New Indole Alkaloids from *Kopsia*. Alkaloid Variation in *Kopsia singapurensis*

by **G. Subramaniam**[1]) and **Toh-Seok Kam***

Department of Chemistry, University of Malaya, 50603 Kuala Lumpur, Malaysia
(phone: +603-79674266; fax: +603-79674193; e-mail: tskam@um.edu.my)

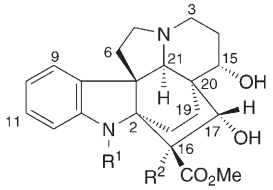
Five new indole alkaloids, *viz*., kopsiloscine G (**2**), kopsidarine (**4**), kopsimaline F (**6**), kopsidine C *N*-oxide (**7**), and aspidophylline B (**9**), in addition to 17 other known alkaloids, were obtained from the leaf and stem-bark extract of the Malaysian *Kopsia singapurensis*. The structures were determined by using NMR and MS analysis. The results are compared with the alkaloidal composition of another sample of *K. singapurensis*, collected from the same location but at a different time of the year, as well as with other samples of *K. singapurensis* collected from different locations.

Introduction. – Plants of the genus *Kopsia* (Apocynaceae) are distributed mainly over Southeast Asia, India, and China but have its stronghold in Southeast Asia. About 16 species occur in Malaysia [1–3]. In our systematic study of the Malaysian representatives of this genus, we have reported many examples of new alkaloids, which are distinguished by their structural novelty, as well as useful bioactivity [4–24]. Recent examples of unusual alkaloids from *Kopsia* which are notable for possessing novel ring systems, and which were postulated to derive from known monoterpenoid indole precursors through pathways involving deep-seated rearrangements and/or loss of key fragments include, *inter alia*, the cage indole arbophylline [8], the three-nitrogen pentacyclic indole arboflorine [9], the tetracyclic indole mersicarpine [11], the tetracyclic quinolinic alkaloid mersilongine [12], and the pair of intriguing regioisomeric tetracyclic indoles arboricine and arboricinine [6]. As part of our continuing studies of this genus, we investigated the alkaloidal composition of *K. singapurensis*. An early study by *Thomas et al.* revealed the presence of kopsingine and kopsaporine [25]. A subsequent study by *Sevenet* and co-workers provided additional aspidofractinine-type alkaloids, including a kopsaporine derivative and the singapurensines [26], congeners of the epoxy-bridged kopsidine-type alkaloids [24][27]. Our sample of *K. singapurensis* was collected in Pahang, Malaysia. In an attempt to investigate the seasonal dependence of the alkaloid composition, two collections were carried out in the same location, but at different times of the year (July and November of the same year). We have reported the alkaloidal composition of the July sample (A), including the isolation of new alkaloids as well as the biological activity of some of the alkaloids [28]. Herein we report the alkaloidal composition of the November sample (B), including the isolation of additional new compounds.

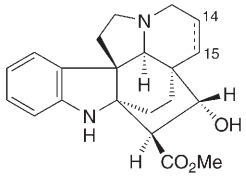
---

[1]) Present address: *MerLion Pharmaceuticals*, 1 Science Park Road, The Capricorn 05-01, Singapore Science Park II, Singapore 117528.

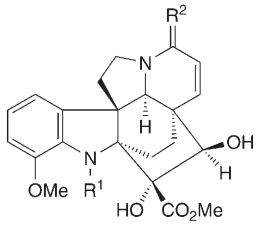
Results and Discussion. – A total of 22 alkaloids (see *Exper. Part*) were obtained, 13 from the leaf extract and 14 from the stem-bark extract. Of these, five are new alkaloids, while kopsingine, kopsinganol, 16-epiakuammiline, 16-epideacetylakuammiline, and akuammidine were common in both the stem and leaf extracts. The kopsiloscines A–F, which represent a group of aspidofractinine-type alkaloids all sharing a common configuration feature, which is an α-oriented OH group at C(17), have been reported from the other sample (A) of *K. singapurensis* [28]. In the present sample (B), only kopsiloscine C (**1**) was isolated, in addition to a new member of this group, kopsiloscine G (**2**).



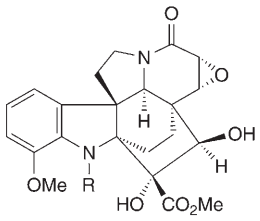
1 R^1 = CO_2Me, R^2 = OH [2])



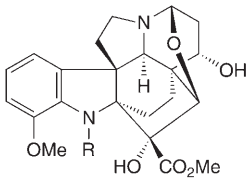
2 [2])
3 Δ^{14} [2])



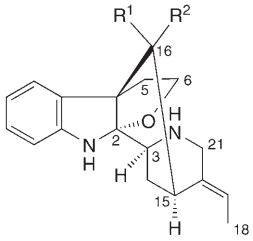
4 R^1 = CO_2Me, R^2 = O
5 R^1 = CO_2Me, R^2 = H_2



6 R = CO_2Me



7 R = CO_2Me, *N*(4)→O
8 R = CO_2Me



9 R^1 = CH_2OAc, R^2 = CO_2Me [2])
10 R^1 = CH_2OH, R^2 = CO_2Me [2])

Kopsiloscine G[2]) (**2**) was obtained as a yellowish oil. The UV spectrum showed three absorption bands (203, 246, and 294 nm), characteristic of a dihydroindole chromophore, while the IR spectrum indicated the presence of OH (3432 cm^{-1}), NH (3350 cm^{-1}), and ester (1728 cm^{-1}) functions. The MS showed a molecular ion at *m/z* 354, which is two mass units higher than that of (17α)-17-hydroxy-Δ^{14}-kopsinine[2]) (**3**) and consistent with the molecular formula $C_{21}H_{26}N_2O_3$. Examination of the ^{1}H- and ^{13}C-NMR data (*Tables 1* and *2*) as well as 2D COSY and HMQC data showed that **2** resembles **3** [29][30], except for the absence of signals due to the C(14)=C(15) bond. The structure of **2** was established as (17α)-17-hydroxykopsinine[2]).

The C(14)=C(15) bond of **3** is replaced by a CH_2CH_2 unit in **2**, the ^{1}H-NMR signals of which were shifted upfield. The same behavior was also shown by the C(14) and C(15) resonances. The other NMR

[2]) Trivial atom numbering; for systematic names, see *Exper. Part*.

Table 1. *^{1}H-NMR Data* (400 MHz, $CDCl_3$) *of Compounds* **2**, **4**, **6**, **7**, *and* **9**[a])[2]). δ in ppm, J in Hz.

	2	**4**	**6**	**7**	**9**
CH_2(3) or H–C(3)	2.92–2.97 (*m*), 3.10–3.15 (*m*)	–	–	5.06 (*d*, *J* = 3.3)	3.21 (*t*, *J* = 3)
CH_2(5)	2.92–2.97 (*m*), 3.10–3.15 (*m*)	3.32 (*td*, *J* = 12, 5), 4.04 (*dd*, *J* = 12, 8)	3.18 (*td*, *J* = 12, 5), 4.18 (*dd*, *J* = 12, 8)	3.70 (*dd*, *J* = 12.5, 7.5), 3.83–3.87 (*m*)	3.50 (*ddd*, *J* = 12, 8, 5.5), 4.06 (*t*, *J* = 8)
CH_2(6)	1.50 (*ddd*, *J* = 14, 8, 6), 2.51 (*ddd*, *J* = 14, 8.5, 5)	1.62 (*dd*, *J* = 12, 5), 2.93 (*td*, *J* = 12, 8)	1.53 (*dd*, *J* = 12, 5), 2.54 (*td*, *J* = 12, 8)	3.47 (*td*, *J* = 14.5, 7.5), 1.75–1.79 (*m*)	2.52 (*dd*, *J* = 12, 5.5), 2.75 (*td*, *J* = 12, 8)
H–C(9)	7.18 (br. *d*, *J* = 7.5)	6.84 (br. *d*, *J* = 7.4)	6.80 (*d*, *J* = 8)	7.17 (*dd*, *J* = 8, 1)	6.59 (*d*, *J* = 7.5)
H–C(10)	6.75 (*td*, *J* = 7.5, 1)	7.09 (*dd*, *J* = 8.3, 7.4)	7.07 (*t*, *J* = 8)	7.12 (*t*, *J* = 8)	6.81 (*t*, *J* = 7.5)
H–C(11)	7.00 (*td*, *J* = 7.5, 1)	6.88 (br. *d*, *J* = 8.3)	6.88 (*d*, *J* = 8)	6.90 (*dd*, *J* = 8, 1)	7.09 (*t*, *J* = 7.5)
H–C(12)	6.66 (br. *d*, *J* = 7.5)	–	–	–	7.50 (*d*, *J* = 7.5)
CH_2(14) or H–C(14)	1.29–1.35 (*m*), 1.82 (*qt*, *J* = 14, 4)	6.18 (*d*, *J* = 9.8)	3.62 (*d*, *J* = 4)	2.25 (*dd*, *J* = 15.5, 8.7), 2.96 (*dd*, *J* = 15.5, 3.3)	1.79 (*dt*, *J* = 15, 3), 2.13 (*dt*, *J* = 15, 3)
CH_2(15) or H–C(15)	1.05 (*td*, *J* = 14, 4), 2.07–2.12 (*m*)	6.11 (*d*, *J* = 9.8)	3.30 (*d*, *J* = 4)	3.77 (*d*, *J* = 8.7)	3.38 (*t*, *J* = 3)
H–C(16)	2.70 (*d*, *J* = 7.5)	–	–	–	–
H–C(17) or CH_2(17)	4.98 (*dd*, *J* = 7.5, 2)	3.80 (*d*, *J* = 5)	4.07 (*d*, *J* = 5)	3.90 (*d*, *J* = 1.3)	3.74 (*d*, *J* = 11.5), 4.23 (*d*, *J* = 11.5)
CH_2(18) or Me(18)	1.23–1.30 (*m*), 1.86–1.95 (*m*)	1.60 (*ddd*, *J* = 13, 11, 8), 2.20 (br. *dd*, *J* = 13, 10.5)	1.57–1.66 (*m*), 1.86–1.92 (*m*)	1.48 (*ddd*, *J* = 13.5, 11, 8.2), 2.14 (*ddd*, *J* = 13.5, 11, 2)	1.71 (*dd*, *J* = 7, 2)
CH_2(19) or H–C(19)	0.89–0.99 (*m*), 1.86–1.95 (*m*)	1.38 (*ddd*, *J* = 12.5, 11, 1), 1.84 (*ddd*, *J* = 12.5, 10.5, 8)	1.57–1.66 (*m*), 2.21 (br. *t*, *J* = 10.5)	1.75 (*ddd*, *J* = 13.5, 11, 8.2), 1.82 (*ddd*, *J* = 13.5, 11, 2)	5.60 (br. *q*, *J* = 7)
H–C(21) or CH_2(21)	3.01 (*s*)	3.91 (br. *s*)	3.74 (br. *s*)	4.26 (*d*, *J* = 1.3)	3.29 (*d*, *J* = 13.5), 3.72 (br. *d*, *J* = 13.5)
MeO–C(12)	–	3.86 (*s*)	3.85 (*s*)	3.84 (*s*)	–
OH–C(16)	–	6.37 (*s*)	6.52 (*s*)	6.08 (*s*)	–
MeOOC–C(16)	3.80 (*s*)	3.78 (*s*)	3.78 (*s*)	3.81 (*s*)	3.76 (*s*)
OH–C(17)	–	3.13 (*d*, *J* = 5)	3.49 (*d*, *J* = 5)	–	–
AcO–C(17)	–	–	–	–	1.97 (*s*)
MeOOC–N	–	3.83 (*s*)	3.84 (*s*)	3.86 (*s*)	–
NH	3.74 (br. *s*)	–	–	–	4.30 (br. *s*)

[a]) Assignments based on COSY and HMQC.

Table 2. *^{13}C-NMR Data* (100 MHz, $CDCl_3$) *of Compounds* **2**, **4**, **6**, **7**, *and* **9**[a])[2]). δ in ppm, J in Hz.

	2	**4**	**6**	**7**	**9**
C(2)	66.7	75.2	75.1	75.1	104.0
C(3)	47.6	162.9	166.9	96.4	53.2
C(5)	50.5	43.3	43.9	67.9	68.0
C(6)	34.8	36.4	35.3	38.1	39.2
C(7)	57.2	58.2	59.0	56.6	55.6
C(8)	139.8	141.0	140.7	141.5	134.6
C(9)	121.4	113.6	114.1	112.6	119.5
C(10)	119.7	125.3	125.5	125.7	126.9
C(11)	126.6	112.1	112.6	113.0	128.8
C(12)	110.6	148.9	149.0	149.1	109.1
C(13)	148.7	127.6	128.4	127.5	148.2
C(14)	17.4	141.5	52.6	34.5	27.8
C(15)	30.7	128.1	56.9	64.4	31.4
C(16)	54.7	79.2	78.9	76.2	56.1
C(17)	68.4	79.3	79.9	77.5	67.4
C(18)	34.0	25.4	24.4	25.6	13.2
C(19)	26.7	25.3	24.4	20.3	123.4
C(20)	37.2	39.3	39.9	39.9	129.6
C(21)	67.6	63.3	58.8	77.1	49.5
MeO–C(12)	–	55.9	56.1	56.2	–
*Me*OOC–C(16)	52.0	51.8	52.4	52.5	51.6
MeOO*C*–C(16)	174.2	170.7	170.8	170.5	173.0
*Me*OOC–N	–	52.9	53.2	53.2	–
MeOO*C*–N	–	155.6	156.3	155.8	–
*Me*COO–C(17)	–	–	–	–	20.7
Me*C*OO–C(17)	–	–	–	–	169.8

[a]) Assignments based on COSY and HMQC.

signals remained essentially similar to those of **3**. As in **3**, the configuration of OH–C(17) of **2** was deduced to be α from the *W*-coupling observed between H–C(17) and H_α–C(19) and the NOEs observed for H–C(17)/H_β–C(5) and H_β–C(6), while the observed NOE for H–C(16)/H_β–C(18) (δ 1.27) established the configuration of the COOMe substituent at C(16) as β.

Kopsidarine[2]) (**4**) was isolated in minute amounts as a colorless oil. The UV spectrum showed absorption maxima at 216, 253, 280, and 291 nm (log ε 4.58, 4.04, 3.54, and 3.45, resp.), indicating the presence of a dihydroindole chromophore. The IR spectrum showed absorption bands at 3349, 1743, and 1659 (broad) cm^{-1} which were assigned to OH, ester, and carbamate or lactam functions, respectively. The EI-MS revealed a molecular ion at m/z 470, corresponding to the formula $C_{24}H_{26}N_2O_8$, differing from kopsingine (=(2α,3α,4β,5α,12*R*,19α)-6,7-didehydro-3,4-dihydroxy-17-methoxy-2,21-cycloaspidospermidine-1,3-dicarboxylic acid demethyl ester; **5**) by 14 mass units, and suggesting an oxo derivative of kopsingine. The presence of a kopsingine-type structure, *i.e.*, of a 3-oxokopsingine[2]) structure, was further confirmed by the ^{1}H- and ^{13}C-NMR data (*Tables 1* and *2*). Kopsidarine (**4**) has been previously prepared from kopsingine (**5**) in connection with a study on the electrochemical

oxidation of aspidofractinine derivatives [31]; however, **4** was now isolated for the first time as a natural product.

The ^{13}C-NMR spectrum of **4** showed the presence of a conjugated lactam carbonyl function at δ 162.9, in addition to the carbamate and ester carbonyl absorptions at δ 155.6 and 170.7, resp. The ^{1}H-NMR spectrum was generally identical to that of **5** [29], except for the signals due to CH_2(3) which were absent, indicating the location of the lactam carbonyl group at C(3). This was further confirmed from the substantial downfield shift of the olefinic H–C(15) resonance to δ 6.11, which is characteristic of the β-positioned H-atom of an α,β-unsaturated carbonyl moiety. The observation of *W*-coupling between H_α–C(17) and H–C(21) in **4** confirmed that the configuration of OH–C(17) is β.

Kopsimaline F[2]) (**6**) had UV and IR spectra, which were generally similar to those of kopsidarine (**4**). The FAB-MS showed the $[M+H]^+$ peak at *m*/*z* 487, corresponding to $C_{24}H_{26}N_2O_9$ and differing from **4** by the addition of an O-atom. The ^{1}H-NMR spectrum (*Table 1*) indicated that **6** is similar to **4**, except for the absence of the C(14)=C(15) moiety, which was replaced by a CH–O–CH epoxy unit (*AB d* at δ 3.62 and 3.30). The ^{13}C-NMR data (*Table 2*) further confirmed the presence of an epoxy unit (C(14) and C(15) at δ 52.6 and 56.9). The configuration of the epoxy group was deduced to be α (NOE H–C(15)/H_β–C(17)). Compound **6** is, therefore, (14α,15α)-14,15-epoxy-14,15-dihydro-3-oxokopsingine[2]).

Kopsidine C *N*-oxide[2]) (**7**) was obtained as a light yellowish oil. The FAB-MS of **7** showed an $[M+H]^+$ at *m*/*z* 489, corresponding to the molecular formula $C_{24}H_{28}N_2O_9$, 16 mass units higher than that of kopsidine C (**8**). The UV and IR spectra were identical to those of **8**. Examination of the ^{1}H-NMR spectrum (*Table 1*) showed downfield shifts for H–C(5), H–C(9), and H–C(21), while the ^{13}C-NMR data (*Table 2*) showed downfield shifts involving C(5) and C(21), when compared to **8** [24][27]. These features are characteristic of *N*-oxides, and this structural feature was demonstrated by conversion of kopsidine C into **7** by treatment with 3-chloroperbenzoic acid. Compound **7** is, therefore, kopsidine C *N*-oxide[2]).

Aspidophylline B[2]) (**9**) was obtained from the bark extract as a yellowish oil. The UV spectrum showed absorption maxima at 205, 242, and 298 nm (log ε 4.52, 3.89, and 3.54, resp.), indicating the presence of a dihydroindole chromophore. The EI-MS of **9** showed a molecular ion at *m*/*z* 412, which corresponded to $C_{23}H_{28}N_2O_5$. Other significant fragments were observed at *m*/*z* 369 ($[M-COMe]^+$), 353 ($[M-OCOMe]^+$), and 108 ($C_7H_{10}N^+$). The ^{1}H-NMR spectrum of **9** (*Table 1*) was generally similar to that of aspidodasycarpine (**10**), which was also obtained, except for the replacement of the CH_2OH unit at C(16) of **10** by a CH_2OAc unit at C(16) of **9**. Thus, the structure of **9** was assigned as 17-*O*-acetylaspidodasycarpine[2]).

The ^{13}C-NMR spectrum (*Table 2*) of **9** gave a total of 23 C-resonances, in agreement with the molecular formula. The ^{1}H-NMR spectrum showed the presence of four aromatic H-atoms from δ 6.59 to 7.50, an NH group at δ 4.30, a COOMe group at δ 3.76, an Ac group at δ 1.97, and an ethylidene side chain (Me at δ 1.71 and olef. H at δ 5.60). The ^{1}H-NMR spectrum also showed the presence of a pair of *AB d* at δ 4.23 and 3.74 due to CH_2(17), and two broad *d* at δ 3.72 and 3.29 due to CH_2(21). The presence of the Ac group was further confirmed by the C resonances at δ 20.7 and 169.8 in the ^{13}C-NMR spectrum. The Me signal of the Ac group were observed upfield (δ(H) 1.97) indicating anisotropic interaction from the benzene ring, and confirming the configuration at C(16), which places the CH_2OAc group above the aromatic ring.

A comparison of the present results with previous results, based on a sample (A) collected in the same location but at a different time of the year [28], does show a variation in the alkaloidal composition. While a number of alkaloids occur in common, others, which were present in the previous sample (A) [28], were absent in the present sample (B) and *vice versa*. Notable differences include the following: the predominance of the kopsiloscine alkaloids in sample A but not in sample B, the presence of vincophylline (the first example of a vincorine alkaloid in *Kopsia*), leuconolam, and rhazinal in sample A only, and the presence of kopsidarine (**4**), kopsimaline F (**6**), kopsiloscine G (**2**), and the cage kopsinitarines and mersingines in sample B but not in sample A.

K. singapurensis also presents another interesting feature in respect of the variation of alkaloid types within the species, in addition to the seasonal dependence mentioned above. The predominant alkaloid skeleton in the two studies mentioned is that of aspidofractinine (=(2α,5α,12*R*,19α)-2,21-cycloaspidospermidine; *e.g.* kopsingine (**5**)), which was also the case in the previous studies of *Thomas et al.* [25] and *Sevenet* and coworkers [26]. In two other recent studies, however, both involved samples, which were initially identified as *K. fruticosa* but subsequently amended to *K. singapurensis* by *Middleton* [3], revealed that the dominant alkaloid skeletons were very different, being mersinine in one sample (*e.g.*, mersinine A = (5*R*,5a*R*,7a*R*,11a*R*,13a*R*)-5,5a,6,7,12,13-hexahydro-5-hydroxy-4*H*,10*H*-1,3-benzodioxolo[4,5-*k*]pyrrolo[3,2,1-*mn*][1,8]phenanthroline-4,5,7a(11a*H*)-tricarboxylic acid trimethyl ester) [15][32–34], and kopsifoline (*e.g.*, kopsifoline A = (3β,5α,12*R*,19α)-6,7-didehydro-2-hydroxy-17-methoxy-3,21-cycloaspidospermidine-3-carboxylic acid methyl ester) in the other [13][14]. Furthermore, both samples are characterized by the absence of kopsingine (**5**) [13][34], which was the major alkaloid present in previous studies of *K. singapurensis*, including samples A and B [25][28]. There appears, therefore, to be three morphologically similar *K. singapurensis*, which are nevertheless distinguishable by the occurrence of different dominant alkaloid-structure types, *viz.*, of aspidofractinine-, mersinine-, or kopsifoline-type alkaloids.

We thank the University of Malaya and *MOSTI*, Malaysia (ScienceFund), for financial support.

Experimental Part

General. Optical rotations: *Jasco DIP-370* digital polarimeter or *Atago-Polax-D* polarimeter. UV Spectra: EtOH solns.; *Shimadzu UV-3101PC* spectrophotometer; λ_{max} in nm (log ε). IR Spectra: *Perkin-Elmer RX1*-FT-IR spectrophotometer; in cm^{-1}. ^1H- and ^{13}C-NMR Spectra: $CDCl_3$ solns. *Jeol JNM-LA 400* spectrometer at 400 and 100 MHz, resp.; δ in ppm rel. to $SiMe_4$, *J* in Hz. MS Measurements were obtained by courtesy of Dr. *Komiyama* of the Kitasato Institute, Tokyo, Japan, and at Organic Mass Spectrometry, Central Science Laboratory, University of Tasmania, Tasmania, Australia.

Plant Material. Plant material was collected in Pahang, Malaysia (November, 1996), and identification was confirmed by Dr. *David Middleton*, Herbarium, Royal Botanic Garden, Edinburgh, 20A Inverleith Row, EH3 5LR Scotland. Herbarium voucher specimens (K 642) are deposited at the Herbarium, University of Malaya, Kuala Lumpur, Malaysia, and at the Rijksherbarium, University of Leiden, Leiden, The Netherlands.

Extraction and Isolation. Extraction of the leaf and stem-bark material was carried out in the usual manner by partitioning the concentrated EtOH extracts with dilute acid, as described in detail elsewhere [35]. The alkaloids were isolated by initial column chromatography (silica gel, $CHCl_3$ with increasing

proportions of MeOH), followed by rechromatography of the appropriate partially resolved fractions by using centrifugal TLC. Solvent systems used for centrifugal TLC were: Et_2O/hexane, Et_2O/cyclohexane, AcOEt/hexane, AcOEt/cyclohexane, AcOEt, AcOEt/MeOH, Et_2O/MeOH, $CHCl_3$/MeOH, and NH_3-sat. $CHCl_3$. The yields (g kg^{-1}) of the alkaloids from the leaf extract were as follows: **4** (0.001), **6** (0.003), **7** (0.005), kopsingine (**5**; 2.65), kopsinganol (0.097), kopsidine A (0.012), kopsidine C (0.015), 16-epiakuammiline (0.005), 16-epideacetylakuammiline (0.009), akuammidine (0.009), kopsinitarine A (0.005), kopsinitarine B (0.002), and mersingine A (0.018). The yields (g kg^{-1}) of the alkaloids from the stem-bark extract were as follows: kopsiloscine C (**1**; 0.003), **2** (0.008), **9** (0.002), kopsinine (0.002), (17α)-17-hydroxy-Δ^{14}-kopsinine (**3**; 0.002), kopsingine (**5**; 0.70), kopsinganol (0.049), rhazinilam (0.007), 5,21-dihydrorhazinilam (0.001), 16-epiakuammiline (0.017), 16-epideacetylakuammiline (0.012), aspidodasycarpine (0.021), lonicerine (0.003), and akuammidine (0.007).

Kopsiloscine G (*= (17α)-17-Hydroxykopsinine = (2α,3β,4α,5α,12*R*,19α)-4-Hydroxy-2,21-cycloaspidospermidine-3-carboxylic Acid Methyl Ester*; **2**): Colorless oil. $[\alpha]_D = -29$ ($c = 0.33$, $CHCl_3$). UV: 203 (4.43), 246 (3.77), 294 (3.32). IR (dry film): 3432, 3350, 1728. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. EI-MS: 354 (100, M^+), 353 (12), 326 (23), 294 (9), 271 (13), 251 (9), 237 (7), 202 (32), 167 (10), 152 (23), 149 (15), 125 (58), 124 (58). HR-EI-MS: 354.1961 ($C_{21}H_{26}N_2O_3^+$; calc. 354.1943).

Kopsidarine (*= 3-Oxokopsingine = (2α,3α,4β,5α,12*R*,19α)-6,7-Didehydro-3,4-dihydroxy-17-methoxy-8-oxo-2,21-cycloaspidospermidine-1,3-dicarboxylic Acid Dimethyl Ester*; **4**): Colorless oil. $[\alpha]_D = -23$ ($c = 0.05$, $CHCl_3$). UV: 216 (4.58), 253 (4.04), 280 (3.54), 291 (3.45). IR (dry film): 3349, 1743, 1659. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. EI-MS: 470 (100, M^+), 441 (7), 411 (8), 383 (7), 353 (15), 319 (7), 260 (13), 232 (9), 244 (15), 231 (7), 149 (6), 93 (10). HR-FAB-MS: 471.1779 ($C_{24}H_{27}N_2O_8^+$; calc. 471.1769).

Kopsimaline F (*= (14α,15α)-14,15-Epoxy-14,15-dihydrokopsidarin = (2α,3α,4β,5α,6α,7α,12*R*,19α)-6,7-Epoxy-3,4-dihydroxy-17-methoxy-8-oxo-2,21-cycloaspidospermidine-1,3-dicarboxylic Acid Dimethyl Ester*; **6**): Colorless oil. $[\alpha]_D = +23$ ($c = 0.16$, $CHCl_3$). UV: 216 (4.52), 252 (3.95), 283 (3.36), 290 (3.32). IR (dry film): 3391, 1743, 1667. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. FAB-MS: 487 (100, $[M+H]^+$), 455 (7), 427 (9), 395 (5), 307 (22), 289 (12). HR-FAB-MS: 487.1722 ($C_{24}H_{27}N_2O_9^+$; calc. 487.1717).

Kopsidine C N-*Oxide* (*= (2α,3α,4β,5α,6α,8β,12*R*,19α)-4,8-Epoxy-3,6-dihydroxy-17-methoxy-2,21-cycloaspidospermidine-1,3-dicarboxylic Acid Dimethyl Ester 1-Oxide*; **7**): Light yellowish oil. $[\alpha]_D = -7$ ($c = 0.37$, $CHCl_3$). UV: 216 (4.60), 254 (4.09), 285 (3.60), 292 (3.56). IR (dry film): 3355, 1743, 1672. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. FAB-MS: 489 (100, $[M+H]^+$), 470 (8), 452 (5), 329 (11), 307 (15), 289 (8), 176 (27), 154 (56), 136 (35), 107 (11). HR-FAB-MS: 489.1888 ($C_{24}H_{29}N_2O_9^+$; calc. 489.1873).

Aspidophylline B (*= (1*S*,4*E*,5*S*,6*R*,6a*S*,11a*R*)-6-[(Acetyloxy)methyl]-4-ethylidene-1,2,3,4,5,6-hexahydro-11α,6α-(epoxyethano)-1,5-methano-11*H*-azocino[3,4-*b*]indole-6-carboxylic Acid Methyl Ester*; **9**): Light yellowish oil. $[\alpha]_D = -16$ ($c = 0.06$, $CHCl_3$). UV: 205 (4.52), 242 (3.89), 298 (3.54). ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. EI-MS: 412 (100, M^+), 369 (6), 353 (12), 305 (10), 267 (19), 232 (9), 194 (11), 156 (11), 144 (10), 122 (12), 108 (100). HR-EI-MS: 412.1997 ($C_{23}H_{28}N_2O_5^+$; calc. 412.1998).

REFERENCES

[1] F. Markgraf, *Blumea* **1972**, *20*, 416.
[2] T. Sevenet, L. Allorge, B. David, K. Awang, A. H. A. Hadi, C. Kan-Fan, J. C. Quirion, F. Remy, H. Schaller, L. E. Teo, *J. Ethnopharmacol.* **1994**, *41*, 147.
[3] D. J. Middleton, *Harvard Pap. Bot.* **2004**, *9*, 89.
[4] T. S. Kam, in 'Alkaloids: Chemical and Biological Perspectives', Ed. S. W. Pelletier, Pergamon, Amsterdam, 1999, Vol. 14, Chap. 2, p. 285–435.
[5] T. S. Kam, Y. M. Choo, in 'The Alkaloids', Ed. G. A. Cordell, Academic Press, Amsterdam, 2006, Vol. 63, p. 181–337.
[6] K. H. Lim, K. Komiyama, T. S. Kam, *Tetrahedron Lett.* **2007**, *48*, 1143.
[7] K. H. Lim, T. S. Kam, *Helv. Chim. Acta* **2007**, *90*, 31.
[8] K. H. Lim, T. S. Kam, *Tetrahedron Lett.* **2006**, *47*, 8653.
[9] K. H. Lim, T. S. Kam, *Org. Lett.* **2006**, *8*, 1733.

[10] K. H. Lim, Y. Y. Low, T. S. Kam, *Tetrahedron Lett.* **2006**, *47*, 5037.
[11] T. S. Kam, G. Subramaniam, K. H. Lim, Y. M. Choo, *Tetrahedron Lett.* **2004**, *45*, 5995.
[12] T. S. Kam, G. Subramaniam, *Tetrahedron Lett.* **2004**, *45*, 3521.
[13] T. S. Kam, Y. M. Choo, *Helv. Chim. Acta* **2004**, *87*, 991.
[14] T. S. Kam, Y. M. Choo, *Tetrahedron Lett.* **2003**, *44*, 1317.
[15] T. S. Kam, G. Subramaniam, T. M. Lim, *Tetrahedron Lett.* **2001**, *42*, 5977.
[16] T. S. Kam, T. M. Lim, Y. M. Choo, *Tetrahedron* **1999**, *55*, 1457.
[17] T. S. Kam, G. Subramaniam, K. M. Sim, K. Yoganathan, T. Koyano, M. Toyoshima, M. C. Rho, M. Hayashi, K. Komiyama, *Bioorg. Med. Chem. Lett*. **1998**, *8*, 2769.
[18] T. S. Kam, K. Yoganathan, W. Chen, *J. Nat. Prod*. **1997**, *60*, 673.
[19] T. S. Kam, K. Yoganathan, W. Chen, *Tetrahedron Lett.* **1996**, *37*, 3603.
[20] T. S. Kam, K. Yoganathan, T. Koyano, K. Komiyama, *Tetrahedron Lett.* **1996**, *37*, 5765.
[21] T. S. Kam, K. Yoganathan, W. Chen, *J. Nat. Prod*. **1996**, *59*, 1109.
[22] T. S. Kam, K. Yoganathan, C. H. Chuah, *Tetrahedron Lett.* **1995**, *36*, 759.
[23] T. S. Kam, K. Yoganathan, C. H. Chuah, *Tetrahedron Lett.* **1994**, *35*, 4457.
[24] T. S. Kam, K. Yoganathan, C. H. Chuah, *Tetrahedron Lett.* **1993**, *34*, 1819.
[25] D. W. Thomas, K. Biemann, A. K. Kiang, R. D. Amarasingham, *J. Am. Chem. Soc.* **1967**, *89*, 3235.
[26] K. Awang, O. Thoison, A. H. A. Hadi, M. Pais, T. Sevenet, *Nat. Prod. Lett.* **1993**, *3*, 283.
[27] T. S. Kam, K. Yoganathan, C. H. Chuah, *Phytochemistry* **1997**, *45*, 623.
[28] G. Subramaniam, O. Hiraku, M. Hayashi, T. Koyano, K. Komiyama, T. S. Kam, *J. Nat. Prod.* **2008**, *71*, 53.
[29] T. S. Kam, T. S. K. Yoganathan, C. H. Chuah, W. Chen, *Phytochemistry* **1993**, *32*, 1343.
[30] T. S. Kam, T. M. Lim, G. Subramaniam, Y. M. Tee, K. Yoganathan, *Phytochemistry* **1999**, *50*, 171.
[31] T. S. Kam, T. M. Lim, G. H. Tan, *J. Chem. Soc., Perkin Trans. 1* **2001**, 1594.
[32] G. Subramaniam, Y. M. Choo, O. Hiraku, K. Komiyama, T. S. Kam, *Tetrahedron* **2008**, *64*, 1397.
[33] G. Subramaniam, T. S. Kam, *Tetrahedron Lett*. **2007**, *48*, 6677.
[34] G. Subramaniam, O. Hiraku, M. Hayashi, T. Koyano, K. Komiyama, T. S. Kam, *J. Nat. Prod.* **2007**, *70*, 1783.
[35] T. S. Kam, P. S. Tan, *Phytochemistry* **1990**, *29*, 2321.

Received October 5, 2007