

Review

¹H and ¹³C-NMR Data of the Simplest Plumeran Indole Alkaloids Isolated from *Aspidosperma* Species

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Abstract: Indole alkaloids are the chemotaxonomic markers of the *Aspidosperma* genera. Those that have the simplest plumeran skeleton are classified as the precursors of biosynthetic routes and the intermediates for several synthetic reactions. This work aims to review the ¹H and ¹³C-NMR data, up to 2011, describing the skeleton of 35 different plumeran indole alkaloids, from a group of 46 of them, and highlight the main spectral differences amongst them.

Keywords: *Aspidosperma*; plumeran indole alkaloids; NMR spectral data

1. Introduction

The Apocynaceae family consists of 424 genera, divided into 17 tribes that are subdivided under five subfamilies: Rauwolfioideae, Apocynoideae, Asclepioideae, Periplocoideae and Secamonioidae [1]. About 400 species of Apocynaceae have been identified and grouped into 41 genera in Brazil, 78% of which are found only in the Amazonia region [2]. Several Apocynaceae are used for landscaping. These include the arboreal *Tabernaemontana* and *Plumeria*, the climbing *Alamanda* and *Mandevilla*, and the shrubby *Catharanthus* and *Nerium* [3]. The wood of some species is of particular commercial importance for local development. Species such as the popularly named “peroba” and “guatambu” are useful for making furniture and buildings [4].

The *Aspidosperma* genus is included in the Rauwolfioideae subfamily, Alstoniae tribe. The genus includes about 57 species, divided according to their chemotaxonomy into eight series—Rigida,

Nitida, Quebranchines, Polyneura, Pyricolla, Nobile, Macrocarpa and Tomentosa, all of which are restricted to the American tropical and subtropical regions [5,6]. *Aspidosperma* is another genus of Apocynaceae of high commercial value due the good quality of its wood.

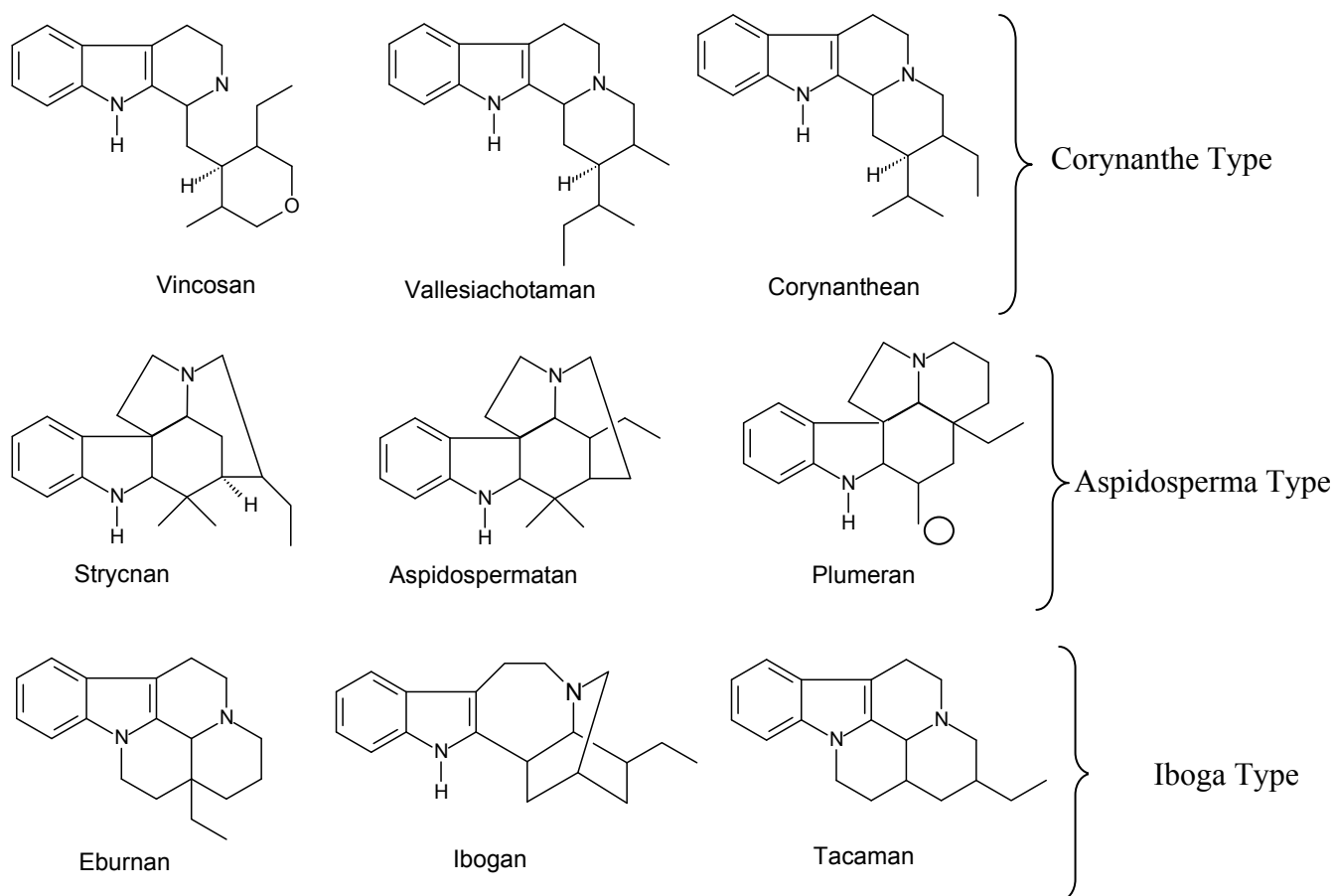
Some species are also used in folk medicine; the infusion of its stem barks is used to treat a number of diseases [7]. For example, the extracts of the *Aspidosperma* stem bark, are characterized by the presence of indole alkaloids with high structural diversity. These alkaloids are responsible for the many pharmacological effects known for the plant [8]. The experimental assays [9–13] involving *A. ramiflorum* Müll. Arg., *A. pyrifolium* Mart., *A. megalocarpon* Müll. Arg., *A. macrocarpon* Mart. and *A. quebracho-blanco* Schltdl. species, attest to their popular use as an antileishmanial and an antimalarial. Alkaloids found in root bark of *A. ulei* Markgr. were used to evaluate and prove pro-erectile effects [14,15]. *A. subincanum* Mart., used in folk medicine to treat *Diabetes mellitus* and hypercholesterolemia [16], was proven to have a low acute toxicity in *in vivo* tests, resulting in its characterization as a non-toxic treatment [17]. *A. ramiflorum* Müll. Arg. exhibited antibacterial activity against the standard strains of Gram-positive (*Bacillus subtilis* and *Staphylococcus aureus*) and Gram-negative (*Escherichia coli* and *Pseudomonas aeruginosa*) [18] bacteria. The extracts of several different parts of *A. polyneuron* Müll. Arg. were tested against a wide range of fungi; only the ethanolic extract of the stem was able to inhibit the growth of *Cladosporium herbarum* [19].

The structural diversity of the indole alkaloids can be classified by examining their biosynthetic origins. The occurrence of complex alkaloids containing indole moieties is restricted to a few families; the best sources appear in the Apocynaceae, Loganiaceae and Rubiaceae families [20]. The indole alkaloids are *N*-methyl derivatives of tryptophan, which has a terpenic unit that originates from the mevalonic acid pathway. The metabolic origins of the remaining portion consist of a chain of ten carbon atoms, the sub-architecture of which is useful for dividing indole alkaloids into three classes: Iboga, Corynanthe and *Aspidosperma*. Usually, the C₉ or the C₁₀ chain units are shown to be of a terpenoid origin and are identified as secologanins (secoiridoids) [20,21].

Each of the three classes mentioned can be subdivided again, giving rise to nine other subclasses (Figure 1), according to Danieli and Palmisano [22]. Vincadifformine (**1**), as well as tabersonine (which presents a double bond between C-14 and C-15) are of *Aspidosperma* type. However, the loss of the carbon atom linked to C-16 (indicated by a circle and corresponding to the carboxylate function of secologanin) by hydrolysis/decarboxylation, originates most of the plumeran indole alkaloids [21].

The alkaloids isolated from *Aspidosperma* species have been exhaustively studied through phytochemical prospection, synthesis and semi-synthesis [23–26], as well the expression of secondary metabolites in cell cultures [27].

This work presents a review of the literature describing the ¹H and ¹³C-NMR data of 35 alkaloids with a simplest plumeran skeleton. This alkaloid subclass was chosen for their structural diversity (the basic ring skeleton is representative of a large number of natural compounds) and the review of Pereira *et al.* (2007) [2] was adopted as a guideline. In this referred work, the criterion of classification is the same as that adopted by Manske [28]. Some structures (nomenclature and structure) were adjusted based on the literature.

Figure 1. Subclasses of indole alkaloids.

The numbering of the structures in this work followed the method proposed by Le Men and Taylor (Figure 2) [20]. In many cases, it was not possible to obtain the NMR data, because the compounds' structures were elucidated by other techniques (Infrared—IR, Ultraviolet—UV, Mass Spectrometry—MS).

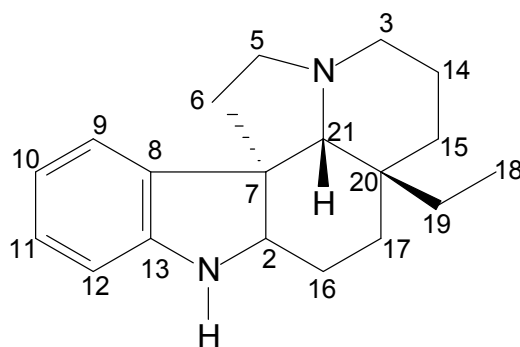
Figure 2. Numbering of the structure proposed by Le Men and Taylor [20].

Table 1 lists the alkaloids, the respective plant origin and the spectral data available for each. The alkaloids listed are shown in Figures 3, 4 and 5.

Table 1. Plumeran indole alkaloids isolated from *Aspidosperma*.

Alkaloid	Species [2]	Data
(–)-Vincadifformine (1)	<i>A. macrocarpon</i> , <i>A. pyrifolium</i>	¹ H-NMR [29], ¹³ C-NMR [30]
Fendlispermene (2)	<i>A. fendleri</i>	*
Spegazzinine (3)	<i>A. chakensis</i>	¹ H-NMR [31]
Spegazzinidine (4)	<i>A. chakensis</i>	¹ H-NMR [31]
1,2-Dehydroaspidospermidine (5)	<i>A. neblinae</i> , <i>A. quebracho-blanco</i>	¹ H-NMR, ¹³ C-NMR [32]
1,2-Dehydro- <i>N</i> -deacetyl-aspidospermin (6)	<i>A. neblinae</i>	IR, UV, MS [33]
(+)-Pyrifolidine (7)	<i>A. cylindrocarpon</i> , <i>A. neblinae</i> , <i>A. pyrifolium</i> , <i>A. refractum</i> , <i>A. quebracho-blanco</i>	¹ H-NMR, ¹³ C-NMR [32]
(+)-Aspidospermene (8)	<i>A. album</i> , <i>A. australe</i> , <i>A. exalatum</i> , <i>A. peroba</i> , <i>A. polyneuron</i> , <i>A. pyricollum</i> , <i>A. pyrifolium</i> , <i>A. quebracho-blanco</i> , <i>A. quirandy</i> , <i>A. sessiflorum</i> , <i>A. rhombeosignatum</i>	¹ H-NMR, ¹³ C-NMR [34,35]
15-Methoxyaspidospermene (9)	<i>A. pyrifolium</i>	¹ H-NMR, ¹³ C-NMR [32]
Aspidospermidine (10)	<i>A. neblinae</i> , <i>A. quebracho-blanco</i> , <i>A. pyrifolium</i> , <i>A. rhombeosignatum</i>	GC/MS [36,37]
Deacetylaspidoaspermene (11)	<i>A. neblinae</i> , <i>A. polyneuron</i> , <i>A. pyrifolium</i> , <i>A. quebracho-blanco</i>	¹ H-NMR [38]
(+)- <i>O</i> -Demethylaspidoaspermene (12)	<i>A. discolor</i> , <i>A. eburneum</i> , <i>A. excelsum</i> , <i>A. neblinae</i> , <i>A. pyricollum</i>	¹ H-NMR [39]
<i>N</i> -Methyl-deacetylaspidoaspermene (13)	<i>A. quebracho-blanco</i>	MS [40]
Demethoxyaspidospermene (14)	<i>A. discolor</i> , <i>A. macgravianum</i> , <i>A. neblinae</i> , <i>A. pyrifolium</i>	¹ H-NMR [39]
Aspidosine (15)	<i>A. quebracho-blanco</i>	IR, MS [41]
10-Methoxy-aspidospermidine (16)	<i>A. pyrifolium</i>	¹ H-NMR, ¹³ C-NMR [42]
Demethoxypalosine (17)	<i>A. discolor</i> , <i>A. exalatum</i> , <i>A. limae</i> , <i>A. rhombeosignatum</i>	MS [39]
Palosine (18)	<i>A. polyneuron</i> , <i>A. pyrifolium</i>	¹ H-NMR, ¹³ C-NMR [42]
<i>O</i> -Demethylpalosine (19)	<i>A. exalatum</i> , <i>A. limae</i> , <i>A. pyrifolium</i>	¹ H-NMR [43]
Aspidocarpine (20)	<i>A. album</i> , <i>A. formosanum</i> , <i>A. limae</i> , <i>A. marcgravianum</i> , <i>A. megalocarpon</i>	¹ H-NMR, ¹³ C-NMR [44]
<i>O</i> -Demethylaspidocarpine (21)	<i>A. album</i> , <i>A. cuspa</i> , <i>A. melanocalyx</i>	¹ H-NMR [45]
Deacetylpyrifolidine (22)	<i>A. neblinae</i> , <i>A. quebracho-blanco</i>	GC/MS [46]
15-Methoxypyrifolidine (23)	<i>A. pyrifolium</i>	¹ H-NMR, ¹³ C-NMR [32]
Aspidolimene (24)	<i>A. limae</i> , <i>A. obscurinervium</i>	¹ H-NMR [47]
<i>N</i> -Propionyl-16,17-dihydroxyaspidospermidine (25)	<i>A. melanocalyx</i>	GC/MS [48]
<i>N</i> -formyl-aspidospermidine (26)	<i>A. pyrifolium</i>	** [49]
<i>N</i> -Methylaspidospermidine (27)	<i>A. quebracho-blanco</i>	¹ H-NMR, ¹³ C-NMR [50]
Limaspermene (28)	<i>A. limae</i>	¹ H-NMR [51]
11-Methoxylimaspermene (29)	<i>A. album</i>	¹ H-NMR [52]
Limaspermidine (30)	<i>A. rhombeosignatum</i>	MS [53]

Table 1. Cont.

Alkaloid	Species [2]	Data
Limapodine (31)	<i>A. album</i> , <i>A. limae</i> , <i>A. marcgravianum</i>	¹ H-NMR [52]
11-Methoxylimapodine (32)	<i>A. limae</i>	¹ H-NMR [52]
Cylindrocarpinol (33)	<i>A. cylindrocarpon</i>	IR, MS [54]
<i>N</i> -Formyleilindrocarpinol (34)	<i>A. cylindrocarpon</i>	¹ H-NMR [55]
<i>N</i> -Acetylcylindrocarpinol (35)	<i>A. cylindrocarpon</i>	¹ H-NMR [25]
Cylindrocarine (36)	<i>A. cylindrocarpon</i>	¹ H-NMR [23,25], ¹³ C-NMR [23]
19-Hydroxycylindrocarine (37)	<i>A. cylindrocarpon</i>	¹ H-NMR [54]
Cylindrocarpidine (38)	<i>A. cylindrocarpon</i>	¹ H-NMR [25], ¹³ C-NMR [32]
Cylindrocarpine (39)	<i>A. cylindrocarpon</i>	¹ H-NMR [56]
<i>N</i> -Methyl-cylindrocarine (40)	<i>A. cylindrocarpon</i>	¹ H-NMR [55]
<i>N</i> -Formyl-cylindrocarine (41)	<i>A. cylindrocarpon</i>	¹ H-NMR, ¹³ C-NMR [23]
<i>N</i> -Benzoylcylindrocarine (42)	<i>A. cylindrocarpon</i>	¹ H-NMR [55]
12-Demethoxy- <i>N</i> -acetylcylindrocarine (43)	<i>A. cylindrocarpon</i>	¹ H-NMR, ¹³ C-NMR [23]
<i>N</i> -Dihydrocinamoil-19-hydroxycylindrocarine (44)	<i>A. cylindrocarpon</i>	¹ H-NMR [55]
<i>N</i> -Formyl-19-hydroxycylindrocarine (45)	<i>A. cylindrocarpon</i>	¹ H-NMR [55]
<i>N</i> -Cinnamoyl-19-hydroxycylindrocarine (46)	<i>A. cylindrocarpon</i>	¹ H-NMR [55]

* Data not found; ** The alkaloid was identified by comparison, after the acetylation of aspidospermidine.

Figure 3. Plumeran indole alkaloids: methyl- β -anilineacrylate (1), fendlispermene (2), spegazzinine (3 and 4), aspidospermidine (5 and 6) and pyrifolidine (7 to 9) skeletons [28].

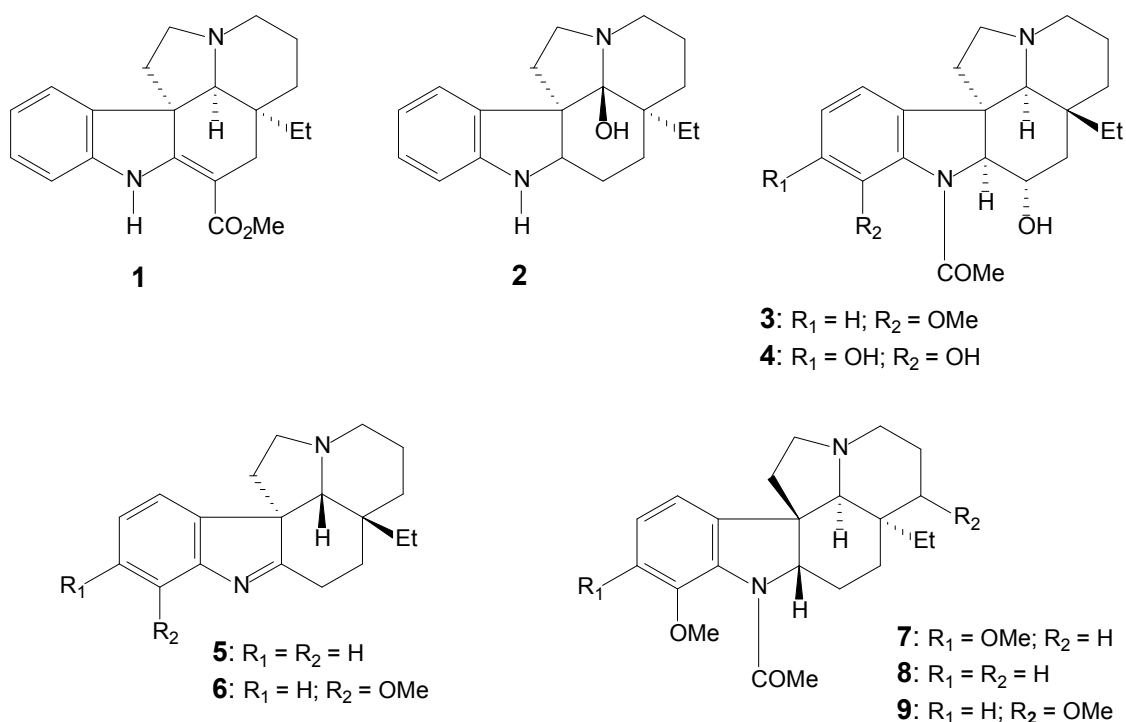
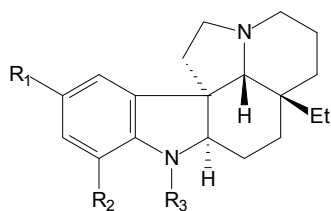
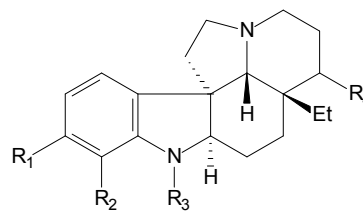
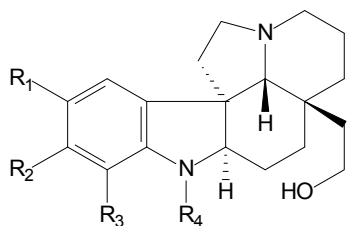


Figure 4. Plumeran indole alkaloids: Aspidospermine (10 to 19) and Aspidoscarpine (20 to 27) types [28].

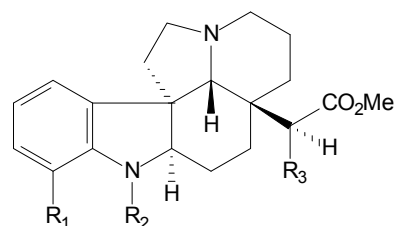
- 10:** $R_1 = R_2 = R_3 = H$
11: $R_1 = H; R_2 = OMe; R_3 = H$
12: $R_1 = H; R_2 = OH; R_3 = COMe$
13: $R_1 = H; R_2 = OMe; R_3 = Me$
14: $R_1 = R_2 = H; R_3 = COMe$
15: $R_1 = R_2 = H; R_3 = OH$
16: $R_1 = OMe; R_2 = R_3 = H$
17: $R_1 = R_2 = H; R_3 = COEt$
18: $R_1 = H; R_2 = OMe; R_3 = COEt$
19: $R_1 = H; R_2 = OH; R_3 = COEt$



- 20:** $R_1 = OMe; R_2 = OH; R_3 = COMe; R_4 = H$
21: $R_1 = R_2 = OH; R_3 = COMe; R_4 = H$
22: $R_1 = R_2 = OMe; R_3 = COMe; R_4 = H$
23: $R_1 = R_2 = OMe; R_3 = COMe; R_4 = OMe$
24: $R_1 = OMe; R_2 = OH; R_3 = COEt; R_4 = H$
25: $R_1 = R_2 = OH; R_3 = COEt; R_4 = H$
26: $R_1 = R_2 = H; R_3 = CHO; R_4 = H$
27: $R_1 = R_2 = H; R_3 = Me; R_4 = H$

Figure 5. Plumeran indole alkaloids: Limaspermine (28 to 35) and Cylindrocarine (36 to 46) skeletons [28].

- 28:** $R_1 = R_2 = H; R_3 = OH; R_4 = COEt$
29: $R_1 = H; R_2 = OMe; R_3 = OH; R_4 = COEt$
30: $R_1 = R_2 = R_3 = H; R_4 = Me$
31: $R_1 = R_2 = H; R_3 = OH; R_4 = COMe$
32: $R_1 = H; R_2 = OMe; R_3 = OH; R_4 = COMe$
33: $R_1 = R_2 = H; R_3 = OMe; R_4 = H$
34: $R_1 = R_2 = H; R_3 = OMe; R_4 = CHO$
35: $R_1 = R_2 = H; R_3 = OMe; R_4 = COMe$



- 36:** $R_1 = OMe; R_2 = R_3 = H$
37: $R_1 = OMe; R_2 = H; R_3 = OH$
38: $R_1 = OMe; R_2 = COMe; R_3 = H$
39: $R_1 = OMe; R_2 = COCH=CHC_6H_5; R_3 = H$
40: $R_1 = OMe; R_2 = Me; R_3 = OH$
41: $R_1 = OMe; R_2 = CHO; R_3 = H$
42: $R_1 = OMe; R_2 = Bz; R_3 = H$
43: $R_1 = H; R_2 = COMe; R_3 = H$
44: $R_1 = OMe; R_2 = COCH_2CH_2C_6H_5; R_3 = OH$
45: $R_1 = OMe; R_2 = CHO; R_3 = OH$
46: $R_1 = OMe; R_2 = COCH=CHC_6H_5; R_3 = OH$

Table 2 shows the 1H -NMR data of some alkaloids. The remarkable feature is the typical aspidospermine signals of hydrogen linked to CH-2, whose chemical shift remains at δ_H 4.5. The multiplicity of this signal, a double-doublet, reveals the spin coupling of H-2 with the two hydrogen atoms at CH₂-16 unsubstituted. This pattern is present in almost all alkaloids, except for those substituted in C-16 vincadifformine (**1**, with double bond between C-2 and C-16) [30], spgazzinine (**3**) and spgazzinidine (**4**) [31], and also 1,2-dehydroaspidospermidine (**5**) and 1,2-dehydro-*N*-deacetyl-aspidospermin (**6**), although the NMR data for the latter is not available.

Table 2. ^1H -NMR data (in CDCl_3) for the plumeran indole alkaloids from *Aspidosperma* species. (Multiplicities, J , in parenthesis).

Hydrogens	Compound/ δ_{H} (ppm), J_{H} (Hz)					
	1	3	4	5	7	
2	-	4.05 (<i>d</i> , 8.0)	4.05 (<i>d</i> , 8.0)	-	4.60 (1H, <i>s</i>)	
3				3.18 (1H, <i>m</i>)	3.02 (1H, <i>d</i> , 11.0)	
				2.48 (1H, <i>dd</i> , 13.2; 3.3)	1.96 (1H, <i>m</i>)	
5				3.20 (1H, <i>m</i>)	3.10 (1H, <i>dt</i> , 8.0)	
				2.80 (<i>ddd</i> , 10.2; 3.3)	2.20 (1H, <i>m</i>)	
6				2.18 (1H, <i>m</i>)	2.00 (1H, <i>m</i>)	
				1.56 (1H, <i>m</i>)	1.56 (1H, <i>m</i>)	
9	6.74–7.5 (4H, <i>m</i>)	6.57 (2H, <i>m</i>)	≈ 7.0 (3H, <i>m</i>)	7.35 (1H, <i>d</i> , 8.0)	6.83 (1H, <i>d</i> , 8.6)	
10	6.74–7.5 (4H, <i>m</i>)	6.57 (2H, <i>m</i>)	≈ 7.0 (3H, <i>m</i>)	7.25–7.30 (1H, <i>m</i>)	6.64 (1H, <i>d</i>)	
11	6.74–7.5 (4H, <i>m</i>)		≈ 7.0 (3H, <i>m</i>)	7.25–7.30 (1H, <i>m</i>)		
12	6.74–7.5 (4H, <i>m</i>)			7.53 (1H, <i>d</i> , 8.0)		
14				1.86 (2H, <i>dq</i> , 12.9)	1.73 (1H, 4.0)	
				1.52 (1H, <i>m</i>)	1.51 (1H, <i>m</i>)	
15				1.58 (1H, <i>m</i>)	1.64 (1H, <i>m</i>)	
				1.0 (1H, <i>dt</i> , 13.5; 2.7)	1.08 (1H, <i>m</i>)	
16				2.60 (1H, <i>m</i>)	1.54 (1H, <i>m</i>)	
				3.1 (1H, <i>ddd</i>)	1.80 (1H, <i>m</i>)	
17				1.28 (1H, <i>m</i>)	1.08 (1H, <i>m</i>)	
				1.47 (1H, <i>m</i>)	2.02 (1H, <i>m</i>)	
18		0.75 (3H, <i>t</i>)	0.75 (3H, <i>t</i>)	0.50 (3H, <i>t</i> , 6.9)	0.63 (3H, <i>t</i> , 7.3)	
19				0.65 (2H, <i>q</i> , 6.9)	0.81 (1H, <i>q</i> , 6.9)	
					1.25 (1H, <i>m</i>)	
21	8.96 (1H, <i>br s</i>)			2.42 (1H, <i>s</i>)	2.18 (1H, <i>s</i>)	
11-OCH ₃					3.98 (3H, <i>s</i>)	
12-OCH ₃					3.78 (3H, <i>s</i>)	
15-OCH ₃						
COCH ₃		2.48 (3H, <i>s</i>)	2.48 (3H, <i>s</i>)		2.15 (3H, <i>s</i>)	
COOCH ₃	3.76 (3H, <i>s</i>)					
OH (C11)		-	5.84 (1H, <i>m</i>)			
OH (C12)		11.1 (1H, <i>s</i>)	11.1 (1H, <i>s</i>)			
OH (C16)		7.3 (1H, <i>s</i>)	7.3 (1H, <i>s</i>)			
	1.6–3.6 (18H, <i>complex m</i>)					

Hydrogens	Compound/ δ_{H} (ppm), J_{H} (Hz)						
	8	9	11	12	14	16	18
2	4,5 (1H, <i>m</i>)	4.70 (1H, <i>q</i>)	3.48 (<i>q</i> , 6; 10)	4.07 (<i>q</i> , 6; 10)	4.08 (1H, <i>q</i> , 6, 10)	3.6 (<i>dd</i> , 6.2, 11)	4.5 (<i>m</i>)
3	3.0 (<i>brd</i> , 10.7)	3.28 (1H, <i>m</i>)				3.0 (<i>dt</i> , 13.1, 4.1)	3.0 (<i>brd</i> , 10.7)
	1.9 (<i>m</i>)	2.22 (1H)				1.95 (<i>dd</i> , 2.9, 10.7)	2.05–1.9 (<i>m</i>)
5	3.11 (<i>td</i> , 9, 3.2)	3.48 (1H, <i>m</i>)				3.1 (<i>td</i> , 7.6, 2.8)	3.1 (<i>td</i> , 9, 3.2)
	2.25 (<i>m</i>)	2.40 (<i>m</i> , 13.8)				2.3 (<i>m</i>)	2.25 (<i>m</i>)

Table 2. Cont.

Hydrogens	Compound/ δ_H (ppm), J_H (Hz)						
	8	9	11	12	14	16	18
6	2.05 (<i>m</i>) 1.55 (<i>m</i>)	2.20 (1H, <i>m</i>) 1.80 (1H, <i>m</i>)				2.4 (<i>m</i>) 1.5 (<i>m</i>)	2.05–1.9 (<i>m</i>) 1.6–1.35 (<i>m</i>)
9	6.83 (<i>d</i> , 8.7)	6.82 (2H, 8.0)	6.56 (3H, <i>m</i>)	6.9 (3H, <i>m</i>)	7.17 (3H, <i>m</i>)	6.75 (<i>d</i> , 1.7)	6.83 (<i>d</i> , 8.7)
10	7.07 (<i>t</i> , 8.7)	7.08 (1H, <i>d</i> , 8.0)	6.56 (3H, <i>m</i>)	6.9 (3H, <i>m</i>)	7.17 (3H, <i>m</i>)		7.07 (<i>t</i> , 8.7)
11	6.8 (<i>d</i> , 8.7)	6.82 (2H, 8.0)	6.56 (3H, <i>m</i>)	6.9 (3H, <i>m</i>)	7.17 (3H, <i>m</i>)	6.6 (<i>dd</i> , 1.7, 7)	6.8 (<i>d</i> , 8.7)
12					8.13 (1H,)	6.7 (<i>d</i> , 7)	
14	1.73 (<i>qt</i> , 13.4) 1.5 (<i>brd</i> , 13)	2.0 (1H) 1.60 (1H, <i>m</i>)				1.8 (<i>qt</i> , 13.1, 4.0) 1.5 (<i>m</i>)	1.7 (<i>qt</i> , 13, 4) 1.6–1.35 (<i>m</i>)
15	1.6 (<i>m</i>) 1.05 (<i>m</i>)	3.22 (<i>m</i> , 14.5, 7.0)				1.6 (<i>m</i>) 1.2 (<i>dd</i> , 4.5, 13.4)	1.6–1.35 (<i>m</i>) 1.3–1.05 (<i>m</i>)
16	1.95 (<i>m</i>) 1.35 (<i>m</i>)	1.30 (1H, <i>m</i>) 2.04 (1H, <i>m</i>)				1.6 (<i>m</i>) 1.4 (<i>dt</i> , 14.0, 3.4)	2.05–1.9 (<i>m</i>) 1.6–1.35 (<i>m</i>)
17	2.0 (<i>m</i>) 1.1 (<i>m</i>)	1.10 (1H, <i>dq</i> , 14.7) 2.18 (1H, <i>m</i>)				2.0 (<i>dd</i> , 2.6, 12.3) 1.1 (<i>dt</i> , 12.6, 2.5)	2.05–1.9 (<i>m</i>) 1.3–1.05 (<i>m</i>)
18	0.6 (<i>t</i> , 7.1)	0.63 (3H, <i>t</i> , 6.9)	0.6 (3H, <i>t</i> , 6)	0.72 (3H, <i>t</i>)	0.73 (3H, <i>t</i>)	0.7 (<i>t</i> , 7.3)	0.6 (<i>t</i> , 7)
19	1.2 (<i>dq</i> , 14, 7.1) 0.8 (<i>dq</i> , 14, 7.1)	0.75 (1H, <i>q</i> , 6.9) 1.22 (1H, <i>m</i>)				1.5 (<i>m</i>) 0.9 (<i>dq</i> , 14.3, 7.3)	1.3–1.05 (<i>m</i>) 0.8 (<i>dq</i> , 14, 7)
21	2.23 (1H, <i>s</i>)	2.20 (1H, <i>s</i>)	2.16 (1H, <i>s</i>)	2.28 (1H, <i>s</i>)		2.2 (1H, <i>s</i>)	2.2 (<i>s</i>)
10-OCH ₃						3.8 (3H, <i>s</i>)	
12-OCH ₃	3.87 (3H, <i>s</i>)	3.90 (3H, <i>s</i>)	3.78 (3H, <i>s</i>)				3.87 (<i>s</i>)
15-OCH ₃		3.33 (3H, <i>s</i>)					
COCH ₃	2.2 (3H, <i>s</i>)	2.22 (3H, <i>s</i>)	-	2.32 (3H, <i>s</i>)	2.27 (3H, <i>s</i>)		
COCH ₂ CH ₃							2.6 (<i>dq</i> , 13, 6.5) 2.4 (<i>m</i>)
COCH ₂ CH ₂ OH				10.83 (<i>s</i>)			1.3 (<i>t</i> , 6.5)
NH			3.35 (1H, <i>s</i>)				

Hydrogens	Compound/ δ_H (ppm), J_H (Hz)						
	19	20	21	23	24	27	28
2	4.07 (<i>q</i> , 6, 10)	4.07 (<i>dd</i> , 11, 6)	4.0 (1H, <i>q</i> , 5)	4.71 (1H, <i>q</i>)	4.12 (1H, <i>q</i>)	3.4 (<i>dd</i> , 5.5, 9)	4.12 (1H, <i>q</i>)

Table 2. Cont.

Hydrogens	Compound/ δ_H (ppm), J_H (Hz)						
	19	20	21	23	24	27	28
3		1.98 (<i>td</i> , 12, 4) 3.04 (<i>dm</i> , 12)		3.26 (1H, <i>m</i>) 2.20 (1H)		3.1–3.05 (<i>m</i>) 1.8–2.0 (<i>m</i>)	
5		2.27 (<i>m</i>), 3.12 (<i>m</i>)		3.48 (1H, <i>m</i>) 2.40 (<i>m</i> , 13.8)		3.1–3.05 (<i>m</i>) 2.3 (<i>m</i>)	
6		1.57 (<i>m</i>), 2.04 (<i>m</i>)		2.20 (1H, <i>m</i>) 1.80 (1H, <i>m</i>)		2.3 (<i>m</i>) 1.4–1.55 (<i>m</i>)	
9	6.78 (<i>m</i> , 8)	6.61 (<i>d</i> , 8)	6.36 (<i>s</i>)	6.65 (1H, <i>d</i> , 8.0)	6.65 (2H, <i>q</i> , 8)	7.0 (<i>d</i> , 7.5)	6.92 (3H, <i>m</i>)
10	6.77 (<i>m</i> , 7)	6.69 (<i>d</i> , 8)	6.36 (<i>s</i>)	6.82 (1H, <i>d</i> , 8.0)	6.65 (2H, <i>q</i> , 8)	6.6 (<i>dt</i> , 1, 7.5)	6.92 (3H, <i>m</i>)
11	7.02 (<i>m</i> , 1.5)					7.05 (<i>dt</i> , 1, 7.5)	6.92 (3H, <i>m</i>)
12						6.35 (<i>d</i> , 7.5)	
14		1.72 (<i>tm</i> , 12) 1.53 (<i>dm</i> , 12)		2.10 (1H) 1.52 (1H, <i>m</i>)		1.75 (<i>m</i>) 1.4–1.55 (<i>m</i>)	
15		1.11 (<i>td</i> , 12, 4) 1.65 (<i>dt</i> , 12, 4)		3.22 (<i>m</i> , 14, 7)		1.65 (<i>m</i>) 1.1–1.2 (<i>m</i>)	
16		1.86 (<i>m</i>) 1.52 (<i>m</i>)		1.25 (1H, <i>m</i>) 2.0 (1H, <i>m</i>)		1.75 (<i>m</i>) 1.25 (<i>m</i>)	
17		2.00 (<i>td</i> , 12, 14) 1.15 (<i>dm</i> , 12)		1.00 (<i>dq</i> , 14.7) 2.15 (1H, <i>m</i>)		1.8–2.0 (<i>m</i>) 1.1–1.2 (<i>m</i>)	
18	0.59 (<i>t</i> , 14)	0.63 (<i>t</i> , 7.5)	0.7 (<i>t</i> , 4)	0.63 (3H, <i>t</i>)	0.62 (3H, <i>t</i> , 6)	0.6 (<i>t</i> , 7.5)	3.52 (<i>t</i> , 2H, 7)
19		0.93 (<i>m</i>), 1.44 (<i>m</i>)		0.76 (1H, <i>q</i> , 7.2) 1.30 (1H, <i>m</i>)		0.85 (<i>dq</i> , 14.5, 7.5) 1.4–1.55 (<i>m</i>)	
21		2.25 (<i>s</i>)		2.28 (1H, <i>s</i>)		2.2 (<i>s</i>)	
11-OCH ₃		3.88 (<i>s</i>)		3.90 (6H, <i>s</i>)	3.88 (3H, <i>s</i>)		
12-OCH ₃				3.90 (6H, <i>s</i>)			
15-OCH ₃				3.22 (3H, <i>s</i>)			
COCH ₃		2.33 (<i>s</i>)	2.25 (<i>s</i>)	2.25 (3H, <i>s</i>)			
COCH ₂ CH ₃	2.53 (<i>q</i> , 14)				2.57 (2H, <i>q</i> , 7) 1.25 (3H, <i>t</i> , 7)		2.57 (2H, <i>q</i> , 7.5)
COCH ₂ CH ₃	1.24 (<i>t</i> , 14.2)						
Ph-OH	10.86 (<i>s</i>)	10.98 (<i>s</i>)	10.85 (1H, <i>s</i>)		10.98 (1H, <i>s</i>)		10.88 (1H, <i>s</i>)
N-CH ₃						2.75 (<i>s</i>)	
	0.3–3.2 (17 hydrogens)						

Table 2. Cont.

Hydrogens	Compound/ δ_H (ppm), J_H (Hz)							
	29	31	32	34	35 **	36 **	37	38 **
2	4.1 (<i>q</i>)	4.1 (1H, <i>q</i>)	4.07 (<i>q</i>)	*	5.5 (1H, <i>bq</i> , 5)	6.7–6.95 (<i>m</i>)	3.60 (1H, <i>m</i>)	5.4 (1H, <i>m</i>)
3								
5					6.8–7.1 (2H, <i>m</i>)	6.7–6.95 (<i>m</i>)		6.5–7.0 (2H, <i>m</i>)
6								
9	6.5–6.9 (2H, <i>q</i> , 8)	6.58–7.25 (3H, <i>m</i>)	6.5–6.9 (2H, <i>q</i> , 8)	6.6–7.1 (3H, <i>m</i>)	7.75 (3H, <i>m</i>)	6.8–6.61 (3H, <i>m</i>)	6.55–6.93 (3H, <i>m</i>)	7.57 (3H, <i>m</i>)
10	6.5–6.9 (2H, <i>q</i> , 8)	6.58–7.25 (3H, <i>m</i>)	6.5–6.9 (2H, <i>q</i> , 8)	6.6–7.1 (3H, <i>m</i>)	7.75 (3H, <i>m</i>)	6.8–6.61 (3H, <i>m</i>)	6.55–6.93 (3H, <i>m</i>)	7.57 (3H, <i>m</i>)
11		6.58–7.25 (3H, <i>m</i>)		6.6–7.1 (3H, <i>m</i>)	7.75 (3H, <i>m</i>)	6.8–6.61 (3H, <i>m</i>)	6.55–6.93 (3H, <i>m</i>)	7.57 (3H, <i>m</i>)
12								
14								
15								
16								
17								
18	3.55 (2H, <i>t</i> , 7)	3.53 (2H, <i>t</i> , 7)	3.53 (2H, <i>t</i> , 7)					
19					6.47 (2H, <i>bt</i> , 7)		4.31 (1H, <i>d</i> , 6)	
21	2.5 (<i>s</i>)	*	*		2.85–3.3 (1H, <i>s</i>)	3.2–3.4	2.93 (1H, <i>s</i>)	2.8–3.2 (1H, <i>s</i>)
11-OCH ₃	3.87 (<i>s</i>)		3.87 (<i>s</i>)					
12-OCH ₃				2.30 (3H, <i>s</i>)	6.15 (3H, <i>s</i>)	6.18 (3H, <i>s</i>)	3.79 (3H, <i>s</i>)	6.13 (3H, <i>s</i>)
18-OCH ₃							3.88 (3H, <i>s</i>)	
N-COCH ₃		2.32 (<i>s</i>)	2.32 (<i>s</i>)		7.84 (3H, <i>s</i>)			7.80 (3H, <i>s</i>)
N-COCH ₂ CH ₃	2.57 (<i>q</i> , 7.5)							
N-COCH ₂ CH ₃	1.27 (<i>t</i> , 7.5)							
N-COOCH ₃						6.44 (3H, <i>s</i>)		6.44 (3H, <i>s</i>)
N-CHO				9.3 (1H, <i>s</i>)				
Ph-OH	10.95 (1H, <i>s</i>)	10.87 (1H, <i>s</i>)	10.95 (1H, <i>s</i>)					
C-19-OH							2.74 (1H, <i>d</i> , 6)	
					7.6–9.2 (15H, <i>m</i>)	7.4–8.9 (14H, <i>m</i>)		7.5–9.1 (14H, <i>m</i>)

Table 2. Cont.

Hydrogens	Compound/ δ_H (ppm), J_H (Hz)							
	39	40	41	42	43	44	45	46
2	4.50 (1H, <i>m</i>)	3.9–4.1 (1H, <i>q</i>)	4.54 (1H, <i>dd</i> , 10.5, 6.1)	4.24–4.44 (1H, <i>q</i>)	4.04 (1H, <i>dd</i> , 10.8, 8)	4.35–4.6 (1H, <i>q</i>)	*	4.50–4.70 (1H, <i>q</i>)
3								
5			3.07 (2H, <i>m</i>)		3.3–2.95 (2H, <i>m</i>)			
6								
9		6.56–6.75 (3H, <i>m</i>)	7.1–6.72 (3H, <i>m</i>)	6.6–7.6 (8H, <i>m</i>)	7.3–6.9 (3H, <i>m</i>)	6.7–7.2 (8H, <i>m</i>)	6.6–7.3 (3H, <i>m</i>)	6.8–7.6 (8H, <i>m</i>)
10		6.56–6.75 (3H, <i>m</i>)	7.1–6.72 (3H, <i>m</i>)	6.6–7.6 (8H, <i>m</i>)	7.3–6.9 (3H, <i>m</i>)	6.7–7.2 (8H, <i>m</i>)	6.6–7.3 (3H, <i>m</i>)	6.8–7.6 (8H, <i>m</i>)
11		6.56–6.75 (3H, <i>m</i>)	7.1–6.72 (3H, <i>m</i>)	6.6–7.6 (8H, <i>m</i>)	7.3–6.9 (3H, <i>m</i>)	6.7–7.2 (8H, <i>m</i>)	6.6–7.3 (3H, <i>m</i>)	6.8–7.6 (8H, <i>m</i>)
12					8.12 (1H, <i>m</i>)			
14								
15								
16								
17								
18								
19						3.98 (1H, <i>s</i>)	4.19 (1H, <i>s</i>)	4.06 (1H, <i>s</i>)
21	2.48 (1H, <i>s</i>)		2.46 (1H, <i>s</i>)	2.50 (1H, <i>s</i>)	2.53 (1H, <i>s</i>)		2.99 (1H, <i>s</i>)	3.00 (1H, <i>s</i>)
12-OCH ₃	3.89 (3H, <i>s</i>)	3.53 (3H, <i>s</i>)	3.56 (3H, <i>s</i>)	3.37 (3H, <i>s</i>)	2.26 (3H, <i>s</i>)	3.80 (3H, <i>s</i>)	3.86 (3H, <i>s</i>)	3.79 (3H, <i>s</i>)
COOCH ₃	3.89 (3H, <i>s</i>)	3.75 (3H, <i>s</i>)	3.88 (3H, <i>s</i>)	3.56 (3H, <i>s</i>)	3.57 (3H, <i>s</i>)	3.86 (3H, <i>s</i>)	3.89 (3H, <i>s</i>)	3.89 (3H, <i>s</i>)
C-19-OH								
N-Me		3.04 (3H, <i>s</i>)						
N-CHO							9.30 (1H, <i>s</i>)	
CH=CHPh	7.85 (1H, <i>d</i>)							7.70 (1H, <i>d</i> , 16)
CH=CHPh	6.88 (1H, <i>d</i>)							6.74 (1H, <i>d</i> , 16)
			2.4–1.2 (15H, <i>m</i>)		2.5–1.2 (14H, <i>m</i>)			

* Data not provided; ** The data of the chemical shifts amongst the CH-21 and the aromatic hydrogens were originally exchanged, and were corrected.

Table 3. ^{13}C -NMR data (in CDCl_3) for the plumeran indole alkaloids from *Aspidosperma* species.

Carbons	Compound/ δ_{C} (ppm)													
	1	5	7	8	9	16	18	20	23	27	36	38	41	43
2	167.8	193.0	69.6	64.0	66.0	66.0	69.4	70.3	69.6	71.6	65.4	68.7	63.6	67.6
3	51.7	51.8	53.5	53.5	52.8	53.7	53.6	53.7	52.9	53.7	53.6	53.2	52.8	53.0
5	50.7	54.4	52.6	52.4	52.8	53.0	52.5	52.4	52.8	52.9	52.7	52.0	52.1	52.2
6	44.3	34.9	38.2	38.0	37.1	38.4	37.9	39.4	38.4	38.9	37.9	37.4	39.4	39.3
7	55.0	58.1	52.6	52.4	53.7	54.1	52.5	52.2	52.6	52.4	54.3	53.2	53.4	53.5
8	138.0	147.2	143.4	128.0	143.3	136.6	125.9	133.1	143.4	137.0	138.9	142.0	140.3	137.4
9	121.0	121.7	117.7	115.4	115.3	115.3	115.4	112.4	117.7	122.0	115.4	115.2	115.9	124.3
10	120.5	125.3	108.8	125.9	126.2	146.0	125.9	110.0	108.8	117.0	197.8	126.3	124.8	122.3
11	127.4	127.6	152.7	110.0	111.8	108.8	111.2	149.4	152.7	127.2	109.6	115.5	111.0	127.9
12	109.3	120.2	152.7	148.0	149.5	119.5	*	137.5	152.7	106.4	146.2	149.0	148.6	118.6
13	143.4	154.5		141.0	130.1	138.3	*	127.5	129.5	150.5	135.5	129.4	127.8	141.1
14	22.2	21.7	21.3	21.5	24.0	21.8	21.6	21.5	24.4	21.9	21.8	21.3	21.6	21.6
15	32.9	32.9	34.6	34.1	75.5	34.5	34.2	34.0	75.5	34.4	35.3	42.3	35.1	34.8
16	92.8	27.0	24.6	24.7	24.5	28.2	24.4	25.1	25.0	21.6	28.4	24.8	24.8	29.7
17	25.6	23.4	22.8	23.0	22.5	23.2	23.1	22.9	24.4	28.8	24.3	34.6	24.3	24.6
18	7.3	6.9	5.6	6.7	6.57	6.8	6.6	6.8	6.90	6.7	172.3	175.7	172.0	171.8
19	29.3	29.5	29.8	29.9	29.9	29.9	30.1	30.0	30.0	30.0	42.5	45.0	42.4	42.4
20	38.2	36.2	35.3	35.4	35.6	35.6	35.5	35.5	35.7	35.5	36.2	35.8	36.1	36.0
21	72.7	69.0	71.1	71.0	71.6	71.3	71.1	70.6	71.5	71.1	70.1	69.5	69.4	69.9
N-COCH ₃				160.0	171.2		161.4	169.3	171.2			172.0		168.3
N-COCH ₃				22.9	23.0			22.7	23.1			23.3		23.2
11-OMe			56.0					56.4	56.2					
12-OMe			56.0	55.3	53.5	55.2	55.6		56.2		55.4	55.4	55.6	
15-OMe					56.3				56.4					
COCH ₂ CH ₃							28.1							
COCH ₂ CH ₃							10.1							
N-CH ₃										31.4				
N-CHO													161.4	
COOCH ₃	50.9										50.9	51.1	51.0	51.0
COOCH ₃	169.2													

* Data not provided.

2. Discussion

The structure of the alkaloid aspidospermine (**8**) has the basic ring skeleton typical of a large number of natural alkaloids. In addition to the characteristic signals for the aromatic methoxyl function, the ^1H -NMR spectrum shows a very common pattern amongst the indole alkaloids the *N*-acetyl group, the C-20 ethyl side chain, and the lone hydrogen atom attached to CH-21, which is not split by any neighboring hydrogen [56].

Thus, the ^1H and ^{13}C -NMR spectra of the alkaloids with the plumeran skeleton present key characteristic signals of the typical aspidospermine pattern. The hydrogen linked at CH-2 shows a signal with a double-doublet multiplicity (due the low sensibility of the 1960's NMR spectrometers, it was originally characterized as a quartet). This feature indicates that CH-2 hydrogen couples its spin

with the two hydrogen atoms at C-16 [56]. The values for chemical shifts of the H-2 vary towards δ_H 4.5 in the 1H -NMR and the CH-2 appears as δ_C 66 in the ^{13}C -NMR spectra.

For the main consulting reference molecule aspidospermine (**8**), the data relative to the hydrogen attached to CH-2 was missing [34] and did not appear on the spectra. Its presence was deduced by the signal of a methine carbon at δ_C 64 in the ^{13}C spectrum. This information was then supplied by another source [35], to the best of our knowledge.

Note that for the alkaloids that present a substituent in C-16, there is a striking difference. Vincadifformine (**1**) is presented here as a precursor of the plumeran skeleton, by the loss of the carbon linked to C16, by a hydrolysis/decarboxylation reaction [21]. For this molecule, the presence of a double bond between C-2 and C-16 justify the absence of the signal of H-2 in the 1H -NMR. In the ^{13}C -NMR, the chemical shift for C-2 appears at δ_C 167.8, what may be explained by the presence of the nitrogenous atom and by its conjugation with the carbonyl group, which is also the reason for the chemical shifts of the of the carbomethoxy at δ_C 169 and the carbon atom C-16 at δ_C 92.8, which is very characteristic of vincadifformine [30].

Spegazzinine (**3**) and spegazzinidine (**4**) are also substituted in C-16. The signal of H-2 in the 1H -NMR spectra appears as a doublet, due to the coupling of the hydrogen attached to CH-2 with the only hydrogen atom in CH-16. The assignments in the 1H -NMR spectrum for spegazzinine showed signals for three aromatic hydrogen atoms, an *N*-acetyl function and a C-ethyl group. The consulted bibliography did not provide detailed information regarding the orientation of the aromatic hydrogen atoms [31]. However, the nature of these hydrogens is better known today due to the modern techniques available, and it is now known that there are two doublets attributed to H-9 and H-11, thus revealing *ortho* interactions and one triplet corresponding to H-10. The 1H -NMR spectrum of spegazzinidine demonstrated a significant difference only in the region corresponding to aromatic hydrogens; the two aromatic hydrogens have an *ortho* interaction.

The relative configuration of the additional asymmetric center, C-16, in spegazzinine and spegazzinidine was defined because of the constant coupling ($J = 8.0$ Hz), a value typical of axial-axial hydrogens at CH-2 and CH-16 [31].

For 1,2-dehydroaspidospermidine (**5**), the presence of a non-substituted carbon at δ_C 193.0, assigned as C-2, linked to the indolic nitrogenous atom by a double bond, characterizes it as an indolenine system. As is the case of vincadifformine (**1**), the presence of a double bond between C-2 and N justify the absence of the signal of H-2 in the 1H -NMR. The aromatic hydrogens have the expected couplings for a non-substituted indolenine system: a doublet attributed to H-9 with chemical shift of δ_H 7.53, two multiplets attributed to H-10 and H-11 (each with a chemical shifts of δ_H 7.25), and a triplet attributed to C-12 with δ_H 6.17.

Another important feature is the displacement of the chemical shift of the H-12 at non-substituted aromatic ring, as was evident for the demethoxyaspidospermidine (**14**) and 12-demethoxy-*N*-acetylcylindrocarine (**43**). The displacement occurs due to the attenuation of the mesomeric effect of the nitrogenous atom by the presence of an acyl group. This, in addition to the anisotropic effect of the carbonyl group at CH-12 results in the deshielding of the hydrogens at CH-10 and CH-12.

The compounds **9** and **23**, isolated and characterized by Oliveira [32] as new alkaloids, were not reported in the guideline literature [2]. For these two alkaloids, the presence of the methoxy group at

CH-15 is deduced by the presence of chemical shifts at δ_H 3.22 (MeO-15) and at δ_C 75.5. This is a new substitution pattern observed among the *Aspidosperma* alkaloids.

Hitherto, all the structures have the same ethyl feature at the C-20 side chain. Due to the optical rotation $[\alpha]_D$, one- (1H -NMR, ^{13}C -NMR- $\{^1H\}$ and ^{13}C -NMR-DEPT or APT) and two-dimensional (homonuclear 1H - 1H -COSY, and 1H - 1H -NOESY; heteronuclear HMQC or HSQC- $^1J_{HC}$ and HMBC- $^nJ_{HC}$, $n = 2$ and 3) NMR experiments, the stereochemistry of this group and the absolute configuration of each of these molecules was well established.

Nevertheless, limaspermine (**28**) and related alkaloids **29** to **35** reveal the oxidative biotransformation of the methyl group present in the carbon atom C-20-ethyl substituent (C-20-CH₂CH₃) to yield C-20-CH₂CH₂OH, presenting an expressive difference at the lateral chain at C-20 [23,53]. The presence of these moieties in such structures may be recognized by a comparative analysis of the 1H -NMR spectra: -CH₂CH₃ deduced by triplet signal at δ_H 0.6 is attributed to the hydrogens of the methyl group (3H-18) and -CH₂CH₂OH is characterized by a triplet signal at δ_H 3.5 corresponding to the carbinolic hydrogens of the methylene CH₂OH (2H-18). The ^{13}C -NMR data for these compounds are not available, so the expected differences of the C-18 and the C-19 in the ^{13}C -NMR spectrum can only be predicted, by considering the displacement that occurs to deshielded fields.

In the alkaloids **36** to **46** (Figure 5) the carbon atom C-20 sustains a -CH₂COOMe containing a carbomethoxy group (COOMe), which may be explained by the additional oxidative biotransformation of CH₂CH₂OH to CH₂COOH, which is then followed by a methylation reaction. The presence of the C-20-CH₂COOMe can be recognized by 1H and ^{13}C -NMR spectra as the anticipated modifications of the 1H and ^{13}C chemical shifts. The singlet signal corresponding to the hydrogen of the methine carbon CH-21 demonstrates the influence induced by the proximity of the carbonyl group at C-20. This contributes to the signal displacement of the CH-21 hydrogen to δ_H 2.4 (s), which is different from the observed value of δ_H 2.23, found for aspidospermine [55]. The most impressive result involves the modifications observed by carbon atoms C-18 and CH₂-19 in the ^{13}C -NMR spectra: δ_C 171.0 and δ_C 42.0, respectively.

Data analysis suggests that for the alkaloids **35**, **36** and **38**, the values attributed to aromatics and the CH-21 hydrogens were exchanged. The chemical shifts for the aromatics remain towards δ_H 6.5–7.0 and for the hydrogen attached at CH-21 remains towards δ_H 2.2, in accordance with all the other compounds for the same series (**36** to **46**).

In regards to the aromatic hydrogens, it is important to notice that for the structures **42**, **44**, and **46**, the aromatic signals are concerning 8 hydrogens—3 being part of the monosubstituted A ring and the other 5 of a benzyl substituent.

The expected differences associated with the *N*-acyl substituent also appear in the cylindrocarpidine (**39**) and its cinnamoyl derivative *N*-cinnamoyl-19-hydroxycylindrocarine (**46**). They exhibit the absence of the acetyl group and the appearance of lines relative to cinnamoyl system [55]. The coupling constant value ($J = 16$ Hz) observed in the olefinic signals of the 1H -NMR spectra was used to confirm the *trans* configuration of the double bond present in the cinnamoyl moiety [50].

3. Conclusions

The initial purpose of this work was to emphasize a review of the ^1H and ^{13}C -NMR spectral data for a small group of alkaloids in a discussion that would highlight the spectral differences amongst them. However, we observed that most of the literature data were reported in the 1960's and the NMR data were incomplete or even unavailable. At that time, the structures of some alkaloids were elucidated on the basis on IR (Infrared) and/or UV (Ultraviolet) and/or MS (Mass Spectrometry) techniques, involving also chemical transformations, before the development of the NMR. Even in the beginning, the NMR spectra presented low sensitivity due to the limited equipment accuracy (frequently about 60 MHz).

Through the modern comparative analysis of ^{13}C -NMR- $\{^1\text{H}\}$ - and DEPT- or APT- ^{13}C -NMR spectra, it becomes easier to differentiate, classify, and count the signals corresponding to quaternary, methane, methylene and methyl carbon atoms. The DEPTQ (Q = quaternary), appears as a new variation of DEPT for the inclusion of signals corresponding to quaternary carbon atoms. The basic skeleton of the plumeran alkaloids (non-substituted molecules) reveals the presence of four non-hydrogenated (two sp^2 and two sp^3), six methine (four sp^2 aromatics and two sp^3), eight methylene (all sp^3) and one methyl carbon atoms, which may be recognized through this comparative analysis. Obviously, the modification of the numbers of CH, CH_2 and CH_3 by presence and type of substituent may be characterized with relative facility. This analysis in collaboration with high resolution mass spectrometry significantly contributes to the deduction of a molecular formula. And so, it is possible to classify an unknown alkaloidic molecule in terms of carbon patterns, which would to facilitate the elucidation of its molecular form. Also, the 2D NMR experiments (^1H - ^1H -COSY, ^1H - ^1H -NOESY, HMQC or HSQC and HMBC) are especially helpful to solve structural problems, allowing especially assignments of methylene groups that frequently appear in region revealing superimpose [34].

So, this work proposes that not only the chemical shifts for the CH-2 hydrogen (absent in the presence of a double bond between this carbon and the N - $1\delta_{\text{C}}$ 193.0 for C-2 as revealed by alkaloid **5**), but also the assignments for the CH-21 and the ethyl group (and its biosynthetic derivatives) sustained by C-20, are the main spectrometric features that characterize the presence of the plumeran alkaloidic skeleton.

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References and Notes

1. Endress, M.E.; Bruyns, P.V. A revised classification of the Apocynaceae s.l. *Bot. Rev.* **2000**, *66*, 1–56.
2. Pereira, M.M.; Jácome, R.L.R.P.; Alcântara, A.F.C.; Alves, R.B.; Raslan, D.S. Alcalóides Indólicos Isolados de Espécies do Gênero *Aspidosperma* (APOCYNACEAE). *Quim. Nova* **2007**, *30*, 970–983. (in Portuguese).

3. CEAP Design. Apocynaceae. Available online: http://www.ceapdesign.com.br/familias_botanicas/apocynaceae.html (accessed on 20 January 2010).
4. UFRRJ—Universidade Federal Rural do Rio de Janeiro. Xiloteca. Available online: <http://www.if.ufrj.br/npqm/xiloteca/index.php> (accessed on 20 January 2010).
5. Bolzani, V.S.; Serur, L.M.; Matos, F.J.A.; Gottlieb, O.R. Indole alkaloids evolution in *Aspidosperma*. *Biochem. Syst. Ecol.* **1987**, *15*, 187–200.
6. Garcia, M.; Ruben, F.; Brown, K.S. Alkaloids of three *Aspidosperma* species. *Phytochemistry* **1976**, *15*, 1093–1095.
7. Oliveira, V.B.; Freitas, M.S.M.; Mathias, L.; Braz-Filho, R.; Vieira, I.J.C. Atividade Biológica e Alcalóides Indólicos de Gênero *Aspidosperma* (Apocynaceae): uma revisão. *Rev. Bras. Pl. Med.* **2009**, *11*, 92–99. (in Portuguese).
8. Bourdy, G.; Oporto, P.; Gimenez, A.; Deharo, E. A search for bioactive compounds in Bolivia through a multidisciplinary approach. Part VI. Evaluation of the antimalarial activity of plants used by Isoceño-Guaraní Indians. *J. Ethnopharmacol.* **2004**, *93*, 269–277.
9. Ferreira, I.C.P.; Lonardoni, M.V.C.; Machado, G.M.C.; Leon, L.L.; Gobbi Filho, L.; Pinto, L.H.B.; Oliveira, A.J.B. Anti-leishmanial activity of alkaloidal extract from *Aspidosperma ramiflorum*. *Mem. Inst. Oswaldo Cruz* **2004**, *99*, 325–327.
10. Mitaine-Offer, A.C.; Sauvain, M.; Valentin, A.; Callapa, J.; Mallié, M.; Zèches-Hanrot, M. Antiplasmodial activity of *Aspidosperma* indole alkaloids. *Phytochemistry* **2002**, *9*, 1442–145.
11. Mesquita, M.L.; Grellier, P.; Mambu, L.; Paula, J.E.; Espindola, L.S. *In vitro* antiplasmodial activity of Brazilian Cerrado plants used as traditional remedies. *J. Ethnopharmacol.* **2007**, *110*, 165–170.
12. Weniger, B.; Robledo, S.; Arango, G.J.; Deharo, E.; Aragón, R.; Muñoz, V.; Callapa, J.; Lobstein, A.; Anton, R. Antiprotozoal activities of Colombian plants. *J. Ethnopharmacol.* **2001**, *78*, 193–200.
13. Bourdy, G.; Oporto, P.; Gimenez, A.; Deharo, E. A search for bioactive compounds in Bolivia through a multidisciplinary approach. Part VI. Evaluation of the antimalarial activity of plants used by Isoceño-Guaraní Indians. *J. Ethnopharmacol.* **2004**, *93*, 269–277.
14. Campos, A.R.; Lima, R.C.P., Jr.; Uchoa, D.E.A.; Silveira, E.R.; Santos, F.A.; Rao, V.S.N. Pro-erectile effects of an alkaloid rich fraction from *Aspidosperma ulei* root bark in mice. *J. Ethnopharmacol.* **2006**, *104*, 240–244.
15. Campos, A.R.; Cunha, K.M.A.; Santos, F.A.; Silveira, E.R.; Uchoa, D.E.A.; Nascimento, N.R.F.; RAO, V.S.N. Relaxant effects of an alkaloid-rich fraction from *Aspidosperma ulei* root bark on isolated rabbit corpus cavernosum. *Int. J. Impot. Res.* **2008**, *20*, 225–263.
16. Souza, L.G.; Alves, N.M.; Paula, J.R.; Valadares, M.C.; Bara, M.T.F.; Cunha, L.C.; Garrote, C.F.D. Estudo Farmacognóstico de *Aspidosperma subcanium* Mart., Apocynaceae. *Rev. Eletr. Farm.* **2005**, *2*, 210–213. (in Portuguese).
17. Goloni, R.; Alves, N.M.; Garrote, C.F.D.; Paula, J.R.; Valadares, M.C.; Bara, M.T.F.; Cunha, L.C. Estudo da Toxicidade Aguda do *Aspidosperma subcanium* Martius. *Rev. Eletr. Farm.* **2005**, *2*, 89–91. (in Portuguese).
18. Tanaka, J.C.A.; Silva, C.C.; Oliveira, A.I.B.; Nakamura, C.V.; Dias Filho, B.P. Antibacterial activity of indole alkaloids from *Aspidosperma ramiflorum*. *Braz. J. Med. Biol. Res.* **2006**, *39*, 387–391.

19. Ferreira, D.T.; Silva, J.V., Jr.; Soeira, L.S.; Filho, L.A.Z.; Ishikawa, N.K.; Barbosa, A.M.; Yassumoto, L. Avaliação da atividade antifúngica dos extratos etanólicos de raiz, caule e folhas de *Aspidosperma polyneuron*. In *XI Encontro de Química da Região Sul*; SBQSUL: Pelotas, Rio Grande do Sul, Brazil, 2003. (in Portuguese).
20. Le Men, J.; Taylor, W.I. A uniform numbering system for indole alkaloids. *Experientia* **1965**, *21*, 508–510.
21. Dewick, P.M. *Medicinal Natural Products: A Biosynthetic Approach*, 3rd ed.; John Wiley & Sons: Chippenhams City, UK, 2009.
22. Danieli, B.; Palmisano, G. Alkaloids from *Tabernaemontana*. In *The Alkaloids*; Brossi, A., Ed.; Academic Press: New York, NY, USA, 1986; pp. 1–130.
23. Brennan, J.P.; Saxton, J.E. A new synthesis of the cylindrocarine group of alkaloids. *Tetrahedron* **1986**, *42*, 6719–6734.
24. Saxton, J.E.; Smith, A.J.; Lawton, G. *Aspidosperma* alkaloids: The total synthesis of (±)-*N,O*-diacetylcylindrocarpinol, (±)-cylindrocarine, (±)-cylindrocarpine, and (±)-cylindrocarpidine. *Tetrahedron Lett.* **1975**, *47*, 4161–4164.
25. Lawton, G.; Saxton, J.E.; Smith, A.J. The synthesis of aspidosperma alkaloids containing a functional group at C-18; The total synthesis of (±)-*N,O*-diacetylcylindrocarpinol, (±)-cylindrocarine, (±)-cylindrocarpine, (±)-cylindrocarpidine, and (±)-20-allyl-20-desethyl-20-epiaspidospermine. *Tetrahedron* **1977**, *33*, 1641–1653.
26. Hugel, G.; Royer, D.; Sigaut, F.; Lévy, J. Flow thermolysis rearrangements in the indole alkaloid series: 1,2-Dehydroaspidospermidine. *J. Org. Chem.* **1991**, *56*, 4631–4636.
27. Fumagali, E.; Gonçalves, R.A.C.; Machado, M.F.P.S.; Vidoti, G.J.; Oliveira, A.J.B. Produção de metabólitos secundários em cultura de células e tecidos de plantas: O exemplo dos gêneros *Tabernaemontana* e *Aspidosperma*. *Rev. Bras. Farmacog.* **2008**, *18*, 627–641. (in Portuguese).
28. Manske, R.H.F.; Rodrigo, R. *The Alkaloids*; Academic Press: New York, NY, USA, 1965.
29. Kuehne, M.E.; Roland, D.M.; Hafter, R. Studies in biomimetic alkaloid syntheses. 2. Synthesis of vincadifformine from tetrahydro- β -carboline through a secodine intermediate. *J. Org. Chem.* **1978**, *43*, 3705–3710.
30. Wenkert, E.; Cochran, D.W.; Hagaman, E.W.; Schell, F.M.; Neuss, N.; Katner, A.S.; Potire, P.; Kan, C.; Plat, M.; Koch, M.; *et al.* Carbon-13 nuclear magnetic resonance spectroscopy of naturally occurring substances. XIX. *Aspidosperma* alkaloids. *J. Am. Chem. Soc.* **1973**, *95*, 4990–4995.
31. Djerassi, C.; Brewer, H.W.; Budzikiewicz, H.; Orazi, O.O.; Corral, R.A. The structures of the *Aspidosperma* alkaloids spagazzinine and spagazzinidine. *J. Am. Chem. Soc.* **1962**, *84*, 3480–3485.
32. Oliveira, A.J.B. Estudo de seis Espécies do Gênero *Aspidosperma* Utilizando GC, GC/MS e HPLC: Análise Qualitativa e Quantitativa. Teste Bioautográfico; Cultura de Tecidos e Células Vegetais e Rota de Preparação dos Compostos Diméricos Ramiflorina A e Ramiflorina B. PhD Thesis. Universidade Estadual de Campinas, Campinas—SP, 1999. (in Portuguese).
33. Djerassi, C.; Gilbert, B.; Schoolery, J.N.; Johnson, L.F.; Biermann, K. Alkaloid studies XXVI. The constitution of pyriferidine. *Experientia* **1961**, *17*, 162–163.
34. Zèches, M.; Mesbah, K.; Richard, B.; Moretti, C.; Nuzillard, J.M.; Le Men-Olivier, L. Alkaloids from leaves and stems of *Vallesia glabra*. *Planta Med.* **1995**, *61*, 89–91.

35. Craveiro, A.A.; Matos, F.J.A.; Serur, L.M. Alkaloids of *Aspidosperma pyrifolium*. *Phytochemistry* **1983**, *22*, 1526–1528.
36. Schnoes, H.K.; Burlingame, A.L.; Biemann, K. Application of mass spectrometry to structure problems: The occurrence of eburnamenine and related alkaloids in *Rhazya stricta* and *Aspidosperma quebracho-blanco*. *Tetrahedron Lett.* **1962**, *22*, 993–999.
37. Biemann, K.; Friedmann-Spiteller, M.; Spiteller, G. An investigation by mass spectrometry of the alkaloids of *Aspidosperma quebracho-blanco*. *Tetrahedron Lett.* **1961**, *14*, 485–492.
38. Walser, A.; Djerassi, C. Alkaloid-Studien LII [1]. Die Alkaloids aus *Vallesia dichotoma* RUIS et PAV. *Helv. Chim. Acta* **1965**, *48*, 391–403. (in German).
39. Ferreira, J.M.; Gilbert, B.; Owellen, R.J.; Djerassi, C. The alkaloids of *Aspidosperma discolor* A. DC. *Experientia* **1963**, *15*, 585–586.
40. Witkop, B.; Patrik, J.B. Aspidospermine II. *J. Am. Chem. Soc.* **1954**, *76*, 5603–5608.
41. Conroy, H.; Brook, P.R.; Rout, M.K.; Silverman, N. Aspidospermine. II. Nuclear magnetic resonance spectra and classical degradations. *J. Am. Chem. Soc.* **1958**, *80*, 5178–5185.
42. Mitaine, A.C.; Mesbah, K.; Richard, B.; Petermann, C.; Arrazola, S.; Moretti, C.; Zèches-Hanrot, M.; Le Men-Olivier, L. Alkaloids from *Aspidosperma* species from Bolivia. *Planta Med.* **1996**, *62*, 458–461.
43. Achenbach, H. O-Demethylpalosin, Ein Neues Alkaloid aus *Tabernaemontana amygdalifolia*. *Tetrahedron Lett.* **1966**, *41*, 507–5030. (in German).
44. McLean, S.; Reynolds, W.F.; Zhu, X. Assignment of the ¹H and ¹³C spectra of aspidocarpine and assignment of the structure and stereochemistry of the von Braun reaction product of aspidocarpine by 2D nmr spectroscopy. *Can. J. Chem.* **1987**, *65*, 200–204.
45. Burnell, R.H.; Medina, J.D. Alkaloids of *Aspidosperma cuspa* Blake. *Phytochemistry* **1968**, *7*, 2045–2051.
46. Djerassi, C.; Archer, A.A.P.G.; George, T.; Gilbert, B.; Antonaccio, L.D. Alkaloid Studies—XXX: Isolation and constitution of three new *Aspidosperma* alkaloids: Cyliandrocarpine, cyliandrocarpidine and pyrifolidine. *Tetrahedron* **1961**, *16*, 212–223.
47. Pinar, M.; Schmid, H. Aspidolimin. *Helv. Chim. Acta* **1962**, *45*, 1283–1286.
48. Miranda, E.C.; Gilbert, B. Alkaloids of *Aspidosperma melanocalyx* Muell-Arg. *Experientia* **1969**, *15*, 575–576.
49. Panas, J.-M.; Morfaux, A.-M.; Olivier, L.; Le Men, J. Alcaloïdes des feuilles de l'*Amsonia Tabernaemontana* Walt., Apocynacées. *Ann. Pharmac. Fran.* **1972**, *30*, 273–276. (in French).
50. Zèches-Hanrot, M.; Nuzzillard, J.-M.; Richard, B.; Schaller, H.; Hadi, H.A.; Sévenet, T.; Le Men-Olivier, L. Alkaloids from leaves and stem bark of *Ervatamia peduncularis*. *Phytochemistry* **1995**, *40*, 587–591.
51. Pinar, M.; Von Philipsborn, W.; Vetter, W.; Schmid, H. Limaspermin. *Helv. Chim. Acta* **1962**, *45*, 2260–2265.
52. Pinar, M.; Schmid, H. 3'-Methoxy-Limaspermin, Limapodin, 3'-Methoxy-Limapodin Und Tubotaiwin Aus *Aspidosperma limae* Woodson. *Liebigs Ann. Chem.* **1963**, *668*, 97–104. (in German).
53. Medina, J.D.; Di Genova, L. Alkaloids of the bark of *Aspidosperma rhombeosignatum*. *Planta Med.* **1979**, *37*, 165–167.

54. Gebreyesus, T.; Djerassi, C. Alkaloid studies. Part LXVI. Reactions of some *Aspidosperma* alkaloids with *m*-chloroperbenzoic acid. Removal of the angular ethyl group of aspidospermine. *J. Chem. Soc. Perkins Transactions I. Org. Bio-Org. Chem.* **1972**, 6, 849–860.
55. Milborrow, B.V.; Djerassi, C. Alkaloid studies. Part LXI. The structure of twelve new alkaloids from *Aspidosperma cylindrocarpon*. *J. Chem. Soc. (C)* **1969**, 417–424.
56. Djerassi, C.; Archer, A.A.; George, T.; Gilbert, B.; Schoolery, J.N.; Johnson, L.F. Alkaloid studies XXV. The structures of the *Aspidosperma* alkaloids cylindrocarpine and cylindrocarpidine. *Experientia* **1960**, 16, 532–534.

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