Intermediates between Uroporphyrinogen-1 and Coproporphyrinogen-1

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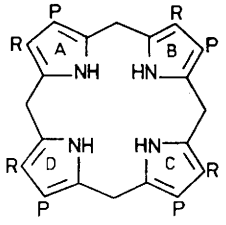
Summary Uroporphyrinogen-I is converted into coproporphyrinogen-I by uroporphyrinogen decarboxylase by two simultaneous routes, in contrast to the apparently specific pathway between uroporphyrinogen-III and coproporphyrinogen-III.

IN certain porphyrias (disorders of haem biosynthesis) isomeric porphyrins of the so-called 'type-I' series may be excreted as well as the normal 'type-III' isomers owing to a defect in the enzyme system catalysing the synthesis of uroporphyrinogen-III (**1a**) from the monopyrrolic porphobilinogen.[1] Uroporphyrinogen-III (**1a**) undergoes a further



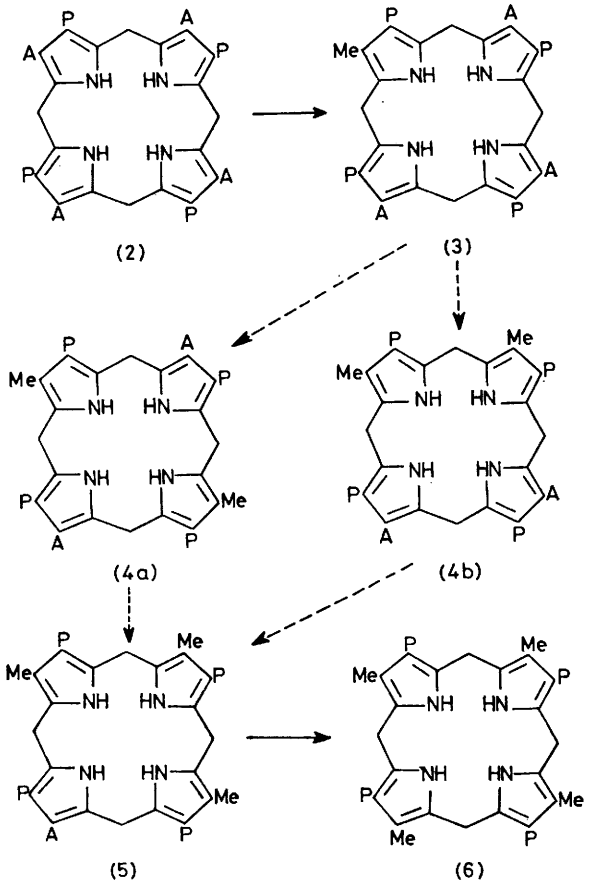
(1)

a; R = A

b; R = Me

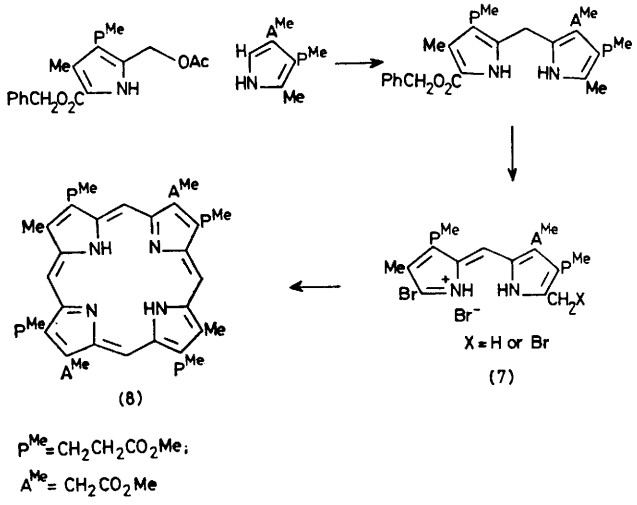
P = $CH_2CH_2CO_2H$; A = CH_2CO_2H

sequence of metabolic changes involving a sequential clockwise decarboxylation of the four acetic acid residues starting with that on the D-ring and ending with that on the C-ring;[2] the resulting coproporphyrinogen-III (**1b**) is then further metabolised[3] to protoporphyrin-IX, the normal branch point for the haem and chlorophyll pathways.



SCHEME 1

In theory there are 24 possible routes from uroporphyrinogen-III (**1a**) to coproporphyrinogen-III (**1b**) but we have shown[2] that nature apparently prefers only one of these routes, both in normal and abnormal metabolism, even though uroporphyrinogen decarboxylase is apparently capable of metabolising any one of the fourteen possible intermediate hepta-, hexa-, and penta-carboxylic porphyrinogens to coproporphyrinogen-III. However owing to the centrosymmetrical character of uroporphyrinogen-I (**2**) there are only two possible routes (Scheme 1) to coproporphyrinogen-I (**6**) with one hepta- two hexa-, and one penta-carboxylic intermediates. We have therefore synthesised the related porphyrins (as their methyl esters) for comparison with the naturally-occurring 'type-I' isomers.



(8)

P^{Me} = $CH_2CH_2CO_2Me$;

A^{Me} = CH_2CO_2Me

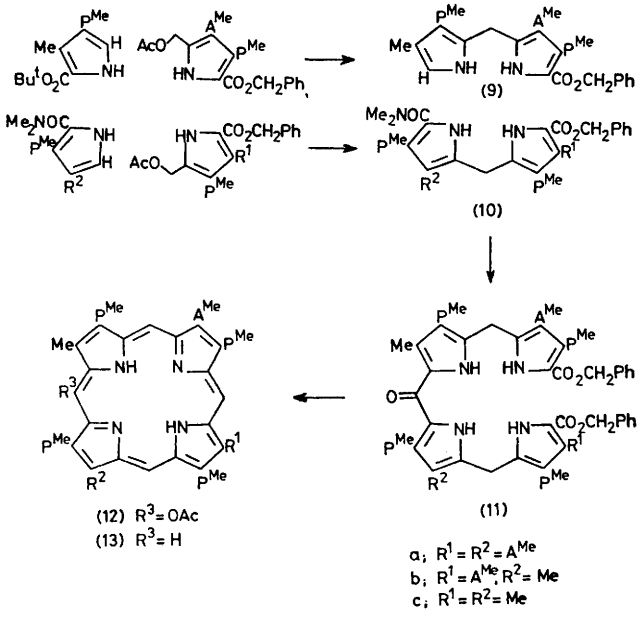
(7)

X = H or Br

SCHEME 2

One of the two hexacarboxylic porphyrins (**8**) has an element of symmetry, and the classical Fischer method[4] was therefore chosen for its synthesis. The required pyrromethene (**7**) was prepared from pyrrolic intermediates as indicated in Scheme 2, and on fusion in methylsuccinic acid, followed by re-esterification, it gave the hexacarboxylic porphyrin hexamethyl ester in 11% yield.

The synthesis of the other three porphyrins (**13a**), (**13b**), and (**13c**) necessitated a route through a rationally constructed open-chain tetrapyrrole. The *b*-oxobilane route[5] was adopted for this purpose, and the synthesis of the required intermediate pyrromethanes is shown in Scheme 3. The close structural relationship of the three porphyrins enabled us to utilise the same pyrromethane (**9**) for two of the pyrrole rings of each porphyrin, and this was coupled with each of the pyrromethane amides (**10a**, **b**, and **c**) to afford the *b*-oxobilanes (**11a**, **b**, and **c**), respectively. Of the eight possible ways this particular route was also chosen on the basis of previous experience[2,6] with the *b*-oxobilane method so that the oxo-bridge was formed by condensation at an α-position flanked by a neighbouring β-methyl group, rather than by a β-propionate or acetate side-chain. The oxobilanes (**11**) were converted in the usual manner into the related *meso*-acetoxyporphyrins (**12a**) (28%), (**12b**) (12%), and (**12c**) (24%) as shown in Scheme 3, and the latter were then transformed into the desired *meso*-unsubstituted porphyrins (**13a**), (**13b**), and (**13c**) by hydrogenolysis and re-oxidation.[5]



SCHEME 3

T.l.c., h.p.l.c.,[7] and m.p. comparisons confirmed the identity of the hepta- and penta-carboxylic esters with the esters of the corresponding type-I porphyrins isolated from the urine of a porphyric bull with congenital erythropoetic protoporphyria.[8] M.p. comparisons with the small amount of natural hexacarboxylic porphyrin hexamethyl ester available were inconclusive, but careful h.p.l.c. analysis showed that both type-I hexacarboxylic porphyrins were present, together with a small amount of the natural type-III hexacarboxylic porphyrin,[2] as shown by comparisons with the synthetic materials (see Figure). We also studied several samples of hexacarboxylic porphyrins obtained from porphyric patients, and each of these likewise showed the presence of all three compounds, although the relative proportions varied somewhat; with one patient (M.D.), for example, very little of the type-III isomer was present, but almost equal amounts of the type-I isomers.

Preliminary kinetic studies of the metabolism of the type-I porphyrinogens by haemolysates of chicken erythrocytes[9] showed that they are all readily converted into coproporphyrinogen-I. The hexacarboxylic porphyrin obtained from short incubations of the type-I heptacarboxylic porphyrinogen was shown by h.p.l.c. to be a mixture of approximately equal amounts of both isomers (**8**) and (**13b**) (see Figure).

We conclude that decarboxylation of uroporphyrinogen-I by uroporphyrinogen decarboxylase occurs by a non-specific route, in contrast to the highly ordered 'clockwise' decarboxylation of uroporphyrinogen-III by the same enzyme.[2]

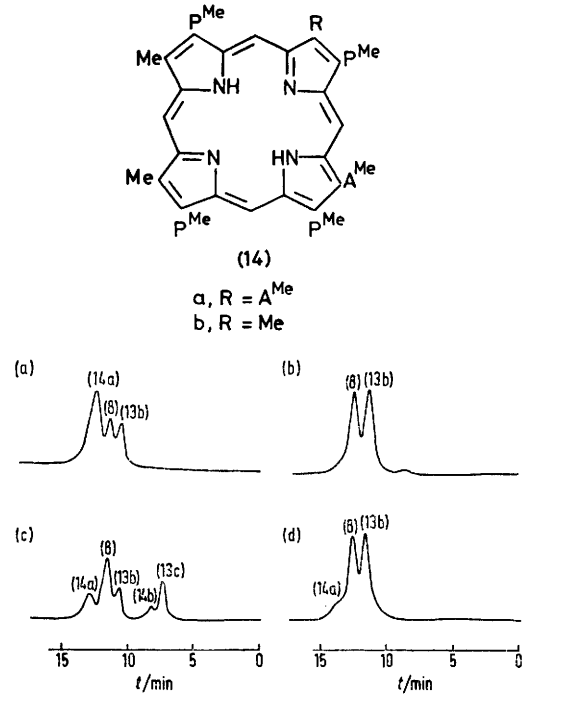


FIGURE. H.p.l.c. analysis of hexacarboxylic porphyrin hexamethyl esters on a 15 × 0·5 cm 5 μ Partisil column in 1% Me_2CO in $CHCl_3$ at a flow rate of 1 ml min^{-1}. (In view of the potential hazards associated with keeping this solvent mixture, it was only made up as required and not stored. 4% Me_2CO in 1,2-dichloroethane also gave satisfactory results. (a) Mixture of synthetic 'type-I' and 'type-III' hexacarboxylic porphyrin hexamethyl esters, (**8**), (**13b**), and (**14a**). (b) Hexacarboxylic porphyrin hexamethyl ester fraction isolated from incubation of type-I heptacarboxylic porphyrinogen (**3**) with haemolysates of chicken red blood cells. [Note the presence of both type-I isomers, (**8**) and (**13b**)]. (c) Hexacarboxylic porphyrin methyl ester fraction obtained by Professor T. K. With from the urine of a porphyric bull. [Note the presence of both type-I and type-III hexacarboxylic porphyrins, and also the presence of some type-I and type-III pentacarboxylic porphyrins, (**13c**) and (**14b**)]. (d) Hexacarboxylic porphyrin methyl ester fraction obtained from a human porphyric (M.D.), kindly supplied by Dr. D. C. Nicholson and Dr. G. H. Elder. [(Note the presence of both type-I porphyrins (**8**) and (**13b**) but only traces of the type-III porphyrin, (**14a**)].

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