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PHOEBEGRANDINES A AND B, PROAPORPHINE-TRYPTAMINE DIMERS, FROM *PHOEBE GRANDIS*

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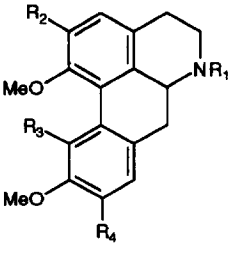
Key Word Index—*Phoebe grandis*; Lauraceae; aporphine alkaloids; proaporphine-tryptamine dimer alkaloids.

Abstract—Four known aporphine alkaloids, boldine, norboldine, laurotetanine and lindecarpine were isolated from the bark of *Phoebe grandis*. The leaves yielded two new alkaloids belonging to the proaporphine-tryptamine dimers series, phoebegrandines A and B. The structure of the new compounds was elucidated by spectral methods.

INTRODUCTION

In the course of our research on Malaysian plants,§ we have investigated the alkaloid extracts of the bark and leaves of *Phoebe grandis* (Nees) Merr. It is a tree of 20 m high with yellowish brown flowers. The plant material was collected from Sik, Kedah in the Northern part of Peninsula Malaya. The bark afforded four known aporphine alkaloids boldine (**1**), norboldine (**2**), lauretanine (**3**) and lindcarpine (**4**). The leaves, however, yielded exclusively two new alkaloids, phoebegrandine A (**5**) and B (**6**) of the rare proaporphine-tryptamine type. Such alkaloids have been found previously only in the *Roemeria hybrida* species (Papaveraceae) [1–3]. Structural elucidation was done mainly by 2D NMR.



	R_1	R_2	R_3	R_4
1	Me	OH	H	OH
2	H	OH	H	OH
3	H	OMe	H	OH
4	H	OH	OH	H

RESULTS AND DISCUSSION

The alkaloids were extracted using conventional methods. The bark alkaloids (**1–4**), as well as phoebegrandines A (**5**) and B (**6**), were separated by column chromatography on silica gel.

Phoebegrandine A (**5**), $[\alpha]_D$ 0°, showed a molecular ion peak in the HREI mass spectrum at *m/z* 473.2715 (Δ 3.65 mmu) corresponding to the molecular formula $C_{29}H_{35}N_3O_3$. The UV spectrum exhibited two maxima at 224 and 277 nm which revealed an indole chromophore. The high M_r indicated that compound **5** is probably a dimeric alkaloid. An intense peak was observed in the mass spectrum at *m/z* 214 (214.1103, $C_{13}H_{14}N_2O$, Δ −0.28 mmu), suggesting a β-carboline type moiety (ion **7**). The ^{1}H NMR showed the signals of two aromatic methoxy groups at δ 3.80 and 3.84 with one belonging to the β-carboline part. The presence of three aromatic protons: two doublets at δ 7.20 and δ 6.93 (J = 8.6 and 1.5 Hz, respectively) and one double doublet at δ 6.78 (J = 8.6 and 1.5 Hz) indicated that the aromatic ring of the β-carboline was methoxylated either at C-3′ or C-4′. A broad exchangeable proton at δ 7.75 was assignable to the NH-1′ and a spin system of two multiplets at δ 2.70 and δ 3.18 was attributed to the CH_2-6′ and CH_2-7′, respectively. In the ^{13}C NMR, the typical signals of the aromatic

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Table 1. ^{13}C (62.5 MHz) and ^{1}H (400 MHz) NMR data[a] for phoebegrandines A (**5**) and B (**6**)

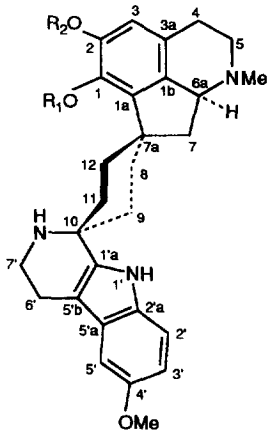
	5[b]				**6**		
Positon	δ C[c]	δ H (*J* Hz)	HMBC	NOESY	δ C[c]	δ H (*J* Hz)[b]	HMBC[c]	NOESY[b]
1	140.5				144.4			
1a	132.5				141.1			
1b	133.4				132.7			
2	147.2				151.2			
3	108.4	6.48 *s*	1,1b,2,3a	4α, OMe-2	115.1	6.56 *s*	1,1b,2,4	4αβ
3a	121.9				127.8			
4	27.3	α 2.70 *m*	3a	4β, 5β	27.7	α 2.70 *m*	1b,3,3a	4β, 5α
		β 2.90 *m*	3a,5			β 3.00 *m*	1b,3,3a	
5	55.4	α 2.45 *dd* (12, 5)	6a	5β, 6a	56.0	α 2.45 *m*	6a	5β, 6a
		β 3.15 *m*	3a,4,6a			β 3.10 *m*		
6a	65.8	3.22 *m*	1a	7α, 8e	66.6	3.23 *m*	1b	7α,8e,NMe
7	42.7	α 2.80 *dd* (11, 6.5)	1a,6a,7a,8	7β,9,11	43.1	α 2.80 *m*	3a,6a,7a,8	7β,9,11
		β 1.55 *dd* (11, 8.9)	6a,7a,8,12	12e		β 1.50 *dd* (10.5, 11)	6a,7a,8,12	
7a	47.1				48.0			
8	27.9	e 1.60 *br d* (13.5)		8ax,9	30.0	e 1.60 *br d* (13.5)		8ax, 9
		ax 2.25 *m*	7,7a,9			ax 2.25 *m*		9,OMe-1
9	33.4	1.95 *m*	7a,8,10,11	1′	33.6	1.95 *m*		1′
10	52.0				54.2			
11	33.9	1.85 *m*		1′, 12e,ax	33.9	1.85 *m*	7a	1′
12	31.0	e 1.38 *br d* (13.8)	7,8a,11	12ax	32.9	e 1.40 *br d* (12)		12ax
		ax 3.10 *m*				ax 3.05 *m*	1a,1b	OMe-1
1′a	141.8				141.2			
2′	111.4	7.20 *d* (8.6)	4′,5′a	1′,3′	112.4	7.21 *d* (8.6)	4′,5′a	
2′a	130.8				132.9			
3′	115.5	6.78 *dd* (8.6, 1.5)	2′a,4′,5′	OMe-4′	111.9	6.79 *dd* (8.6, 1.5)	2′a,4′,5′	
4′	154.2				155.0			
5′	100.5	6.93 *d* (1.5)	2′a,3′,4′,5′b	6′, OMe-4′	101.2	6.93 *d* (1.5)	2′a,3′,4′,5′	6′, OMe-4′
5′a	128.1				128.1			
5′b	109.0				107.8			
6′	23.1	2.70 *m*	1′a,5′a,5′b,7′	7′	22.8	2.70 *m*		7′
7′	39.4	3.18 *m*	5′b,6′,10		40.4	3.20 *m*	10, 6′,5′b	
OMe-1					61.3	3.91 *s*	1	
OMe-2	56.2	3.80 *s*	2					
OMe-4′	56.6	3.84 *s*	4′		56.3	3.84 *s*	4′	
NMe	43.8	2.42 *s*	5,6a		43.3	2.42 *s*	5,6a	
NH-1′		7.74 *br s*				7.67 *br s*		

[a] Assignments based on 2D experiments.
[b] In $CDCl_3$.
[c] In CD_3OD.

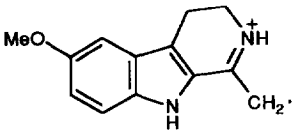
quaternary carbons of the β-carboline skeleton [4] were observed (Table 1). The HMBC experiment (Table 1) showed that the methoxy was located at C-4′ and confirmed the assignments of all the carbons.

In addition, the presence of seven methylene peaks between δ 27.3 and δ 55.4 in the ^{13}C NMR was reminiscent of a proaporphine skeleton. Ring A contained an OH, a methoxy and one aromatic proton (δ 6.48, *s*). The ring B nitrogen was methylated (NMe group at δ 2.42). The 2D NMR spectra showed that ring D contained four methylene groups and was attached to the β-carboline moiety by C-10 (δ 52.0) as in previously reported known proaporphine-tryptamine dimers. Four spin systems were observed in the COSY experiment: CH_2-4 (δ 2.90 and 2.70, 2*m*) and CH_2-5 (δ 3.15 and 2.45, 2*m*); CH-6a (δ 3.22, *m*) and CH_2-7 (δ 3.80 and 1.55, 2*m*); 3. CH_2-8 (δ 2.25 and 1.60, 2*m*) and CH_2-9 (δ 1.95 *m*); CH_2-11 (δ 1.85, *m*) and CH_2-12 (δ 3.10 and 1.38, 2*m*). The HMBC spectrum (Table 1) verified the relative position of the substituents of the aromatic ring and further supported the connectivities and the assignments of all the carbons of the proaporphine moiety.

The relative configuration at C-6a, C-7a and C-10 of alkaloid **1** was deduced from the NOESY spectrum (Table 1). The cross peaks 6a/8e, 6a/7α and 7β/12e established the relative stereochemistry between C-6a and C-7a as shown. The spectrum further exhibited correlations between NH-1′ and the protons of both CH_2-9 and CH_2-11. H-11e and H-11ax were superimposed, as well as H-9e and H-9ax. However, the Dreiding models show that the correlations were with H-9ax and H-11ax and could be observed only if NH-1′ lies on the same side of ring D as the aporphine nitrogen, that is the alkaloid belongs to the *syn* series [2].



5 $R_1 = H, R_2 = Me$
6 $R_1 = Me, R_2 = H$



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Phoebegrandine B (**6**), $[\alpha]_D$ 0°, exhibited a M^+ peak in the HREI mass spectrum at *m/z* 473.2663 (Δ −1.57 mmu) corresponding to the molecular formula $C_{29}H_{35}N_3O_3$, which is isomeric to alkaloid **5**. The base peak *m/z* 214 (214.1087, $C_{13}H_{14}N_2O$, Δ −1.95 mmu) indicated that the β-carboline moiety was monomethoxylated. The UV, 1D and 2D NMR spectra were very close to those of compound **5**. However, significant differences were observed in the chemical shifts of the aporphine methoxy and aromatic signals (Table 1). The HMBC spectrum revealed clearly that the methoxy was attached to C-1 instead of C-2 as in phoebegrandine A, while the OH was at C-2.

The NOESY spectrum (Table 1) indicated that the stereochemistry of phoebegrandine (**6**) was similar to the one of alkaloid **5**. The correlations 6a/8e and 6a/7a established the stereochemistry of C-6a and C-7a, which was further confirmed by the cross peaks OMe-1/8ax and OMe-1/12ax. Alkaloid **6** also belonged to the *syn* series as shown by the correlations NH-1′/H-9 and NH-1′/H-11.

The absolute configuration of the dimeric alkaloids of *Roemeria hybrida* was determined by the fact that all aporphine alkaloids isolated from the same species possess an *S* configuration at C-6a [1]. The configuration shown for the alkaloids **5** and **6** is only relative. Both compounds appear as racemates, since no optical rotation and no CD curve were observed.

To our knowledge, *Phoebe grandis* and *Roemeria hybrida* are the sole plants containing proaporphine-tryptamine dimers. The substitution pattern of the aromatic rings of all those alkaloids are very similar. However two methoxy groups were found in the aromatic ring of the tryptamine part of some *Roemeria* alkaloids. The main difference between the dimers of these two species is that phoebegrandines A and B lack the aliphatic methoxy group, which is present in ring D of all *Roemeria* alkaloids.

EXPERIMENTAL

General. UV: MeOH; ^1H NMR: 250 or 400 MHz; ^{13}C NMR: 62.5 MHz; 2D experiments: 400 MHz; CC: Merck silica gel H 60.

Plant material. Bark and leaves of *Phoebe grandis* (Nees) Merr. (Lauraceae) were collected at Sik, Kedah (1994) by G. Perromat (Institut de Chimie des Substances Naturelles, CNRS, Gif sur Yvette). Identification was made by Dr K. M. Kochummen (Forest Research Institute of Malaysia, Kepong, Malaysia). Voucher specimens (KL 4318) are deposited at the Laboratoire de Phanérogamie, Muséum National d'Historie Naturelle in Paris, at the Herbarium of Department of Chemistry, University of Malaya, Kuala Lumpur, Malaysia and at the Herbarium of the Forest Research Institute, Kepong, Malaysia.

Extraction and isolation of the alkaloids. The alkaloids were extracted by the classical method after alkalinisation of the plant material. A total of 11.5 g of crude alkaloids was obtained from the bark (1 kg). The crude product underwent column chromatography on silica gel with CH_2Cl_2 containing increasing amounts of MeOH. The alkaloids purified were boldine (**1**) (15 mg after subsequent purification by prep. TLC), norboldine (**2**) (20 mg), laurotetanine (**3**) (4 mg) and lindcarpine (**4**) (3.2 mg). The crude alkaloids (2.5 g) from the leaves (500 g) were purified by a similar column chromatography yielding phoebegrandine B (**6**) (15 mg) CH_2Cl_2–MeOH, 49:1 and phoebegrandine A (**5**) (12 mg) CH_2Cl_2–MeOH, 24:1. Identification of the known compounds was made by comparison of their spectral data with lit. data [5].

Phoebegrandine A (**5**). Amorphous gum, $[\alpha]_D$ 0° ($CHCl_3$, *c* 0.5) UV λ_{max} nm (log ε) 224 (4.55), 277 (3.97), 308 sh (3.51). EIMS: *m/z* (rel. int.) 473 $[M]^+$ (23), 230 (88), 229 (100), 227 (87), 214 (81). ^1H and ^{13}C NMR, see Table 1.

Phoebegrandine B (**6**). Amorphous gum, $[\alpha]_D$ 0°

($CHCl_3$, *c* 0.5) UV λ_{max} nm (log ε) 225 (4.55), 278 (3.97), 308 sh (3.51). EIMS: *m/z* (rel. int.) 473 $[M]^+$ (42), 230 (42), 227 (40), 214 (100). ^1H and ^{13}C NMR, see Table 1. Since the alkaloid was not very soluble in $CDCl_3$ the HMQC and HMBC spectra were run in CD_3OD; ^1H NMR (CD_3OD): δ 6.54 (*s*, H-3), 2.77 (*m*, H-4α), 2.95 (*m*, H-4β), 2.56 (*m*, H-5α), 3.18 (*m*, H-5β), 3.38 (*m*, H-6a), 3.16 (*m*, H-7α), 1.55 (*m*, H-7β), 1.69 (*br d*, J = 13.5 Hz, H-8e), 2.10 (*m*, H-8ax), 2.10, 2.20 (*m*, H-9), 1.90, 2.15 (*m*, H-11), 1.53 (*m*, H-12e), 2.90 (H-12ax), 7.20 (*d*, J = 8.6 Hz, H-2′), 6.74 (*dd*, J = 8.6 and 1.5 Hz, H-3′), 6.92 (*d*, J = 1.5 Hz, H-5′), 2.80 (*m*, H-6′), 3.30 (*m*, H-7′), 3.81 (*s*, OMe-1), 3.88 (*s*, OMe-4′), 2.48 (*s*, NMe).

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