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ALKALOID STUDIES—LVIII.

THE ALKALOIDS OF SIX *ASPIDOSPERMA* SPECIES

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Abstract—The alkaloids of a number of *Aspidosperma* species were isolated and identified. In one species the lignanes (−)-lirioresinol-C and (+)-O,O-dimethyllirioresinol-B were also encountered.

IN CONTINUATION of previous work[1] directed towards a complete chemo-taxonomic survey[2] of the genus *Aspidosperma* a further series of species has been studied. In general the alkaloids isolated belong to well-defined groups, already found in the genus and normally characteristic of the botanical sub-groups.[2] Thus, for example, the sub-group Nitida (*A. rigidum, nitidum,* and *marcgravianum*) is characterized by indolic alkaloids of the heteroyohimbine and corynantheol types, while the Pyricolla (*A. pyricollum* and *nigricans*) and Tomentosa sub-groups contain as principal constituents uleine and related alkaloids. The appearance of (+)-stemmadenine (I) previously found in the genera *Stemmadenia*[3] and *Melodinus*[4] is noteworthy for although it is a natural precursor of the aspidospermatidine group of alkaloids based on structure (II), it had not been previously encountered in *Aspidosperma.*

Picraline (III),[5,6] isolated for the first time from this genus, is closely related to alkaloids found earlier, for example, aspidodasycarpine (IV).[7] A list of the alkaloids is given in the Table.

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[1] For leading references see B. GILBERT, A. P. DUARTE, Y. NAKAGAWA, J. A. JOULE, S. E. FLORES, J. AGUAYO BRISSOLESE, J. CAMPELLO, E. P. CARRAZZONI, R. J. OWELLEN, E. C. BLOSSEY, K. S. BROWN, JR., and C. DJERASSI, *Tetrahedron* **21**, 1141 (1965).

[2] B. GILBERT, *Anais Acad. Bras. Cienc.* **38** suppl., 315 (1966).

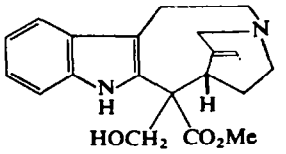
[3] F. WALLS, O. COLLERA and A. SANDOVAL, *Tetrahedron* **2**, 173 (1958).

[4] H. H. A. LINDE, *Helv. Chim. Acta* **48**, 1822 (1965).

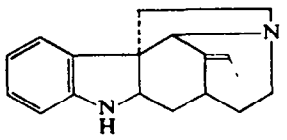
[5] A. Z. BRITTEN, G. F. SMITH and G. SPITELLER, *Chem Ind.* (*London*) 1492 (1963).

[6] L. OLIVIER, J. LÉVY, J. LE MEN, M.-M. JANOT, H. BUDZIKIEWICZ and C. DJERASSI, *Bull. Soc. Chim. France* 868 (1965).

[7] J. A. JOULE, M. OHASHI, B. GILBERT and C. DJERASSI, *Tetrahedron* **21**, 1717 (1965).



(I)



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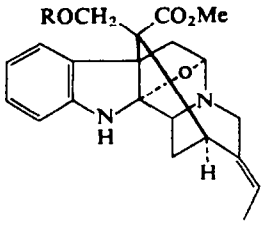
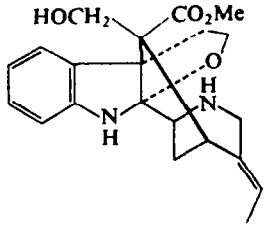
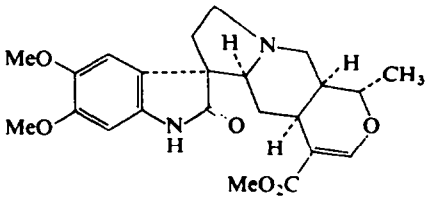


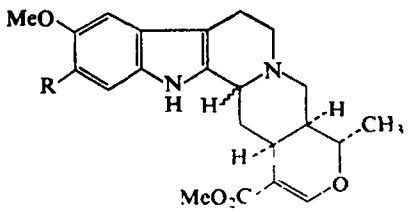
(III) R = Ac
(V) R = H



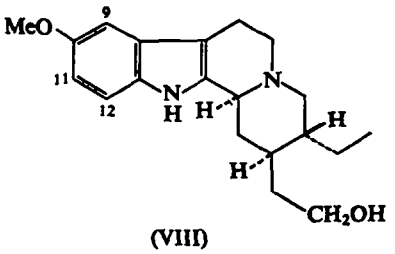
(IV)



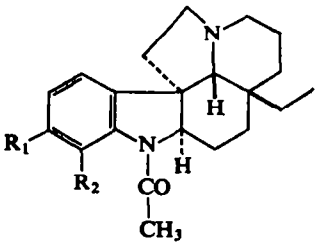
(VI)



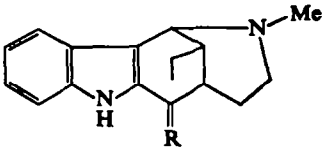
(VII) R = OMe; 3β-H
(IX) R = H; 3α-H



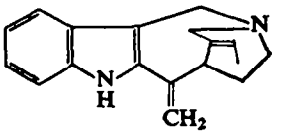
(VIII)



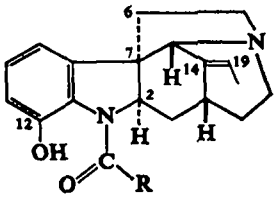
(X) $R_1 = R_2 = H$
(XI) $R_1 = OMe$, $R_2 = OH$



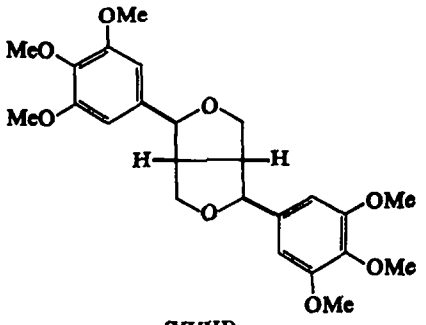
(XII) $R = CH_2$
(XIV) $R = O$
(XV) $R = H, CH_3$



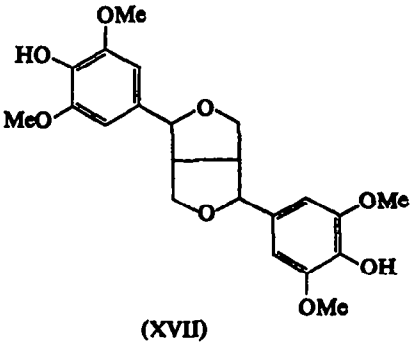
(XIII)



(XVI) $R = CH_3$
(XIX) $R = CH_2CH_3$
(XX) $R = CH_3$;14,19-dihydro-



(XVIII)



(XVII)

TABLE 1.

Species	Alkaloid (with principal references)[8]
A. rigidum Rusby* (bark) (= *A. laxiflorum* Kuhlm.)	Picraline (III)[5, 6] Deacetylpicraline (V)[5, 6, 9] (−)-Carapanaubine (VI)[10] Reserpiline (VII)[11]
A. nitidum Benth ex Müll.-Arg.† (bark)	10-Methoxydihydrocorynantheol (VIII)[11]
A. marcgravianum Woodson‡[12] (wood)	Aricine (IX)[12] Reserpiline (VII) (−)-Demethoxyaspidospermine (X)[1, 13, 14] (+)-Aspidocarpine (XI)[15]
A. pyricollum Mull.-Arg.§ (bark)	Uleine (XII)[16, 17] Apparicine (XIII)[18] Dasycarpidone (XIV)[7, 19]
A. pyricollum [20] (fruits)	(+)-Stemmadenine (I)[21]
A. nigricans Handro¶	(+)-Uleine (XII) Dihydrouleine (XV)[22]
A. tomentosum Mart.**	(+)-Uleine (XII) (+)-12-Hydroxy-*N*-acetylaspidospermatidine (XVI)†† (limatinine)[23]

* *A. rigidum* was collected beside the BR29 highway near Rio Jaru in the Rondonia Territory and registered under Rio de Janeiro Botanical Garden No. 116846.

† *A. nitidum* grows in mud beside the Igarapé da Cachoeira Baixa do Tarumã, Manaus, Amazonas, and has distinctive yellow wood. (Rio de Janeiro Herbarium No. 119069; I.N.P.A. Herbarium, Manaus reg. No. 1240.)

‡ *A. marcgravianum* was collected in inundated ground near Igarapé do Passarinho, Manaus, Amazonas, I.N.P.A. Herbarium no. 6150; Rio de Janeiro Botanical Garden No. 119072.

§ *A. pyricollum* is normally a maritime species. The present specimen from an atypical inland habitat showed slight morphological differences. It was collected at km 18 of the Porto Seguro–BR5 road, Bahia.

‖ *A. pyricollum* Müll.-Arg. fruits were collected from trees on the sandy coastal strip, Rio de Janeiro. This variety is that previously studied.[17, 20]

¶ *A. nigricans* collected in Porto Seguro, Bahia, has been studied previously.[1] The present specimen collected in Garanhuns, Pernambuco (Instituto de Pesquisas Agronômicas, Recife Herbarium No. 1371), appears to be the same species and yields olivacine, olivacine-*N*-oxide and guatambuine as principal constituents. Uleine and dihydrouleine now encountered, were not found in the Porto Seguro specimen, and in view of the difference the firm identification of the present species as *nigricans* must await further botanical examination.

** *A. tomentosum* was collected on the Fazenda da Mãe D'água, Várzea da Palma, Minas Gerais, and is distinguished from co-occurrent species by possessing bark free of cork. We thank Dr. José Vicente Gonçalves Pinto and the Belgo-Mineira Iron Company for assistance in their collection.

†† This alkaloid has been isolated simultaneously in two other laboratories, from species of *Aspidosperma* collected by us: *A. limae* Woodson,[23] and *A. subincanum* Mart.[24]

[8] For more complete bibliography see R. H. MANSKE, *The Alkaloids*, Vol. VIII. Academic Press, New York (1965).

[9] L. OLIVIER, J. LÉVY, J. LE MEN and M. M. JANOT, *Ann. Pharm. Franç.* **20**, 361 (1962).

[10] B. GILBERT, J. AGUAYO BRISSOLESE, N. FINCH, W. I. TAYLOR, H. BUDZIKIEWICZ, J. M. WILSON and C. DJERASSI, *J. Am. Chem. Soc.* **85**, 1523 (1963).

[11a] N. DASTOOR and H. SCHMID, *Experientia* **19**, 297 (1963);

[11b] N. J. DASTOOR, A. A. GORMAN and H. SCHMID, *Helv. Chim. Acta* **50**, 213 (1967). We are indebted to Professor H. Schmid for i.r. and TLC comparison, and mixture m.p. of our 10-methoxydihydrocorynantheol with his alkaloid AD-IV from *A. discolor*. The two are identical.

[12] B. GILBERT, L. D. ANTONACCIO and C. DJERASSI, *J. Org. Chem.* **27**, 4702 (1962).

[13] J. M. FERREIRA, B. GILBERT, R. J. OWELLEN and C. DJERASSI, *Experientia* **19**, 585 (1963).

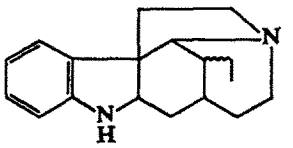
[14] A. WALSER and C. DJERASSI, *Helv. Chim. Acta* **48**, 391 (1965).

[15] S. MCLEAN, K. PALMER and L. MARION, *Can. J. Chem.* **38**, 1547 (1960).

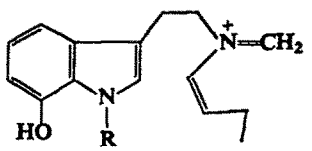
A. marcgravianum Woodson was the object of an earlier study[12] when aricine and dihydrocorynantheol were isolated from the bark. The former again appeared as a major constituent of the wood. In addition to the alkaloids recorded in the table, the lignanes (−)-lirioresinol-C (XVII)[25,26] and (+)-O,O-dimethyllirioresinol-B (XVIII)[25,27,28] were isolated.

12-Hydroxy-*N*-acetylaspidospermatidine (XVI) (see Table note††) the *N*-acetyl analogue of limatine (XIX)[29] was characterized as a 7-hydroxy-*N*-acyldihydroindole by its i.r. and u.v. absorption.[23] NMR and mass spectral data were consistent with structure (XVI), the precisely determined mass of the molecular ion, 324·18367 confirming the molecular formula, $C_{20}H_{24}N_2O_4$. Location of the ethylidene side-chain at position 14 was confirmed by the appearance of the C-3 proton as a singlet (4·52 ppm) superimposed on the C-2 proton quartet, as well as by the mass spectral breakdown of the hydrogenation product, 14,19-dihydro-12-hydroxy-*N*-acetylaspidospermatidine (XX). The diagnostic peaks in dihydro-aspidospermatidine (XXI) have been shown[30] to occur at m/e 199 and 227, the latter having twice the intensity of the former. These peaks appear in the spectrum of (XX) at m/e 257·12921 ($C_{15}H_{17}N_2O_2^+$) and 285·16019 ($C_{17}H_{21}N_2O_2^+$, fragment *a*; accompanied by *b*, m/e 243) with the same relative intensities as found in the case of (XXI).

The absolute configuration of alkaloids of the aspidospermatidine group has been related to their ORD curves.[31] The negative sign of the main Cotton effect (below 290 nm) of (+)-12-hydroxy-*N*-acetylaspidospermatidine (XVI) is indicative of C-6 alpha at C-7 as in (+)-limatine (XIX)[29,31] (see Ref. 23).



(XXI)



a, R=$COCH_3$ m/e 285
b, R=H m/e 243

[16] G. A. Büchi and E. W. Warnhoff, *J. Am. Chem. Soc.* **81**, 4433 (1959).
[17] B. Gilbert, L. D. Antonaccio, A. A. P. G. Archer and C. Djerassi, *Experientia* **16**, 61 (1960).
[18] J. A. Joule, H. Monteiro, L. J. Durham, B. Gilbert and C. Djerassi, *J. Chem. Soc.* 4773 (1965).
[19] M. Ohashi, J. A. Joule, B. Gilbert and C. Djerassi, *Experientia* **20**, 263 (1964).
[20] R. R. Arndt and C. Djerassi, *Experientia* **21**, 566 (1965).
[21] A. Sandoval, F. Walls, J. N. Shoolery, J. M. Wilson, H. Budzikiewicz and C. Djerassi, *Tetrahedron Letters* 409 (1962). We thank Dr. F. Walls of the Universidad Nacional Autónoma de Mexico, and Drs. A. Hofmann and D. Stauffacher of Sandoz A.G., Basel, for samples of stemmadenine and its hydrochloride.
[22] J. Schmutz, F. Hunzicker and R. Hirt, *Helv. Chim. Acta* **40**, 1189 (1957).
[23] M. Pinar and H. Schmid, *Helv. Chim. Acta* **50**, 89 (1967).
[24] J. A. Joule and A. J. Gaskell, Private communication.
[25] E. E. Dickey, *J. Org. Chem.* **23**, 179 (1958); I. A. Pearl, D. L. Beyer and E. E. Dickey, *J. Org. Chem.* **23**, 705 (1958).
[26] L. A. Elyakova, A. K. Dzizenko and G. B. Elyakov, *Doklady Akad. Nauk SSSR* **165**, 562 (1965).
[27] We thank Professor P. R. Jefferies (University of Western Australia, Nedlands, W.A.) for an authentic sample of (XVIII); P. R. Jefferies, J. R. Knox and D. E. White, *Australian J. Chem.* **14**, 175 (1961).
[28] E. N. Maslen, C. Nockolds and M. Paton, *Australian J. Chem.* **15**, 161 (1962).
[29] M. Pinar, B. W. Bycroft, J. Seibl and H. Schmid, *Helv. Chim. Acta* **48**, 822 (1965).
[30] K. Biemann, M. Spiteller-Friedmann and G. Spiteller, *Tetrahedron Letters* 485 (1961); *J. Am. Chem. Soc.* **85**, 631 (1963).
[31] W. Klyne, R. J. Swan, B. W. Bycroft and H. Schmid, *Helv. Chim. Acta* **49**, 833 (1966).

EXPERIMENTAL*

Isolation and identification of known alkaloids. Alkaloids were isolated following procedures described previously[1] and by preparative TLC on silica gel. Identification was made by i.r., u.v., NMR and mass spectral data. Chromatographic comparison and mixture m.p. with authentic samples was made wherever possible.

A. nitidum. 10-Methoxydihydrocorynantheol (VIII) isolated from the pH 7 methylene chloride extract by gradient elution with CH_2Cl_2—MeOH on silica gel followed by successive thin-layer chromatography on alumina using acetone–water–hexane (51:4:50) then chloroform–methanol had m.p. 161–162°, $[\alpha]_D^{26.5}$ −10·8 (pyridine), identical[11] to alkaloid AD-IV from *A. discolor* A.DC. The NMR spectrum ($CDCl_3$, 60 Mc) showed 0·95 (terminal methyl of ethyl group), 3·85 (3H, singlet, OCH_3), 6·90 (1H, quartet, J=9 and 3 c/s, C-11 proton; and 1H, doublet, J=3 c/s, C-9 proton), 7·16 (1H, doublet, J=9 c/s, C-12 proton) and 8·0 ppm (1H, broad, indole N—H). The high resolution mass spectrum showed principal peaks at m/e: 328·21459 ($C_{20}H_{28}N_2O_2$, molecular ion), 313 (M—CH_3), 299·17632 ($C_{18}H_{23}N_2O_2$, M—C_2H_5), 283·18055 ($C_{18}H_{23}N_2O$, M—CH_2CH_2OH), 255·14998 ($C_{16}H_{19}N_2O$), 214·11050 ($C_{13}H_{14}N_2O$), 200·09545 ($C_{12}H_{12}N_2O$), 199·08752 ($C_{12}H_{11}N_2O$) and 186 ($C_{12}H_{12}NO$). All of these peaks are consistent with the fragmentation patterns previously established.[12, 32]

A. marcgravianum. Ethanolic wood extract (200 g) gave a pH 4 benzene extract (3·5 g) from which aricine (175 mg) was isolated. In addition this fraction yielded O,O-dimethyllirioresinol-B (154 mg), recrystallized repeatedly from methanol, m.p. 121–122°, $[\alpha]_D^{24.7}$ +46·0 (c, 1·61 in chloroform), λ_{max}^{EtOH} 210, 230 (sh) and 272 nm (log ϵ 4·57, 4·01, 3·35). The mass spectrum showed principal peaks at m/e: 446 (M^+, base peak), 416, 397, 386, 279, 250, 235, 224, 219, 207, 195, 181. I.r. and TLC comparison and mixture m.p. determination show identity to authentic O,O-dimethyllirioresinol-B.[27]

A small amount of (−)-lirioresinol C was also isolated by TLC on silica-gel, and recrystallized from acetone–hexane when it had m.p. 177–183° $[\alpha]_D$ −34·8 (chloroform), NMR spectrum in $CDCl_3$: 3·11 (2H, broad multiplet, half width 12 c/s, CH—C*H*—CH_2), 3·85 and 4·22 (2H, double doublets with fine structure, CH—CH_2—O), 3·88 (12H, singlet, OCH_3), 4·73 (2H, doublet, J= 5 c/s, O—C*H*(Ar)—CH), 5·60 (2H, singlet, exch. by D_2O, O*H*) and 6·59 ppm (4H, singlet, aromatic H).[26] The high resolution mass spectrum gave the molecular formula $C_{22}H_{26}O_8$ and principal peaks (low resolution) were observed at m/e: 418 (M^+, base peak), 388, 387, 319, 280, 251, 236, 221, 210, 193, 181, 167, 161. The i.r. spectrum showed identity to (+)-lirioresinol C (syringaresinol) and non-identity with lirioresinols-A and -B.[25]

A. tomentosum. (+)-12-Hydroxy-*N*-acetylaspidospermatidine (XVI) was isolated from the pH 7 chloroform extracts by gradient elution from silica gel with ethyl acetate–methanol, and after recrystallization from methanol had mp. 162–163°, Ce^{IV} colour pink, $[\alpha]_D^{22}$ +170 (c, 0·24 in ethanol), ORD in methanol: c, 0·05. $|\Phi|_{max}^{trough}$ +3120°, $|\Phi|_{272}^{trough}$ −3015°, $|\Phi|_{245}^{infl}$ 32000°; ν_{max}^{nujol} 1626 (s), 1595 (m), 1570 (m), 787 (m), 740 (m) cm^{-1}, λ_{max}^{EtOH} 214, 254 and 290 nm (24950, 7940, 4500); λ_{min}^{EtOH} 242 and 282 nm (5320, 4250); $\lambda_{max}^{EtOH-NaOH}$ 310 nm (6210) $\lambda_{min}^{EtOH-NaOH}$ 282 nm (1740), NMR (60 Mc in $CDCl_3$), 1·63 (3H, doublet, J=6·5 c/s, =CH.C*H*$_3$), 2·37 (3H, singlet, COC*H*$_3$), 2·8–3·3 (3H, multiplet), 4·52 (1H, singlet superimposed on 1H, quartet, C-3 and C-2*H*), 5·40 (1H, quartet, J=6·5 c/s, =C*H*.CH_3), 6·55–7·30 (3H, multiplet, aromatic *H*) and 10·7 ppm (1H, singlet, hydrogen bonded O*H*). The last signal was eliminated by D_2O treatment. The mass spectrum showed principal peaks at m/e: 324 (60, M^+, composition by high resolution, $C_{20}H_{24}N_2O_2$), 281 (6, M—$COCH_3$), 238 (3), 160 (8), 159 (5), 146 (7), 137 (10), 136 (100 per cent), 123 (10).

Methylation with dimethyl sulphate, and K_2CO_3 in dry acetone gave only a very low yield of the *O*-methyl derivative (aspidospermatine), λ_{max}^{EtOH} 220, 255 and 290 nm unchanged in alkaline solution, with principal mass spectral peaks at: 338 (M^+, 30), 323 (M—CH_3, 4), 295 (M—Ac, 6), 174 (18), 160 (11), 136 (100 per cent), 123 (16.)

Dihydro-12-hydroxy-N-acetylaspidospermatidine (XXI). 12-Hydroxy-*N*-acetylaspidospermatidine (15 mg) was hydrogenated in ethanol (8 ml) over Pt (from Adam's PtO_2 28 mg) at room temperature and pressure for 20 hr. Filtration of the catalyst and recrystallization from ethanol and sublimation at 170°/10^{-3} gave dihydro-12-hydroxy-*N*-acetylaspidospermatidine, m.p. 222–224° dec. with i.r. and u.v. spectra practically superimposable upon those of the starting material. The R_f (Silica gel G, ethyl acetate methanol, 1:1) and Ce^{IV} colour are also identical to those of (XVI). The mass spectrum showed principal peaks at m/e (ion source inlet): 326 (M^+, 30, composition by high resolution, $C_{20}H_{26}N_2O_2$), 298 (2), 285 (6, $C_{17}H_{25}N_2O_2^+$), 270 (5), 257 (3, $C_{15}H_{17}N_2O_2^+$), 243 (1), 240 (2), 226 (2), 201 (3), 174 (3), 160 (10), 146 (5), 138 (100 per cent), 125 (3), 123 (3), 110 (14).

Acknowledgement—We are indebted to the Rockefeller Foundation, the National Science Foundation, the Conselho Nacional de Pesquisas and the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior for financial support during this investigation.

* M.ps are uncorrected. NMR spectra were determined by Dr. Lois J. Durham and mass spectra by Drs. H. Budzikiewicz and A. Duffield.

[32] L. D. ANTONACCIO, N. A. PEREIRA, B. GILBERT, H. VORBRUEGGEN, H. BUDZIKIEWICZ, J. M. WILSON, L. J. DURHAM and C. DJERASSI, *J. Am. Chem. Soc.* **84**, 2161 (1962).