Pergamon

Phytochemistry, Vol. 42, No. 5, pp. 1485–1488, 1996

0031-9422/96 $15.00 + 0.00

S0031-9422(96)00120-3

TWO PYRROLOQUINAZOLINES FROM *ADHATODA VASICA*

RAJINDER K. THAPPA, SHRI G. AGARWAL,* KANYA L. DHAR, VIVEK K. GUPTA† and KEDAR N. GOSWAMI†

Regional Research Laboratory (CSIR), Jammu Tawi-180 001, India; †Department of Physics, University of Jammu, Jammu Tawi-180 001, India

(*Received in revised form* 18 *December* 1995)

Key Word Index—*Adhatoda vasica*; Acanthaceae; leaves; alkaloids; desmethoxyaniflorine; 7-methoxyvasicinone; 5-methoxyvasicinone.

Abstract—Two new pyrroloquinazoline alkaloids, *viz.* 1,2,3,9-tetrahydropyrrolo(2,1-b)-quinazolin-9-one-3*R*-hydroxy-3(2′-dimethylamino phenyl (desmethoxyaniflorine) and 7-methoxy-3*R*-hydroxy-1,2,3,9-tetrahydropyrrolo-[2,1-b]-quinazolin-9-one (7-methoxyvasicinone), together with several known compounds were isolated from the leaves of *Adhatoda vasica*. Their structures were established by spectroscopic and X-ray diffraction analyses.

INTRODUCTION

Adhatoda vasica is a highly valued Ayurvedic medicinal plant used for the treatment of asthma, coughs, bronchitis and tuberculosis. The plant is a rich source of the quinazoline alkaloids, vasicine, vasicinone, deoxyvasicinone, vasicol, adhavasicinone and some minor compounds in the same series [1–8]. This paper describes the isolation and structural elucidation of two new quinazolines, together with other known constituents from *A. vasica* leaves.

RESULTS AND DISCUSSION

The ethanolic extract of dried leaves was fractionated as reported in the Experimental to give the two new pyrroloquinazolines, **1** and **2**, together with 5-methoxyvasicinone, vasicine and vasicinone.

Compound **1**, analysed for $C_{19}H_{19}O_2N_3$, $[M]^+$ at *m/z* 321. ^1H and ^{13}C NMR spectra combined with mass spectral data established the existence of a 3-substituted quinazoline system containing one dimethylamino and one phenyl group. A fragment at *m/z* 303 and a signal at δ 83.5 in the ^{13}C NMR indicated the presence of a tertiary hydroxyl group, because it did not form an acetate with Ac_2O–pyridine. By comparison of ^1H and ^{13}C NMR signals of **1** with those of vasicinone, the phenyl ring was placed at C-3, as the signal due to the H-3 of vasicinone is absent in the new compound **1**. The phenyl ring also carried a dimethylamino group (δ 2.75) which should be at C-2, *ortho* to the carbon joining the C-3 of the vasicinone moiety; the ^1H and ^{13}C NMR of compound **1** are very similar to those of aniflorine [9]. This position would also explain the steric hindrance of the C-3 OH towards dehydrating and acetylating agents. The presence of a doublet at δ 8.26 (J = 7.96 Hz) was characteristic of a C-8 proton *peri* to the amide carbonyl of a quinazoline moiety. Ring protons could be assigned unequivocally from its homonuclear COSY and chemical shift-correlated spectral data. Two separate multiplets appearing at δ 4.28 and 4.31 were assigned to H-1a and H-1b, while a diffuse multiplet at δ 2.65 was assigned to H-2ab. ^1H–^1H COSY also indicated interaction of H-1a and H-1b with the H-2 protons. The region between δ 7.0 and 8.20 contained all the aromatic protons. A two-proton signal at δ 7.61 having the multiplicity of an overlapped doublet and a triplet, was assigned to H-5 and H-6; this signal showed linkages with signals at δ 8.26 (H-8) and 7.4 (multiplet). The multiplet at δ 7.4 was thus assigned to H-7. The spectra also clearly exhibited that H-5 is weakly coupled to the neighbouring protons and has the same chemical shift as that of H-6, indicating possibly magnetically equivalent nuclei. The protons of the ring D were also assigned similarly. A triplet at δ 7.13 (H-4′) showed linkages with a doublet at δ 7.0 (H-3′) and a triplet at δ 7.31 (H-5′). Furthermore, the signal at δ 7.31 (H-5′) also showed linkage with a doublet at δ 7.41 (H-6′), confirming all the assignments. Thus, the structure of **1** was established as 1,2,3,9-tetrahydropyrrolo-(2,1-b)-quinazolin-9-one-3*R*-hydroxy-3(2-dimethylamino)phenyl or desmethoxy-aniflorine. Spectral data were in agreement with those reported for aniflorine [9], except for the methoxyl.

The structure of the substituents at C-3 was assigned by X-ray crystallographic studies of **1**. The structure was solved by direct methods and refined to an *R*-index of 0.041 for 1688 observed reflections. The atoms constituting the ring D are coplanar with a maximum deviation of +0.012(4) Å for C-4 from the mean plane,

*Author to whom correspondence should be addressed.

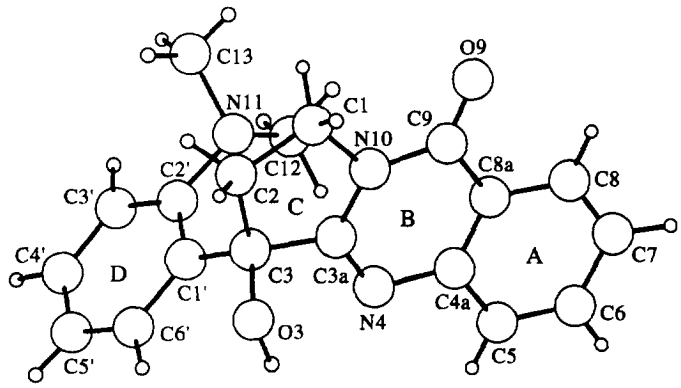

Fig. 1. PLUTO (12) plot showing the atomic arrangement and numbering in compound **1**.

thereby confirming the aromatic character of the D ring. The smaller values of torsion angles for rings A and D support rigid configurations of the benzene rings. The dihedral angle between the planes of the phenyl and pyrroloquinazoline rings is 90.12(7)° showing that they are approximately perpendicular to each other. The short intramolecular distance C-8-O-9 = 2.904(4) Å indicates a possible C–H–O interaction. The molecular structure of **1** is illustrated in Fig. 1; bond-distances and bond-angles involving non-hydrogen atoms are given in Table 1.

Compound **2** analysed for $C_{12}H_{12}N_2O_3$, $[M]^+$ *m/z* 232. IR bands at 1675 and 1630 cm^{-1} indicated the presence of a pyrroloquinazolone moiety [4] carrying a methoxyl group as shown by the mass spectral fragment at *m/z* 217 $[M-15]^+$. The ^1H NMR, IR and

Table 1. Bond distances (Å) and bond angles (°), with e.s.d.s in parentheses, for the non-hydrogen atoms of compound **1**.

C1-C2	1.518(5)	C8a-C9	1.449(3)
C1-N10	1.467(3)	C9-O9	1.236(4)
C2-C3	1.542(4)	C9-N10	1.372(4)
C3-O3	1.421(4)	N10-C3a	1.376(4)
C3-C1′	1.527(4)	N11-C12	1.461(5)
C3-C3a	1.508(4)	N11-C13	1.472(5)
C3a-N4	1.283(3)	C1′-C6′	1.394(5)
C4a-N4	1.395(4)	C1′-C2′	1.392(4)
C4a-C8a	1.409(4)	C2′-C3′	1.388(5)
C5-C4a	1.399(4)	C2′-N11	1.436(5)
C5-C6	1.374(5)	C3′-C4′	1.369(7)
C6-C7	1.392(5)	C4′-C5′	1.369(7)
C7-C8	1.363(5)	C5′-C6′	1.396(6)
C8-C8a	1.404(4)		
N10-C1-C2	104.3(2)	C8-C8a-C9	120.9(3)
C1-C2-C3	108.1(2)	C8a-C9-N10	113.8(2)
C3a-C3-C2	104.0(2)	C8a-C9-O9	125.9(3)
C2-C3-C1′	113.6(2)	O9-C9-N10	120.3(2)
C2-C3-O3	107.3(2)	C9-N10-C1	123.3(2)
C3a-C3-C1′	112.0(2)	C9-N10-C3a	122.9(2)
C3a-C3-O3	107.5(2)	C3a-N10-C1	113.5(2)
O3-C3-C1′	111.9(3)	C2′-N11-C12	112.6(3)
N10-C3a-C3	109.8(2)	C2′-N11-C13	112.4(3)
N10-C3a-N4	125.2(3)	C12-N11-C13	111.4(3)
N4-C3a-C3	124.8(2)	C3-C1′-C2′	120.4(3)
C4a-N4-C3a	116.2(2)	C3-C1′-C6′	120.6(3)
C5-C4a-N4	118.6(2)	C6′-C1′-C2′	118.9(3)
C5-C4a-C8a	119.3(3)	C1′-C2′-C3′	119.5(3)
C8a-C4a-N4	122.1(2)	C3′-C2′-N11	121.9(3)
C6-C5-C4a	119.5(3)	C1′-C2′-N11	118.6(3)
C7-C6-C5	121.5(3)	C4′-C3′-C2′	121.3(4)
C8-C7-C6	119.7(3)	C5′-C4′-C3′	119.9(4)
C7-C8-C8a	120.5(3)	C6′-C5′-C4′	120.0(4)
C8-C8a-C4a	119.5(3)	C1′-C6′-C5′	120.3(3)
C4a-C8a-C9	119.5(3)		

mass spectra of **2** were very similar to those of vasicinone, except for the appearance of the signal arising from a methoxyl group. Compound **2** was thus identified as 7-methoxyvasicinone. The methoxyl group was placed at C-7 because of the presence of a H-8 *meta*-coupled doublet at δ 7.6 (J = 2 Hz). In 5-methoxy-vasicinone, the H-8 proton appeared as a doublet at δ 8.0 (J = 10 hz). Incidently, 5-methoxy-vasicinone was also isolated from another of the fractions after repeated chromatography. The two compounds, 7-methoxyvasicinone and 5-methoxy-vasicinones had similar ^{1}H NMR spectra, except for H-8, and differ only in the position of the signals in the aromatic region. X-ray crystallographic data also confirmed the position of the methoxyl at C-7 and the compound was identified as 7-methoxy-3*R*-hydroxy-1,2,3,9-tetrahydro-pyrrolo [2,1-b]-quinazolin-9-one.

EXPERIMENTAL

General.. Mps uncorr. optical rotations $CHCl_3$. UV in MeOH. IR in KBr disc. ^{1}H NMR (90 MHz, 500 MHz, $CDCl_3$), ^{13}C NMR (25 MHz) with TMS as int. standard. (2D expts (^{1}H-^{1}H COSY) were carried out using a standard pulse sequence. EI-MS probe, 70 eV. CC, silica gel and neutral alumina.

Plant material and isolation procedure. Whole plants of *A. vasica* (Nees), predominantly containing leaves, were collected from Jammu in January and February and dried in the shade. A voucher specimen is deposited at RRL-Herbarium (RRL Acc. No. 16987). Dried leaves (50 kg) were ground to a coarse powder and extracted with EtOH (200 l × 4) by cold percolation. The EtOH extract was concd and the residue treated with dil. HCl for separation of alkaloids. The acid layer was extracted ×3 with $CHCl_3$ to remove non-alkaloidal material. The acid layer was then basified with NH_4OH and the soln extracted with $CHCl_3$. The residue obtained upon recrystallization from MeOH gave vasicine (200 g). The mother liquor remaining was slurried with silica gel and extracted with *n*-hexane, benzene, $CHCl_3$, EtOAc and MeOH in a Soxhlet apparatus. The benzene and $CHCl_3$ extracts were mixed and chromatographed over neutral alumina. Frs eluted with benzene–$CHCl_3$ (1:1) were pooled and rechromatographed over silica gel. Elution with benzene–$CHCl_3$ (1:3) resulted in frs rich in **1**. Pooled frs on recrystallization from Me_2CO gave thick crystals (230 mg). The $CHCl_3$ frs from the alumina column were concd and upon recrystallization from MeOH gave **2** (needles, 450 mg). The mother liquor remaining showed two spots on TLC which on rechromatography over alumina gave **3** (350 mg) after elution with $CHCl_3$.

Desmethoxyaniflorine. Mp 128–129°. UV λ_{max}^{MeOH} nm, 203, 223, 236, 305, 314. IR ν_{max}^{KBr} cm^{-1} 3490, 1610, 1660. EI-MS (rel. int.) *m/z*: 321, [M]$^+$ (89), 303 (54), 302 (49), 287 (41), 271 (50), 183 (10), 162 (15). ^{1}H NMR: δ 2.60–2.70 (*m*, 2H, C-2H), 2.76 [*s*, 6H, N Me_2], 4.17–4.22 (*m*, 1H, C-1H$_A$), 4.27–4.32 (*m*, 1H, C-1H$_B$, 7.00 (*d*, *br*, J = 7.62 Hz, 1H, 3-H), 7.13 (*t*, *br*, J = 7.62 Hz, 1H, 4′-H), 7.31 (*t*, *br*, 1H, 5′-H), 7.41 (*t*, J = 8.61, 1H, 6′-H), 7.6 (*d*, J = 2.26 Hz, 1H, 5-H), 7.6 (*t*, J = 8.6 Hz, 1H, 6-H), 7.40 (*m*, 8 lines, 1H, 7-H), 8.26 (*dd*, J = 7.96, 2.26, 1H, 8-H). ^{13}C NMR: δ 37.6 (*t*, C-1), 42.9 (*t*, C-2), 45.1 [2 × *q*, N $(CH_3)_2$], 83.5 (*s*, C-3), 121.1 (*s*, N = C), 123.2 (*d*), 125.2 (*d*), 126.1 (*d*), 126.5 (*d*), 127 (*d*), 128.8 (*d*), 133.2 (2 × *d*), 137.2 (*s*), 149.8 (*s*), 152.1 (*s*), 161.3 (*s*), 162.2 (*s*).

7-Methoxyvasicinone. Mp 199–202°. UV λ_{max}^{MeOH} 212, 230, 288, 315, 326 nm. IR ν_{max}^{KBr} cm^{-1} 3210, 1670, 1630, 1500, 1350. ^{1}H NMR: δ 2.41 (2H, *m*, C-2H), 3.93 (3H, *m*, (C-1H), 4.25 (2H, *m*, C-1H), 5.26 (1H, *t*, C-3H), 7.41–7.76 (4 aromatic protons) ^{13}C NMR: δ 29.7 (*t*), 43.8 (*t*) 56.1 (*q*, OMe), 71.9 (*d*, C-3), 106.5 (*d*) 125.0 (*d*) 128.4 (*d*) 122.0 (*s*), 143.5 (*s*), 169.7 (*s*), 171.1 (*s*).

5-Methoxyvasicinone. Recrystallized from MeOH, mp 220–222°. Analysed for $C_{12}H_{12}N_2O_3$, [M]$^+$ *m/z* 232. UV λ_{max}^{MeOH} nm 214, 238, 284, 314, 330. IR ν_{max}^{KBr} cm^{-1} 1675, 1630. ^{1}H NMR: δ 2.35 (*m*, 1H), 3.88 (*s*, 3H, OMe) 4.25 (*m*, 2H), 5.2 (*t*, 1H, C-3H) 7.2–7.4 (2 aromatic protons) 7.99 (*dd*, J = 8 Hz and 2 Hz).

Three-dimensional X-ray intensity data were collected on a Enraf-Noniius CAD-4 diffractometer with Mo$K\alpha$ radiation using a crystal of dimensions 0.32 × 0.32 × 0.08 mm ($\sin\theta/\lambda$ = 0.595 Å^{-1}; a $\omega/2\theta$ scan

Table 2. Crystal data and other experimental details of compound **1**

Chemical formula	$C_{19}H_{19}N_3O_2 \cdot H_2O$
Molecular weight	339.39
Cell parameters	a = 8.537(2) Å
	b = 16.766(3) Å
	c = 12.859(7) Å
	β = 107.04(2)°
Unit cell volume	1759.74 Å^3
Crystal system	Monoclinic
Space group	$P2_1/c$
Density (measured)	D_m = 1.297 g cm^{-3}
Density (calculated)	D_c = 1.281 g cm^{-3}
No. of molecules per unit cell	Z = 4
Wavelength (λ)	0.71073 Å
Absorption coefficient	μ(Mo$K\alpha$) = 0.082 mm^{-1}
F(000)	720
Crystal size	0.32 × 0.32 × 0.08 mm
Refinement of unit cell	25 reflections, 7.1 ≤ θ ≤ 12.6°
Scan mode	ω-2θ
Range of θ	2 ≤ θ ≤ 25°
No. of standard reflections	2($\bar{4}$ $\bar{5}$ 3, $\bar{3}$ $\bar{6}$ 2)
No. of measured reflections	3443
No. of unique reflections	2668
No. of observed reflections	1688[$I > -2.5\sigma(I)$]
R_{int}	0.013
No. of parameters refined	310
R	0.041
R_w	0.042
Weight	$2.147/[\sigma^2(F_0) + 0.00112(F_0)^2]$
Final residual electron density	$-0.146 \leq \Delta\rho \leq 0.215$ e Å^{-3}
Max. Δ/σ in the final cycle for all atoms	0.174

Table 3. Atomic coordinates and equivalent isotropic temperature factors $(Å)^2$ with e.s.d.s in parentheses, for the nonhydrogen atoms of compound **1**

Atom	x	y	z	U^*_{eq}
C1	0.6895(4)	0.6935(2)	0.0606(2)	0.0513(12)
C2	0.6042(4)	0.7702(2)	0.0758(2)	0.0518(12)
C3	0.6617(3)	0.7921(2)	0.1976(2)	0.0416(9)
C3a	0.7735(3)	0.7243(2)	0.2495(2)	0.0371(11)
O3	0.7595(3)	0.8619(1)	0.2083(2)	0.0561(9)
N4	0.8490(3)	0.7187(1)	0.3513(2)	0.0419(8)
C4a	0.9564(3)	0.6545(2)	0.3827(2)	0.0420(10)
C5	1.0489(4)	0.6481(2)	0.4919(3)	0.0575(12)
C6	1.1579(4)	0.5862(2)	0.5239(3)	0.0693(14)
C7	1.1756(4)	0.5283(2)	0.4506(3)	0.0669(15)
C8	1.0860(4)	0.5336(2)	0.3440(3)	0.0564(13)
C8a	0.9759(3)	0.5969(2)	0.3077(2)	0.0426(10)
C9	0.8864(3)	0.6054(2)	0.1936(2)	0.0411(10)
O9	0.8916(3)	0.5590(1)	0.1200(2)	0.0589(9)
N10	0.7920(3)	0.6730(1)	0.1704(2)	0.0382(8)
N11	0.4530(3)	0.6625(2)	0.2137(2)	0.0557(11)
C12	0.5096(6)	0.6048(2)	0.3019(4)	0.0730(17)
C13	0.3142(6)	0.6310(3)	0.1263(4)	0.0861(20)
C1′	0.5214(3)	0.8028(2)	0.2476(2)	0.0425(10)
C2′	0.4184(3)	0.7392(2)	0.2515(2)	0.0507(10)
C3′	0.2864(4)	0.7513(3)	0.2920(3)	0.0702(15)
C4′	0.2547(5)	0.8248(3)	0.3277(3)	0.0769(17)
C5′	0.3569(5)	0.8875(3)	0.3266(3)	0.0711(15)
C6′	0.4903(4)	0.8771(2)	0.2860(2)	0.0564(13)
O1w	0.9321(5)	0.5789(2)	−0.0923(2)	0.0816(13)

$^*U_{eq} = (1/3)\Sigma_i \Sigma_j u_{ij} a_i^* a_j^* a_i . a_j$.

mode was employed. Cell parameters were refined from accurately determined 2θ values of 25 reflections in the range $7.1 \leqslant \theta \leqslant 12.6°$. A total of 3443 reflections were collected for values of θ up to 25 and out of those reflections 2668 were unique ($0 \leqslant h \leqslant 19$, $0 \leqslant k \leqslant 19$, $-15 \leqslant l \leqslant 14$); 1668 reflections have intensities greater than 2.5 $\sigma(1)$ and were used in the subsequent analysis. Two monitoring reflections (4 5 3, 3 6 2) showed no significant intensity variations throughout data collection. Intensity data were corrected for Lorentz polarization factors but no absorption and extinction corrections were made. Crystallographic data are summarized in Table 2. The structure was solved by direct methods using SHELXS86 [10]. Isotropic refinement of the structure by least-squares methods using SHELX76 [11] was followed by anisotropic refinement of all non-H atoms. All H atoms were located from a difference-Fourier map and their positions and isotropic displacement parameters were refined. Finally, the R-factor lowered down to 0.041 and $wR = 0.042$. Atomic scattering factors were as contained in SHELX76. Atomic coordinates and thermal parameters for nonhydrogen atoms are given in Table 3.

REFERENCES

1. Atal, C. K. (1981) *Chemistry and Pharmacology of Vasicine – A New Oxytocic and Abortifacient*. Regional Research Laboratory Jammu, India.
2. Pandita, K., Bhatia, M. S., Thappa, R. K., Agarwal, S. G., Dhar, K. L. and Atal, C. K. (1983) *Planta Med.* **48**, 81.
3. Dhar, K. L., Jain, M. P., Kaul, S. K. and Atal, C. K. (1981) *Phytochemistry* **20**, 319.
4. Jain, M. P., Kaul, S. K., Dhar, K. L. and Atal, C. K. (1980) *Phytochemistry* **19**, 188.
5. Chowdhury, B. K. and Bhattacharyya, P. K. (1987) *Chem. Ind.* 35.
6. Chowdhury, B. K. and Bhattacharyya, P. K. (1985) *Phytochemistry* **24**, 3082.
7. Johne, S. (1984) Quinazoline Alkaloids. In *Progress in the Chemistry of Organic Natural Products*, Vol. 46 (L. Zeichmeister, ed.) Springer, p. 174. New York.
8. Hermecz, I. and Vasvari-Debreczy, L. (1986) In *Advances in Heterocyclic Chemistry* Vol. 39 (A. A. Katritzky, ed.), p. 317. Academic Press.
9. Arndt, R. R., Eggers, S. H. and Jordan A. (1967) *Tetrahedron* **23**, 3521.
10. Sheldrick, G. M. (1986) SHELXS86. Program for the solution of crystal structures, University of Gottingen, Germany.
11. Sheldrick, G. M. (1976) SHELX76. Program for crystal structure determination, University of Cambridge, U.K.
12. Motherwell, W. D. S. and Clegg, W. (1978) PLUTO, Program for plotting molecular and crystal structures, University of Cambridge, U.K.