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ALKALOIDS OF *KOPSIA LAPIDILECTA*[1]

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ABSTRACT.—From the bark and leaves of *Kopsia lapidilecta* seven indole alkaloids were isolated. Three are known: venalstonine [**1**], lapidilectine A [**2**], and lapidilectine B [**3**]. The other four are new and possess the same skeleton as lapidilectine A [**2**]; the new alkaloids are isolapidilectine [**4**], lapidilectam [**5**], lapidilectinol [**6**], and epilapidilectinol [**7**]. The configuration of C-2 and C-20 of lapidilectine A [**2**] is revised. All the structures were elucidated chiefly by 2D nmr analysis.

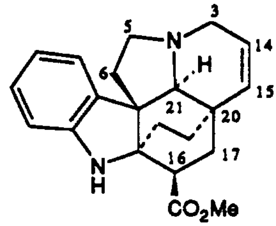
The genus *Kopsia* (Apocynaceae, subfamily Plumerioideae, tribe Rauvolfieae) comprises about thirty species which grow from India to Vanuatu and from Yunan to Java (1). Various medicinal uses have been reported; *Kopsia pitardii* (=*Kopsia officinalis*) is used for the treatment of rheumatoid arthritis, dropsy, and tonsillitis in China. The *Kopsia* plants are also known to possess interesting varieties of indole skeletons, e.g., the five-membered ring, kopsijasminilam (2). For the above reasons, our laboratory has undertaken the investigation of the bark and leaves of the Malayan *Kopsia lapidilecta* van der Sleesen, a tree of 2 m to 5 m in height with yellow flowers and red fruits.

Stems and bark of *K. lapidilecta* were extracted with CH_2Cl_2 using the conventional alkaloid extraction method. Seven alkaloids were afforded, three of which were known: (−)-venalstonine [**1**] (3–5), (−)-lapidilectine A [**2**] (6), and (+)-lapidilectine B [**3**] (6). Four were new. The new alkaloids were named (+)-isolapidilectine [**4**], (+)-lapidilectam [**5**], (−)-lapidilectinol [**6**], and (+)-epilapidilectinol [**7**]. All new alkaloids were isolated in the amorphous form, and the elucidation of their structures was performed using spectral methods, primarily 2D nmr techniques (COSY, HMQC, and NOESY). Lapidilectine A and lapidilectine B have been reported recently (6); however, in this present paper, a correction is suggested on the configuration of C-2 and C-20 in lapidilectine A, based on biogenetic arguments. The four new alkaloids are similar to lapidilectine A [**2**].

Since we have found venalstonine [**1**] in the same plant, the stereochemistry of lapidilectine A [**2**], which is probably biogenetically related to **1**, has to be as designated as in **2**. The change in stereochemistry does not alter the position of the C-16 carbomethoxy above the aromatic plane, which is a shielded zone, as mentioned in the previous publication (6), in order to reduce steric hindrance. This explains the upfield shift of its MeO (δ 2.93). The NOESY experiment is in accordance with structure **2** (Figure 1).

Isolapidilectine A [**4**], $[\alpha]_D$ +54° ($CHCl_3$, c=0.72), revealed uv maxima typical of a dihydroindole chromophore: 207, 249, and 284 nm (log ε 4.26, 4.01, and 3.44). The hreims showed a molecular ion at m/z 440.1955 $[M]^+$ (calcd 440.1948) corresponding to a molecular formula of $C_{24}H_{28}N_2O_6$, which is the same as that of lapidilectine A [**2**]. Their fragmentation patterns were also identical (m/z 381, 354, and 295), implying that they belong to the same skeletal type. The ir spectrum exhibited absorptions at 1717 and 1730 cm^{-1}, indicating the presence of several carbonyls. The ^{13}C nmr revealed three MeO

[1]This work has been done in the framework of a collaborative program between CNRS (France) and the University of Malaya (Kuala Lumpur, Malaysia).



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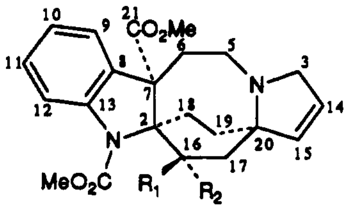
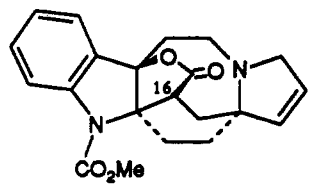


2 R_1=CO_2Me, R_2=H
4 R_1=H, R_2=CO_2Me



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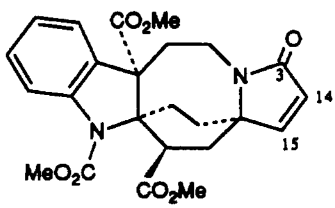

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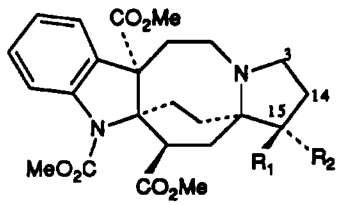

6 R_1=OH, R_2=H
7 R_1=H, R_2=OH

signals between δ 51 and 53, two carbonyl peaks at δ 171.9 (C-21) and δ 172.6 typical of methyl esters, and another one at δ 153.43 indicative of a urethane. In the ^{1}H nmr, the MeO protons resonated at δ 3.49 (methyl ester attached to C-16), δ 3.58 (methyl ester 21) and δ 3.79 (urethane). All the signals observed were similar to those of lapidilectine A [**2**] (Table 1) except for the MeO proton peak of the methyl ester attached to C-16 which is deshielded to about 0.5 ppm. This proved that the MeO group is no longer situated on the aromatic plane, as in the case of lapidilectine A [**2**] (δ 2.93). Hence,

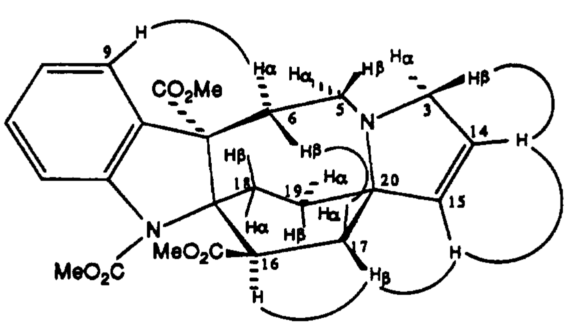

FIGURE 1. NOESY correlations for lapidilectine A [**2**].

TABLE 1. ^{13}C-nmr (62.5 MHz) and ^{1}H-nmr (400 MHz) Data for Lapidilectine A [**2**], Isolapidilectine A [**4**], and Lapidilectam [**5**] ($CDCl_3$).[a]

Position	Compound					
	2		**4**		**5**	
	δ C	δ H (*J* in Hz)	δ C	δ H (*J* in Hz)	δ C	δ H (*J* in Hz)
2	75.7		74.5		73.6	
3α	62.4	3.45 m	63.5	3.37 brd (15)	170.7	
3β		3.75 m		3.89 brd (15)		
5	49.2	3.20 m	49.2	3.35 m	39.8	4.81 dd (8,15)
		3.20 m		3.09 m		3.28 dd (10,15)
6α	29.8	2.77 m	33.0	2.93 m	25.3	3.17 dd (8,15)[b]
6β		2.77 m		2.15 m		2.84 dd (10,15)[b]
7	62.4		60.8		64.7	
8	132.0		132.3		129.9	
9	123.0	7.01 dd (7.5,1)	122.9	7.12 d (7.5)	123.0	7.12 d (7.5)
10	122.8	6.87 ddd (7.5,7.5,1)	123.4	7.02 dd (7.5,7.5)	123.7	7.02 dd (7.5,7.5)
11	129.1	7.21 ddd (7.5,7.5,1)	129.0	7.25 dd (7.5,7.5)	129.9	7.25 dd (7.5,7.5)
12	115.6	7.63 brd (7.5)	117.6	7.58 brd (7.5)	116.0	7.54 brd (7.5)
13	143.1		142.4			
14	125.7	5.71 dt (6,1)	123.6	5.62 brd (6)	124.1	6.09 d (6)
15	138.4	5.49 brd (6)	138.9	5.52 brd (6)	154.4	6.81 d (6)
16	41.0	3.87 m	42.9	3.21 dd (7,11.5)	41.1	3.85 m
17α	32.5	2.05 dd (10,15)	36.9	2.61 dd (11.5,14)	33.7	2.18–2.38 m[c]
17β		2.63 m		1.69 dd (7,14)		
18α	29.3	3.10 m	24.6	3.28 m[b]	27.1	2.38 m[b]
18β		2.63 m		2.22 m[b]		3.42 m[b]
19	31.0	1.75 m	22.4	2.02 m	29.8	1.65 m
		1.61 m		1.61 m		2.31 m
20	67.3		65.4		65.0	
21	173.0		171.9		172.4	
CO ester	174.2		172.6		173.4	
CO urethane	154.0		153.4		154.4	
OMe 21	51.6[d]	3.57 s	52.3[d]	3.58 s	52.7[d]	3.48 s
OMe ester	52.1[d]	2.93 s	51.5[d]	3.49 s	52.1[d]	2.88 s
OMe urethane	52.4[d]	3.95 s	52.3[d]	3.79 s	52.8[d]	3.91 s

[a]Assignments based on COSY, HMQC, HMBC, and NOESY experiments.
[b]Configuration (α or β) cannot be assigned.
[c]δ for H-17α and H-17β (partly overlapped).
[d]In the same column assignments can be reversed.

this observation suggests a C-16α carbomethoxy. Another point that supports this conclusion is the more upfield shift of the urethane methoxy at δ 3.79 rather than δ 3.95 in lapidilectine A [**2**], which may be due to steric hindrance (7). The pseudoaxial vicinal coupling between H-16 and H-17 (ca. 11 Hz), present in both lapidilectine A [**2**] and its isomer, can be explained by the transformation to the other twist boat conformation (Figure 2) in order to permit the carbomethoxy to point out of ring C, thus reducing steric constraint. The characteristic five-membered ring olefin protons of the lapidilectine type alkaloids resonated at δ 5.62 and δ 5.52 with a ^{3}J value of 6 Hz. Finally, the NOESY data (Figure 2) confirmed the stereochemistry of isolapidilectine A [**4**] and particularly the C-16α configuration (H-16/H-17β cross peak).

Lapidilectam [**5**], [α]D +77° ($CHCl_3$, c=0.55), also showed uv maxima indicative of a dihydroindole chromophore: 209, 253, and 290 nm (log ε 4.38, 4.10, and 3.45). The hreims exhibited a molecular ion at *m/z* 454.1730, which corresponded to a molecular

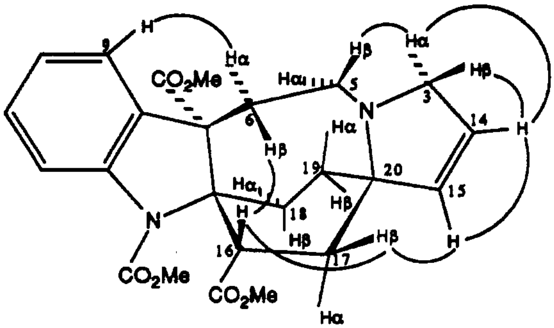

FIGURE 2. NOESY correlations for isolapidilectine A [4].

formula of $C_{24}H_{26}N_2O_7$ (calcd 454.1740), indicating an incorporation of an oxygen accompanied by the loss of two hydrogens and suggesting the presence of a carbonyl function. This was further supported by the ir spectrum which revealed an absorption typical of a five-membered ring lactam at 1680 cm^{-1} (8) and the ^{13}C-nmr spectrum which exhibited a lactam carbonyl peak at δ 170.7. In addition, the normal H-3 protons (δ 3.4–3.8) and their corresponding carbon (δ 48–49) signals have disappeared, thus signifying that it is C-3 that bears the carbonyl oxygen. As expected, the olefinic protons (H-14 and H-15) have moved downfield to δ 6.01 and δ 6.81, respectively, due to the deshielding effect of the carbonyl C-3. Apart from the differences cited above, all the other spectral data were similar to those of lapidilectine A [**2**].

Lapidilectinol [**6**], [α]D −5° ($CHCl_3$, c=0.5), exhibited uv maxima at 208, 253, and 287 nm (log ε 4.46, 4.07, and 3.49) and a shoulder at 224 nm (log ε 4.26). The hreims revealed a molecular peak at *m/z* 458.2066 (calcd 458.2054), corresponding to a molecular formula of $C_{24}H_{30}N_2O_7$. Lapidilectinol differs from lapidilectine A [**2**] by the presence of an alcohol function and the hydrogenation of the C-14–C-15 olefin. In fact, the ^1H nmr and ^{13}C nmr do not show evidence of the olefin. Moreover, a peak at δ_C 81.4 was observed which is reminiscent of an oxymethine. The COSY experiment indicated that the corresponding proton (δ 3.75) correlated with the hydrogens on C-14. The latter were also coupled to CH_2-3, which showed a ^{13}C-nmr signal at δ 53.1 indicative of its vicinal position to N-4. The OH is thus situated on C-15. Its relative configuration was resolved to be β by the NOESY experiment (Table 2) and manipulation of the Dreiding molecular model.

Epilapidilectinol [**7**], [α]D +19° ($CHCl_3$, c=0.8), showed uv maxima similar to those of lapidilectinol [**6**] at 209, 254, and 288 nm (log ε 4.33, 3.94, and 3.37) and a shoulder at 226 (log ε 4.13). Its molecular formula was deduced to be $C_{24}H_{30}N_2O_7$ ($[M]^+$ found *m/z* 458.2055, calcd 458.2053) with the aid of hreims. The COSY and HMQC experiments again demonstrated that the OH is attached on C-15 (Table 1). The NOESY spectrum (Table 2) and observation from the Dreiding molecular model indicated that the OH has an α configuration.

The relative configuration at C-16 of lapidilectam [**5**], lapidilectinol [**6**], and epilapidilectinol [**7**] is assumed to be the same as that of lapidilectine A [**2**] due to the fact that the carbomethoxy attached to C-16 resonated consistently at about δ 2.90 (^1H nmr) as in the case of the latter.

Based on biogenetic reasons and cd spectra of the alkaloids **1**, **2**, and **4**–**6**, which show a positive Cotton effect in the region of 250 nm (9), we concluded that all these alkaloids have the absolute stereochemistry as designated in the formulas.

TABLE 2. ^{13}C-nmr (62.5 MHz), ^{1}H-nmr (400 MHz), and NOESY (400 MHz) Data for Lapidilectinol [6] and Epilapidilectinol [7] ($CDCl_3$).[a]

Position	Compound					
	6			7		
	δC	δH (*J* Hz)	NOESY	δC	δH (*J* Hz)	NOESY
2	75.3			75.4		
3α	53.1	3.15 m[b]	5β	50.7	2.92 m	5α, 14α
3β		2.93 m[b]	5β		3.17 m	
5α	50.3	3.18 m	18α	49.6	2.72 m	
5β		3.42 dt (15, 4)	6α		3.19 m	6α
6α	28.0	2.90 m	9	28.4	2.80 m	9
6β		2.90 m			3.09 m	17β
7	64.0			62.9		
8	131.5			131.3		
9	122.8	7.02 d (7.5)		122.7	7.02 d (7.5)	
10	123.1	7.18 dd (7.5, 7.5)		123.0	6.92 dd (7.5, 7.5)	
11	131.5	6.93 dd (7.5, 7.5)		129.0	7.18 dd (7.5, 7.5)	
12	115.6	7.52 brd		115.6	7.57 brd	
13	142.3			143.2		
14α	29.9	1.83 m[b]		29.7	1.69 m	
14β		2.18 m[b]			2.13 m	15
15	81.4	3.73 dd (4, <1)	17α, 19β	77.7	3.75 dd (13, 5)	
16	40.8	3.82 m		41.2	3.74 d (8)	18β
17α	31.6	2.20 m	17β	27.7	2.71 m	
17β		2.54 ddd (14, 11, 2)			2.38 m	
18α	29.5	2.32 m[b]		28.4	2.39 m	
18β		2.11 m[b]			1.99 ddd (15, 10, <1)	
19α	29.7	1.53 m		23.9	1.54 m	
19β		1.63 m			1.78 m	
20	63.4			62.1		
21	173.0			172.9		
CO ester	174.2			174.1		
CO urethane	154.2			154.1		
OMe 21	51.8[c]	3.52 s		51.6[c]	3.54 s	
OMe ester	52.8[c]	2.99 s		52.1[c]	2.95 s	
OMe urethane	53.5[c]	3.97 s		53.0[c]	3.98 s	

[a]Assignments based on COSY, HMQC, and NOESY experiments.
[b]Configuration (α or β) cannot be assigned.
[c]In the same column assignments can be reversed.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Mp's (uncorrected) were determined on a micro hot-stage apparatus. Optical rotations at 20° were taken on a Perkin-Elmer 241 polarimeter. Spectra were recorded on: uv, Shimadzu UV-161 uv-visible spectrophotometer; ir, Nicolet 205 FT-IR spectrometer; cd, Jobin-Yvon Mark 5; eims (70 eV), Kratos MS 50; nmr, Bruker AC 250 (^{13}C spectra), AM 400 (^{1}H and 2D spectra). Uv and cd spectra were recorded in MeOH. Cc was performed using Si gel Merck H60, and tlc with Si gel 60 F_{254}. Visualization was by viewing under uv light and spraying with Dragendorff's reagent followed by 50% H_2SO_4.

PLANT MATERIAL.—Stem and bark of *K. lapidilecta* were collected in Mersing, Malaysia, February 14, 1990. Identification was made by Dr. B. David. Voucher specimens (KL 3832) were deposited at the Museum National d'Histoire Naturelle in Paris and at the Herbarium of the Department of Chemistry, University of Malaya, Kuala Lumpur, Malaysia.

EXTRACTION AND ISOLATION OF THE ALKALOIDS.—The dried, ground stems and bark of *K. lapidilecta* (2.4 kg) were moistened with 40% NH_4OH and extracted exhaustively with CH_2Cl_2 at room temperature.

The concentrated CH_2Cl_2 extract was diluted with Et_2O and reextracted with 5% aqueous HCl. The aqueous layer was basified to ca. pH 11 with NH_4OH and re-extracted with CH_2Cl_2 until a negative Mayer test was obtained. The CH_2Cl_2 extracts, when pooled, washed with H_2O, dried over anhydrous Na_2SO_4, and evaporated, yielded a crude alkaloid fraction (5.1 g). A portion of the crude product (4.4 g) underwent extensive chromatography and preparative tlc. The alkaloids were eluted in the following order from the cc (solvent): (−)-venalstonine [**1**] (20 mg) [CH_2Cl_2-MeOH (99:1)]; (−)-lapidilectine [**2**] (300 mg) [CH_2Cl_2-MeOH (98:2)]; (−)-lapidilectam [**5**] (7 mg) [CH_2Cl_2-MeOH (98:2), then CH_2Cl_2 100% and NH_3 vapor (preparative tlc)]; (+)-isolapidilectine [**3**] (90 mg) [CH_2Cl_2-MeOH (95:5), then CH_2Cl_2-MeOH (99:1)]; (−)-lapidilectinol [**6**] (36 mg) [CH_2Cl_2-MeOH (95:5), then CH_2Cl_2-MeOH (saturated with NH_3) (99.5:0.5)]; (+)-epilapidilectinol [**7**] (26 mg) [CH_2Cl_2-MeOH (95:5), then CH_2Cl_2-MeOH (saturated with NH_3) (99.5:0.5)]. The leaves (1.7 kg) were extracted with the same procedure as above and yielded 7 g of crude alkaloids. Routine cc of 1 g of the crude alkaloids afforded lapidilectine B [**3**], 38 mg [CH_2Cl_2-MeOH (99:1), then CH_2Cl_2-MeOH (99.5:0.5)].

Venalstonine [**1**].—Cd λ ext 224 ($\Delta\epsilon$ −0.92), 250 nm ($\Delta\epsilon$ +5.3).

Lapidilectine A [**2**].—Cd λ ext 220 ($\Delta\epsilon$ −27), 257 ($\Delta\epsilon$ +27), 290 nm ($\Delta\epsilon$ −0.43); ^{1}H and ^{13}C nmr see Table 1.

Isolapidilectine A [**4**].—Uv λ max 207 (log ϵ 4.26), 225 (sh) (log ϵ 4.01), 249 (log ϵ 3.86), 284 nm (log ϵ 3.44) nm; eims *m/z* (% rel. int.) 440 (48), 409 (11), 381 (21), 354 (60) 295 (100); cd λ ext 233 ($\Delta\epsilon$ −6.5), 257 ($\Delta\epsilon$ +3.7), 289 nm ($\Delta\epsilon$ +1.4); ^{1}H and ^{13}C nmr see Table 1 and Figure 2.

Lapidilectam [**5**].—Uv λ max 209 (log ϵ 4.38), 254 (log ϵ 4.10), 256 (sh) (log ϵ 4.14), 290 nm (log ϵ 3.45) nm; ir ν max ($CHCl_3$) cm^{-1} 1680, 1691, 1728; eims *m/z* (% rel. int.) 454 (32), 423 (3), 395 (100), 368 (44), 309 (19); cd λ ext 228 ($\Delta\epsilon$ −25), 257 nm ($\Delta\epsilon$ +21); ^{1}H and ^{13}C nmr see Table 1.

Lapidilectinol [**6**].—Uv λ max 208 (log ϵ 4.46), 224 (sh) (log ϵ 4.26), 253 (log ϵ 4.07), 287 nm (log ϵ 3.49) nm: ir ν max ($CHCl_3$) cm^{-1} 1700, 1730; eims *m/z* (% rel. int.) 458 (45), 440 (9), 427 (10), 399 (100), 372 (9), 371 (21), 354 (13), 313 (35), 295 (30); cd λ ext 234 ($\Delta\epsilon$ −8.4), 262 ($\Delta\epsilon$ +14), 291 nm ($\Delta\epsilon$ −0.7); ^{1}H and ^{13}C nmr see Table 2.

Epilapidilectinol [**7**].—Uv λ max 209 (log ϵ 4.33), 226 (sh) (log ϵ 4.13), 254 (log ϵ 3.94), 288 nm (log ϵ 3.37) nm; ir ν max ($CHCl_3$) cm^{-1} 1700, 1730; eims *m/z* (% rel. int.) 458 (57), 440 (4), 427 (10), 399 (100), 372 (8), 371 (15), 352 (23), 313 (33), 295 (17); cd λ ext 228 ($\Delta\epsilon$ −19) and 258 ($\Delta\epsilon$ +16); ^{1}H and ^{13}C nmr see Table 2.

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