Biosynthesis. Part 25.[1] Proof that Hasubanonine and Protostephanine are Biosynthesised from the 1-Benzylisoquinoline System

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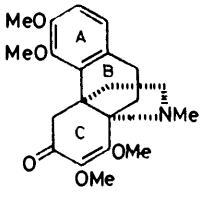
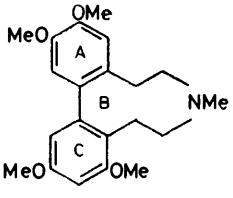
The syntheses are described of a set of sixteen 1-benzylisoquinolines and four bisphenethylamines built from 2-(3,4-dihydroxy-5-methoxyphenyl)ethylamine (3), a known biosynthetic precursor to the alkaloids hasubanonine (1) and protostephanine (2) in *Stephania japonica* plants, and an acid component with a systematically varied oxygenation pattern. Administration of this set of compounds in labelled form to whole plants has resulted in incorporation of six of the 1-benzylisoquinolines into the alkaloids. The earliest of these precursors is a triphenolic tetraoxygenated isoquinoline, and subsequent oxygenation and *O*-methylation steps are indicated; the timing of *N*-methylation is not critical. These results necessitate subtle modification of earlier biogenetic speculations and rejection of some possibilities. Hasubanonine (1) and protostephanine (2) are revealed as 'disguised' members of the 1-benzylisoquinoline group, unique in requiring *two* phenolic oxygens in one of the rings undergoing oxidative coupling during their biosynthesis.

THE unusual structural features present in the alkaloids hasubanonine (1) and protostephanine (2), isolated from *Stephania japonica*, have stimulated considerable interest in the biosynthesis of these natural products.[2] Our initial precursor incorporation studies, detailed in the preceding paper,[1] uncovered the origin of the carbon skeletons of the two alkaloids; rings A and C in both cases arise from the shikimic acid pathway *via* the amino-acid tyrosine. More specifically, ring C and its asso-



(1)



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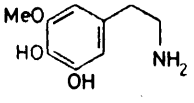
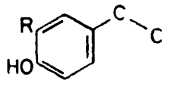
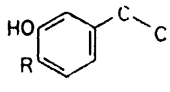


(3)



(4)
a; R = H
b; R = OMe



(5)
a; R = OH
b; R = OMe

ciated ethanamine unit were found to originate from the diphenolic trioxygenated phenethylamine (3). Further *O*-methylation does not occur before formation of more advanced precursors containing both aromatic rings.

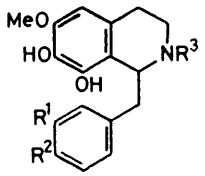
With these requirements in mind, a set of advanced precursors of the 1-benzylisoquinoline or bisphenethylamine types was envisaged by combination of the amine (3) with a C_6–C_2 unit (4) or (5). The syntheses of these potential precursors, and the results of incorporation experiments with them, are reported herein.[3]

RESULTS AND DISCUSSION

The finding that *O*-methylation of (3) does not occur in the biosynthesis of hasubanonine (1) and protostephanine (2), and the lack of any information about possible 'masking' of the extra phenolic oxygen derived from (3) during any later oxidative coupling steps,[4] led us to incorporate the structure of amine (3) intact into the set of compounds planned for new biosynthetic experiments. In other alkaloid groups that have been shown to incorporate phenethylamines into one part of a 1-benzylisoquinoline-derived system, it has been found that the amine forms the isoquinoline nucleus.[5] It was reasonable to assume that, if they are being formed by modification of a benzylisoquinoline, the same will apply to alkaloids (1) and (2).

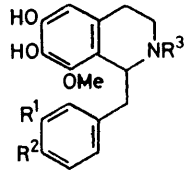
Consideration of possible 1-benzylisoquinoline precursors for hasubanonine (1) and protostephanine (2) that include the intact C_6C_2N phenethylamine moiety of (3) in this fashion, *i.e.* in a 6,7,8-trioxygenated isoquinoline, shows the two orientations possible, with the methoxy-group of (3) at the 6- or the 8-position; our efforts had to include both possibilities. The incorporation of tyrosine into ring A of alkaloids (1) and (2) [1] defined the minimum requirement of the 'benzyl' ring of possible 1-benzylisoquinoline precursors as a 4-hydroxy-substituent [*cf.* (4a)]. The various biogenetic speculations,[1] and the structures of the alkaloids themselves, pointed to a 3,4-dioxygenation pattern appearing in this ring at some stage, either 3-hydroxy-4-methoxy [*cf.* (5b)] or 4-hydroxy-3-methoxy [*cf.* 4b)]. Both of these arrangements could arise from a 3,4-dihydroxy-intermediate [*cf.* (5a)], and it was decided to include 1-benzylisoquinolines with all four arrangements in our incorporation studies. The timing of *N*-methylation was the remaining uncertainty; since it seemed that both NH and NMe compounds would be available from the same synthetic route, it was decided to examine both possibilities. These considerations generated a set of sixteen 1-benzylisoquinolines (6a—h) and (7a—h) to be synthesised in labelled form for incorporation experiments with *S. japonica*. It was also decided to examine the set of four secondary amines (8a—d) with similar substituent variation to explore further the biosynthetic proposals involving bisphenethylamines.[6]

The task of synthesising all twenty compounds in labelled form was completed economically as shown in Scheme 1. Benzyl ethers were chosen as the protecting groups for phenolic functions throughout the synthesis. It was anticipated that the two patterns of isoquinoline ring substitution [7,8-dihydroxy-6-methoxy (6) and 6,7-dihydroxy-8-methoxy (7)] would be available in protected form as a mixture from Bischler–Napieralski ring-closure of the amides (11a—d) which differ only in



(6)

a; R^1 = H, R^2 = OH, R^3 = H
b; R^1 = R^2 = OH, R^3 = H
c; R^1 = OH, R^2 = OMe, R^3 = H
d; R^1 = OMe, R^2 = OH, R^3 = H



(7)

e; R^1 = H, R^2 = OH, R^3 = Me
f; R^1 = R^2 = OH, R^3 = Me
g; R^1 = OH, R^2 = OMe, R^3 = Me
h; R^1 = OMe, R^2 = OH, R^3 = Me

(8)

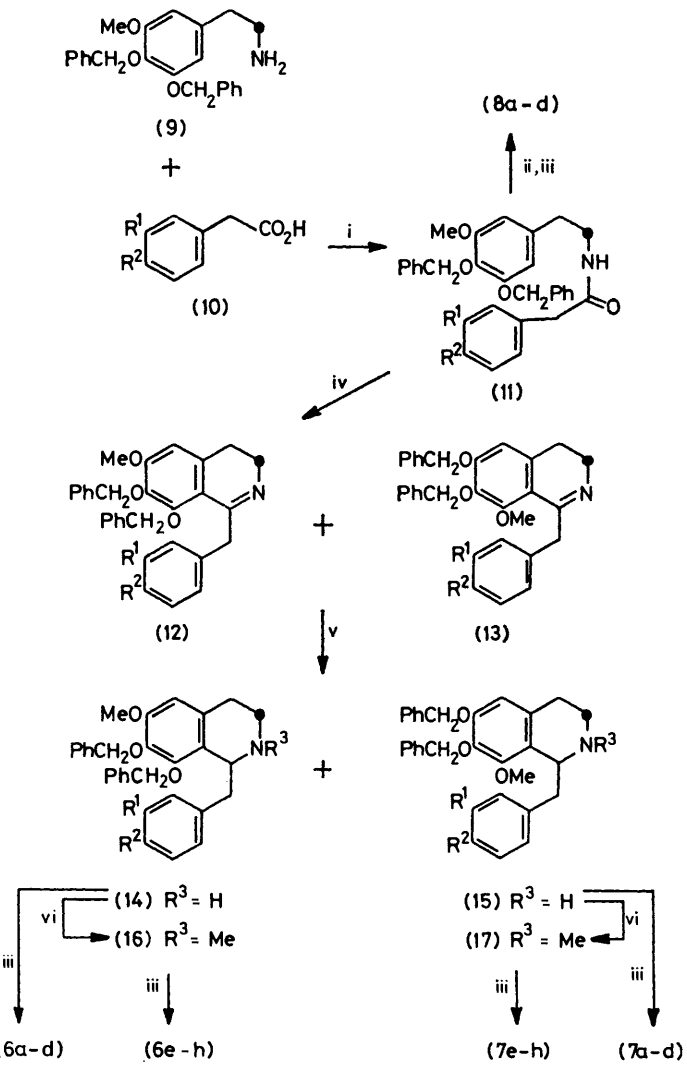
a; R^1 = H, R^2 = OH
b; R^1 = R^2 = OH
c; R^1 = OH, R^2 = OMe
d; R^1 = OMe, R^2 = OH

their acid portions. Further, reduction of the same four amides should provide the required bisphenethylamines (8a—d). The amine portion of the amides (11) is common to all twenty compounds. It was therefore decided to introduce radiochemical labelling into these molecules using the 2-phenyl[1-^{14}C]ethylamine (9),* prepared with only minor modification by the published method.[1] The amine (9) was combined separately with the acid chloride of each of four phenylacetic acids (10a—d) to produce the four required amides (11a—d). Reduction of these with diborane and hydrogenolytic debenzylation led directly to the bisphenethylamines (8a—d) carrying a ^{14}C-label α to nitrogen.

Cyclisation of the amides (11) in a Bischler–Napieralski fashion with phosphorus oxychloride in acetonitrile gave, in each case, the expected mixture of two 3,4-dihydroisoquinoline isomers (12) and (13) as their hydrochlorides in good yield and *ca.* 1 : 1 ratio. From the cyclisation of (11a), one product (13a) was isolated as a crystalline solid by treatment of the crude products with ethanol and ether, leaving the other isomer (12a) as a gum from the mother-liquors. The separated isomers were converted by reduction with sodium borohydride into the corresponding 1,2,3,4-tetrahydroisoquinolines (14a) and (15a) which were characterised as

* For a discussion of the reasons for choosing ^{14}C skeletal labelling, see preceding paper.[1]

crystalline hydrochlorides. The product mixture from cyclisation of the amide (11c) could be separated in similar fashion if the crude dihydroisoquinoline hydrochlorides were first converted into the oxalate salts *via*



SCHEME 1 *Reagents:* i, (9) + acid chloride of (10); ii, B_2H_6; iii, H_2–Pd; iv, $POCl_3$; v, $NaBH_4$; vi, H_2CO–$NaBH_4$

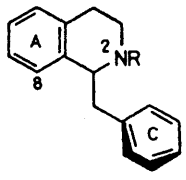
the free bases. A solid was isolated consisting of a 4 : 1 mixture of the imine oxalates; recrystallisation afforded the pure oxalate salt of the 8-methoxy-isomer (13c), and the 6-methoxy-isomer (12c) was recovered from the mother-liquors. Again the 1,2,3,4-tetrahydroisoquinolines (14c) and (15c), obtained by reduction of the separated isomers, were used for characterisation (as the hydrochlorides) and structural assignment. In the

case of the remaining two amides (11b) and (11d), the crude cyclisation mixture proved inseparable by fractional crystallisation of various salts or by chromatography. The mixtures were reduced as above to the tetrahydroisoquinolines, at which point trituration with ether gave a solid. Conversion of this solid in each case into the hydrochloride afforded one pure isomer (15b) and (15d); the other (14b) and (14d) was obtained as the pure hydrochloride salt by treatment of the ethereal mother-liquors with ethereal hydrogen chloride.

Synthesis of the hydroxytetrahydroisoquinolines (6a—d) and (7a—d) was completed by hydrogenolysis of the benzyloxy-compounds (14a—d) and (15a—d) respectively. Alternatively these benzyloxy-compounds were methylated (formaldehyde–sodium borohydride) to give (16a—d) and (17a—d) before debenzylation to afford the corresponding set of *N*-methyl derivatives (6e—h) and (7e—h), thus completing the set of sixteen 1-benzyl-[3-^{14}C]isoquinolines for biosynthetic studies.

The basis of the structural assignments for the isomeric pairs (14a—d) and (15a—d), and of the compounds prepared from them, was ^{1}H n.m.r. spectroscopy of their hydrochloride salts. 1-Benzyltetrahydroisoquinolines can exist in a range of conformations, two extremes of which are (18) and (19), with the nature of the sub-

(18)



(19)

stituents at C-8 and nitrogen determining which arrangement is thermodynamically favoured.[7] For each isomeric pair, the ring-A methoxy-substituent in one isomer showed a methyl resonance in the range δ 3.78—3.89, as expected for a simple methoxylated isoquinoline,[7,8] whereas in the other isomer, the ring-A methoxy-signal was observed at lower field (δ 3.92—3.98). In the former case, the methylene group of one of the benzyloxy-substituents was seen as an AB quartet, whereas in the latter case all the benzyloxy-methylene resonances were singlets. This behaviour is in accord with deshielding of the ring-A methoxy-group in the 8-methoxy-series (15) caused by the magnetic anisotropy of the 'benzyl' ring C in a conformation where the C-8 substituent lies alongside ring C, *i.e.* near to (18). Such a conformation is reasonable, despite the presence of C-8 substitution, because of the quaternary nature of the isoquinoline nitrogen.[8] Effects on the C-6 substituent would be expected to be negligible, as is indeed observed in the other, 6-methoxy, series (14); in any case, the 6-methoxy-isomers would be expected to exist in a conformation nearer to (19) as the 8-substituent is now the bulky benzyloxy-group. The methylene group of this C-8 benzyloxy-substituent in isomers (14) can now be assigned to the observed AB quartet from this series, the protons being non-equivalent by virtue of proximity to the chiral centre at C-1.

Support for this structural assignment was obtained from ^{1}H n.m.r. spectra of the *N*-methyltetrahydroisoquinolines (16) and (17). Conformation (19) is favoured for this class of compounds when C-8 is substituted, and the *N*-methyl resonance is expected to be shifted upfield from its normal position (δ *ca.* 2.5) as the protons lie over the 'benzyl' ring C, the extent of the shift increasing with increased size of the C-8 substituent.[1] This effect was indeed observed in (16) and (17); all the *N*-methyl signals were recorded at chemical-shift values of less than δ 2.5, the signals for the 6-methoxy-isomers (16) in each case lying upfield of those from the corresponding 8-methoxy-isomer (17). The n.m.r. results are thus self-consistent and on the basis of the quoted literature precedents, the structural conclusions should be sound. A derivative of one of the isoquinolines (6a—h) and (7a—h) suitable for *X*-ray analysis is being sought to provide rigour from a different method.

The twenty compounds (6a—h), (7a—h), and (8a—d) were administered separately as hydrochlorides to *S. japonica* whole plants by the published method;[1] [3-^{14}C]tyrosine was fed in parallel as a control to check for alkaloid synthesis. After a growth period of 10 days, the plants were harvested and the alkaloids hasubanonine (1) and protostephanine (2) isolated.

None of the bisphenethylamines (8a—d) was incorporated into alkaloids (1) and (2).* In contrast, six of the 1-benzylisoquinolines did act as precursors of (1) and (2), and were well incorporated into the alkaloids (Table); all were 6-methoxy-isomers (6a—c and e—g). The remaining ten compounds (6d and h) and (7a—h) were not significantly incorporated.*,†

The hasubanonine (1) isolated from feedings of the three isoquinolines (6a, e, and g) was degraded according to a scheme developed earlier [2b] in which the ethanamine bridge of (1) is lost, and the results show (Table) that the radioactivity was located specifically in this bridge, *i.e.* the ring-C ethanamine unit. Degradation of protostephanine (2) [2b] from the feeding of isoquinoline (6f) to isolate the ring-A ethanamine carbons also demonstrated (Table) a specific incorporation of radioactivity into the ring-C ethanamine side-chain. These results are consistent with those reported earlier from incor-

* Incorporations were less than 1% of the values found for (2*RS*)-[3-^{14}C]tyrosine in the parallel feeding [3% for (6d)]. Purification of alkaloids (1) and (2) was discontinued at this point, although their specific activities were still decreasing and had not reached a constant value,

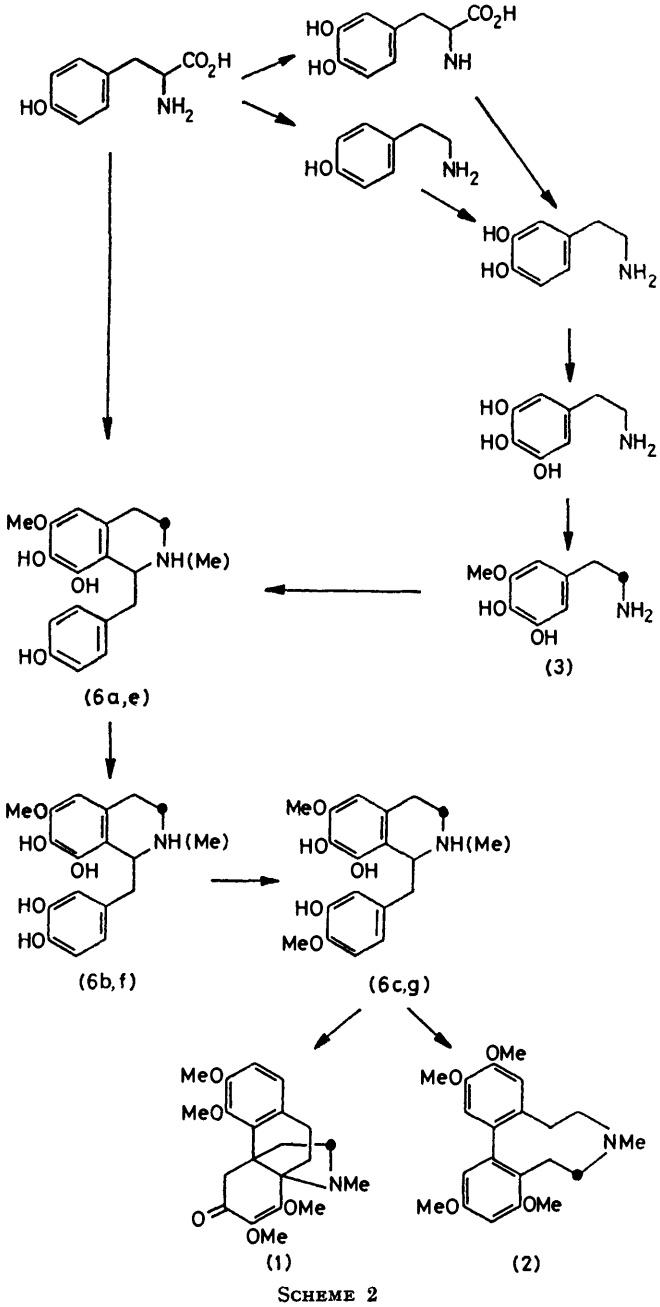
† Two of the 8-methoxyisoquinolines, (7a) and its *N*-methyl derivative (7e), had apparent incorporations of greater than 1% of the values for tyrosine in the parallel feeding, although much lower than their 6-methoxy-counterparts (6a and e). It is possible that the biosynthetic enzymes have some lack of specificity, or that some degradation–reincorporation is occurring; more probable, however, is that these values represent contamination by the 6-methoxy-isomers owing to incomplete separation in the radiochemical synthesis of the isoquinoline mixture (12a) and (13a) from which all of (6a and e) and (7a and e) were prepared. N.m.r. analysis showed that, in this run, the 8-methoxy-tetrahydroisoquinoline (15a) contained *ca.* 5% of the 6-methoxy-isomer (14a).

Tracer experiments on *S. japonica* plants

Precursor	% Incorpn. into (1)	% Incorpn. into (2)	% of radioactivity in ring-c ethanamine chain	
			Base (1)	Base (2)
(2*RS*)-[3-^{14}C]Tyrosine	1.2	0.1	57 ± 1	*a*
[3-^{14}C]Isoquinoline (6a)	0.33	0.020	100 ± 1	*a*
[3-^{14}C]Isoquinoline (6b)	0.33	0.022	*a*	*a*
[3-^{14}C]Isoquinoline (6c)	0.24	0.017	*a*	*a*
[3-^{14}C]Isoquinoline (6e)	0.43	0.022	100 ± 1	*a*
[3-^{14}C]Isoquinoline (6f)	0.26	0.023	*a*	100 ± 1
[3-^{14}C]Isoquinoline (6g) [b]	0.10	0.005	98 ± 1	*a*

[a] Not determined. [b] Isolation difficulties caused losses and necessitated considerable dilution with unlabelled material for this experiment; incorporation values are certainly minimal ones.

poration experiments with phenethylamines [1] and with the expected construction of the isoquinoline nucleus from C_6C_2N phenethylamine precursors.



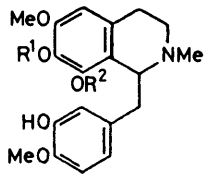
SCHEME 2

These positive incorporations of a number of 1-benzylisoquinolines and the negative results obtained with the bisphenethylamines (8) combine to rule out any biosynthetic speculation for alkaloids (1) and (2) that involves bisphenethylamine intermediates.[6] Hasubanonine (1) and protostephanine (2) have finally been revealed as ' disguised ' members of the group of 1-benzylisoquinoline-derived alkaloids, and the precursors established here are subtly modified versions of those included in earlier biosynthetic speculations and incorporation studies.[1] The results are fully consistent with the earlier investigations.

The known part of the biosynthetic pathway [1] to alkaloids (1) and (2) can now be extended as illustrated in Scheme 2. The amine (3), a known precursor,[1] combines with another tyrosine-derived unit to give the 1-benzyl-6-methoxyisoquinoline (6a or e), before a second hydroxylation takes place in the ' benzyl ' ring. It is likely that this other tyrosine-derived building block will be a carbonyl compound such as 4-hydroxyphenylpyruvic acid, and these results are in agreement with an earlier failure to incorporate 3,4-dihydroxyphenylpyruvic acid.[1] The incorporation of all the isoquinolines as both secondary amines and their *N*-methyl derivatives indicates that the timing of *N*-methylation is not critical; enzymes controlling the pathway can accept both types of amine. The *N*-methylating enzyme may also be non-specific.

The next steps are hydroxylation of the ' benzyl ' ring to give (6b or f) and methylation of the original phenolic oxygen to form (6c or g), a sequence that has precedent, *e.g.* in the biosynthesis of colchicine.[9] Incorporation of (6c or g), and the negative results with isoquinolines (6d and h), defines the substitution in the ' benzyl ' ring sufficiently to rule out biosynthetic proposals involving a 4-hydroxy-3-methoxy-pattern in this ring, the ' bis-dienone ' route.[1] Earlier failures to incorporate more-highly methylated derivatives of (6g), such as (20a and b),[1] indicate that further methylation does not take place; it is likely that (6c or g) possesses the correct oxygenation–methylation pattern for oxidative coupling. As far as we know, the cases here are unique in requiring *two* phenolic hydroxy-groups in one of the rings undergoing coupling.

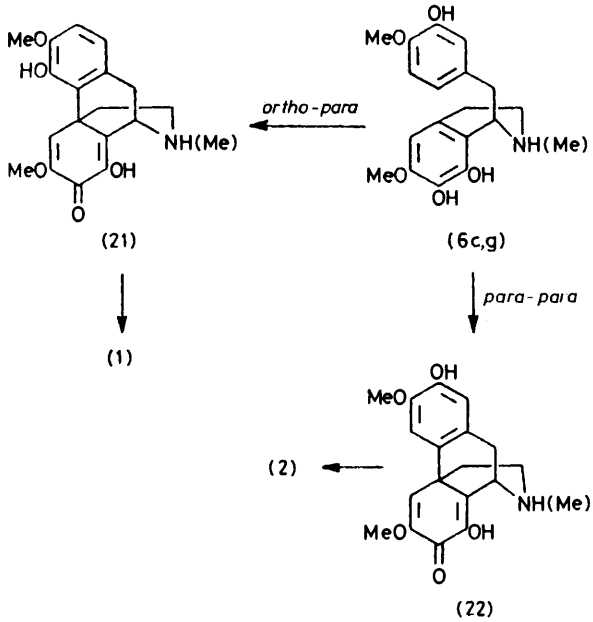
Later stages in the sequence cannot be defined exactly at present, but our results are consistent with the branching pathway shown in Scheme 3. Oxidative coupling directed by the 7-hydroxy-substituent of the 1-benzyliso-



(20)

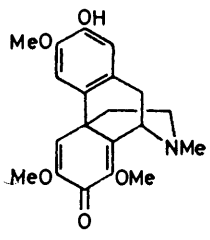
a; R^1 = Me, R^2 = H
b; R^1 = H, R^2 = Me

quinoline precursor (6c or g) leads to hasubanonine *via* the dienone (21) and protostephanine *via* the dienone (22). This is a simple modification of an earlier proposal [10] to accord with the successful incorporation of the



SCHEME 3

tri- and not di-hydroxyisoquinolines. Of relevance here is an earlier feeding experiment with the closely related dienone (23) where an apparent incorporation



(23)

of 1.38% into protostephanine (2) was observed.[11] This result must, however, be regarded as tentative at present; the isolated protostephanine (2) was extensively purified by several different methods but totally satisfactory evidence for having reached constant specific activity could not be obtained because of lack of material, nor could a check on the specificity of incorporation be made. Further progress requires synthesis of ^{14}C-labelled dienones (21) and (22) to test rigorously their efficacy as precursors of hasubanonine (1) and protostephanine (2).

EXPERIMENTAL

General directions are detailed in Part 24.[1] The synthesis of labelled compounds is described only where the procedure used differed from the radioinactive synthesis. Plant cultivation, administration of precursors, and isolation of the alkaloids are as detailed in Part 24.[1]

2-(4,5-Bisbenzyloxy-3-methoxyphenyl)[1-^{14}C]ethylamine (9) was prepared from methyl 3-methoxy-4,5-dibenzyloxybenzoate as published previously,[1] that is *via* the corresponding benzyl alcohol and benzyl chloride, with the following modifications.

4,5-Bisbenzyloxy-3-methoxyphenyl[1-^{14}C]*acetonitrile*.—The corresponding benzyl chloride (58.2 mg, 0.16 mmol) was dissolved in dimethyl sulphoxide (DMSO) (10 ml; freshly distilled) and potassium cyanide (4.5 mg, 0.07 mmol) was added. The mixture was stirred at room temperature for 20 h and K^{14}CN (1.14 mg, 0.017 mmol; 1 mCi) was then added with DMSO (4 ml). After stirring for a further 3 days, radioinactive KCN (14.7 mg, 0.23 mmol) was added, and the mixture was left to stir for 2 days and then poured into saturated brine (150 ml). The resultant emulsion was extracted with ethyl acetate (3 × 30 ml); the combined organic phases were washed with brine (3 × 100 ml), dried, and evaporated to give a residue, which was recrystallised from ethyl acetate as the pure nitrile, identical to radioinactive samples having m.p. 72—75 °C (lit.,[12] 72.5—75 °C), $\nu_{max.}$ (Nujol) 2 250, 1 595, and 1 500 cm^{-1}; δ(60 MHz) 3.60 (2 H, s, $ArCH_2CN$), 3.79 (3 H, s, OCH_3), 4.99 and 5.04 (each 2 H, s, *O*-benzyl-CH_2), 6.33br (2 H, s, ArH), and 7.37 (10 H, s, ArH); *m/e* 359 (M^+).

2-(*4,5-Bisbenzyloxy-3-methoxyphenyl)ethylamine*.—The nitrile (25 g, 0.069 mol, prepared as above) in dry tetrahydrofuran (THF) (300 ml) was treated with diborane in THF (1M; 75 ml) and the solution was heated under reflux for 7 h. The cooled mixture was then cautiously acidified with dilute hydrochloric acid, evaporated to low volume, diluted with water (150 ml), and basified with sodium carbonate. The mixture was extracted with dichloromethane (1 × 250, 2 × 75 ml) and the combined organic extracts were washed with water, dried, and evaporated to afford the crude amine as a gum (25.0 g); this was converted in ethyl acetate into its oxalate salt (19.7 g; 63%), identical to a sample prepared by sodium borohydride–boron trifluoride reduction of the phenylacetonitrile.[1]

N-(*4,5-Bisbenzyloxy-3-methoxyphenethyl*)-2-(*4-benzyloxyphenyl)acetamide* (11a) (*with S. A. surgenor*).—4-Benzyloxyphenylacetic acid (1.21 g) in dry benzene (100 ml) was treated with oxalyl chloride (2 ml) and DMF (10 drops). After 30 min the mixture was evaporated to dryness, the residue taken up successively in dry benzene (2 × 20 ml) and dry dichloromethane (2 × 20 ml) and each time re-evaporated to remove the excess of oxalyl chloride. The phenylacetyl chloride remaining was dissolved in dichloromethane (80 ml) and added to a vigorously stirred two-phase system of water (100 ml) and dichloromethane (100 ml) containing sodium hydrogen carbonate (8 g) and the oxalate salt of 2-(4,5-bisbenzyloxy-3-methoxyphenyl)ethylamine (2.36 g). After 1 h, water (100 ml), and dichloromethane

(100 ml) were added, the layers were separated, and the aqueous layer was further extracted with dichloromethane (3 × 100 ml). The combined organic solutions were washed with water, dried, and evaporated to leave the *amide* (2.93 g, 98%), m.p. 114.5—115.5 °C [from ethyl acetate–light petroleum (b.p. 60—80 °C)] (Found: C, 77.35; H, 6.55; N, 2.2. $C_{38}H_{37}NO_5$ requires C, 77.6; H, 6.3; N, 2.4%), $\nu_{max.}$ (Nujol) 3 420, 1 660, 1 595, and 1 510 cm^{-1}; $\lambda_{max.}$ 234, 258, 264, 268, 276sh, and 283sh nm; δ 2.65 (2 H, t, $ArCH_2CH_2$), 3.41 (2 H, t, $ArCH_2CH_2$), 3.46 (2 H, s, $ArCH_2CO$), 3.74 (3 H, s, OCH_3), 4.99 (6 H, s, 3 × *O*-benzyl-CH_2), 5.37br (1 H, NH), 6.23—6.34 (2 H, m, ArH), 6.84 and 7.01 (each 2 H, d, *J* 9 Hz, ArH), and 7.2—7.5 (15 H, m, ArH); *m/e* 587 (M^+).

The following amides were prepared by similar methods: N-(4,5-*bisbenzyloxy*-3-*methoxyphenethyl*)-2-(3,4-*bisbenzyloxyphenyl*)*acetamide* (11b), from the same amine oxalate and 3,4-dibenzyloxyphenylacetic acid, m.p. 113—115 °C [from ethyl acetate–light petroleum (b.p. 60—80 °C)] (Found: C, 77.9; H, 6.3; N, 2.0. $C_{45}H_{43}NO_6$ requires C, 77.9; H, 6.2; N, 2.2%), $\nu_{max.}$ (Nujol) 3 340, 1 635, and 1 590 cm^{-1}; δ 2.58 (2 H, t, *J* 6 Hz, $ArCH_2CH_2$), 3.36 (2 H, s, $ArCH_2CO$), 3.72 (3 H, s, OCH_3), 4.95, 4.98, 5.04, and 5.07 (each 2 H, s, *O*-benzyl-CH_2), 5.30br (1 H, NH), 6.30 (2 H, m, ArH), 6.56 and 6.80 (each 1 H, d, *J* 10 Hz, ArH), 6.76 (1 H, s, ArH), and 7.28 (20 H, m, ArH); *m/e* 693 (M^+); N-(4,5-*bisbenzyloxy*-3-*methoxyphenethyl*)-2-(3-*benzyloxy*-4-*methoxyphenyl*)*acetamide* (11c), from the same amine oxalate and 3-benzyloxy-4-methoxyphenylacetic acid, m.p. 129—130 °C [from ethyl acetate–light petroleum (b.p. 30—40 °C)] (Found: C, 75.4; H, 6.55; N, 2.15. $C_{39}H_{39}NO_6$ requires C, 75.6; H, 6.4; N, 2.25%), $\nu_{max.}$ (Nujol) 3 299 and 1 661 cm^{-1}; $\lambda_{max.}$ 265, 269, 274, and 279 nm; δ 2.6 (2 H, t, $ArCH_2CH_2$), 3.38 (4 H, m, $ArCH_2CH_2$ and $ArCH_2CO$), 3.74 and 3.78 (each 3 H, s, OCH_3), 4.97, 4.99, and 5.10 (each 2 H, s, *O*-benzyl-CH_2), 5.4br (1 H, t, NH), 6.30 (2 H, s, ArH), 6.6—6.8 (3 H, m, ArH), and 7.2—7.55 (15 H, m, ArH); *m/e* 617 (M^+); and N-(4,5-*bisbenzyloxy*-3-*methoxyphenethyl*)-2-(4-*benzyloxy*-3-*methoxyphenyl*)*acetamide* (11d) (*with S. A. surgenor*) from the same amine oxalate and 4-benzyloxy-3-methoxyphenylacetic acid, had m.p. 120—122 °C [from ethyl acetate–light petroleum (b.p. 60—80 °C) or aqueous ethanol] (Found: C, 75.6; H, 6.1; N, 2.2. $C_{39}H_{39}NO_6$ requires C, 75.6; H, 6.4; N, 2.25%), $\nu_{max.}$ (Nujol) 3 300, 1 640, and 1 590 cm^{-1}; δ 2.47 (2 H, t, *J* 6 Hz, $ArCH_2CH_2$), 3.43 (2 H, s, $ArCH_2CO$), 3.76 and 3.82 (each 3 H, s, OCH_3), 4.97, 5.03, and 5.08 (each 2 H, s, *O*-benzyl-CH_2), 5.40br (1 H, NH), 6.35 (2 H, m, ArH), 6.56 and 6.78 (each 1 H, d, *J* 8 Hz, ArH), 6.75 (1 H, s, ArH), and 7.33 (15 H, m, ArH); *m/e* 617 (M^+).

Synthesis of the Bisphenethylamines (8a—d). N-(4,5-*Dihydroxy*-3-*methoxyphenethyl*)-N-(4-*hydroxyphenethyl*)-*amine* (8a).—The amide (11a) (1.21 g) in dry THF (70 ml) was stirred at 0 °C during the addition of diborane in THF (0.8M; 75 ml). The solution was heated under reflux for 2 h and then the reaction was quenched with aqueous sodium hydroxide (20% w/v; 100 ml). The mixture was filtered and the filtrate extracted with ether (3 × 50 ml). The organic extracts were dried and evaporated to leave an oil that was treated in ethyl acetate with ethereal hydrogen chloride to afford N-(4-*benzyloxyphenethyl*)-N-(4,5-*bisbenzyloxy*-3-*methoxyphenethyl*)*amine hydrochloride* (0.93 g, 77%), m.p. 159—161 °C (from ethanol–ether) (Found *m/e*, 571.275. $C_{38}H_{37}NO_4$ requires $M - H_3Cl$, 571.272), $\nu_{max.}$ (Nujol) 3 380, 1 610, and 1 500 cm^{-1}; $\lambda_{max.}$ 258, 265, 269, 277, and 283 nm; δ([$^2H_4$]methanol) 2.8—3.0 (4 H, m, 2 × $ArCH_2CH_2$), 3.1—3.3 (4 H, m, 2 × $ArCH_2CH_2N$), 3.82 (3 H, s, OCH_3), 4.94, 5.05, and 5.10 (each 2 H, s, *O*-benzyl-CH_2), 6.60 (2 H, 2 × s, ArH), 6.90 and 7.15 (each 2 H, d, *J* 8 Hz, ArH), and 7.2—7.5 (15 H, m, ArH). The tribenzyloxyamine hydrochloride (60 mg) in methanol (18 ml) containing concentrated hydrochloric acid (5 drops) was stirred under hydrogen (1 atm) with 10% palladium–charcoal (30 mg) for 3 h. The suspension was filtered and the filtrate evaporated to dryness, taken up in the minimum volume of methanol, and treated with ether until the solution became cloudy. On standing at 0 °C, the trihydroxyamine hydrochloride separated (37 mg; 97%), m.p. 195—196 °C, $\nu_{max.}$ (KBr) 3 600—3 000br, 1 610, and 1 510 cm^{-1}; $\lambda_{max.}$ 277 nm; δ([$^2H_4$]methanol) 2.74—3.30 (8 H, m, 2 × $ArCH_2CH_2N$), 3.83 (3 H, s, OCH_3), 6.40 (2 H, s, ArH), and 6.74 and 7.08 (each 2 H, d, *J* 8 Hz, ArH); *m/e* 301 ($M^+ - H_3Cl$).

N-(4,5-Dihydroxy-3-methoxyphenethyl)-*N*-(3-hydroxy-4-methoxyphenethyl)amine (8c) was prepared from amide (11c) by the same procedures used to prepare (8a), *via* reduction to N-(3-*benzyloxy*-4-*methoxyphenethyl*)-N-(4,5-*bisbenzyloxy*-3-*methoxyphenethyl*)*amine hydrochloride*, m.p. 104—105 °C (Found: C, 73.1; H, 6.7; N, 1.8; *m/e* 603.297. $C_{39}H_{41}NO_5$·HCl requires C, 73.25; H, 6.55; N, 2.1%; M − HCl, 603.298), $\nu_{max.}$ (Nujol) 3 400br, 1 590, and 1 500 cm^{-1}; $\lambda_{max.}$ 256, 265, 269, and 277 nm; δ([$^2H_6$]DMSO) 2.8—3.3 (complex aliphatics), 3.78, 3.82 (each 3 H, s, OCH_3), 4.91, 5.08, and 5.11 (each 2 H, s, *O*-benzyl-CH_2), 6.62 and 6.72 (each 1 H, d, *J* 2 Hz, ArH), 6.85—7.0 (3 H, m, ArH), and 7.2—7.5 (15 H, m, ArH). Hydrogenolysis afforded the trihydroxyamine hydrochloride, m.p. 199—200 °C, $\nu_{max.}$ (KBr) 3 600—2 700br, 1 610, and 1 530 cm^{-1}; $\lambda_{max.}$ 277 nm; δ([$^2H_4$]methanol) 2.74—3.3 (complex aliphatics), 3.84 (6 H, s, 2 × OCH_3), 6.40 (2 H, s, ArH), and 6.72 and 6.84 (3 H, 2s, ArH); *m/e* 334 and 333 (M^+ — HCl).

N-(4,5-*Dihydroxy*-3-*methoxyphenethyl*)-N-(3,4-*dihydroxyphenethyl*)*amine* (8b).—The amide (11b) was treated with diborane solution as in the preparation of (8a) above, heating under reflux for 1 h. The reaction was then quenched by cautious addition of hydrochloric acid (6M) and the mixture was evaporated to low volume. The residue was partitioned between sodium carbonate solution (50 ml) and ethyl acetate (3 × 30 ml); the organic layers were washed with water, dried, and evaporated to leave a gum that was treated in ethyl acetate with ethereal hydrogen chloride to yield N-(4,5-*bisbenzyloxy*-3-*methoxyphenethyl*)-N-(3,4-*bisbenzyloxyphenethyl*)*amine hydrochloride*, m.p. 144—145 °C (from ethanol–ether) (Found: C, 75.25; H, 6.65; N, 1.75. $C_{45}H_{45}NO_5$·HCl requires C, 75.5; H, 6.45; N, 1.95%), $\nu_{max.}$ (KBr) 3 400br, 1 590, and 1 505 cm^{-1}; δ([$^2H_4$]methanol) 2.18—3.35 (8 H, m, 2 × $ArCH_2CH_2N$), 3.82 (3 H, s, OCH_3), 4.93 and 5.11 (each 2 H, s, *O*-benzyl-CH_2), 5.07 (4 H, s, 2 × *O*-benzyl-CH_2), 6.60 (2 H, s, ArH), 6.79—7.01 (3 H, m, ArH), and 7.34 (20 H, m, ArH); *m/e* 680 and 679 (M^+ — HCl). The tetrabenzyloxyamine hydrochloride was debenzylated by hydrogenolysis as above (for 2 h) to give the tetrahydroxyamine hydrochloride, m.p. 110 °C (from ethanol–ether), $\nu_{max.}$ (KBr) 3 700—2 700br, 1 630, and 1 530 cm^{-1}; δ([$^2H_6$]DMSO) 3.0br (8 H, 2 × $ArCH_2CH_2N$), 3.50br (4 H, OH), 3.76 (3 H, s, OCH_3), 6.36 (2 H, s, ArH), and 6.48—6.76 (3 H, m, ArH); *m/e* 319 (M^+ — HCl).

N-(4,5-Dihydroxy-3-methoxyphenethyl)-*N*-(4-hydroxy-

3-methoxyphenethyl)amine (8d) was prepared from the amide (11d) by the same methods described above for preparation of (8b), *via* reduction to N-(4-*benzyloxy*-3-*methoxyphenethyl*)-N-(4,5-*bisbenzyloxy*-3-*methoxyphenethyl*)-*amine hydrochloride*, m.p. 114 °C (from ethanol–ether) (Found: C, 73.2; H, 6.75; N, 2.0. $C_{39}H_{41}NO_5$·HCl requires C, 73.2; H, 6.55; N, 2.2%), $\nu_{max.}$ (KBr) 3 400br, 1 590, and 1 510 cm^{-1}; δ([$^2H_4$]methanol) 2.80—3.30 (8 H, m, 2 × ArCH_2CH_2N), 3.82 and 3.83 (each 3 H, s, OCH_3), 4.93, 5.04, and 5.08 (each 2 H, s, *O*-benzyl-CH_2), 6.64 (2 H, m, ArH), 6.76—6.96 (3 H, m, ArH), and 7.35 (15 H, m, ArH); *m/e* 604 and 603 (M^+ — HCl). Debenzylation as usual gave the trihydroxyamine hydrochloride, m.p. 183 °C (from ethanol–ether), $\nu_{max.}$ (KBr) 3 600—2 700vbr, 1 640, 1 620, 1 605, 1 530, and 1 515 cm^{-1}; δ([$^2H_6$]DMSO) 3.0br (8 H, 2 × ArCH_2CH_2N), 3.44 (4 H, s, OH), 3.75 and 3.77 (each 3 H, s, OCH_3), 6.36 (2 H, s, ArH), and 6.75 (3 H, m, ArH); *m/e* 334 and 333 (M^+ — HCl), and 319 and 318 (M^+ —HCl — CH_3).

Synthesis of 1-*Benzyltetrahydroisoquinolines* (6a—h) *and* (7a—h). 1-(4-*Benzyloxybenzyl*)-6,7-*bisbenzyloxy*-8-*methoxy*-1,2,3,4-*tetrahydroisoquinoline Hydrochloride* (15a) *and the* 7,8-*Bisbenzyloxy*-6-*methoxy-isomer* (14a) (*with S. A. Surgenor*).—A solution of the amide (11a) (3.61 g; 6 mmol) in dry acetonitrile (160 ml; freshly distilled) was flushed with nitrogen, heated to 60 °C, when phosphorus oxychloride (5.4 ml; freshly distilled) was added, and then heated under reflux under nitrogen for 20 min. The cooled solution was evaporated to dryness, this operation being repeated from toluene to remove the excess of phosphorus oxychloride, to yield a mixture of 3,4-dihydroisoquinoline hydrochlorides. The residue in the minimum volume of hot ethanol was treated with ethereal hydrogen chloride, to give a solution of pH 2—3, followed by ether until cloudiness appeared. On standing at 0 °C, 1-(4-benzyloxybenzyl)-6,7-bisbenzyloxy-8-methoxy-3,4-dihydroisoquinoline hydrochloride (13a) separated (1.5 g, 41%), m.p. 163—164 °C (from ethanol–ether), $\nu_{max.}$ (Nujol) 2 600br, 1 610, and 1 590 cm^{-1}; $\lambda_{max.}$ 246 and 320 nm; δ 2.84 (2 H, t, *J* 7 Hz, ArCH_2-CH_2N), 3.8—3.9 (5 H, s + t, ArCH_2CH_2N, OCH_3), 4.75 (2 H, s, ArCH_2C=N), 4.91, 4.96, and 5.11 (each 2 H, s, *O*-benzyl-CH_2), 6.64—6.86 (5 H, m, ArH), and 7.2—7.4 (15 H, m, ArH); *m/e* 569 (M^+ — HCl). The 7,8-bisbenzyloxy-6-methoxy-3,4-dihydroisoquinoline hydrochloride (12a) was obtained as a yellow gum on evaporation of the mother-liquors from above.

To 1-(4-benzyloxybenzyl)-6,7-bisbenzyloxy-8-methoxy-3,4-dihydroisoquinoline hydrochloride (600 mg) in methanol (40 ml) at 0 °C under nitrogen was added sodium borohydride (260 mg) with stirring over 15 min. After stirring for a further 30 min at 20 °C, the solution was evaporated to dryness and the residue partitioned between saturated brine (100 ml) and ethyl acetate (100 ml). The aqueous layer was further extracted with ethyl acetate (3 × 100 ml) and the combined organic extracts were washed with water, dried, and reduced to a small volume. Ethereal hydrogen chloride was added to give pH 2—3, when the 1-(4-*benzyloxybenzyl*)-6,7-*bisbenzyloxy*-8-*methoxy*-1,2,3,4-*tetrahydroisoquinoline hydrochloride* (15a) separated (510 mg, 84%), m.p. 194—195 °C (from ethanol–ether) (Found: C, 74.85; H, 6.45; N, 2.15; Cl, 5.55. $C_{38}H_{37}NO_4$·HCl requires C, 75.0; H, 6.3; N, 2.3; Cl, 5.8%), $\nu_{max.}$ (Nujol) 1 600 and 1 585 cm^{-1}; $\lambda_{max.}$ 238, 258, 264, 268, 276, and 283 nm; δ([$^2H_4$]methanol) 3.0—3.3 (complex aliphatics), 3.98 (3 H, s, OCH_3), 5.04, 5.12, and 5.14 (each 2 H, s, *O*-benzyl-CH_2), 6.77 (1 H, s, ArH of ring A), 7.02 and 7.20 (each 2 H, d, *J* 8 Hz, ArH of ring c), and 7.4 (15 H, m, ArH); *m/e* 569 (M^+ — H_3Cl).

1-(4-*Benzyloxybenzyl*)-7,8-*bisbenzyloxy*-6-*methoxy*-1,2,3,4-*tetrahydroisoquinoline hydrochloride* (14a) was prepared in the same way from the corresponding 3,4-dihydroisoquinoline isomer (12a), and had m.p. 158—160 °C (from ethanol–ether) (Found: C, 74.9; H, 6.4; N, 2.2; Cl, 5.6. $C_{38}H_{37}NO_4$·HCl requires C, 75.0; H, 6.3; N, 2.3; Cl, 5.8%), $\nu_{max.}$ (Nujol) 1 600 and 1 590 cm^{-1}; $\lambda_{max.}$ 229, 258, 264, 268, and 280 nm; δ([$^2H_4$]methanol) 2.7—3.0 (complex aliphatics), 3.78 (3 H, s, OCH_3), 5.05 (4 H, s, 2 × *O*-benzyl-CH_2), 5.00 and 5.30 (each 1 H, d, *J* 10 Hz, AB system of C-8 *O*-benzyl-CH_2), 6.67 (1 H, s, ArH of ring A), 6.84 (4 H, s, ArH of ring c), and 7.36 (15 H, m, ArH); *m/e* 571 (M^+ — HCl).

1-(3-*Benzyloxy*-4-*methoxybenzyl*)-6,7-*bisbenzyloxy*-8-*methoxy*-1,2,3,4-*tetrahydroisoquinoline Hydrochloride* (15c) *and the* 7,8-*Bisbenzyloxy*-6-*methoxy-isomer* (14c).—The amide (11c) (1 g) was cyclised by treatment with phosphorus oxychloride in acetonitrile as described above for (11a), but heating at reflux for 50 min, to give a mixture of 3,4-dihydroisoquinoline hydrochlorides that was treated with sodium carbonate solution (1M; 80 ml). The solution was extracted with dichloromethane (1 × 30 ml, 2 × 40 ml) and the combined extracts were washed with brine (1 × 50 ml), dried, and evaporated. To the residue in warm methanol (10 ml) was added oxalic acid dihydrate (430 mg). Treatment of the solution with ether (25 ml) in portions caused precipitation of a solid that was recrystallised from ethanol to yield 1-(3-*benzyloxy*-4-*methoxybenzyl*)-6,7-*bisbenzyloxy*-8-*methoxy*-3,4-*dihydroisoquinoline oxalate* (13c) (330 mg, 39%), m.p. 180—182 °C (Found: C, 71.15; H, 5.85; N, 1.9. $C_{41}H_{39}NO_9$ requires C, 71.4; H, 5.65; N, 2.05%); $\nu_{max.}$ (Nujol) 1 600 and 1 510 cm^{-1}; $\lambda_{max.}$ 218, 227, and 325 nm; δ([$^2H_6$]DMSO) 2.8br (2 H, t, ArCH_2-CH_2N), 3.6—3.9 (2 H, s, ArCH_2CH_2N), 3.76 and 3.84 (each 3 H, s, OCH_3), 4.24 (2 H, s, ArCH_2C=N), 4.93, 5.04, and 5.22 (each 2 H, s, *O*-benzyl-CH_2), 6.84 (1 H, s, ArH of ring A), 6.98 (3 H, m, ArH of ring c), and 7.2—7.5 (15 H, m, ArH); *m/e* 599 (M^+ — $C_2H_2O_4$). The 7,8-dibenzyloxy-6-methoxy-3,4-dihydroisoquinoline oxalate (12c) was obtained as a gum from the mother-liquors.

The dihydroisoquinoline isomers were separately reduced by sodium borohydride in methanol as described earlier, but using dichloromethane as the extraction solvent in the work-up. 1-(3-*Benzyloxy*-4-*methoxybenzyl*)-6,7-*bisbenzyloxy*-8-*methoxy*-1,2,3,4-*tetrahydroisoquinoline* (15c) was converted in methanol with ethereal hydrogen chloride into its *hydrochloride*, m.p. 193—194 °C (from ethanol–ether), $\nu_{max.}$ (KBr) 3 400br, 1 590, and 1 500 cm^{-1}; $\lambda_{max.}$ 258, 270, and 280 nm; δ([$^2H_4$]methanol) 2.8—3.4 (complex aliphatics), 3.84 and 3.94 (each 3 H, s, OCH_3), 5.01, 5.02, and 5.20 (each 2 H, s, *O*-benzyl-CH_2), 6.7—7.0 (4 H, m, ArH), and 7.2—7.5 (15 H, m, ArH); *m/e* 601 (M^+ — HCl). 1-(3-*Benzyloxy*-4-*methoxybenzyl*)-7,8-*bisbenzyloxy*-6-*methoxy*-1,2,3,4-*tetrahydroisoquinoline* (14c) was converted in methanol–di-isopropyl ether into its *hydrochloride*, m.p. 140—142 °C (from ethanol–di-isopropyl ether) (Found: C, 73.2; H, 6.2; N, 2.1; Cl, 5.6. $C_{39}H_{39}NO_5$·HCl requires C, 73.4; H, 6.25; N, 2.2; Cl, 5.55%), $\nu_{max.}$ (KBr) 3 400br, 1 590, and 1 500 cm^{-1}; $\lambda_{max.}$ 258, 270, and 280 nm; δ([$^2H_4$]methanol) 2.8—3.4 (complex aliphatics), 3.81 and 3.89 (each 3 H, s, OCH_3), 4.86 and 5.06 (each 2 H, s, *O*-benzyl-CH_2), 5.10 and 5.30 (each 1 H, d, *J* 9 Hz, AB system of C-8 *O*-benzyl-

CH_2), 6.65—6.85 (4 H, m, ArH), and 7.25—7.5 (15 H, m, ArH); *m/e* 601 (M^+ − HCl).

6,7-*Bisbenzyloxy*-1-(3,4-*bisbenzyloxybenzyl*)-8-*methoxy*-1,2,3,4-*tetrahydroisoquinoline Hydrochloride* (15b) *and the* 7,8-*Bisbenzyloxy*-6-*methoxy-isomer* (14b).—The amide (11b) was cyclised as usual with phosphorus oxychloride in acetonitrile, heating at reflux for 1 h. The residue obtained on evaporation crystallised from ethanol–ether to give a mixture of 3,4-dihydroisoquinoline hydrochloride isomers (91%) that was reduced directly with sodium borohydride in ethanol as described earlier, using dichloromethane as extraction solvent in the work-up, to yield a mixture of 1,2,3,4-tetrahydroisoquinolines. This residue was triturated with ether and kept at 0 °C, when a solid separated that was converted, in dichloromethane, with ethereal hydrogen chloride into 6,7-*bisbenzyloxy*-1-(3,4-*bisbenzyloxybenzyl*)-8-*methoxy*-1,2,3,4-*tetrahydroisoquinoline hydrochloride* (15b) (31%), m.p. 162—163 °C (from methanol–di-isopropyl ether) (Found: C, 75.35; H, 6.25; N, 1.85. $C_{45}H_{43}NO_5$·HCl requires C, 75.65; H, 6.15; N, 1.95%), $\nu_{max.}$ (KBr) 3 400br, 1 600, 1 580, and 1 490 cm^{-1}; δ 2.4—3.4br (6 H, m), 3.92 (3 H, s, OCH_3), 5.12 (4 H, s, 2 × *O*-benzyl-CH_2), 5.14 and 5.18 (each 2 H, s, *O*-benzyl-CH_2), 6.39 (1 H, s, ArH of ring A), 6.74 (3 H, s, ArH of ring c), and 7.35 (20 H, m, ArH); *m/e* 677 (M^+ − HCl) and 374. The ethereal mother-liquor from the separation of the free-base isomers above afforded a residue on evaporation that was treated in methanol with ethereal hydrogen chloride to yield 7,8-*bisbenzyloxy*-1-(3,4-*bisbenzyloxybenzyl*)-6-*methoxy*-1,2,3,4-*tetrahydroisoquinoline hydrochloride* (14b) (29%), m.p. 137—138 °C [from methanol–di-isopropyl ether (×3)] (Found: C, 75.35; H, 6.15; N, 1.75. C_{45}-$H_{43}NO_5$·HCl requires C, 75.65; H, 6.15; N, 1.95%), $\nu_{max.}$ (KBr) 3 400br and 1 600br cm^{-1}; δ 2.4—3.4br (6 H, m), 3.81 (3 H, s, OCH_3), 4.93 (2 H, s, *O*-benzyl-CH_2), 5.04 (4 H, s, 2 × *O*-benzyl-CH_2), 5.14 (2 H, ABq, *J* 11 Hz, C-8 *O*-benzyl-CH_2), 6.34 (1 H, s, ArH of ring A), 6.68 (3 H, m, ArH of ring c), and 7.8 (20 H, m, ArH); *m/e* 677 (M^+ − HCl), 374.

6,7-*Bisbenzyloxy*-1-(4-*benzyloxy*-3-*methoxybenzyl*)-8-*methoxy*-1,2,3,4-*tetrahydroisoquinoline Hydrochloride* (15d) *and the* 7,8-*Bisbenzyloxy*-6-*methoxy-isomer* (14d) (*with S. A. Surgenor*).—These compounds were prepared from the amide (11d) by the methods detailed above for (15b) and (14b). The 6,7-*bisbenzyloxy*-8-*methoxy-isomer* (15d) was isolated as the *hydrochloride* (26%), m.p. 215—217 °C (from dichloromethane–ether) (Found: C, 73.3; H, 6.2; N, 2.4. $C_{39}H_{39}NO_5$·HCl requires C, 73.4; H, 6.3; N, 2.2%), $\nu_{max.}$ (KBr) 3 400, 1 640, 1 615, and 1 510 cm^{-1}; δ 2.4—3.4br (6 H, m), 3.72 and 3.96 (each 3 H, s, OCH_3), 5.01, 5.04, and 5.06 (each 2 H, s, *O*-benzyl-CH_2), 6.41 (1 H, s, ArH of ring A), 6.7 (3 H, m, ArH of ring c), and 7.36 (15 H, m, ArH); *m/e* 601 (M^+ − HCl) and 374. The 7,8-*bisbenzyloxy*-6-*methoxy-isomer* (14d) gave a *hydrochloride* (30%), m.p. 94—96 °C (from methanol–ether) (Found: C, 71.1; H, 6.0; N, 2.2. $C_{39}H_{39}NO_5$·HCl·H_2O requires C, 71.4; H, 6.3; N, 2.1%), $\nu_{max.}$ (KBr) 3 400, 1 605, 1 590, and 1 515 cm^{-1}; δ 2.5—3.4br (6 H, m), 3.67 and 3.85 (each 3 H, s, OCH_3), 5.07 (4 H, s, 2 × *O*-benzyl-CH_2), 5.18 (2 H, ABq, *J* 12 Hz, C-8 *O*-benzyl-CH_2), 6.40 (1 H, s, ArH of ring A), 6.6 (3 H, m, ArH of ring c), and 7.36 (15 H, m, ArH); *m/e* 601 (M^+ − HCl) and 374.

The relatively stable 1,2,3,4-tetrahydroisoquinoline hydrochlorides (14a—d) and (15a—d) were debenzylated by hydrogenolysis, as described earlier for preparation of the bisphenethylamine (8a), until hydrogen uptake ceased, to afford the following more labile tri-, or tetra-phenolic isoquinolines. These were prepared just before they were required and were characterised spectroscopically: 6,7-dihydroxy-1-(4-hydroxybenzyl)-8-methoxy-1,2,3,4-tetrahydroisoquinoline (7a), from (15a), as the hydrochloride, m.p. 260—262 °C (from methanol–ether), $\nu_{max.}$ (Nujol) 3 465, 1 615, and 1 595 cm^{-1}; $\lambda_{max.}$ 238, 279, and 286sh nm; δ([$^2H_4$]methanol), 3.0 (complex aliphatics), 3.98 (3 H, s, OCH_3), 6.8 (1 H, s, ArH of ring A), 6.85 and 7.18 (each 2 H, d, *J* 8 Hz, ArH of ring C); 7,8-dihydroxy-1-(4-hydroxybenzyl)-6-methoxy-1,2,3,4-tetrahydroisoquinoline (6a), from (14a), as the hydrochloride, m.p. 288—290 °C (from methanol–ether), $\nu_{max.}$ (Nujol) 3 480, 1 610, and 1 590 cm^{-1}; $\lambda_{max.}$ 229, 283, and 290sh nm; δ([$^2H_4$]methanol) 3.3—3.4 (complex aliphatics), 3.78 (3 H, s, OCH_3), 6.38 (1 H, s, ArH of ring A), 6.85 and 7.18 (each 2 H, d, *J* 8 Hz, ArH of ring c); 6,7-dihydroxy-1-(3,4-dihydroxybenzyl)-8-methoxy-1,2,3,4-tetrahydroisoquinoline (7b), from (15b), as the hydrochloride, m.p. 171—180 °C (from methanol–ether), $\nu_{max.}$ (KBr) 3 600—2 900br, 1 615, and 1 530 cm^{-1}; δ(D_2O) 3.88 (3 H, s, OCH_3), 6.58 (1 H, s, ArH of ring A), and 7.02 (3 H, m, ArH of ring c); 7,8-dihydroxy-1-(3,4-dihydroxybenzyl)-6-methoxy-1,2,3,4-tetrahydroisoquinoline (6b), from (14b), as the hydrochloride, m.p. 235—236 °C (decomp.; sinters *ca.* 130 °C) (from aqueous methanol–ether), $\nu_{max.}$ (KBr) 3 600—2 700br, 1 640, 1 610, 1 540, and 1 520 cm^{-1}; δ(D_2O) 3.83 (3 H, s, OCH_3), 6.5 (1 H, s, ArH of ring A), and 6.84 (3 H, m, ArH of ring c); 6,7-dihydroxy-1-(3-hydroxy-4-methoxybenzyl)-8-methoxy-1,2,3,4-tetrahydroisoquinoline (7c), from (15c), as the hydrochloride, m.p. 180—181 °C (from ethanol–ether), $\nu_{max.}$ (Nujol) 3 600—3 000br, 1 600, and 1 570 cm^{-1}; $\lambda_{max.}$ 274 nm; δ([$^2H_4$]methanol) 3.0—3.5 (complex aliphatics), 3.96 (6 H, s, 2 × OCH_3), 6.50 (1 H, s, ArH of ring A), and 6.78 (3 H, m, ArH of ring c); 7,8-dihydroxy-1-(3-hydroxy-4-methoxybenzyl)-6-methoxy-1,2,3,4-tetrahydroisoquinoline (6c), from (14c), as the hydrochloride, m.p. 314—318 °C (from ethanol–ether), $\nu_{max.}$ (Nujol) 3 600—3 000br, 1 600, and 1 510 cm^{-1}; $\lambda_{max.}$ 273 nm; δ([$^2H_4$]methanol) 3.0—3.4 (complex aliphatics), 3.83 (6 H, s, 2 × OCH_3), 6.38 (1 H, s, ArH of ring A), and 6.78 (3 H, m, ArH of ring c); 6,7-dihydroxy-1-(4-hydroxy-3-methoxybenzyl)-8-methoxy-1,2,3,4-tetrahydroisoquinoline (7d) (with *S. A. Surgenor*), from (15d), as the hydrochloride, m.p. 220—221 °C (decomp.) (from methanol–ether), $\nu_{max.}$ (KBr) 3 600—2 700br, 1 600, and 1 515 cm^{-1}; δ([$^2H_6$]-DMSO) 3.76 and 3.88 (each 3 H, s, OCH_3), 6.44 (1 H, s, ArH of ring A), and 6.74 (3 H, m, ArH of ring c); and 7,8-dihydroxy-1-(4-hydroxy-3-methoxybenzyl)-6-methoxy-1,-2,3,4-tetrahydroisoquinoline (6d) (*with S. A. Surgenor*), from (14d), as the hydrochloride, m.p. 234—235 °C (decomp.) (from methanol–ether), $\nu_{max.}$ 3 600—2 700br, 1 610, and 1 510 cm^{-1}; δ([$^2H_6$]DMSO) 3.78 and 3.83 (each 3 H, s, OCH_3), 6.35 (1 H, s, ArH of ring A), 6.78 (2 H. s, ArH of ring c), and 6.86 (1 H, s, ArH of ring c).

6,7-*Dihydroxy*-1-(4-*hydroxybenzyl*)-8-*methoxy*-2-*methyl*-1,2,3,4-*tetrahydroisoquinoline* (7e).—The corresponding tribenzyloxyisoquinoline hydrochloride (15a) (60 mg) in dichloromethane (30 ml) was shaken with saturated sodium hydrogen carbonate solution (30 ml). The aqueous layer was further extracted with dichloromethane (3 × 20 ml) and the combined organic phases were washed with water, dried, and evaporated. The residue in methanol (20 ml) and aqueous formaldehyde (40% w/v; 5 ml) was stirred under nitrogen for 12 min, then cooled to 0 °C and treated

with sodium borohydride (500 mg). After stirring at room temperature for 1 h the solvents were removed, the residue was partitioned between saturated brine (40 ml) and ethyl acetate (1 × 40, 3 × 20 ml), and the combined organic layers were washed with brine, dried, and evaporated to afford 6,7-bisbenzyloxy-1-(4-benzyloxybenzyl)-8-methoxy-2-methyl-1,2,3,4-tetrahydroisoquinoline (17a) (56 mg, 96%), $\nu_{max.}$ 1 595 and 1 500 cm^{-1}; δ 2.40 (3 H, s, NCH_3), 2.8—3.4 (complex), 3.92 (3 H, s, OCH_3), 4.04 (1 H, t, 1-H), 5.01—5.02 (6 H, 3 × s, 3 × *O*-benzyl-CH_2), 6.46 (1 H, s, ArH of ring A), 7.00 (4 H, m, ArH of ring c), and 7.2—7.5 (15 H, m, ArH). Debenzylation by the usual method gave the *2-methyltrihydroxyisoquinoline hydrochloride* (7e) (30 mg; 93%), m.p. 218—220 °C (Found: C, 61.35; H, 6.2; N, 4.05; Cl, 10.0. $C_{18}H_{21}NO_4$,HCl requires C, 61.45; H, 6.25; N, 4.0; Cl, 10.1%), $\nu_{max.}$ (Nujol) 3 480br, 1 610, and 1 592 cm^{-1}; $\lambda_{max.}$ 229, 283, and 290 nm; δ([$^2H_4$]methanol) 3.05 (3 H, s, NCH_3), 3.3 (complex), 3.98 (3 H, s, OCH_3), 6.49 (1 H, s, ArH of ring A), and 6.88 and 7.15 (each 2 H, d, ArH of ring c); *m/e* 315 (M^+ — HCl).

The following 2-methylhydroxyisoquinolines were prepared in the same way, except that dichloromethane rather than ethyl acetate was used as extraction solvent in the isolation of all the 2-methylbenzyloxyisoquinolines other than (16a), and with other modifications as stated.

7,8-*Dihydroxy*-1-(4-*hydroxybenzyl*)-6-*methoxy*-2-*methyl*-1,2,3,4-*tetrahydroisoquinoline* (6e). Prepared from the isoquinoline (14a) *via* the 2-methyltribenzyl ether (16a), $\nu_{max.}$ 1 610 and 1 500 cm^{-1}; δ 2.20 (3 H, s, NCH_3), 2.85 (complex), 3.40 (2 H, t), 3.84 (3 H, s, OCH_3), 4.03 (1 H, t, 1-H), 5.00—5.10 (2 H, m, C-8 *O*-benzyl-CH_2), 4.97 and 5.06 (each 2 H, s, *O*-benzyl-CH_2), 6.40 (1 H, s, ArH of ring A), 6,75 and 6.96 (each 2 H, d, *J* 9 Hz, ArH of ring c), and 7.4 (15 H, m, ArH). Debenzylation as usual gave the *trihydroxyisoquinoline hydrochloride* (6e), m.p. 225—226 °C (Found: C, 61.4; H, 6.25; N, 4.1; Cl, 9.95. $C_{18}H_{21}NO_4$,HCl requires C, 61.45; H, 6.25; N, 4.0; Cl, 10.1%), $\nu_{max.}$ (Nujol) 3 400br, 1 600, and 1 505 cm^{-1}; $\lambda_{max.}$ 229, 283, and 290 nm; δ([$^2H_4$]methanol) 3.1 (complex aliphatics), 2.80 (3 H, s, NCH_3), 3.80 (3 H, s, OCH_3), 6.39 (1 H, s, ArH of ring A), and 6.90 and 7.16 (each 2 H, d, *J* 9 Hz, ArH of ring c); *m/e* 315 (M^+ — HCl).

6,7-*Dihydroxy*-1-(3,4-*dihydroxybenzyl*)-8-*methoxy*-2-*methyl*-1,2,3,4-*tetrahydroisoquinoline* (7f). Prepared from (15b) *via* the 2-*methyltetrabenzyl ether* (17b), isolated as the *picrate*, m.p. 134—135 °C (from ethyl acetate–ether) (Found: C, 67.65; H, 5.25; N, 6.15. $C_{52}H_{48}N_4O_{12}$ requires C, 67.8; H, 5.25; N, 6.1%). The picrate in chloroform was passed down a column of alumina (neutral grade III; *ca.* 10 g : 200 mg picrate) and the eluate evaporated to recover the free base, $\nu_{max.}$ 1 605, 1 595, and 1 500 cm^{-1}; δ 2.34 (3 H, s, NCH_3), 2.5—3.3 (6 H, m), 3.92 (3 H, s, OCH_3), 5.02 and 5.05 (each 2 H, s, *O*-benzyl-CH_2), 5.12 (4 H, s, 2 × *O*-benzyl-CH_2), 6.45 (1 H, s, ArH of ring A), 6.84 (3 H, m, ArH of ring c), and 7.35 (20 H, m, ArH); *m/e* 691 (M^+) and 388. Hydrogenolysis of this free base afforded the tetrahydroxyisoquinoline hydrochloride (7f), m.p. 228—229 °C (from methanol–ether), $\nu_{max.}$ (KBr) 3 600—2 900br, 1 625, 1 600, 1 525, and 1 505 cm^{-1}; δ([$^2H_6$]DMSO) 3.19 (3 H, s, NCH_3), 3.81 (3 H, s, OCH_3), 6.36 (1 H, s, ArH of ring A), and 6.7 (3 H, m, ArH of ring c).

7,8-*Dihydroxy*-1-(3,4-*dihydroxybenzyl*)-6-*methoxy*-2-*methyl*-1,2,3,4-*tetrahydroisoquinoline* (6f). Prepared from (14b) *via* the 2-*methyltetrabenzyl ether* (16b), isolated as the *picrate*, m.p. 76—78 °C (from ethyl acetate–ether) (Found: C, 67.7; H, 5.4; N, 6.15. $C_{52}H_{48}N_4O_{12}$ requires C, 67.8; H, 5.25; N, 6.1%). The free base was recovered as above, $\nu_{max.}$ 1 605, 1 595, and 1 500 cm^{-1}; δ 2.16 (3 H, s, NCH_3), 2.3—3.3 (6 H, m), 3.79 (3 H, s, OCH_3), 4.95, 5.04, and 5.07 (each 2 H, s, *O*-benzyl-CH_2), 5.11 (2 H, AB q, *J* 10 Hz, C-8 *O*-benzyl-CH_2), 6.38 (1 H, s, ArH of ring A), 6.65 (3 H, m, ArH of ring c), and 7.3 (20 H, m, ArH); *m/e* 691 (M^+) and 388. Debenzylation of the free base gave the tetrahydroxyisoquinoline hydrochloride (6f), m.p. 220—221 °C (from methanol–ether), $\nu_{max.}$ (KBr) 3 700—2 900br, 1 615, 1 605, and 1 525 cm^{-1}; δ([$^2H_6$]DMSO) 3.21 (3 H, s, NCH_3), 3.81 (3 H, s, OCH_3), 6.49 (1 H, s, ArH of ring A), and 6.7 (3 H, m, ArH of ring c).

6,7-*Dihydroxy*-1-(3-*hydroxy*-4-*methoxybenzyl*)-8-*methoxy*-2-*methyl*-1,2,3,4-*tetrahydroisoquinoline* (7g). Prepared from (15c) *via* the 2-methyltribenzyl ether (17c), $\nu_{max.}$ (film) 1 605, 1 590, and 1 500 cm^{-1}; $\lambda_{max.}$ 258 and 283 nm; δ(60 MHz) 2.7—3.2 (complex aliphatics), 2.32 (3 H, s, NCH_3), 3.86 and 3.92 (each 2 H, s, OCH_3), 5.05 (4 H, s, 2 × *O*-benzyl-CH_2), 5.12 (2 H, s, *O*-benzyl-CH_2), 6.46 (1 H, s, ArH of ring A), 6.8 (3 H, m, ArH of ring c), and 7.2—7.5 (15 H, m, ArH). Debenzylation as usual afforded the *trihydroxyisoquinoline hydrochloride* (7g) m.p. 206—208 °C, $\nu_{max.}$ (KBr) 3 600—2 700, 1 600, and 1 510 cm^{-1}; $\lambda_{max.}$ 273 nm; δ([$^2H_4$]methanol) 3.0—3.3 (complex aliphatics), 2.88 (3 H, s, NCH_3), 3.96 (6 H, s, 2 × OCH_3), 6.48 (1 H, s, ArH of ring A), and 6.8 (3 H, m, ArH of ring c) [Found: *m/e*, 208.0976. $C_{11}H_{14}NO_3$ (isoquinoline fragment) requires *m/e*, 208.0975].

7,8-*Dihydroxy*-1-(3-*hydroxy*-4-*methoxybenzyl*)-6-*methoxy*-2-*methyl*-1,2,3,4-*tetrahydroisoquinoline* (6g). Prepared from (14c) *via* the 2-methyltribenzyl ether (16c), $\nu_{max.}$ (film) 1 600 and 1 510 cm^{-1}; δ(60 MHz) 2.9—3.3 (complex aliphatics), 2.28 (3 H, s, NCH_3), 3.89 (6 H, s, 2 × OCH_3), 5.00 and 5.20 (each 1 H, d, *J* 8 Hz, AB system of C-8 *O*-benzyl-CH_2), 5.15 (4 H, s, 2 × *O*-benzyl-CH_2), 6.45 (1 H, s, ArH of ring A), 6.82 (3 H, m, ArH of ring c), and 7.15—7.48 (15 H, m, ArH). The usual debenzylation procedure afforded the *trihydroxyisoquinoline hydrochloride* (6g), m.p. 218—225 °C, $\nu_{max.}$ (KBr) 1 600 and 1 510 cm^{-1}; $\lambda_{max.}$ 273 nm; δ([$^2H_4$]-methanol) 2.78 (3 H, s, NCH_3), 3.0—3.2 (complex aliphatics), 3.83 (6 H, s, 2 × OCH_3), 6.38 (1 H, s, ArH of ring A), and 6.78 (3 H, m, ArH of ring c) [Found: *m/e*, 208.0977. $C_{11}H_{14}NO_3$ (isoquinoline fragment) requires *m/e*, 208.0975].

6,7-*Dihydroxy*-1-(4-*hydroxy*-3-*methoxybenzyl*)-8-*methoxy*-2-*methyl*-1,2,3,4-*tetrahydroisoquinoline* (7h). Prepared from (15d) *via* the 2-*methyltribenzyl ether* (17d), isolated as the *picrate*, m.p. 109—110 °C (from ethyl acetate–ether) (Found: C, 65.25; H, 5.3; N, 6.7. $C_{46}H_{44}N_4O_{12}$ requires C, 65.4; H, 5.2; N, 6.65%). The free base was recovered as detailed earlier, $\nu_{max.}$ 1 590 and 1 505 cm^{-1}; δ 2.39 (3 H, s, NCH_3), 2.6—3.4 (6 H, m), 3.81 and 3.95 (each 3 H, s, OCH_3), 5.03 (4 H, s, 2 × *O*-benzyl-CH_2), 5.09 (2 H, s, *O*-benzyl-CH_2), 6.45 (1 H, s, ArH of ring A), 6.78 (3 H, m, ArH of ring c), and 7.35 (15 H, m, ArH); *m/e* 615 (M^+) and 388. Debenzylation gave the trihydroxyisoquinoline hydrochloride (7h), m.p. 218—219 °C (decomp.), $\nu_{max.}$ (KBr) 3 400br, 3 100br, 1 620, 1 610, and 1 530 cm^{-1}; δ([$^2H_6$]-DMSO) 3.18 (3 H, s, NCH_3), 3.69, 3.78 (each 3 H, s, OCH_3), 6.45 (1 H, s, ArH of ring A), and 6.68 (3 H, m, ArH of ring c).

7,8-*Dihydroxy*-1-(4-*hydroxy*-3-*methoxybenzyl*)-6-*methoxy*-2-*methyl*-1,2,3,4-*tetrahydroisoquinoline* (6h). Prepared from (14d) *via* the 2-*methyltribenzyl ether* (16d), isolated as a *picrate*, m.p. 123—124 °C (from ethyl acetate–ether) (Found: C, 65.15; H, 5.3; N, 6.8. $C_{46}H_{44}N_4O_{12}$ requires

C, 65.4; H, 5.2; N, 6.65%). The free base, recovered as usual, had $\nu_{max.}$ 1 590 and 1 505 cm^{-1}; δ 2.23 (3 H, s, NCH_3), 2.4—3.4 (6 H, m), 2.70 and 2.83 (each 3 H, s, OCH_3), 5.06 and 5.09 (each 2 H, s, *O*-benzyl-CH_2), 5.14 (2 H, AB q, *J* 10 Hz, C-8 *O*-benzyl-CH_2), 6.41 (1 H, s, ArH of ring A), 6.65 (3 H, m, ArH of ring C), and 7.3 (15 H, m, ArH); *m/e* 615 (M^+) and 388. Hydrogenolysis gave the trihydroxyisoquinoline hydrochloride (6h), m.p. 200 °C (decomp.), sinters 105 °C, $\nu_{max.}$ (KBr) 3 600—3 000br, 1 620, and 1 520 cm^{-1}; δ([$^2H_6$]DMSO) 2.72 (3 H, s, NCH_3), 3.72 and 3.83 (each 3 H, s, OCH_3), 6.37 (1 H, s, ArH of ring A), and 6.76 (3 H, m, ArH of ring C).

We thank the S.R.C. for Research Studentships (to R. C, F, J, and A. P, O.), the Salters Company for an Award (to R. C. F. J.), the Commonwealth Scholarship Plan for an Award (to A. M.), and the staff of the Botanic Garden, University of Cambridge, for cultivation of plants. We also thank the Nuffield Foundation, the S.R.C., and Roche Products for financial support, and Mr. S. A. Surgenor for the experiments indicated.

[0/1596 *Received*, 20*th October*, 1980]

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