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ALKALOID STUDIES—LIII[1]

THE STRUCTURES OF NINE NEW ALKALOIDS FROM *ASPIDOSPERMA DASYCARPON* A. DC.

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Abstract—The structures of twelve alkaloids (I–XII) isolated from *Aspidosperma dasycarpon* A. DC. are deduced. Nine of these indole alkaloids are new.

THE bark of the species *Aspidosperma dasycarpon* A. DC. has been found to contain twelve alkaloids, which we describe as a group in this paper. Uleine[2] (I), (+)-guatambuine[3] (N_b-methyltetrahydroolivacine) (II) and N_b-methyltetrahydroellipticine[4] (III) had been isolated and studied previously. These three compounds together with olivacine,[3b,5,6] 1,2-dihydroolivacine, ellipticine,[4] methoxyellipticine,[4a] and 1,2-dihydroellipticine,[3a,6] and metho salts of two of these bases[4c] were the only complex indole alkaloids lacking the tryptamine two carbon bridge, known before this study of *A. dasycarpon*. It was thus of considerable interest and biogenetic significance to isolate, from this plant, seven new indolic compounds (IV–X) all of which also lack this feature. The structure determination of one of these, apparicine (IX), has been dealt with in detail[7] and only the essential features of the proof will be mentioned in this paper. Bases IV–VIII have been the subject of a preliminary communication.[8] Both remaining alkaloids (XI and XII) possess the usual two carbon function at the indolic β-position, though in aspidodasycarpine[9] (XI) the second carbon has become detached from the basic nitrogen atom, as in the cinchona bases;[10] the co-occurrence of such a structure with ten alkaloids lacking the tryptamine bridge will be commented on later in this paper.

[1] Part LII, A. Walser and C. Djerassi, *Helv. Chim. Acta* **48**, 391 (1965).

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[3a] J. Schmutz and F. Hunziker, *Helv. Chim. Acta* **41**, 288 (1958); [b] M. A. Ondettie and V. Deulofeu, *Tetrahedron Letters* No. 7, 1 (1959); *Tetrahedron* **15**, 160 (1961); [c] P. Carvalho-Ferreira, G. B. Martini-Bettolo and J. Schmutz, *Experientia* **15**, 179 (1959).

[4a] S. Goodwin, A. F. Smith and E. C. Horning, *J. Amer. Chem. Soc.* **81**, 1903 (1959); [b] R. B. Woodward, G. A. Iacobucci and F. A. Hochstein, *Ibid.* **81**, 4434 (1959); [c] G. Büchi, D. W. Mayo and F. A. Hochstein, *Tetrahedron* **15**, 167 (1961).

[5] J. Schmutz and F. Hunziker, *Pharm. Acta Helv.* 341 (1958).

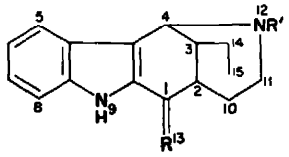
[6] G. B. Marini-Bettolo and J. Schmutz, *Helv. Chim. Acta* **42**, 2146 (1959).

[7] J. A. Joule, H. Monteiro, L. J. Durham, B. Gilbert and C. Djerassi, *J. Chem. Soc.* in press (1965).

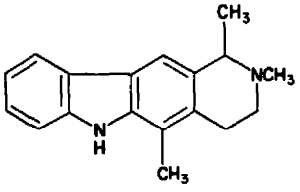
[8] M. Ohashi, J. A. Joule, B. Gilbert and C. Djerassi, *Experientia* **20**, 263 (1964).

[9] M. Ohashi, J. A. Joule and C. Djerassi, *Tetrahedron Letters* 3899 (1964).

[10] R. B. Turner and R. B. Woodward, *The Alkaloids* (Edited by R. H. Manske and H. L. Holmes) Vol. III; Chap. 1b. Academic Press, New York (1953).



	R	R′
I	CH_2	CH_3
IV	CH_2	H
V	O	CH_3
VI	O	H
VII	H, OH	CH_3
VIII	H, CH_2OH	CH_3
XIII	CH_3	CH_3CO
XIV	CH_2	CH_3CH_2
XV	CH_2	CH_3, CH_2CH_3
XVI	CH_2	CH_2CH_3, CH_3



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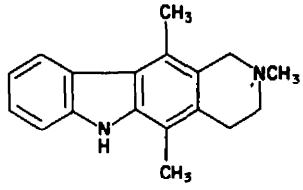

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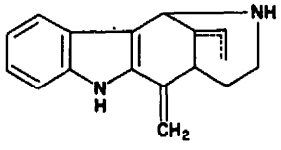


X

Des-N-*methyluleine* (IV) *and des*-N-*methyldehydrouleine* (X)

The alkaloid (IV) had UV absorption and ceric sulphate colour reaction identical with those of uleine (I). Its mol. wt. (252) by mass spectrometry, was fourteen mass units less than that of uleine (I) and the spectrum displayed a characteristic fragmentation pattern[11] totally in accord with that to be expected from an uleine molecule lacking the N-methyl function. Chemical confirmation for this postulate was obtained by reacting IV with methyl iodide. Separation of the resulting mixture gave uleine (I) of the same sign of rotation as the naturally occurring alkaloid together with uleine methiodide.

Acetylation of IV gave N-acetyl-des-N-methyluleine (XIII), LAH reduction of which then gave N-ethyl-des-N-methyluleine (XIV), both compounds still possessing the typical uleine UV chromophore and both showing the characteristic mass spectrometric fragmentation patterns.[11] Reaction of the N-ethyl compound (XIV) with

[11] For the mass spectra of compounds I and V and a detailed discussion of the fragmentation behaviour of uleine and its congeners see Ref. 2c.

methyl iodide yielded a crystalline quaternary salt which was *not* identical with the iodide obtained by reaction of uleine with ethyl iodide. These two salts (XV and XVI) must represent stereoisomers about the nitrogen atom of the type encountered with codeine[12a] and the tropane alkaloids.[12b] Lack of material however precluded a more detailed examination[13] of the conformations of the N-alkyl groups in this pair of salts.

The dehydro analog (X) of des-N-methyluleine (IV) possessed the same UV absorption as uleine and des-N-methyluleine (IV) the mol. wt. being two mass units less than that of IV. The new double bond is thus not conjugated with the chromophore and can only be located between carbon atoms 3–14, 14–15, or 10–11. Position 10–11 can be eliminated on the grounds of the similarity between the mass spectrometric fragmentation pattern shown by the base and that of des-N-methyluleine (IV), which characteristic pattern also speaks strongly for the alkaloid having the same skeleton as IV and I. No rigorous distinction can be made between the two remaining possibilities since insufficient material was available for NMR examination which would have settled this point.

Dasycarpidone (V) *and des-*N-*methyldasycarpidone* (VI)

Dasycarpidone (V) and des-N-methyldasycarpidone (VI) both possess UV absorption characteristics of α-keto indole chromophores[14] and both have peaks corresponding to such conjugated carbonyl groups[14] in their IR spectra (6·10 and 6·07 μ). Des-N-methyldasycarpidone the mol. wt. (254) of which was fourteen mass units less than that of dasycarpidone (V) was easily converted to the latter by the method described for the interconversion of des-N-methyluleine (IV) and uleine (I).

The general resemblance between the mass spectrum[2c] of dasycarpidone and that of uleine (I),[2c] together with the mol. wt. of the compound (two mass units higher than that of uleine) and UV and IR data suggested structure V for the base. The NMR spectrum of dasycarpidone (V) provided clear-cut confirmation for this postulate, showing resonances for four aromatic protons, an indolic N-hydrogen, benzylic proton (doublet at 4·35 ppm, J = 2·5 c/s) at C-4,[2b,2c] N–methyl singlet and methyl (of an ethyl) functions. Significantly absent were the absorptions shown by the exocyclic methylenic protons of uleine (I).

Chemical verification for the structural assignment was provided by converting uleine (I) to dasycarpidone (V). This was achieved by low temperature ozonolysis of uleine when up to 15% of dasycarpidone (V) of the same magnitude and sign of rotation as the natural material, could be isolated and identified. Oxidation of uleine with iodine or chromium trioxide pyridine also produced dasycarpidone in low yield.

Dasycarpidol (VII)

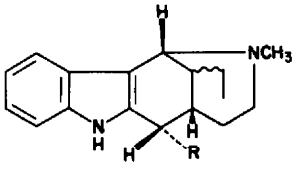
Dasycarpidol showed typical indole UV absorption and a mass spectrometrically determined mol. wt. (270) two units higher than that of dasycarpidone (V). The absence of ketonic IR absorption and the non-conjugated nature of the UV absorption

[12a] K. Koczka and G. Bernath, *Chem. Ind.* 1401 (1958); [b] G. Fodor, K. Koczka and J. Lestyan, *J. Chem. Soc.* 1411 (1956); [c] R. Bognar and S. Szabo, *Tetrahedron Letters* 2867 (1964).

[13] J. McKenna, J. White and A. Tulley, *Tetrahedron Letters* 1097 (1962); J. K. Becconsall and R. A. Y. Jones, *Ibid.* 1103 (1962).

[14] J. A. Ballantine, C. B. Barrett, R. J. S. Beer, B. G. Boggiano, S. Eardley, B. E. Jennings and A. Robertson, *J. Chem. Soc.* 2227 (1957).

strongly suggested that dasycarpidol is dihydrodasycarpidone (VII), as did the similarity in mass spectrometric fragmentation pattern to that of dihydrouleine.[11] The NMR spectrum was completely consistent with this structure. In addition to the usual features (see above)[2b,2c] the spectrum showed no exocyclic methylene proton absorption, but did exhibit a doublet (5·1 ppm, J = 6 c/s) corresponding to a benzylic proton situated on a carbon atom (C-1) which also carries an oxygen substituent. The coupling constant (C-1 hydrogen coupling with C-2 hydrogen) necessitates a dihedral angle of ca. 30° between the two C–hydrogen bonds a restriction which according to Dreiding models seems only possible when the hydroxyl group at C-1 is equatorial leading to the expression XVII for the relative stereochemistry of the molecule (alternative dihedral angle is 90°). The conversion of dasycarpidol, by mild oxidation with chromium trioxide pyridine, to dasycarpidone (V) settled the constitution as VII.



XVII R = OH
XVIII R = CH_2OH

1,13-*Dihydro*-13-*hydroxyuleine* (VIII)

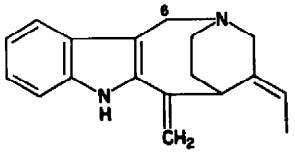
This alkaloid had a mol. wt. of 284, eighteen mass units higher than that of uleine (I). Since the UV absorption is indolic, the conjugation provided by the 1–13 double bond of uleine is absent. These facts suggested that the molecule is a 1–13 hydrated uleine. Further evidence in favour of this suggestion was derived from the NMR spectrum which displayed two active hydrogen signals (eliminated by exchange with heavy water), one of the indolic N–hydrogen and the second an alcoholic O–hydrogen (3·40 ppm). That the hydroxyl group is situated at C-13 and not at C-1 was shown by the absence of a singlet methyl signal (C-13) and the presence of signals for three protons in the region 3·6–4·1 ppm (one at C-4 and two at C-13). The partial synthesis of 1,13-dihydro-13-hydroxyuleine (VIII) from uleine (I) confirmed the identity of the skeletons. The addition of the elements of water to uleine in the desired orientation was achieved by hydroboration.[15] Treatment of uleine boron trifluoride salt with an equivalent of sodium borohydride and two equivalents of boron trifluoride-etherate with subsequent peroxidic oxidation led to VIII in 79% yield. In addition to establishing the gross structure of the alkaloid, this highly stereospecific synthesis suggests the relative configuration XVIII for the molecule, since the reagent would be expected to attack uleine from the β-face (opposite side to the piperidine ring) of the molecule.[16] Since the rotation of the synthetic material was the same as that of the naturally occurring alcohol (VIII) the absolute configuration of the molecule is the same as that of uleine.

Apparicine (IX)[7]

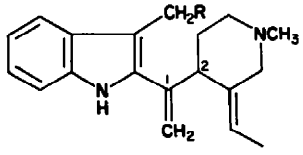
This interesting base is the first example of a new type of indole alkaloid. It possesses the same chromophoric system as uleine but unlike uleine contains an ethylidene side chain. These features, easily distinguished in the NMR spectrum of apparicine, together with the two mass unit lower mol. wt. (264) and a general similarity of

[15] G. Zweifel and H. C. Brown, *Organic Reactions*, Vol. 13; Chap. 1. J. Wiley, New York (1963).
[16] G. Zweifel and H. C. Brown, *J. Amer. Chem. Soc.* **86**, 393 (1964).

fragmentation pattern to that of uleine, suggested a structure for the molecule rather similar to uleine. The NMR spectrum differed from that of uleine by lacking an N–methyl singlet, having instead a two proton AB quartet corresponding to the two hydrogens on C-6. Clear-cut structural information was obtained[7] by breaking the C-6–N_b bond of apparicine methiodide by nucleophilic attack (methoxide or hydride) at C-6 with displacement of N_b^+. The ring opened compounds (XIX, R = MeO or H) so obtained were subjected[7] to detailed NMR analysis using decoupling. This analysis, together with the simple mass spectral fragmentation of these molecules (XIX) (fission between C-1 and C-2 accompanied by hydrogen transfer) and the identification of pyridine-3,4-dicarboxylic acid from dehydrogenation and oxidation of apparicine, led[7] to the structure IX for the alkaloid.



IX



XIX

Aspidodasycarpine (XI)

The structure determination of perhaps the most interesting base of the group, aspidodasycarpine (XI), has been briefly described.[9] The alkaloid possesses a molecular formula $C_{21}H_{26}N_2O_4$ as indicated by combustion analysis and mass spectrometry (see Fig. 1, which also lists plausible assignments to principal diagnostic peaks). Its UV spectrum is typical of a dihydroindole unsubstituted on the benzene ring, a chromophore confirmed by the presence of signals for four aromatic protons in its NMR spectrum. Bands for N–hydrogen and/or O–hydrogen (2·90 μ) and ester (5·76 μ) appear in the IR spectrum and the ester carbonyl function could be more particularly identified as due to carbomethoxyl by the presence of a three proton methoxy singlet (3·72 ppm) in the NMR spectrum of the base. This latter technique also showed the existence of an ethylidene side chain, by the presence of the characteristic three proton doublet (1·69 ppm, J = 7 c/s) and one proton quartet (5·5 ppm, J = 7 c/s). The nature of the two remaining oxygen atoms and the substitution of the aliphatic nitrogen atom, N_b, could be inferred from an examination of aspidodasycarpine-N_b,O-diacetate (XX) prepared from the alkaloid by treatment with acetic anhydride–pyridine. This diacetate showed IR absorption due to amide carbonyl (6·18 μ) as well as ester absorption and its NMR spectrum now had two acetyl three-hydrogen singlets (2·16 and 1·92 ppm). The diacetate was non-basic and did not react with methyl iodide at room temperature. These facts, combined with the unchanged UV absorption (N–acetyl not involved in the chromophoric system), necessitate that the basic nitrogen, N_b, of the alkaloid itself be secondary and that the third oxygen atom be alcoholic. The remaining oxygen must by inference be ethereal, a conclusion supported by data cited later in this paper.

More specific information on the environment of the alcoholic hydroxyl function was obtained by base catalysed retroaldolization of the diacetate (XX) to desformo-aspidodasycarpine-N_b-acetate (XXI), a compound still possessing indoline UV absorption, ester and amide IR bands, but only one acetyl methyl singlet (2·19 ppm) in its

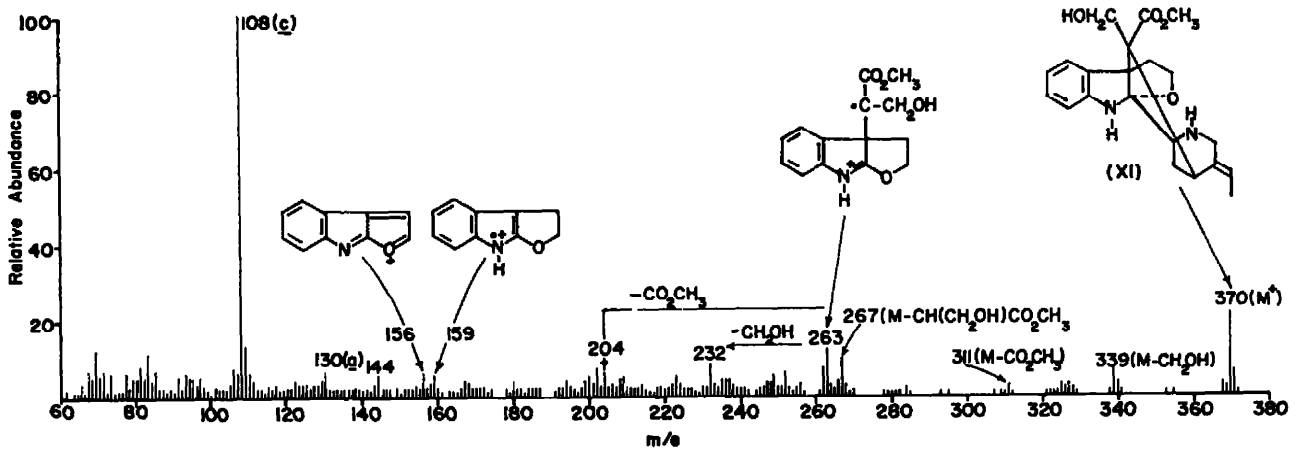
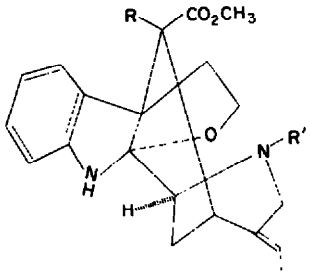
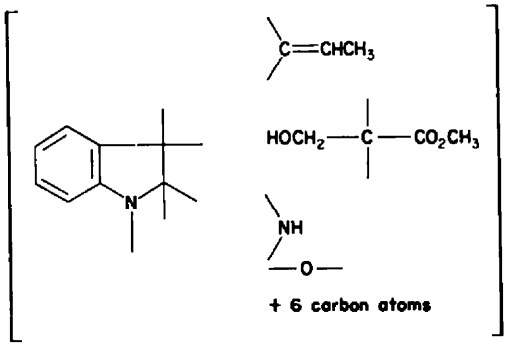


FIG. 1. Mass spectrum of aspidodasycarpine (XI).



	R	R′
XI	CH_2OH	H
XX	CH_2OCOCH_3	$COCH_3$
XXI	H	$COCH_3$
XXIII	H	CH_2OH
XXIV	H	H
XXVII	CH_2OH	CH_3

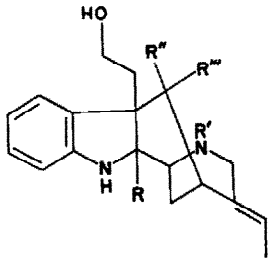
NMR spectrum. The mol. wt., by mass spectrometry, (382) indicated loss of $CH_3CO{\cdot}OCH_2{\cdot}$ and suggested the presence, in aspidodasycarpine-N_b,O-diacetate of the grouping $CH_3CO{\cdot}OCH_2{\cdot}C{\cdot}CO_2CH_3$. These data can be summarized in expression XXII for the alkaloid.



XXII

Information on the nature of the ring containing N_b and definitive evidence as to the ethereal nature of the fourth oxygen atom and its site of attachment could be obtained from N_b-hydroxymethyl desformoaspidodasycarpine (XXIII) and its reduction products. When aspidodasycarpine was treated with potassium hydroxide in 80% methanol at room temperature desformoaspidodasycarpine (XXIV) was produced which could be converted to the previously obtained desformoaspidodasycarpine-N_b-acetate (XXI) by acetylation. However, when desformoaspidodasycarpine (XXIV) was treated with alkali and formaldehyde in 66% aqueous methanol at room temperature *or* aspidodasycarpine itself treated with alkali in 66% aqueous methanol, the highly crystalline methylol (XXIII) separated in high yield from the reaction mixture. The methylol was easily reconverted to desformoaspidodasycarpine by treatment with acetic acid. This easy reversal and the physical properties of this somewhat surprising compound are consistent with the structure (XXIII) assigned to it. Most importantly a pair of doublets (4·15 and 4·40 ppm, J = 9 c/s) representing the signals due to the $N_b{\cdot}CH_2OH$ grouping appear in the NMR spectrum of the compound as do signals due to four aromatic protons, an ethylidene and a methoxyl function. Reduction of the methylol (XXIII) with LAH gave an N_b-methyl derivative (XXV), the N-methyl grouping being identified as aliphatic by the chemical shift of the three-hydrogen singlet (2·32 ppm).

From the mol. wt. of the N-methyl reduction product (XXV) (328, corresponding to $C_{20}H_{28}N_2O_2$) it followed that as well as reducing the NCH_2OH group and the ester



	R	R′	R″	R‴
XXV	H	CH_3	H	CH_2OH
XXVI	D	CDH_2	H	CD_2OH
XXIX	H	$COCH_3$	$CH_3CO_2CH_2$	CO_2CH_3
XXXVII	H	CH_2CH_3	CH_2OH	CH_2OH
XXXVIII	D	CD_2CH_3	H	CD_2OH

function, the ether linkage had also been cleaved. This could be neatly confirmed by reduction of the methylol with LAD to give XXVI the mass spectrometrically determined mol. wt. of which showed the presence of four deuterium atoms ($C_{20}H_{24}D_4N_2O_2$). It thus seemed likely that the methylol and hence the alkaloid itself contained a carbinolamine ether system, a grouping well known to be split with LAH.[17,18] That this is indeed the situation was deduced from the shift of the indole peak,[19] *m/e* 130 (*a*) to *m/e* 131 (*b*) in the mass spectrum of the LAD product (XXVI), a technique already used with pseudoakuammigine[18,20a] and picraline[18,21] to establish the point of connection of such an ether linkage. The indole peaks also indicate that the aromatic nitrogen is unsubstituted in aspidodasycarpine.

With the establishment of a carbinolamine ether system at N_a, it became clear why this nitrogen was not acetylated during the treatment of the alkaloid with acetic anhydride–pyridine. The UV absorption of the base in strong acid (benzenoid) however was inconsistent with the spectral behaviour of carbinolamine ether systems as exemplified by pseudoakuammigine[20b] and picraline.[21] Precedent for such behaviour in a carbinolamine ether was found in a study[20c] of simple systems containing such a grouping, some of which were found to protonate partly or completely on the nitrogen rather than on the oxygen atom.

The base peaks of the mass spectra of aspidodasycarpine (XI) (Fig. 1) or desformoaspidodasycarpine (XXIV) occur at *m/e* 108 and indicate that the basic nitrogen atom forms part of a piperidine ring with a two-carbon substituent, which by analogy with a large number of indole alkaloids[19] might be expected to be the ethylidene group, the structure of the *m/e* 108 ion then being the stable pyridinium species *c*. Consistent with this assignment is the observation that the base peak in the spectra of the N_b–methyl reduction product (XXV) (or of N-methyl aspidodasycarpine, XXVII) occurs at *m/e* 122 (*d*) and at *m/e* 123 (*e*) in the LAD reduction product (XXVI). Similarly, in the spectra of the acetates XX and XXI, two important peaks appear at *m/e* 150 (*f*) and *m/e* 108 (*c*), the latter being formed from the former by the familiar expulsion of

[17] K. S. Brown, H. Budzikiewicz and C. Djerassi, *Tetrahedron Letters* 1731 (1963); C. Djerassi, L. D. Antonnaccio, H. Budzikiewicz, J. M. Wilson and B. Gilbert, *Ibid.* 1001 (1962); M. P. Cava, S. K. Talapatra, K. Namura, J. A. Weisbach, B. Douglas and E. C. Shoop, *Chem. Ind.* 1242 (1963).

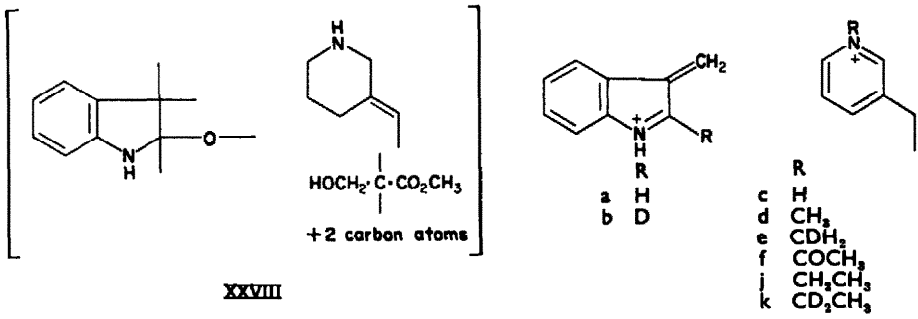
[18] L. Olivier, J. Levy, J. LeMen, M.-M. Janot, C. Djerassi, H. Budzikiewicz, J. M. Wilson and L. J. Durham, *Bull. Chem. Soc. Fr.* 646 (1963).

[19] H. Budzikiewicz, C. Djerassi and D. H. Williams, *Structure Elucidation of Natural Products by Mass Spectrometry*, Vol. 1 *Alkaloids*, Holden-Day, San Francisco (1964).

[20a] J. Levy, J. LeMen and M.-M. Janot, *Bull. Chem. Soc. Fr.* 1658 (1961); A. Z. Britten, P. N Edwards, J. A. Joule, G. F. Smith and G. Spiteller, *Chem. Ind.* 1120 (1963); [b] J. A. Joule and G. F. Smith, *J. Chem. Soc.* 312 (1962); [c] W. Bardsley and G. F. Smith, unpublished work.

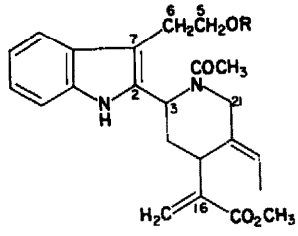
[21] A. Z. Britten, G. F. Smith and G. Spiteller, *Chem. Ind.* 1492 (1964).

ketene,[22] while the N_b–ethyl (XXXVII) N_b–d_2–ethyl (XXXVIII) derivatives show *m/e* 136 (*j*) respectively *m/e* 138 (*k*) peaks. Expansion of the part structure for aspidodasycarpine to XXVIII is now possible.



XXVIII

Treatment of aspidodasycarpine-N,O-diacetate (XX) with zinc and hydrochloric acid gave, in addition to the anticipated dihydroindolinic compound (XXIX) (N·C·O· → N·C·H· + HO·), an indole (UV), secoaspidodasycarpine-N-acetate ($C_{23}H_{28}N_2O_4$) to which the structure XXX is attributed.

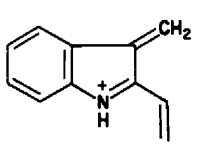


XXX R = H
XXXI R = $COCH_3$

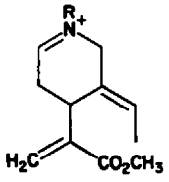
The NMR spectrum of the compound confirms the chromophoric system as a 2,3-disubstituted indole (four aromatic protons and an indolic N–hydrogen signal (8·55 ppm)), and the presence of ethylidene, *one* acetyl methyl (2·02 ppm) and ester methoxyl (3·72 ppm) groups. The absence of the original acetoxy function (part of $CH_3CO·O·CH_2·C·CO_2CH_3$) together with the presence of two one-hydrogen singlets (6·20 and 5·63 ppm) in the NMR spectrum and a conjugated ester carbonyl band (5·85 μ) in its IR spectrum led to the conclusion that the molecule contains $CH_2{=}C·CO_2CH_3$. The presence of a $CH_2·CH_2OH$ chain attached to the aromatic system was suggested by the NMR two-hydrogen signal at 3·0 ppm (C-6, benzylic protons) and a two-hydrogen triplet (3·85 ppm, J = 7 c/s) for the C-5 protons, the latter being shifted to 4·30 ppm in the diacetate (XXXI). The mass spectra of secoaspidodasycarpine-N-acetate (XXX) and secoaspidodasycarpine-N O-diacetate (XXXI) showed peaks corresponding to loss of CH_2OH ($OCOCH_3$) (*m/e* 365, fission between C-5 and C-6) and loss of the entire CH_2CH_2OH ($OCOCH_3$) side chains (*m/e* 351, fission between C-3 and C-6). This oxygenated side chain must, barring rearrangements, represent the other end of the ether linkage already established as being attached to C-2 (in XXVIII). Strong indications as to the sites of attachment of the indole system and the unsaturated ester moiety to the piperidine nucleus can be gained

[22] H. Budzikiewicz, C. Djerassi and D. H. Williams, *Interpretation of Mass Spectra of Organic Compounds*, Holden-Day, San Francisco (1964).

from the NMR spectrum of XXX. A one-hydrogen multiplet (5·0 ppm) corresponds to the C-3 proton,[23,24] while a one-hydrogen multiplet (2·20 ppm) represents the doubly allylic hydrogen at C-15. Further, the A part of the AB system caused by the two isolated hydrogen atoms on C-21 (4·49 ppm, J = 13 c/s) can be clearly seen, the B part being obscured by the methoxyl signal. Finally peaks in the mass spectrum of this interesting and significant product at *m/e* 156 (*g*), 194 (*h*) and 236 (*i*) find ready explanation in the proposed structure XXX, the latter two again being related by expulsion of ketene.[22] The structures of the ions *h* and *i* were confirmed by four-mass unit shifts (to *m/e* 198 and 240) in the tetrahydro catalytic reduction product of secoaspidodasycarpine-N-acetate.

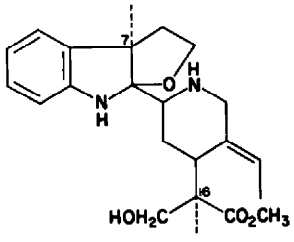


g



	R
h	H
i	$COCH_3$

On the assumption that no fundamental skeletal rearrangement occurs during the production of the indole (XXX) (the mechanism of the formation of this compound will be discussed later) the part structure for aspidodasycarpine can be extended to XXXII in which necessarily C-7 and C-16 must be joined to arrive at the structure XI for aspidodasycarpine.



XXXII

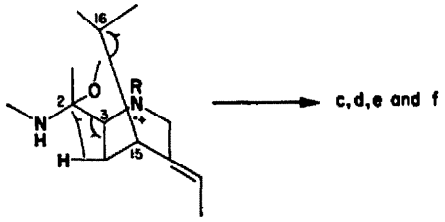
The facile production of piperidine fragments from aspidodasycarpine and closely related derivatives are nicely explained by the processes shown in XXXIII, fission of the 2–3 bond with hydrogen transfer followed by homolysis of the then double allylically activated 15–16 bond to give fragments *c*, *d*, *e* and *f*.

Chemical confirmation for the structure of XI for aspidodasycarpine was obtained by interconversion with picraline (XXXIV), whose structure[18,21] and absolute configuration[25] have recently been established. The highly crystalline N_b–hydroxymethyl desformoaspidodasycarpine (XXIII) was shown to be identical, in all respects, including rotation, with the N_b–hydroxymethyl derivative of the borohydride reduction

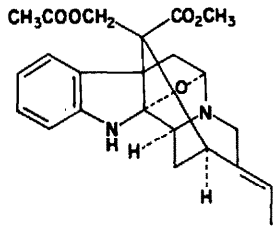
[23] E. Wenkert, B. Eickberg and C. L. Leicht, *J. Amer. Chem. Soc.* **83**, 5037 (1961).

[24] M. Uskokovic, H. Bruderer, C. von Planta, T. Williams and A. Brossi, *J. Amer. Chem. Soc.* **86**, 3364 (1964).

[25] L. Olivier, J. Levy, J. LeMen, M.-M. Janot, H. Budzikiewicz and C. Djerassi, *Bull. Chem. Soc. Fr.* 868 (1965).



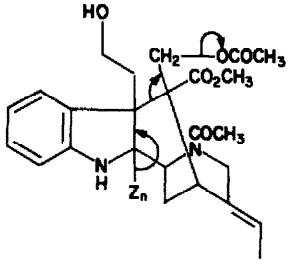
XXXIII



XXXIV

product[25] of picraline (XXXIV). The reduction of picraline (to desformoaspidodasycarpine (XXIV)) involves base catalysed hydrolysis and retroaldol loss of the $CH_3CO{\cdot}OCH_2$ grouping and reduction of the oxidation state of the C-5 carbon atom to that of an alcohol with fission of the C-5–N_b bond.

The mechanism of formation of secoaspidodasycarpine-N_b-acetate (XXX) appears to represent an interesting example of a fragmentation reaction.[26] The intermediate (XXXV) derived by complexing of the zinc with the C-2 carbon atom of an acid-catalysed, ring open form of the starting material, could either remain as such, giving rise to dihydroaspidodasycarpine-N_b,O-diacetate (XXIX) on hydrolytic work up, or undergo the fragmentation indicated in XXXV. The change is both the nitrogen analog and the ethylogue of the well known α-fission which occurs in Clemmensen reductions.[27] The proposed scheme is consistent with the isolation, albeit in poor yield of secoaspidodasycarpine itself from zinc–hydrochloric acid treatment of the alkaloid. A similar fragmentation has been observed during the zinc–hydrochloric acid reduction of picraline[28] and the phenomenon in general is under study.[29] The alternate mechanism[9] involving attack by zinc at the acetate or carbomethoxyl carbonyl oxygen is, of course, not excluded.



XXXV

Polyneuridine (or akuammidine) aldehyde (XII)

The proposed structure of this compound is based on its indolic UV absorption and characteristic fragmentation pattern[19] which is completely consistent with the structure XII, where the orientations of the aldehyde and ester groups are unspecified.

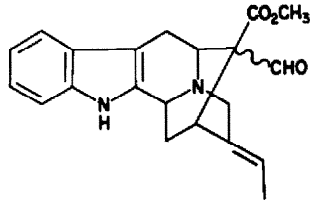
[26] C. A. Grob, *Experientia* **13**, 126 (1957), *Bull. Chem. Soc. Fr.* 1360 (1960); *Gazz. Chim. Ital.* 902 (1962).

[27] D. Staschewski, *Angew. Chem.* **71**, 726 (1959); J. H. Brewster, *J. Amer. Chem. Soc.* **76**, 6361, 6364 (1954).

[28] G. F. Smith and J. A. Joule, unpublished work.

[29] J. A. Joule and A. Jackson, unpublished work.

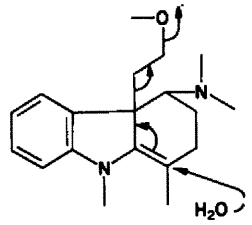
The minute quantity of the alkaloid isolated did not permit further chemical proof of the proposed structure.



XII

Biogenesis

The co-occurrence of uleine (I) type alkaloids with a base (aspidodasycarpine) in which the tryptamine bridge–N_b bond has been broken is interesting in view of Wenkert's suggestion[30] that the uleine type molecule is formed from an indole moiety which does not yet contain such a two atom bridge. Although several α-indole alkaloids are known which are oxygenated α to N_b on the tryptamine bridge and sometimes (e.g. aspidodasycarpine) have the N_b bond broken, there are no examples of β-indole alkaloids (akuammicine or condylocarpine types) which possess this feature. It may well be that should such a biogenetic oxidation take place, the molecule fragments, with loss of the bridge to give uleine types by some scheme[31] such as that illustrated in XXXVI.



XXXVI

EXPERIMENTAL

M.ps were determined on a Kofler micro-hot-stage and are not corrected. NMR spectra were run on a Varian A60 spectrometer by Dr. L. J. Durham in $CDCl_3$ with tetramethylsilane as internal standard Mass. spectra were determined by Drs. H. Budzikiewicz, J. M. Wilson and M. Ohashi, using a CEC-103C mass spectrometer equipped with heated all-glass inlet system or a direct inlet system.[32] Mass spectra are identified as h (= heated) or d (= direct), depending upon which system was employed. Thin-layer chromatography (TLC) was performed on silica gel "G" using ethyl acetate–benzene–ethanol (4:4:2) and bands or spots detected by spraying with 2% $Ce_2(SO_4)_3$ in 1 M H_2SO_4 or by their fluorescence under UV light. Rotations (in $CHCl_3$) and UV spectra were carried out by Mrs. Aguilar and microanalysis were determined by Messrs. E. Meier and J. Consul.

Isolation of alkaloids

The total methanolic extract (800 g) of the bark in glacial acetic acid (600 ml) was added with vigorous stirring to water (4 l.). The slurry was filtered after stirring for 6 hr. The filtrate was successively washed with pet. ether (no material), extracted with benzene (3 × 500 ml), fraction A (1·9 g),

[30] E. Wenkert, *J. Amer. Chem. Soc.* **84**, 98 (1962).

[31] Ethylogue of scheme proposed by: W. I. Taylor, *Proc. Chem. Soc.* 247 (1962).

[32] J. F. Lynch, J. M. Wilson, H. Budzikiewicz and C. Djerassi, *Experientia* **19**, 211 (1963).

$CHCl_3$ (3 × 500 ml) after adjusting the pH to 7, fraction B (6·1 g) and finally $CHCl_3$ (3 × 500 ml) at pH 11, fraction C (3·4 g).

Fraction A. Filtration through alumina (act. I) in ether yielded the only crystalline alkaloid obtainable from this fraction, *uleine* (I), m.p. 75–98° (ex MeOH), $[\alpha]_D^{25}$ +16·5° (*c*, 0·91); IR and UV spectra identical with those of a genuine sample; 0·84 (3H triplet J = 6 c/s, $C\underline{H}_3CH_2$), 2·29 (3H singlet, $C\underline{H}_3N$), 4·11 (1H doublet J = 3 c/s, β-indole $C\underline{H}$·N), 4·98 and 5·28 (2 × 1H singlets, $C\underline{H}_2$=), 7·00–7·65 (4H, $\underline{H}$Ar), 8·58–8·75 ppm (1H, $\underline{H}$N); for mass spectrum see Ref. 2*c*.

Fraction B. Chromatography over alumina (act. IV) gave 10 cuts from benzene to ether. From all fractions uleine (8·3 g) was crystallized separately. The filtrates from the first five fractions were evaporated and the residues crystallized from acetone to give (+)-*appariceine* (IX) (400 mg), m.p. 192–194°, $[\alpha]_D^{27}$ +176·0° (*c*, 2·16). (Found: C, 81·48; H, 7·50; N, 10·71; CH_3O, 0·00; CH_3C, 5·21, mol. wt. by mass spectrometry, 264; calc. for $C_{18}H_{20}N_2$: C, 81·78; H, 7·63; N, 10·60; CH_3O, 0·00; 1 × CH_3C, 5·68%, mol. wt., 264); λ_{max} ($CHCl_3$) 2·90 μ; λ_{max} (EtOH) 303 (4·67), λ_{infl} 312, 230 (4·61, 4·70), λ_{min} 265 mμ (3·79), unchanged in acid and base; 1·47 (3H doublet J = 8 c/s, $C\underline{H}_3CH_2$), 4·19 and 4·56 (2H AB quartet J = 18 c/s, β-indole-$C\underline{H}_2$N), 5·24 and 5·37 (2 × 1H singlets, $C\underline{H}_2$=), 7·90–8·20 ppm (1H, $\underline{H}$N), for mass spectrum see Ref. 7.

By thin-layer chromatography of the second fraction of the above chromatography, there was isolated N_b-*methyltetrahydroellipticine* (III) (20 mg; zone at R_f 0·5), crystallized from acetone, m.p. 198–200° dec; IR and UV spectra identical with those of an authentic sample; mass spectrum (h) showed a mol. ion at 264 (53%) as well as *m/e* 263 (100), 249 (32), 233 (6), 221 (69), 206 (11), 205 (10), 204 (12), 191 (6), 133 (24), 132 (18), 124 (22), 123 (14), and 117 (11).

By thin-layer chromatography of the third fraction of the alumina chromatography, *dasycarpidone* (V; 100 mg), amorphous, R_f 0·5, was isolated, $[\alpha]_D^{26}$ +64·7° (*c*, 1·02). (Found: mol. wt. by mass spectrometry: 268; calc. for $C_{17}H_{20}N_2O$: mol. wt., 268); λ_{max} ($CHCl_3$) 2·93, 6·10 μ; λ_{max} (EtOH) 316, 237 (4·29, 4·15), λ_{min} 265, 223 mμ (2·20, 3·07), unchanged in acid and base; 0·88 (3H triplet J = 7 c/s, $C\underline{H}_3CH_2$), 2·34 (3H singlet, $C\underline{H}_3N$), 4·25 (1H doublet J = 2·5 c/s, β-indole-$C\underline{H}$—N), 7·00–7·90 (4H, $\underline{H}$Ar), 10·20–10·50 ppm (1H, $\underline{H}$N), for mass spectrum see Ref. 2*c*.

Thin-layer chromatography of fraction 7 of the original alumina chromatography allowed separation of four alkaloids. The zone at R_f 0·65 was crystallized from ether to give *polyneuridine* (or *akuammidine*) *aldehyde* (XII; 2 mg), m.p. 231–233°. (Found: mol. wt. by mass spectrometry: ⁶350; calc. for $C_{21}H_{22}N_2O_3$: mol. wt., 350); λ_{max} (EtOH) 290, 282, 226 (3·65, 3·67, 4·50), λ_{min} 288, 252 mμ (3·63, 3·46); mass spectrum (h) showed a mol. ion at 350 (70%) as well as *m/e* 349 (53), 321 (5), 319 (5), 306 (7), 291 (12), 281 (9), 263 (17), 246 (14), 223 (30), 207 (33), 197 (23), 184 (32), 183 (48), 182 (86), 169 (96), 168 (100), 157 (92), 156 (67), 143 (28), 130 (36), and 128 (40).

The zone at R_f 0·5 gave (+)-*guatambuine* (ex acetone), m.p. 236–238° dec, $[\alpha]_D^{25}$ +88° (*c*, 0·67, dioxan), IR and UV spectra identical with those of an authentic sample, mass spectrum (h) showed a mol. ion at 264 (10%) as well as *m/e* 263 (25), 249 (500), 247 (33), 233 (22), 221 (21), 206 (23), 205 (26), 204 (37), 167 (7), 132 (21), 124 (38) and 117 (36).

The zone at R_f 0·4 was crystallized from $CHCl_3$ to give *des*-N-*methyldasycarpidone* (VI; 30 mg), m.p. 208–210°. (Found: mol. wt. by mass spectrometry: 254; calc. for $C_{16}H_{18}N_2O$: mol. wt., 254), λ_{max} ($CHCl_3$) 2·90, 3·05, 6·07 μ; λ_{max} (EtOH) 317, 238 mμ (4·23, 4·15); mass spectrum (h) showed a mol. ion at 254 (53%) as well as *m/e* 225 (21), 211 (45), 197 (97), 184 (65), 169 (100), 157 (29) and 130 (29).

Concentration of fraction 8 *in vacuo* gave a crystalline alkaloid *dehydro-des*-N-*methyluleine* (X; 3 mg), m.p. 220°. (Found: mol. wt. by mass spectrometry: 250; calc. for $C_{17}H_{18}N_2$: mol. wt., 250); λ_{max} (nujol) 3·5, 6·12, 6·20, 11·6, 12·2, 12·3, 13·4 μ, λ_{max} (EtOH) 310, 303 mμ (4·21, 4·21); mass spectrum (h) showed a mol. ion at 250 (86%) as well as *m/e* 230 (67), 220 (77), 218 (57), 205 (67), 204 (77), 181 (100) and 180 (51).

Fraction C. The crude material from this extract was chromatographed over alumina (act. I) with $CHCl_3$ up to $CHCl_3$–MeOH (9:1) giving 5 cuts. This third cut was rechromatographed over alumina (act. I), elution with $CHCl_3$–MeOH (98:2) giving 60 fractions. Fractions 15–53 were evaporated and separated by TLC (C_6H_6—Me_2CO—Et_2NH, 80:15:5) to give 1,13-*dihydro*-13-*hydroxyuleine* (VIII; 35 mg; R_f 0·27), amorphous, $[\alpha]_D^{27}$ −96° (*c*, 0·25, EtOH). (Found: mol. wt. by mass spectrometry: 284; calc. for $C_{18}H_{24}N_2O$: mol. wt., 284); λ_{max} ($CHCl_3$) 2·90, 9·90 μ; λ_{max} (EtOH) 289, 282, 211 mμ (3·87, 3·91, 4·56); 0·90 (3H triplet, $C\underline{H}_3CH_2$), 2·34 (3H singlet, $C\underline{H}_3N$), 3·40 (1H), 3·8–4·1 (3H, β-indole-$C\underline{H}$—N and $C\underline{H}_3O$), 6·9–7·8 (4H, $\underline{H}$Ar), 9·15 ppm (1H, $\underline{H}$N); mass spectrum

(h) showed a mol. ion at 284 (2%) as well as *m/e* 266 (100), 237 (9), 230 (10), 223 (13), 209 (10), 194 (31), 180 (16), 167 (9) and 143 (12).

Rechromatography of cut 4 from the first chromatography over alumina (act. II) with $CHCl_3$—MeOH (98:2) gave, in fractions 12–15, *des*-N-*methyluleine* (IV; 125 mg), m.p. 143–145° (ex $CHCl_3$), $[\alpha]_D^{26}$ −20° (*c*, 1·18 EtOH). (Found: mol. wt. by mass spectrometry: 252; calc. for $C_{17}H_{20}N_2$: mol. wt., 252), λ_{max} ($CHCl_3$) 2·90, 6·12 μ; λ_{max} (EtOH) 312, 305 mμ (4·23, 4·24); mass spectrum (d) showed a mol. ion at 252 (100%) as well as *m/e* 237 (15), 223 (49), 209 (24), 195 (33), 194 (30), 180 (26) and 167 (11).

Fractions 26–32 gave, from $CHCl_3$, *dasycarpidol* (VII; 210 mg), m.p. 118–122°, $[\alpha]_D^{27}$ −54° (*c*, 1·03, EtOH). (Found: mol. wt. by mass spectrometry: 270; calc. for $C_{17}H_{22}N_2O$: mol. wt., 270); λ_{max} (KBr) 2·92, 6·15 μ; λ_{max} (EtOH) 290, 282, 220 mμ (3·81, 3·89, 4·54); 0·89 (3H triplet J = 5·5 c/s CH_3CH_2), 2·21 (3H singlet, CH_3N), 4·05 (1H doublet, J = 2 c/s, β-indole-CH—N), 5·1 (1H doublet J = 6 c/s, α-indole-CH—O), 6·9–7·7 ppm (4H, HAr) (in deuterioacetone); mass spectrum (d) showed a mol. ion at 270 (21%) as well as *m/e* 252 (19), 223 (29), 213 (19), 209 (27), 195 (100), 187 (38), 183 (22), 180 (42), 168 (24), 167 (27) and 157 (36).

Cut 5 was rechromatographed over alumina (act. II) with $CHCl_3$—MeOH (99:1) as eluate, giving, from fractions 9–21, *aspidodasycarpine* (XI; 150 mg), m.p. 207–209° (ex Me_2CO), $[\alpha]_D^{25}$ −101° (*c*, 1·42). (Found: C, 67·7, H, 7·13, N, 7·67%, mol. wt, by mass spectrometry: 370; calc. for $C_{21}H_{26}N_2O_4$: C, 68·1, H, 7·07, N, 7·56, mol. wt., 370); λ_{max} ($CHCl_3$) 2·90, 5·76, 6·25, 9·70 μ; λ_{max} (EtOH) 297, 240, 205 mμ (3·63, 3·96, 4·16); 1·69 (3H doublet J = 7 c/s, CH_3CH=), 3·72 (3H singlet, CH_3O_2C), 5·5 (1H quarter J = 7 c/s, CH_3CH=), 6·5–7·5 ppm (4H, HAr); mass spectrum (d) showed a mol. ion at 370 (21%) as well as *m/e* 339 (6), 311 (3), 267 (10), 263 (12), 232 (8), 204 (6), 159 (5), 156 (6), 144 (5), 130 (6), and 108 (100). Structural assignments for these peaks are given in Fig. 1.

Reaction of des-N-*methyluleine* (IV) *with methyl iodide*

The alkaloid (IV; 30 mg) was treated with MeI (0·5 ml) in refluxing acetone–benzene (1:1; 6 ml) for 30 min to give a crystalline material, m.p. 196–198° (10 mg), identified as uleine methiodide by IR (KBr pellet) comparison. The evaporated filtrate was treated with NaOH aq and extracted with $CHCl_3$. Evaporation of the dried $CHCl_3$-extract gave a residue from which uleine (I; 11 mg) could be crystallized in MeOH, m.p. 70–75°, $[\alpha]_D^{26}$ +6·7 (*c*, 0·6), identified by IR, mass spectrometric and TLC comparisons.

Reaction of des-N-*methyluleine* (IV) *with acetic anhydride–pyridine*

Treatment of IV with acetic anhydride–pyridine (1:1) at room temp overnight with subsequent removal of solvents *in vacuo* and partition of the residue between ether and K_2CO_3 aq, gave, from the organic layer, *des*-N-*methyl*-N-*acetyluleine* (XIII), m.p. 214–215°. (Found: mol. wt. by mass spectrometry, 294; calc. for $C_{19}H_{22}N_2O$: mol. wt., 294); λ_{max} ($CHCl_3$) 2·87, 6·17 μ; λ_{max} (EtOH) 313, 308 (4·29, 4·29), λ^{infl} 237 (4·10), λ_{min} 265 mμ (3·77); mass spectrum (h) showed a mol. ion at 294 (100%) as well as *m/e* 279 (2), 265 (18), 251 (22), 235 (18), 223 (39), 222 (50), 208 (42), 206 (44), 194 (54), 180 (51) and 167 (20).

Reduction of des-N-*methyl*-N-*acetyluleine* (XIII)

The acetate (15 mg) was reduced with excess LAH in refluxing tetrahydrofuran overnight. Excess reagent was decomposed with water and the mixture filtered and the filtrate evaporated. The residue was partitioned between 2 N HCl and ether, the aqueous layer basified (K_2CO_3) and extracted with ether to give a glass (9 mg), further purified by preparative TLC to give *des*-N-*methyl*-N-*ethyluleine* (XIV; 4 mg) as a colourless glass. (Found: mol. wt. by mass spectrometry: 280; calc. for $C_{19}H_{24}N_2$: mol. wt., 280); mass spectrum (h) showed a mol. ion at 280 (93%) as well as *m/e* 265 (42), 251 (65), 236 (41), 223 (38), 209 (100), 194 (89), 180 (81), 167 (31), 140 (15), and 118 (26).

The above product was reacted with MeI in benzene at room temp for 0·5 hr. The solvents were removed and the residue crystallized from MeOH to give *des*-N-*methyl*-N-*ethyluleine methiodide*, m.p. 190–191°, mixture m.p. with *uleine ethiodide* m.p. 198–201° (ex MeOH). (Found: C, 55·98, H, 6·20, N, 6·91; calc. for $C_{20}H_{27}N_2I \cdot \frac{1}{2}H_2O$: C, 55·80, H, 6·28, N, 6·51%) 187–201°.

Reaction of des-N-*methyldasycarpidone* (VI) *with methyl iodide*

The ketone (VI; 7 mg) in benzene–acetone (1:1; 2 ml) was heated with MeI (0·3 ml) under reflux for 1 hr. The solvents were removed under red. press. and the residue treated with 0·1 N

NaOH and ether. The ethereal extract was evaporated to give dasycarpidone (V; 2 mg), identified by mass spectrum (h) and thin layer mobility comparison with the naturally occurring base.

Ozonolysis of uleine (I)

The alkaloid (350 mg) in ethyl acetate–MeOH (20 ml–1 ml) was treated with 4 portions of ethyl acetate (20 ml) into which ozonized oxygen had been passed for 5 min (6 psi O_2 press., flow rate 0·15, Model T4 Wellsbach ozonator). Acetic acid (5 ml) and Zn (3 g) were added and the mixture allowed to come to room temp whilst stirring (1·5 hr). After filtration, basification (K_2CO_3) and ether extraction a gum (73 mg) was obtained. An ethereal solution of this material was filtered through a column of alumina (act. I) and the eluate evaporated to give dasycarpidone (47 mg), $[\alpha]_D^{27}$ +61·2° (*c*, 0·91), identical with the naturally occurring material by thin-layer, IR and mass spectrometric comparison.

Oxidation of dasycarpidol (VII)

To the alcohol (30 mg) in pyridine (2 ml) was added, with stirring, CrO_3 (60 mg) in pyridine (1 ml). After 2 min the solution was poured into water. Extraction with $CHCl_3$ and chromatography on alumina (act. II), elution with $CHCl_3$, gave a colourless solid (10 mg), identical by IR, UV, mass spectral and thin-layer comparison with V.

Hydroboration of uleine (I)

To uleine boron trifluoride salt (170 mg; m.p. 205° dec, prepared by reaction of uleine with BF_3–etherate in ether) in suspension in tetrahydrofuran (3 ml) was added $NaBH_4$ (210 mg) and BF_3–etherate (0·2 ml) with vigorous stirring and the resulting mixture stirred overnight. NaOH (30%, 1 ml) and H_2O_2 (30%, 2 ml) were added and the mixture refluxed with stirring for 5 hr. Purification by alumina chromatography (act. II, elution with $CHCl_3$—MeOH (99:1)) gave a solid (115 mg), $[\alpha]_D^{26}$ −97° (*c*, 0·7, EtOH), which was identical with VIII by thin-layer mobility, IR and mass spectral comparisons.

Acetylation of aspidodasycarpine (XI)

The alkaloid (XI) was treated with acetic anhydride–pyridine (1:1) at room temp overnight. Removal of solvents under red. press. and partition of the residue between ether and K_2CO_3 aq gave, from the organic layer, *aspidodasycarpine*-N,O-*diacetate* (XX), m.p. 111–114°, $[\alpha]_D^{29}$ −34·5° (*c*, 1·42). (Found: C, 63·81, H, 6·90, N, 6·28, mol. wt. by mass spectrometry: 454; calc. for $C_{25}H_{30}N_2O_5 \cdot H_2O$: C, 63·54, H, 6·83, N, 5·93%, mol. wt., 454, anhydrous); λ_{max} ($CHCl_3$) 5·47, 6·18 μ; λ_{max} (EtOH) 295, 238 (3·30, 3·78), λ_{min} 242, 227 mμ (2·51, 3·68); 1·73 (3H doublet J = 7 c/s, CH_3CH=), 1·92 (3H singlet, CH_3COO), 2·16 (3H singlet, CH_3CON), 3·75 (3H singlet, CH_3O_2C), 5·67 (1H quartet J = 7 c/s, CH_3CH=), 6·55–7·70 ppm (4H, HAr).

Action of akali on aspidodasycarpine-N,O-*diacetate* (XX)

The diacetate (XX; 34 mg) in MeOH (2 ml) was treated with KOH aq (100 mg in 1 ml) at room temp for 48 hr. The solution was poured into water and extracted with ether. The dried evaporated ethereal layer gave a gum (15 mg) purified by preparative TLC (band at R_f 0·7), to give *desformo-aspidodasycarpine*-N-*acetate* (XXI), amorphous, $[\alpha]_D^{25}$ −86° (*c*, 0·98 in EtOH). (Found: mol. wt. by mass spectrometry: 382; calc. for $C_{22}H_{26}N_2O_4$: mol. wt., 382), λ_{max} ($CHCl_3$) 2·95, 5·75, 6·26 μ: λ_{max} (EtOH) 295, 240 (3·59, 3·98), λ_{min} 268 mμ (3·13); 1·58 (3H doublet J = 7 c/s, CH_3CH=), 2·20 (3H singlet, CH_3CON), 3·70 (3H singlet, CH_3O_2C), 4·7 (1H, HN), 5·58 (1H quartet J = 7 c/s, CH_3CH=), 6·5–7·3 ppm (4H, HAr); mass spectrum (h) showed a mol. ion at 382 (53%) as well as *m/e* 351 (16), 339 (13), 323 (17), 263 (18), 207 (21), 194 (13), 180 (19), 172 (29), 167 (19), 159 (20), 156 (24), 150 (43), 144 (23), 130 (22), 125 (30) and 108 (100).

Action of alkali on aspidodasycarpine (XI)

The alkaloid (20 mg) was refluxed with excess MeONa in MeOH (5 ml) for 6 hr under N_2. The mixture was poured into water and extracted with $CHCl_3$. The residue after removal of organic solvent was chromatographed over alumina (act. II) and eluted with $CHCl_3$ to give a colourless solid (7 mg.) *desformoaspidodasycarpine* (XXIV). (Found: mol. wt. by mass spectrometry: 340; calc. for $C_{20}H_{24}N_2O_3$: mol. wt., 340); λ_{max} ($CHCl_3$) 2·95, 5·75, 6·22 μ; λ_{max} (EtOH) 296, 240 mμ (3·77, 3·94);

mass spectrum showed a mol. ion at 340 (13%) as well as *m/e* 233 (6), 194 (6), 180 (7), 174 (9), 172 (7), 167 (5), 159 (4), 156 (5), 154 (4), 144 (7), 130 (7), 122 (6) and 108 (100).

Acetylation of XXIV with acetic anhydride–pyridine in the manner described above gave XXI identical with the sample obtained above, by comparison of mass and IR spectra.

Preparation of N-*hydroxymethyl desformoaspidodasycarpine* (XXIII)

(a) Aspidodasycarpine (40 mg) in MeOH (2 ml) was treated with KOH (100 mg) in water (1 ml) at room temp for 30 hr. The crystalline precipitate was filtered to give N-*hydroxymethyl desformoaspidodasycarpine* (XXIII; 27 mg), m.p. 175–182° dec, $[\alpha]_D^{30}$ −50° (*c*, 0·18). (Found: M-18 by mass spectrometry: 352; calc. for $C_{21}H_{26}N_2O_4$: mol. wt. 370); λ_{max} ($CHCl_3$) 2·95, 5·75, 6·20 μ; λ_{max} (EtOH) 298, 242 mμ (3·45, 3·83); 1·51 (3H doublet J = 7 c/s, CH_3CH=), 4·15 and 4·40 (2H AB quartet J = 9 c/s, N_bCH_2O), 5·50 (1H quartet J = 7 c/s, CH_3CH=), 6·4–7·2 ppm (4H, HAr); mass spectrum (h) showed no mol. ion but an M-18 peak at 352 (21%) as well as *m/e* 340 (16), 233 (7), 232 (7), 194 (4), 180 (6), 172 (12), 167 (7), 159 (6), 156 (7), 152 (9), 144 (10), 130 (10), 122 (41), 120 (46) and 108 (100).

(b) Desformoaspidodasycarpine (XXIV; 6 mg) in MeOH (1 ml) and water (0·5 ml) was treated with KOH (80 mg) and formaldehyde (30%, 1 drop) at room temp for 1 hr. The crystalline precipitate (quantitative) was identical with XXIII as prepared above by IR, mixture m.p., and TLC comparison.

Reaction of N-*hydroxymethyl desformoaspidodasycarpine* (XXIII) *with acetic acid*

The methylol (5 mg) in MeOH (0·5 ml) was treated with acetic acid (0·3 ml) at room temp for 3 hr. The mixture was poured onto water, basified and extracted with $CHCl_3$ to give a solid (3 mg) identical with XXIV by IR and TLC comparison.

Reduction of N-*hydroxymethyl desformoaspidodasycarpine* (XXIII) *with lithium aluminium hydride*

The methylol (85 mg) was heated under reflux with excess LAH in tetrahydrofuran for 8 hr. The mixture was cooled and decomposed with saturated $MgSO_4$ solution, filtered, diluted with $CHCl_3$, dried and evaporated. The residue was purified by preparative TLC (zone at R_f 0·1) to give a gum (30 mg; XXV). (Found: mol. wt. by mass spectrometry: 328; calc. for $C_{20}H_{28}N_2O_2$: mol. wt, 328); λ_{max} ($CHCl_3$) 2·95, 3·50, 6·25, 9·70 μ; λ_{max} (EtOH) 297, 246 mμ (3·42, 3·80); 1·74 (3H doublet J = 7 c/s, CH_3CH=), 2·32 (3H singlet, CH_3N), 5·5 (1H quartet J = 7 c/s, CH_3CH=), 6·5–7·2 ppm (4H, HAr); mass spectrum (d) showed a mol ion at 328 (1%) as well as *m/e* 167 (5), 152 (5), 150 (5), 145 (5), 144 (4), 136 (15), 130 (17), 124 (14), 122 (100) and 110 (18).

The *tetradeuterio compound* (XXVI) was prepared as above but using LAD; mass spectrum (d) showed a mol. ion at 332 (1%) (corresponding to $C_{20}H_{24}D_4N_2O_2$) as well as *m/e* 169 (3), 136 (17)) 131 (14), 125 (18) and 123 (100).

Preparation of N-*methyl aspidodasycarpine* (XXVIII)

The alkaloid (XI; 10 mg) in acetone–benzene (1:2, 3 ml) was treated with MeI at room temp. The precipitate formed was washed with ether and treated with 0·1 N NaOH and extracted with $CHCl_3$. The dried evaporated $CHCl_3$-layer gave N-*methyl aspidodasycarpine* (XXVII; 4 mg). (Found: mol. wt. by mass spectrometry: 384; calc. for $C_{23}H_{28}N_2O_4$: mol. wt., 384); λ_{max} ($CHCl_3$) 2·95, 5·78, 6·26, 9·58 μ; mass spectrum (d) showed a mol. ion at 384 (10%) as well as *m/e* 354 (5), 194 (5), 180 (5), 172 (4), 167 (4), 159 (3), 156 (3), 150 (3), 144 (4), 130 (4) and 122 (100).

Reduction of aspidodasycarpine-N,O-*diacetate* (XX)

Reduction using LAH was carried out as described above to give amorphous XXXVII. 1·1 (3H triplet J = 9 c/s, CH_3CH_2), 1·8 (3H doublet J = 7 c/s, CH_3CH=), 5·55 (1H quartet J = 7 c/s, CH_3CH=), 6·5–7·3 ppm (4H, HAr); mass spectrum (d) showed a mol. ion at 372 (calc. 372 for $C_{22}H_{32}N_2O_3$) as well as a base peak at *m/e* 136.

Reduction of desformoaspidodasycarpine-N-*acetate* (XXI)

Reduction with LAD was carried out as described above to give the alcohol XXXVIII; λ_{max} ($CHCl_3$) 3·0, 4·55, 4·80, 6·25, 9·85 μ, whose mass spectrum (d) showed a mol. ion at 347 (calc. for $C_{21}H_{25}D_6N_2O_2$) as well as a base peak at *m/e* 138.

Action of zinc-hydrochloric acid on aspidodasycarpine-N,O-*diacetate* (XX)

To the diacetate (100 mg) in EtOH (4 ml) was added Zn dust (500 mg) and then conc. HCl (0·5 ml) dropwise with vigorous stirring at room temp during 2 min. The slurry was filtered, concentrated *in vacuo* and then poured into water. The precipitate was filtered and purified by preparative TLC to give *secoaspidodasycarpine*-N-*acetate* (XXX; R_f 0·67; 45 mg), m.p. 196–199° (ex pyridine–water). (Found: mol. wt. by mass spectrometry: 396; calc. for $C_{23}H_{28}N_2O_4$: mol. wt., 396); λ_{max} (KBr) 2·94, 5·85, 6·17 μ; λ_{max} (EtOH) 292, 284, 224 (3·73, 3·79, 4·40) λ_{infl} 277 mμ (3·75); 1·49 (3H doublet J = 6 c/s, CH_3CH=), 2·02 (3H singlet, CH_3CON), 2·20 (1H multiplet, =C·CH·C=), 3·0 (2H triplet J = 7 c/s, β-indole·CH_2CH_2O), 3·72 (3H singlet, CH_3O_2C), 3·85 (2H triplet J = 7 c/s, CH_2O), 4·0 (1H, HO), 4·49 (1H doublet J = 13 c/s, one of N_b·CH_2C=), 5·0 (1H multiplet, α-indole·CH·N), 5·5 (1H quartet J = 6 c/s, CH_3CH), 5·63 and 6·20 (2 × 1H singlets, CH_2), 6·9–7·7 (4H, HAr), 8·55 ppm (1H, HN); mass spectrum (d) showed a mol. ion at 396 (55%) as well as *m/e* 378 (65), 365 (100), 353 (13), 351 (7), 335 (21), 323 (46), 309 (14), 306 (20), 236 (22), 194 (27), 156 (46), 144 (16) and 130 (26). In addition there was produced *dihydroaspidodasycarpine*-N,O-*diacetate* (XXIX; R_f 0·50; 11 mg) as a glass. (Found: mol. wt. by mass spectrometry: 456; calc. for $C_{25}H_{32}N_2O_6$: mol. wt., 456); λ_{max} ($CHCl_3$) 2·95, 5·75, 6·15 μ; λ_{max} (EtOH) 293, 244 mμ (3·34, 3·68); mass spectrum (d) showed a mol. ion at 456 (5%) as well as *m/e* 438 (2), 424 (8), 396 (7), 382 (7), 365 (4), 351 (11), 194 (14), 180 (11), 156 (20), 150 (41), 144 (25), 130 (55) and 108 (100).

Secoaspidodasycarpine-N,O-*diacetate* (XXXI) was prepared from the above product by acetylation with acetic anhydride–pyridine as described previously, m.p. 187–190° (ex MeOH—H_2O). (Found: mol. wt. by mass spectrometry: 438; calc. for $C_{25}H_{30}N_2O_4$: mol. wt., 438); λ_{max} ($CHCl_3$) 2·89, 5·83, 5·86, 6·18 μ; λ_{max} (EtOH) 292, 283, 223 (3·91, 3·96, 4·61), λ_{infl} 276 mμ (3·95), 1·50 (3H doublet J = 6 c/s, CH_3CH=), 1·80 (3H singlet, CH_3COO), 2·03 (3H singlet, CH_3CON), 3·07 (2H triplet J = 8 c/s, β-indole·CH_2CH_2), 3·75 (3H singlet, CH_3O_2C), 4·30 (2H triplet J = 8 c/s, CH_2O), 5·60 (1H quartet, J = 6 c/s, CH_3CH=), 5·70 and 6·25 (2 × 1H singlets, CH_2=), 6·9–7·7 (4H, HAr), 9·5 ppm (1H, HN); mass spectrum (d) showed a mol. ion at 438 (50%) as well as *m/e* 421 (14), 395 (24), 378 (34), 365 (10), 351 (26), 335 (56), 323 (26), 319 (18), 309 (43), 236 (35), 194 (35), 183 (19), 168 (64), 156 (42), 144 (21), 143 (22), 130 (30) and 71 (100).

Action of zinc–hydrochloric acid on aspidodasycarpine (XI)

To the alkaloid (60 mg) in EtOH (2 ml) Zn dust (400 mg) and then conc. HCl (2 ml) were added and the mixture stirred at room temp for 5 min. The mixture was then filtered and the filtrate poured into water (20 ml), basified (NaOH), extracted with $CHCl_3$ and the residue purified by preparative TLC to give: *compound* A (R_f 0·03; 7 mg); λ_{max} ($CHCl_3$) 2·95, 5·75, 6·24 μ; λ_{max} (EtOH) 300, 246 mμ, λ_{max} (EtOH–0·5 N HCl) 270, 253 mμ; *compound* B (R_f 0·08; 3 mg); λ_{max} ($CHCl_3$) 2·94, 5·77, 6·24 μ; λ_{max} (EtOH) 298, 245 mμ, λ_{max} (EtOH–0·5 N HCl) 272 mμ, λ_{infl} 250 mμ; and *seco-aspidodasycarpine* (R_f 0·33; 5 mg). (Found: mol. wt. by mass spectrometry: 354; calc. for $C_{21}H_{26}N_2O_3$: mol. wt., 354); λ_{max} ($CHCl_3$) 2·9, 3·5, 5·85 μ; λ_{max} (EtOH) 291, 283, 225 (3·79, 3·85, 4·51), λ_{infl} 276 mμ (3·83); mass spectrum (d) showed a mol. ion at 354 (100%) as well as *m/e* 336 (35), 324 (38), 323 (31), 309 (32), 269 (45), 255 (20), 194 (32), 187 (29), 186 (26), 183 (40), 169 (77), 158 (50), 157 (54), 156 (80), 144 (44), 130 (54) and 108 (54).

Tetrahydrosecoaspidodasycarpine-N-*acetate*

The indole (XXX; 13 mg) was reduced in EtOH at room temp and atm. press. with H_2 over Pd—C (10%). The resulting glass was purified by preparative TLC to give the *tetrahydrocompound* (4 mg; R_f 0·7). (Found: mol. wt. by mass spectrometry: 400; calc. for $C_{23}H_{32}N_2O_4$: mol. wt., 400); λ_{max} ($CHCl_3$) 2·95, 5·79, 6·15 μ; λ_{max} (EtOH) 292, 282, 273, 224 mμ (3·59, 3·67, 3·67, 4·23); mass spectrum (d) showed a mol. ion at 400 (32%) as well as *m/e* 385 (41), 383 (18), 369 (27), 357 (24), 355 (26), 339 (18), 327 (41), 313 (29), 295 (13), 240 (44), 225 (19), 198 (21), 175 (19), 156 (17), 152 (33), 150 (19), 130 (19), 110 (100) and 108 (70).

Conversion of picraline (XXXIV) *to* N-*hydroxymethyl desformoaspidodasycarpine* (XXIII)

A mixture of picraline (98 mg) and KBH_4 (2 g) in MeOH (20 ml) was heated under reflux for 5 hr. The cooled solution was acidified with 2 N HCl and concentrated *in vacuo*. The solution was made alkaline (NaOH) and extracted with ether. Removal of solvent gave a glass whose IR spectrum

and TLC mobility were identical with those of XXIV. Half of the material was dissolved in 1 ml MeOH and water (0·5 ml), KOH (80 mg) and 37% aqueous formaldehyde (2 drops) were added and allowed to stand for 3 hr to give 22 mg of N-hydroxymethyl desformoaspidodasycarpine (XXIII) (m.p. 174–180°, $[\alpha]_D^{28}$ −42° (*c*, 0·20)), which was identical by IR, UV, mixture m.p. and TLC mobility with the substance obtained from aspidodasycarpine (XI). The IR spectrum and TLC mobility of N_b–acetyl desformoaspidodasycarpine (XXI) prepared from the borohydride reduction product (XXVI) of picraline (XXXIV) by pyridine-acetic anhydride was also identical with XXIV derived from aspidodasycarpine (XI).

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