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## <sup>1</sup>H- AND <sup>13</sup>C-NMR ASSIGNMENTS FOR SOME PYRROLO[2,1b]- QUINAZOLINE ALKALOIDS OF *ADHATODA VASICA*

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**ABSTRACT.**—From the leaves of *Adhatoda vasica*, the pyrrolo[2,1b]quinazoline alkaloids *l*-vasicine [1], *l*-vasicinone [2], *l*-vasicol [3], anisotine [7], 3-hydroxyanisotine [8], and a new alkaloid, vasnetine [9] have been isolated and their structures established by <sup>1</sup>H- and <sup>13</sup>C-nmr spectral studies. The chemical shift assignments for these alkaloids were confirmed by <sup>1</sup>H homonuclear COSY, DEPT, HETCOR, selective INEPT and HMBC nmr experiments.

A number of plants belonging to the families Acanthaceae, Cruciferae, Malvaceae, and Rutaceae are known to contain quinazoline alkaloids (1). Of these, the leaves, roots, and the young plants of *Adhatoda vasica* Nees (Acanthaceae) have been extensively investigated and the alkaloids *l*- and *d*-vasicine (peganine) [1] (2–8), 7-hydroxyvasicine (9), 5-methoxyvasicine (7), *l*- and *dl*-vasicinone [2] (4,6,8), 3-deoxyvasicinone (8), vasicinone (7-hydroxyvasicinone) (6), adhavaicinone (5-methoxyvasicinone) (10), vasicol [3] (11), vasicoline [4] (5), vasicolinone [5] (5), adhatodine [6] (5), and anisotine [7] (5) have been reported.

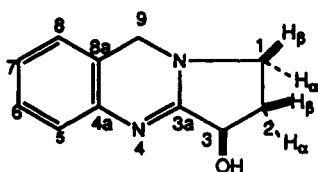
In connection with chemical studies of Indian plants for their biological activity (12), we have investigated the crude alkaloidal fractions from the leaves of *A. vasica* (Sanskrit: "Vasaka"). Extracts of the leaves are a commonly used medicine in India as an expectorant and a bronchodilator. Vasicine has been reported to be a respiratory stimulant, bronchodilator, and hypotensive (13). It has also been claimed to be a uterine stimulant and abortifacient (14). We have isolated from the leaves of *A. vasica*, the pyrrolo[2,1b]quinazoline alkaloids *l*-vasicine [1], *l*-vasicinone [2], *l*-vasicol [3], anisotine [7], 3-hydroxyanisotine [8], and a new alkaloid, vasnetine [9]. The alkaloid 8 was prepared in earlier studies (5) by the KMnO<sub>4</sub> oxidation of anisotine [7].

Varying optical rotations have been reported for *l*-vasicine: [α]<sub>D</sub> –254°, –210° (CHCl<sub>3</sub>) (15), –61.5° (EtOH) (15), –173° to –177° (CHCl<sub>3</sub>) (4,16); for *d*-vasicine (peganine) [α]<sub>D</sub> +162.5° (CHCl<sub>3</sub>), isolated from *Galega officinalis* (17), and [α]<sub>D</sub> +163° to +203° (CHCl<sub>3</sub>), obtained by resolution (18). Vasicine isolated by us had mp 211–212° and [α]<sub>D</sub> –210° (CHCl<sub>3</sub>). Variations of these optical rotations are due to the instability of vasicine in CHCl<sub>3</sub> solution as it is known to give mixtures of *l*- and *dl*-vasicinone (4). We observed that a CDCl<sub>3</sub> solution of *l*-vasicine kept overnight in an nmr tube gave a combined spectrum of vasicine and vasicinone.

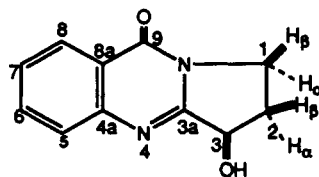
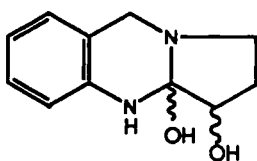
### RESULTS AND DISCUSSION

The <sup>1</sup>H-nmr spectrum of *l*-vasicine [1] is reported to exhibit signals for four aromatic protons at δ 6.8–7.3, a one-proton triplet at δ 4.80, two proton multiplets representing two protons centered at δ 2.80 and 3.50 assigned to the C-2 and C-1 protons, respectively, and a two-proton singlet at δ 4.62 assigned to the C-9 protons (19). All these protons have not been accurately assigned. The <sup>13</sup>C-nmr spectral assignments of 1 recorded earlier (20) agree with our present findings. Table 1 gives the <sup>1</sup>H- and <sup>13</sup>C-nmr chemical shift assignments of 1 and these have been confirmed by DEPT, <sup>1</sup>H-<sup>1</sup>H

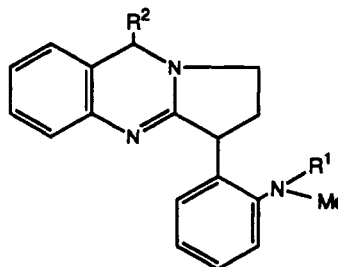
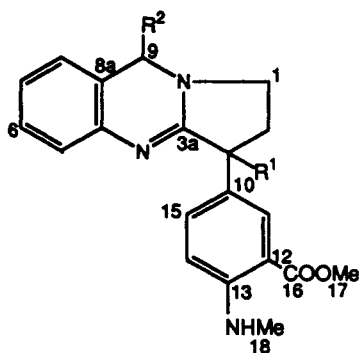
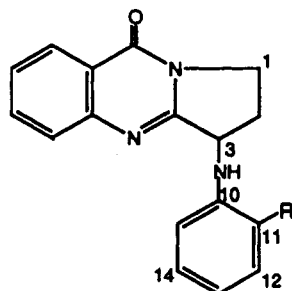
<sup>1</sup>Schering-Plough Research Institute, 2015 Galloping Hill Road, Kenilworth, New Jersey 07033.



1 Vasicine

2 Vasicinone ( $R^1 = \beta\text{-OH}$ ;  $R^2 = R^3 = \text{H}$ )

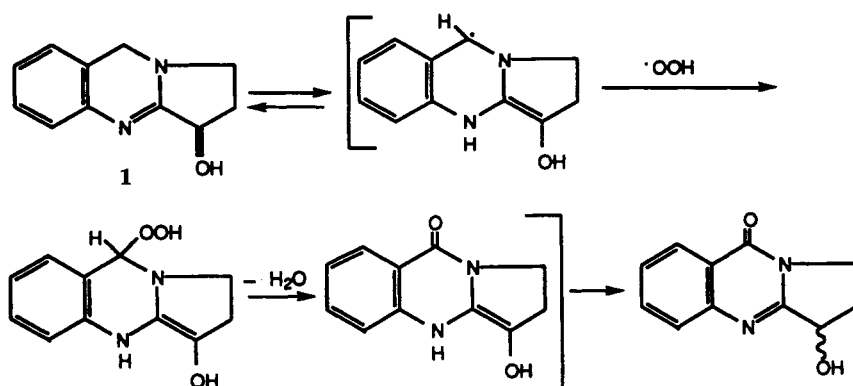
3 Vasicol

4 Vasicoline ( $R^1 = R^2 = \text{H}$ )5 Vasicolinone ( $R^1 = \text{Me}$ ;  $R^2 = \text{O}$ )6 Adhatodine ( $R^1 = R^2 = \text{H}$ )7 Anisotine ( $R^1 = \text{H}$ ;  $R^2 = \text{O}$ )8 3-Hydroxyanisotine ( $R^1 = \text{OH}$ ;  $R^2 = \text{O}$ )9 Vasnetine ( $R = \text{COOMe}$ )10 Anisessine ( $R = \text{COOEt}$ )

COSY, and HETCOR experiments. The absolute stereochemistry of *l*-vasicine is reported to be 3*R* as determined by an X-ray crystal structure determination of its hydrochloride (16).

Different optical rotations have been given for *l*-vasicinone [2]:  $[\alpha]_D -100^\circ$  (EtOH) (4),  $-58^\circ$  ( $\text{CHCl}_3$ ) (4), and  $-129^\circ$  (21). *l*-Vasicine on autoxidation or with 30%  $\text{H}_2\text{O}_2$  is known to give a mixture of *l*- and *dl*-vasicinone indicating that racemization and oxidation take place simultaneously (4). A plausible mechanism is probably that the benzylic radical stabilized by resonance picks up molecular oxygen of air and autoxidizes to the hydroperoxide which by the loss of an  $\text{H}_2\text{O}$  molecule gives racemic vasicinone as indicated in Scheme 1.

The isolation of (+)-vasicinone [2] from the leaves of *A. vasica* has been recently claimed for the first time (22). The  $^1\text{H}$ -nmr spectrum of 2 [ $\text{CF}_3\text{COOH}$  (1),  $\text{CD}_3\text{SOCD}_3$  (11)] is reported to show the aromatic protons at ca.  $\delta$  7.30–7.80, the H-8 proton at ca.  $\delta$  8.05, the H-3 proton at  $\delta$  5.56, the H-2 protons at ca.  $\delta$  2.21, 2.64, and the H-1 protons at ca.  $\delta$  3.92, 4.32. In the present study, all the protons and carbon chemical shifts of 1 and 2 have been unambiguously assigned and these assignments have been confirmed by DEPT, COSY, and HETCOR nmr experiments as shown in Tables 1 and 2. The selective INEPT nmr technique provides important information, since by selection of the pulse delay, three-bond  $^1\text{H}$ - $^{13}\text{C}$  connectivities can be established (23). Accurate assignments



SCHEME 1

of the non-protonated carbons of *l*-vasicinone have been deduced using this technique and are given in Table 3. The high-field  $^1\text{H}$ -nmr spectra of *l*-vasicine [**1**] and *l*-vasicinone [**2**] for the protons H-1 $_{\alpha}$ , H-1 $_{\beta}$ , H-2 $_{\alpha}$ , H-2 $_{\beta}$ , and H-3 $_{\alpha}$  show a first-order splitting pattern as shown in Figure 1. The  $^1\text{H}$ -nmr spectrum of vasicine [**1**] was calculated utilizing the spin simulation program (24) provided by Varian Associates and the observed and calculated spectra are portrayed in Figure 1 (A) and (B). The calculated and observed spectra are in excellent agreement. An analysis of the multiplicities for these alkaloids is given in Tables 1 and 2.

Vasicol [**3**] was isolated by Dhar and coworkers (11) from the roots of *A. vasica* and they derived its structure on the basis of spectral and chemical studies. We isolated this alkaloid as a viscous liquid and carried out detailed  $^1\text{H}$ - and  $^{13}\text{C}$ -nmr studies as shown in Table 4. The multiplet at  $\delta$  3.18 (H-1 $_{\alpha}$ ) was shown to be coupled with the vicinal

TABLE 1.  $^1\text{H}$ - and  $^{13}\text{C}$ -Nmr Chemical Shift Assignments of Vasicine [**1**] ( $\text{CDCl}_3$ ).<sup>a</sup>

Position	$^{13}\text{C}$ $\delta$	DEPT	$^1\text{H}$	$\delta$	Multiplicity ( $J$ , Hz)	COSY
1 . . . . .	48.1	t	1 $_{\alpha}$	3.33	dt or ddd $J_{1_{\alpha},1_{\beta}}=9.9$ $J_{1_{\alpha},2_{\beta}}=7.1$ $J_{1_{\alpha},2_{\alpha}}=7.3$	H-1 $_{\beta}$ , H-2 $_{\alpha}$ , H-2 $_{\beta}$
			1 $_{\beta}$	3.43	ddd $J_{1_{\beta},1_{\alpha}}=9.9$ $J_{1_{\beta},2_{\beta}}=8.6$ $J_{1_{\beta},2_{\alpha}}=3.9$	H-1 $_{\alpha}$ , H-2 $_{\alpha}$ , H-2 $_{\beta}$
2 . . . . .	28.8	t	2 $_{\alpha}$	2.43	dddd $J_{2_{\alpha},2_{\beta}}=13.1$ $J_{2_{\alpha},1_{\alpha}}=7.3$ $J_{2_{\alpha},3_{\alpha}}=7.8$ $J_{2_{\alpha},1_{\beta}}=3.9$	H-1 $_{\alpha}$ , H-1 $_{\beta}$ , H-2 $_{\beta}$ , H-3 $_{\alpha}$
			2 $_{\beta}$	2.12	dddd $J_{2_{\beta},2_{\alpha}}=13.1$ $J_{2_{\beta},1_{\beta}}=8.6$ $J_{2_{\beta},1_{\alpha}}=7.1$ $J_{2_{\beta},3_{\alpha}}=6.8$	H-2 $_{\alpha}$ , H-1 $_{\beta}$ , H-1 $_{\alpha}$ , H-3 $_{\alpha}$
3 . . . . .	70.2	d	3 $_{\alpha}$	4.83	dd $J_{3_{\alpha},2_{\alpha}}=7.8$ $J_{3_{\alpha},2_{\beta}}=6.8$	H-2 $_{\alpha}$ , H-2 $_{\beta}$
3a . . . . .	163.9	s	3a	—		
4a . . . . .	142.3	s	4a	—		
5 . . . . .	123.6	d	5	7.14	m	
6 . . . . .	128.3	d	6	7.14	m	H-7
7 . . . . .	124.0	d	7	6.96	m	H-6, H-8
8 . . . . .	125.7	d	8	6.84	d $J_{8,7}=7.5$	H-8, H-9
8a . . . . .	119.0	s	8a	—		
9 . . . . .	47.0	t	9	4.53	br s	H-8

<sup>a</sup> $^1\text{H}$ - and  $^{13}\text{C}$ -nmr assignments were established by HETCOR experiment.

TABLE 2.  $^1\text{H}$ - and  $^{13}\text{C}$ -Nmr Chemical Shift Assignments of Vasicinone [2] ( $\text{CDCl}_3$ ).<sup>a</sup>

Position	$^{13}\text{C}$ $\delta$	DEPT	$^1\text{H}$	$\delta$	Multiplicity ( $J$ , Hz)	COSY
1	43.4	t	1 <sub>a</sub>	4.02	dt or ddd $J_1 1_{a,1_{\beta}}=12.3$ $J_2 1_{a,2_{\beta}}=7.6$ $J_3 1_{a,2_{\alpha}}=7.6$	H-1 <sub><math>\beta</math></sub> , H-2 <sub><math>\alpha</math></sub> , H-2 <sub><math>\beta</math></sub>
			1 <sub><math>\beta</math></sub>	4.38	ddd $J_1 1_{\beta,1_{a}}=12.3$ $J_2 1_{\beta,2_{\beta}}=8.8$ $J_3 1_{\beta,2_{\alpha}}=4.0$	H-1 <sub><math>\alpha</math></sub> , H-2 <sub><math>\alpha</math></sub> , H-2 <sub><math>\beta</math></sub>
2	29.3	t	2 <sub><math>\alpha</math></sub>	2.68	dtd or dddd $J_1 2_{\alpha,2_{\beta}}=13.2$ $J_2 2_{\alpha,1_{a}}=7.6$ $J_3 2_{\alpha,3_{\alpha}}=7.6$ $J_4 2_{\alpha,1_{\beta}}=4.0$	H-2 <sub><math>\beta</math></sub> , H-1 <sub><math>\alpha</math></sub> , H-1 <sub><math>\beta</math></sub> , H-3 <sub><math>\alpha</math></sub>
			2 <sub><math>\beta</math></sub>	2.31	dddd $J_1 2_{\beta,2_{\alpha}}=13.2$ $J_2 2_{\beta,1_{\beta}}=8.8$ $J_3 2_{\beta,1_{a}}=7.6$ $J_4 2_{\beta,3_{\alpha}}=7.6$	H-2 <sub><math>\alpha</math></sub> , H-1 <sub><math>\beta</math></sub> , H-1 <sub><math>\alpha</math></sub> , H-3 <sub><math>\alpha</math></sub>
3	71.6	d	3 <sub><math>\alpha</math></sub>	5.25	dd or t $J_1 3_{\alpha,2_{\beta}}=7.6$ $J_2 3_{\alpha,2_{\alpha}}=7.6$	H-2 <sub><math>\alpha</math></sub> , H-2 <sub><math>\beta</math></sub>
3a	160.7	s	3a	—		
4a	148.3	s	4a	—		
5	126.4	d	5	7.75	m	H-7
6	134.4	d	6	7.75	m	H-7, H-8
7	126.9	d	7	7.48	dt $J_1 7,8=7.5$ $J_2 7,6=7.2$ $J_3 7,5=2.0$	H-6, H-8
8	126.5	d	8	8.28	d $J 8,7=7.5$	H-6, H-7
8a	120.8	s	8a	—		
9	160.5	s	9	—		

<sup>a</sup> $^1\text{H}$ - and  $^{13}\text{C}$ -nmr assignments were established by HETCOR experiment.

methine at  $\delta$  3.29 (H-1 <sub>$\beta$</sub> ) and the adjacent methylene protons at  $\delta$  1.92 (H-2 <sub>$\beta$</sub> ) and  $\delta$  2.39 (H-2 <sub>$\alpha$</sub> ) by double resonance experiments. Irradiation at  $\delta$  3.29 (H-1 <sub>$\beta$</sub> ) showed changes of the signals assigned to H-1 <sub>$\alpha$</sub> , H-2 <sub>$\alpha$</sub> , and H-2 <sub>$\beta$</sub> . Similarly, double resonance experiments showed that H-2 <sub>$\beta$</sub>  ( $\delta$  1.92) is coupled with H-2 <sub>$\alpha$</sub> , H-1 <sub>$\alpha$</sub> , H-1 <sub>$\beta$</sub> , and H-3 ( $\delta$  4.46); also, irradiation of H-2 <sub>$\alpha$</sub>  showed changes in H-2 <sub>$\beta$</sub> , H-1 <sub>$\alpha$</sub> , H-1 <sub>$\beta$</sub> , and H-3. Double resonance experiments showed that H-3 is coupled to H-2 <sub>$\alpha$</sub>  and H-2 <sub>$\beta$</sub>  protons. Irradiation at  $\delta$  4.35 did not bring about change of any of the signals, indicating that these protons may be assigned to H-9. The H-9 protons are seen as two doublets at  $\delta$  4.31 and  $\delta$  4.38 ( $J=15$  Hz) constituting an AB spectrum ascribable to geminal coupling between the two non-equivalent protons (25). Double resonance experiments established the assignments of the aromatic protons H-5, H-6, H-7, and H-8. Selective INEPT nmr data given in Table 5 confirm the non-protonated and some protonated carbon assignments.

Adhatodine [6], the C-3 aryl derivative of vasicine, was obtained from the young plants of *A. vasica* and its autoxidation product anisotine [7] was also isolated (5). The isolation of anisotine from the branches and leaves of *Anisotes sessiflorus* was reported in an earlier investigation (19). The structure of 7 was based on mass and  $^1\text{H}$ -nmr spectral

TABLE 3. Nmr Data of Vasicinone [2] from Selective INEPT Experiments.

Proton Irradiated	$\delta$	Carbon Signal Enhanced
H-8	8.28	160.5 (C-9), 148.3 (C-4a)
H-5, H-6	7.75	148.3 (C-4a), 126.5 (C-8)
H-7	7.48	126.5 (C-8), 120.8 (C-8a)
H-3	5.25	160.7 (C-3a)
H-1 <sub><math>\beta</math></sub>	4.38	160.5 (C-9), 71.6 (C-3), 29.3 (C-2)
H-1 <sub><math>\alpha</math></sub>	4.02	160.5 (C-9), 29.3 (C-2)
H-2 <sub><math>\alpha</math></sub>	2.68	160.7 (C-3a)
H-2 <sub><math>\beta</math></sub>	2.31	160.7 (C-3a), 71.6 (C-3), 43.4 (C-1)

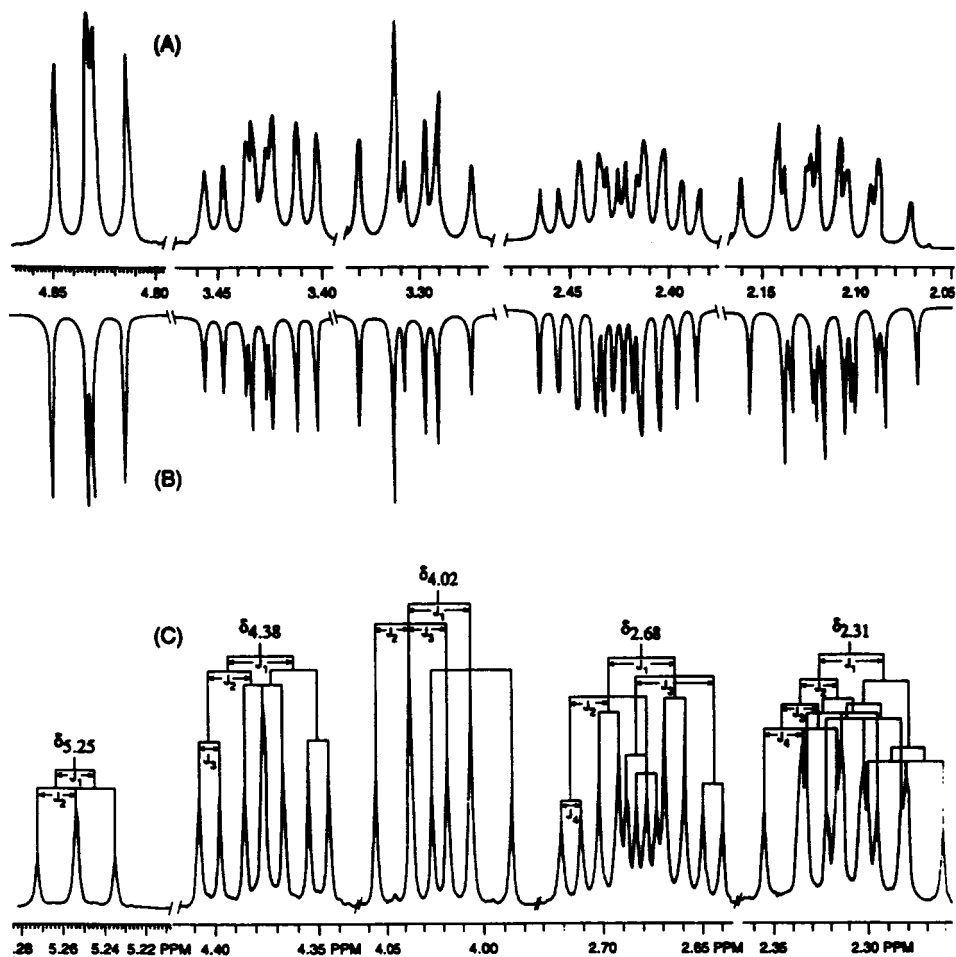


FIGURE 1. 400 MHz Spectrum (Partial) of (A) Vasicine [1] Observed, (B) Vasicine [1] Calculated, (C) Vasicinone [2] Observed.

TABLE 4.  $^1\text{H}$ - and  $^{13}\text{C}$ -Nmr Chemical Shift Assignments of Vasicol [3] ( $\text{CDCl}_3$ ).<sup>a</sup>

Position	$^{13}\text{C}$ $\delta$	DEPT	$^1\text{H}$	$\delta$	Multiplicity ( $J$ , Hz)	COSY
1 .....	43.1	t	$1_\alpha$	3.18	m	H- $1_\beta$ , H- $2_\alpha$ , H- $2_\beta$
			$1_\beta$	3.29	m	H- $2_\alpha$ , H- $2_\beta$
2 .....	27.6	t	$2_\alpha$	2.39	m	H- $1_\alpha$ , H- $1_\beta$
			$2_\beta$	1.92	m	H- $2_\beta$ , H-3
						H- $1_\alpha$ , H- $1_\beta$
						H- $2_\alpha$ , H-3
3 .....	69.9	d	3	4.46	t	H- $2_\alpha$ , H- $2_\beta$
					$3,2_\beta = 8.4$	
					$3,2_\alpha = 8.4$	
3a .....	175.2	s	3a	—		
4a .....	145.7	s	4a	—		
5 .....	115.7	d	5	6.64	d	H-6
6 .....	129.5	d	6	7.11	t	H-7, H-5
7 .....	117.3	d	7	6.66	t	H-6, H-8
8 .....	131.2	d	8	7.02	d	H-7
8a .....	118.4	s	8a	—		
9 .....	44.7	t	9	4.31, 4.38	each d	H-9a to H-9b
					( $J$ )=15	

<sup>a</sup> $^1\text{H}$ - and  $^{13}\text{C}$ -nmr assignments were established by HETCOR experiment.

TABLE 5. Nmr Data of Vasicol [3] from Selective INEPT Experiments.

Proton Irradiated	$\delta$	Carbon Signal Enhanced
H-6	7.11	145.7 (C-4a), 131.2 (C-8)
H-8	7.02	145.7 (C-4a), 44.7 (C-9)
H-5, H-7	6.64	131.2 (C-8), 117.3 (C-7), 115.7 (C-5)
H-3	4.46	175.2 (C-3a)
H-1 <sub><math>\alpha</math></sub>	3.18	27.6 (C-2), 69.9 (C-3), 175.2 (C-3a)
H-1 <sub><math>\beta</math></sub>	3.29	175.2 (C-3a)
H-2 <sub><math>\alpha</math></sub>	2.39	69.9 (C-3), 43.1 (C-1)
H-2 <sub><math>\beta</math></sub>	1.92	69.9 (C-3), 175.2 (C-3a)

studies. We have carried out  $^1\text{H}$ ,  $^1\text{H}$ - $^1\text{H}$  COSY,  $^{13}\text{C}$ , DEPT, HETCOR, and selective INEPT nmr spectral studies to establish the structure and nmr assignments of **7**. These results are summarized in Tables 6 and 7. 3-Hydroxyanisotine [**8**] was prepared by  $\text{KMnO}_4$  oxidation of anisotine [**7**] (19). We isolated **8** by chromatographic separation of the crude alkaloidal mixture and established its structure by detailed nmr investigations summarized in Tables 8 and 9. A new alkaloid, vasnetine [**9**], was isolated during chromatographic separation of the crude alkaloidal mixture. It showed the molecular ion peak at  $m/z$  335. A carbomethoxy group substituted ortho to the amino substituent of the aromatic ring was seen from the loss of  $\text{MeOH}$  ( $m/z$  303,  $\text{M}^+ - 32$ ) and  $m/z$  302 ( $\text{M}^+ - \text{H} - 32$ ), fragments arising through the "ortho effect" (26). These and some other prominent fragments **a-e** resemble the fragmentations observed in anisessine [**10**] (19) (Scheme 2). The upfield shift of the H-3 proton by ca. 0.7 ppm and C-3 by ca. 7 ppm compared with the corresponding chemical shifts for anisotine [**7**] are indicative of the imino substituent at C-3 of the pyrroloquinazoline ring. An HMBC nmr (27) experiment showed correlations analogous to the selective INEPT correlations for alkaloids **7**

TABLE 6.  $^1\text{H}$ - and  $^{13}\text{C}$ -Nmr Chemical Shift Assignments of Anisotine [**7**] ( $\text{CDCl}_3$ ).<sup>a</sup>

Position	$^{13}\text{C}$ $\delta$	DEPT	$^1\text{H}$	$\delta$	Multiplicity ( $J$ , Hz)	COSY
1	44.7	t	1 <sub><math>\alpha</math></sub>	4.12	m	H-1 <sub><math>\beta</math></sub> , H-2 <sub><math>\alpha</math></sub> , H-2 <sub><math>\beta</math></sub>
			1 <sub><math>\beta</math></sub>	4.41	m	H-1 <sub><math>\alpha</math></sub> , H-2 <sub><math>\alpha</math></sub> , H-2 <sub><math>\beta</math></sub>
2	29.9	t	2 <sub><math>\alpha</math></sub>	2.73	m	H-1 <sub><math>\alpha</math></sub> , H-1 <sub><math>\beta</math></sub>
			2 <sub><math>\beta</math></sub>	2.29	m	H-2 <sub><math>\beta</math></sub> , H-3
						H-1 <sub><math>\beta</math></sub> , H-1 <sub><math>\alpha</math></sub>
						H-2 <sub><math>\alpha</math></sub> , H-3
						H-2 <sub><math>\alpha</math></sub> , H-2 <sub><math>\beta</math></sub>
3	49.1	d	3	4.40	m	
3a	124.7	s	3a	—		
4a	149.2	s	4a	—		
5	127.4	d	5	7.64	m	H-7
6	134.0	d	6	7.64	m	H-7, H-8
7	126.3	d	7	7.48	m	H-6, H-8
8	126.3	d	8	8.33	d	8,7=10.2
8a	120.5	s	8a	—		H-6, H-7
9	160.8	s	9	—		
10	109.9	s	10	—		
11	130.9	d	11	7.83	d	11,15=2.5
12	151.3	s	12	—		H-15
13	151.3	s	13	—		
14	111.5	d	14	6.70	d	14,15=10.5
15	134.2	d	15	7.32	m	H-15
16	168.7	s	16	—		H-11, H-14
17	51.5	q	17	3.82	s	
18	29.6	q	18	2.90	d	NH, Me=6.0
			NH	7.64	m	NH
						H-18

<sup>a</sup> $^1\text{H}$ - and  $^{13}\text{C}$ -nmr assignments were established by HETCOR experiment.

TABLE 7. Nmr Data of Anisotine [7] from Selective INEPT Experiments (CDCl<sub>3</sub>).

Proton Irradiated	$\delta$	Carbon Signal Enhanced
H-8	8.33	149.2 (C-4a), 160.8 (C-9)
H-11	7.83	168.7 (C-16), 151.3 (C-12, C-13)
H-7	7.48	127.4 (C-5), 120.5 (C-8a)
H-15	7.32	151.3 (C-13), 130.9 (C-11)
H-14	6.70	109.9 (C-10)
H-1, H-3	4.40	160.8 (C-9), 134.2 (C-15), 130.9 (C-11) 124.7 (C-3a), 49.1 (C-3), 29.9 (C-2)
H-1	4.12	29.9 (C-2)
H-2 <sub>a</sub> , H-2 <sub><math>\beta</math></sub>	2.29, 2.73	124.7 (C-3a), 49.1 (C-3), 44.7 (C-1)

TABLE 8. <sup>1</sup>H- and <sup>13</sup>C-Nmr Chemical Shift Assignments of 3-Hydroxyanisotine [8] (CDCl<sub>3</sub>).<sup>a</sup>

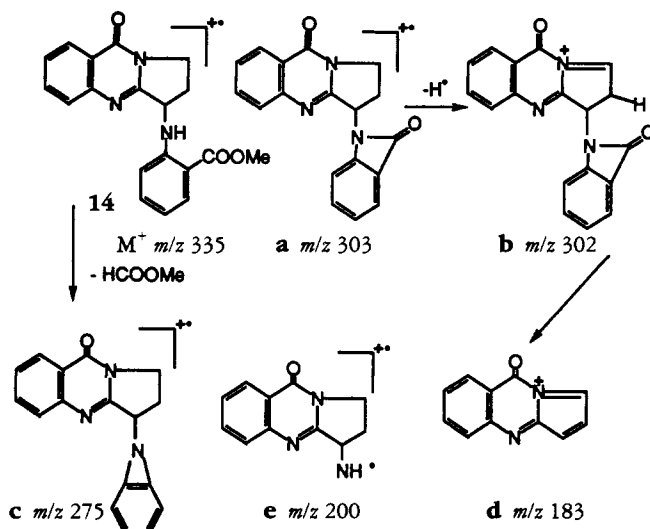
Position	<sup>13</sup> C $\delta$	DEPT	<sup>1</sup> H	$\delta$	Multiplicity ( <i>J</i> , Hz)	COSY
1	42.8	t	1 <sub>a</sub>	3.99	m	H-1 <sub><math>\beta</math></sub> , H-2
			1 <sub><math>\beta</math></sub>	4.29	m	H-1 <sub>a</sub> , H-2
2	37.8	t	2	2.60	m	H-1 <sub>a</sub> , H-1 <sub><math>\beta</math></sub>
3	81.1	s	3	—		
3a	161.1	s	3a	—		
4a	148.8	s	4a	—		
5	127.3	d	5	7.67	m	H-7
6	134.2	d	6	7.67	m	H-7, H-8
7	126.8	d	7	7.49	m	H-5, H-6, H-8
8	126.4	d	8	8.29	d	8,7=9.6 H-6, H-7
8a	120.9	s	8a	—		
9	160.7	s	9	—		
10	109.4	s	10	—		
11	128.6	d	11	7.97	d	11,15=2.5 H-15
12	151.7	s	12	—		
13	151.7	s	13	—		
14	110.9	d	14	6.60	d	14,15=8.9 H-15
15	131.9	d	15	7.38	dd	15,11=2.5 15,14=9.6 H-11, H-14
16	168.6	s	16	—		
17	51.5	q	17	3.77	s	
18	29.5	q	18	2.87	d	NH, Me=6.0 NH
			NH	7.67	m	CH <sub>3</sub> -18
			OH	1.90	br s	

<sup>a</sup><sup>1</sup>H- and <sup>13</sup>C-nmr assignments were established by HETCOR experiment.

TABLE 9. Nmr Data of 3-Hydroxyanisotine [8] from Selective INEPT Experiments.

Proton Irradiated	$\delta$	Carbon Signal Enhanced
H-8	8.29	160.7 (C-9), 148.8 (C-4a)
H-11	7.97	168.6 (C-16), 151.7 (C-12, C-13) 81.1 (C-3)
H-5, H-6	7.67	148.8 (C-4a), 127.3 (C-5)
H-7	7.49	127.3 (C-5), 120.9 (C-8a)
H-15	7.38	151.7 (C-13), 128.6 (C-11), 81.1 (C-3)
H-1 <sub><math>\beta</math></sub>	4.29	161.1 (C-3a), 81.1 (C-3)
H-2	2.60	161.1 (C-3a)



TABLE 10. <sup>1</sup>H- and <sup>13</sup>C-Nmr Chemical Shift Assignments of Vasnetine [9].<sup>a</sup>

Position	<sup>13</sup> C δ	DEPT	<sup>1</sup> H	δ	Multiplicity (J, Hz)	COSY
1	43.5	t	1 <sub>α</sub> 1 <sub>β</sub>	4.10 4.44	m m	H-1 <sub>β</sub> , H-2 <sub>α</sub> , H-2 <sub>β</sub> H-1 <sub>α</sub> , H-2 <sub>α</sub> , H-2 <sub>β</sub>
2	29.7	t	2 <sub>α</sub> 2 <sub>β</sub>	2.17 3.00	m m	H-1 <sub>α</sub> , H-1 <sub>β</sub> , H-2 <sub>β</sub> H-1 <sub>β</sub> , H-1 <sub>α</sub> H-2 <sub>α</sub> , H-3 H-2 <sub>α</sub> , H-2 <sub>β</sub>
3	56.1	d	3	5.10	m	
3a	119.6	s	3a	—		
4a	149.0	s	4a	—		
5	127.8	d	5	7.82	m	H-7
6	134.2	d	6	7.70	m	H-7
7	126.8	d	7	7.50	m	H-6, H-8
8	126.3	d	8	8.33	d	8,7=7.5 H-7
8a	121.0	s	8a	—		
9	160.8	s	9	—		
10	111.5	s	10	—		
11	149.5	s	11	—		
12	131.8	d	12	7.98	d	12,13=8.0
13	116.2	d	13	6.74	t	12,13,14=8.0
14	134.6	d	14	7.44	m	H-13, H-14 H-12, H-13 H-15
15	112.1	d	15	6.91	d	15,14=7.5 H-14
16	168.9	s	16	—		
17	51.7	q	17	3.88	s	

<sup>a</sup><sup>1</sup>H- and <sup>13</sup>C-nmr assignments were established by HETCOR experiment.

and **8**, further supporting this structure proposal. The nmr spectral data of **9** are summarized in Tables 10 and 11.

### EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Mps were determined on a Thomas-Kofler hot stage equipped with a microscope and polarizer. Nmr spectra were determined in CDCl<sub>3</sub> solutions on Bruker 250, 300 MHz and Varian 400 MHz spectrometers. Mass spectra were recorded on a Finnegan Quadrupole 4023 mass spectrometer at 70 eV. Optical rotations were measured on a Perkin-Elmer Model 141 polarimeter. Vlc (28) was carried out on Merck SiO<sub>2</sub> 60H (EM 7736) and Merck Al<sub>2</sub>O<sub>3</sub> (EM 1085). Chromatographic separations

TABLE 11. Selected Pertinent HMBC Correlations for Vasnetine [9].

Proton	Carbon Signal Correlated
H-7 . . . . .	C-5, C-8a
H-8 . . . . .	C-4a, C-6, C-9
H-12 . . . . .	C-11, C-14, C-16
H-13 . . . . .	C-15
H-14 . . . . .	C-12
H-15 . . . . .	C-10, C-13

on a Chromatotron (29) were carried out on rotors coated with 1 mm thick Si gel (HF-254+366; EM 7749). Tlc was carried out on Si gel 60H (EM 7741).

PLANT MATERIAL.—The plant material was identified and collected by Mr. M.R. Almeida, Botanist, CIBA-Geigy Research Centre, Bombay, India. A voucher specimen is deposited in the herbarium collection of the CIBA-Geigy Research Centre Goregaon East, Bombay 400063, India.

EXTRACTION AND ISOLATION.—The dried and ground leaves of *A. vasica* (5 kg) were extracted with hot EtOH (4×10 liters) and the extract evaporated to dryness to give a green gummy residue which was extracted with hot H<sub>2</sub>O (3×500 ml), cooled, and filtered. The green chlorophyllic residue was discarded and the aqueous solution extracted with CHCl<sub>3</sub> (5×250 ml). The aqueous layer was basified with 5% NaOH (pH 8–9) and extracted with CHCl<sub>3</sub> (5×250 ml). The CHCl<sub>3</sub> layer was extracted with 5% HCl (3×200 ml) and the acidic solution basified with NH<sub>3</sub> and extracted with CHCl<sub>3</sub> until the organic layer was free of alkaloids (Dragendorff's reagent). The CHCl<sub>3</sub> layer gave a solid (9 g) which on crystallization from MeOH/CH<sub>2</sub>Cl<sub>2</sub> afforded long rods of *l*-vasicine (**1**; 2.5 g), mp 213–214°; [α]<sub>D</sub><sup>20</sup> –210° (c=2, CHCl<sub>3</sub>). *Anal.*, found: C, 70.38, 69.95; H, 6.58, 6.68; N, 14.55; calcd for C<sub>11</sub>H<sub>12</sub>N<sub>2</sub>O: C, 70.18; H, 6.43; N, 14.88%. Uv (EtOH) λ max 213, 218, 289 nm (log ε, 4.24, 4.24, 3.81). Ms *m/z* 189 (M<sup>+</sup>+1, 7%), 188 (M<sup>+</sup>, 57), 187 (M<sup>+</sup>–1, 100), 169 (5), 159 (15), 131 (18), 116 (4), 104 (6), 89 (7), 77 (12). For <sup>1</sup>H- and <sup>13</sup>C-nmr spectral data, see Table 1.

Similar extractions were carried out in six batches (30 kg of leaves) and *l*-vasicine isolated. The mother liquors after separation of **1** gave on evaporation a crude alkaloidal mixture A (40 g). A solution of the crude alkaloid A (6.5 g) in CH<sub>2</sub>Cl<sub>2</sub> (100 ml) was extracted with 5% H<sub>2</sub>SO<sub>4</sub> (5×50 ml) and the acidic solution basified with NH<sub>3</sub> and extracted with CH<sub>2</sub>Cl<sub>2</sub> to afford an alkaloidal fraction (5.05 g). Crystallization of this fraction (500 mg) from MeOH (15 ml) gave fine needles of *l*-vasicinone (**2**, 122 mg), mp 201–202°; [α]<sub>D</sub><sup>20</sup> –122° (c=1.1, CHCl<sub>3</sub>). Uv λ max (EtOH) 225, 276, 300, 313 nm (log ε, 4.43, 3.97, 3.73, 3.66). Ms *m/z* 203 (M<sup>+</sup>+1, 14%), 202 (M<sup>+</sup>, 100), 174 (4), 147 (13), 146 (69), 130 (11), 119 (42). For <sup>1</sup>H- and <sup>13</sup>C-nmr spectral data, see Tables 2 and 3.

The CH<sub>2</sub>Cl<sub>2</sub> fraction was again extracted with 10% H<sub>2</sub>SO<sub>4</sub> (5×50 ml) and the organic layer dried (Na<sub>2</sub>SO<sub>4</sub>) to give a weakly basic gummy alkaloid B (215 mg). The acidic layer was basified with Na<sub>2</sub>CO<sub>3</sub> and the crude alkaloid isolated. Another batch of the alkaloidal fraction A (6.2 g) was similarly processed to afford B (200 mg). The combined fractions of the fraction were purified twice on a SiO<sub>2</sub> rotor of a Chromatotron and eluted with CHCl<sub>3</sub>-hexane (1:1) with increasing percentages of CHCl<sub>3</sub>. The separation was monitored by the bluish fluorescent bands seen under uv light and 50 ml fractions were collected. Fractions 7–9 (CHCl<sub>3</sub>-hexane, 65:35) afforded vasnetine (**9**, 7 mg), mp 185–187°; tlc (SiO<sub>2</sub>; CHCl<sub>3</sub>-MeOH, 98:2) R<sub>f</sub> 0.55. Uv λ max (EtOH) 205, 225, 255, 302, 314, 342 nm (log ε 4.55, 4.73, 4.15, 3.68, 3.60, 3.79). Ms *m/z* 336 (M<sup>+</sup>+1, 14), 335 (M<sup>+</sup>, 100), 303 (**a**, 6), 302 (**b**, 6), 276 (**c**, 51), 274 (28), 200 (**e**, 8), 183 (**d**, 26), 130 (11), 77 (25). For <sup>1</sup>H- and <sup>13</sup>C-nmr spectral data, see Tables 10 and 11. Fraction 13 gave anisotine (**7**, 11 mg), mp 184–186°; tlc (SiO<sub>2</sub>; CHCl<sub>3</sub>-MeOH, 98:2), R<sub>f</sub> 0.48. For <sup>1</sup>H- and <sup>13</sup>C-nmr spectral data, see Tables 6 and 7. Fractions 24–25 afforded 3-hydroxyanisotine (**8**, 15 mg), mp 182–184°; tlc (SiO<sub>2</sub>; CHCl<sub>3</sub>-MeOH, 98:2), R<sub>f</sub> 0.38. Uv λ max (EtOH) 226, 263, ca. 302, ca. 314, 353 nm (log ε, 4.74, 4.46, 3.98, 3.87, 3.69). Ms *m/z* 366 (M<sup>+</sup>+1, 32), 365 (M<sup>+</sup>, 100), 347 (19), 336 (32), 192 (36), 160 (12). For <sup>1</sup>H- and <sup>13</sup>C-nmr spectral data, see Tables 8 and 9. In another separation, the basic fraction A (3.37 g) was purified by vlc (SiO<sub>2</sub>) and eluted with hexane and increasing percentages of CHCl<sub>3</sub> and MeOH. The fractions which eluted with CHCl<sub>3</sub>-MeOH (96:4) were again separated by vlc on an Al<sub>2</sub>O<sub>3</sub> column and eluted with CHCl<sub>3</sub>-MeOH (96:4) (600 ml) to give a crude fraction (600 mg) which was separated on a SiO<sub>2</sub> rotor and eluted with CHCl<sub>3</sub>-MeOH, 99.5:0.5 to afford vasicol as a viscous alkaloid (**3**, 25 mg). Tlc (SiO<sub>2</sub>; CHCl<sub>3</sub>-MeOH, 90:10), R<sub>f</sub> 0.5. [α]<sub>D</sub><sup>20</sup> –12° (c=0.67, CHCl<sub>3</sub>). Uv (EtOH) λ max 210, 236, 288 nm (log ε, 4.33, 4.01, 3.46). Ms *m/z* 207 (M<sup>+</sup>+1, 7), 206 (M<sup>+</sup>, 100), 187 (2), 162 (10), 161 (44), 147 (59), 133 (22), 106 (76). For <sup>1</sup>H- and <sup>13</sup>C-nmr spectral data, see Tables 4 and 5.

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## LITERATURE CITED

1. S. Johnne, in: "The Alkaloids, Chemistry and Pharmacology." Ed. by A. Brossi, Academic Press, New York, Vol. 29, 1986, p. 99.
2. D. Hooper, *Pharm. J.*, **18**, 841 (1888).
3. J.N. Sen and T.P. Ghose, *J. Ind. Chem. Soc.*, **1**, 315 (1924).
4. D.R. Mehta, J.S. Naravane, and R.M. Desai, *J. Org. Chem.*, **28**, 445 (1963).
5. S. Johnne, D. Groger, and M. Hesse, *Helv. Chim. Acta*, **54**, 826 (1971).
6. M.P. Jain and V.K. Sharma, *Planta Med.*, **46**, 250 (1982).
7. B.K. Chowdhury and P. Bhattacharyya, *Phytochemistry*, **24**, 3080 (1985).
8. A.H. Amin and D.R. Mehta, *Nature*, **184**, 1317 (1959).
9. E. Späth and F. Keszler-Gandini, *Monatsch. Chem.*, **91**, 1150 (1960).
10. B.K. Chowdhury and P. Bhattacharyya, *Chem. Ind.*, 35 (1987).
11. K.L. Dhar, M.P. Jain, S.K. Koul, and C.K. Atal, *Phytochemistry*, **20**, 319 (1981).
12. H.K. Desai, D.H. Gawad, T.R. Govindachari, B.S. Joshi, P.C. Parthasarathy, K. S. Ramachandran, K.R. Ravindranath, A.R. Sidhaye, and N. Viswanathan, *Ind. J. Chem.*, **14B**, 473 (1975).
13. O.P. Gupta, M.L. Sharma, B.J. Ray Ghatak, and C.K. Atal, *Ind. J. Med. Res.*, **66**, 680 (1977).
14. C.K. Atal, "Chemistry and Pharmacology of Vasicine—A New Oxytocic and Abortifacient," Raj Bandhu Industrial Co., New Delhi, 1980.
15. E. Späth and F. Keszler, *Chem. Ber.*, **69**, 384 (1936).
16. K. Szulzewsky, E. Hohne, S. Johnne, and D. Gröger, *J. Prakt. Chem.*, **318**, 463 (1976).
17. K. Schreiber, O. Aurich, and K. Pufahl, *Arch. Pharm.*, **295**, 271 (1962).
18. E. Späth, F. Kuffner, and N. Platzer, *Chem. Ber.*, **68**, 1384 (1935).
19. R.R. Arndt, S.H. Eggers, and A. Jordaan, *Tetrahedron*, **23**, 3521 (1967).
20. S. Johnne, B. Jung, D. Gröger, and R. Radeaglia, *J. Prakt. Chem.*, **319**, 919 (1977).
21. G.A. Cordell, J.E. Saxton, M. Shamma, and G.F. Smith (Eds.), "Dictionary of Alkaloids," Chapman and Hall, London, 1989, p. 1113.
22. R. Poi and N. Adityachaudhury, *J. Ind. Chem. Soc.*, **65**, 814 (1988).
23. A. Bax, *J. Magn. Reson.*, **57**, 314 (1984).
24. J.D. Swalen, in: "Progress in Nuclear Magnetic Resonance Spectroscopy" Ed. by J.W. Emsley, J. Feeney, and L.H. Sutcliffe, Pergamon Press, Oxford, 1966, Vol. 1, p. 205.
25. L.M. Jackman, "Applications of Nuclear Magnetic Resonance Spectroscopy," Pergamon Press, New York, 1959, p. 85.
26. K. Biemann, *Angew. Chem.*, **74**, 102 (1962).
27. A. Bax and M.F. Summers, *J. Am. Chem. Soc.*, **108**, 2093 (1986).
28. S.W. Pelletier, H.P. Chokshi, and H.K. Desai, *J. Nat. Prod.*, **49**, 892 (1986).
29. H.K. Desai, B.S. Joshi, A.M. Panu, and S.W. Pelletier, *J. Chromatogr.*, **322**, 223 (1985).

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