Pyrrolidinoindoline Alkaloids from Psychotria o leoides and Psychotria l yciiflora

ARTICLE *in* JOURNAL OF NATURAL PRODUCTS · JULY 1999

Impact Factor: 3.8 · DOI: 10.1021/np9805387 · Source: PubMed

Pyrrolidinoindoline Alkaloids from *Psychotria oleoides* and *Psychotria lyciiflora*

Valérie Jannic, Françoise Guéritte,* Olivier Laprévote, Laurent Serani, Marie-Thérèse Martin, Thierry Sévenet, and Pierre Potier

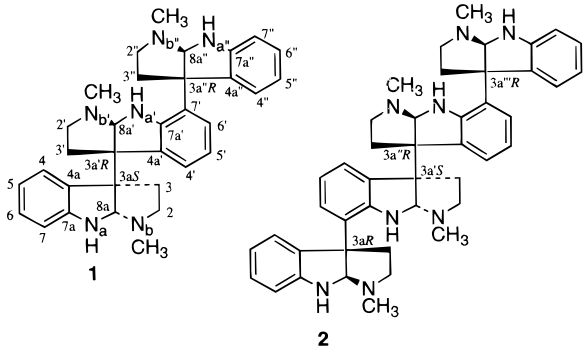
Institut de Chimie des Substances Naturelles, Centre National de la Recherche Scientifique, 1, Avenue de la Terrasse, 91198 Gif-sur-Yvette Cedex France

Received November 30, 1998

The chemical study of two Rubiaceae from New Caledonia, *Psychotria lyciiflora* and *Psychotria oleoides*, led to the isolation of several pyrrolidinoindoline alkaloids. Two dimers, the known *meso*-chimonanthine (**9**) and the new N_b-desmethyl-*meso*-chimonanthine (**5**), and a known trimer, hodgkinsine (**1**), have been isolated from *P. lyciiflora*. Hodgkinsine (**1**), quadrigemine C (**2**), isopsychotridine B (**3**), psychotridine (**4**), and three new alkaloids, quadrigemine I (**6**), oleoidine (**7**), and caledonine (**8**), have been isolated from *P. oleoides*. Structural assignments of the compounds were based on mass spectra analysis and 2D NMR experiments. A tentative stereochemical determination is made from 2D NMR experiments, circular dichroism study and chemical correlations. Some of these compounds are functional antagonists of somatostatine (SRIH).

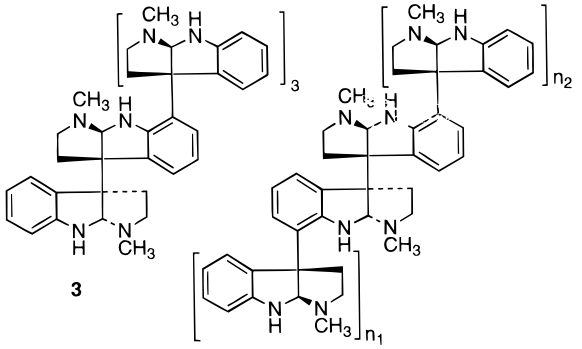
Our interest in the genus *Psychotria* followed the discovery of its significant activity on rat pituitary hormone secretion.[1] Originating from New Caledonia, *Psychotria lyciiflora* Schlecht. and *Psychotria oleoides* Schlecht. are two Rubiaceae possessing the same type of pyrrolidinoindoline alkaloids. The isolation of hodgkinsine (**1**),[2,3] quadrigemine C (**2**),[4−7] isopsychotridine B (**3**),[4,8] and psychotridine (**4**)[8,9] from *P. oleoides* has been reported previously by Libot et al.[4,10] Then we described the bioassay-guided purification of *P. oleoides*[7] and, more recently, we reported the somatostatin antagonistic activity of **2** and of its rearranged product, psycholeine, on GH secretion by pituitary cells.[11] This original activity on the neuroendocrine system prompted us to reexamine the alkaloid content of *P. oleoides* as well as of another species, *P. lyciiflora*.

This investigation has resulted in the isolation and characterization of four new pyrrolidinoindoline alkaloids, N_b-desmethyl-*meso*-chimonanthine (**5**), quadrigemine I (**6**), oleoidine (**7**), and caledonine (**8**) along with the known *meso*-chimonanthine (**9**),[12−16] **1**,[2,3] **2**,[4,6] **3**,[4] and **4**.[9] These



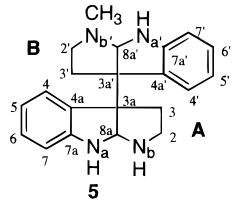
1

2

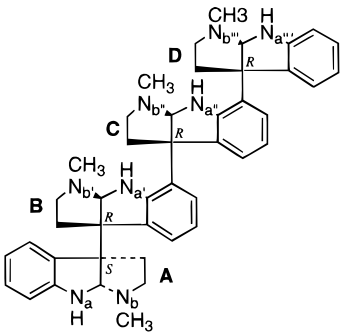


3

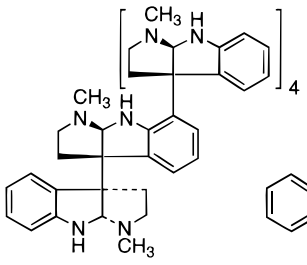
4 (n_1= 1, n_2= 2 or n_1= 2, n_2= 1)



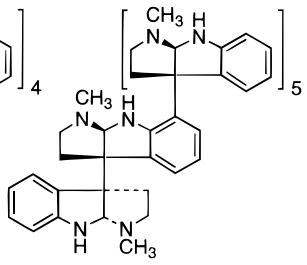
5
(3a*S*, 3a'*R* or 3a*R*, 3a'*S*)



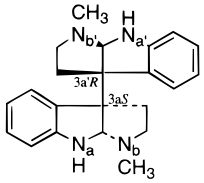
6



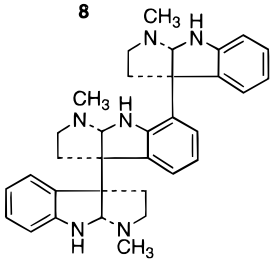
7



8



9



10

* Corresponding author. Tel.: +33 (0)169 82 4580. Fax: +33 (0)169 07 7247. E-mail: gueritte@icsn.cnrs-gif.fr.

10.1021/np9805387 CCC: $18.00
Published on Web 05/15/1999

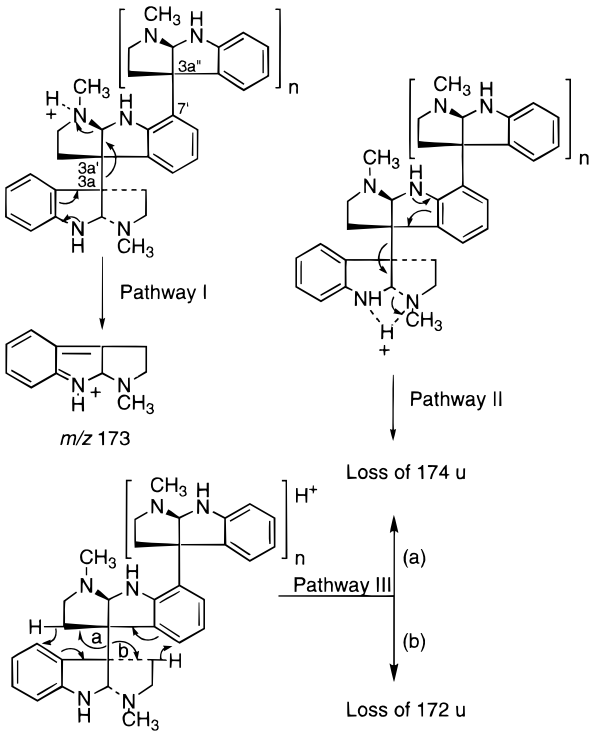


Figure 1. Mechanisms proposed to explain the fragmentations of pyrrolidinoindoline alkaloids.

alkaloids have structures made up from two to seven linked N_b-methyltryptamine units. The known dimer **9**, trimer **1**, tetramer **2**, and pentamers **3** and **4** were identified by comparison of their spectral data with literature. Among these products, the stereochemistry of **1** (3a(*S*), 3a′(*R*), 3a″-(*R*)) was established by X-ray analysis[3] whereas that of **2** (3a(*R*), 3a′(*S*), 3a″(*R*), 3a‴(*R*)[17] was suggested from NMR and CD studies.[7] In this paper, we made a tentative stereochemical assignment for **3** and **4** of unknown stereochemistry and for the new isolated compounds **5**−**8** on the basis of 2D NMR experiments, circular dichroism data, chemical correlations and biogenetic hypothesis. It should be noted that most of the signals in the ^{1}H NMR spectra of these alkaloids are unresolved due to the presence of several conformers in solution. For that reason, the NMR experiments were performed at low temperature as in the case of the previous NMR analysis of **1**[7,18] and **2**.[7]

Results and Discussion

The total alkaloid extracts of the leaves of *P. oleoides* and *P. lyciiflora* were obtained using a protocol similar to that described in ref 4. **1**, **9**, and unknown **5** were obtained after purification by chromatography of the alkaloid extract of *P. lyciiflora*. The fractionation and purification of the crude extract of *P. oleoides* led to **1**, **2**, **3**, and **4** and to the three new alkaloids **6**, **7**, and **8**.

The isolated alkaloids generated, under liquid−secondary ion mass spectrometry (LSIMS), an ion peak corresponding to the $[M + H]^+$ species. The primary structure of the alkaloids **6**, **7**, and **8** was easily deduced from their mass spectra (LSIMS) in which the presence of an ion peak at *m*/*z* 173 indicates that the terminal *Nb*-methyltryptamine unit is linked by a C-3a−C-3a′ bond to the next one (Figure 1, pathway I) such as in **1** and **3**. In contrast, this ion is not observed in the mass spectra of **2** and **4**. The $[M + H]^+$ ions, analyzed by tandem mass spectrometry (MS/MS) and submitted to collisional activation, fragmented mainly by cleavage of the C-3a−C-3a′ bond. Other low-intensity fragment ion peaks, arising from the loss of a methylamine molecule from the precursor ions, were also observed in the high-mass range of the MS/MS spectra. In the case of the *n*+1 arrangement of the pyrrolidinoindoline units, the relative contribution of the *m*/*z* 173 ion to the fragment ion current decreased strongly with increasing the molecular mass of the compound under investigation (Figure 2). The complementary part of the molecules led to two ions corresponding to the loss of 172 and 174 u from the $[M + H]^+$ ions (Figure 1, pathways II and III). The continuous increase of the $[M + H - 172]^+$ ion relative intensity when increasing the size of the molecules is particularly noteworthy (Figure 2). Such a behavior, under collisional activation conditions, suggests the occurrence of two competitive fragmentation processes, depending on the location of the charge site on the protonated molecules. The presence of the protonation site on a tryptamine unit linked by a C-3a−C-3a′ bond could induce a charge-directed fragmentation mechanism as shown in Figure 1 (pathways I and II). When other subunits are added by C-7−C-3a bonding, the presence of the charge at a site remote from the C-3a−C-3a′ bond could lead, under high-energy CID, to an alternative charge−remote fragmentation process (Figure 1, pathway III).[19] The fragmentation mechanism involved in this gas-phase reaction could be either a hydrogen rearrangement process (Figure 1, pathways IIIa and IIIb) or a homolytic cleavage of the C-3−C-3a′ bond followed by a hydrogen transfer between the radical species present in an intermediate ion−neutral complex.

N_b-Desmethyl-*meso*-chimonanthine (**5**) showed a $[MH^+]$ ion peak at *m*/*z* 333 with major fragments at *m*/*z* 173 and 159. The NMR experiments (COSY, HETCOR, HMBC, NOESY) performed at low temperature allowed the characterization of two conformers in solution and the identification of the chemical shifts for all hydrogen and carbon atoms of each subunits A and B of these conformers (Table 1). However, it was not possible to assign the absolute

Table 1. ^{1}H and ^{13}C NMR Assigments of the Two Stable Conformers at 233 K of Compound **5** (3a(*S*), 3a′(*R*) or 3a(*R*) 3a′(*S*))

	major conformer		minor conformer	
position	δ ^{1}H	δ ^{13}C	δ ^{1}H	δ ^{13}C
subunit A				
N_b-H	5.02 (s)		5.02 (s)	
2	3.18−2.73 (m)	44.87	3.18 (m)	44.57
3	2.60−2.40 (m)	35.73	2.60 (m)	37.59
3a		62.85		63.63
4a		132.22		131.31
4	7.28 (d)	123.91	5.62 (d)	124.25
5	6.80 (t)	118.45	6.28 (t)	117.73
6	7.10 (t)	128.19	6.91 (t)	128.65
7	6.46 (d)	109.10	6.49 (d)	108.19
7a		151.75		151.04
N_a-H	3.80 (s)		4.64 (s)	
8a	4.32 (s)	79.30	5.42 (s)	82.36
subunit B				
N_b′-CH_3	2.32 (s)	35.12	2.47 (s)	35.12
2′	2.82 (m)	51.85	2.82−2.42 (m)	51.66
3′	2.10 (m)	38.06	2.10 (m)	36.37
3a′		63.95		63.30
4a′		130.04		131.31
4′	5.67 (d)	124.43	7.32 (d)	124.06
5′	6.30 (t)	117.97	6.82 (t)	118.83
6′	6.91 (t)	128.43	7.10 (t)	127.95
7′	6.48 (d)	108.19	6.48 (d)	109.10
7a		150.30		151.51
N_a′-H	4.64 (s)		3.80 (s)	
8a′	5.42 (s)	82.36	4.32 (s)	82.36

[a] ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz) spectra were run in $CDCl_3$ (ppm from TMS).

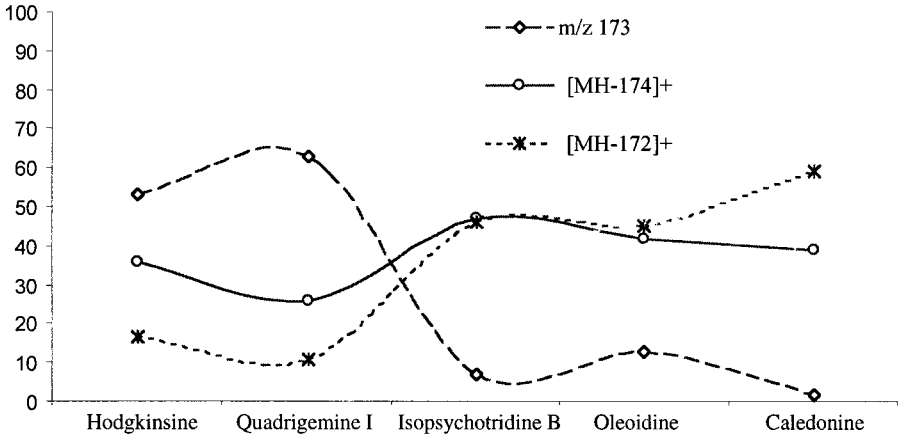


Figure 2. Relative contribution of ions *m*/*z* 173, $[MH - 174]^+$, $[MH - 172]^+$ to the fragment ion current under CID conditions for hodgkinsine (**1**), quadrigemine I (**6**), isopsychotridine B (**3**), oleoidine (**7**), and caledonine (**8**).

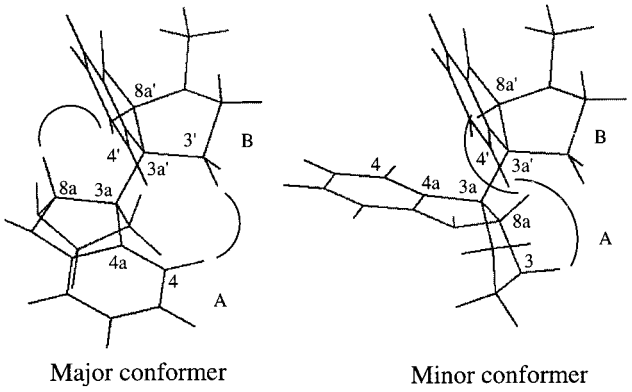


Figure 3. Energy-minimized structure of **5** with NOEs indicated.

configuration of **5** and subunit A and B can possess the 3a(*S*) or 3a(*R*) and 3a′(*R*) or 3a′(*S*) configuration, respectively. The upfield shifts of the aromatic proton H-4 (minor conformer) and H-4′ (major conformer) at 5.62 and 5.67 ppm, respectively, are due to their locations below, or above, the planes of the aromatic rings in units B and A, respectively (see Figure 3). This spatial arrangement was confirmed by the observation of NOE correlations between H-8a, H-8a′ in the major and minor conformers, between H-4′, H-3 in the minor conformer, and between H-4, H-3′ in the major conformer. Figure 3 shows the two energy-minimized structures of **5** obtained from molecular modeling. The difference between these two stable conformers is the value of the C4a−C3a−C3a′−C4a′ dihedral angle which is +59° for the major conformer and −61° for the minor conformer. Finally, the structure of **5** was confirmed by chemical correlation with **9** after methylation of **5** into **9**.

Compound **6** (named quadrigemine I) is a tetrameric isomer of previously described **2**,[4,7] quadrigemine A, and quadrigemine B.[6] It should be noted that several other quadrigemine type alkaloids have been reported,[20] but full details of their structures have never been published. In the mass spectra of **6**, the main fragment ions at *m*/*z* 173 and 517 indicate the association of one tryptamine unit to a group of three. This arrangement of the pyrrolidinoindoline units has also be reported for quadrigemine B.[6] The optical rotation of **6** ($[\alpha]_D$ +199° (EtOH)) being different from that of quadrigemine B ($[\alpha]_D$ +263° (EtOH)),[6] these two alkaloids are therefore stereoisomers. The NMR experiments performed at low temperature (260 K) showed the presence of a major (60%) and a minor (40%) conformer. The ^{1}H NMR data of the aromatic hydrogens of each

Table 2. ^{1}H Assigments of the Two Stable Conformers at 260 K of **6**[a]

	unit[b]					
	A		B			
	M[c]	m[c]	M	m	C	D
H-4	7.35	5.37	5.57	7.20	7.03	7.20
H-5	6.80	6.10	6.20	6.72	6.63	6.85
H-6	7.12	6.85	7.03	7.20	6.95	7.15
H-7	6.45	6.48				6.69

[a] ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz) spectra were run in $CDCl_3$ (ppm from TMS). [b] Pyrrolidinoindoline unit. [c] Major conformer = M, minor conformer = m.

Table 3. Circular Dichroism Data of **1**, **3**, **4**−**8**, and **10**

	1	**3**	**4**	**6**	**7**	**8**	**10**[a]
λ nm	230	223	223	221	222	221	
$\Delta\epsilon$	+3.2	+95.4	+96.6	+81.9	+191	+280	
λ nm	246	246	246	246			
$\Delta\epsilon$	−6.6	−12.8	−17.1	−5.8			
λ nm		263	263	263	263	262	271
$\Delta\epsilon$		+8.6	+16	+12.4	+23.2	+34.4	−3.2
λ nm	314	316	318	318	316	315	322
$\Delta\epsilon$	+4.4	+15.8	+18.3	+12.7	+32.9	+47.4	−4.9

[a] Data from ref 21.

conformer are summarized in Table 2. The major conformer is characterized by the upfield shift at 5.57 ppm of the hydrogen at C-4′ (H-4 of unit B) whereas the signal of the hydrogen at C-4 (H-4 of unit A) of the minor conformer appears at 5.37 ppm. This effect, similar to that observed for **5**, suggests that compound **6** also possesses a *meso*-chimonanthine unit which adopts a similar conformation to that shown in Figure 3. Moreover, the CD curve of **6** is similar to that of hodgkinsine **1** which possess one 3a(*R*) pyrrolidinoindoline unit linked to a *meso*-chimonanthine unit (Table 3). On the contrary, the cd spectra of **6** is different from that of (−)-idiospermuline **10** (Table 3), a natural trimeric alkaloid in which one pyrrolidinoindoline unit of *S* configuration is linked to a chimonanthine unit.[21] On the basis of these NMR and cd comparisons, we can suggest that **6** is formed by the association of two (*R*) pyrrolidine units with a *meso*-chimonanthine part. To confirm this hypothetical stereochemistry, **6** was reacted with methyl iodide and treated with base.[6] The crude material so obtained was reduced to give the indolylbisindoline **11** (Figure 4). The CD spectrum of **11** was found to be similar to that of the indolyl-indoline derivative **12** obtained from **1**[6] and **2** (see Experimental Section). This confirms that the configuration of the pyrrolidinoindoline

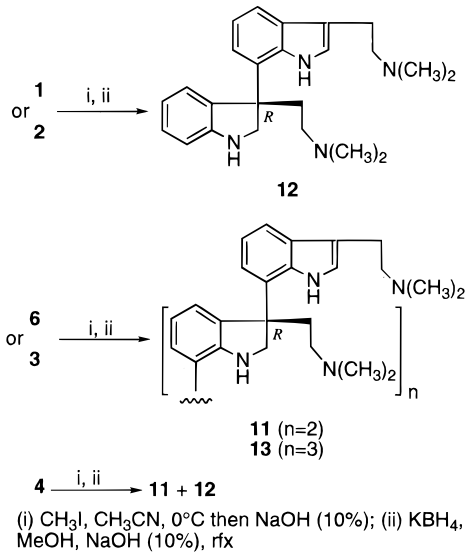


Figure 4. Chemical degradation of hodgkinsine (**1**), quadrigemine C (**2**), quadrigemine I (**6**), isopsychotridine (**3**), and psychotridine (**4**).

units linked to the *meso*-chimonanthine part of **6** is identical to that of the 3a″(*R*) pyrrolidinoindoline unit of **1**. Two configurations are thus possible for **6**: 3a(*S*), 3a′-(*R*), 3a″(*R*), 3a‴(*R*) or 3a(*R*), 3a′(*S*), 3a″(*R*), 3a‴(*R*). From a biogenetical point of view, one can assume that **6** is formed from **1** by the addition of one 3a(*R*) pyrrolidinoindoline unit to the terminal 3a(*R*) unit of hodgkinsine. Thus **6** would likely possess the 3a(*S*), 3a′(*R*), 3a″(*R*), 3a‴(*R*) configuration as shown in structure (**6**).

Although partly described, the stereochemistry of **3**[4] and **4**[9] is not known. We thus attempted to characterize the configuration of the pyrrolidino units of the pentamers **3** and **4** by 2D NMR studies, circular dichroism analysis and chemical correlations. The 2D NMR experiments performed on **3** and **4** did not give satisfactory results, the signals being unresolved even at low temperature. The CD data of these two compounds are identical to those of **1** and **6** (Table 3). This suggests a similar configuration of the pyrrolidinoindoline units of **3** and **4** to those of **1** and **6**. To assess the stereochemistry of **3** and **4**, these two compounds were subjected to chemical degradation as above (Figure 4). Thus, **4** led to the compounds **11** and **12**, which are the degradation products of **6** and **1**, respectively. Two configurations could thus be proposed for **4**: 3a(*R*), 3a′(*S*), 3a″-(*R*), 3a‴(*R*), 3a⁗(*R*) or 3a(*R*), 3a′(*R*), 3a″(*S*), 3a‴(*R*), 3a⁗(*R*). Despite all our efforts, the indolyltrisindoline **13** could not be obtained in a pure form from **3**. However the fact that its cd spectrum is similar to those of compounds **11** and **12**, suggests that the terminal units of **3** possess a 3a (*R*) configuration. Assuming as above that compound (**3**) could derive from **1** by addition of two pyrrolidinoindoline (3a*R*) units, its configuration should be as shown in structure (**3**) (3a*S*, 3a′*R*, 3a″*R*, 3a‴*R*, 3a⁗*R*).

The FAB mass spectrum of oleoidine **7** showed a [M + H$^+$] ion peak at *m*/*z* 1035 and two CID fragments at *m*/*z* 861 and 172, indicating the presence of a 5+1 disposition of the pyrrolidinoindoline units as expressed in structure **7**. The hexamer oleoidine **7** is thus different from the structure of vatine, an alkaloid of unknow configuration isolated from *Calycodendron milnei*, which possesses a 4+2 arrangement of the pyrrolidinoindoline units.[22] In the case of **8**, the mass spectrum indicated the heptamere nature of the compound with a [M + H$^+$] ion peak at *m*/*z* 1207 and a 6+1 disposition of the pyrrolidino units deduced by the presence of fragments at *m*/*z* 1033 and 173. This structure differs from the structure of the heptamer vatamine[22] which possess a 5+2 disposition of the pyrrolidinoindoline units. The NMR spectra of oleoidine **7** and caledonine **8** showed unresolved signals even at low temperature and the chemical degradation of **7** and **8** as described above, did not lead to identifiable compounds. However, the similarity of the CD spectrum of **7** and **8** (Table 3) with those of the pyrolidinoindoline alkaloids described above suggests that compound **7** possesses five *R* and one *S* configuration pyrrolidinoindoline units whereas compound **8** is characterized by the presence of six *R* and one *S* configuration units as indicated in structures **7** and **8**. In view of the fact that **1**−**4** and **6**−**8** are the main components of the alkaloid extract of *P. oleoides*, it is reasonable to suggest that these alkaloids are formed by the addition of one to five N_b-methyltryptamine units of (*R*) configuration to a *meso*-chimonanthine unit. It is however surprising that **9** was not found in *P. oleoides*.

In conclusion, four new pyrrolidinoindoline alkaloids, **6**−**8**, have been isolated from *P. lyciiflora* and *P. oleoides*. Comparative spectrometric studies combined with chemical degradation and biogenetical hypothesis allowed us to propose the configuration of the pyrrolidinoindoline units which composed these alkaloids. The effect of all these compounds on the growth hormone release by pituitary cells has been analyzed in detail.[23] This study showed that some of these alkaloids act as functional antagonists of somatostatin and that they could possess a growth hormone-releasing peptide (GHRP)-like activity.[24] These results will be reported elsewhere in due time.[25]

Experimental Section

General Experimental Procedures. Optical rotations have been measured on a Perkin-Elmer 141 MC polarimeter. Infrared spectra were recorded on a Nicolet FT-IR 205 and UV spectra on a Perkin-Elmer lambda 5 spectrometer. ^{1}H and ^{13}C NMR spectra were recorded on Bruker, AM-250, AM-300, or AM-400 spectrometers using tetramethylsilane as internal standard. Chemical shifts are expressed in parts per million (ppm); s, d, d, t and m indicate singlet, doublet, triplet and multiplet signals, respectively, and M and m indicate major conformer and minor conformer, respectively. Mass spectra were measured on a AEI MS-9 (CI), and exact mass measurements have been carried out, when possible, in the chemical-ionization mode using methane as reagent gas. LSIMS and MS/MS experiments were performed with a Zabspec-T tandem mass spectrometer (Micromass, Manchester, UK). Detailed experimental conditions have been described previously.[26] CD spectra have been measured on a Jobin-Yvon Mark V or VI dichrograph. TLC were realized with Si gel 60 F_{254} and spots revealed by spraying with CAS (Cerium Ammonium Sulfate) or Dragendorff reagents. Silica gel or neutral alumina (activity II−III) were used for column chromatography. HPLC purification was carried out on a Ultrabase C-18 column (15 μm, 10 × 250 mm, flow rate: 3 mL/min) or on a Delta Pak column (15 μm, 40 × 250 mm, flow rate: 80 mL/min). HPLC analyses were realized on a Ultrabase C-18 column (15 μm, 4 × 250 mm) or on a Kromasil C-18 column (15 μm, 3.6 × 250 mm) at a flow rate of 1 mL/min. Molecular modeling studies were performed on a Silicon Graphics Indigo II (R10000) workstation, using MacroModel (version 3.1). Conformational searches and comparison of conformers were performed with the Monte Carlo procedure using MM2 force field parameters.

Plant Material. *P. lyciiflora* leaves were collected at Plateau de Dogny (cote 88) in New-Caledonia by M. Litaudon (lot LIT 029) in September 1995. *P. oleoides* leaves were collected in July 1987 at Montagne des Sources (cote 900) by T. Sévenet and S. Labarre (lot Sévenet-Labarre 8). Voucher specimens are deposited to Centre ORSTOM of Nouméa (New-

Caledonia) and to the Museum National d'Histoire Naturelle (Paris, France).

Purification and Isolation Procedures. Dried and ground *P. lyciiflora* leaves (1.6 kg) were moistened with 20% aqueous NH_4OH. After extraction with methanol in a Soxhlet apparatus, the mixture was filtered, and the methanolic solution was concentrated. The crude extract was taken up with 5% aqueous HCl. The solution was then washed with methylene chloride. To the aqueous phase was added NH_4OH to pH 9−10. After extraction with CH_2Cl_2, the organic phases were washed with water, dried over Na_2SO_4 and evaporated to dryness yielding the alkaloid extract (13 g, 0.8% yield). HPLC analyses of the alkaloid extract (mobile phase: MeOH−H_2O−Et_3N 75:25:0.5) gave the retention time and yield of the alkaloids **5** (8 min, 15%), **9** (9.5 min, 65%), and **1** (23.5 min, 10% yield). The alkaloid extract (200 mg) was chromatographed on TLC (CH_2Cl_2−MeOH−NH_4OH 90:10:1) to give pure **9**, **1**, and **5**. A crude alkaloid extract (91 g, 2.27%) of *P. oleoides* was obtained from 4 kg of dried leaves using the same procedure as above. HPLC analyses of the alkaloid extract of *P. oleoides* (mobile phase: MeOH−H_2O−Et_3N 90:10:0.5) allow the estimation of the yields of each alkaloid which are given following their retention time: **1** (6.30 min, 5.2%), **6** (8 min, 8.7%), **2** (9.30 min, 45.3%), **3** (11.40 min, 5.5%), **4** (12.50 min, 9.9%), **7** (14.10 min, 11%), **8** (22.10 min, 8%). The fractionation of the alkaloid extract of *P. oleoides* (20 g) was realized by column chromatography performed on alumina using a step gradient of MeOH (0% to 100%) in CH_2Cl_2. This led to 14 fractions some of which being further purified by HPLC using MeOH−H_2O−Et_3N 85:15:5 as mobile phase to give pure alkaloids.

***N*$_b$-Desmethyl-*meso*-chimonanthine (5):** amorphous white powder: $[\alpha]^{25}_D$ 0.5° (c = 1, EtOH); UV (EtOH) λ_{max} (log ϵ) 245 (4.00), 303 (3.60) nm; IR ($CHCl_3$) ν_{max} 3430, 2965, 2940, 1610 cm^{-1}; ^1H and ^{13}C NMR ($CDCl_3$) at 233 K (see Table 1); CIMS (isobutane) m/z 333 $[M + 1]^+$ (100), 159, 173; *anal*. C 75.80%, H 7.33%, calcd for $C_{21}H_{24}N_4$, C 75.87%, H 7.28%.

To a solution of **5** (10 mg, 3×10^{-5} mol) in CH_3CN (1 mL) was added a 10% solution of MeI in CH_3CN (20μL) at 0 °C. After standing at room temperature for 5 h, the mixture was concentrated to dryness. MeOH and water were added and the solution was extracted with EtOAc. The organic phase was dried over Na_2SO_4 and evaporated to dryness. The crude material was purified by preparative TLC (90:10:0.5 CH_2Cl_2−MeOH−NH_4OH) to give *meso*-chimonanthine **9** (2.5 mg, 24%) and **5** (6 mg).

Quadrigemine I (6): amorphous white powder: $[\alpha]^{25}_D$ +16° (c = 0.34, $CHCl_3$) and +199° (c = 0.34, EtOH); UV (EtOH) λ_{max} (log ϵ): 244 (4.45) and 302 (4.13) nm; IR ($CHCl_3$) ν_{max} 3430, 2965, 2940, 1610 cm^{-1}; CD (see Table 3); ^1H NMR ($CDCl_3$, 260 K, 400 MHz) δ 1.85−3.15 (m, 16 H, 8×CH_2), 2.30, 2.40, and 2.45 (3s, 12 H, 4×N−CH_3), 3.70 (br s, NH), 4.20 (br s, NH), 4.95, 5.00, and 5.05 (3s, 4 H, H-8a, H-8a′, H-8a″, and H-8a‴), for aromatic hydrogens see Table 2; ^{13}C NMR ($CDCl_3$, 260 K, 100 MHz) δ 36.0 (N−CH_3), 38.0 and 39.0 (C-3), 53.0 (C-2), 60.0 and 63.0 (C-3a), 83.0 and 88.0 (C-8a), 109.0 and 110.0 (C-7), 117.0, 118.5, 119.0, and 119.5 (C-5), 124.0, 126.0, 127.5, 128.5, and 129.5 (C-4 and C-6), 132.0 (C-4a), 150.0 (C-7a); LSIMS m/z 691 $[M + 1]^+$, 517, 173, 130; MS/MS m/z 691, 519 $[MH - 172]^+$, 517 $[MH - 174]^+$, 173. HRMS m/z 691.4223 (calcd for $C_{44}H_{51}N_8$, 691.4236).

Oleoidine (7): amorphous yellow powder: $[\alpha]^{25}_D$ +89° (c = 0.4, $CHCl_3$) and +371° (c = 0.2, EtOH); UV (EtOH) λ_{max} (log ϵ): 244 (4.55) et 302 (4.26) nm; IR ($CHCl_3$) ν_{max} 3430, 2970, 2940, 2880, 1610; CD see Table 3; ^1H NMR ($CDCl_3$, 250 MHz) δ 1.90−3.05 (m, 24 H, CH_2), 2.25, 2.30, 2.35, and 2.40 (4s, 18 H, N−CH_3), 3.40−5.05 (br s, NH and H-8a), 5.25−7.15 (m, 20 H, H-aromatic); ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 35.17, 35.59, and 35.73 (N−CH_3), 38.68 and 38.73 (C-3), 52.59, 52.78, and 52.99 (C-2), 60.44, 60.98, and 63.34 (C-3a), 82.84, 83.05, and 86.60 (C-8a), 109.30 (C-7), 116.3 and 119.38 (C-5), 122.68, 124.15, 125.74, 126.17, 128.43, and 128.68 (C-4 and C-6), 132.78 (C-4a), 150.35 and 150.7 (C-7a); LSIMS m/z 1035 $[M + 1]^+$, 861, 173; MS/MS m/z 1035, 863 $[MH - 172]^+$, 861 $[MH - 174]^+$.

Caledonine (8): amorphous yellow powder: $[\alpha]^{25}_D$ +125° (c = 0.4, $CHCl_3$) and +462° (c = 0.2, EtOH); UV (EtOH) λ_{max} (log ϵ): 243 (4.62), 303 (4.35) nm; IR ($CHCl_3$) ν_{max} 3430, 2965, 2940, 1610; CD see Table 3; ^1H NMR ($CDCl_3$, 250 MHz) δ 1.90−3.10 (m, 28 H, CH_2), 2.15, 2.25, 2.35, 2.42, and 2.45 (5s, 21 H, N−CH_3), 3.95−5.05 (br s, NH and H-8a), 5.30−7.20 (m, 23 H, H-aromatic); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 35.37, 35.63 (N−CH_3), 38.30 and 38.56 (C-3), 52.08, 52.38 (C-2), 60.00, 60.55, and 63.05 (C-3a), 85.98 and 86.94 (C-8a), 107.69 and 108.83 (C-7), 117.3 and 118.9 (C-5), 123.23, 123.61, 124.09, 125.21, 125.35, 128.07 (C-4 and C-6), 132.11 and 132.38 (C-4a), 148.59, 148.95, and 150.47 (C-7a); LSIMS m/z 1207 $[M + 1]^+$, 1035, 1033, 173. MS/MS m/z 1207, 1035 $[MH - 172]^+$.

Chemical Degradation of 1−4 and 6. **1** (45 mg), **2** (100 mg), **6** (37 mg), **3** (36 mg), and **4** (30 mg), in CH_3CN (1 mL), were treated separately with methyl iodide (0.5 mL). The solutions were left at 0 °C for 2 h. After hydrolysis, the aqueous phases were treated with 10% NaOH and extracted with EtOAc. The organic layers were dried over Na_2SO_4 and evaporated under reduced pressure after filtration. The crude materials were dissolved in methanol (1 mL) and treated with KBH_4 (200 mg) and 10% NaOH (50 μL). After reflux for 3 h, the solutions were extracted with Et_2O, and the organic layers were dried over Na_2SO_4 and evaporated under reduced pressure after filtration. The crude materials were purified by preparative TLC (CH_2Cl_2−MeOH−NH_4OH (90:10:1)) to give compound **12** (8.5 mg) from **1**, compound **12** (22 mg) from **2**, compound **11** (9 mg) from **6**, compound **13** (2 mg) from **3**, and compounds **11** (3 mg) and **12** (2 mg) from **4**.

Compound (11): amorphous white powder: $[\alpha]^{25}_D$ +181° (c = 0.25, EtOH); UV (EtOH) λ_{max} (log ϵ): 230 (4.55), 288 (4.22), 293 (4.2), λ_{infl} 262 (4.09) nm; IR ($CHCl_3$) ν_{max} 3430, 2965, 2940, 1610; CD (EtOH) λ nm ($\Delta\epsilon$) 217 (+25.9), 229 (−30.8), 243 (+17.7), 287 (−1.9), 305 (+9.9); ^1H NMR ($CDCl_3$, 250 MHz) δ 2.20−2.92 (m, 12 H), 2.29, 2.31 and 2.38 (3s, 18 H), 3.68 (s, 1H), 3.71 and 3.82 (dd, J = 10 Hz, 4H), 4.57 (br s, 1H), 6.67 and 7.43 (m, 10 H), 6.83 (s, 1H), 9.17 (br s, 1H); cims m/z 565 $[M + 1]^+$, 237, 131; HRMS m/z 564.3924 (calcd for $C_{36}H_{48}N_6$, 564.3940).

Compound 12 was identical with the indolyl-indoline derived from hodgkinsine and quadrigemine A[6]: CD (EtOH) λ nm ($\Delta\epsilon$) 212 (+29.1), 228 (−76), 242 (+22.1), 283 (−2.5), 304 (+7.5); HRMS m/z 376.2620 (calcd for $C_{24}H_{32}N_4$, 376.2626).

Compound 13: UV (EtOH) λ_{max}: 230, 288, 293, λ_{infl} 262 nm; IR ($CHCl_3$) ν_{max} 3420, 2965, 2935, 1610; cd (EtOH) λ nm ($\Delta\epsilon$) 211 (+49.4), 225 (−108.7), 239 (+30.3), 302 (+12.3); ^1H NMR ($CDCl_3$, 250 MHz) δ 2.16−2.90 (m), 2.46, 2.50 and 2.54 (3s), 3.76 and 4.36 (2 m), 3.94 and 4.68 (2s), 6.70−7.50 (m), 9.65 (br s); cims m/z $[M + 1]^+$ 754.

References and Notes

(1) Rasolonjanahary, R.; Sévenet, T.; Kordon, C. In *Advances in Medicinal Phytochemistry*; Barton, D., Ollis, W. D., Eds.; 1986; pp 81−87.
(2) Anet, E. F. L.; Hughes, G. K.; Ritchie, E. *Aust. J. Chem.* **1961**, *14*, 173−174.
(3) Fridrichsons, J.; Mackay, M. F.; Mathieson, A. McL. *Tetrahedron* **1974**, *36*, 85−92.
(4) Libot, F.; Miet, C.; Kunesch, N.; Poisson, J. E.; Pusset, J.; Sévenet, T. *J. Nat. Prod.* **1987**, *50*, 468−473.
(5) Quadrigemine C is possibly one of the diastereoisomers of the quadrigemine A mixture reported by Parry and Smith (see ref 6). The configuration of quadrigemine C was suggested as shown in structure **2** from NMR and CD spectra analysis (see ref 7).
(6) Parry, K. P.; Smith, G. F. *J. Chem. Soc., Perkin Trans. 1* **1978**, 1671−1682.
(7) Guéritte-Voegelein, F.; Sévenet, T.; Pusset, J.; Adeline M.−T.; Gillet, B.; Beloeil, J.-C.; Guénard, D.; Potier, P.; Rasolonjanahary, R.; Kordon, C. *J. Nat. Prod.* **1992**, *55*, 923−930.
(8) The stereochemistry of **3** and **4** was deduced from this work.
(9) Hart, N. K.; Johns, S. R.; Lamberton, J. A.; Summons, R. E. *Aust. J. Chem.* **1974**, *27*, 639−646.
(10) Libot, F. Plantes à alcaloïdes indoliques de Nouvelle-Calédonie. Ph.D. Thesis, Université de Paris−Sud Orsay, France, 1982.
(11) Rasolonjanahary, R.; Sévenet, T.; Guéritte-Voegelein, F.; Kordon, C. *Eur. J. Pharm.* **1995**, *285*, 19−23.
(12) Hendrickson, J. B.; Göschke, R.; Rees, R. *Tetrahedron* **1964**, *20*, 565−579.
(13) Hall, E. S.; McCapra, F.; Scott, A. I. *Tetrahedron* **1964**, *23*, 4131−4141.
(14) Hino, T.; Kodato, S.; Takahashi, K.; Yamagushi, H.; Nakagawa, M. *Tetrahedron Lett.* **1978**, *19*, 4913.

(15) Link, J. T.; Overman, L. *J. Am. Chem. Soc.* **1996**, *118*, 8166−8167.
(16) Adjibade, Y.; Weniger, B.; Quirion, J.-C.; Kuballa, B.; Cabalion, P.; Anton, R. *Phytochemistry*, **1992**, *31*, 317−319.
(17) For clarity, the numbering used for the pyrrolidinoindoline alkaloids is based on that of **1** as indicated in the figure. This is different from the numbering used in our preliminary paper.[7]
(18) Very recently, the NMR spectra of **1** at low temperature has also been described: Verotta, L.; Pilati, T.; Tato, M.; Elisabetsky, E.; Amador, T. A.; Nunes, D. S. *J. Nat. Prod.* **1998**, *61*, 392−396.
(19) Gross, M. L. *Int. J. Mass Spectrom.* **1992**, *118/119*, 137−165.
(20) (a) Saad, H.-E. A.; El-Sharkawy, S. H.; Shier, W. T. *Planta Med.* **1995**, 313−316. (b) Saad, H.-E. A. Contribution à l'étude d'alcaloïdes d'espèces nouvelles de Rubiacées: *Calycodendron milnei* (A. Gray) A. C. Smith et de *Lindenia austro-caledonica* Brongn, et de quelques propriétés pharmacologiques. Ph.D. Thesis, Université Louis Pasteur de Strasbourg, France, 1986. (c) Adjibade Y. Pharmacognosie du *Psychotria forsteriana* A. Gray (Rubiaceae). Ph.D. Thesis, Université Louis Pasteur de Strasbourg, France, 1989.
(21) Duke, R. K.; Allan, R. D.; Johnston, G. A. R.; Mewett, K. N.; Mitrovic, A. D.; Duke, C. C.; Hambley, T. W. *J. Nat. Prod.* **1995**, *58*, 1200−1208.
(22) Adjibadé, Y.; Saad, H.; Sévenet, T.; Kuballa, B.; Quirion, J.-C.; Anton, R. *Planta Med.* **1990**, *56*, 212−215.
(23) Jannic, V. *Psychotria oleoides* et *P. lyciiflora* Schlecht., Rubiaceae de Nouvelle-calédonie: Alcaloïdes poly-indoliniques modulateurs de la sécrétion d'hormone de croissance chez le rat. Ph.D. Thesis, Université Paris XI, France, 1997.
(24) Bowers, C. Y.; Sartor, A. O.; Reynolds, G. A.; Badger, T. M. *Endocrinology* **1991**, *128*, 2027−2035.
(25) The biological studies have been realized in collaboration with Dr. M. T. Bluet-Pajot and Dr. C. Kordon (INSERM, Unité 159, Paris).
(26) Queiroz, E. F.; Roblot, F.; Laprévote, O.; Serani, L.; Cavé, A. *J. Nat. Prod.* **1997**, *56*, 760−765.

NP9805387