, mp 124–125° (lit.[46] mp 125°) was reproducibly formed as a by-product.

*Step* 18. *Diethyl* 3-*methylpyrrole*-2,4-*dicarboxylate* (**18**). The monocarboxylic acid (**17**) (100 g) and copper bronze powder (5 g) were heated at 230–240° for 1 h in a flask equipped with a condenser, to allow sublimed material to be retained. The flask was then equipped for distillation, and the product distilled at *ca.* 185°/15 mm. When the yellow distillate was redistilled from a small amount of copper bronze, a colourless liquid was obtained, which soon solidified. Crystallisation from aqueous ethanol gave colourless prisms (60 g, 72%), mp 91–92° (lit.[46] mp 91°).

*Step* 19. 4-*Ethoxycarbonyl*-3-*methylpyrrole*-2-*carboxylic acid* (**19**). The method of Corwin and Viohl[48] (80% aqueous ethanolic KOH) was successfully carried out on a 100–360 g scale to give 90% of the monocarboxylic acid, mp 228–231° (lit.[48] mp 230°).

*Step* 20. *Ethyl* 4-*methylpyrrole*-3-*carboxylate* (**20**). The acid (**19**) (125 g) and copper bronze powder (15g) were heated to 220–240°, slowly at first, when a vigorous reaction began. Heating at 220–240° was continued for a total reaction time of 1 h. The flask was equipped for distillation, and

the product was distilled at the water pump. The colourless distillate was dissolved in hot hexane, from which the product (**20**) crystallised as prisms (77 g, 80%), mp 72–74° (lit.[48] mp 73°). $\lambda_{max}$ (EtOH) ($\varepsilon$) 231 (9 500), 253 sh. nm (4 500). $\nu_{max}$ ($CH_2Cl_2$) 3560, 1700 $cm^{-1}$.

9.5. *Synthesis of ring D*

*Step* 21. *5-Ethoxycarbonyl-2,4-dimethylpyrrole-3-acrylic acid* (**21**). This was prepared by the decarboxylative condensation of ethyl 4-formyl-3,5-dimethylpyrrole-2-carboxylate (**4**) with malonic acid in the presence of aniline according to Fischer and Andersag.[49] In those parts of the procedure in which basic media are used, excess basicity should be avoided to prevent hydrolysis of the α-ester group. The crude product was recrystallised from ethanol to give the pure acid, mp 245°, decomp. (lit.[49] mp 240°, decomp.) in 70–75% yield.

*Step* 22. *5-Ethoxycarbonyl-2,4-dimethylpyrrole-3-propionic acid* (**22**). The corresponding carboxyvinyl compound (**21**) (53 g) dissolved in an equivalent amount of 10% aqueous sodium hydroxide was shaken with hydrogen over Raney nickel (3 g) at room temperature in a Parr hydrogenation apparatus at an initial pressure of 30 lb $in^{-2}$ until hydrogen uptake ceased (*ca.* 12 h). The catalyst was removed by filtration through Celite, the latter being washed with water. The combined filtrates were acidified (5 M-HCl) whereupon a voluminous crystalline precipitate formed. The product was washed with water and recrystallised from aqueous ethanol giving pale pink leaflets (48 g, 90%) of the product (**22**), mp 155–156° (lit.[50] mp 156–158°, 152°, dimorphic).

The reaction was carried out reproducibly on a 50–300 g scale giving yields of 90–92%.

*Step* 23. *2-Carboxy-5-ethoxycarbonyl-4-methylpyrrole-3-propionic acid* (**23**). The pyrrole propionic acid (**22**) (70 g) suspended in dry ether (600 ml) at −15° was treated dropwise with freshly distilled colourless sulphuryl chloride (122 g) with stirring during 1 h (temp ≯ −15°) giving a clear yellow solution. Stirring was continued for 1 h more at −15° and then for 4 h at 0° (separation of crystals). The mixture was treated with water (200 ml) at 0–5°, and allowed to stand at room temperature overnight (crystalline precipitate). The ether was removed under reduced pressure, and the precipitate was removed by filtration. Trituration of the solid with hot acetone (1 l) left a first crop of the product (**23**) as colourless to very pale yellow prisms (30.5 g, mp 242°, decomp. lit.[52] mp 243°). Concentration of the acetone successively to 300 ml and to 100 ml gave two further crops (23 g, mp 243°, decomp.; 15.5 g, mp 242°, decomp.) of the product as very pale yellow needles. Total yield 69 g (88%).

This reaction was carried out reproducibly on a 50–200 g scale giving yields of 85–90%.

*Step* 24. *Methyl 4-methylpyrrole-3-propionate* (**24**). The following procedure is essentially that of Fischer[52,53] but modified in detail.

A solution of the pyrrole dicarboxylic acid (**23**) (80 g) in 10% aqueous sodium hydroxide (700 ml) was heated in an autoclave at 180° for 90 min. The cooled solution was evaporated under reduced pressure to a small volume and carefully acidified with 50% aqueous sulphuric acid to pH2, with vigorous stirring and cooling. The precipitated solids (= product + $Na_2SO_4$) were filtered off, and the filtrate was extracted with ether (200 ml). The solids were added to the ether extract, which was diluted with more ether (400 ml). Water (100 ml) was added, the phases were separated, and the ethereal extract was washed with a further portion of water (100 ml). The combined aqueous extracts were back-washed with a small quantity of ether, and the combined organic extracts were dried over $Na_2SO_4$. Methanol (50 ml) was added, followed by a slight excess of ethereal diazomethane. The solution was washed with aqueous sodium hydrogencarbonate and with water, and taken to dryness.

The material from two such runs was combined and distilled (bp 91–94° at 0.05 mm, lit.[70] bp 152° at 11.0 mm) giving methyl 4-methylpyrrole-3-propionate (**24**) (78 g, 78%). The reaction was carried out reproducibly on a 50–200 g scale to give yields of 75–80%.

The corresponding ethyl ester (**118**) was prepared in comparable yield by substituting ethanol/

excess diazoethane for methanol/diazomethane in the above procedure. Bp 100–101° at 0.04 mm, $\eta_D^{23}$ 1.4901.

The corresponding free acid (**117**) was obtained from the above ether extract following hydrolysis-decarboxylation: the ether was removed and the residue was crystallised from chloroform-hexane giving colourless prisms (82%) of 4-methylpyrrole-3-propionic acid (**117**, opsopyrrole carboxylic acid) mp 116–118° (lit.[52] mp 119°).

*Step* 25. *Methyl 5-formyl-4-methylpyrrole-3-propionate* (**25**). To freshly distilled dimethylformamide (10 ml) at 0° was added freshly distilled phosphorus oxychloride (10 ml). To the resulting viscous yellow complex (which sometimes solidified) was added freshly distilled 1,2-dichloroethane (80 ml). The mixture was allowed to warm up to room temperature giving a clear orange solution. This was cooled to −10° and methyl 4-methylpyrrole-3-propionate (**24**) (10.3 g) in freshly distilled 1,2-dichloroethane (15 ml) was added dropwise over 20 min with vigorous stirring. The residual ester in the addition funnel was washed in with a further portion (5 ml) of 1,2-dichloroethane. The cooling bath was then removed, and the clear orange reaction mixture was stirred for an additional 10 min. The mixture was then transferred to a 500 ml r-b flask ($CH_2Cl$–$CH_2Cl$ washings) and the bulk of the solvent was removed under reduced pressure (temp ⩾30°). The dark orange residue was transferred (MeOH washing) to a 1 l beaker, ice-cold 2M-NaOH (500 ml) was added, and the mixture was heated almost to boiling for *ca.* 20 min in order to hydrolyse the reaction product and volatilise residual solvents and methanol. A trace of dark oily material which did not pass into solution was removed by extraction of the cooled aqueous mixture with ether (2 × 100 ml). The resulting orange solution was heated briefly to remove ether, treated with charcoal, cooled and filtered. Careful acidification of the resulting yellow solution at 0° to pH 3.5 with 5 M-HCl gave a crystalline precipitate which was collected after 15 min, washed well with several portions of cold water, and dried *in vacuo* at room temperature giving 7.04 g (63%) of 5-formyl-4-methylpyrrole-3-propionic acid (**24A**) as off-white microneedles, mp 150–152°. The acid was recrystallised from ether–ethanol with excellent recovery giving very pure material as needles, mp 154–155° (lit.[54] mp 151°).

The combined aqueous filtrate and washings from the separation of the above acid were continuously extracted with ether for 18 h. The yellow extract was dried ($Na_2SO_4$), treated with charcoal, and evaporated. The residue was dissolved in 2M-NaOH and acidified to pH 3.5 at 0° as described for its isomer. A pale tan solid (1.61 g, 15%, mp 112–116°) separated. Recrystallisation from ether gave pure 2-formyl-4-methylpyrrole-3-propionic acid (**119**) as large colourless prisms, mp 125°.

Pure 5-formyl-4-methylpyrrole-3-propionic acid (0.47 g, mp 153–155°) in dry methanol (45 ml) was treated with excess ethereal diazomethane and kept for 15 min at room temperature. The solvents were removed under reduced pressure at room temperature, the residual oil was dissolved in ether, and hexane was added to the boiling solution giving 0.21 g of methyl 5-formyl-4-methylpyrrole-3-propionate (**25**) as colourless needles, mp 76°. A further quantity of product (0.15 g, mp 74–75°) was obtained from the mother liquor. Total yield 71%. For analysis, crystallised thrice from light petroleum (bp 30–60°), colourless needles, mp 77° (lit.[54] mp 77°). (Found: C, 61.55; H, 6.45; N, 6.9. Calcd. for $C_{10}H_{13}NO_3$: C, 61.5; H, 6.7; N, 7.2%). [August 1989, $\nu_{max}$ (KBr) 3240, 1740, 1640, 1435, 1365, 1200, 800, 618 $cm^{-1}$. $\delta$ ($CDCl_3$) 9.60 (s, CHO), 9.14 (b, NH), 6.88 (d, J ~ 1 Hz, 2-H), 3.68 (s, OMe), 2.76 (t, J = 8 Hz, $CH_2CH_2CO_2Me$), 2.56 (t, J = 8 Hz, $CH_2CH_2CO_2Me$), 2.30 (s, 4-Me). nOe: irrad. 9.60 $\delta$, positive nOe at 2.30. $R_f$ = 0.63 ($CHCl_3$: acetone = 2:1)].

The isomeric acid (**119**, 0.52 g) was similarly esterified with diazomethane in ether/methanol giving *methyl 2-formyl-4-methylpyrrole-3-propionate* (**116**) which crystallised from hexane as glistening plates (0.45 g, 86%), mp 91°. Mixed mp of (**25**) and (**116**) *ca.* 50°. (Found: C, 61.45; H, 6.5; N, 7.1%). [August 1989. $\nu_{max}$ (KBr) 3240, 1730, 1655, 1440, 1360, 1295, 1180, 775 $cm^{-1}$. $\delta$ ($CDCl_3$) 9.62 (s, CHO), 9.08 (b, NH), 6.84 (bs, 5-H), 3.66 (s, OMe), 3.05 (t, J = 8 Hz, $CH_2CH_2CO_2Me$), 2.58 (t, J = 8 Hz, $CH_2CH_2CO_2Me$), 2.06 (s, 4-Me). nOe: irrad. 9.62 $\delta$, strongly positive effect at 3.05, weak positive effect at 2.58. $R_f$ = 0.63 ($CHCl_3$: acetone = 2:1)].

The ethyl ester (**120**) corresponding to the major isomer (**25**) is best obtained by esterification of the pure acid (**24A**) mp 154–155°, prepared as described above, with diazoethane in ethanol/ether. Bold prisms, mp 71–72°, from ether–hexane.

9.6. *Left-hand component*

*Step* 26. *4-(β-Aminoethyl)-4′-(β-methoxycarbonylethyl)-3,3′-dimethylpyrromethene dihydrobromide monohydrate* (**26**), *(8-(β-aminoethyl)-2-(β-methoxycarbonylethyl)-3,7-dimethyldipyrrin dihydrobromide monohydrate*). 3-(β-Aminoethyl)-4-methylpyrrole (**11**) (0.71 g) was dissolved in methanol (5 ml). Freshly prepared 3.15 M-aqueous methanolic hydrogen bromide (5 ml; $H_2O$: MeOH = 2.5 : 100) was added, and the mixture was rapidly cooled to −25° ($CO_2$-acetone). Without delay a few seed crystals of product were added to the vigorously stirred solution, followed by a solution of methyl 5-formyl-4-methylpyrrole-3-propionate (**25**) (1.11 g) in methanol (5 ml) and ether (1 ml). An orange yellow precipitate formed at once and the internal temperature rose to *ca.* 0°. The mixture was cooled to −25° over a few min, and treated with 1.6 M-aqueous methanolic hydrogen bromide (*ca.* 20 ml, $H_2O$: MeOH = 1 : 100) previously cooled to −60°. The slurry was then filtered as rapidly as possible, and the solid was washed with several small portions of the cold (−60°) 1.6 M-aqueous methanolic HBr. During the washing process the filter cake was broken up and ground with a spatula to a homogenous paste in order to facilitate rapid removal of the wash liquors. As soon as the cake had been sucked dry it was washed with ether–methanol (10 : 1, −60°), and finally with anhydrous ether. The dry crystalline pyrromethene (1.8 g) was recrystallised from hot 1.3 M-methanolic hydrogen bromide (60 ml) giving the pyrromethene dihydrobromide (**26**) as orange red crystals (1.55 g, 56%), mp 200–202°, decomp. (Found : C, 42.55 ; H, 5.55 ; N, 8.6 ; Br, 34.1. $C_{17}H_{25}Br_2N_3O_2 . H_2O$ requires C, 42.45 ; H, 5.65 ; N, 8.75 ; Br, 33.2%). $\lambda_{max}$ (0.05 M-HBr/MeOH) (ε) 373 (8700), 467 nm (84 500). $\nu_{max}$ (KBr) 3495, 1745, 1639, 1543, 1285, 1185, 1135, 920, 810 $cm^{-1}$.

*4-(β-Aminoethyl)-4′-(β-ethoxycarbonylethyl)-3,3′-dimethylpyrromethene dihydrobromide monohydrate* (**121**). *(8-(β-aminoethyl)-2-(β-ethoxycarbonylethyl)-3,7-dimethyldipyrrin dihydrobromide monohydrate*). The corresponding ethyl ester was prepared analogously from the corresponding pyrrole propionic acid ethyl ester (**120**), using ethanol in place of methanol. The pyrromethene dihydrobromide crystallised from 1 M-HBr/EtOH containing a small amount of water as small pale orange leaflets, mp 219–221°, decomp. Analytical sample, mp 224–225°, decomp. after four recrystallisations from 1 M-HBr/EtOH containing a small amount of water. (Found : C, 43.65 ; H, 5.95 ; N, 8.7 ; Br, 31.35. $C_{18}H_{27}Br_2N_3O_2 . H_2O$ requires C, 43.65 ; H, 5.9 ; N, 8.5 ; Br, 32.3%). $\lambda_{max}$ (1 M-HBr/EtOH) (ε) 370 (7 400), 469 nm (82 000). $\nu_{max}$ (KBr) 3450, 1735, 1630, 1465, 1425, 920, 810 $cm^{-1}$.

*4,4′-Di-(β-ethoxycarbonylethyl)-3,3′-dimethylpyrromethene hydrobromide* (**122**), *(2,8-di-(β-ethoxycarbonylethyl)-3,7-dimethyldipyrrin hydrobromide*). Ethyl 5-formyl-4-methylpyrrole-3-propionate (**120**, 621 mg) and ethyl 4-methylpyrrole-3-propionate (**118**, 538 mg) were dissolved in absolute ethanol (12 ml) at room temperature. 1 M-Ethanolic hydrogen bromide (6 ml) was added in one lot with shaking, when a bulky orange precipitate separated at once. The mixture was chilled in ice (5 min) and the product was collected by filtration, washed free of the dark red mother liquor with 1 M-ethanolic hydrogen bromide (5 × 1 ml) and then with anhydrous ether (4 × 5 ml), giving matted orange leaflets (930 mg, 69%) of the pyrromethene hydrobromide, mp 130–133°. For analysis the compound was recrystallised with substantial loss from hot 1 M-HBr/EtOH ; mp 134–137° decomp. (Found : C, 55.8 ; H, 6.65 ; N, 5.95 ; OEt, 18.65. $C_{21}H_{29}BrN_2O_4$ requires C, 55.65 ; H, 6.45 ; N, 6.2 ; OEt, 19.85%. $\lambda_{max}$ (M-HBr/EtOH) (ε) 373 (7 700), 471 nm (76 500). $\nu_{max}$ ($CHCl_3$) 3460, 3140, 2950, 1735, 1630, 1470, 1280, 1165, 1130, 910 $cm^{-1}$.

9.7. *Right-hand component*

*Step* 27. *5′-(β,β-Dicyanovinyl)-4-ethoxycarbonyl-3′-ethyl-3,4′-dimethyldipyrrylmethane* (**27**), *(1-(β,β-dicyanovinyl)-8-ethoxycarbonyl-3-ethyl-2,7-dimethyldipyrrane*). A mixture of the chloromethyl-

pyrrole (**16**) (66.4 g), ethyl 4-methylpyrrole-3-carboxylate (**20**, 47.8 g), 95% ethanol (600 ml), water (300 ml) and conc. HCl (30 ml) was slowly heated to reflux with swirling. The suspension was refluxed for 1 h, cooled to 40°, and the solid material was removed by filtration. The filter cake was washed with 60% ethanol (100 ml) and the combined dark coloured filtrates were stored in the refrigerator for 3 h. The dark brown prisms which separated were recrystallised from ethanol and combined with the main solid product.

The solid was suspended in boiling 95% ethanol (3 l) and heated until most of the material dissolved. The insoluble material was boiled with fresh 95% ethanol (1 l) and the insoluble material (*ca.* 3 g), mp 248–252°, consisting largely of the tripyrrane (**124**) was filtered off. The combined ethanolic filtrates were concentrated to *ca.* 1500 ml and allowed to cool overnight at room temperature in the dark. The total crystalline product was then subjected to systematic fractional dissolution and crystallisation from ethanol giving:

(i) the more soluble, crystalline, component, the dipyrrylmethane (**27**), fine golden prisms, 54.5 g (55%) mp 191–193°. (Found: C, 68.55; H, 6.5; N, 15.7. $C_{20}H_{22}N_4O_2$ requires C, 68.55; H, 6.35; N, 16.0%. $\lambda_{max}$ (EtOH) 402 nm. $\nu_{max}$ (KI) 3380, 3225, 2205, 1665, 1585, 1530, 1410, 1325, 1240, 1185, 1160, 1070, 970, 910, 775, 765 $cm^{-1}$.

(ii) the less soluble material, which was recrystallised from chloroform–ethanol giving the by-product, the tripyrrane (**124**), (1,14-bis(*β*,*β*-dicyanovinyl)-7-ethoxycarbonyl-3,12-diethyl-2,8,13-trimethyltripyrrane) as small yellow prisms, mp 252–255°, decomp., in 7.5 g (10%) yield. (Found: C, 70.4; H, 6.2. Calcd. for $C_{32}H_{33}N_7O_2$ requires C, 70.2; H, 6.1%). $\nu_{max}$ ($CHCl_3$) 3300, 2220, 1680 $cm^{-1}$.

For comparison purposes, the tripyrrane was prepared as follows. The dipyrrylmethane (**27**) (1 g) and the chloromethylpyrrole (**16**) (0.8 g) were refluxed on a steam bath in ethanol (20 ml) containing conc. HCl (1 ml). A clear reddish solution was obtained, from which yellow prisms began to crystallise. After 1 h reflux the solution was cooled and the product filtered off and washed with aqueous ethanol giving 1.2 g (77%) of the tripyrrane (**124**) as bright yellow prisms, mp 248–254°. Recrystallisation ($CHCl_3$-EtOH) raised the mp to 252–254°. The substance was indistinguishable from the by-product of the above reaction.

*Step* 28. *5′(β,β-Dicyanovinyl)-4-ethoxycarbonyl-3′-ethyl-5-(β-methoxycarbonylpropionyl)-3,4′-dimethyldipyrrylmethane* (**28**), *(1-(β,β-dicyanovinyl)-8-ethoxycarbonyl-3-ethyl-9-(β-methoxycarbonylpropionyl)-2,7-dimethyldipyrrane*). To the α-unsubstituted dipyrrylmethane (**27**) (70 g) and freshly distilled *β*-methoxycarbonylpropionyl chloride (70 g) in dichloromethane (2 l) was added quickly, in one lot, zinc chloride (40 g, previously fused and poured hot into hexane). The mixture was stirred vigorously under reflux for 2 h, when it had become dark red in colour. With continued stirring the mixture was cooled in ice and treated dropwise with saturated aqueous sodium hydrogencarbonate (750 ml) (care: frothing). The dark organic layer was separated, and washed thoroughly with aqueous sodium hydrogencarbonate and with water. The extract was dried ($MgSO_4$) and concentrated until not quite all the solvent had been removed. The residue was taken up in hot ether (2 × 1 l), from which the product shortly began to crystallise. After 24 h the first crop (69 g) was obtained which on recrystallisation from ethanol gave the acyldipyrrylmethane (**28**) as yellow prisms (65 g), mp 133–135°. Concentration of the original ethereal solution gave, after recrystallisation from ethanol, further pure material, 8 g, mp 134–135°. Total yield = 73 g (79%). (Found: C, 65.3; H, 6.1; N, 11.95. $C_{25}H_{28}N_4O_5$ requires C, 64.65; H, 6.1; N, 12.05%) $\lambda_{max}$ (95% EtOH) (*ε*) 311 (17 700), 406 nm (43 500). $\nu_{max}$ ($CHCl_3$) 3380, 2200, 1725, 1640, 1590 $cm^{-1}$. [August 1989. *δ* ($CDCl_3$) 9.57, 9.26 (2 × 1 Hbs, NH), 7.36 (s, = CH—), 4.40 (q, J = 8 Hz, O $CH_2$Me), 4.00 (s, *meso*-$CH_2$), 3.66 (s, O*Me*), 3.30 (t, J = 8 Hz, $COCH_2CH_2CO_2Me$), 2.64 (t, J = 8 Hz, $COCH_2CH_2CO_2Me$), 2.43 (q, J = 8 Hz, $ArCH_2Me$), 2.22, 2.17 (2 × 3 Hs, Ar-*Me*), 1.40 (t, J = 8 Hz, $OCH_2CH_3$), 1.06 (t, J = 8 Hz, Ar $CH_2Me$). nOe: irrad 4.00 *δ*, strongly positive effect at 9.57, 9.26, 2.43, 2.22 and 1.06 *δ*.]

*Step* 29. *3′-Ethyl-5′-formyl-4-methoxycarbonyl-5-(β-methoxycarbonylpropionyl)-3,4′-dimethyldipyrrylmethane* (**29**), (*3-ethyl-1-formyl-8-methoxycarbonyl-9-(β-methoxycarbonylpropionyl)-2,7-dimethyldipyrrane*). Water (675 ml) in a 2 l Ehrlenmeyer flask was deoxygenated with a vigorous stream of deoxygenated nitrogen for 15 min. Sodium hydroxide (225 g) was added, and the solution was heated to 95° (steam bath) with nitrogen flushing. The dicyanovinyldipyrrylmethane (**28**) (15 g, finely powdered) was added in one lot, and the suspension was heated under nitrogen for 15 min with occasional swirling. The yellow starting material dissolved within 2 min to give a dark brown solution, but at the end of the heating period the colour had become paler. The mixture was cooled rapidly to 10°, carefully acidified with 5 M-hydrochloric acid (*ca.* 1250 ml, temperature ≯20° during neutralisation) while nitrogen was flushed through the suspension. The precipitate was filtered off, washed with water (500 ml) and dried thoroughly *in vacuo* to give the crude off-white dicarboxylic acid. The dried acid was finely powdered, suspended in anhydrous methanol (200 ml) and treated with excess freshly prepared ethereal diazomethane (not more than 90 min, with stirring). The acid went into solution gradually and was replaced by new crystalline material. The ether was removed under reduced pressure, the methanol suspension was cooled, and the white product (11.5 g, mp 197–200°) was collected. Recrystallisation from benzene gave the dipyrrylmethane dimethyl ester (**29**) as colourless fine needles (10.7 g, 84%) mp 199–201°. Analytical sample from benzene, mp 202–204°. (Found: C, 62.8; H, 6.6; N, 6.8; O, 23.65. $C_{21}H_{26}N_2O_6$ requires C, 62.7; H, 6.5; N, 6.95; O, 23.85%). $\lambda_{max}$ (EtOH) (ε) 270 sh (10 800), 320 nm (26 700). $\nu_{max}$ ($CH_2Cl_2$) 3255, 1730, 1705, 1640, 1615 $cm^{-1}$.

The corresponding diethyl ester (**125**), mp 163.5–164° was obtained analogously but using diazoethane/ethanol. Analytical sample, mp 165–165.5° from benzene. (Found: C, 64.3; H, 7.2; N, 6.3. $C_{23}H_{30}N_2O_6$ requires C, 64.15; H, 7.0; N, 6.5%) $\nu_{max}$ ($CH_2Cl_2$) 3390, 3245, 1730, 1645, 1620 $cm^{-1}$.

*Step* 30. *3′-Ethyl-5′-(N-ethylformimino)-4-methoxycarbonyl-5-(β-methoxycarbonylpropionyl)-3,4′-dimethyldipyrrylmethane hydrobromide* (**30**), (*3-ethyl-1-(N-ethylformimino)-8-methoxycarbonyl-9-(β-methoxycarbonylpropionyl)-2,7-dimethyldipyrrane hydrobromide*). The aldehyde (**29**) (10.4 g) was dissolved in anhydrous ethylamine (200 g) and glacial acetic acid (3 ml) at 0°, and kept at 0° for 45 min. Rapid evaporation (water pump) gave a partly crystalline residue which was shaken vigorously with dichloromethane (130 ml) and 2 M-HBr (200 ml) for 2 min. (Check aqueous layer is acidic: protracted exposure to ethylamine must be avoided since it causes some amidation of the alkanoic ester function). After further extractions with dichloromethane (2 × 40 ml) the combined organic extracts were dried ($Na_2SO_4$, 20 min) without preliminary washing. The solvent was removed from the filtrate at the water pump, and the residual white foam was crystallised from benzene giving colourless very fine needles (12.4 g, 94%) of the azomethine hydrobromide (**30**), mp 100°. (Found: C, 54.1; H, 6.35; N, 8.35; Br, 15.2. $C_{23}H_{32}BrN_3O_5$ requires C, 54.1; H, 6.3; N, 8.25; Br, 15.65%). $\lambda_{max}$ (2 M-HBr: EtOH = 1:4) (ε) 306 (19 500), 350 nm (33 700). $\nu_{max}$ ($CH_2Cl_2$) 3100 (b), 1725, 1665 $cm^{-1}$.

The corresponding free Schiff's base was readily obtained when the benzene solution of the above hydrobromide was shaken briefly with M-KOH. Colourless fine needles from cyclohexane, mp 85–87°. $\lambda_{max}$ (95% EtOH) (ε) 306 nm (26 600). $\nu_{max}$ ($CH_2Cl_2$) 3400, 3245, 1705, 1625 $cm^{-1}$.

*Step* 31. *3′-Ethyl-4-methoxycarbonyl-5-(β-methoxycarbonylpropionyl)-3,4′-dimethyl-5′-thioformyldipyrrylmethane* (**31**), (*3-ethyl-8-methoxycarbonyl-9-(β-methoxycarbonylpropionyl)-2,7-dimethyl-1-thioformyldipyrrane*). A stirred solution of the Schiff's base hydrobromide (**30**) (7.1 g) in benzene (350 ml) and anhydrous methanol (350 ml) was saturated with hydrogen sulphide at room temperature. Methanolic sodium methoxide (1.5 M, 0.5 ml) was added (deep red colouration), and thioaldehyde formation was complete within 5 min, as evidenced by the electronic spectrum of an aliquot (0.1 ml → 25 ml EtOH) which showed $A_{400}/A_{315} = 2.28$. Without delay, most of the dissolved hydrogen sulphide was removed by passing a stream of nitrogen through the mixture under reduced pressure. The solution was diluted with benzene (300 ml) and extracted with ice water (1 × 400 ml,

2 × 250 ml) to remove methanol and ethylamine hydrobromide. The benzene solution was dried ($Na_2SO_4$, 10 min) and evaporated quickly at the water pump. (Occasionally the thioaldehyde crystallised during the drying stage: it was redissolved by warming.) The crystalline residue was dissolved in boiling benzene (90 ml), and cyclohexane (40 ml) was added to the hot solution: further portions of cyclohexane (total 70 ml) were added as the solution cooled. The thioaldehyde (**31**) crystallised as yellow–orange needles (5.04 g, 86%), mp 145–146°. (The elemental analysis has not been located). $\lambda_{max}$ (95% EtOH) ($\varepsilon$) 315 (16 000), 400 nm (37 000). $\nu_{max}$ ($CH_2Cl_2$) 3450, 3335, 1725, 1645, 1555 $cm^{-1}$. [August 1989. $R_f = 0.59$ with streaking ($CHCl_3$: MeOH = 30:1). The mass spectrum (FAB, *p*-nitrobenzyl alcohol) indicated that the sample was no longer pure, but included a strong peak at $m/z = 419.164$. (Calcd. for $C_{21}H_{26}N_2O_5S+H$, M = 419.164)].

9.8. *Porphyrin ring formation*

*Steps* 32,33,34,35. 8-(*β-Acetylaminoethyl*)-13-*ethyl*-18-*methoxycarbonyl*-3,7,12,17-*tetramethyl*-*porphyrin*-2,20-*dipropionic acid dimethyl ester* (**35**). (*Porphyrin* **35**). The following procedure was established through very extensive experimentation. For its success, adherence to detail is essential. This arises because of the extreme sensitivity of the dipyrrylmethanes (**32**) and (**33**), which must be manipulated as rapidly as possible in the absence of oxygen and avoiding premature contact with acid. The method has been used many times with reproducible results in the hands of several investigators, for the preparation in all of more than 50 g of the porphyrin (**35**).

*Reagents and apparatus.* Nitrogen was deoxygenated by passage through Fieser's solution, and was then passed successively through saturated aqueous lead acetate, conc. $H_2SO_4$, and sodium hydroxide/Drierite. Dichloromethane was washed with aqueous $NaHCO_3$, distilled and stored under nitrogen. Distilled water was boiled and then allowed to cool under nitrogen flush, and was used throughout. Sodium carbonate solution was made by dissolving anhydrous sodium carbonate (10 g) in nitrogen-flushed distilled water (100 ml). Methanol contained 0.01–0.1% water. All solvents and solutions were flushed with nitrogen immediately before use.

Methanolic hydrogen chloride (12–13 M) was prepared just before use by saturating methanol, cooled in an ice bath, with gaseous hydrogen chloride purified by passage through conc. $H_2SO_4$. An aliquot was titrated to check that the correct concentration had been achieved. The quantity required for step 34 was measured by pouring into a graduated cylinder, the correct amount being then poured into a 500 ml two-necked reaction flask, equipped with calcium chloride drying tube. (While these pouring operations seem to negate other operations conducted anhydrous under nitrogen, it was found by experience that yields were depressed by 10–15% without them.)*

The apparatus shown in Fig. 19 was assembled, and thoroughly flushed with nitrogen.

*Step* 32. A solution of the pyrromethene dihydrobromide (**26**) (1.00 g, 2.08 mmoles) in deoxygenated distilled water (60 ml) was placed in the top separating funnel A. The stopcock at B was partially opened to permit a slow stream of $N_2$ to bubble through the solution. Fresh sodium borohydride (1.6–1.7 g) was dissolved in water (30 ml), and the solution was poured rapidly into the pyrromethene solution. Considerable effervescence occurred, and the orange–red colour was discharged almost instantaneously. Dichloromethane (100 ml) and 10% aqueous $NaHCO_3$ (30 ml) were then added rapidly, the funnel was removed, shaken vigorously four times, and replaced. As rapidly as the lower phase separated, it was allowed to pass through the anhydrous sodium sulphate

* "The necessity of this whimsical prerequisite for the successful execution of the reaction series with the stated yield was repeatedly verified. Without it, final yields of porphyrin lower by 10–15% were obtained. One of us, who swore a colorful oath that he would never, by pouring the acid, be party to such an offence against what he regarded (wrongly) as sound scientific procedure, was forced by cruel experience to suffer the ignominy of having a colleague pour his acid for him in order to achieve results comparable with his fellows. Numerous experiments designed to elucidate the cause of the effect led us to no conclusive rationalisation, nor did they enable us to substitute a more respectable—if not easier—alternative procedure." RBW.

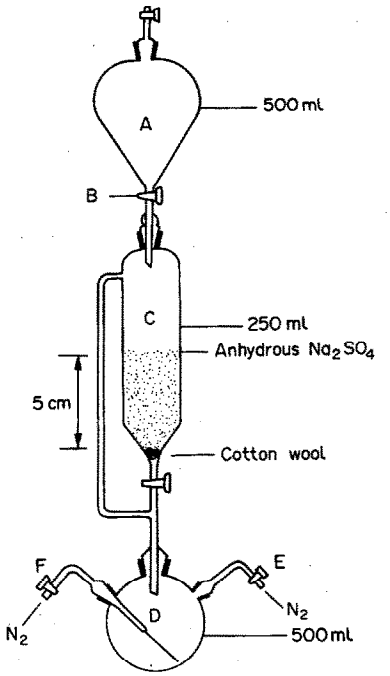



Fig. 19. Apparatus for the preparation of the dipyrrylmethane (**32**) and the Schiff's base (**33**).

in funnel C to the reaction flask D. The reaction mixture was then extracted rapidly with dichloromethane (2 × 50 ml) in the same manner.

*Step* 33. Funnels A and C were removed and, while a vigorous stream of nitrogen was kept passing through flask D, finely powdered thioaldehyde (**31**) (809 mg, 1.93 mmoles) was added all at once. The flask was swirled briefly to effect dissolution, equipped with a non-splash take-off head, the nitrogen inlet E was closed, and the dichloromethane was removed as rapidly as possible at the water pump, using a large $CO_2$/acetone trap between flask and pump, wide bore connections, and a water bath temperature of 50–55°. The evaporation was completed in about 5 min: occasional foaming at the outset was controlled by the rate of introduction of nitrogen at the capillary inlet F, or by delaying the introduction of the flask into the warm water bath. The flask was removed from the water bath just before the solvent has disappeared, and was then disconnected from the distillation train while a vigorous stream of nitrogen was introduced through stopcock E. Benzene (10 ml) was immediately added (pipette), the flask was swirled quickly and methanol (40 ml) was added (pipette) to give a solution of the Schiff's base (**33**). This was transferred (pipette) to a nitrogen-filled addition funnel (Fig. 20). The flask was washed with a further portion of methanol (40 ml), again pipetted into the addition funnel.

One drop of the Schiff's base solution was removed for spectroscopic control, the remainder being used immediately for the next step.

The Schiff's base solution (1 drop) was added to methanol (10 ml). The solution was swirled (not shaken), one drop of M-HCl/EtOH (85 ml conc. HCl made up to 1 l with ethanol) was added, and the electronic spectrum was determined without delay (Fig. 21). When the preparation of the Schiff's base (**33**) had been satisfactorily carried out, the ratio $A_{502}/A_{311}$ fell within the range 4.0–4.5.

*Step* 34. The tip of the addition funnel (Fig. 20) was introduced well below the surface of well-stirred (magnetic stirring) freshly prepared, poured (see above) saturated methanolic HCl (12–13

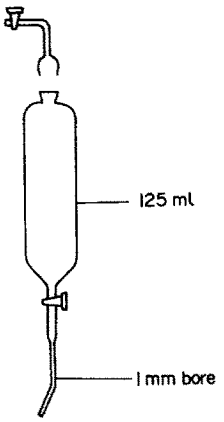



Fig. 20. Addition funnel for step 34.

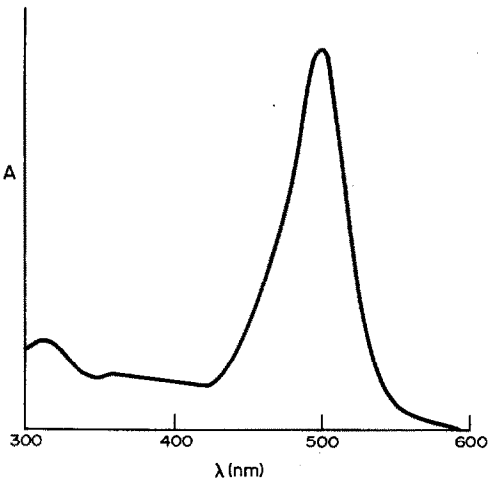



Fig. 21. Spectroscopic evaluation of step 33: conversion of Schiff's base (**33**) to corresponding bilene-*b* salt (**131**).

M, 360 ml). The methanolic solution of the Schiff's base (**33**) was rapidly and uniformly displaced from the funnel using a slight positive pressure of nitrogen, taking care that no hydrogen chloride gained prior access to the Schiff's base solution. Colour change: yellow → orange → red → dark reddish brown; (a persistent bright cherry red colour was indicative of a relatively unsuccessful run). The mixture was kept at room temperature for 30 min.

For control purposes, the reaction mixture (0.06 ml) was diluted to 10 ml with M-HCl/MeOH, and the electronic spectrum was determined (Fig. 22). The theoretical concentration of this control solution was *ca.* $3.5 \times 10^{-5}$ M. The main features of the spectrum are associated with the phlorin salt (**34**): a successful run was characterised by (i) a sharp well defined band at 425 nm, ε 47,000–50,000; (ii) relatively low absorption 450–500 nm; and (iii) the long wavelength band had $A_{713} > 0.50$. An estimate of the yield of the phlorin salt (**34**) was obtained from : % yield = $A_{713}V/2.90$, where V is the volume of the solution in ml measured after the conclusion of the oxidation step, which follows.

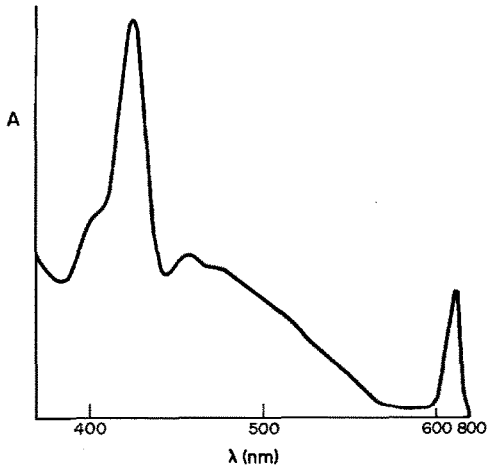



Fig. 22. Spectroscopic evaluation of step 34.

*Step* 35. Except in experiments where it was desired to isolate the phlorin salt (**34**, see below iodine (1.7 g) was added to the above solution and the mixture was stirred for 1 h at roo temperature. The total volume (V, *ca.* 350 ml) of the solution was measured for use in the phlori stage estimation (above) and in the estimation for the final porphyrin stage, as follows.

A small sample (0.03 ml) of the mixture was diluted to 10 ml with M-HCl/MeOH, and th electronic spectrum was measured (Fig. 23). The main feature of the spectrum was a very stron

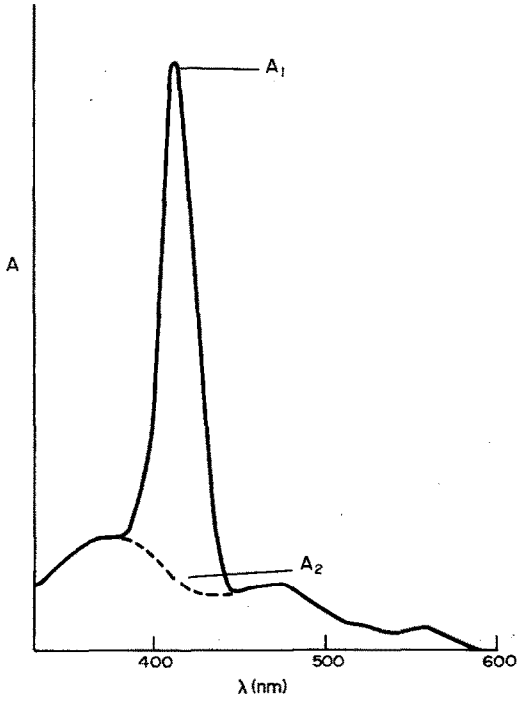



Fig. 23. Spectroscopic evaluation of step 35: assay of phlorin salt used as starting material.

Soret band at 418 nm due to the porphyrin ring. The theoretical concentration was *ca.* $1.26 \times 10^{-5}$ M, and $\varepsilon_{418}$ was in the range 151,000–153,000. An estimate of the yield of porphyrin at this stage could be obtained by constructing the dotted line as shown, and using $A_1$–$A_2$ as the corrected value for $A_{418}$, in conjunction with the value $\varepsilon_{418} = 269\ 000$ for the pure porphyrin salt under the conditions of measurement.

The reaction mixture was taken to dryness (water pump) using a capillary air inlet and a water bath (55–60°). The last traces of solvent were removed at the oil pump (30 min). Pyridine (40 ml) and acetic anhydride (20 ml) were added to the dry residue; solid material was dissolved by swirling, and the mixture was heated (50–55°) for 1 h. The solvent was removed as before. The dry residue was completely dissolved in freshly prepared M-HCl/MeOH (200 ml) and the solution was kept at room temperature for 40 minutes (re-esterification of any free carboxyl groups). It was then poured into ether (1 l, in a 2 l separating funnel), ice-cold 1.5 M-aqueous HCl (200 ml) was added, (order of addition important), and the mixture was shaken. The aqueous acidic layer, containing almost all the product, was quickly separated and transferred to a 4 l separating funnel. The ether layer was washed with more ice-cold aqueous HCl (20 ml), the acidic washings being combined, and layered over with ether (2.4 l). An ice-cold saturated solution (*ca.* 600 ml) of $Na_2CO_3$ in distilled water was added until the aqueous layer was just alkaline (pH *ca.* 8). The mixture was shaken vigorously for a short time, and then allowed to stand to allow the compaction of flocculent material at the interface. The lower aqueous phase was largely removed, and the ethereal phase was decanted from flocculent material. The latter was extracted with several portions of ether until virtually no more coloured material was extracted. The separated layer was extracted once with ether. The combined ethereal extracts were dried over $Na_2SO_4$ (20–30 min), filtered, and made up to 4 l with ether. When a portion of this solution (5 ml) was made up to 10 ml with ether, the visible spectrum appeared as shown in Fig. 24. The yield of crude porphyrin was calculated from $\varepsilon_{510} = 12\ 500$: in properly executed runs this estimate was about 60%. A rough estimate of purity was afforded by the ratio $A_{510}/A_{463}$ which usually fell in the range 4.5–5.5.

The ether solution of porphyrin was evaporated and the residue was taken up in dichloromethane

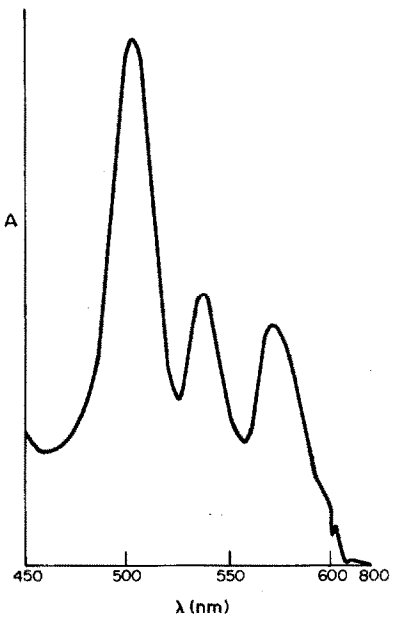



Fig. 24. Spectroscopic evaluation of step 35: assay of product.

(30 ml) and applied to a column (3.2 cm internal diameter) of Florisil (100–200 mesh, 70 g) which had been made up in benzene/ether and then washed with ether/methanol (9 : 1) and then with dichloromethane. The solution was washed on to the top of the column with dichloromethane (30 ml). Development and elution with ether/methanol (9 : 1) gave, after removal of early yellow fractions, a well defined major band which was completely eluted with *ca.* 4 l solvent. Occasionally a minor by-product remained on the column at this stage and could be eluted with ether/methanol = 85 : 15. This substance was identified as the 15-propion-N-ethylamide corresponding to (**35**) and is attributed to those runs of step 30 where the conditions were insufficiently mild.

The solutions of the major product were concentrated to about 200 ml. From the hot solution (very largely methanol) the porphyrin began to crystallise. The concentrate was allowed to cool, then kept at 4° overnight to give the porphyrin (**35**) (635 mg, 48%) as beautiful purple crystals, mp 226–228°. A further small amount of porphyrin could be obtained by rechromatography of the material in the mother liquors. The isolation of the porphyrin was conveniently carried out on the combined material from five such runs, with no loss in yield or purity, using a column with internal diameter 10.4 cm packed with Florisil (700 g) to a height of 14 cm.

The porphyrin (**35**) crystallised in violet needles from methanol–dichloromethane. For analysis it was recrystallised twice from methanol, and dried in high vacuum overnight over $P_2O_5$. Mp 233.5–234°. (Found: C, 67.55; H, 6.8; N, 9.6; O, 15.95. $C_{40}H_{47}N_5O_7$ requires C, 67.7; H, 6.65; N, 9.9; O, 15.8%). $\lambda_{max}$ ($CH_2Cl_2$) ($\varepsilon$) 510 (12 600), 546 (7 500), 575.5 (6 800), 628 nm (1 200). $\nu_{max}$ (KBr) 3435, 3280, 1720, 1650, 1540, 1435, 1360 $cm^{-1}$.

[August 1989. *m/z* (FAB, thioglycerol) 710.355 (100%, $M+H^+$ requires 710.355). $\delta$ ($CDCl_3$, $1.2\times10^{-3}$ M) 10.12, 10.09, 9.91 (3×1 Hs, *meso* H), 5.66 (bs, 1H, NH), 5.17 (bt, 2H, *meso*-$CH_2CH_2CO_2Me$), 4.42 (bt, 2H, $CH_2CH_2N$), 4.35 (s, 3H, Ar-$CO_2Me$), 4.13 (m, 6H, $CH_2Me$, $\beta$-$CH_2CH_2NHAc$, $\beta$-$CH_2CH_2CO_2Me$), 3.77, 3.75, 3.73 (3×3 Hs, Ar*Me* on rings A, B, and D), 3.64, 3.65 (2×3 Hs, 2×$CH_2CH_2CO_2Me$), 3.57 (s, 3H, Ar*Me* on ring C), 3.24, (2 Hbt, *meso*-$CH_2CH_2CO_2Me$), 3.04 (2 Hbt, $\beta$-$CH_2CH_2CO_2Me$), 1.90 (s, 3H, *Me*CON), 1.87 (t, J = 7.5 Hz, $CH_2Me$), −3.0 (b, 2H, 2×NH). $R_f = 0.54$ ($CHCl_3$ : acetone = 2 : 1); $R_f = 0.30$ on $AgNO_3$ impregnated plate ($CHCl_3$ : acetone = 2 : 1).]

*Step* 34. 8-(*β-Aminoethyl*)-13-*ethyl*-18-*methoxycarbonyl*-3,7,12,17-*tetramethyl*-20,22-*dihydroporphyrin*-2,20-*dipropionic acid dimethyl ester dihydrobromide* (**34**). A portion (40–45 ml) of the acid condensation mixture containing the phlorin (step 34 above) or, alternatively, the total reaction mixture from a preparation run on one eighth the scale described above, was poured into nitrogen-swept dichloromethane (100 ml). The mixture was extracted with ice (75 g) and water (25 ml) under nitrogen. The organic layer was separated, dried ($Na_2SO_4$, several h), concentrated to 20 ml, and applied to a column of Florisil (60 g) made up in 0.5 M-HCl/MeOH and washed with 0.05 M-HCl/MeOH (500 ml), set up to run and collect under a nitrogen atmosphere. The column was developed and eluted with 0.05 M-HCl/MeOH. There separated a fast moving pink band (probably pyrromethenes, $\lambda_{max}$ 487 nm), an olive-green zone, and a dark red immobile band. The olive green component was collected in 50 ml portions: those having $A_{714}/A_{459} > 10$ and $A_{428}/A_{714} \sim 2.7$–2.9 were combined and taken to dryness. These criteria guard against contamination by pyrromethene salts ($\lambda_{max}$ *ca.* 480 nm) and by porphyrin ($\lambda_{max}$ 421 nm under the conditions of the measurement, the band being three times as intense as the 428 nm band of the phlorin salt).

The residue containing the phlorin dihydrochloride (*ca.* 60 mg = 33%) and some inorganic material from the column, was treated with dichloromethane (40 ml). The solution was shaken briefly under nitrogen with an equal volume of 1 M-$Na_2CO_3$. The resulting pure blue solution of the free phlorin base was then immediately shaken with an equal volume of 2 M-HBr (none of the phlorin salt enters the aqueous phase). The chloromethane solution of the phlorin dihydrobromide was dried ($Na_2SO_4$, several h), filtered, and taken to dryness under nitrogen. The residue was crystallised from acetone (1 ml) giving 30 mg (15%) of the pure phlorin dihydrobromide (**34**) as dark green crystals. Analytical sample, from acetone, well formed very dark bluish-green rhombs,

did not melt, decomp. >200°. (Found: C, 55.25; H, 6.3; N, 8.45. $C_{38}H_{47}N_5O_6$. 2 HBr requires C, 54.9; H, 5.95; N, 8.4%). $\lambda_{max}$ ($CH_2Cl_2$) (ε) 303 (12 500), 362 (22 300), 431 (57 800), 483 (4 100), 518 (6 100), 553 (4 900), 723 nm (17 400). (Our earlier data[1] refer to methanolic hydrogen chloride as solvent, not dichloromethane as inadvertently stated.) $\lambda_{max}$ ($CH_2Cl_2$, sat. HBr) 447 nm. $\lambda_{max}$ ($CH_2Cl_2$, sat. HBr, diluted with MeOH) 427, 513, 546, 710 nm. Free base: $\lambda_{max}$ ($CH_2Cl_2$) 291, 387, 620 nm, but solution not stable.

12-*Ethoxycarbonyl*-17-*ethyl*-3,7,13,18-*tetramethylporphyrin*-2,8,10-*tripropionic acid triethyl ester* (**130**). A mixture of the pyrromethene salt (**122**) (113 mg), 10% Pd on charcoal (25 mg), ethanol (5 ml) and 1 M-NaOH (0.30 ml) was shaken under hydrogen until absorption stopped and the solution was colourless (*ca.* 5 min). The mixture was filtered at once through Celite directly into a 100 ml three-necked flask and evaporated to dryness under reduced pressure (no oxygen). The residue, under hydrogen, was treated rapidly with a solution of the aldehyde (**125**) (108 mg) in glacial acetic acid (30 ml) followed by a mixture of 38% aqueous HBr (0.25 ml) and glacial acetic acid (20 ml). The mixture was heated under hydrogen at 80° for 1 h, transferred to a distilling flask, and evaporated to dryness under reduced pressure. The residue was treated for 30 min at room temperature with 1 M-HCl/anhydrous EtOH (25 ml) and the resulting dark brown solution was poured into ether (250 ml). The ether solution was washed with water (2 × 100 ml), the washings being back-extracted with a small amount of ether. The combined ether solutions were treated with iodine (0.5 g) and kept at room temperature for 15 min. The mixture was then shaken vigorously with aqueous sodium thiosulphate, washed with water, and extracted with 2 M-HCl (3 × 70 ml). The acid extracts were quickly layered over with ether (300 ml) and neutralised by the addition of solid $Na_2CO_3$. The ether extract was dried ($Na_2SO_4$) and taken to dryness and the residue in benzene was chromatographed on Florisil (25 g). The product was eluted rapidly with ether : methanol = 98 : 2. The solvent was removed and the residue was crystallised from light petroleum giving the porphyrin tetra-ethylester (**130**) as red–brown needles (43 mg, 40%), mp 128–130°. (Found: C, 68.7; H, 7.2; N, 7.7; OEt, 23.45. $C_{44}H_{54}N_4O_8$ requires C, 68.9; H, 7.1; N, 7.3; OEt, 23.5%). $\lambda_{max}$ ($Et_2O$) (ε) 503 (14 500), 538 (7 000), 573 (6 200), 626 nm (1 000). The compound was also obtained as a polymorph, mp 174–175°, from benzene : hexane = 1 : 4.

9.9. *Porphyrin reactions: a porphyrin–phlorin equilibrium*

*Step* 36. 8-(*β-Acetylaminoethyl*)-13-*ethyl*-18-*methoxycarbonyl*-20-(*β-methoxycarbonylvinyl*)-3,7,12,17-*tetramethylporphyrin*-2-*propionic acid methyl ester* (**36**). (*Porphyrin* **36**). A solution of the porphyrin *meso*-propionic acid ester (**35**) (500 mg) in glacial acetic acid (165 ml) was stirred vigorously under a drying tube ($CaCl_2$) in an oil bath at 95° for 1 h. The acetic acid was then removed at the rotary evaporator, the last traces of solvent being removed by several evaporations with benzene. The residue was dissolved in boiling dichloromethane (*ca.* 200 ml) and methanol was added dropwise. Crystallisation was completed at 4° giving the porphyrin *meso*-acrylic acid ester (**36**) (456 mg, 91%) as purple crystals, mp 245–247°. Analytical sample from dichloromethane/methanol large violet spears, mp 251–252°. (Found: C, 67.6; H, 6.55; N, 9.75; O, 16.2; OMe, 12.5%; $M^+$, 707.333. $C_{40}H_{45}N_5O_7$ requires C, 67.85; H, 6.4; N, 9.9; O, 15.8; OMe, 13.15% M, 707.332). $\lambda_{max}$ ($CH_2Cl_2$) (ε) 512.5 (11 200), 549 (7 500), 578.5 (7 400), 633.5 nm (2 300). $\nu_{max}$ (Nujol) 3300, 1720, 1635, 1520 $cm^{-1}$. [August 1989. δ ($CDCl_3$) 10.23 (d, 1H, J = 15.5 Hz, *meso*-C*H*=CH—$CO_2$Me), 10.18, 10.04, 9.96 (3 × 1 Hs, *meso*-H). 6.25 (d, 1H, J = 15.5 Hz, *meso*-CH=C*H*—$CO_2$Me), 5.70 (bs, 1H, NH), 4.39 (m, 2H, β-$CH_2$—$CH_2$—N), 4.16 (s, 3H, Ar-$CO_2$*Me*), 4.10 (m, 6H, $CH_2$—Me, β-$CH_2$—$CH_2$—NHAc, β-$CH_2$—$CH_2$—$CO_2$Me), 4.00 (s, 3H, *meso* CH=CH—$CO_2$*Me*), 3.76 (s, 3H, β-$CH_2CH_2$—$CO_2$*Me*), 3.68, 3.66, 3.64 (3 × 3 Hs, Ar *Me* on rings A, B, and D), 3.57 (s, 3H, Ar *Me* on ring C), 3.03 (bt, 2H, β-$CH_2$—$CH_2$—$CO_2$Me), 1.90 (s, 3H, *Me*CON), 1.84 (t, 3H, $CH_2$-*Me*), −3.3 (b, 2H, 2 × NH). $R_f$ = 0.52 ($CHCl_3$ : acetone = 2 : 1), $R_f$ = 0.24 on $AgNO_3$ impregnated plate ($CHCl_3$ : acetone = 2 : 1). $\nu_{max}$ (KBr) 3285, 1725, 1635, 1560, 1435, 1355, 1310, 1270, 1195, 1175, 1080, 1060, 740 $cm^{-1}$.]

*Quantitative determination of oxygen consumption in Step* 36. A 250 ml flask containing pure glacial acetic acid (100 ml) and a magnetic stirring bar was attached through a Liebig condenser to a gas-measuring burette (semi-micro hydrogenation apparatus). The system was filled with pure oxygen, and the acetic acid was stirred for 1 h to equilibrate the system. The porphyrin (**35**) (298 mg, 0.42 mmoles) was added, and the apparatus was evacuated and refilled with oxygen. The solution was stirred for 30 min at a constant temperature (29°, 760 mm), and then stirred and heated for 1 h in an oil bath at 100°. The flask was then removed from the bath, and cooled to 29° and allowed to equilibrate for 4 h. The observed oxygen uptake was 5.2 ml (29°, 760 mm) corresponding to 0.21 mmoles of oxygen.

The solution was diluted with dichloromethane (250 ml) and washed with saturated NaCl (130 ml; washings showed negative KI/starch test for peroxides). The porphyrin acrylic ester (**36**), mp 245–248°, was isolated (247 mg, 83%).

*Catalytic reduction of the porphyrin meso-acrylic ester* (**36**) *to the corresponding porphyrin meso-propionic ester* (**35**). Catalytic reduction of the porphyrin (**36**) (25.8 mg) was effected in hydrogen over 10% Pd/C (113 mg) in acetic acid (20 ml). Colour change: blue violet → green → orange → colourless over 45 min. The catalyst was removed and washed with dichloromethane (20 ml). The combined solutions were diluted with ether (150 ml) and extracted with ice water (3 × 80 ml), the water being back-extracted with ether (2 × 50 ml). The combined organic solutions were dried ($Na_2SO_4$) and treated with iodine (170 mg) in methanol (50 ml). After 18 h the porphyrin was extracted with 0.7 M-HCl, the acid extract was washed once with an equal volume of ether, and then neutralised over dichloromethane (100 ml) by careful addition of solid $Na_2CO_3$. The porphyrin solution in dichloromethane was reduced to a small volume and applied to a column (5.5 cm diameter × 3 cm) of Florisil. The first fraction eluted with ether–methanol = 9 : 1 contained almost all the porphyrin present (spectroscopic yield 30.2%). The porphyrin propionic ester (**35**) was crystallised (6.4 mg, 25%) from methanol as characteristic violet needles, mp 227–229°, mixed mp 232.5–234°, quantitative electronic spectrum indistinguishable from that of the authentic material.

*An equilibrium between the porphyrin meso-propionic ester* (**35**) *and the phlorin meso-acrylic ester* (**136**). Oxygen must be excluded throughout: all nitrogen is oxygen-free.

Nitrogen was passed through boiling acetic acid for 1 h; the acetic acid was allowed to cool down under nitrogen, and used at once. The porphyrin (**35**) (50.7 mg, 71 μmoles) was placed in a 70 ml flask, which was then twice evacuated and filled with nitrogen. Freshly prepared acetic acid (50 ml) was added, and the flask was thrice evacuated and filled with nitrogen. The solution was heated on the steam bath for 1 h, cooled, and made up to 500 ml with ether. The visible spectrum of this solution was determined (Fig. 8) and was consistent with presence of, essentially, only one porphyrin and one phlorin. On the basis of $\varepsilon_{507} = 13{,}600$ for the porphyrin (**35**) and $\varepsilon_{721} = 24{,}000$ for the phlorin (**136**) the solution contained 44.5 μmoles (62.2%) of porphyrin and 26.5 μmoles (37%) of phlorin, accounting for 99.2% of the starting material and giving $K_{(\text{phlorin/porphyrin})} = 0.60$.

A portion (28.3 ml) of the solution was set aside (see below). The remainder was made up to 500 ml with ether (blue–violet solution) and extracted ($N_2$) with 0.7 M-HCl. The acid extract was neutralised (solid $Na_2CO_3$), and the liberated free porphyrin was extracted into dichloromethane. The organic extract was made up to 250 ml with dichloromethane, and the spectrum was determined (Fig. 9): the presence of maxima at 510, 546, 577, and 629 nm showed that this solution contained essentially only porphyrin (**35**). Using $\varepsilon_{510} = 12\ 500$, the amount was determined as 37.5 μmoles (52.5%).

The ether layer remaining after extraction of the porphyrin was made up to 500 ml with ether, and its spectrum was determined at once (Fig. 10), and showed that 23.8 μmoles of phlorin (33%) was present. Since 5.6% of the original mixture had been set aside, the total recovery was 91%, and the equilibrium constant was 0.57. The ethereal phlorin solution was extracted with 5 M-HCl in air. The aqueous phase was removed and neutralised with saturated aqueous $Na_2CO_3$. During the neutralisation a greenish blue precipitate separated. Dichloromethane was added, with shaking in

air, until the precipitate had dissolved, and the colour had changed from bluish-green to red. The resulting solution after drying ($Na_2SO_4$) showed $\lambda_{max}$ 512, 549, 578, 631 nm, essentially that of the porphyrin acrylic ester (**36**) in this solvent.

The aliquot (28.3 ml) was taken to dryness and the residue was heated in acetic acid in air for 1 h giving pure porphyrin acrylic ester (**36**) in a spectroscopic yield of 80%.

9.10. *Entry into the chlorin series*

*Step* 37. [2*S*(*R*),3*S*(*R*)]-8-(*β-Acetylaminoethyl*)-13-*ethyl*-2$^1$,18-*dimethoxycarbonyl*-3,7,12,17-*tetramethyl-cyclopentano*[*a,t*]*chlorin*-2-*propionic acid methyl ester* (**37**). (*Purpurin* **37**). All nitrogen is oxygen-free. To the porphyrin *meso*-acrylic acid ester (**36**) (2.2 g) in a 1 l r-b flask was added nitrogen-flushed acetic acid (440 ml). The flask was evacuated and filled with nitrogen four times, and the contents were refluxed (oil bath 140°) for 31.5 h under a slow stream of nitrogen, with magnetic stirring and protection from light. The cold solution was transferred to a 5 l r-b flask, and ether (3 l) was added dropwise over a period of *ca.* 1 h (colour change: green → purple). The mixture was kept at room temperature for 17 h. The precipitate was collected giving 0.953 g (43%) of the pure purpurin (**37**), with $A_{704}/A_{502} = 9.8$ and $A_{704}/A_{575} = 2.63$.

The filtrate was concentrated, and the concentrate was subjected to a similar equilibration; the cycle was repeated, as follows:

| Substrate | HOAc (ml) | Time (h) | Ether (l) | Time (h) | Yield (g) |
|---|---|---|---|---|---|
| First Filtrate | 250 | 30 | 1.70 | 34 | 0.334 |
| Second Filtrate | 180 | 30 | 1.25 | 29 | 0.216 |
| Third Filtrate* | 170 | 30 | 1.20 | 41 | 0.138 |

* Plus mother liquors from earlier recrystallisations

The purpurin was recrystallised by dissolution in dichloromethane under reflux, and concentrating to a volume of approximately 250 ml per gram. Methanol (150 ml per gram) was added dropwise to the boiling solution, and the mixture was reduced in volume until purpurin (**37**) began to crystallise (bp of solvent mixture *ca.* 55°). Slow cooling to room temperature and overnight refrigeration gave *ca.* 85% recovery. In this way 1.30 g (59%) of purpurin (**37**) was obtained as dark blue rhombic plates, mp 310° decomp. (Found: C, 67.75; H, 6.5; N, 9.85; MeO, 12.95. $C_{40}H_{45}N_5O_7$ requires C, 67.85; H, 6.4; N, 9.9; MeO, 13.5%). $\lambda_{max}$ ($CH_2Cl_2$) (ε) 502 (5 000), 543 sh (7 000), 575 (19 500), 649 (8 700), 704.5 nm (52 200). $\nu_{max}$ (KBr) 3400, 3320, 1740 sh, 1720 sh, 1705 sh, 1695, 1665, 1600, 1540, 1510, 1435, 1410, 1300, 1125, 900, 810, 735 $cm^{-1}$. [August 1989 *m/z* (FAB, *p*-nitrobenzyl alcohol) 708.340 (100%). $C_{40}H_{45}N_5O_7+H$ requires 708.340].

9.11. *Modification of peripheral substituents of purpurins*

*Step* 38. [2*S*(*R*),3*S*(*R*)]-13-*Ethyl*-2$^1$,18-*dimethoxycarbonyl*-3,7,12,17-*tetramethyl*-8-*vinyl-cyclopentano*[*a,t*]*chlorin*-2-*propionic acid methyl ester* (**38**). (*Vinylpurpurin* **38**). All nitrogen is oxygen free. The crystalline purpurin (**37**) (1.33 g) was dissolved in a mixture of conc. HCl (340 ml) and nitrogen-flushed methanol (2.1 l) in a 5 l three necked flask ($N_2$ inlet, efficient condenser, dropping funnel); the solution was flushed with nitrogen with stirring for 30 min. The solution was then heated to boiling and hot, nitrogen-flushed, water (1.65 l) was added gradually in such a way that no precipitation occurred. The $N_2$ inlet was repositioned at the top of the condenser (safety valve) and the apparatus was covered with aluminium foil to exclude light and heated in an oil bath (120°, 18 h) with continuous magnetic stirring. When necessary, nitrogen-flushed methanol was added from the dropping funnel to keep the volume constant.

The solution was evaporated to dryness (rotary evaporator attached via a $CO_2$-acetone cold trap to a mercury diffusion pump; water bath temperature 35–40°). The residue was evaporated to

dryness with a little methanol, dissolved in methanol, and treated with excess diazomethane in ether (5–10 min). After removal of solvent the residue was dissolved in dichloromethane (300 ml) and ether (1.5 l), and extracted with 0.5 M-HCl (3 × 1.5 l) and then 5 M-HCl (2 × 0.5 l). The two acid extracts were separately washed with ether (1.5 l), neutralised with solid $NaHCO_3$, and extracted with dichloromethane. The extracts from the 0.5 M-HCl afforded the aminoethylpurpurin (**37A**), (935 mg, 75%), $\lambda_{max}$ ($CH_2Cl_2$) (ε rel) 499 (0.10), 536 (sh) (0.15), 571 (0.40), 645 (0.14), 698 nm (1.00). The extracts from the 5 M-HCl gave the starting material (7%).

The β-aminoethyl compound (**37A**) is unstable and should be used without delay. The β-aminoethylpurpurin (**37A**) (537 mg) was dissolved in 2 M-HCl/MeOH (1 ml) and nitrogen-flushed methanol (500 ml) in a 2 l three necked flask equipped with stirrer, reflux condenser, and two dropping funnels which had been flushed with nitrogen. With exclusion of light and air the solution was stirred under reflux while freshly distilled dimethyl sulphate (83 ml) and 2 M-NaOH in nitrogen-flushed methanol (380 ml) were simultaneously added over 2.5 h. The addition was made in such a way that during the first part of the reaction dimethyl sulphate was in excess (green solution) whereas in the second part of the reaction the alkali was in excess (purple solution). Finally dimethyl sulphate (*ca.* 10 ml) was added, and boiling was continued for a further 5 min. The solution was then evaporated (water bath 35–40°) to a thick slurry at the rotary evaporator (cold trap, diffusion pump). The residue was dissolved in dichloromethane (100 ml) and water (70 ml), and ether (1 l) was added. The organic phase was extracted with 1 M-HCl (3 × 1 l) and 7 M-HCl (3 × 500 ml). The latter acid extract was cooled in ice, and worked up rapidly (solid $NaHCO_3$, $CH_2Cl_2$) giving the vinylpurpurin (**38**) as narrow prisms, mp: decomp. ~ 285–290° without melting, from dichloromethane–ether (385 mg). Chromatography of the mother liquors on Florisil (5 g, 1% MeOH in $CH_2Cl_2$) gave a further 54 mg. Total yield 84%. (Found: C, 70.45; H, 6.25; N, 8.6; O, 15.0. $C_{38}H_{40}N_4O_6$ requires C, 70.35; H, 6.2; N, 8.65; O, 14.8%). $\lambda_{max}$ ($CH_2Cl_2$) (ε) 504 (4 700), 547 (7 400), 581 (19 800), 656.5 (8 400), 713.5 nm (55 000). $\nu_{max}$ (Nujol) 3320, 1735, 1710, 1695, 1600 $cm^{-1}$.

*Step* 39. [2*R*(*S*),3*S*(*R*)]-13-*Ethyl*-20-*formyl*-18-*methoxycarbonyl*-2-*methoxyoxalyl*-3,7,12,17-*tetramethyl*-8-*vinylchlorin*-2-*propionic acid methyl ester* (**39**). (*Methoxalylpurpurin* **39**). The vinylpurpurin (**38**) (50.4 mg) in dichloromethane (200 ml, freshly distilled from $Na_2CO_3$) was stirred vigorously at 0° and irradiated in air (750 watt tungsten lamp, 35 min). The spectroscopic yield at this stage was 74%. The solvent was removed and the product was crystallised from dichloromethane–ether to give 41 mg of the crude product. Recrystallisation gave 31 mg (59%) of the methoxalylpurpurin, (**39**), decomp. ~250–255°. Analytical sample, dark crystals from two further recrystallisations, had the same decomp. point, and was dried *in vacuo* at 55–60° for 48 h. (Found: C, 66.5; H, 6.0; N, 8.2; O, 18.85. $C_{38}H_{40}N_4O_8$ requires C, 67.05; H, 5.9; N, 8.25; O, 18.80%). $\lambda_{max}$ ($CH_2Cl_2$) (ε) 486 (4 000), 512 (6 500), 551 (15 800), 559 sh (2 400), 644 (7 000), 701 nm (39 500). $\nu_{max}$ ($CH_2Cl_2$) 3390, 1730 sh, 1715, 1660, 1600 $cm^{-1}$. δ ($CDCl_3$, 60 MHz, semi-systematic numbering) 11.22 (s, *meso*-CHO), 9.45, 9.00 (2 × 1 Hs, *meso*-H at C-5, C-10), 8.28 (1 Hs, C-20 *meso*-H), ~2.4, 6.0 (m, m, —CH=$CH_2$), 4.20 (s, $ArCO_2Me$).

Spectroscopic yields of 68–79% were consistently obtained on a 15–50 mg scale.

*Step* 40. [2*S*(*R*),3*S*(*R*)]-18-*Carboxy*-13-*ethyl*-20-(*hydroxy-methoxymethyl*)-3,7,12,17-*tetramethyl*-8-*vinylchlorin*-2-*propionic acid methyl ester* δ-*lactone* (**40**). *rac-Isopurpurin 5 methyl ester.*

Freshly prepared methoxalylpurpurin (**39**) (102 mg), dichloromethane (16 ml, freshly distilled and filtered through $Na_2CO_3$) and 2 M-KOH/MeOH (49 ml) were stirred for 1 h, poured into ether (500 ml), washed with water (twice), and briefly treated with diazomethane. The ethereal layer was washed with 3.3 M-HCl (3 × 500 ml) the acid being back-extracted with ether (500 ml). The combined ether solutions (34% yield at this stage) were concentrated and chromatographed on Florisil (3 g), eluting with dichloromethane and then MeOH: $CH_2Cl_2$ = 4:96 giving 19 mg (22%) of *rac*-isopurpurin 5 methyl ester (**40**), mp 220–221° decomp. from ether–methanol (20:1). $\lambda_{max}$ ($CH_2Cl_2$) (ε) 500 (11 600), 532 (9 500), 562 (2 800), 612 (7 000), 668 nm (49 200), spectroscopically identical with isopurpurin 5 methyl ester derived from natural sources. $\nu_{max}$ ($CH_2Cl_2$) 3370, 2920, 1720, 1610,

1075, 1035 $cm^{-1}$, identical with the spectrum of natural isopurpurin 5 methyl ester measured at the same concentration. [August 1989 $R_f = 0.83$ ($CHCl_3$ : acetone = 2 : 1)].

*Step* 41. [2*S*(*R*),3*S*(*R*)]-18-*Carboxy*-20-(*dihydroxymethyl*)-13-*ethyl*-3,7,12,17-*tetramethyl*-8-*vinylchlorin*-2-*propionic acid δ-lactone* (**41**). *rac-Chlorin* 5.

To synthetic racemic isopurpurin 5 methyl ester (**40**) (66.5 mg) dissolved in dioxan (80 ml) was added water (35 ml) and 1 M-NaOH (5 ml). The solution was flushed with nitrogen for a few minutes and kept at room temperature for 12 h. A sample diluted with methanol showed the purpurin 5 dianion spectrum ($\lambda_{max}$ 504, 538, 578, 633, 687 nm). The solution was poured into water containing 1.5 M-HCl (5 ml) and extracted with ether. All the pigment went into the ether layer. Careful acid fractionation with 1.5 M-HCl and 3.3 M-HCl gave, on basification of the 3.3 M-HCl extract and transfer to ether, chlorin 5 in a spectroscopic yield of 37.8 mg (60%), $\lambda_{max}$ ($Et_2O$) 499, 528, 558, 611, 668 nm. Visible spectrum identical with that of an optically active sample from natural sources. [August 1989 $R_f = 0.28$ ($CHCl_3$ : MeOH = 10 : 3), *m/z* (FAB, *p*-nitrobenzyl alcohol) 567.261 (40%, $C_{33}H_{34}N_4O_5$+H requires 567.261)].

9.12. *Optical resolution to enter natural series*

*Step* 42. *Resolution of chlorin* 5. Quinine was purified by the method of Thron and Dirscherl.[62] Fractional crystallisation of the quinine salt from dichloromethane–ether did not lead to resolution. However, when the crystallised salt (23.1 mg, equivalent to 14.7 mg chlorin 5) was recrystallised slowly overnight from dichloromethane–ether, in the presence of one additional mol. proportion of quinine, crystals separated [8.17 mg, spectroscopic yield based on $\varepsilon_{667}$ ($Me_2CO$) = 50 000] with $[\alpha]_{546}^{23} = +1000°$. Recrystallisation from the same solvent system yielded a salt (2.78 mg) having $[\alpha]_{546}^{23} = +1236°$ ($c = 9.72 \times 10^{-3}$, $Me_2CO$). The quinine salt from naturally-derived chlorin 5 had $[\alpha]_{546}^{23} = +1215°$ ($c = 1.19 \times 10^{-2}$, $Me_2CO$). The quinine salt crystals from the synthetic chlorin 5 had a visible spectrum identical with that of the naturally-derived material [$\lambda_{max}$ ($Me_2CO$) ($\varepsilon$) 498 (11 000), 528 (7 800), 558 (2 000), 610 (5 800), 667 nm (47 600).]

The crystalline quinine salt was shaken with ether and 1.5 M-HCl, the ether layer was washed with water and dried ($Na_2SO_4$). The chlorin 5 solution was evaporated to dryness and the residue was dissolved in acetone. The rotation was $[\alpha]_{546}^{23} + 1810°$ ($c = 8.02 \times 10^{-3}$, $Me_2CO$). The naturally-derived sample of chlorin 5 had $[\alpha]_{546}^{23} + 1823°$ ($c = 8.83 \times 10^{-3}$, $Me_2CO$). The visible spectra of the synthetic active sample and the naturally-derived sample were identical [$\lambda_{max}$ ($Me_2CO$) ($\varepsilon$) 498 (10 300), 529 (7 100), 560 (2 500), 610 (5 500), 667 nm (46 900)].

The mother liquor from the quinine salt crystals with $[\alpha]_{546}^{23} = +1000°$ (above) gave a further crop of crystals. The resulting mother liquor, (equivalent to 3.8 mg of chlorin 5) then had $[\alpha]_{546}^{23} = -1146°$. Treatment with acid as before gave [17*R*,18*R*]-chlorin 5 (3.25 mg) with $[\alpha]_{546}^{23} = -1340°$ ($c = 6.3 \times 10^{-3}$, $Me_2CO$) corresponding to 74% optical purity.

9.13. *Relay position and final elaboration of peripheral substituents*

*Step* 43. (2*S*,3*S*)-13-*Ethyl*-20-*formyl*-18-*methoxycarbonyl*-3,7,12,17-*tetramethyl*-8-*vinylchlorin*-2-*propionic acid methyl ester*, (**43**). *Purpurin* 5 *dimethyl ester*. The chlorin 5 solution from step 42 was evaporated to dryness, and the residue was dissolved in ether containing some methanol. The solution was treated with a large excess of diazomethane in ether (dark, 20 min). The product was crystallised from ether–methanol giving synthetic purpurin 5 dimethyl ester (*ca.* 1 mg) as needles, mp 191.5–195°. The naturally-derived compound had mp 192–195.5° (lit.[67] mp 194°); the mixed mp showed no depression. (*rac*-Purpurin 5 dimethyl ester had mp 221–222.5°). The visible spectrum of the synthetic sample was identical with that of the natural one: $\lambda_{max}$ ($CH_2Cl_2$) 480, 506, 543, 638, 691 nm.

The infrared spectra of the natural and synthetic samples in the needle crystalline form were identical: $\nu_{max}$ (KBr) 1710, 1690, 1580, 1560, 1480, 1420, 1240, 1210, 1070, 1050, 1030, 1020, 800 $cm^{-1}$. Occasionally the sample crystallised in the form of plates, and the infrared spectrum of this

form was slightly different: $\nu_{max}$ (KBr) 1720, 1660, 1590, 1570, 1490, 1430, 1300, 1250, 1080, 1060, 1030, 980, 900, 800 $cm^{-1}$. $\delta$ (0.17 M, $CDCl_3$, natural, semi-systematic numbering) 11.48 (1 Hs, *meso*-CHO), 9.27, 8.90 (2 × 1 Hs, C-5, C-10 *meso*-H), 8.35 (1 Hs, C-20 *meso*-H), ~7.60, ~6.0 (m, m, —CH=$CH_2$), 4.08 (3 Hs, $CO_2Me$ at C-13), 3.50 (3 Hs, —$CH_2CO_2Me$), 3.40, 3.09, 2.50 (3 × 3 Hs, 3 × Ar*Me*).

*Step* 44. (2*S*,3*S*)-18-*Carboxy*-20-(*cyanohydroxymethyl*)-13-*ethyl*-3,7,12,17-*tetramethyl*-8-*vinyl*-*chlorin*-2-*propionic acid methyl ester* δ-*lactone* (**44**). (*Cyanolactone* **44**).[64] An ice-cold solution of purpurin 5 methyl ester (**43**) (75 mg) in acid-free dichloromethane (45 ml) under nitrogen was treated with freshly distilled anhydrous hydrogen cyanide (*ca.* 3 ml). Triethylamine (0.25 ml) in dichloromethane (2 ml) was added, and the mixture was shaken at room temperature for 3.5 min (colour change: brown → green). The solvent was at once removed under reduced pressure, and the residue was dissolved in a small amount of dichloromethane and diluted with ether. The solution was extracted with 3.3 M-HCl (5 ×, to remove starting material), with 4 M-HCl (4 ×) and with 4.75 M-HCl (1 ×). The cyanolactone (**44**) was then extracted with 23% HCl, the acid extract being separated, diluted with water, and reextracted into ether–dichloromethane (*ca.* 2:1). The solution was dried ($Na_2SO_4$), concentrated, and the residue was crystallised from dichloromethane–acetone–methanol to give 58 mg (78%) of the cyanolactone (**44**), mp 264–266°. For analysis the product was recrystallised from the same solvent, then from dichloromethane–methanol and dried *in vacuo* for 5 days at room temperature, mp 269–270°. (Found: C, 71.35; H, 5.95; N, 11.8. $C_{35}H_{35}N_5O_4$ requires C, 71.3; H, 6.0; N, 11.9%). $\lambda_{max}$ ($CH_2Cl_2$) (ε) 470 (3 900), 500 (10 550), 531.5 (10 400), 557 (2 500), 608.5 (6 850), 665 nm (44 000). $\nu_{max}$ ($CH_2Cl_2$) 3355, 1730, 1610, 1580, 1520, 1490 $cm^{-1}$.

The experiment has been carried out on the scale of 40–480 mg with yields in the range of 68–88%.

*Step* 45. (2*S*,3*S*)-20-*Cyanomethyl*-13-*ethyl*-18-*methoxy-carbonyl*-3,7,12,17-*tetramethyl*-8-*vinyl*-*chlorin*-2-*propionic acid methyl ester* (**45**). (*Chlorin* $e_6$ *nitrile* **45**). The cyanolactone (**44**) (21 mg) was dissolved in dichloromethane (2 ml) and acetic acid (11 ml). Zinc dust (2.5 g) was mixed with acetic acid (5 ml) and after 5 min the suspension was added to the cyanolactone solution. The mixture was vigorously shaken until the initial blue colour had changed to intense green (1 min). The zinc dust was immediately removed by filtration (filter aid) and the filter cake was washed with acetic acid and finally with a little dichloromethane. All these operations were conducted under a blanket of nitrogen. The filtrate was taken to dryness under reduced pressure. The residue was dissolved in dichloromethane–ether (1:4) and extracted with 23% HCl. The organic layer was discarded. To demetallate the zinc complex, the acid solution was kept at room temperature for 5 min. After dilution with water, the chlorin components were extracted with dichloromethane. Spectroscopic yield:* 6.7 mg (31%).

To the dry concentrated dichloromethane solution was added methanol (several drops) and freshly distilled ethereal diazomethane (10 min). The chlorin $e_6$ nitrile (**45**) was extracted from ether–dichloromethane (8:1) with 15% HCl, while the unchanged cyanolactone (spectroscopic recovery 1.2 mg, 6%) remained in the organic layer. The acid extract was neutralised with saturated $KHCO_3$. Extraction with dichloromethane gave 4.6 mg* (21%) of chlorins, which were chromatographed on a Florisil column (5 mm diam, 50 mm high). Elution with dichloromethane, followed by dichloromethane–methanol (1000:1) yielded 2.29 mg of material which was crystallised twice from dichloromethane–acetone–methanol giving 1.9 mg (8.8%) of chlorin $e_6$ nitrile (**45**) as attractive crystals, mp 207°. Analytical sample from $CH_2Cl_2$—$Me_2CO$—MeOH, mp 207°. (Found: C, 71.2; H, 6.6. $C_{36}H_{39}N_5O_4$ requires C, 71.4; H, 6.5%). $\lambda_{max}$ ($CH_2Cl_2$) (ε) 499.5 (14 900), 529.5 (6 600), 555 (2 500), 608.5 (6 300), 663.5 nm (48 900). $\nu_{max}$ ($CH_2Cl_2$) 3335, 2240, 1725, 1710 sh, 1600, 1575 sh, 1490. Yields of 7–10% were consistently obtained on a scale of 20–115 mg.

* Estimated assuming ε for chlorin peak at *ca.* 660 nm to be 50,000.

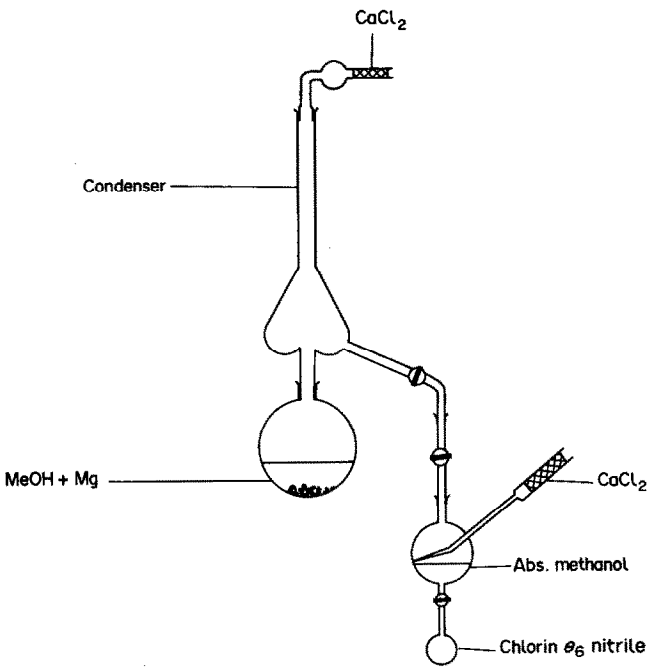



Fig. 25. Apparatus of step 46.

*Step* 46. (2*S*,3*S*)-13-*Ethyl*-18-*methoxycarbonyl*-20-(*methoxycarbonylmethyl*)-3,7,12,17-*tetramethyl*-8-*vinylchlorin*-2-*propionic acid methyl ester* (**46**). *Chlorin* $e_6$ *trimethyl ester*. Glassware was flame-dried in a nitrogen stream and cooled under vacuum. Freshly purified methanol (10 ml, from magnesium) was saturated at 0° with anhydrous hydrogen chloride and poured on to crystalline chlorin $e_6$ nitrile (**45**) (9.4 mg) in the apparatus shown (Fig. 25). After 16 h at room temperature the ice-cooled mixture was poured into ice-water and the green solution was extracted with ether–dichloromethane (8 : 1). If the acidic layer was slightly blue it was diluted further and re-extracted to remove all pigment. The organic solution was washed twice with water and once with 0.5 M-HCl. The chlorins were then extracted with 9% HCl. This acid extract was neutralised with saturated $KHCO_3$ and the pigment was transferred into dichloromethane (spectroscopic yield 7 mg, 71%). The solution was concentrated and chromatographed on a Florisil column (5 mm diam, 55 mm high). Dichloromethane–methanol (ratios 1500 : 1, 750 : 1, 500 : 1, 25 ml of each) eluted 4 mg (40%) of chlorins. This material was combined with like material from a preliminary run (total chlorin = 5.5 mg) and crystallised thrice from dichloromethane–acetone–methanol containing a very small percentage (2–8% of the methanol component) of water to give 3.6 mg of chlorin $e_6$ trimethyl ester (**46**), mp 207.5–208.5°; naturally-derived sample, mp 208–209.5° (lit.[68] mp 211°), mixed mp 207.5–209°. $\lambda_{max}$ ($CH_2Cl_2$) ($\varepsilon$) (nat.) 501 (13 000), 530 (5400), 559 (2 300), 608.5 (5400), 664.5 nm (45 600). The synthetic sample had a visible spectrum identical with that of the naturally-derived sample. $\nu_{max}$ (Nujol) (synth.) (1.3 mg/0.01 ml) 3320, 1730 sh, 1705, 1585, 1575 sh, 1485, 1230, 1155, 1110, 1055, 980, 890, 850, 795, 725 $cm^{-1}$. $\nu_{max}$ (Nujol) (nat.) (1.5 mg/0.01 ml) 3320, 1730 sh, 1710, 1585, 1575 sh, 1485, 1230, 1155, 1110, 1055, 980, 890, 795, 725 $cm^{-1}$. $\nu_{max}$ ($CH_2Cl_2$, 3.8 mg in 0.3 ml, 0.47 mm): synthetic and naturally-derived samples gave identical spectra (Fig. 18).

*Acknowledgements*—We wish to express our warm appreciation to the National Institutes of Health, the National Science Foundation, and the Research Corporation for generous support; to Chas. Pfizer and Co. Inc. for support and invaluable assistance in the preparation of intermediates in quantity; to Dr Arthur Stoll and Sandoz A. G. for a handsome gift of

methyl pheophorbide *a*, and to Dr Stephan Sallay and Dr Wolfgang H. F. Sasse for their cooperation in certain phases of the work.

The writing of this paper was supported by a grant from the Maxwell Foundation which is gratefully acknowledged, and was undertaken by one of us (RB) at the Chemistry Department, Queen Mary and Westfield College, London with the assistance of Miss Diana Garside, Mr Aslam Galia and Dr Maria Asuncion Valles (Barcelona). Spectroscopic and other observations were carried out by Dr Valles and Miss Garside on some of the original samples which were made available through the courtesy of Dr Alfred Bader. We are grateful to Teresa McCarthy for the typescript, and to Dr Mark Baynham for the numerous complex structures.

## REFERENCES

1. Woodward, R. B., Ayer, W. A., Beaton, J. M., Bickelhaupt, F., Bonnett, R., Buchschacher, P., Closs, G. L., Dutier, H., Hannah, J.; Hauck, F. P., Ito, S.; Langemann, A.; Le Goff, E., Leimgruber, W.; Lwowski, W.; Sauer, J.; Valenta, Z.; Volz, H. *J. Am. Chem. Soc.* **1960,** *82*, 3800.
2. Woodward, R. B. *Angew. Chem.* **1960,** *72*, 651.
3. Woodward, R. B. *Pure Appl. Chem.* **1961,** *2*, 383.
4. Lwowski, W. in *The Chlorophylls* (Eds. Vernon, L. P.; Seely, G. R.), Academic Press, New York 1966, p. 119.
5. Fleming, I. *Selected Organic Syntheses*, Wiley, London 1972, p. 112.
6. Stoll, A.; Wiedemann, E. *Fortschr. Chem. Forsch.* **1952,** *2*, 538.
7. Willstätter, R.; Stoll, A. *Untersuchungen über Chlorophyll, Methoden und Ergebnisse*, Springer, Berlin, 1913.
8. For one iteration (ring C reduced) see Fischer, H. *Chem. Rev.* **1937,** *20*, 41.
9. Fischer, H.; Wenderoth, H. *Justus Liebig's Ann. Chem.* **1939,** *537*, 170; **1940,** *545*, 140. Ref. 10, Vol. II.2. p. 23.
10. Fischer, H.; Orth, H.; (Stern, A.) *Die Chemie des Pyrrols* Vol. 1, Vol. II.1, (Vol. II.2), Akademische Verlagsgesellschaft, Leipzig, 1934, 1937, 1940.
11. Ficken, G. E.; Johns, R. B.; Linstead, R. P. *J. Chem. Soc.* **1956,** 2272.
12. Burrell, J. W. K.; Jackman, L. M.; Weedon, B. C. L. *Proc. Chem. Soc.* **1959,** 263. Burrell, J. W. K.; Garwood, R. F.; Jackman, L. M.; Oskay, E.; Weedon, B. C. L. *J. Chem. Soc. C.* **1966,** 2144.
13. Richter, I. referred to in Brockmann, H. in *The Porphyrins* (Ed. Dolphin, D.), Vol. 2, Academic Press, New York 1978, p. 294.
14. Brockmann, H.; Bode, J. *Justus Liebig's Ann. Chem.* **1974,** 1017.
15. Fleming, I. *Nature (Lond).* **1967,** *216*, 151; *J. Chem. Soc. C.* **1968,** 2765. Brockmann, H. *Justus Liebig's Ann. Chem.* **1971,** *754*, 139.
16. Chow, H.-C.; Serlin, R.; Strouse, C. E. *J. Am. Chem. Soc.* **1975,** *97*, 7230. Kratky, C.; Dunitz, J. D. *Acta Cryst.* **1975,** *B31*, 1586.
17. Strell, M.; Kalojanoff, A. *Justus Liebig's Am. Chem.* **1952,** *577*, 97. Treibs, A.; Schmidt, R. *Justus Liebig's Ann. Chem.* **1952,** *577*, 105.
18. Fischer, H.; Balaz, F. *Justus Liebig's Ann. Chem.* **1942,** *553*, 166.
19. IUPAC–IUB Joint Commission on Biochemical Nomenclature, Nomenclature of Tetrapyrroles, Recommendations 1986 in *Eur. J. Biochem.* **1988,** *178*, 277.
20. Fischer, H.; Lautsch, W. *Justus Liebig's Ann. Chem.* **1937,** *528*, 265.
21. Fischer, H.; Oestreicher, A. *Justus Liebig's Ann. Chem.* **1941,** *546*, 49.
22. Willstätter, R.; Forsén, L. *Justus Liebig's Ann. Chem.* **1913,** *396*, 180. Fischer, H.; Goebel, S. *Justus Liebig's Ann. Chem.* **1936,** *524*, 269.

22a. Smith, K. M.; Lewis, W. M. *Tetrahedron* **1981,** *37*, W 399.

23. Fischer, H.; Spielberger, G. *Justus Liebig's Ann. Chem.* **1934,** *510*, 156. Ref. 10, Vol. II.2, p. 53.
24. Willstätter, R.; Stoll, A. *Justus Liebig's Ann. Chem.* **1910,** *378*, 18; **1911,** *380*, 148.
25. For reviews see Johnson, A. W.; and Clezy, P. S.; Jackson, A. H. in *The Porphyrins* (Ed. Dolphin, D.), Vol. 1, Academic Press, New York, 1978, p. 235 and p. 265.
26. Fischer, H.; Süs, O.; Klebs, G. *Justus Liebig's Ann. Chem.* **1931,** *490*, 38.
27. Fischer, H.; Heckmaier, J., Hagert, W. *Justus Liebig's Ann. Chem.* **1933,** *505*, 209.
28. Golden, J. H.; Linstead, R. P.; Whitham, G. H. *J. Chem. Soc.* **1958,** 1725.
29. Bonnett, R.; Gale, I. A. D.; Stephenson, G. F. *J. Chem. Soc. C.* **1966,** 1600.
30. MacDonald, S. F. *J. Chem. Soc.* **1952,** 4176.
31. Knorr, L. *Justus Liebig's Ann. Chem.* **1886,** *236*, 290; Fisher, H. *Org. Synth.* **1943,** Coll. Vol. II, 202.
32. We are much indebted to Mr A. T. Gilman of Chas. Pfizer & Co., Inc. (Groton) for adapting this procedure for pilot plant use, and for preparing large quantities of this intermediate for us.
33. Fischer, H.; Walach, B. *Ber.* **1925,** *58*, 2818.
34. Chu, E. J.-H.; Chu, T. C. *J. Org. Chem.* **1954,** *19*, 266.
35. Fischer, H.; Weiss, B. *Ber.* **1924,** *57*, 602.
36. Fischer, H.; Zeile, K. *Justus Liebig's Ann. Chem.* **1930,** *483*, 251.
37. Fischer, H. *Org. Synth.* **1955,** Coll. Vol. III, 513.
38. Johnson, A. W.; Markham, E.; Price, R.; Shaw, K. B. *J. Chem. Soc.* **1958,** 4254.
39. *Cf.* Fischer, H.; Schubert, F. *Hoppe-Seylers' Z. Physiol. Chem.* **1926,** *155*, 99.
40. Treibs, A.; Schmidt, R. *Justus Liebig's Ann. Chem.* **1952,** *577*, 105.
41. Fischer, H.; Neber, M. *Justus Liebig's Ann. Chem.* **1932,** *496*, 1.
42. Fischer, H.; Wiedemann, O. *Hoppe Seylers' Z. Physiol. Chem.* **1926,** *155*, 59.

43. Kleinspehn, G. G; Corwin, A. H. *J. Am. Chem. Soc.* **1953,** *75*, 5295.
44. Triebs, A., Schmidt, R.; Zinmeister, R. *Chem. Ber.* **1957,** *90*, 84.
45. Cheng, D. O.; Bowman, T. L.; Le Goff, E. *Heterocyclic Chem.* **1976,** *13*, 1145.
46. Corwin, A. H.; Bailey, W. A.; Viohl, P. *J. Am. Chem. Soc.* **1942,** *64*, 1267.
47. Corwin, A. H.; Straughn, J. L. *J. Am. Chem. Soc.* **1948,** *70*, 1416.
48. Corwin, A. H.; Viohl, P. *J. Am. Chem. Soc.* **1944,** *66*, 1137.
49. Fischer, H.; Andersag, H. *Justus Liebig's Ann. Chem.* **1926,** *450*, 201.
50. Morsingh, F.; MacDonald, S. F. *J. Am. Chem. Soc.* **1960,** *82*, 4381.
51. Bullock, E.; Johnson, A. W.; Markham, E.; Shaw, K. B. *J. Chem. Soc.* **1958,** 1430; Johnson, A. W.; Kay, I. T., Markham; E.; Price, R.; Shaw, K. B. *J. Chem. Soc.* **1959,** 3416.
52. Fischer, H.; Lamatsch, W. *Justus Liebig's Ann. Chem.* **1928,** *462*, 240.
53. Reference 10, Vol. 1, p. 282.
54. Fischer, H.; Csukas, Z. *Justus Liebig's Ann. Chem.* **1934,** *508*, 167.
55. Arsenault, G. P.; Bullock, E.; MacDonald, S. F. *J. Am. Chem. Soc.* **1960,** *82*, 4384.
56. Treibs, A.; Fritz, G. *Justus Liebig's Ann. Chem.* **1958,** *611*, 162; Paine, J. B. in *The Porphyrins* (ed. Dolphin, D.) Vol. 1, Academic Press, New York, 1978, p. 101 and p. 170; Bonnett, R.; Buckley, D. G.; Hamzetash, D.; McDonagh, A. F. *Isr. J. Chem.* **1983,** *23*, 173.
57. Fischer, H.; Strell, M. *Justus Liebig's Ann. Chem.* **1939,** *538*, 157.
58. Bonnett, R.; Stewart, J. C. M. *J. Chem. Soc., Perkin Trans. 1.* **1975,** 224.
59. Wolf, H.; Brockmann, H.; Biere, H.; Inhoffen, H. H. *Justus Liebig's Ann. Chem.* **1967,** *704*, 208.
60. Fischer, H.; Strell, M. *Justus Liebig's Ann. Chem.* **1939,** *540*, 232.
61. Fischer, H.; Conrad, M. *Justus Liebig's Ann. Chem.* **1939,** *538*, 143.
62. Thron, H.; Dirscherl, W. *Justus Liebig's Ann. Chem.* **1934,** *515*, 252.
63. Reference 10, Vol. II.2, p. 115.
64. See Fischer, H.; Strell, M. *Justus Liebig's Ann. Chem.* **1940,** *543*, 143.
65. Bellamy, L. J. *The Infra-red Spectra of Complex Molecules*, Methuen, London, 1958, p. 266.
66. Inhoffen, H. H.; Jäger, P.; Mählhop, R. *Justus Liebig's Ann. Chem.* **1971,** *749*, 109.
67. Fischer, H.; Kahr, K. *Justus Liebig's Ann. Chem.* **1937,** *531*, 209.
68. Fischer, H.; Siebel, H. *Justus Liebig's Ann. Chem.* **1932,** *499*, 84.
69. Fischer, H.; Schubert, M. *Ber.* **1923,** *56*, 1202.
70. Fischer, H.; Hussong, M. *Justus Liebig's Ann. Chem.* **1931,** *492*, 128.