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Indole alkaloids from *Rauvolfia bahiensis* A.DC. (Apocynaceae)

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Abstract

Four indole alkaloids, 12-methoxy-N_a-methyl-vellosimine, demethoxypurpeline, 12-methoxyaffinisine, and 12-methoxy-vellosimine, in addition to picrinine, vinorine, raucaffrinoline, normacusine B, norseredamine, seredamine, 10-methoxynormacusine B, norpurpeline and purpeline, were isolated from the bark or leaf extracts of *Rauvolfia bahiensis*. © 2002 Published by Elsevier Science Ltd.

Keywords: *Rauvolfia bahiensis*; Apocynaceae; Indole alkaloids; ^{1}H NMR; ^{13}C NMR; 12-Methoxy-N_a-methyl-vellosimine; Demethoxypurpeline; 12-Methoxyaffinisine; 12-Methoxy-vellosimine

1. Introduction

Bioactive indole alkaloids have been isolated from several plant species of the Apocynaceae family, including many belonging to the *Rauvolfia* genus, among which *Rauvolfia serpentina* Benth, from India, has been a most important source of reserpine (Stöckigt, 1995). The chemistry of the *Rauvolfia* species has been exhaustively investigated for the presence of indole alkaloids over a long period of time (Siddiqui et al., 1987; Stöckigt, 1995; Court, 1983; Libot et al., 1986). In Brazil, there are 17 species of *Rauvolfia,* but only three, *R. sellowii* (Batista et al., 1996; Belém-Pinheiro et al., 1988), *R. grandiflora* (Cancelieri et al., 2001) and *Rauvolfia ligustrina* (Müller, 1957) have been previously investigated for alkaloids.

As part of a research program on the occurrence of indole alkaloids in Brazilian species of *Rauvolfia*, a phytochemical analysis from the extracts from leaves and bark of *R. bahiensis* A.DC. is described. Thirteen indole alkaloids were isolated, of which four are new. Three new alkaloids possess the sarpagine-type skeleton and the other has the ajmalan type skeleton, with a new substitution pattern. The structures of all indolic alkaloids isolated were determined by a detailed analysis of the ^{1}H NMR, ^{13}C NMR, HMBC and COSY spectral data.

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2. Results and discussion

Plant material was collected at Reserva da Una (Ilhéus, Bahia, Brazil) and air-dried. The ethanolic extract, from the leaves or bark, was submitted to acid-base treatment and fractions corresponding to different pH ranges were purified by flash-chromatography, CC or preparative TLC, leading to the isolation of thirteen indole alkaloids (Fig. 1). Picrinine (**1**) (Batista et al., 1996), vinorine (**2**) (Libot et al., 1986), raucaffrinoline (**3**) (Batista et al., 1996), normacusine B (**4**) (Jokela and Lounasmaa, 1996a), norseredamine (**5**), seredamine (**6**) (Hanaoka et al., 1970), 10-methoxynormacusine B (**7**) (Belém-Pinheiro et al., 1988), norpurpeline (**8**) and purpeline (**9**) (Iwu and Court, 1978) were previously isolated from other sources and their structures were confirmed by spectroscopic analysis.

The alkaloids 12-methoxy-N_a-methyl-vellosimine (**10**), demethoxypurpeline (**11**), 12-methoxyaffinisine (**12**), and 12-methoxy-vellosimine (**13**) are described for the first time (Fig. 1).

A detailed analysis of the ^{1}H NMR, ^{13}C NMR, HMBC and COSY spectral data was conducted for the new alkaloids. The initial ^{1}H NMR assignments were achieved using a ^{1}H–^{13}C correlation experiment, and the chemical shifts and spin-coupling constants were determined from the ^{1}H NMR spectrum. The COSY experiment was used in order to obtain structural information via the spin connectivities revealed by the cross peaks. The chemical shifts were assigned by comparison with other sarpagine-type (Braga and Reis,

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1987; Jokela and Lounasmaa, 1996a) or other ajmalan-type (Jokela and Lounasmaa, 1996b) derivatives.

2.1. 12-Methoxy-N_a-methyl-vellosimine (**10**)

The HREIMS showed a molecular ion $[M]^+$ at m/z 336.1836 corresponding to $C_{21}H_{24}N_2O_2$ (required $M^+ = 336.1838$). The mass spectrum shows peaks at $m/z = 335$ $[M-1]^+$, 321 $[M-CH_3]^+$, 307 $[M-CHO]^+$, and the fragments at $m/z = 293$ and $m/z = 213$ are consistent with a sarpagine-type skeleton with a methoxy and a methyl group on an indolic ring, (Djerassi et al., 1964; Banerji and Chakrabarty, 1974) as shown in Fig. 2

The UV spectrum showed a typical indolic absorption at λ_{max} (MeOH), 284 nm (log ε 3.10), while the IR spectrum showed the presence of a C=O function (1721 cm^{-1}). The ^1H NMR spectrum (Table 1) showed resonances corresponding to three vicinal aromatic hydrogens at δ 7.04 (*dd*, $J = 8$ and 1 Hz), δ 6.61 (*dd*, $J = 8$ and 1 Hz) and δ 6.96 (*t*, $J = 8$ Hz). The δ values suggested the presence of a 12-substituted or 9-substituted indole ring. Absorptions for two methyl groups (*s*, δ 3.90, 6H), an aldehydic hydrogen (*d*, δ 9.61, $J = 1$ Hz) and an ethylidene group (1H, *q*, δ 5.35, $J = 7$ Hz, and 3H, *dt*, δ 1.61, $J = 7$ and 2 Hz) were also observed. The ^{13}C NMR spectrum (Table 2) was in accordance with a sarpagine-type skeleton (Braga and Reis, 1987; Jokela and Lounasmaa, 1996a). The chemical shifts observed at δ 32.5 and δ 55.4 correlated with the chemical shift at δ 3.90 in the HMBC spectra and suggest the presence of a N_a-methyl group and an O–CH_3 group, respectively. The chemical shifts of C-6 (δ 27.2) and C-14 (δ 32.3) demonstrated that the CHO group at C16 must be situated away from the aromatic nucleus (Jokela et al., 1996a). The chemical shifts of C-9 (δ 111.1), C-10 (δ 119.4), C-11 (δ 102.6), and C-12 (147.6) corroborate a 12-methoxy substituted indole ring (Braga and Reis, 1987). The geometry of the ethylidene side chain of **10** was deduced from the chemical shifts of C-15 (δ 26.5) and C-21 (δ 56.0), which are consistent with *E* geometry (Clivio et al., 1991; Schun and Cordell, 1987; Aimi et al., 1978)

2.2. Demethoxypurpeline (**11**)

The HREIMS showed a molecular ion $[M]^+$ at m/z 306.1732 corresponding to $C_{20}H_{22}N_2O$ (required $M^+ = 306.1732$). UV absorptions were seen at λ_{max} (MeOH) 246 nm (log ε 3.61), 280 nm (3.32) and 294 nm (3.31), while the IR spectrum showed the presence of a C=O function (1738 cm^{-1}). The ^1H NMR spectrum (Table 1) showed signals for a N_a-methyl group (*s*, δ 2.82) and an ethylidene group (1H, q, δ 5.34, $J = 7$ Hz, and 3H, *d*, δ 1.66, $J = 7$ Hz). The ^{13}C NMR spectrum showed peaks corresponding to a quaternary carbon at δ 58.3 assigned to C-7 and a CH carbon at δ 78.8 assigned to C-2, suggesting an ajmaline-type alkaloid, with a carbonyl group at C-17 (δ 213.2), and a *N*-methyl group at δ 34.4. Comparison with the NMR spectral data for rauflorine (Jokela and Lounasmaa, 1996b) corroborates structure **11** for this alkaloid, since they have the same ajmaline skeleton, although rauflorine does not have the *N*-methyl group on the indolic ring.

2.3. 12-Methoxyaffinisine (**12**)

The HREIMS showed a molecular ion $[M]^+$ at m/z 338.1997 corresponding to $C_{21}H_{26}N_2O_2$ (required $M^+ = 338.1994$). The UV spectrum showed an absorption maximum at λ_{max} 274 nm (log ε 3.25), while the IR spectrum showed the presence of an OH function (3368

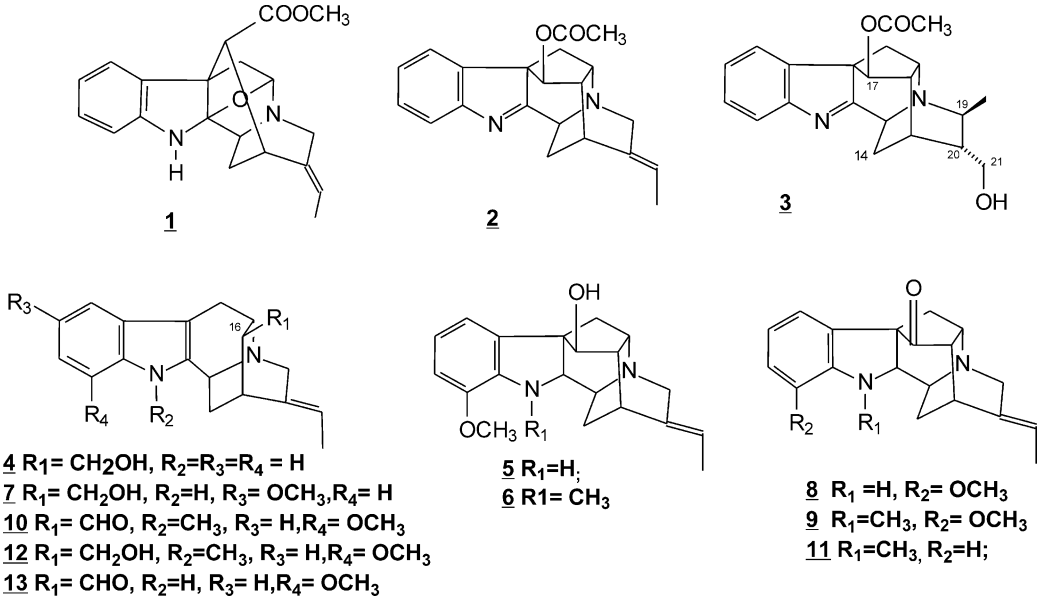

Fig. 1. Indole alkaloids from *Rauvolfia bahiensis* A.DC.

Table 1
^{1}H NMR spectroscopic data values for alkaloids **10**, **11**, **12**, and **13** (500 MHz, $CDCl_3$/0.03%TMS)[ab]

H	**10**	**11**	**12**	**13**
H-3	4.23 (1H, *dd*, *J* = 10, 2 Hz)	3.77 (1H, d, *J* = 10 Hz)	4.19 (1H, *d*, *J* = 8 Hz)	4.28 (1H, *d*, *J* = 9 Hz)
H-5	3.63 (1H, *br s*)	3.31 (1H, t, *J* = 6 Hz)	2.80 (1H, *br s*)	3.65 (1H, *t*, *J* = 6 Hz)
H-6α H-6β	2.56 (1H, *dd*, J = 2, 16 Hz)	1.73 (1H, *dd*, *J* = 12, 5 Hz)	2.59 (1H, *d*, *J* = 15 Hz)	2.58 (1H, *dd*, *J* = 16, 2 Hz)
	3.11 (1H, *dd*, *J* = 6, 16 Hz)	2.50 (1H, *d*, *J* = 12 Hz)	3.04 (1H, *dd*, *J* = 15, 5 Hz)	3.18 (1H, *dd*, *J* = 16, 5 Hz)
H-9	7.04 (1H, *dd*, *J* = 1, 8 Hz)	7.24 (1H, *br d*, *J* = 7 Hz)	7.05 (1H, *d*, *J* = 8 Hz)	7.07 (1H, *d*, *J* = 8 Hz)
H-10	6.61 (1H, *dd*, *J* = 1, 8 Hz)	6.86 (1H, *br t*, *J* = 7 Hz)	6.96 (1H, *t*, *J* = 8 Hz)	7.02 (1H, *t*, *J* = 8 Hz)
H-11	6.96 (1H, *t*, *J* = 8 Hz)	7.18 (1H, *br t*, *J* = 8 Hz)	6.61 (1H, *d*, *J* = 8 Hz)	6.64 (1H, *d*, *J* = 8 Hz)
H-12	–	6.68 (1H, *br d*, *J* = 8 Hz)	–	–
H-14α H-14β	2.11 (1H, *ddd*, *J* = 12, 4, 2 Hz)	1.95 (1H, *br t*, *J* = 14 Hz)	1.67 (1H, *dt*, *J* = 12, 3 Hz)	1.83 (1H, *d*, *J* = 12 Hz)
	1.75 (1H, *dd*, *J* = 12, 2 Hz)	1.45 (1H, *dd*, *J* = 14, 4 Hz)	2.07 (1H, *td*, *J* = 12, 2 Hz)	2.10 (1H, *t*, *J* = 12 Hz)
H-15	3.18 (1H, *m*)	3.23 (1H, *t*, *J* = 6 Hz)	2.80 (1H, *br s*)	3.20 (1H, *br s*)
H-16	2.46 (1H, *d*, *J* = 7 Hz)	2.66 (1H, *m*)	1.80 (1H, *dd*, *J* = 7, 2 Hz)	2.51 (1H, *d*, *J* = 7 Hz)
H-17	9.61 (1H, *d*, *J* = 1 Hz)	–	3.50(1H, *dd*, *J* = 11, 9 Hz)	9.61 (1H, *s*)
			3.57 (1H, *dd*, *J* = 11, 6Hz)	
H-18	1.61 (1H, *dt*, *J* = 7, 2 Hz)	1.66 (3H, *d*, *J* = 7 Hz)	1.64 (3H, *d*, *J* = 7 Hz)	1.60 (3H, *d*, *J* = 7 Hz)
H-19	5.35 (3H, *q*, *J* = 7 Hz)	5.34 (1H,*q*, *J* = 7 Hz)	5.41 (1H, *q*, *J* = 7 Hz)	5.34 (1H, *q*, *J* = 7 Hz)
H-20	–	–	–	–
H-21	3.63 (2H, *br s*)	3.55 (2H, *br s*)	3.63 (2H, *br s*)	3.60∼3.70 (2H, *m*)
N_a-CH_3	3.90 (3H, *s*)	2.82 (3H, *s*)	3.91 (3H, *s*)	–
O-CH_3	3.90 (3H, s)	–	3.93 (3H, *s*)	3.96 (3H, *s*)
N-H	–	–	–	8.20 (1H, *s*)

[a] Chemical shifts (δ) in ppm from TMS, number of H, multiplicities, and coupling constants (*J*) in Hz are in parentheses.
[b] Signal assignments based on 2D experiments.

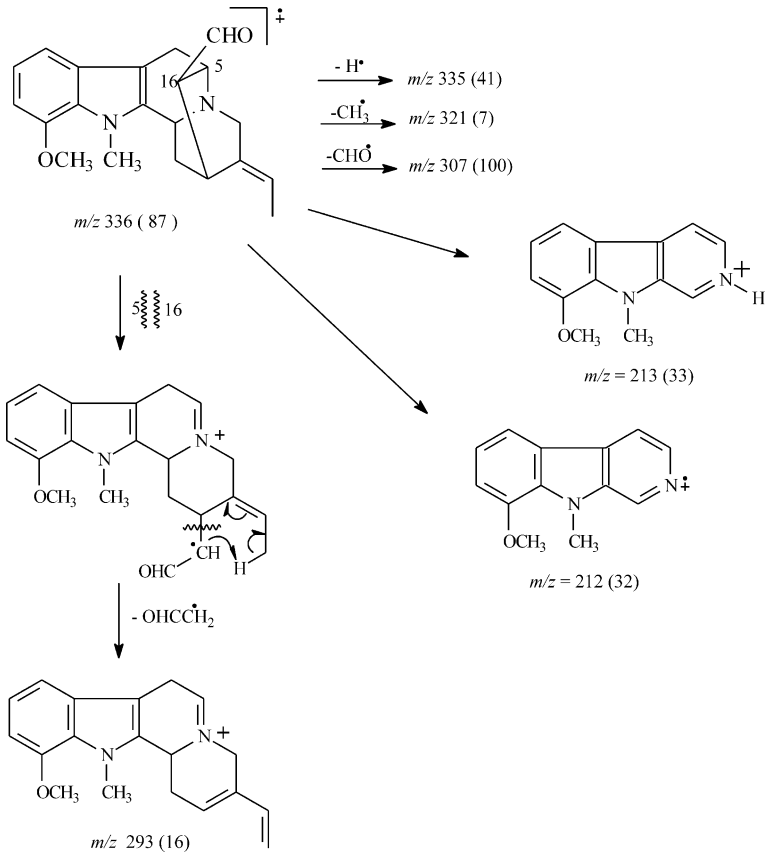

Fig. 2. Fragmentation pathway for compound **10**.

Table 2
^{13}C NMR spectroscopic data for alkaloids **10**, **11**, **12**, and **13** (75 MHz, $CDCl_3$)[ab]

C	10	11	12	13
2	134.5	78.8	135.8	133.5
3	49.3	50.0	49.4	50.2
5	50.4	53.3	54.2	50.4
6	27.2	34.8	27.0	27.1
7	103.3	58.3	103.8	104.6
8	126.7	129.0	126.6	126.6
9	111.1	122.5	111.1	111.1
10	119.4	120.1	119.2	120.0
11	102.6	128.2	102.5	102.0
12	147.6	110.0	147.5	146.0
13	129.2	154.0	129.4	133.5
14	32.3	31.4	32.9	32.8
15	26.5	28.5	27.5	26.8
16	54.6	50.3	44.3	54.8
17	202.6	213.2	65.0	202.3
18	12.6	13.0	12.8	12.6
19	117.0	116.7	116.7	117.3
20	139.3	136.5	139.7	136.8
21	56.0	55.4	56.2	55.3
N_a–$\underline{C}H_3$	32.5	34.4	32.4	–
–O–$\underline{C}H_3$	55.4	–	55.4	55.4

[a] Chemical shifts (δ) in ppm from TMS.
[b] Signal assignments based on 2D experiments.

cm^{-1}). The ^1H NMR spectrum (Table 1) showed a substituted indolic ring, a *N*-methyl group, an aromatic methoxy group (3H, *s*, δ 3.93), an ethylidene group (1H, *q*, δ 5.41, $J=7$ Hz, and 3H, *d*, δ 1.64, $J=7$ Hz), and a hydroxymethyl function (1H, *dd*, δ 3.50, $J=11$, and 9 Hz, and 1H, *dd*, δ 3.57, $J=11$, and 6 Hz). These data and the ^{13}C NMR spectrum (Table 2) suggested a sarpagine-type alkaloid. Comparison with the NMR spectroscopic data furnished by affinisine (Braga and Reis, 1987), and the alkaloids normacusine B (**4**), and 10-methoxynormacusine B (**7**), revealed **12** to be the 12-methoxy substituted derivative of the known alkaloid affinisine.

2.4. 12-Methoxy-vellosimine (*13*)

The HREIMS showed a molecular ion $[M]^+$ at *m*/*z* 322.1677 corresponding to $C_{20}H_{22}N_2O_2$ (required $M^+=322.1681$). The UV spectrum showed absorption maxima at λ_{max} 254 nm (log ε 3.78) and 230 nm (4.20), while the IR spectrum showed the presence of a C=O function (1729 cm^{-1}). The ^1H NMR spectrum (Table 1) and the ^{13}C NMR spectrum (Table 2) suggested a sarpagine-type alkaloid similar to **10**, except that the signal of a *N*-methyl group, was not present in **13**.

The alkaloids isolated from *Rauvolfia bahiensis* show a systematic relationship with sarpagine and ajmaline alkaloids. The overall enzymatic pathway for ajmaline type alkaloids was shown by Stöckigt (1995), where the formation of vinorine **2** and raucaffrinoline **3** and related alkaloids from sarpagine type alkaloids are proposed.

The new alkaloids isolated from *Rauvolfia bahiensis* have a 12-methoxy substitution pattern in indolic nucleus. It is interesting to note that the same substitution pattern was found for the indolic alkaloids isolated from *Peschiera fuchsiaefolia* (Apocynacae) (Braga and Reis, 1987) from Brazil and from *Rauvolfia vomitoria* from Africa (Iwu and Court, 1977).

3. Experimental

3.1. General

UV spectra were recorded on a HP 8452A Diode Array Spectrophotometer. The IR spectra were recorded as a CH_2Cl_2 film with a Bomen MB FT/IR spectrometer. The ^1H NMR and ^{13}C NMR spectra were obtained at 300/75 and 500/125 MHz on Gemini 300 BB/(Varian) and Inova 500 (Varian) spectrometers with $CDCl_3+0.03\%$ TMS solutions in 5 mm probe tubes at room temperature. The chemical shifts are recorded in δ (ppm) based on δ TMS = 0, and coupling constants (*J*) are in Hz. EIMS and HREIMS (70 eV, direct probe) experiments were performed using a VG Auto Spec-Fisions Instrument using electron ionisation at 70 eV (linked scan from 8 keV collisions with helium). Optical rotations were measured with a Jasco model J-720 digital polarimeter.

Silica-gel (0.040–0.063 mm, 60) (Merck) and silica-gel GF_{254} (Merck) were used for flash-chromatography CC and TLC, respectively. Detection of alkaloids was made by UV (254 and 365 nm) and spraying with Dragendorff's reagent followed by H_2SO_4–MeOH (1:1) and heating for 5 min.

3.2. Plant material

Leaves and bark of *Rauvolfia bahiensis* were collected in Reserva da Una (Ilhéus, Bahia, Brazil) and were identified by one of us (IK). Voucher specimens are deposited at the Herbarium of the Instituto de Biologia—UNICAMP (I. Koch 640 and R. Belinello).

3.3. Extraction and isolation

Powdered, air-dried leaves (400 g) and bark (265 g) were separately extracted with 95% aq. EtOH, until a negative Mayer test, at room temperature (leaves) or in a Soxhlet apparatus (bark). The EtOH extract was concentrated to dryness under reduced pressure. The leaves furnished 60 g extract and the bark furnished 11.8 g, extract which were kept separate, to which was then added a 10% HOAc soln. (250 ml, 3×) and each suspension was kept at 5 °C overnight. The suspension was filtered and the acidic aqueous phase was partitioned with CH_2Cl_2. The non-basic fraction was discarded. The combined organic layers were washed with

satd. $NaHCO_3$ solution, then with water. The acidic water phase was neutralized (pH = 7) and partitioned again with CH_2Cl_2. The aqueous phase was made alkaline with $NaHCO_3$ (pH = 9) and extracted with CH_2Cl_2 and then with EtOAc. The alkaloids of each fraction were isolated by repeated fractionation using flash-chromatography CC (CH_2Cl_2–MeOH, or $CHCl_3$–MeOH%) followed by preparative TLC on SiO_2 ($CHCl_3$ was utilized instead of CH_2Cl_2 to avoid artefacts).

The neutral CH_2Cl_2 and basic EtOAc extracts (< 100 mg) were not studied. Flash-chromatography with CH_2Cl_2–MeOH (90–10) of the basic CH_2Cl_2 extract from the leaves (340 mg) furnished 77 fractions (20 ml each), which were reduced to 13 groups after TLC analysis. Further purification by successive prep. TLC [CH_2Cl_2–MeOH (97–3), and (96–4), both NH_3 atm] of some of these fractions furnished: picrinine (**1**, 20.9 mg), raucaffrinoline (**3**, 9.1 mg), and normacusine B (**4**, 18.0 mg). Successive prep. TLC [CH_2Cl_2–MeOH (97–3) NH_3 atm] from the acidic CH_2Cl_2 extract (890 mg) of the leaves furnished vinorine (**2**, 31.0 mg). Flash-chromatography with $CHCl_3$–MeOH (92–8) of the basic CH_2Cl_2 extract of bark (460 mg) furnished 72 fractions of 20 ml each, which were reduced to 17 groups after TLC analysis. Further purification by successive prep. TLC [CH_2Cl_2–MeOH (95–5), and (96–4), both NH_3 atm] of some of these fractions furnished norseredamine (**5**, 15.5 mg), seredamine (**6**, 14.4 mg), and 10-methoxy-normacusine B (**7**, 10.0 mg).

Flash-chromatography with $CHCl_3$–MeOH (95–5) of the acidic CH_2Cl_2 extract of bark (1.16 g) furnished 70 fractions of 30 ml each, which were reduced to 15 groups after TLC analysis. Further purification by successive prep. TLC [Hept–EtOAc (50–50) and CH_2Cl_2–MeOH (92–8), NH_3 atm] of some of these fractions furnished norpurpeline (**8**, 102 mg), purpeline (**9**, 18 mg), 12-methoxy-N_a-methyl-vellosimine (**10**, 40 mg), 12-demethoxypurpeline (**11**, 8 mg), 12-methoxyaffinisine (**12**, 9 mg), and 12-methoxy-vellosimine (**13**, 10 mg).

3.3.1. 12-Methoxy-N_a-methyl-vellosimine (**10**)

Yellow amorphous solid; UV λ_{max} (MeOH) nm (log ε) 284 (3.10). $[\alpha]_D^{20}$ +14 ($CHCl_3$, *c* 0.59). IR ν (cm^{-1}) (CH_2Cl_2 film) 3376, 2935, 1721, 1617. ^1H NMR spectral data (500 MHz, $CDCl_3$/TMS): Table 1. ^{13}C NMR spectral data (75 MHz, $CDCl_3$): Table 2. EIMS (probe) 70 eV, *m*/*z* (rel. int.) 336 $[M]^+$ (87), 335 $[M-1]^+$ (43), 321 $[M-Me]^+$ (7), 307 $[M-29]^+$ (100), 293 $[M-43]^+$, 213 $[M-123]^+$ (33), 212 $[M-124]^+$ (32), 197 $[M-Me-124]^+$ (22). HR-EIMS *m*/*z* : found 336.1836 ($C_{21}H_{24}N_2O_2$ requires 336.1838).

3.3.2. Demethoxypurpeline (**11**)

Yellow amorphous solid; UV λ_{max} (MeOH) nm (log ε) 246 (3.61), 280 (3.32), 294 (3.31). $[\alpha]_D^{25}$ +22 ($CHCl_3$, *c* 0.57). IR ν (cm^{-1}) (CH_2Cl_2 film) 1738. ^1H NMR spectral data (500 MHz, $CDCl_3$/TMS): Table 1. ^{13}C NMR spectral data (75 MHz, $CDCl_3$): Table 2. EIMS (probe) 70 eV, *m*/*z* (rel. int.) 306 $[M]^+$ (100), 277 $[M-29]^+$ (11), 219 $[M-87]^+$ (12), 183 $[M-123]^+$ (6), 144 $[M-162]^+$ (34). HR-EIMS *m*/*z*: found 306.1732 $[M]^+$ ($C_{20}H_{22}N_2O$ requires 306.1732).

3.3.3. 12-Methoxyaffinisine (**12**)

Yellow amorphous solid; UV λ_{max} (MeOH) nm (log ε) 274 (3.25). $[\alpha]_D^{20}$ +3 ($CHCl_3$, *c* 0.72). IR ν (cm^{-1}) (CH_2Cl_2 film) 3368, 2924, 1616, 1572 . ^1H NMR spectral data (500 MHz, $CDCl_3$/TMS): Table 1. ^{13}C NMR spectral data (75 MHz, $CDCl_3$): Table 2. EIMS (probe) 70 eV, *m*/*z* (rel. int.) 338 $[M]^+$ (100), 337 $[M-1]^+$ (86), 323 [M−Me] (44), 307 $[M-31]^+$ (44), 213 $[M-125]^+$ (38), 212 [M−126]+, 197 $[M-Me-126]^+$. HR-EIMS *m*/*z* : found 338.1997 $[M]^+$ ($C_{21}H_{26}N_2O_2$ requires 338.1994).

3.3.4. 12-Methoxy-vellosimine (**13**)

Yellow amorphous solid; UV λ_{max} (MeOH) nm (log. ε) 254 (3.78), 230 (4.20). $[\alpha]_D^{20}$ +5 ($CHCl_3$, *c* 0.59). IR ν (cm^{-1}) (CH_2Cl_2 film), 2923, 1729, 1630, 1578. ^1H NMR spectral data (500 MHz, $CDCl_3$/TMS): Table 1. ^{13}C NMR spectral data (75 MHz, $CDCl_3$): Table 2. EIMS (probe) 70eV, *m*/*z* (rel. int.) 322 $[M]^+$ (100), 321 $[M-1]^+$ (28), 293 $[M-29]^+$ (81), 199 $[M-123]^+$, 181 $[M-141]^+$ (36). HR-EIMS *m*/*z*: found 322.1677 ($C_{20}H_{22}N_2O_2$ requires 322.1681).

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References

Aimi, N., Yamaguchi, K., Sakai, S.I., Haginiwa, J., Kubo, A., 1978. *Gardneria* alkaloids. XII. Carbon magnetic resonance spectra of gardneria alkaloids. A study on the configuration of the side chain double bonds of indole alkaloids. Chemical and Pharmaceutical Bulletin 26, 3444–3449.

Batista, C.V.F., Schripsema, J., Verpoorte, R., Rech, S.B., Henriques, A.T., 1996. Indole alkaloids from *Rauwolfia sellowii*. Phytochemistry 41, 969–973.

Banerji, A., Chakrabarty, M., 1974. Lochvinerine: a new alkaloid of *Vinca major*. Phytochemistry 13, 2309–2312.

Belém-Pinheiro, M.L., Imbiriba da Rocha, A.F., Quentin-Leclercq, J., Wauters, J.N., Angenot, L., 1988. Alkaloids of *Rauvolfia sellowii* from Amazonia. J. Pharm. Belg. 43, 379–381.

Braga, R.M., Reis, F.A.M., 1987. Quaternary alkaloids from *Peschiera fuchsiaefolia*. Phytochemistry 26, 833–836.

Cancelieri, N. M., Vieira, I. J. C., Braz Filho, R., Schripsema, J.,

Mathias, L., 2001. Alcalóides indólicos de *Rauvolfia grandiflora* (Apocynaceae). Resumos da 24ª Reunião Anual da Sociedade Brasileira de Química—Poços de Caldas (MG) Brazil, 28–31 de maio de 2001, PN-002.

Clivio, P., Richard, B., Deverre, J.R., Sévenet, T., Zeches, M., Le Men-Oliver, L., 1991. Alkaloids from leaves and root bark of *Ervatamia hirta*. Phytochemistry 30, 3785–3792.

Court, W.E., 1983. Alkaloid distribution in some African *Rauvolfia* species. Planta Medica 48, 228–233.

Djerassi, C., Budzikiewicz, H., Williams, D.H., 1964. Alkaloids. In: Structure Elucidation of Natural Products by Mass Spectrometry, Vol. 1. Holden-Day, San Francisco, USA, pp. 77–88.

Hanaoka, M., Hesse, M., Schmid, H., 1970. $N_{(a)}$-Demethylseredamin, ein neues Alkaloid aus *Rauwolfia sumatrama*; absolute Konfiguration von Seredamin. Helvetica Chimica Acta 53, 1723–1732.

Iwu, M.M., Court, W.E., 1977. Root alkaloids of *Rauwolfia vomitoria*. Planta Medica 32, 88–89.

Iwu, M.M., Court, W.E., 1978. The alkaloids of *Rauwolfia mombasiana* leaves. Planta Medica 33, 232–236.

Jokela, R., Lounasmaa, M.A., 1996a. ^{1}H and ^{13}C-NMR spectral data of five sarpagine-type alkaloids. Heterocycles 43, 1015–1020.

Jokela, R., Lounasmaa, M.A., 1996b. A ^{1}H and ^{13}C-NMR study of seven ajmaline-type alkaloids. Planta Medica 62, 577–579.

Libot, F., Miet, C., Kunesch, N., Poisson, J.E., Pusset, J., Sévenet, T., 1986. Alcaloides indoliques de *Rauvolfia neo-calédoniens*. Ann. Pharmaceut Fr. 44, 477–485.

Müller, J.M., 1957. Über die Alkaloide von *Rauvolfia ligustrina* R. & S. Raugustin, ein neues reserpinähnliches Alkaloid. Experientia 13, 479–481.

Schun, Y., Cordell, G.A., 1987. Revision of the stereochemistry of koumidine. Phytochemistry 26, 2875–2876.

Siddiqui, S., Haider, S.I., Ahmad, S.S., Sultana, N., 1987. The Alkaloids of *Rauvolfia*, Pakistan. J. Sci. Ind Res. 30, 71–80.

Stöckigt, J., 1995. Biosynthesis in *Rauvolfia serpentina* Modern aspects of an old medicinal plant. In: Cordell, G.A. (Ed.), The Alkaloids: Chemistry and Pharmacology. Academic Press, San Diego, pp. 115–172.