Synthesis along Biosynthetic Pathways. Part 2.[1,2] Synthesis of Protostephanine

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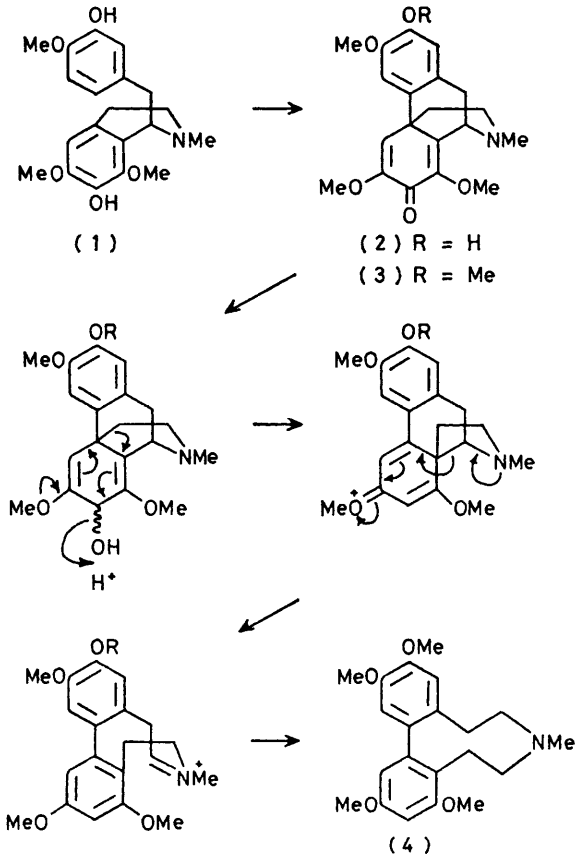
The dienone protostephanone (3) is synthesised by phenol coupling or by Pschorr cyclisation from readily prepared tetrahydroisoquinolines, and the corresponding dienols (17) and (18) are converted by rearrangement, fragmentation, and reduction into protostephanine (4). This sequence mimics the natural pathway to the alkaloid.

BIOSYNTHETIC studies (see the following papers) on the unusual dibenzazonine structure of protostephanine[3] (4) sparked our interest in achieving its synthesis by a route analogous to the biosynthetic pathway; a synthesis which was not biomimetic had already been described.[4] Experimental work on the living plant had been guided at the outset by Barton's proposal[5] that phenol oxidation is a key step in the formation of protostephanine (4) and his seminal hypothesis is shown in Scheme 1.

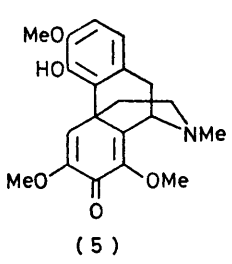


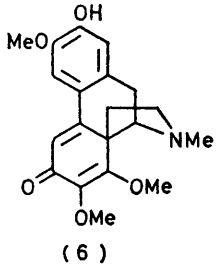
SCHEME 1

Our synthetic study fell into two parts. First, development of the best route for synthesis of the dienone (3) and secondly, studies of the steps involving rearrangement and fragmentation (Scheme 1) which, it was hoped, would generate the nine-membered nitrogenous ring.

The diphenol (1) required for the first part was synthesised by standard methods; these were also used for synthesis of labelled materials and are described in one of the following papers.[6] The present Experimental section includes useful methods for the preparation of intermediates and for characterisation of the products.

When the diphenol (1) was oxidised with alkaline ferricyanide in a two-phase system, a complex mixture resulted which was fractionated by partition chromatography followed by preparative thin-layer chromatography. Four products were isolated and the structures of two were rigorously established. The first, in 2% yield, was the desired phenolic dienone (2), $C_{20}H_{23}NO_5$ by accurate mass measurement, which showed i.r. and n.m.r. spectra characteristic of a cross-conjugated dienone. N.m.r. spectroscopy also distinguished the dienone (2) from its isomer (5) which could, in principle, be formed in that the protons on the aromatic ring of the product gave rise to two 1H-singlets. Those from the



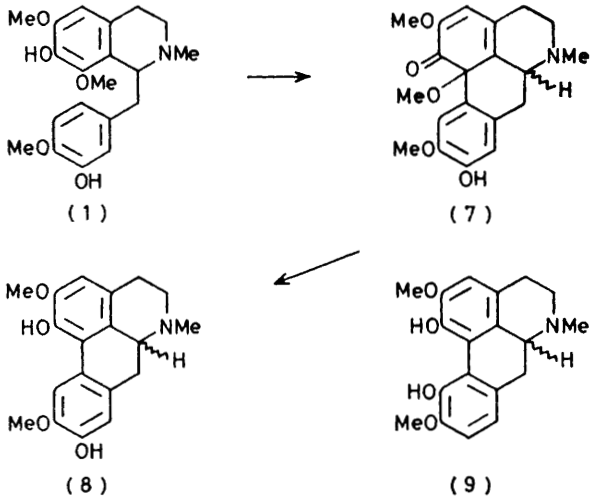


isomer (5) would appear either as an AB double doublet or as a 2H-singlet if the chemical shifts happened fortuitously to be identical. Finally, the u.v. spectrum of the product corresponded to an isolated aromatic ring (280 nm) which eliminated structures having extended conjugation such as (6) which plausibly could arise by phenol coupling followed by rearrangement (see later).

A second product was the racemic form of isoboldine (8), a known alkaloid, and this was identified by full spectroscopic and chromatographic comparison with an (*RS*)-sample synthesised earlier by Jackson and Martin.[7] A possible alternative structure (9) corresponds to (*RS*)-corytuberine[8] and this was eliminated by showing that the synthetic product differed from natural corytuberine by t.l.c., u.v., and mass spectrometry. A search was made for (*RS*)-corytuberine (9) in the total products

from the phenol coupling step but none was detected which means that <0.1% had been produced.

It is probable that the isoboldine (8) arises by breakdown of a conjugated dienone (7) (Scheme 2) but further work would be needed to establish this point; acid-catalysed loss of such a methoxy-group is known.[9]



Scheme 2

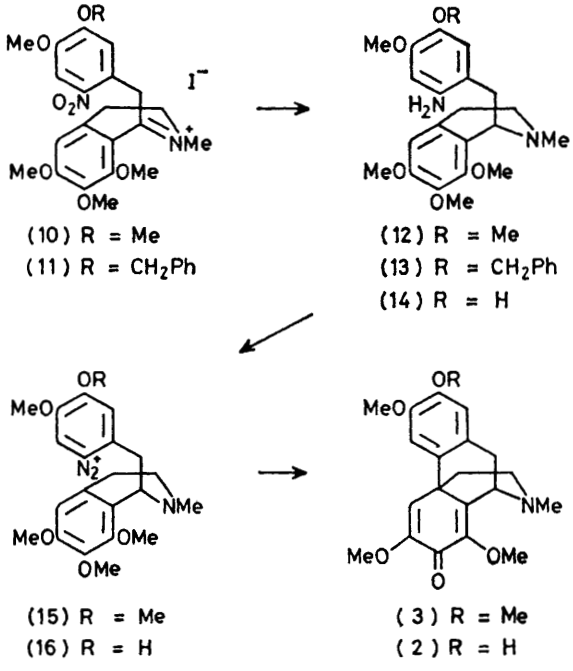
The structures of the remaining two products were not fully elucidated. However, they were isomeric and their compositions corresponded to the removal of 2 H from the diphenol (1), *i.e.* they were products from intramolecular coupling. All the spectroscopic data were in keeping with these substances being stereoisomers of structure (7) formed by *o,p* coupling, though the alternative *o,o* coupling cannot be excluded. In particular, the base peak in the mass spectrum of both substances ($M^+ - CH_2O - H$) corresponded to the base peak of isoboldine ($M^+ - H$) and the fragments of lower molecular weight matched those from isoboldine. A breakdown of (7) to (8) in the mass spectrometer is a very reasonable fragmentation.

In order to obtain greater quantities of the dienone (2) and its *O*-methyl ether (3), Pschorr coupling of aromatic rings was investigated. Hey and his co-workers [10] first observed dienone formation in Pschorr reactions and the value of this approach for our purpose was clear from the preparation of morphinandienones in this way by Gregson-Allcott and Osbond.[11] Accordingly, the nitrodihydroisoquinolinium salt (10) was synthesised by well known methods. It was reduced to the aminotetrahydroisoquinoline (12) and the corresponding diazo-derivative (15) was treated with copper powder (Scheme 3). This afforded the non-phenolic dienone (3), named protostephanone and the yield under the best conditions in several runs was 18—25%. Though this yield is not high, it will stand comparison with the likely overall yield for any multistage construction of protostephanone (3).

Analogous Pschorr coupling of the phenolic diazo-derivative (16) [obtained by the sequence (11) ⟶ (13) ⟶ (14) ⟶ (16) (Scheme 3)] gave the phenolic dienone (2) in 3% yield. This step was not optimised and in our view, further experimentation would increase the yield; the initial yield of protostephanone (3) was 5% and this was eventually raised to 25%.

The phenolic dienone (2) from the Pschorr route was identical with the product obtained earlier by phenol oxidation. Also, *O*-methylation of the phenol (2) with diazomethane gave protostephanone (3). The interlocking set of dienones was thus complete and attention turned to the second phase of the synthesis.

Controlled reduction of protostephanone (3) with borohydride quantitatively afforded a mixture of the dienols (17) and (18), which were separated for spectroscopic characterisation. Since, however, the stereochemistry at the asterisked carbon was unlikely to be

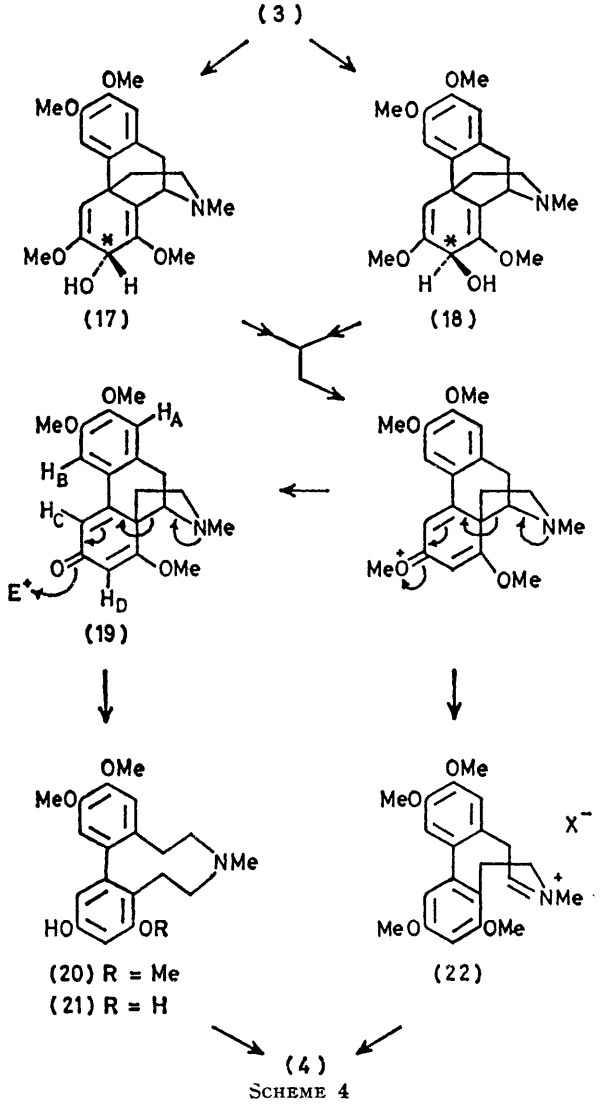


Scheme 3

synthetically important (*cf.* related examples in the morphine series [12]), the dienols were used together for the next step. Initially this involved treatment of the mixed dienols (17) and (18) with sulphuric acid in methanol when a new dienone-B, $C_{20}H_{23}NO_4$, was formed in 89% yield; the molecular formula corresponds to loss of OH and Me from the dienols. The n.m.r. spectrum of dienone-*B* was in agreement with structure (19) by showing 1H-singlets (δ 7.0 and 6.78) for H_A and H_B and 1H-doublets (*J* 2 Hz, δ 6.28 and 5.61) for H_C and H_D, respectively. The appearance of coupling for the dienone protons H_C and H_D and the size of *J* was as expected.[13] Also, the long-wavelength absorption of dienone-B at $\lambda_{max.}$ 352 nm showed extended conjugation.

The evidence so far does not conclusively rule out the mechanistically plausible structure (23) for dienone-B though the u.v. absorption for structure (23) would be

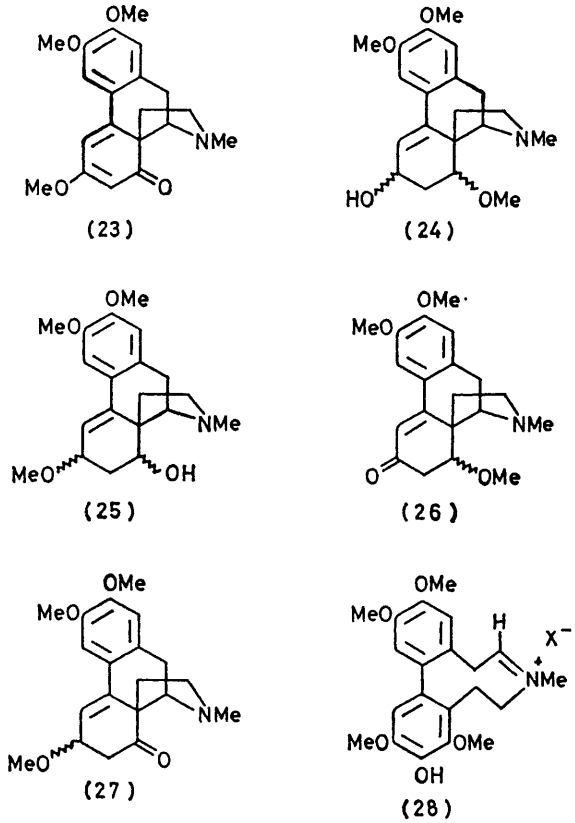
expected, by approximate calculation,[14] to lie around 400 nm (*cf.* observed value above). Clear support for structure (19) was obtained by reduction of protostephanone (3) with borodeuteride followed by acid-catalysed rearrangement to give monodeuterio-dienone-B [as (19)]. The high-field olefinic signal (δ 5.61) was now absent and that at low-field (δ 6.28) was a singlet.



SCHEME 4

In addition, borohydride reduction of unlabelled dienone-B afforded the enols (24), or possibly (25), M^+ 345. This mixed product showed $\lambda_{max.}$ at 297 nm in agreement with a styrene-like chromophore. Jones oxidation of the mixture yielded an enone, M^+ 343, having structure (26) rather than (27) because the long-wavelength band in its u.v. spectrum had shifted to 345 nm; the n.m.r. spectra of the enols (24) and enone (26) were also in agreement with the assigned structures (see Experimental section).

Having obtained dienone-B (19), the desired rearrangement step had been achieved and it remained to bring about the fragmentation (19) ⟶ (20) shown in Scheme 4. Dienone-B was stable to mineral acids; *e.g.* it was recovered unchanged after treatment with concentrated sulphuric acid at 20 °C. We therefore reasoned that it should be heated with anhydrous magnesium iodide to encourage co-ordination at oxygen whilst leaving at least some of the basic centres free to participate in the fragmentation process. This ploy successfully gave a phenolic mixture which was treated with lithium aluminium hydride to reduce the C=$\overset{+}{N}$Me residue. The product was shown by mass spectrometry to contain the phenols (20) and (21) in a combined yield of 46%. *O*-Methylation of the mixture with diazomethane then



afforded protostephanine (4), identified by full comparison with the natural alkaloid.

Although successfully yielding the product, this sequence from the dienols (17) and (18) to protostephanine (4) was experimentally capricious and the last two stages gave unsatisfactory yields. A lead to better conditions came from the observation that the n.m.r. spectrum of protostephanone (3) in neat trifluoroacetic acid (TFA) showed a broad signal (*ca.* 0.5 H) at δ 8.1; this was interpreted as arising from the imine proton of (28), the salt being produced by acid-catalysed rearrangement of the dienone (3) followed by fragmentation. The use of TFA by Kirby *et al.*[15] to promote an analogous rearrangement–fragmentation of thebaine gave added encouragement. Accordingly, the mixture of dienols (17) and (18) was treated with TFA, the acid was evaporated,

and the residue reduced with borohydride. Protostephanine (4) was then isolated in 60% overall yield from the starting dienols.

The sequence (12) ⟶ (3) ⟶ (17) and (18) ⟶ (4) represents an efficient synthesis of protostephanine (4) which mimics the key stages of its biosynthesis. This method has already proved useful for the preparation of specifically labelled alkaloid.[16]

There have been a number of developments relating to protostephanine since the completion of our work (see ref. 2). The most relevant are (a) the use of the Pschorr reaction for synthesis of a range of dienone alkaloids,[17] (b) the synthesis of protostephanone (3) by anodic oxidation,[18] and formation of dibenzazonines by direct coupling,[18b] and (c) the observation of reactions closely similar to those described above leading to other dibenzazonine systems.[19]

EXPERIMENTAL

For general directions, see refs. 6 and 16.

O-Benzylsyringaldehyde from Gallic Acid.—(a) Gallic acid was methylated in the standard way with dimethyl sulphate and the resultant methyl 3,4,5-trimethoxybenzoate (20 g) was stirred with concentrated sulphuric acid (100 ml) at 40 °C for 8 h and then at 20 °C for 12 h. The precipitate which formed when the acidic solution was poured onto ice (500 g) was collected, washed with water, and recrystallised from benzene and then water to give 4-hydroxy-3,5-dimethoxybenzoic acid (syringic acid), m.p. 203 °C (lit.,[20] 204—206.5 °C) (9.2 g, 53%).

This acid (10 g) was converted as earlier [6] into *O*-benzylsyringic acid (12.6 g), m.p. 159 °C (lit.,[21] 140—150 °C). Part of it (5.8 g) in dry benzene (80 ml) was stirred for 1 h with oxalyl chloride (2 g) and dimethylformamide (2 drops). The solution was evaporated repeatedly from benzene and the resultant acid chloride in dry xylene (70 ml) was mixed with 10% palladium–barium sulphate (poisoned as usual with sulphur–quinoline). The mixture was stirred and heated under reflux while hydrogen was bubbled through it and into a water trap. Titration of the latter with sodium hydroxide solution monitored the course of the Rosenmund reduction which was complete in 12 h. After the solution had been filtered, it was evaporated and the residue was recrystallised from benzene–heptane and from methanol to give *O*-benzylsyringaldehyde (5 g), m.p. 62 °C (lit.,[22] 63 °C), M^+ 272, identical with material prepared [6] by standard *O*-benzylation of syringaldehyde.

(b) (*With Dr. A. P. Ottridge*). An alternative method involved reduction of *O*-benzylsyringic acid (56 g) with lithium aluminium hydride [6] to give 4-benzyloxy-3,5-dimethoxybenzyl alcohol (53 g).

Chromium trioxide (23 g, 0.23 mol) was slowly added to a stirred solution of pyridine (38 ml, 0.23 mol) in dry dichloromethane (500 ml). After the mixture had been stirred for 15 min, a solution of the above benzyl alcohol (10 g, 0.036 mol) in dichloromethane (200 ml) was added and the mixture was stirred for 1 h. The solution was then decanted and the solids were extracted with dichloromethane (3 × 250 ml), these extracts being added to the main solution which was then evaporated to *ca.* 1 l. This was washed with 2N-sodium hydroxide (3 × 300 ml), 2N-hydrochloric acid (3 × 300 ml), saturated aqueous sodium hydrogen carbonate (300 ml), and saturated brine (300 ml). Evaporation of the dried organic layer gave *O*-benzylsyringaldehyde (8.2 g, 89%), m.p. 60 °C, $\nu_{max.}$ 1 680 and 1595 cm^{-1}, which was identical with material prepared by the Rosenmund route.

7-Benzyloxy-1-(3-benzyloxy-4-methoxybenzyl)-6,8-dimethoxy-2-methyl-1,2,3,4-tetrahydroisoquinoline and the Corresponding Diphenol (1).—The *O*-benzylated isoquinoline was synthesised by methods described in ref. 6; the corresponding picrate had m.p. 143.5—144.5 °C (decomp.) (from methanol). Recovery of the base from the picrate, by passing a chloroformic solution down a column of basic alumina, showed *m/e* 539.269 (required mass 539.267).

A solution of the foregoing base (from 384 mg of picrate) in ethanol (10 ml), water (2 ml), and concentrated hydrochloric acid (0.38 ml) was shaken with 10% palladium–charcoal (140 mg) and hydrogen at room temperature and pressure. Uptake of hydrogen being complete after 3 h, the solution was then filtered and evaporated to leave the diphenolic base hydrochloride (210 mg), shown by n.m.r. (in TFA) to be fully debenzylated.

Dienone Formation by Phenol Oxidation.—The foregoing phenolic hydrochloride (210 mg) in a two-phase mixture of methylene chloride (190 ml) and saturated aqueous sodium hydrogen carbonate was shaken under nitrogen with potassium ferricyanide (350 mg) for 35 min. The separated aqueous layer was extracted with methylene chloride (3 × 50 ml), the combined solutions in methylene chloride were washed with water, dried, and evaporated to yield an oil shown (t.l.c.) to contain at least 10 components; initial separation was done by partition chromatography.

Celite (18 g) was washed with 2M-acid, water, methanol, and ether, dried at 100 °C, and cooled. This was then thoroughly shaken with 0.5M-phosphate buffer (9 ml), pH 5.8, which had been equilibrated with hexane containing 17% (by vol.) methylene chloride; the ' dry ' powder was packed into a column. The crude oxidation products were dissolved in the minimum mixture of the above phosphate buffer and organic phase and the two phases were absorbed on Celite (1 g) which was packed onto the top of the Celite column. Elution was then carried out with 17% methylene chloride in hexane which had previously been equilibrated with the aqueous buffer. The promising fractions (by t.l.c. control) were combined (total 14.5 mg) and fractionated by preparative t.l.c. on silica with 10% methanol in chloroform to give *de-O-methylprotostephanone* (2) (3 mg), R_F 0.25 (Found: *m/e*, 357.157 7 and 342.133 1. $C_{20}H_{23}NO_5$ requires *M*, 357.157 6 and *M* − 15, 342.1341); $\lambda_{max.}$ 212, 264, and 280 nm, shifted in base to 219, 259, and 295 nm; $\nu_{max.}$ 3 550, 1 663, 1 625, and 1 595 cm^{-1}; δ 6.73 (1 H, s), 6.66 (1 H, s), 6.30 (1 H, s), 3.83 (3 H, s), 3.77 (3 H, s), 3.75 (3 H, s), and 2.47 (3 H, s); *m/e* 357 (M^+, 100%), 342 (43), 326 (20), 315 (20), 314 (94), 299 (91), and 272 (43); and *isoboldine* (8) (4.4 mg), R_F 0.4 (Found: M^+, 327.1471. $C_{19}H_{21}NO_4$ requires *M*, 327.1470); $\lambda_{max.}$ 280 and 306 nm, shifted in base to 285 and 337 nm; δ 7.97 (1 H, s), 6.66 (1 H, s), 6.40 (1 H, s), 3.86 (3 H, s), 3.84 (3 H, s), and 2.69 (3 H, s); identified by comparison with authentic material.[7]

Two products of molecular weight 357 had higher R_F than isoboldine. One (1.1 mg) showed $\lambda_{max.}$ 217, 270—280, 303, and 315 nm shifted in base to 219, 250, 283, 300sh, and 335—345 nm; $\nu_{max.}$ 3 520, 1 655, and 1 625 cm^{-1}; *m/e* 357 (M^+, 30%), 327 (79), 326 (100), 314 (40), 312 (45), 310 (40), 284 (44), 256 (46), 253 (34), and 222 (93). The other (1.3 mg) showed $\lambda_{max.}$ 218, 270—280, 303sh, and

315sh nm shifted in base to 221, 250, 280, 302sh, and 325 (infl.) nm; $\nu_{max.}$ 3 520, 1 655, and 1 625 cm^{-1}; *m/e* 357 (M^+, 5%), 327 (90), 326 (100), 312 (40), 284 (42), 253 (29), and 222 (100).

Dienone Formation by Pschorr Reaction.—(a) *Synthesis of the aminotetrahydroisoquinoline* (12). 4,5-Dimethoxy-2-nitrophenylacetic acid [23] (10 g) suspended in anhydrous benzene (500 ml) was treated with oxalyl chloride (6 g) in benzene (15 ml) followed by dimethylformamide (5 drops). When effervescence slowed, the mixture was stirred at 20 °C for 15 min and then evaporated at <30 °C. The residue in chloroform (30 ml) was added to a vigorously stirred mixture of 3,4,5-trimethoxyphenethylamine (mescaline) (9.5 g) in ether (300 ml) and excess of saturated aqueous sodium hydrogen carbonate. Stirring was continued for 15 min when solid precipitated. After addition of ether (300 ml), the solid was collected and recrystallised from ethyl acetate to give *4,5-dimethoxy-2-nitro*-N-(*3,4,5-trimethoxyphenethyl*)*phenylacetamide* (16.7 g, 93%), m.p. *159*—160 °C (Found: C, 58.1; H, 5.0; N, 6.5. $C_{21}H_{26}N_2O_8$ requires C, 58.1; H, 6.0; N, 6.5%); δ_C 169.3 (CONH), 153.2, 134.3, 125.3, 114.5, 108.2, 105.6, 60.7, 56.4, 56.2, 56.0, 41.3, 40.7, and 35.8.

A solution of the foregoing amide (5 g) in dry chloroform (100 ml) [or, alternatively, acetonitrile (150 ml)] was heated under reflux with freshly distilled phosphorus oxychloride (3 ml) for 25 min. Evaporation left a gum which in methanol (6 ml) was made alkaline with 2M-ammonium hydroxide (at 0 °C) and this mixture, with added water (200 ml), was extracted with chloroform (3 × 100 ml). This gave a product which crystallised from methanol to yield 1-(*4,5-dimethoxy-2-nitrobenzyl*)-*3,4-dihydro*-*6,7,8-trimethoxyisoquinoline* (3.9 g, 81%), m.p. 146—148 °C. The corresponding *picrate* crystallised from methanol, m.p. 106—108 °C (Found: C, 50.0; H, 4.2; N, 11.0. $C_{27}H_{27}N_5O_{14}$ requires C, 50.2; H, 4.2; N, 10.9%). The isoquinoline ring closure was also successful in benzene, 1 h reflux, 75% yield. The dihydroisoquinoline hydrochloride, m.p. 162—165 °C showed as sharp signals δ 7.70, 6.95, and 6.62 (each 1 H, ArH), 4.72 (2 H, d, *J* 1.5 Hz, $ArCH_2$), 4.03 (6 H), 3.97 (3 H), 3.94 (3 H), and 3.80 (3 H) (5 × OMe), and 2.91 (2 H, t, *J* 7 Hz, $ArCH_2$).

The foregoing dihydroisoquinoline (5 g) and methyl iodide (100 ml) were heated under reflux in the dark for 3 h and the solvent was then evaporated to low volume and the solid was collected (5.6 g, 84%). This methiodide crystallised from methanol, m.p. 179—181 °C (decomp.). The corresponding *perchlorate* crystallised from methanol, m.p. 195—196 °C (Found: C, 49.6; H, 5.2; N, 5.1. $C_{22}H_{26}ClN_2O_{11}$ requires C, 49.9; H, 5.0; N, 5.3%). The methiodide showed δ_C 175.1 (C=$\overset{+}{N}$), 136.0, 124.7, 114.3, 107.8, 106.6, 62.4, 60.8, 58.0, 56.7, 56.2, 53.9, 46.9, 38.6, and 27.3.

The methiodide (3 g) was partly dissolved and partly suspended in concentrated hydrochloric acid (100 ml) and water (50 ml) and zinc dust was gradually added with stirring until the organic material was in solution and almost colourless. The filtrate from removal of the zinc was basified with ammonium hydroxide (cooling) and then extracted thrice with ether. The base (12) so obtained was converted into its *dipicrate* (2.8 g, 61%), m.p. 162—164 °C (from methanol) (Found: C, 47.4; H, 4.5; N, 12.9. $C_{34}H_{36}N_8O_{19}$ requires C, 47.4; H, 4.2; N, 13.0%). This product could also be prepared in two stages as follows.

(*With Dr. A. P. Ottridge*). A solution of the above dihydroisoquinolinium methiodide (1.2 g) in ethanol (50 ml) at 0 °C was treated with an excess of sodium borohydride. After acidification with 2M-sulphuric acid, the solution was evaporated to remove the ethanol and the mainly aqueous solution was extracted with ethyl acetate. The aqueous acidic solution was basified with sodium hydrogen carbonate and extracted with ether and then with ethyl acetate. The combined organic phases were washed with water, dried, and evaporated. Picric acid (0.5 g) was added to a solution of the residue in methanol to give 1-(*4,5-dimethoxy-2-nitrobenzyl*)-*1,2,3,4-tetrahydro*-*6,7,8-trimethoxy-2-methylisoquinoline picrate* (1.37 g), m.p. 161—162 °C (Found: C, 50.65; H, 4.9. $C_{28}H_{31}N_5O_{14}$ requires C, 50.8; H, 4.7%).

The *free base*, recovered as usual by the alumina method, crystallised from light petroleum (b.p. 60—80 °C), m.p. 112—113 °C; δ_H 7.40, 6.40, and 6.31 (each 1 H, s, ArH), 3.89, 3.86, 3.79, 3.77, and 3.74 (each 3 H, s, OMe), 3.34 (2 H, d, *J* 6 Hz, $ArCH_2$), 2.35 (3 H, s, NMe), plus unresolved signals; δ_C (C_6D_6) 115.0, 108.4, 107.8, 60.9 (Ar*C*HN), 60.5, 55.7, 55.5, 47.2, 42.9, 36.8, and 26.2 (Found: M^+, 432.1898. $C_{22}H_{28}N_2O_7$ requires *M*, 432.1896).

A solution of the foregoing nitrotetrahydroisoquinoline (1.2 g) in methanol (250 ml) was shaken with hydrogen and platinum oxide (85 mg) at room temperature and pressure; quantitative uptake was complete in 15 min. The filtered solution was treated with 1.1 mol equiv. of picric acid to give the dipicrate salt, m.p. 161—162 °C, identical with the sample prepared above by reduction with zinc. The free base, recovered as usual, showed δ_H 6.53, 6.34, 6.25 (each 1 H, s, ArH), 4.30br (2 H, ArCH_2), 3.94, 3.85, 3.80, 3.78, and 3.72 (each 3 H, s, OMe), 2.78 (2 H, m, CH_2CH_2), and 2.38 (3 H, s, NMe); δ_C (C_6D_6) 152.7, 141.5, 119.5, 119.1, 108.0, 102.7, 61.9 (Ar*C*HN), 60.6, 57.7, 56.1, 55.7, 46.8, 42.9, 39.1, and 24.7.

(b) *The Pschorr ring closure.* The aminotetrahydroisoquinoline (12) (650 mg) in 1M-sulphuric acid (100 ml) at −3 °C was treated with stirring with sodium nitrite (120 mg) and stirring was continued for 30 min. Acetone (400 ml) and 1M-sulphuric acid (400 ml), both at −3 °C, were added followed by copper powder [24] (10 g) and the mixture was stirred at −3 °C for 2 h before filtering off the solids and concentrating the filtrate to 400 ml. This solution was basified with ammonia (sufficient to redissolve precipitated copper hydroxide) and then extracted with methylene chloride to yield the crude Pschorr products.

These were fractionated on alumina (Grade I) using first benzene and later 5% steps from 5% chloroform–benzene to 15% chloroform–benzene. Protostephanone (3) was detected by t.l.c. control (intense red colour with concentrated sulphuric acid) and the product from the appropriate combined fractions was further chromatographed on silica gel (0.05—0.2 mm; 30 g). The solvents, in sequence, were benzene, benzene +1% ethyl acetate +0.5% diethylamine, benzene +1.5% ethyl acetate +0.5% diethylamine, benzene +2% ethyl acetate +1.5% diethylamine. Combination of the appropriate fractions gave *protostephanone* (3) in yields of 18—25% (here 110 mg, 18%), m.p. 188—189 °C (from ethanol) (Found: C, 67.7; H, 6.7; N, 3.6. $C_{21}H_{25}NO_5$ requires C, 67.9; H, 6.8; N, 3.8%); $\nu_{max.}$ 1 660 and 1 625 cm^{-1}; δ 6.85, 6.67, and 6.40 (each 1 H, s, ArH and olefinic), 4.40br (1 H, CHN), 3.90 and 3.84 (each 6 H, s, 4 × OMe), and 2.48 (3 H, s, NMe); *m/e* 371 (M^+).

Protostephanone picrate crystallised from methanol, m.p. 228—230 °C.

Reduction of Protostephanone (3) *and Rearrangement of the Dienols* (17) *and* (18) *to give Dienone–B* (19).—A solution of protostephanone (110 mg) in ethanol (10 ml) at 0 °C was reduced with an excess of sodium borohydride. After 1 h, water (20 ml) was added and the ethanol was evaporated off. Methylene chloride then extracted the two dienols (95—100% yield) which were separable by preparative t.l.c. on silica using benzene–ethyl acetate–diethylamine (7 : 2 : 1 v/v/v) as eluant. They were detected by spraying with concentrated sulphuric acid which instantly gave a bright orange colour, R_F (dienol-I) 0.35, R_F (dienol-II) 0.15; δ (dienol-I) 6.72, 6.55 (each 1 H, s, ArH), 5.27 (1 H, s, olefinic), 4.85 (1 H, s, CHOH), 3.81, 3.79, 3.70, and 3.68 (each 3 H, s, OMe), and 2.44 (3 H, s, NMe). The corresponding values for dienol-II were 6.77, 6.58, 5.30, 4.72, 3.84, 3.82, 3.71, 3.68, and 2.46; *m/e* for dienols 373 (M^+, 33%), 355 (M^+ − 18, 100), 340 (27), 286 (97), and 168 (98) (Found: *m/e* 286.143 5. $C_{17}H_{20}NO_3$ requires M − 87, 286.144 3).

A solution of the mixed dienols (40 mg) in methanol (25 ml) at 20 °C was mixed with 6M-sulphuric acid (10 ml) and kept at 20 °C for 40 min. The mixture was then basified with 2M-sodium hydroxide, diluted with water (100 ml) and extracted with methylene chloride (3 × 50 ml) to give dienone-B as a gum (33 mg, 89%) which was homogeneous by t.l.c.; $\lambda_{max.}$ 352 nm (ε 17 500); $\nu_{max.}$ 1 647, 1 611, and 1 591 cm^{-1}; δ 7.00 and 6.78 (each 1 H, s, ArH), 6.28 (1 H, d, *J* 2 Hz, H_C), 5.61 (1 H, d, *J* 2 Hz, H_D), 3.94 (6 H, s, 2 × OMe), 3.86 (3 H, s, OMe), and 2.54 (3 H, s, NMe); *m/e* 341 (100%), 326, 298, and 284 (doubly charged ion at 170.5). *Dienone-B picrate* crystallised from methanol, m.p. 230—235 °C (decomp.) (Found: C, 55.0; H, 4.6; N, 9.8. $C_{26}H_{26}N_4O_{11}$ requires C, 54.7; H, 4.6; N, 9.8%).

To prepare monodeuteriodienone-B the above experiment was repeated with protostephanone (100 mg) except that CH_3OD (5 ml) was the solvent and sodium borodeuteride was the reducing agent. The mixed deuterio-dienols were rearranged as previously to yield monodeuteriodienone-B (38 mg); δ 7.00 and 6.78 (each 1 H, s, ArH), 6.28 (1 H, s, olefinic H_C), 3.94 (6 H, s, OMe), 3.86 (3 H, s, OMe), and 3.54 (3 H, s, NMe); *m/e* 342 (100%), 327, 299, 285, and 268.

Reduction of Dienone-B (19).—Dienone-B (40 mg) in methanol (15 ml) at 0 °C was treated over 10 min with sodium borohydride (150 mg) and the mixture was stirred for a further 30 min. After the solvent had been evaporated off, the residue was mixed with water (20 ml) and extracted with chloroform (3 × 15 ml). The product was purified by preparative t.l.c. on silica using 1 : 4 (v/v) methanol–chloroform and was ' one-spot ' material (22 mg); however, no attempt was made to separate any stereoisomers; $\lambda_{max.}$ 297, 258, and 223 nm; $\nu_{max.}$ 1 603 and 1 520 cm^{-1}; δ 6.86 and 6.69 (each 1 H, s, ArH), 5.60 (1 H, d, olefinic), 3.86 (6 H, s, OMe), 3.42 (3 H, s, CHO*Me*), and 2.39 (3 H, s, NMe); *m/e* 245 (M^+, 60%), 330, 312, 286, and 256 (100%).

This product (22 mg) in AnalaR acetone (10 ml) was oxidised with Jones reagent (8 drops) and the reaction was quenched by addition of ammonia solution (*d* 0.88; 25 drops) as in a related case.[25] The resultant enone (8 mg) was extracted into chloroform and purified by preparative t.l.c. with 1 : 4 (v/v) methanol–chloroform as eluant and showed $\lambda_{max.}$ 345, 295, and 239 nm; $\nu_{max.}$ 1 658 and 1 592 cm^{-1}; δ 6.70 and 6.56 (each 1 H, s, ArH), 5.92 (1 H, s, olefinic), 3.90 and 3.94 (each 3 H, s, OMe), 3.46 (3 H, s, OMe), and 2.52 (3 H, s, NMe); *m/e* 343 ($C_{20}H_{25}NO_4$).

Conversion of Dienone-B (19) *into Protostephanine* (4).—Magnesium iodide was prepared by treating magnesium turnings (170 mg) with iodine (490 mg) in dry ether (15 ml). This product was added over 15 min to a stirred solution of dienone-B (250 mg) in dry benzene (10 ml). A precipitate formed which was stirred and heated under reflux for 1.5 h, and the cooled solution (20 °C) was treated during 10 min with lithium aluminium hydride (300 mg) in ether (25 ml). The mixture was then heated under reflux for 2 h and to the cooled stirred mixture was added 2M-hydrochloric acid. The separated aqueous layer was mixed with 10% (w/v) aqueous ammonium chloride, basified to pH 9 with ammonia and extracted with chloroform (4 × 25 ml). After being washed with water, the chloroform solution was dried and evaporated to leave a phenolic residue which was fractionated by preparative t.l.c. on silica with 7 : 2 : 1 (v/v/v) benzene–ethyl acetate–diethylamine. This gave a monophenol (20), *m/e* 343 (100%), 328, 313, 298, and 285 (98%), and a diphenol (21), *m/e* 329 (100%), 314, 299, 285, and 271. Methylation of either phenol with an excess of diazomethane in ether gave a single spot on t.l.c. inseparable from that of protostephanine (4). Accordingly, the mixture of phenols (20) and (21) in methanol (5 ml) was treated with ethereal diazomethane [from *N*-nitrosomethylurea (1 g)]. After 36 h at 20 °C, the solvent was evaporated off and the residue in 2M-hydrochloric acid (5 ml) was extracted with ether (3 × 10 ml). The aqueous solution was basified with sodium hydroxide and extracted (5 × 10 ml) with chloroform to yield crude protestephanine which was purified by preparative t.l.c. with benzene–ethyl acetate–diethylamine as above. Final purification was *via* the picrate (23 mg), m.p. and mixed m.p. 207 °C, identical (i.r., n.m.r., and mass spectrometry) with natural protestephanine picrate.

Conversion of the Dienols (17) *and* (18) *into Protostephanine* (4).—A solution of the mixed dienols (15 mg) in trifluoroacetic acid (3 ml) was kept for 16 h at 20 °C and then evaporated, finally in high vacuum to remove the excess of acid. The residue, whilst being stirred in ethanol (10 ml), was treated portionwise with an excess of sodium borohydride and, after 1 h, the solution was acidified with 2M-hydrochloric acid and the methanol was evaporated off. After the aqueous solution had been basified with sodium hydrogen carbonate, it was extracted with methylene chloride (3 × 5 ml), and the product, by preparative t.l.c. on silica [7 : 2 : 1 (v/v/v) benzene–ethyl acetate–diethylamine], gave protostephanine (8 mg), identical (u.v., i.r., n.m.r., and mass spectra and chromatography) with the natural alkaloid (Found: *m/e* 357.1954. $C_{21}H_{27}NO_4$ requires *M*, 357.1939); $\lambda_{max.}$ 283 and 277 nm; δ 6.77 and 6.66 (each 1 H, s, ArH), 6.48 and 6.33 (each 1 H, s, ArH), 3.90, 3.81, and 3.78 (3 H, 6 H and 3 H, s, 4 × OMe), and 2.47 (3 H, s, NMe). The mass spectra of the synthetic and natural samples were identical, within experimental error, and both showed strong peaks at *m/e* 357, 342, 326, 301, 300, and 299.

5-Benzyloxy-4-methoxy-2-nitrophenylacetic Acid (*with Dr. E. Ruveda*).—A stirred solution of 3-benzyloxy-4-methoxyphenyl acetic acid (11.5 g) in acetic acid (100 ml) was maintained at <30 °C whilst concentrated nitric acid (18 ml) was added in 5 portions. After the final mixture had been kept at 20 °C for 15 min, it was poured into water (300 ml) and the precipitated solid was dried and recrystallised from ethanol to yield the *nitro-acid* (11.1 g, 83%), m.p. 176—178 °C (Found: C, 60.15; H, 5.0; N, 4.3. $C_{16}H_{15}NO_6$ requires C, 60.55; H, 4.8; N, 4.4%), M^+ 317.

5-Benzyloxy-4-methoxy-2-nitro-N-(*3,4,5-trimethoxyphenethyl*)-*phenylacetamide*.—A stirred suspension of the foregoing nitro-acid (6 g) in dry benzene (20 ml) was treated with oxalyl chloride (4 g) and then with dimethylformamide (few drops). After having been stirred for 30 min at 20 °C, the solution was evaporated and the acid chloride, without purification, was dissolved in chloroform (50 ml) and added to a vigorously stirred mixture of mescaline (4.9 g), ether (240 ml), and 10% (w/v) aqueous potassium hydroxide (200 ml). A solid precipitated and after 15 min ether (500 ml) was added, and the solid was collected, dried, and crystallised from ethyl acetate to give the *phenylacetamide* (8.2 g, 79%), m.p. 172—173 °C (Found: C, 63.5; H, 5.9; N, 5.6. $C_{27}H_{30}N_2O_8$ requires C, 63.5, H, 5.9; N, 5.5%).

1-(5-Benzyloxy-4-methoxy-2-nitrobenzyl)-3,4-dihydro-6,7,8-trimethoxyisoquinoline.—The foregoing amide (13 g) was dissolved in chloroform (200 ml) which had previously been passed over alumina (activity I). Freshly distilled phosphorus oxychloride (15 ml) was added, the solution was heated under reflux for 1.25 h and then evaporated. Methanol (50 ml) was added over 15 min to the residue which was cooled to 0 °C and the resultant solution was basified with ammonia solution (*d*, 0.88) (cooling). After addition of water (300 ml), the product was extracted with chloroform (3 × 150 ml); it crystallised from methanol as needles (9.1 g, 73%), m.p. 136—137 °C. The product was characterised as the *dihydroisoquinoline picrate*, m.p. 208—210 °C (from methanol) (Found: C, 55.1; H, 4.4; N, 9.7. $C_{33}H_{31}N_5O_{14}$ requires C, 54.9; H, 4.3; N, 9.7%).

1-(2-Amino-5-benzyloxy-4-methoxybenzyl)-1,2,3,4-tetrahydro-6,7,8-trimethoxy-2-methylisoquinoline (13).—The foregoing dihydroisoquinoline (7.5 g) in methyl iodide (150 ml) was heated under reflux for 3 h in the dark. Evaporation of the methyl iodide left the methiodide salt as a resin (9.7 g) of which part (8 g) was suspended in powdered form in concentrated hydrochloric acid (250 ml) and water (120 ml). Zinc dust was gradually added to this stirred suspension until, after 1 h, a very pale green solution had been formed. This was basified with ammonia solution (*d*, 0.88) (cooling) and then extracted with ether (3 × 300 ml) to yield a gum (3.1 g, 52%) which was homogeneous by t.l.c. This gum in methanol with picrolonic acid gave the *aminotetrahydroisoquinoline dipicrolonate*, m.p. 152—155 °C (from methanol) (Found: C, 56.7; H, 5.1. $C_{48}H_{50}N_{10}O_{15}\cdot CH_3OH$ requires C, 56.6; H, 5.2%).

Pschorr Ring Closure to Demethylprotostephanone (2).—The foregoing base (1 g), recovered from the dipicrolonate, in ethanol (25 ml) and concentrated hydrochloric acid (6 drops) was shaken with 10% palladium–charcoal (0.3 g) and hydrogen at room temperature and pressure. Uptake (1 mol) was complete in 8 h and the filtered solution was then evaporated to give the phenolic base (14) as its dihydrochloride.

The base (14) (1 g), without further purification, was dissolved in 1M-sulphuric acid (20 ml) and stirred at −3 °C whilst sodium nitrite (1 g) in water (3 ml) was added over 15 min. After 4 h at −3 °C, the solution was diluted with acetone (20 ml), previously cooled to −10 °C and then copper powder [24] (1 g) was added. The mixture was stirred at 0 °C for 2 h, basified with saturated aqueous sodium hydrogen carbonate, and filtered. Acetone was evaporated at 20 °C from the filtrate and the resultant suspension was extracted with chloroform (3 × 50 ml) to give a gum (0.55 g). This was fractionated by (*a*) chromatography on alumina (activity I) with 1 : 4 (v/v) chloroform–benzene, (*b*) preparative t.l.c. on silica with 5% (v/v) methanol–chloroform, and (*c*) re-chromatography of the low-running major band from (*b*) on alumina (activity I) in chloroform. This gave the phenolic dienone (2) as a resin (26 mg, 3.2%), identical (full chromatographic and spectroscopic comparison) with the same product described above.

Part of the phenolic dienone (10 mg) in methanol (2 ml) was treated with a large excess of ethereal diazomethane at 0 °C. After 3 days, the solution was evaporated and the residue was purified chromatographically as earlier. It crystallised from ethanol to yield protostephanone (3), m.p. and mixed m.p. 188—189 °C, identical (i.r. and chromatography) with the earlier sample.

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