Phytochemistry, 1976, Vol. 15, pp. 551–553. Pergamon Press. Printed in England

20-EPIHEYNEANINE, AN IBOGA ALKALOID FROM *PESCHIERA AFFINIS*

FRANCISCO J. ABREU MATOS, RAIMUNDO BRAZ F°.,* OTTO R. GOTTLIEB,†
F. WELBANEIDE L. MACHADO and M. IRACEMA L. M. MADRUGA

Departamento de Química Orgânica e Inorgânica, Universidade Federal do Ceará, Fortaleza and †Instituto de Química, Universidade de São Paulo, Brasil

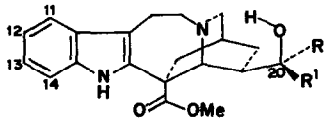
(*Received* 13 *September* 1975)

Key Word Index—*Peschiera affinis*; Apocynaceae; 20-epiheyneanine; heyneanine; coronaridine; coronaridine pseudoindoxyl; affinisine; olivacine.

Abstract—20-Epiheyneanine, an epimer of the known iboga alkaloid heyneanine, is the major base of the root bark of *Peschiera affinis*. It is accompanied by lesser quantities of coronaridine, coronaridine pseudoindoxyl, affinisine and olivacine, as well as sitosterol, β-amyrin, lupeol and 3-*O*-acetyllupeol.

INTRODUCTION

Antitumor [1] and spasmolytic [2] properties of a crude extract and the major alkaloid [3] from the root bark of *Peschiera affinis* (Muell. Arg.) Miers (= *Tabernaemontana affinis* Muell. Arg., Apocynaceae Plumieroideae) were reported. The fractionation of this extract has now led to the terpenoids sitosterol, β-amyrin, lupeol and 3-*O*-acetyllupeol, besides the alkaloids coronaridine [4], coronaridine pseudoindoxyl, whose natural occurrence was anticipated [5], affinisine [6] and olivacine [7]. The major alkaloid was characterized as 20-epiheyneanine (1b).



(1a) R = Me, R¹ = H
(1b) R = H, R¹ = Me

RESULTS

The molecular formula $C_{21}H_{26}N_2O_3$, determined by high resolution MS, the qualitative composition of the low resolution MS (Table 1) and the UV of the major alkaloid were registered previously for heyneanine (20-hydroxycoronaridine) [8,9]. The mp, however, was depressed upon admixture of an authentic sample, kindly supplied by Dr. S. M. Kupchan, and the IR spectra, though very similar, were not superimposable. The sole significant difference of the ^{1}HMR spectra (Table 2) referred to the frequencies due to the protons of the side chain. This is locked in a fixed conformation by the intramolecular hydrogen bridge, whose existence was ascertained through the invariance of the IR 3470 cm^{-1} band upon dilution in $CHCl_3$. Thus the substituents at C-20 are exposed to two possible environments, according to their configuration. The $C\underline{H}_3$-20 MR signal of heyneanine and the H-20 signal of the novel isolate appear upfield relative to the corresponding signals of the respective epimers. These substituents (R in 1) may thus be located in the anisotropically protected region above the ring systems, as tentatively proposed in the formulae 1a for heyneanine and 1b for epiheyneanine.

Mp's, as well as UV, IR, ^{1}HMR and MS, of four additional alkaloids were identical with lit. data for coronaridine [4,10], coronaridine pseudoindoxyl [5], affinisine [6,11] and olivacine [7,12]. The identification of coronaridine was confirmed by saponification of the base with KOH followed by decarboxylation in HCl to ibogamine [4], compared with an authentic sample kindly supplied by Dr. E. Wenkert. The identification of affinisine and of olivacine was confirmed by direct comparison with authentic samples kindly supplied, respectively, by Dr. M. P. Cava and Dr. C. W. Mosher.

DISCUSSION

The major alkaloids of *P. affinis* are, according to previous work [6,11], of the vobasine (vobasine and affinine) and the sarpagine (affinisine) types. In contrast, *P. lundii* (D. C.) Miers from Porto Seguro, Bahia, Brasil, contains

Table 1. MS of heyneanine (1a) and 20-epiheyneanine (1b)

	m/e	1a %	1b %	*m/e*	1a %	1b %
M	354	100	100			
	339	47	64	214	26	48
	336	50	64	195	10	16
	310	20	16	168	16	20
	309	14	20	154	34	27
	295	6	8	152	26	46
	253	6	12	140	23	48
	224	8	12	138	11	18

* Present address: Departamento de Química, Universidade Federal Rural do Rio de Janeiro.

Table 2. 220 MHz ^{1}HMR spectra of heyneanine (1a) and 20-epiheyneanine (1b) in $CDCl_3$

1a	τ		J(Hz)	1b	τ		J(Hz)
NH	2·07	*s*	—	NH	2·00	*s*	—
H-11	2·57	*d*	7·5	H-11	2·55	*d*	7·5
H-14	2·78	*d*	7·5	H-14	2·77	*d*	7·5
H-13	2·88	*t*	7·5	H-13	2·88	*t*	7·5
H-12	2·94	*t*	7·5	H-12	2·92	*t*	7·5
OH-20	6·10	*s*	—	OH-20	5·87	*s*	—
H-20	5·88	*q*	7·0	H-20	6·08	*m*	—
Me-20	8·89	*d*	7·0	Me-20	8·73	*d*	6·5
OMe	6·32	*s*	—	OMe	6·32	*s*	—
H	6·18–6·37	*m*	—	H	6·17–6·37	*m*	—
H	6·53	*m*	—	H	6·56	*m*	—
4 H	6·81–7·06	*m*	—	4 H	6·77–7·06	*m*	—
H	7·20	*d*	10	H	7·21	*d*	10
H	7·45	*d*	12·5	H	7·45	*d*	12·5
3 H	7·93–8·31	*m*	—	2 H	7·93–8·13	*m*	—
2 H	8·38–8·63	*m*	—	2 H	8·13–8·33	*m*	—
				H	8·60	*t*	9

substantial amounts of iboga alkaloids, as well as olivacine, usually associated with the genera *Tabernaemontana*, *Tabernanthe* and *Voacanga*; only trace amounts of vobasine and no sarpagine type alkaloids were isolated. Thus, it appeared to the authors [5] that *P. lundii* might be better viewed as a *Tabernaemontana* species than as a species of *Peschiera*. The present re-investigation of *P. affinis*, however, again revealed the presence of iboga alkaloids (20-epiheyneanine, coronaridine, coronaridine pseudoindoxyl) and olivacine in the genus *Peschiera*, and confirmed the presence of affinisine. This seems to suggest that the indicated chemical differences are of little systematic significance. It should, nevertheless, not be forgotten that the close botanical relationship between these two genera has often led to confusion and either generic name has been assigned to a species, according to a botanist's individual preferences [5].

Furthermore, the previous study on *P. affinis* [11] employed an EtOH extract of the "whole plant" in which our root bark constituents could have played a relatively minor role and escaped detection. Both analyses refer to specimens collected in north-eastern Brazil. This origin eliminates doubts about correctness of identifications of the plant material. *P. affinis* is one of the more frequent and characteristic shrubs of the sandy plateaus around Fortaleza and no other species of these two genera is known to grow in Ceará State. The identification was based on studies by Adolfo Ducke [13], and confirmed by Dr. G. M. Barroso (Rio de Janeiro Botanical Garden) and Dr. J. Monachino (New York Botanical Garden).

EXPERIMENTAL

Isolation of the constituents. Dry powdered root bark (200 g) was percolated successively with hexane and 0·1 N HCl. The hexane soln. was evap. and the residue (3 g) extd. with hot MeOH. The MeOH soln. was evap. and the residue (420 mg) separated by SiO_2 column chromatography into a solid hydrocarbon, sitosterol, β-amyrin, lupeol and 3-*O*-acetyllupeol. The HCl soln. was basified with NH_4OH. The ppt. (9·1 g) was separated by centrifugation and extd. with C_6H_6–MeOH (3 : 1). Evap. of the solvent gave a residue (6·5 g) which was separated by SiO_2 column chromatography into fractions A_1, A_2 and A_3. A_1 (1 g) was separated by PLC (SiO_2) into sitosterol, 3-*O*-acetyllupeol, lupeol and an alkaloid mixture. The mixture, in anh. C_6H_6–Et_2O, was treated with dry HCl gas. The ppt. was collected and separated into the chlorohydrates of coronaridine, mp 225–227° dec. (lit. [4] 235° dec.), and coronaridine pseudoindoxyl, mp 260° dec. (lit. [5] 278–279° dec.), by fractional crystallization from anh. Me_2CO. The free bases were obtained by alkalinization of the salt solutions and ext. with C_6H_6. A_2 (2·5 g) was separated by SiO_2 column chromatography into an additional quantity of coronaridine, a vitreous mass which pptd crude 1b (1·2 g) upon addition of Me_2CO, and olivacine (100 mg) mp 315° dec. (C_6H_6–MeOH, 9 : 1) (lit. [13] 314–316°). A_3 (2 g) was chromatographed on an Al_2O_3 column. Elution with C_6H_6 gave a fraction which, redissolved in MeOH, was treated with conc. aq. picric acid. The ppt. was collected and recryst. from MeOH. Ion exchange chromatography gave affinisine (30 mg), mp 194–197° (lit. [11] 194–196° dec.).

Epiheyneanine (1b). Colourless crystals, mp 170–172° (cyclohexane).* M found 354·1918; $C_{21}H_{26}N_2O_3$ requires 354·1943. λ_{max}^{EtOH}(nm): 226, 280, 285, 293 (ε 26 900, 5500, 6200, 5300). ν_{max}^{KBr} (cm^{-1}): 3333, 1730, 1618, 1488, 1250, 1080, 1019, 746. $[\alpha]_D^{24}$ −46° (c 10 mg/ml, $CHCl_3$). *Acetate.* Mp 214–215° (MeOH). ν_{max}^{KBr} (cm^{-1}): 1730, 1709, 1626, 1493, 1279, 1250, 1075, 1044, 750. ^{1}HMR (60 MHz, $CHCl_3$, τ): 2·10 (*s*, NH), 2·40–3·10 (*m*, 4 ArH), 4·90 (*m*, CHOAc), 6·30 (*s*, OMe), 6·50–7·60 (*m*, 8H), 7·95 (*s*, OAc), 7·80–8·70 (*m*, 5H), 8·8 (*d*, *J* 6·5 Hz, Me-20).

Acknowledgements—This work was sponsored by Ministério do Planejamento (Financiadora de Estudos e Projetos S.A.) through Academia Brasileira de Ciências and Conselho Nacional de Pesquisas. The authors are indebted to Dr. A. Aragão Craveiro for the MS and to Miss M. H. Torres de Araujo for technical assistance.

* *Note added in proof*: A report on (−)-epiheyneanine [Bellefon, M. de, Debray, M.-M., Men-Oliver, L. le and Men, le (1975) *Phytochemistry* **14**, 1649], which appeared after the present paper had been submitted, gives mp 112° (Me_2CO). Acc. to our experience, samples cryst. from Me_2CO show two CO IR bands of nearly equal intensity at 1710 and 1730 cm^{-1} and a double mp 120–130° and 170–172°.

REFERENCES

1. Fonteles, M. C., Sampaio, M. C., Cardoso, J. H., Pereira, M. E. and Matos, F. J. A. (1969) *Ciência & Cultura* (São Paulo) **21**, 508.
2. Viana, G. S. B., Matos, F. J. A., Vieira, J. E. B., Sousa, M. P. and Medeiros, M. C. (1970) *J. Pharm. Pharmacol.* **22**, 116.

3. Fonteles, M. C., Jerram, D., Matos, F. J. A. and Alquist, R. P. (1974) *Planta Med.* **25,** 175.
4. Gorman, M., Neuss, N., Cone, N. J. and Deyrup, J. A. (1960) *J. Am. Chem. Soc.* **82,** 1142.
5. Hwang, B., Weisbach, J. A., Douglas, B., Raffauf, R. F., Cava, M. P. and Bessho, K. (1969) *J. Org. Chem.* **34,** 412.
6. Cava, M. P., Talapatra, S. K., Weisbach, J. A., Douglas, B., Raffauf, R. F. and Ribeiro, O. (1964) *Chem. Ind. (London)* 1193.
7. Schmutz, J. and Wittwer, H. (1960) *Helv. Chim. Acta* **43,** 793.
8. Govindachari, T. R., Joshi, B. S., Saksena, A. K., Sathe, S. S. and Viswanathan, N. (1965) *Tetrahedron Letters* 3873.
9. Kupchan, S. M., Cassady, J. M. and Telang, S. A. (1966) *Tetrahedron Letters* 1251.
10. Verkey, E. T., Pillay, P. P., Bose, A. K. and Das, K. G. (1966) *Indian J. Chem.* **4,** 332.
11. Weisbach, J. A., Raffauf, R. F., Ribeiro, O., Macko, E. and Douglas, B. (1963) *J. Pharm. Sci.* **52,** 350.
12. Ondetti, M. A. and Deulofeu, V. (1961) *Tetrahedron* **15,** 160.
13. Ducke, A. (1959) *Anais Acad. Brasil. Ciênc.* **31,** 211.