Aspidosperma Alkaloids. Conversion of Tabersonine into Vindoline

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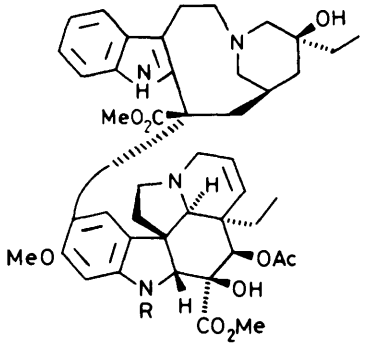
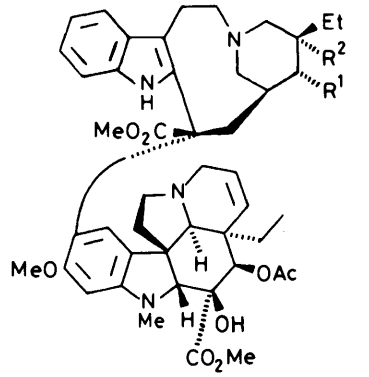
An efficient synthesis of vindoline, the indoline portion of the clinically useful antitumour agent vinblastine, starting from tabersonine is described.

The widespread clinical use of the antitumour agent vinblastine (vincaleucoblastine, VBL) (**1**), an indole alkaloid of the Madagascan periwinkle *Catharanthus roseus*, has generated great interest in the synthesis of compound (**1**) and analogues, such as vincristine (leurocristine, VCR) (**2**), leurosine (**3**), and leurosidine (**4**).



(**1**) R = Me
(**2**) R = CHO



(**3**) R^1R^2 = =O
(**4**) R^1 = H, R^2 = OH

Successful routes to these compounds have been devised by several authors by means of coupling the catharanthine component ('upper half') with the vindoline moiety ('lower half').[1,2] To date, however, insufficient data from structure modification studies are available to draw definite conclusions regarding structure–activity relationships. Thus, synthetic routes to structurally modified compounds are especially desirable.

Recently, we described the synthesis of vindoline (VDL) (**5**), starting from the rare alkaloid 11-methoxytabersonine (**6**),[3,†] *via* alcohol (**7**), by virtue of a regio- and stereo-selective reaction sequence.[4] The incorporation of a hydroxy function at the 17-position could be also viewed as a versatile point of entry into the synthesis of 17-substituted derivatives of VDL and, thence, of VBL. In none of the previous structure–activity relationship studies has the 17-position of VDL or VBL been examined as a modified site leading to potentially active compounds.

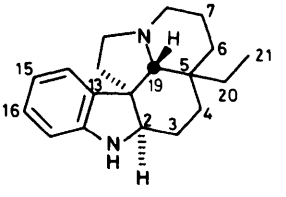
From a chemical standpoint, at least six different synthetic approaches to VDL (**5**) have been reported.[5] However, despite the diversity of these strategies, none of them refers to the preparation of enantiomerically pure VDL starting from (−)-tabersonine (TBS) (**8**). This was of interest since compound (**8**) is more readily available than is its 11-methoxy derivative (**6**), and might constitute a convenient precursor for VDL and its 17-modified analogues (*vide infra*).

Results and Discussion

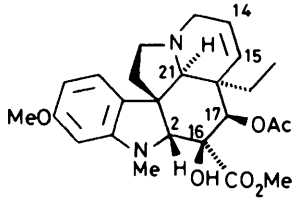
In principle, a simple approach to a precursor of VDL involves the electrophilic substitution of TBS (**8**) at the 11-position. In practice, however, this route is limited by the propensity of TBS (**8**) to react toward electrophilic reagents preferentially at C-10 and/or C-16.[6] Since we were unable to functionalise the intact TBS molecule in the desired manner, we planned to mask the β-anilinoacrylic moiety in TBS (**8**) at the indoline oxidation level by reduction with sodium cyanoborohydride in acetic acid to give the known compound 2β,16β-dihydro-TBS (**12**).[7] Few systematic analyses of the products of electrophilic substitution in indolines have been reported and the literature is conflicting in that in some cases monosubstitution is stated to occur at C-5 (indole numbering) (*i.e.* our C-10) while in others it is claimed to be at C-6 (our C-11).[8] By analogy with reported nitration of some indolines,[9] we hoped that 2β,16β-dihydro-TBS (**12**) could be functionalised at C-11 (C-6, indole numbering).

In the event, however, nitration of compound (**12**) under the same conditions as described by Ikan *et al.*[9a] resulted in a complex mixture, which was impractical for our purposes. During this investigation we were aware of a report by Miyake and Kikugawa[10] describing a regioselective bromination at C-6 (indole numbering), *i.e.* at C-11 in compound (**12**), with bromine in 97% sulphuric acid in the presence of Ag^I sulphate or with bromine in superacidic medium (HF:SbF_5,

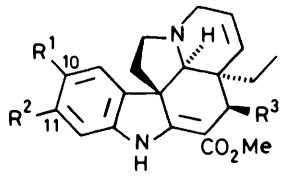
† The numbering system used throughout this paper is one proposed by J. Le Men and W. I. Taylor, *Experientia*, 1965, **21**, 508. All the cited compound have been also named (see Experimental section) as derivatives of aspidospermidine (i) as required by *Chemical Abstracts* nomenclature rules.



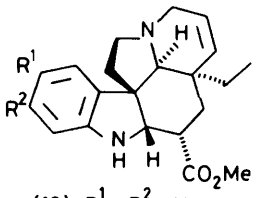
(i)



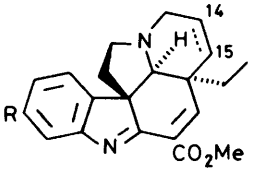
(5)



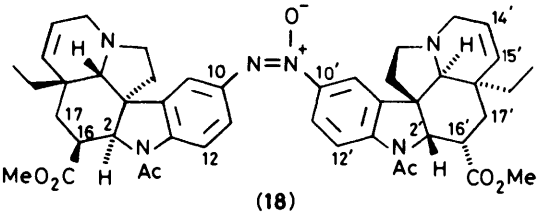
(6) $R^1 = R^3 = H$, $R^2 = OMe$
(7) $R^1 = H$, $R^2 = OMe$, $R^3 = OH$
(8) $R^1 = R^2 = R^3 = H$
(9) $R^1 = Br$, $R^2 = R^3 = H$
(10) $R^1 = R^2 = H$, $R^3 = OH$
(11) $R^1 = H$, $R^2 = OMe$, $R^3 = Nu$



(12) $R^1 = R^2 = H$
(13) $R^1 = H$, $R^2 = Br$
(14) $R^1 = NH_2$, $R^2 = H$
(15) $R^1 = Br$, $R^2 = H$



(16) R = OMe, 14,15-double bond
(17) R = H

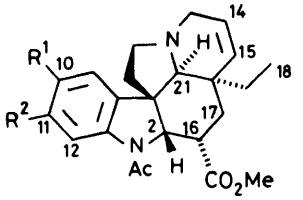


(18)

1:1). Despite numerous attempts and strict adherence to prescribed procedures, bromination of compound (**12**) led to a mixture of products which resulted presumably from random electrophilic attack at ring *A* and/or peripheral 14,15-double bond. In any case, the desired 11-bromo derivative (**13**) could not be identified.

We have been unsuccessful so far in finding conditions suitable for straightforward functionalisation of the indoline (**12**) at the 11-position, so a more circuitous route was employed. For the introduction of the oxygenated function at the proper position, we elected to do this *via* a specific *ortho*-rearrangement of the 10-*O*-(methylsulphonyl)-*N*-(acetyl)-hydroxylamine (**19**) as recently reported by Gassman and Granrud.[11] Compound (**19**) would be available by reduction of its corresponding 10-nitro derivative (**20**). When 96% nitric acid in trifluoroacetic acid (TFA) was used to effect nitration of *N*-acetyl-2β,16β-dihydro-TBS (**21**), a particularly clean and preparatively useful reaction took place and nitro derivative (**20**) was isolated in 86% yield. The salient features of the ^1H n.m.r. spectrum of compound (**20**) were the resonances due to the aromatic protons, which can be analysed as an AMX system [δ 8.20 (d, *J* 2.4 Hz, 9-H), 7.51 (d, *J* 8.5 Hz, 12-H), and 8.04 (dd, *J* 8.5 and 2.4 Hz, 11-H)]. The location of the nitro group at C-10 in compound (**20**) was confirmed by nuclear Overhauser effect difference spectrum (n.O.e.d.s.) For instance, a 15% enhancement in the doublet at δ 7.51 (12-H) was observed upon irradiation of the three-proton singlet at δ 2.35 due to NCOMe. Attempts to reduce compound (**20**) to the required 10-hydroxylamino derivative (**22**) by means of Zn dust and ammonium chloride in aqueous ethanol[12] resulted in the isolation of the azoxy compound (**18**), apparently formed from the rapid condensation of compound (**22**) and the intermediate nitroso derivative.[13] Similar results have ben noted under other conditions.[14]

Attention was therefore directed toward electrophilic functionalisation of the 10-amino indoline (**23**) at C-11, a transformation whose regiochemical outcome is a predictable consequence of stereoelectronic control of electrophilic aromatic substitution. Subsequent reduction of the nitro group in



(19) $R^1 = N(Ac)O_3SMe$, $R^2 = H$
(20) $R^1 = NO_2$, $R^2 = H$
(21) $R^1 = R^2 = H$
(22) $R^1 = NHOH$, $R^2 = H$
(23) $R^1 = NH_2$, $R^2 = H$
(24) $R^1 = NHAc$, $R^2 = H$
(25) $R^1 = NH_2$, $R^2 = Br$
(26) $R^1 = H$, $R^2 = Br$
(27) $R^1 = Br$, $R^2 = H$

compound (**20**) was brought about by activated zinc in acetic acid at ambient temperature to give the 10-amino compound (**23**) (75—89%). As expected, the ^1H n.m.r. spectrum of the product (**23**) exhibited upfield shifts of 0.41 p.p.m. for 9-H, 1.52 p.p.m. for 11-H, and 1.14 p.p.m. for 12-H with respect to the signals for nitro compound (**20**). On occasions, a variable amount of the 10-acetamido compound (**24**) [$M^{+\cdot}$ 437; δ 2.28 and 2.13 (3 H each, NCOMe)] was obtained from the above reduction but the formation of this polar compound could be prevented by careful monitoring (t.l.c.) of the reaction. It was hoped that a halogen X at C-11 would serve as a useful leaving group thus providing the site for aromatic nucleophilic substitution in ring *A* (X ⟶ OMe). Thus, the amine (**23**) was regioselectively brominated with *N*-bromosuccinimide (NBS) in *NN*-dimethylformamide (DMF)[15] at room temperature in the dark to furnish the bromide (**25**) as the only product (81%) (Scheme 1). Compound (**25**) [$M^{+\cdot}$ 475/473 ($^{81}Br/^{79}Br$)] was identified by spectral methods; in particular the *two* aromatic

protons appeared as singlets at δ 7.42 (12-H) and 6.55 (9-H) in the ^{1}H n.m.r. spectrum, thus confirming that substitution had occurred at the 11-position. The conditions of this reaction were somewhat critical to ensure a good yield; use of alternative conditions produced a range of other products. It was recognised from the outset that N-1 in compound (**14**) must be protected temporarily as the acetamide derivative, since the 9-, 11-, 12-position in compound (**14**) would not differ in electrophilic activity enough to allow specific substitution at C-11.

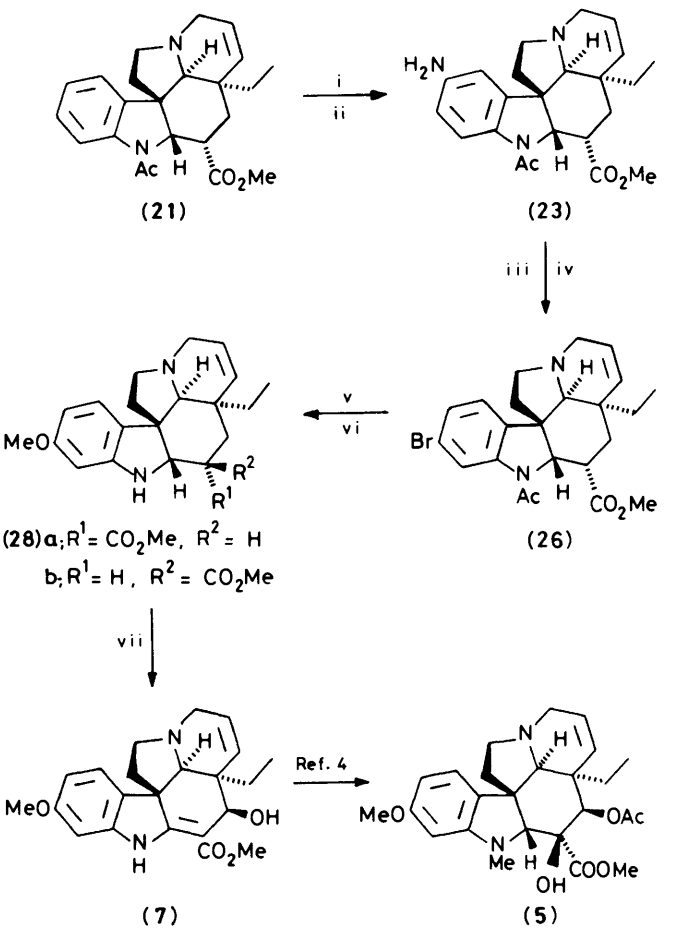
Since the 11-bromo compound (**26**) appeared to be the key intermediate in the synthesis outlined in Scheme 1, efforts were made to optimise the protodeamination (hydro-dediazonization)[16] of amine (**25**). Reduction of its diazonium salt with hypophosphorous acid gave a low yield of nitro compound (**20**), whilst treatment of amine (**25**) with isopentyl nitrite in refluxing tetrahydrofuran (THF) according to the procedure of Cadogan and Molina[17] proceeded smoothly to get the expected bromide (**26**) as the sole product (84%). Deamination of ring *A* caused downfield shifts for all the aromatic protons in the ^{1}H n.m.r. spectrum which exhibited an AMX system [δ 7.50 (br s, 12-H), 7.17 (dd, *J* 8.1 and 1.6 Hz, 10-H), and 6.96 (d, *J* 8.1 Hz, 9-H)].

From the known compound 10-bromo-TBS (**9**)[6] the isomeric 10-bromo indoline (**27**) was prepared by reduction with sodium cyanoborohydride–acetic acid to give compound (**15**), followed by standard acetylation at N-1. ^{1}H N.m.r. spectroscopy of compound (**27**) showed a close resemblance to that of compound (**26**) except that the aromatic protons were nearly isochronous.

These findings establish beyond doubt the correctness of the orientation assigned to compound (**26**) produced in our synthesis reported in Scheme 1.

Since our ultimate goal was to prepare VDL (**5**), the incorporation of the 11-OMe group *via* nucleophilic aromatic substitution appeared mandatory. In 1969, Bacon and Rennison[18] reported an efficient copper(I) iodide-assisted synthesis of aryl alkyl ethers from the appropriate aryl halides using alkali metal alkoxides in 2,4,6-collidine (2,4,6-trimethylpyridine). This reaction was apparently complicated by hydrolysis of both acetamido and ester functions and, only after treatment with methanolic hydrogen chloride, the required methyl ethers (**28a,b**) were obtained as a mixture of 16-epimers in 66% yield (Scheme 1). Duplication of signals detected in the ^{1}H n.m.r. and proton-noise-decoupled (p.n.d.) ^{13}C n.m.r. spectra was consistent with the presence of two closely similar compounds. The ^{1}H n.m.r. spectrum was particularly revealing and showed, *inter alia*, an AMX pattern at δ 6.80 (d, 9-H), 6.18 (d, 10-H) and 6.12 (d, 12-H) [$J_{9,10}$ 8.1, $J_{10,12}$ 2.4 Hz], two olefinic protons at δ 5.74 (ddd, *J* 9.8, 3.8, and 2.3 Hz, 14-H), and 5.34 (br d, *J* 9.8 Hz, 15-H), and an aromatic OMe group at δ 3.70 for the 16β-H epimer (**28a**). Further, the loss of stereochemical integrity at C-16 is not unexpected since the kinetically favoured product (**28a**) is actually thermodynamically less stable in the presence of bases, giving epimer (**28b**). Kutney *et al.* also observed base-catalysed equilibration in the 14,15-saturated series.[19] However, it was unimportant whether we obtained a mixture of 16-epimers since the target molecule requires the obliteration of this stereocentre.

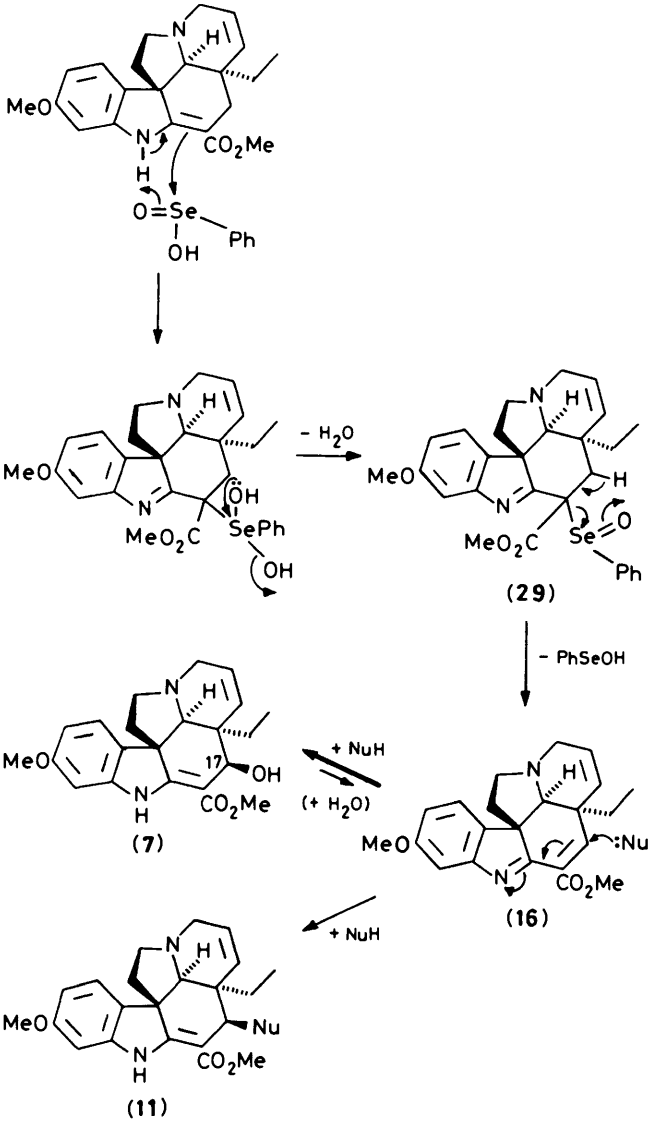
As anticipated, the final stages of VDL synthesis entailed essentially the dehydrogenation of epimers (**28**) to 11-methoxy-TBS (**6**). For dehydrogenation of epimers (**28**), a series of reagents was evaluated as, for example, manganese dioxide, lead(IV) acetate, copper(II) chloride–pyridine, chloranil, dichlorodicyano-*p*-benzoquinone (DDQ) and Pd/C in refluxing mesitylene, but none was very promising with respect to completeness of reaction.[20] The only effective, reliable method proved to be reaction with benzeneseleninic anhydride (BSA), a methodology that has been successfully employed for the oxidation of dihydroergolines providing the corresponding



Scheme 1. *Reagents and conditions:* i, 96%HNO_3–TFA, 0 °C to room temp.; ii, Zn–AcOH, room temp.; iii, NBS, DMF, room temp.; iv, $Me_2CHCH_2CH_2$, ONO, THF, reflux; v, MeONa, CuI, 2,4,6-collidine, 120 °C; vi, MeOH–HCl; vii, BSA or PhSe(O)OH, benzene, 40 °C

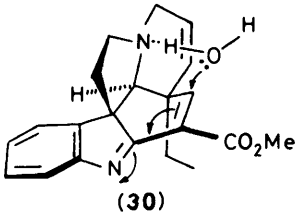
indoles with high chemoselectivity.[21] Gratifyingly, we have found that dehydrogenation of epimers (**28**) and 17-hydroxylation[→ (**7**)] could be effected concurrently in the presence of BSA or its acid. When compounds (**28**) were treated with an equimolar amount of BSA in benzene (30 min at 40 °C), an essentially clean reaction was observed and the acrylate (**7**) was isolated in 81% yield. During the course of the reaction, t.l.c. (diethyl ether–diethylamine, 24:1) indicated the formation of a transient product of lower polarity, which we identified as 11-methoxy-TBS (**6**) by comparison with a sample generously supplied by Dr. Potier (Gif-sur-Yvette). Indeed, when the reaction of epimers (**28**) with BSA was interrupted prior to completion, a mixture of compounds (**6**) and (**7**) was obtained. If the same reaction is performed in the presence of benzeneseleninic acid, the acrylate (**7**) is produced in 69% yield. Assignment of structure (**7**) was made by m.s. and ^{1}H n.m.r. spectroscopy in strict analogy to 17-hydroxy-TBS (**10**).[4] While conventional electron-impact m.s. failed to detect the molecular ion of compound (**7**), owing to easy dehydration, the f.a.b.-m.s. (fast atom bombardment-m.s.) technique, employing glycerol as matrix and 7 keV xenon neutral as primary beam, permitted detection of the pseudo-molecular ion at *m/z* 383 $(M + H)^+$. The ^{1}H n.m.r. spectrum ($CDCl_3$) of compound (**7**) showed, *inter alia*, the presence of a NH singlet at δ 9.13, two multiplets at δ 5.80 and 6.03 due to 15-H and 14-H, an exchangeable singlet at δ 6.45 (OH), and a triplet at δ 0.63 (*J* 6.5 Hz) for 18-H_3. The signal for 17-H in acrylate (**7**) occurs as a doublet at δ 4.68. The

observed coupling constant of 2 Hz to the bridging 21-H is consistent with a 17α-H orientation and hence the structure is that shown in (7). The two prochiral hydrogens at C-17 in 11-OMe-TBS (6) provided a useful structural probe for compound (7). In the 400 MHz ^{1}H n.m.r. of compound (6), only the *pro-S* hydrogen [δ 2.54 (dd, *J* 15 and 2 Hz)] showed a 2 Hz coupling between 17α-H and 21α-H, transmitted through four σ bonds arranged in a 'W' conformation.[22]



Scheme 2.

It is likely that this remarkably efficient conversion, (**28**) ⟶ (**7**), is initiated by formation of compound (**6**) and is continued by an ene-reaction of BSA or its acid to lead to the indolenine (**29**) (Scheme 2). Thermal elimination of benzeneselenenic acid introduces the 16,17-double bond and the reaction sequence is then terminated by capture of water by the reactive azadiene (**16**) thus formed.[23] Such a marked proclivity for nucleophilic addition of water could be explained in terms of subtle stereoelectronic and conformational effects. The reverse effect, *i.e.* stabilisation of the azadiene, was found in the vincadifformine analogue (**17**).[4] The concave–convex nature of the skeleton of compound (**16**) was expected to provide the geometrical bias required to introduce the hydroxy group in a stereocontrolled fashion. In addition, attack of (**16**) by water on the β-face is likely to be the result of delivery of the nucleophile by the neighbouring bridgehead N_b, as shown in structure (**30**).



(**30**)

The acrylate (**7**) is a particularly versatile synthon not only because it possesses the proper chirality at C-17 present in VDL (**5**), but also because, through the intermediacy of the enimine (**16**) (Scheme 2), a series of 17-substituted compounds (**11**) could be available for further elaboration to VDL-like analogues.

Since compound (**7**) has recently been transformed into vindoline (**5**) by us,[4] the foregoing route constitutes the first conversion of the abundant and accessible alkaloid tabersonine (**8**) into VDL (**5**).

Experimental

I.r. spectra were recorded on a Perkin-Elmer 681 spectrometer for chloroform solutions; u.v. spectra on a Perkin-Elmer model 554 for solutions in methanol. ^{1}H N.m.r. spectra were recorded on a Bruker WP-80 (80 MHz) with deuteriochloroform as solvent unless otherwise stated and tetramethylsilane as internal standard. ^{13}C N.m.r. spectra were taken in deuteriochloroform on a Varian XL-100 at 25.2 MHz, with tetramethylsilane as internal standard. Mass spectra (electron-impact and fast-atom bombardment) were determined on a VG 70-70 EQ (70 eV). Compounds were detected on developed chromatograms by fluorescence quenching (λ 254 or 365 nm) or visualised with cerium(IV) ammonium sulphate (CAS) (1% in 85% phosphoric acid); R_F and colours (CAS spray on t.l.c.) of products are given. Flash chromatography (f.c.) was carried out as described by Still *et al.*[24] and performed with Silica gel S (230—400 mesh). Preparative t.l.c. (p.l.c.) was performed on 1 mm thick layers of Merck silica gel HF_{254} coated on 20 × 20 cm glass plates. Sodium sulphate was employed as the drying agent. 2β,16β-Dihydro-TBS (**12**) was prepared by a literature procedure.[7]

Acetylation of Compound (**12**) *to* 1-*Acetyl*-2β,16β-*dihydro-TBS* [*Methyl* 1-*Acetyl*-6,7-*didehydro*-(2β,5α,12β,19α)-*aspidospermidine*-3α-*carboxylate*] (**21**).—Anhydrous sodium acetate (2.46 g, 30 mmol) was added to a stirred solution of compound (**12**) (1.014 g, 3.0 mmol) in acetic anhydride (5 ml). After being stirred at room temperature for 8 h, the reaction mixture was worked up to give 1-acetyldihydro-TBS (**21**) (1.13 g, 99%) as needles, m.p. 128—129 °C (from dichloromethane–di-isopropyl ether); R_F 0.17 (diethyl ether–cyclohexane, 4:1; colourless); δ_H* 5.85 (ddd, *J* 10.3, 5.2, and 1.4 Hz, 14-H), 5.17 (br d, *J* 10.3 Hz, 15-H), 4.80 (d, *J* 4.8 Hz, 2-H), 3.63 (s, CO_2Me), 2.30 (s, NCOMe), and 0.68 (t, *J* 6.6 Hz, 18-H); *m/z* (100 °C) 380 ($M^{+\cdot}$, 18%), 349 (8), 294 (50), 252 (10), 186 (10), 144 (45), 122 (100), and 107 (67).

Nitration of Compound (**21**) *to* 1-*Acetyl*-10-*nitro*-2β,16β-*dihydro-TBS* [*Methyl* 1-*Acetyl*-6,7-*didehydro*-15-*nitro*-(2β,5α,12β,19α)-*aspidospermidine*-3α-*carboxylate*] (**20**).—To a solution of (**21**) (2.52 g, 6.63 mmol) in TFA (20 ml) at 0 °C under nitrogen, was added 96% nitric acid (0.6 ml, 13.2 mmol) dropwise during 5 min. The syrupy brown solution was kept at

* Locants for all n.m.r. spectra refer to the TBS numbering scheme.

room temperature for 30 min and then poured into conc. ammonia–ice. Dichloromethane (80 ml) was added to the golden yellow reaction mixture and the organic layer was washed with brine. The dried solution was evaporated under reduced pressure to give a foam. The residue was purified by f.c. to yield 1-*acetyl* 10-*nitrodihydro-TBS* (**20**) (2.42 g, 86%) as beautiful yellow needles, m.p. 177—178 °C (from diethyl ether); R_F 0.21 (diethyl ether; pale yellow); δ_H 8.20 (d, *J* 2.4 Hz, 9-H), 8.04 (dd, *J* 8.4 and 2.4 Hz, 11-H), 7.51 (d, *J* 7.5 Hz, 12-H), 5.88 (ddd, *J* 9.4, 5.3, and 2.6 Hz, 14-H), 5.18 (br d, *J* 9.4 Hz, 15-H), 3.67 (s, CO_2Me), 2.35 (s, NCOMe), and 0.77 (t, *J* 5.3 Hz, 18-H_3); *m/z* (100 °C) 425 ($M^{+\bullet}$, 13%), 394 (9), 339 (100), 322 (56), 189 (94), 135 (94), 122 (93), and 107 (33) (Found: C, 65.0; H, 6.4; N, 9.75. $C_{23}H_{27}N_3O_5$ requires C, 64.92; H, 6.39; N, 9.87%).

Reduction of Compound (**20**) *to* 1-*Acetyl*-10-*amino*-2β,16β-*dihydro-TBS* [*Methyl* 1-*Acetyl*-15-*amino*-6,7-*didehydro*-(2β,5α,-12β,19α)-*aspidospermidine*-3α-*carboxylate* (**23**) *and its* 10-N-*Acetyl Derivative* (**24**).—A solution of the foregoing nitro derivative (**20**) (714 mg, 1.68 mmol) in acetic acid (5 ml) was treated with zinc dust (activated in 2M-hydrochloric acid, and washed with methanol and light petroleum) (1.066 g, 0.016 g-atom) and then stirred at room temperature for 2 h. T.l.c. examination (ethyl acetate–conc. ammonia, 49:1) revealed complete disappearance of starting material and emergence of two new compounds, R_F 0.40 (major) and 0.27, corresponding to compounds (**23**) and (**24**), respectively. The reaction mixture was diluted with water (100 ml) and filtered through Celite. The filtrate was neutralised with conc. ammonia and extracted with dichloromethane (2 × 30 ml). Evaporation of the dried organic phase to dryness, and purification of the residue by f.c. (ethyl acetate–conc. ammonia, 100:1) gave two fractions.

The first eluted material, isolated as needles (530 mg, 89%) was 1-*acetyl*-10-*aminodihydro-TBS* (**23**), m.p. 208—209 °C (from ethyl acetate); R_F 0.40 (pale blue); δ_H 7.06 (d, *J* 9.2 Hz, 12-H), 6.52 (dd, *J* 9.2 and 2.4 Hz, 11-H), 6.47 (d, *J* 2.4 Hz, 9-H), 5.85 (ddd, *J* 9.7, 5.3, and 1.4 Hz, 14-H), 5.18 (br d, *J* 9.7 Hz, 15-H), 4.76 (d, *J* 4.8 Hz, 2-H), 3.63 (s, CO_2Me), 2.28 (s, NCOMe), and 0.75 (t, *J* 5.3 Hz, 18-H); *m/z* (120 °C) 395 ($M^{+\bullet}$, 31%), 364 (25), 159 (18), 135 (18), and 122 (100) (Found: C, 69.9; H, 7.4; N, 10.55. $C_{23}H_{29}N_3O_3$ requires C, 69.84; H, 7.39; N, 10.62%).

The second eluted material, isolated as a glass (47 mg, 6%), was 10-acetamido compound (**24**), δ_H 7.63 (br s, 9-H), 7.40 (br s, NH), 7.02 (dd, *J* 9.4 and 2.9, 11-H), 5.83 (ddd, *J* 11.6, 5.6, and 1.5 Hz, 14-H), 5.13 (br d, *J* 11.6 Hz, 15-H), 4.80 (d, *J* 5.2 Hz, 2-H), 3.65 (s, CO_2Me), 2.28 (s, NCOMe), 2.13 (s, NCOMe), and 0.70 (t, *J* 5.3 Hz, 18-H); *m/z* (110 °C) 437 ($M^{+\bullet}$), 351 ($M^+ - C_4H_7O_2$), 201, 135, 122, and 107; *m** at *m/z* 281.9 (437→351).

Bromination of Compound (**23**) *to* 1-*Acetyl*-10-*amino*-11-*bromo*-2β,16β-*dihydro-TBS* [*Methyl* 1-*Acetyl*-15-*amino*-16-*bromo*-6,7-*didehydro*-2β,5α,12β,19α)-*aspidospermidine*-3α-*carboxylate*] (**25**).—A solution of freshly crystallised NBS (96 mg, 0.53 mmol) in dry DMF (1 ml) was added to a solution of compound (**23**) (210 mg, 0.53 mmol) in dry DMF (4 ml). The reaction mixture was kept at room temperature under nitrogen in the dark for 15 min. T.l.c. indicated the formation of a single component (R_F 0.46, ethyl acetate–conc. ammonia, 99:1; pale blue). The mixture was diluted with water (50 ml) and extracted with diethyl ether (3 × 10 ml). Removal of the dried solvent gave a glass, which was purified by passage through a short column of silica gel, with diethyl ether as eluant, to give the title compound (**25**) (204 mg, 81%) as a glass, $\lambda_{max.}$ 264 and 321 nm; δ_H 7.42 (br s, 12-H), 6.55 (s, 9-H), 5.85 (ddd, *J* 10.0, 5.2, and 1.4 Hz, 14-H), 5.17 (br d, *J* 10 Hz, 15-H), 4.72 (d, *J* 5.2 Hz, 2-H), 3.65 (s, CO_2Me), 2.25 (s, NCOMe), and 0.68 (t, *J* 5.6 Hz, 18-H); *m/z* (90 °C) 475 ($M^{+\bullet}$, ^{81}Br), 473 ($M^{+\bullet}$, ^{79}Br), 387 (7), 385 (7), 239 (8), 237 (8), 135 (50), and 121 (100) (Found: M^+, 475.1299. $C_{23}H_{28}N_3O_3\ ^{81}Br$ requires *M*, 475.1293).

Proto-deamination of Compound (**25**) to 1-*Acetyl*-11-*bromo*-2β,16β-*dihydro-TBS* [*Methyl* 1-*Acetyl*-16-*bromo*-6,7-*didehydro*-(2β,5α,12β,19α)-*aspidospermidine*-3α-*carboxylate*] (**26**).—*Method A*. To a solution of isopentyl nitrite (0.8 ml, 2.98 mmol) in dry THF (10 ml) under nitrogen was added dropwise a solution of amino compound (**25**) (640 mg, 1.36 mmol) in dry THF (15 ml), and the reaction mixture was refluxed for 1 h. The solvent was removed under reduced pressure and water (20 ml) was added to the residue before extraction with dichloromethane (3 × 15 ml). After evaporation of the dried organic phase the residue was purified by f.c. (diethyl ether as eluant) to yield the title compound (**26**) (520 mg, 84%) as an amorphous glass, R_F 0.44 (ethyl acetate; colourless); $\lambda_{max.}$ 250, 285, and 290 nm; δ_H 7.50 (d, *J* 1.6 Hz, 12-H), 7.17 (dd, *J* 8.1 and 1.6 Hz, 10-H), 6.96 (d, *J* 8.1 Hz, 9-H), 5.86 (br dd, *J* 9.3 and 5.3 Hz, 14-H), 5.18 (br d, *J* 9.3 Hz, 15-H), 4.78 (d, *J* 5.3 Hz, 2-H), 3.63 (s, CO_2Me), 2.32 (s, NCOMe), and 0.75 (t, *J* 6 Hz, 18-H_3); *m/z* (120 °C) 460 ($M^{+\bullet}$, ^{81}Br), 458 ($M^{+\bullet}$, ^{79}Br), 429, 427, 374, 372, 224, 222, 143, 135, 122, and 107 (Found: M^+, 460.1192. $C_{23}H_{27}N_2O_3\ ^{81}Br$ requires *M*, 460.1184).

Method B. The amine (**25**) (450 mg, 0.94 mmol) was dissolved in 85% phosphoric acid (10 ml). The resultant homogeneous solution was cooled to −10 °C and a solution of sodium nitrite (39 mg, 5.7 mmol) in water (10 ml) was added slowly such that the temperature was not allowed to rise above −5 °C. The resulting dark yellow solution was added to a stirred, pre-chilled (0 °C) 50% hypophosphorous acid solution (3 ml) dropwise, and the mixture was then allowed to warm at room temperature. After 3 h, the solution was diluted with ice-cold water (30 ml), neutralised with conc. ammonia, and extracted with dichloromethane (2 × 20 ml). The combined dried extracts were evaporated under reduced pressure and the residue was purified by p.l.c. (ethyl acetate as eluant) to give compound (**26**) (100 mg, 23%).

Reduction of Nitro compound (**20**) *to Azoxy Derivative* (**18**).—Compound (**20**) (500 mg, 1.18 mmol) and ammonium chloride (136 mg, 2.55 mmol) were added to a mixture of ethanol (20 ml) and water (5 ml) at room temperature. After deoxygenation of the solution with argon for 30 min, zinc dust (173 mg, 2.55 mg-atom) was added. The yellow mixture was stirred for 20 min under argon and the suspension was filtered. The filtrate was basified with conc. ammonia and extracted rapidly with dichloromethane (3 × 20 ml). The combined extracts were dried, filtered, and evaporated to leave a reddish glass which contained several components on t.l.c. examination. The major component was separated by p.l.c. (ethyl acetate as eluant) and azoxy derivative (**18**) was isolated as a yellow foam (481 mg, 51%), R_F 0.29 (ethyl acetate; red spot after spraying with conc. sulphuric acid); δ_H 8.35 (d, *J* 2 Hz, 9′-H), 8.19 (dd, *J* 8.9 and 2 Hz, 11′-H), 7.95 (d, *J* 8.9 Hz, 12′-H), 7.41 (dd, *J* 9 and 2.7 Hz, 11-H), 5.85 (m, 14- and 14′-H), 5.18 (br d, *J* 10 Hz, 15- and 15′-H), 4.82 (d, *J* 5.3 Hz, 2- and 2′-H), 3.66 and 3.64 (s, CO_2Me), 2.37 and 2.35 (s, NCOMe), and 0.75 (m, 18- and 18′-H); *m/z* (100 °C) 802 ($M^{+\bullet}$), 786 ($M^+ - O$), 700, 408, 394, and 308.

11-*Methoxy*-2,16-*dihydro-TBS* [*Methyl* 6,7-*Didehydro*-16-*methoxy*-(2β,5α,12β,19α)-*aspidospermidine*-3α *and* β-*carboxylates*] (**28a,b**).—Sodium (52 mg, 2.28 mg-atom) was dissolved in absolute methanol (1 ml) and the cooled solution was diluted with dry 2,4,6-collidine (5 ml). Freshly purified copper(I) iodide (72 mg, 0.38 mmol) and compound (**26**) (350 mg, 0.76 mmol) were added and the mixture was stirred under nitrogen at 120 °C. After 4 h, further copper(I) iodide (36 mg, 0.19 mmol) was added and the mixture was stirred for 2 h, then filtered

through a Celite pad, diluted with water (50 ml), and neutralised with 5% aqueous phosphoric acid. The aqueous solution was thoroughly extracted with chloroform (5 × 10 ml) and the combined extracts were dried and concentrated under reduced pressure (oil-pump). The oily residue was taken up in dry methanol (10 ml) and a saturated methanolic solution of hydrochloric acid (5 ml) was added. After the mixture had been refluxed for 2 h, the solvent was evaporated off. Diethyl ether (50 ml) and 10% aqueous sodium hydrogen carbonate (10 ml) were added. The organic phase was washed with copper(II) sulphate (2 × 10 ml) and dried. Evaporation of the solvent left the title compounds (**28**) (185 mg, 66%) as a mixture of 16-epimers which were carried on to the next stage without separation. The mixture of esters (**28a**) and (**28b**) showed two spots (red) on t.l.c. (diethyl ether–cyclohexane, 4:1); $\nu_{max.}$ 3 470, 1 720, 1 660, and 1 620 cm^{-1}; $\lambda_{max.}$ 208 log ε 4.55), 245 (3.75), and 302 nm (3.71; *m/z* (85 °C) 368 ($M^{+\cdot}$, 11%), 337 (7), 282 (38), 144 (40), 135 (100), 122 (62), and 107 (48) (Found: M^+, 368.2089, $C_{22}H_{28}N_2O_3$ requires *M*, 368.2099).

Oxidation of Compounds (**28a,b**) *to* 17-*Hydroxy*-11-*methoxy-TBS* [*Methyl* 2,3,6,7-*Tetrahydro*-4β-*hydroxy*-16-*methoxy*-(4α,5α,12β,19α)-*aspidospermidine*-3-*carboxylate*] (**7**).—(a) *With BSA*. The foregoing mixture (**28a,b**) (125 mg, 0.34 mmol) in dry benzene (5 ml) was treated with BSA (90 mg, 0.25 mmol) and the mixture was heated under nitrogen at 40 °C. After 10 min, t.l.c. (diethyl ether–diethylamine, 24:1) indicated that most of the indolines (**28a,b**) had reacted to produce two new products, at R_F 0.57 and 0.21. Further BSA (32 mg, 0.09 mmol) was added, and at 30 min conversion was complete and the less polar compound had disappeared. The residue obtained after evaporation was dissolved in dichloromethane (50 ml) and extracted with 5% hydrochloric acid (2 × 15 ml). The combined aqueous extracts were basified with 10% aqueous sodium hydrogen carbonate and extracted with dichloromethane (2 × 15 ml). Evaporation of the dried extracts left a yellowish mass which was purified by f.c. (elution with diethyl ether–diethylamine, 24:1) to afford the pure 17-hydroxy-11-methoxy-TBS (**7**) (105 mg, 81%), R_F 0.21 (sky-blue); δ_H 9.13 (br s, NH), 6.03 (m, 14-H), 5.80 (m, 15-H), 6.45 (br s, OH), 4.68 (d, *J* 2 Hz, 17-H), 3.78 (s, OMe + CO_2Me), 2.87 (d, *J* 2 Hz, 21-H), and 0.62 (t, *J* 6.5 Hz, 18-H); f.a.b.-m.s. *m/z* 383 (M + H)$^+$; e.i.-m.s. (80 °C) 382 (absent), 364, 349, 335, 273, 135, and 122.

(b) *With benzeneseleninic acid*. Compounds (**28a,b**) (50 mg, 0.13 mmol) were dissolved in dry benzene (5 ml), benzeneseleninic acid (38 mg, 0.06 mmol) was added, and the mixture was heated at 40 °C under nitrogen, with monitoring as above. Additional benzeneseleninic acid (12 mg, 0.06 mmol) was added after 30 min, and all starting material was used up by 1 h. After being cooled, diluted with diethyl ether (5 ml), and washed successively with 5% aqueous sodium hydrogen carbonate and then water, the solution was dried and then concentrated to afford a yellow mass. P.l.c. of this residue gave the title compound (64 mg, 69%).

10-*Bromo-TBS* [*Methyl* 15-*Bromo*-2,3,6,7-*tetradehydro*-(5α,12β,19α)-*aspidospermidine*-3-*carboxylate*] (**9**).—This compound was obtained according to the method of Lewin *et al.*[6] by bromination (NBS–TFA) of TBS (**8**); R_F 0.44 (heptane–diethyl ether, 1:1; blue-violet; δ_H 8.97 (br s, NH), 7.33 (d, *J* 1.9 Hz, 9-H), 7.23 (dd, *J* 8 and 1.9 Hz, 11-H), 6.67 (d, *J* 8 Hz, 12-H), 5.70 (m, 14- and 15-H), and 3.75 (s, CO_2Me), and 0.73 (t, *J* 6 Hz, 18-H).

10-*Bromo*-2β,16β-*dihydro-TBS* [*Methyl* 15-*Bromo*-6,7-*didehydro*-(2β,5α,12β,19α)-*aspidospermidine*-3α-*carboxylate*] (**15**).—To a solution of compound (**9**) (200 mg, 0.48 mmol) dissolved in acetic acid (10 ml) was added sodium cyanoborohydride (45 mg, 0.70 mmol) and the resulting mixture was stirred at room temperature until t.l.c. (heptane–diethyl ether, 1:1) indicated reduction to be complete. After 2 h, the mixture was diluted with water (50 ml), basified with conc. ammonia (to pH 8), and extracted repeatedly with diethyl ether. Evaporation of the dried extracts gave pure 10-bromo-2,16-dihydro-TBS (**15**) (185 mg, 92%) as a glass, R_F 0.33 (red-orange); $\nu_{max.}$ 3 420, 1 722, and 1 620 cm^{-1}; δ_H 7.88 (br s, NH), 7.08 (d, *J*, 2 Hz, 9-H), 7.07 (dd, *J* 9 and 2 Hz, 11-H), 6.39 (d, *J* 9 Hz, 12-H), 5.76 (ddd, *J* 9.8, 5.1, and 1.5 Hz, 14-H), 5.43 (br dd, *J* 9.8 and 2.3 Hz, 15-H), 3.72 (s, CO_2Me), and 0.65 (t, *J* 6.7 Hz, 18-H) (Found: M^+, 418.1095. $C_{21}H_{25}N_2O_2$ ^{81}Br requires *M*, 418.1079).

1-*Acetyl*-10-*bromo*-2β,16β-*dihydro-TBS* [*Methyl* 1-*Acetyl*-15-*bromo*-6,7-*didehydro*-(2β,5α,12β,19α)-*aspidospermidine*-3α-carboxylate] (**27**).—The foregoing compound (**15**) (150 mg, 0.36 mmol) was dissolved in pyridine (5 ml) and the solution was treated with acetic anhydride (0.5 ml) overnight. The solution was poured onto ice–water and, after neutralisation with 5% aqueous sodium hydrogen carbonate, the title compound was recovered in diethyl ether (2 × 10 ml). The extracts were washed successively with 5% aqueous copper(II) sulphate and brine, and dried. The solvent was evaporated off to afford the title compound (158 mg, 95%), homogeneous on t.l.c. (chloroform–ethyl acetate, 2:1); R_F 0.25 (colourless); δ_H 7.30 (m, 12-, 11-, and 9-H), 5.85 (ddd, *J* 9.3, 5.3, and 2.5 Hz, 14-H), 5.20 (br dd, *J* 9.3 and 2.5 Hz, 15-H), 4.73 (d, *J* 2.5 Hz, 2-H), 3.63 (s, CO_2Me), 2.30 (NCOMe), and 0.78 (t, *J* 6.7 Hz, 18-H); *m/z* (100 °C) 460/458 ($M^{+\cdot}$, ^{81}Br/^{79}Br), 429, 427, 374, 372, 224, 222, 143, and 135.

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