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Cytotoxic Acylphenols from *Myristica maingayi*

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Abstract—From the cytotoxic AcOEt extract of the fruits of *Myristica maingayi*, a Myristicaceae, five new acylphenols (promalabaricones B and C, maingayic acids B and C, and maingayone) were isolated, together with the known malabaricones A–C. The structures were determined from spectral analysis, including mass spectrometry and 2D NMR. The cytotoxicity of the new compounds and that of malabaricones was assessed against KB cells. A biosynthetic pathway for malabaricones, via the corresponding promalabaricones as precursors, is suggested.

Introduction

Myristica maingayi is a tropical tree of the nutmeg family (Myristicaceae) occurring in Malaysia and Vietnam. The family is known to produce lignans and acylphenols mainly of the 2,6-dihydroxyphenyl type, such as malabaricones A–C from *M. malabarica* and *M. fragans*.[1–6] This aromatic substitution type is less common than the 3,5-dihydroxyphenyl type which is widespread in Anacardiaceae, Gingkoaceae and Proteaceae.[7,8] In addition to their antimicrobial properties, malabaricones have been recently shown to have nematocide actitivity.[9]

We have previously reported the isolation from the Myristicaceae species *Knema furfuracea*, of cytotoxic and antimicrobial acylphenols, knerachelins A and B,[5] which are diarylpentanoids. In continuation of our search for cytotoxic compounds from Myristicaceae,[5,10] we have investigated *M. maingayi* and observed for the AcOEt extract of the fruits a cytotoxic activity on human nasopharynx carcinoma KB cells (IC_{50} 23 μg ml^{-1}) and murine leukemia P 388 cells (100% inhibition at 10 μg ml^{-1}).

We now describe the bioguided isolation from *M. maingayi* and the structure of two new 2-acylcyclohexane-1,3-diones, named promalabaricones B (**1**) and C (**2**), of two structurally related arylnonanoic acids, maingayic acids (**3**, **4**) and of a diarylnonanoid dimer termed maingayone (**8**), along with known malabaricones A (**5**), B (**6**) and C (**7**). The bioactivities are discussed and a biosynthetic relationship between the isolated compounds is suggested.

Keywords: cytotoxicity; diarylnonanoids; promalabaricones B and C; maingayic acids B and C; maingayone; Myristicaceae.

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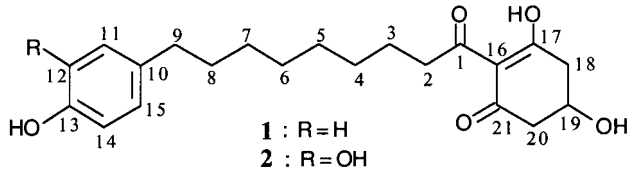
Results and Discussion

The EtOAc extract of the dried fruits of *M. maingayi* was chromatographed on silica gel to yield bioactive fractions from which were separated, by a combination of chromatographic procedures, five new compounds, **1**–**4** and **8**, along with malabaricones A–C (**5**–**7**).



Promalabaricone B (**1**) was obtained as a microcrystalline solid, without optical activity. Its HR FAB mass spectrum showed the $[M+H]^+$ ion at *m/z* 361.2024 leading to the molecular formula $C_{21}H_{28}O_5$. The IR spectrum had strong absorptions for hydroxyls at 3418, methylenes at 2922 and 2852 and carbonyls at 1722 and 1640 cm^{-1}.

The ^1H NMR (Table 1), ^1H–^1H and ^1H–^{13}C COSY spectra (CD_3OD) depicted characteristic spin systems for a 1,4-disubstituted benzene ring and for ten methylenes, eight of them forming together an octyl linear chain [–$(CH_2)_8$–]: four methylenes giving signals between δ_H 1.50 and 3.00 ppm and four methylenes at δ_H 1.29 and 1.32 in a broad singlet. A methinoxy proton at δ_H 4.27, vicinal to the two remaining methylenes, formed with them a symmetrical AA′BB′X spin system (–CH_2–CH(OH)– CH_2–); the chemical shift of these latter methylenes at δ_H 2.63 and 2.87 suggested they were vicinal to a carbonyl (Table 1). The *J*-modulated ^{13}C NMR spectrum displayed only 15 signals, which suggested symmetrical moieties in the molecule. In addition to the *p*-disubstituted benzene ring

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Table 1. ^{13}C and ^{1}H NMR data for promalabaricones B (**1**) and C (**2**) (^{1}H: 400.13 MHz, ^{13}C: 100.61 MHz CD_3OD; carbon numbering does not follow the official rule and is used here for simplicity)

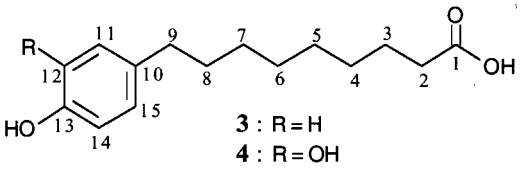
	1			**2**		
C number	δ_C	δ_H	m (*J* (Hz))	δ_C	δ_H	m (*J* (Hz))
1	206.3	–		206.3	–	
2	41.5	2.96	t (7.5)	41.5	2.97	t (7.6)
3	25.8	1.60	m	25.9	1.60	m
4	30.4	1.32	m	30.4	1.33	m
5	30.4	1.32	m	30.4	1.33	m
6	30.4	1.32	m	30.4	1.33	m
7	30.2	1.29	m	30.2	1.30	m
8	33.0	1.55	m	32.9	1.55	m
9	36.0	2.49	t (7.6)	36.2	2.47	t (7.7)
10	134.9			135.7	–	
11	130.2	6.97	m (8.5)	116.5	6.60	d (2.0)
12	116.0	6.67	m (8.5)	145.8		
13	156.2	–		144.0	–	
14	116.0	6.67	m (8.5)	116.2	6.65	d (8.0)
15	130.2	6.97	m (8.5)	120.6	6.47	dd (8.0, 2.0)
16	114.1	–		114.0		
17	197.1	–		197.0		
18	47.6	2.63	dd (16.6, 7.6)	47.8	2.65	dd (16.5, 7.7)
		2.87	dd (16.6, 3.6)		2.85	dd (16.5, 3.8)
19	64.1	4.27	m	(64.1)	4.28	*m*
20	47.6	2.63	dd (16.6, 7.6)	47.8	2.65	dd (16.5, 7.7)
		2.87	dd 16.6, 3.6		2.85	dd (16.5, 3.8)
21	197.1				197.0	–

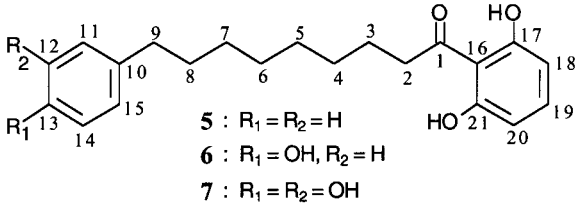
and the *n*-octyl chain, were depicted signals for CH_2 at δ_C 47.6, one CHOH at δ_C 64.1, one quaternary carbon at δ_C 114.1, one CO at δ_C 206.3 and two keto–enol carbons at δ_C 197.1. In the HMBC spectrum, the carbonyl at δ_C 206.3 was correlated to the methylene at δ_H 2.96, thus linking the octyl chain on one side. The aromatic quaternary carbon at δ_C 134.9 was, on one hand, ^{3}J-correlated to the aromatic protons at δ_H 6.67 vicinal to the phenolic hydroxyl and, on the other hand, to the methylenes at 2.49 and 1.55, thus ending the *n*-octyl chain. The carbons at δ_C 197.1 were correlated to the methinoxy proton at δ_H 4.27 and to the methylenes displaying non-equivalent protons at 2.63 and 2.87, leading to the symmetrical substructure –CO–CH_2–CH(OH)–CH_2–CO–. Finally, the quaternary carbon atom at δ_C 114.1 was correlated to both the methylene at δ_H 2.96 and those at δ_H 2.63 and 2.87: it was thus linked to the three carbonyls, leading for compound **1** to a 2-acylcyclohexane-1,3-dione structure. However, this cyclic β-triketonic structure is, as usual,[11] not observed and the presence of the quaternary carbon at δ_C 114.1 instead of a methine showed that the enolic tautomer (**1**) was predominant, inasmuch as the ^{1}H NMR spectrum recorded in DMSO-d_6 showed a hydroxyl signal at δ_H 18.10, indicating a very strong intramolecular hydrogen bonding. HMBC cross peaks of carbon at δ_C 197.1 indicated that enolization occurred into the ring, with formation of symmetrical enols. Broadening of this signal evidenced tautomeric equilibrium for the carbonyls in the ring.

As a result, signals for CO at 17 and 21 were identical, as well as signals for the methylenes at 18 and 20, producing symmetry of the cyclohexanedione ring.

Carbon atoms at 1, 16, 17 and 21 of structure **1**, are coplanar, leading for carbons at 18, 19 and 20, to an average conformation in which the substituents are eclipsed, in agreement with the *cis* (3.6 Hz) and *trans* (7.6 Hz) coupling constant values determined for the methinoxy H-19.[16] Promalabaricone B (**1**) is thus 3,5-dihydroxy-2-[9-(4-hydroxyphenyl)-nonanoyl]-cyclohexen-1-one, according to the official nomenclature.

Promalabaricone C (**2**) was also obtained as a microcrystalline solid from MeOH, without optical activity. Its HR CIMS showed the protonated molecular ion $[M+H]^+$ at *m/z* 377, corresponding to the molecular formula $C_{21}H_{28}O_6$, which differed from that of **1** only by an oxygen atom more. The ^{1}H and ^{13}C NMR spectra were similar to those of **1**, showing the same substructures, especially the *n*-octyl chain and the 1-acyl-4-hydroxycyclohexane-2,6-dione ring, but differed in the aromatic part because a 3,4-dihydroxyphenyl ring was observed instead of the 4-hydroxyphenyl one, as established from the observed aromatic spin system. The final structure and the ^{1}H and ^{13}C chemical shift assignments resulted from the HMBC spectrum correlations (Table 1). Thus, promalabaricone C (**2**) is 3,5-dihydroxy-2-[9-(3,4,-dihydroxyphenyl)nonanoyl]-cyclohexen-1-one.



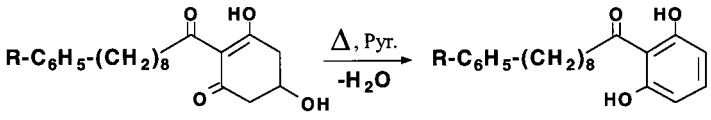


The IR spectra of **3** and **4** showed strong absorptions at 1705 cm^{-1}, suggesting they were carboxylic acids, in agreement with their ^{13}C NMR spectra which displayed carbonyl groups at δ_C 177.8 and 178.1, respectively. The HR CIMS indicated the molecular formulae $C_{15}H_{22}O_3$ for **3** and $C_{15}H_{22}O_4$ for **4**: the two compounds differed from each other by an oxygen atom. Their ^{1}H NMR spectra were similar in the aliphatic proton region, only showing spin systems characteristic of an Ar–$(CH_2)_8$–CO– substructure, but differed in the aromatic region: the NMR spectra indicated they have a 4-hydroxyphenyl ring and a 3,4-dihydroxyphenyl ring, respectively. The substitution of the aromatic rings and the final structures were confirmed by HMBC experiments and mass spectral fragmentations.

On these bases, the structures of 9-(4′-hydroxyphenyl)-nonanoic acid and 9-(3′,4′-dihydroxyphenyl)-nonanoic acid were assigned to **3** and **4**, respectively, which were named maingayic acids B and C.

Compounds **5**, **6** and **7** were identified from their mass spectral and NMR characteristics as malabaricones A, B and C, respectively.[1,9]

Confirmation of the structures of **1** and **2** arose from their complete transformation by heating in pyridine at 110°C for



Scheme 1.

Table 2. NMR data for maingayone (**8**) (^{1}H: 300.13 MHz, ^{13}C: 75.47 MHz; CD_3OD); a, b, c may be reversed within the same line

8 (substructure I)				**8** (substructure II)			
C number	δ_C	δ_H	m (*J* (Hz))	C number	δ_C	δ_H	m (*J* (Hz))
1	210.0	–		1′	34.7	4.54	dd (9.1, 6.8)
2	45.6	3.08	t (7.2)	2′	32.1	2.04, 2.41	m
3	25.6	1.50	m	3′	29.3	1.50	m
4	30.1	1.30	m	4′	30.1	1.30	m
5	30.1	1.30	m	5′	30.1	1.30	m
6	30.3	1.30	m	6′	30.3	1.30	m
7	30.3	1.30	m	7′	30.3	1.30	m
8	32.6	1.50	m	8′	32.6	1.50	m
9	36.0	2.42[a]	t (7.0)	9′	36.0	2.40[a]	t (7.0)
10	135.8	–		10′	135.8	–	
11	116.4	6.59	d (2.3)	11′	116.4	6.59	d (2.3)
12	145.5	–		12′	145.5	–	
13	143.5	–		13′	143.5	–	
14	116.1	6.64[b]	d (8.0)	14′	116.1	6.63[b]	d (8.0)
15	120.6	6.45[c]	dd (8.0, 2.3)	15′	120.6	6.43[c]	dd (8.0, 2.3)
16	110.8	–		16′	118.0	–	
17	160.2	–		17′	157.1	–	
18	107.8	6.30	d (8.5)	18′	108.7	6.28	d (8.0)
19	137.4	7.57	d (8.5)	19′	127.9	6.79	t (8.0)
20	123.3	–		20′	108.7	6.28	d (8.0)
21	160.4	–		21′	157.1	–	

30 min, into malabaricones B and C. The 3,5-dihydroxycyclohexen-1-one ring underwent a dehydration and subsequent aromatisation, due to the enolic tautomerisation to form the 2,6-dihydroxyphenyl ring of malabaricones (Scheme 1).

Maigayone (**8**) was obtained as a colourless, not crystallized powder, with a weak optical activity ($[\alpha]_D=+3°$, MeOH). Its IR spectrum indicated absorption bands of a conjugated carbonyl at 1702 cm^{-1} together with hydroxyl bands at 3385 cm^{-1} and its FAB MS showed the $[M+H]^+$ ion at *m/z* 701 in agreement with the molecular formula $C_{42}H_{52}O_9$. By acetylation with Ac_2O/Pyr, compound **8** gave an octa-acetyl derivative, as shown from its NMR spectrum where eight acetyl groups forming sharp singlets were observed between 2.07 and 2.23 ppm, and confirmed by its HR (+)FAB MS indicating the molecular formula $C_{58}H_{68}O_{17}$. The ^{1}H NMR spectrum, ^{1}H–^{1}H and ^{13}C–^{1}H COSY of **8** depicted four substituted benzene rings: two 1,3,4-trisubstituted (a and a′), one 1,2,3,4-tetrasubstituted (b) and one 1,2,3-trisubstituted (b′) showing two ortho protons at δ_H 6.30 and 7.57 (*J*=8.5 Hz); in the aliphatic region, signals for 16 methylenes were depicted (Table 2). The NMR spectra were similar to those of malabaricone C, with three main differences: (i) a tetrasubstituted benzene ring (b′) instead of a trisubstituted one; (ii) only one carbonyl at 210.0 ppm; and (iii) a methine signal at 4.54 ppm, strongly coupled with a non-equivalent methylene at 2.04 and 2.41 ppm and ending one polymethylene chain (Fig. 1).

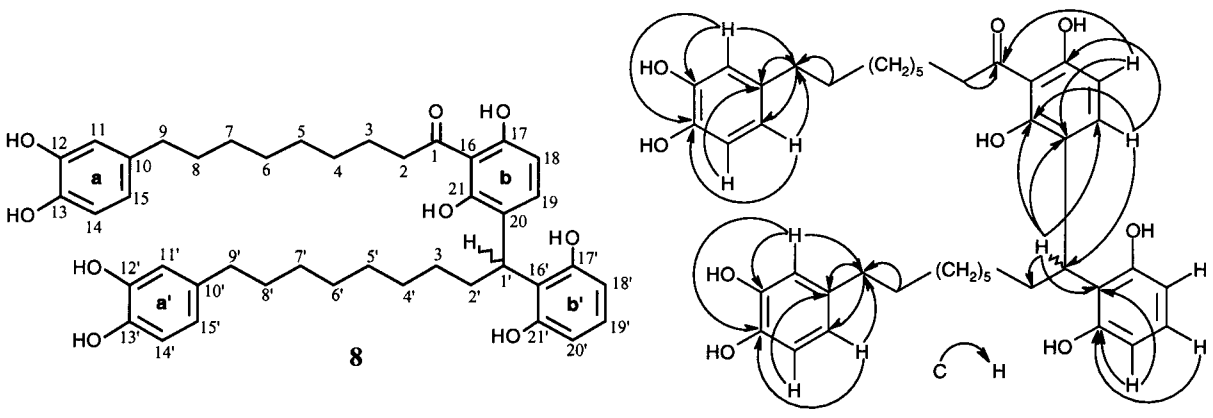


Figure 1. HMBC correlations of **8**.

In the HMBC spectrum, carbons at δ_C 135.8 ppm (C-10 and 10′ on a and a′ rings) were correlated with protons at δ_H 2.40 and 2.42 (9 and 9′) and carbons at 36.0 ppm (C-9 and 9′) with aromatic protons at 6.59, 6.43 and 6.45 ppm (a and a′-ring): two *n*-alkyl-dihydroxyphenyl groups were thus defined, the aromatic a and a′-rings ending two aliphatic chains. The carbonyl at 210.0 ppm (C-1) was correlated with the methylene at 3.08 ppm and with the aromatic proton of b-ring at δ_H 6.30 (H-18), defining thus the substructure I.

The methine proton at 4.54 ppm (H-1′) was correlated in the HMBC spectrum with the aromatic carbons at 118.0 (C-16′) and 157.1 ppm (C17′ and 21′) of b′-ring, allowing to define substructure II (Table 2). It was also correlated with carbons at 123.3 (C-20), 137.4 (C-19) and 160.4 (C-21) of b-ring, indicating a diarylmethane structure and allowing the linkage of substructures I and II as indicated in structure **8**. This was confirmed by the long range correlation between proton at 7.57 ppm (H-19) and the methine carbon 1′. The resulting dimeric structure **8** for maingayone was in agreement with mass spectral fragmentations. Compound **8** appears as a dimeric product of malabaricone C (**2**) which could result from the aldol condensation of one molecule **2** with another molecule of malabaricone C, followed by the reduction of the aldol hydroxyl.

The biosynthesis of promalabaricones presumably results from the elongation of a cinnamoyl type precursor, originating from an amino acid such as phenyl alanine and its hydroxy-derivatives (tyrosine or DOPA) by six acetate (malonate) units, followed by reduction of the first three acetate units and cyclisation of the last three acetate units into a triketonic cyclohexane ring according to the phloroglucinol type cyclisation; then the reduction of the *para*-carbonyl group into an alcohol yields promalabaricones and further dehydration of the ring hydroxyl leads to malabaricones (Fig 2). This suggested mixed biosynthesis is similar to that now well established for flavonoids, for which the cinnamoyl moiety is elongated by only three acetate units, followed by cyclisation. Acids **3** and **4** could result either from the cleavage of promalabaricones at the triketonic methine level, or from an unachieved promalabaricone biosynthesis.

When tested on human tumoral KB cells, all the compounds showed significant cytotoxicity, malabaricone B being the more active with an IC_{50} of 3 μg ml^{-1} (Table 3). Malabaricones were more toxic than the related promalabaricones, and *p*-hydroxyphenyl compounds were generally more toxic than the *o*,*p*-dihydroxyphenyl ones. These compounds were responsible for the toxicity of the plant extract. Tubulin could be the target for cytotoxicity of the active components as an IC_{50} value of 10 μg ml^{-1} was observed for the crude extract. The mechanism of action of the *Myristica* phenols may thus differ from that described for the 3,5-dihydroxyacylphenols which cleave DNA or inhibit DNA polymerase β.[8,12,13]

All the isolated compounds had moderate or no activity against *Plasmodium falciparum*, only malabaricone B

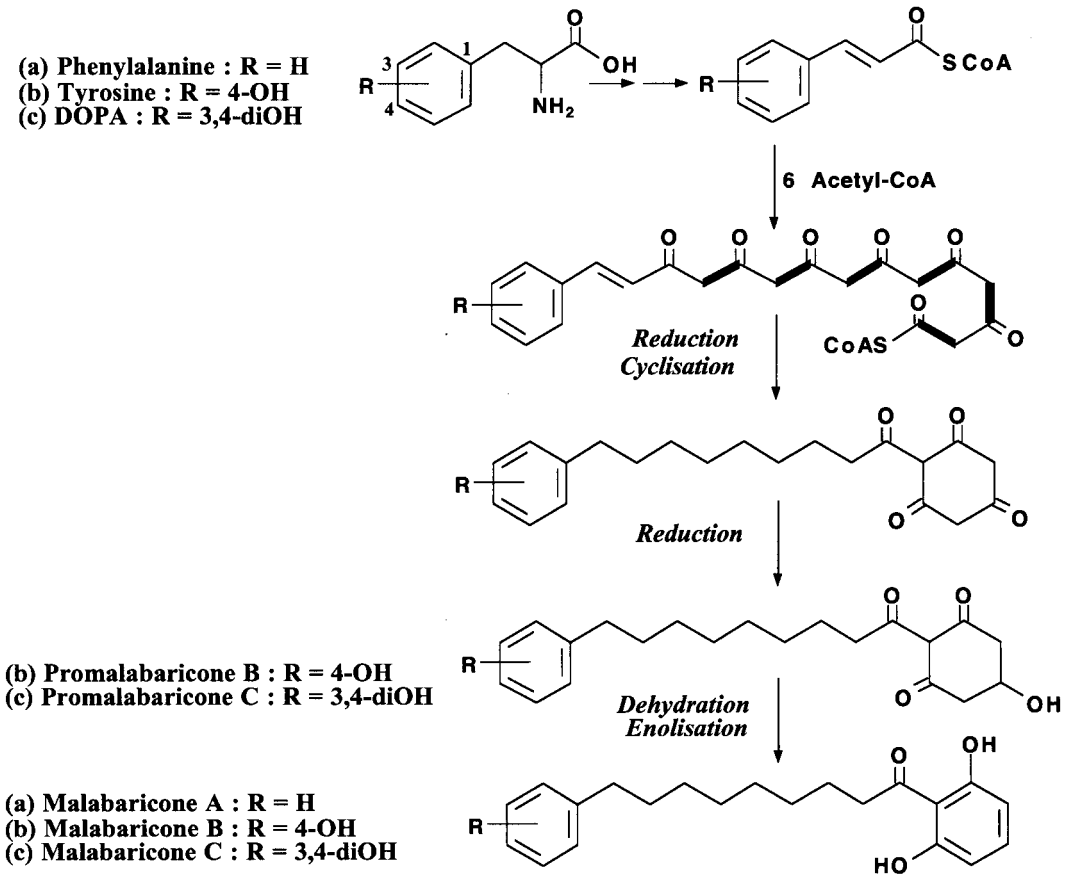


Figure 2. Hypothetical pathway for promalabaricones and malabaricones biosynthesis.

Table 3. Biological activity of acylphenols from *M. maingayi:* cytotoxicity against KB cells, and inhibition of *Plasmodium falciparum* growth

Compound	KB cells IC_{50} $\mu g\ \mu l^{-1}$ (μM)	*Plasmodium falciparum* IC_{50} $\mu g\ \mu l^{-1}$ (μM)
Crude extract	23	–
Promalabaricone B (**1**)	6 (16)	>100 (>277)
Promalabaricone C (**2**)	9 (24)	16 (43)
Maingayic acid B (**3**)	30 (120)	45 (180)
Maingayic acid C (**4**)	8 (30)	56 (210)
Malabaricone A (**5**)	50 (153)	32 (98)
Malabaricone B (**6**)	3 (9)	>100 (>292)
Malabaricone C (**7**)	4 (11)	20 (56)
Maingayone (**8**)	18 (26)	>100 (>143)

showing a significative growth inhibition with an IC_{50} value of 16 $\mu g\ ml^{-1}$ (Table 3).

Experimental

General experimental procedures

Melting points were determined on a Reichert microscope apparatus and are uncorrected. Optical rotations were measured on a Perkin–Elmer 141 polarimeter at room temperature. IR spectra were recorded in KBr disks on a Nicolet Impact 400D spectrophotometer, and UV spectra with an Uvikon 930 Kontron spectrophotometer. Mass spectra were obtained either on a Nermag R 10-10 in the CI or EI mode or on a ZAB2-SEQ (VG analytical) for the FAB MS.

^{1}H and ^{13}C spectra were recorded on a Bruker AC 300 spectrometer operating at 300.13 and 75.43 MHz, respectively, or on an Avance 400 Bruker (400.13: ^{1}H and 100.61 MHz: ^{13}C). ^{1}H chemical shifts were referenced relative to CHD_2OD at 3.31 ppm or $CHCl_3$ at 7.24 ppm and ^{13}C chemical shifts to central peak of CD_3OD at 49.0 or $CDCl_3$ at 77.0 ppm. For the HMBC spectra, the delay was optimized for a coupling constant of 7 Hz.

Plant material

The fruits of *Myristica maingayi* Hk, f. were collected at Gua Musang (Malaysia) in August 1997. A voucher specimen (no. KL 4525) is on deposit in the Herbarium of the Department of Chemistry, University of Malaysia (Kuala-Lumpur).

Extraction and isolation

The powder of dried fruits (1.3 kg) was extracted with EtOAc at room temperature. The residue (83 g) obtained after evaporation of the solvent was chomatographed on a silica gel column with a CH_2Cl_2/MeOH gradient starting from 95/5 to pure MeOH and yielding 36 fractions (F1–F36). Fractions F1–F5 were combined (2.34 g) and further chromatographed on silica gel with a cyclohexane/AcOEt gradient starting from 95/5 to pure AcOEt and 28 fractions were collected: fraction 20 yielded malabaricone A (**5**, 0.2 g). Fractions F9 and F12 corresponded to pure malabaricones B (**6**, 1.76 g) and C (**7**, 11.42 g), respectively. Fractions F18–F21 were combined (6.01 g) and further chromatographed on silica gel with a AcOEt/MeOH gradient starting from 9/1 to pure MeOH and 12 fractions (f1–f12) were collected: fraction f9 yielded promalabaricone B (**1**, 70 mg) and fractions f4–f6 were combined (0.42 g) and submitted to repetitive column chromatography to yield maingayic acid B (**3**, 42 mg).

Fraction F22 was chomatographed on silica gel with an AcOEt/MeOH gradient starting from 9/1 to pure MeOH: 18 fractions (f1–f18) were collected. Fraction f11 yielded maingayic acid C (**4**, 20 mg) and fraction f17 yielded promalabaricone C (**2**, 120 mg). Fractions f3–5 (0.70 g) were combined and chromatographed on a reversed phase (RP8) silica gel column eluted with a MeOH/H_2O gradient from 1/1 to pure MeOH and yielded maingayone (**8**, 250 mg).

Determination of cytotoxic activity

The cytotoxicity assays were carried out in 96-well microtiter plates in triplicate against human nasopharynx carcinoma KB cell lines (3×10^3 cells ml^{-1}) using a modification of the published method.[14] KB cells were maintained in Dulbecco's D-MEM medium, supplemented with 10% fetal calf serum, $NaHCO_3$ (3.7 g l^{-1}), l-glutamine (2 mM), penicillin G (100 units ml^{-1}), streptomycin (100 $\mu g\ ml^{-1}$) and gentamycin (10 $\mu g\ ml^{-1}$). After 72 h incubation at 37°C in air/CO_2 (95/5) with or without test compounds, cell growth was estimated by colorimetric measurement of stained living cells by neutral red. Optical density was determined at 540 nm on a Titertek Multiscan photometer. The IC_{50} value was defined as the concentration of sample necessary to inhibit the cell growth to 50% of the control.

Determination of antimalarial activity

The assays on *Plasmodium falciparum* were performed on the chloroquine resistant-strain FcB1/Colombia. *P. falciparum* was maintained in vitro on human type O^+ erythrocytes in RPMI 1640 culture medium supplemented with 27.5 mM $NaHCO_3$, 25 mM HEPES, 11 mM glucose and 7.5% heat treated human O^+ serum in an atmosphere of 3% CO_2, 6% O_2, 91% N_2 at 37°C and pH 7.4. The experiments were carried out in 96-well plates according to the semi-automated microdilution technique.[15] Stock solutions of acylphenols were prepared in DMSO. Different concentrations were tested on parasite cultures (2% parasitaemia, 2% haematocrite) for 24 h, at 37°C prior to the addition of 0.5 μCi per well of [^{3}H]hypoxanthine. DMSO concentration never exceeded 0.05%, a concentration which has no effect upon parasite growth. After further incubation for 24 h, the cells were harvested onto filter paper. The dried disks were counted with a scintillation spectrometer. The growth inhibition was determined by comparison of the incorporated radioactivity of treated culture to control culture without drug. The concentrations causing 50% of growth inhibition (IC_{50}) were obtained from the drug concentration–response curves and the results expressed as the mean determined from several independent experiments.

Promalabaricone B (1). $C_{21}H_{28}O_5$=360. Colorless microcrystals mp: 140–142°C (MeOH); $[\alpha]_D^{22}$=0° (*c* 0.5, MeOH). UV (MeOH) λ_{max} (log ϵ): 203 (4.1), 226 (4.0), 274 (4.1). IR (KBr) ν_{max} (cm^{-1}): 3418, 2922, 2852, 1722 (sh), 1640, 1582, 1517, 1447, 1406, 1239, 1064, 824, 783, 611, 511. CI MS (NH_3), *m/z* (%): 361 ($[M+H]^+$, 100), 343 ($[M-H_2O+H]^+$, 94), 284 (10), 268 (12), 233 (15). HR FAB MS, *m/z*: 361.2024 ($[M+H]^+$), calcd $C_{21}H_{29}O_5$= 361.2015. EI MS *m/z* (%): 360 ($M^{\cdot+}$, 5), 342 (23), 232 (20), 204 (25), 137 (95), 107 (100). ^{1}H NMR (300.13 MHz, DMSO-d_6): 1.25 (8H, br.s, CH_2-4 to 7), 1.48 (4H, br.s, CH_2-3 and 8), 2.42 (2H, t, *J*=7.5 Hz, CH_2-9), 2.93 (2H, t, *J*=7.4 Hz, CH_2-2), 2.81 (2H, m) and 2.51 (2H, m) CH_2-18 and 20), 4.14 (1H, br.s, H-19), 6.64 (2H, m, *J*=8.6 Hz, H-12, 14), 6.94 (2H, m, *J*=8.6 Hz, H-11, 15), 9.08 (1H, br.s, OH-13), 18.10 (1H, br.s, OH-19).

Promalabaricone C (2). $C_{21}H_{28}O_6$=376. Mp: 162–164°C (MeOH); $[\alpha]_D^{22}$=0° (*c* 0.5, MeOH). UV (MeOH) λ_{max} (log ϵ): 204 (4.3), 222 (2.4), 272 (3.9). IR (KBr) ν_{max} (cm^{-1}): 3420, 2930, 2854, 1729, 1637, 1585, 1400, 1262, 1038, 815. CI MS (NH_3), *m/z* (%): 377 ($[M+H]^+$, 29), 359 $[M-H_2O+H]^+$, 100). EI MS *m/z* (%): 376 ($M^{\cdot+}$, 5), 358 (20), 248 (25), 221 (21), 149 (100), 137 (85), 123 (70), 110 (80).

Maingayic acid B (3). $C_{15}H_{22}O_3$=250. Colorless crystals (MeOH), mp: 106–107°C. UV (MeOH) λ_{max} (log ϵ): 202 (3.6), 224 (3.6), 278 (2.9). IR (KBr) ν_{max} (cm^{-1}): 3400, 2925, 2856, 1778 (sh.), 1702, 1620, 1602, 1520, 1473, 1251, 1239, 1219, 859, 818, 725. CI MS *m/z* (%): 268 ($[M+NH_4]^+$, 95), 250 ($M^{\cdot+}$, 57), 233 (100), 205 (10), 107 (20). HRMS, *m/z*: 250.1589 ($[M]^{\cdot+}$), calcd $C_{15}H_{22}O_3$= 250.1569. ^{13}C NMR (75.47 MHz, CD_3OD): 26.0 (C-3), 30.2 (C-4), 30.2 (C-5), 30.3 (C-6), 30.4 (C-7), 32.9 (C-8), 35.0 (C-2), 36.0 (C-9), 116.0 (C-12), 116.0 (C-14), 130.2 (C-11), 130.2 (C-15), 134.8 (C-10), 156.1 (C-13), 177.8 (C-1). ^{1}H NMR (300.13 MHz, CD_3OD): 1.26 (8H, br.s, CH_2-4 to 7), 1.56 (4H, m, CH_2-3, 8), 2.26 (2H, t, *J*=7.5 Hz, CH_2-2), 2.47 (2H, t, *J*=7.6 Hz, CH_2-9), 6.68 (2H, m, *J*=8.3 Hz, H-12, 14), 6.95 (2H, m, *J*=8.3 Hz, H-11, 15).

Maingayic acid C (4). $C_{15}H_{22}O_4$=266. Colorless crystals (MeOH), mp: 133–135°C. UV (MeOH) λ_{max} (log ϵ): 204 (4.1), 220 (3.8), 278 (3.5). IR (KBr) ν_{max} (cm^{-1}): 3433, 2929, 2856, 1713, 1611, 1529, 1389, 1207, 1117, 871, 812. CI MS (NH_3), *m/z* (%): 284 ($[M+NH_4]^+$, 85), 266 ($M^{\cdot+}$, 50), 249 (100), 233 (10), 205 (5), 151 (5), 123 (15). HRMS, *m/z*: 266.1499 ($[M]^{\cdot+}$), calcd $C_{15}H_{22}O_4$=266.1518. ^{13}C NMR (75.47 MHz, CD_3OD): 26.2 (C-3), 30.2 (C-4), 30.2 (C-5), 30.4 (C-6), 30.4 (C-7), 32.9 (C-8), 35.2 (C-2), 36.2 (C-9), 116.2 (C-14), 116.5 (C-11), 120.6 (C-15), 135.7 (C-10), 144.0 (C-13), 146.0 (C-12), 178.1 (C-1). ^{1}H NMR (300.13 MHz, CD_3OD): 1.30 (8H, br.s, CH_2-4 to 7), 1.53 (2H, t, *J*=6.5 Hz, CH_2-3), 1.58 (2H, p, *J*=7.1 Hz, CH_2-8), 2.27 (2H, t, *J*=7.2 Hz, CH_2-2), 2.43 (2H, t, *J*=7.4 Hz, CH_2-9), 6.46 (1H, dd, *J*=8.0, 2.0 Hz, H-15), 6.58 (1H, d, *J*=2.2 Hz, H-11), 6.64 (1H, d, *J*=8.0 Hz, H-14).

Malabaricone A (5). ($C_{21}H_{26}O_3$)=326. Colorless crystals (MeOH), mp: 80–82°C (lit.[1]: mp 81–82°C). UV (MeOH) λ_{max} (log ϵ): 205 (4.2), 221 (3.9), 272 (3.6), 340 (3.1). IR (KBr) ν_{max} (cm^{-1}): 3262, 2930, 2914, 2854, 1632, 1603, 1510, 1459, 1347, 1242, 1045, 965, 874, 722. CI MS (NH_3) *m/z* (%): 344 ($[M+NH_4]^+$, 20), 327 ($[M+H]^+$, 100), 137 (20), 91 (9). ^{13}C NMR (75.47 MHz, CD_3OD): 24.4 (C-3), 29.1 (C-7), 29.3 (C-4), 29.3 (C-5), 29.3 (C-6), 31.4 (C-8), 35.9 (C-9), 44.7 (C-2), 108.2 (C-18), 108.2 (C-20), 110.0 (C-16), 125.5 (C-13), 128.1 (C-12), 128.1 (C-14), 128.3 (C-11), 128.3 (C-15), 135.9 (C-19), 142.9 (C-10), 161.3 (C-17), 161.3 (C-21), 208.5 (C-1). ^{1}H NMR (300.13 MHz, CD_3OD): 1.33 (8H, br.s, CH_2-4 to 7), 1.61 (2H, p, *J*=7.5 Hz, CH_2-8), 1.71 (2H, p, *J*=7.5 Hz, CH_2-3), 2.60 (2H, t, *J*=7.5 Hz, CH_2-9), 3.15 (2H, t, *J*=7.5 Hz, CH_2-2), 6.40 (2H, d, *J*=8.3 Hz, H-18, 20), 7.17 (3H, m, H-11, 13, 15), 7.26 (3H, m, H-12, 14, 19).

Malabaricone B (6). $C_{21}H_{26}O_4$=342. Colorless crystals (MeOH), mp: 133–135°C (lit.[1]: mp 81–82°C). UV (MeOH) λ_{max} (log ϵ): 203 (4.1), 224 (4.2), 270 (3.9), 341 (3.3). IR (KBr) ν_{max} (cm^{-1}): 3572, 3348, 2921, 2848, 1637, 1588, 1517, 1460, 1394, 1341, 1255, 1222, 1193, 1045, 959, 830, 808, 782, 722. CI MS (NH_3) *m/z* (%): 360 ($[M+NH_4]^+$, 10), 343 ($[M+H]^+$, 100), 137 (10), 107 (10). ^{13}C NMR (75.47 MHz, CD_3OD): 24.4 (C-3), 29.0 (C-5), 29.0 (C-6), 29.2 (C-7), 29.5 (C-4), 31.6 (C-8), 35.0 (C-9), 44.7 (C-2), 107.8 (C-18), 107.8 (C-20), 110.1 (C-16), 115.0 (C-12), 115.0 (C-14), 129.3 (C-11), 129.3 (C-15), 134.6 (C-10), 135.7 (C-19), 153.6 (C-13), 161.6 (C-17), 161.6 (C-21), 208.6 (C-1), ^{1}H NMR (300.13 MHz, CD_3OD): 1.29 (8H, br.s, CH_2-4 to 7), 1.51 (2H, p, *J*=7.4 Hz, CH_2-8), 1.64 (2H, p, *J*=7.4 Hz, CH_2-3), 2.46 (2H, t, *J*=7.4 Hz, CH_2-9), 3.10 (2H, t, *J*=7.4 Hz, CH_2-2), 6.32 (2H, d, *J*=8.2 Hz, H-18, 20), 6.69 (2H, m, *J*=8.4 Hz, H-12, 14), 6.96 (2H, m, *J*=8.4 Hz, H-11, 15), 7.14 (1H, t, *J*=8.2 Hz, H-19).

Malabaricone C (7). $C_{21}H_{26}O_5$=358. Colorless crystals (MeOH), mp: 127–128°C; (lit.[1]: mp 123–124°C). UV (MeOH) λ_{max} (log ϵ): 206 (4.5), 222 (4.3), 272 (4.1), 341 (3.4). IR (KBr) ν_{max} (cm^{-1}): 3467, 3296, 2934, 2855, 1683 (ép.), 1637, 1604, 1532, 1460, 1393, 1367, 1255, 1124, 1045, 955, 801, 742, 538. CI MS (NH_3) *m/z* (%): 376 ($[M+NH_4]^+$, 12), 359 ($[M+H]^+$, 100), 137 (10), 123 (8). ^{13}C NMR (75.47 MHz, CD_3OD): 24.3 (C-3), 28.9 (C-7), 29.2 (C-4), 29.2 (C-5), 29.2 (C-6), 31.4 (C-8), 35.0 (C-9), 44.6 (C-2), 107.5 (C-18), 107.5 (C-20), 110.1 (C-16), 114.9 (C-14), 115.2 (C-11), 120.0 (C-15), 135.3 (C-10), 135.7 (C-19), 141.8 (C-13), 143.8 (C-12), 161.8 (C-17), 161.8 (C-21), 208.7 (C-1).

^{1}H NMR (300.13 MHz, CD_3OD): 3.09 (2H, t, *J*=7.4 Hz, CH_2-2), 1.62 (2H, p, *J*=7.4 Hz, CH_2-3), 1.30 (2H, m, CH_2-4), 1.26 (6H, br.s, CH_2-5 to 7), 1.49 (2H, p, *J*=7.4 Hz, CH_2-8), 2.39 (2H, t, *J*=7.4 Hz, CH_2-9), 6.62 (1H, d, *J*=2.0 Hz, H-11), 6.67 (1H, d, *J*=8.0 Hz, H-14), 6.50 (1H, dd, *J*=8.0, 2.0 Hz, H-15), 6.31 (2H, d, *J*=8.1 Hz, H-18, 20), 7.12 (1H, t, J=8.1 Hz, H-19).

Maingayione (8). $C_{42}H_{52}O_9$=700. Amorphous solid. $[\alpha]_D^{22}$=+3° (*c* 0.5, MeOH). UV (MeOH) λ_{max} (log ϵ): 206 (4.8), 277 (4.2), 354 (3.4). IR (KBr) ν_{max} (cm^{-1}): 3385, 2929, 2856, 1702, 1611, 1523, 1468, 1441, 1362, 1283, 1242, 1117, 994, 815, 789. MS FAB$^+$ *m/z* (%): 701 ($[M+H]^+$, 5), 629 (3), 591 (45), 441 (9) 473 (16), 343

(25), 123 (100). CI MS (NH_3), *m/z* (%): 591 (1), 495 (1), 416 (2), 399 (6), 387 (18), 359 (100), 343 (23).

Maingayione octa-acetate. SM FAB^+ *m/z* (%): 1059 ($[M+Na]^+$, 100); HR MS obs. 1059.4275, calcd for $C_{58}H_{68}O_{17}Na$: 1059.4354. ^1H NMR (300.13 MHz, $CDCl_3$): 1.23 (16H, br.s, 8 CH_2), 1.55 (8H, m, 4 CH_2: 3, 8, 3′ and 8′), 2.07 (3H, s), 2.13 (6H, s), 2.20 (3H, s) and 2.23 (12H, s): 8-CO–CH_3, 2.55 (8H, m, 4 CH_2: 2, 9, 2′ and 9′), 4.21 (1H, t, *J*=7.5 Hz, H-1′), 6.88 (2H, d, *J*=8.0 Hz, H-18′ and 20′), 6.90–7.10 (6H, m, arom. a and a′-ring), 7.10 (1H, d, *J*=8.7 Hz, H-18), 7.20 (1H, t, *J*=8.0 Hz, H-19′), 7.50 (1H, d, *J*=8.7 Hz, H-19).

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