A Novel Stereoselective Synthesis of the Macrocycle of Haem d_1 that establishes its Absolute Configuration as 2*R*,7*R*

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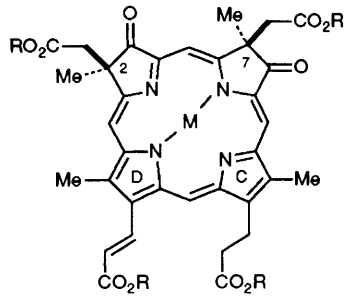
A novel route to isobacteriochlorins is developed that allows the stereoselective synthesis of the macrocycle of haem d_1 and so establishes its absolute configuration 2*R*,7*R*.

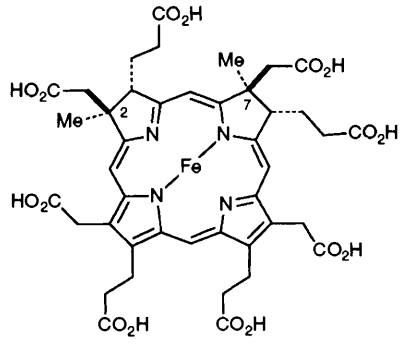
Haem d_1 is the iron-containing prosthetic group of bacterial reductase–cytochrome oxidase enzymes which carry out the reduction of nitrite. It was isolated by Timkovich *et al.*[1] and Chang[2] suggested that the ligand holding the iron is a dioxoisobacteriochlorin, see **2**. That the ester of the ligand has the gross structure **2**, without definition of the stereochemistry, was established by Wu and Chang's synthesis[3] of a mixture of racemic diastereoisomers corresponding to **2**, one of which was identical, apart from being a racemate, with the esterified metal-free ligand from haem d_1. Then Montforts *et al.*[4] showed that the *C*-methyl groups at C-2 and C-7 are *syn*-oriented by a partial synthesis yielding all the diastereoisomers of a related macrocycle (as **25**) followed by X-ray analysis of the racemic diastereoisomer known[3] to correspond to haem d_1. Thus haem d_1 has the absolute configuration **1** or it is the corresponding enantiomer.

It is important to determine which is the true configuration in order to know whether haem d_1 is related stereochemically, and so probably biosynthetically also, to sirohaem (the cofactor for sulfite reductases) and F-430 (the cofactor for methane production). Sirohaem,[5] which is related stereochemically to F-430,[6] has the absolute configuration **3**.

Our plan was to achieve a stereochemically controlled synthesis of the macrocycle of haem d_1, as its ester **2**, by the photochemical approach developed in Cambridge.[7,8] The initial target was the isobacteriochlorin **24**, which requires the synthesis of **23** from the lactams **15** and **19**, Scheme 2. Previously,[7,8] the best way to build **15** and **19** was *via* the nitriles **16** and **20** with subsequent removal of the cyano group by difficult chemistry. The present synthesis eliminates this



1; R = H, M = Fe^{II}
2; R = Me, M = H,H



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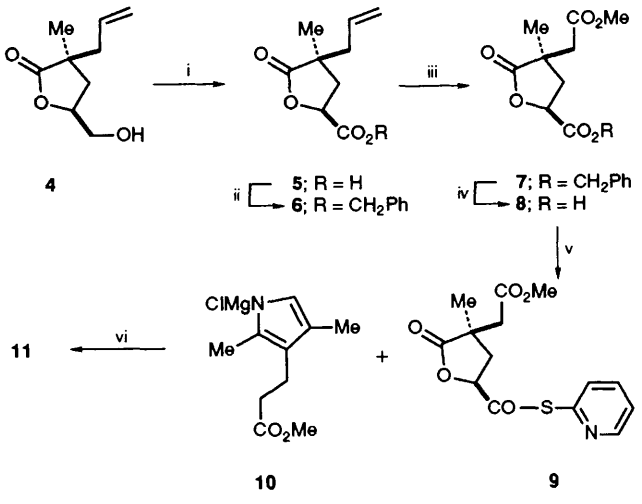


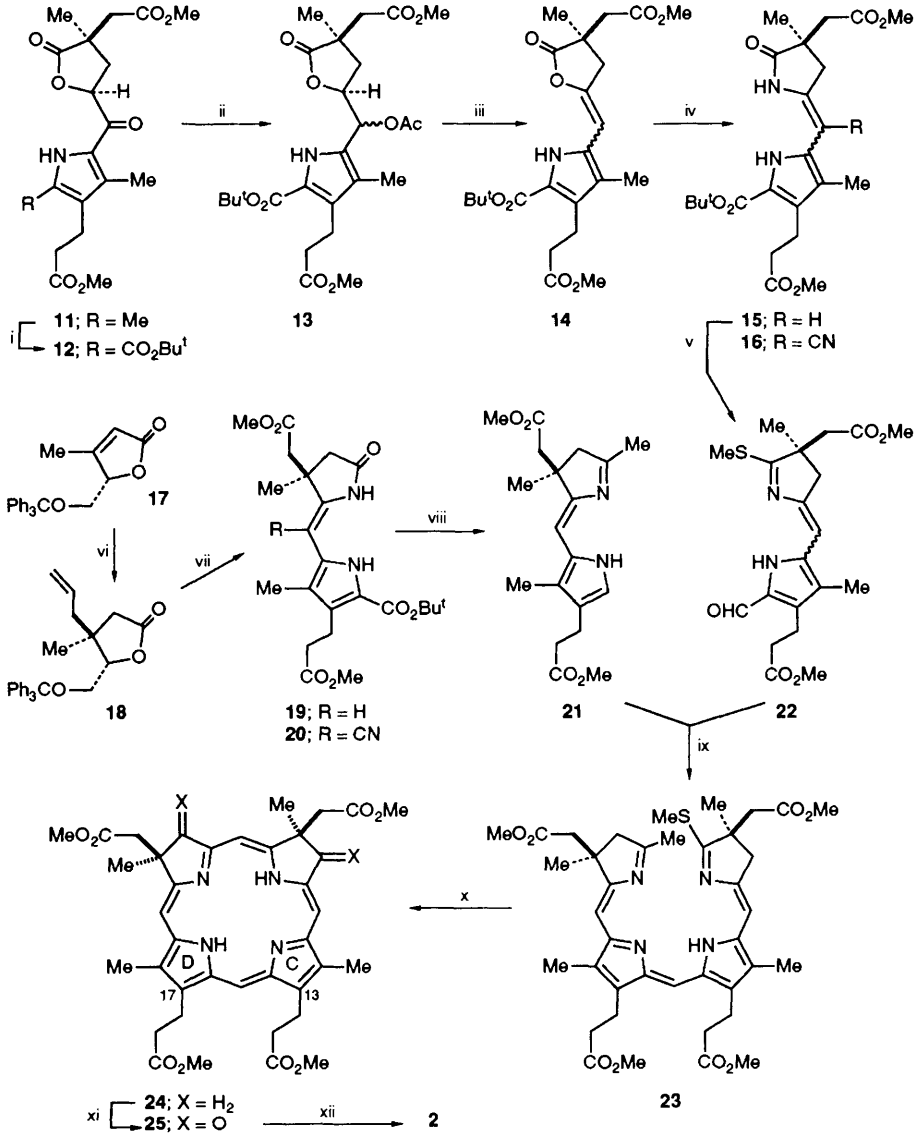
Scheme 1 *Reagents and conditions*: i, CrO_3; ii, $PhCH_2Br$, K_2CO_3; iii, RuO_2, $NaIO_4$, then CrO_3, then CH_2N_2; iv, H_2, Pd/C; v, 2,2′-dipyridyl disulfide, PPh_3; vi, react at −78 °C in toluene

problem by a novel approach readily amenable to large-scale work.

The alcohol[8,9] **4**, available from cheap L-glutamic acid and hence of known absolute configuration,[9] was converted as in Scheme 1 into the thioester **9** which acylated the magnesium pyrrole derivative **10** to yield the ketone **11**. The stabilizing keto function allowed oxidation of the pyrrolic α-methyl group and the keto group of the derived *tert*-butyl ester **12** was reduced and then acetylated to yield two diastereoisomeric *O*-acetates **13**, Scheme 2. Thermal elimination of acetic acid from both acetates gave the enol lactone system **14** as a mixture of *E* and *Z* isomers. These were converted into the *E* and *Z* lactams **15** by the key step which introduced the nitrogen atom.

Synthesis of the complementary lactam **19** involved reaction of an allyl cuprate with the lactone **17**, also of known absolute configuration (Scheme 2), prepared from L-glutamic acid.[10] The product **18** was then converted into the lactam **19** by steps strictly analogous to those used in Schemes 1 and 2 for the isomeric series, **4** → **15**, but in this series essentially only the *Z* forms of the intermediates were produced.

Synthesis of the eastern block **22** as an *E*–*Z* mixture was



Scheme 2 *Reagents and conditions*: i, SO_2Cl_2, then H_2O, then isobutene-H^+; ii, BH_4^-, then Ac_2O, 4-dimethylaminopyridine (DMAP); iii, heat at 200 °C; iv, NH_3, then *p*-$MeC_6H_4SO_3H$; v, Lawesson's reagent, then $(MeO)_3CH$, trifluoroacetic acid (TFA); vi, $(allyl)_2Cu(CN)Li_2$; vii, *cf.* **4** → **15**; viii, as in refs. 7 and 8; ix, H^+; x, *hν*; xi, SeO_2; xii, OsO_4, then H^+

completed, Scheme 2, by conversion of **15** by Lawesson's reagent into the corresponding thiolactams, followed by one-pot formylation and *S*-methylation using trimethyl orthoformate–trifluoroacetic acid. The previously developed methodology[7,8] allowed preparation of the imine **21** from the lactam **19** to act as the western block. The yields in Schemes 1 and 2 leading to **21** and **22**, except for **17** → **18** which requires optimisation, are at least good (≥ 65%) and most are very good (≥ 80%). Condensation of **21** and **22** gave the *seco*-system **23**, which on irradiation underwent an 18π-electron electrocyclic ring-closure (after a presumed imine–enamine tautomerisation, generating an *exo*-methylene group at the site of cyclisation) to afford the key isobacteriochlorin **24**, 45–50% from the building blocks **21** and **22**.

Before the development of this new route, the macrocycle **24**, identical with the present product, had been synthesised in small quantity by the original 'nitrile route' alluded to at the outset; this chemistry will be described in our full papers.

Selenium dioxide smoothly converted **24** into the dioxo-system **25** and the last step to form **2** followed Wu and Chang's method[3] developed on racemic **25**. This involves formation of the diol on the β-position of ring-*D* of **25** using OsO_4 and then acid-catalysed, reversible elimination of water (giving the exocyclic double bond at C-17) followed by a 1,3-allylic rearrangement and elimination of a second water molecule to give the acrylate **2** as the end product. The same process on ring-*C* generates a minor separable isomer having the acrylate residue on C-13, both isomers being enantiomerically pure and of known absolute configuration.

The former product **2** was identical by ^{1}H NMR and UV–VIS spectroscopy and TLC comparison with an authentic sample of the ligand tetramethyl ester prepared from haem d_1, kindly supplied by Professor R. Timkovich. Importantly, the circular dichroism spectra of the synthetic and natural samples matched closely.

It is thus established that haem d_1 has the illustrated 2*R*,7*R* absolute configuration **1** and is related stereochemically, and so probably biosynthetically also, to sirohaem **3** and F-430. The biological mechanism for removal of the propionate side-chains from some earlier precursor leading finally to haem d_1 poses interesting problems.

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