

QUASSINOIDS AND LIMONOIDS FROM *HARRISONIA PERFORATA*

Kunio Kamiuchi, Katsuyoshi Mitsunaga, Kazuo Koike, Yishan Ouyang, Taichi Ohmoto, and Tamotsu Nikaido*

School of Pharmaceutical Sciences, Toho University, 2-2-1, Miyama, Funabashi, Chiba 274, Japan

Abstract ---- Three new quassinoids, perforaquassins A, B and C, and a new limonoid, perforin A, were isolated from the bark of *Harrisonia perforata* (Simaroubaceae). Their structures were determined by spectral techniques.

During our investigation of Simaroubaceae plants, recently, chemical constituents of two *Harrisonia* species, *H. brownii* and *H. perforata*, have been reported.¹ In earlier studies, chromones and limonoids had been reported, but no quassinoid in spite of their classification as Simaroubaceae. This paper describes the isolation and structural elucidation of three new C₂₀-type quassinoids, perforaquassin A (1), B (2) and C (3), a new limonoid, perforin A (4), along with eight known compounds, obacunone (5),² harrisonin (6),² ptaerochromenol,³ umtatin,³ physcion,⁴ alloptaeroxylin-5-methyl ether (perforatin A),⁵ 2-hydroxymethylalloptaeroxylin-5-methyl ether⁶ and scopoletin,⁷ from the bark of *H. perforata*. There are two new points of view as follows; one is that this is the first time that quassinoids were isolated from the genus *Harrisonia*, and the other involves the viewpoint of biosynthesis in that quassinoids were isolated together with limonoids from the same simaroubaceous plant.

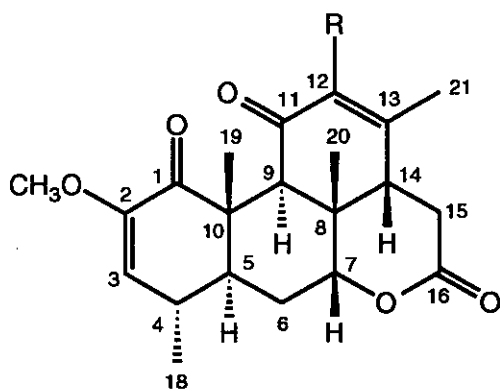
Perforaquassin A (1) was obtained as a white powder, $[\alpha]_D^{20} +38.2^\circ$ (MeOH). The molecular formula, C₂₁H₂₆O₅, was determined by high-resolution mass spectroscopy (HR-ms). The infrared (ir) and ultraviolet (uv) spectra showed absorption bands due to δ -lactone ($\nu_{\max}$ 1734 cm⁻¹) and α,β -unsaturated ketone ($\nu_{\max}$ 1675 cm⁻¹ and $\lambda_{\max}$ 236 nm). The ¹H nmr spectrum of 1 showed signals due to one secondary methyl at δ 1.12 (d, $J=6.6$ Hz, 4-Me), three tertiary methyls at δ 1.19 (s, 8-Me), 1.53 (s, 10-

Me) and 1.93 (d, $J=1.5$ Hz, 13-Me) and two olefinic protons at δ 5.30 (d, $J=2.6$ Hz, H-3) and 5.84 (d, $J=1.5$ Hz, H-12). The ^1H and ^{13}C nmr spectra showed that **1** has a picrasane skeleton and, from a comparison with those of quassin,⁸ the methoxyl group at C-12 in quassin was thought to be replaced by an olefinic proton (δ 5.84) in **1**. In addition, the proton at δ 5.84 was long-range coupled ($J=1.5$ Hz) with a methyl proton at δ 1.93 assigned to 13-Me in the ^1H - ^1H COSY spectrum, which also suggested that the olefinic proton was assigned to 12-H. The complete assignments of the ^1H and ^{13}C nmr signals for **1**, shown in Tables 1 and 2, were based on ^1H - ^1H COSY, HMBC and our experience.⁸ The relative stereochemistry was established by a phase-sensitive NOESY spectrum as in the Figure 1, which was compatible with that of the usual picrasane skeleton. The absolute stereochemistry of **1** was assigned based on the circular dichroism (cd) spectrum. The cd spectrum of **1** showed a distinct positive split Cotton effect ($\Delta\epsilon_{254} +25.49$ and $\Delta\epsilon_{332} -11.93$), due to exciton coupling by two enones, indicating the absolute stereochemistry of **1** to be the same as that of quassin. The structure of perforaquassin A was determined to be **1**.

Perforaquassin B (**2**) was obtained as a white powder, $[\alpha]_D -31.4^\circ$ (MeOH). The molecular formula, $\text{C}_{21}\text{H}_{28}\text{O}_5$, was determined by HR-ms. The ir and uv spectra of **2** indicated the presence of ketone (ν_{max} 1724 cm^{-1}) and α,β -unsaturated ketone (ν_{max} 1672 cm^{-1} and λ_{max} 236 nm) groups. The ^1H and ^{13}C nmr spectra of **2** were similar to those of **1**; however, **2** has no olefinic signals due to C-2 (H-2) and C-3 (H-3). The ^1H - ^1H COSY spectrum suggested the presence of an isolated structure unit, $-\text{C}(2)\text{H}-\text{C}(3)\text{H}_2-\text{C}(4)\text{H}(\text{Me})-\text{C}(5)\text{H}-\text{C}(6)\text{H}_2-\text{C}(7)\text{H}-$, in the structure of **2**. These data indicated that the α,β -unsaturated ketone group in **1** was replaced by the 2,3-saturated ketone group in **2**. The stereochemistry was established by a phase-sensitive NOESY spectrum to be compatible with that of **1** (Figure 2). Furthermore, the proton at C-2 (δ 4.62) was assigned as β -axial from the coupling constants ($J=12.7$ and 7.3 Hz) and by the presence of a strong NOE with β -axial methyl proton signals at C-10, indicating that the configuration of the methoxyl group at C-2 was α . The structure of perforaquassin B was determined to be **2**.

Perforaquassin C (**3**) was obtained as needles, mp 202-204°C, $[\alpha]_D +25.4^\circ$ (MeOH). The molecular formula, $\text{C}_{21}\text{H}_{30}\text{O}_5$, was determined by HR-ms. The ir and uv spectra of **3** indicated the presence of hydroxyl (ν_{max} 3325 cm^{-1}), δ -lactone (ν_{max} 1736 cm^{-1}) and α,β -unsaturated ketone (ν_{max} 1649 cm^{-1} and λ_{max} 243 nm) groups. The ^1H and ^{13}C nmr spectra of **3** were similar to those of **2**, except that the

carbonyl carbon signal at C-1 in **2** was replaced by a methine carbon signal (δ 82.8) in **3**. Furthermore, the ^1H - ^1H COSY experiment with **3** revealed the presence of a spin system, $-\text{C}(1)\text{H}-\text{C}(2)\text{H}-\text{C}(3)\text{H}_2-\text{C}(4)\text{H}(\text{Me})-\text{C}(5)\text{H}-\text{C}(6)\text{H}_2-\text{C}(7)\text{H}-$. Compound (**3**) was acetylated with acetic anhydride in pyridine to give a monoacetate (**3a**). The ^1H nmr spectrum of **3a** showed an extra methyl signal due to an acetyl group at δ 2.01 (3H, s) and the signal of H-1 at δ 4.56 showed a clear downfield shift ($\Delta\delta = 1.4$ ppm), when compared with the corresponding signal in **3**. These data indicated that the hydroxyl group in **3** was located at C-1. The stereochemistry was established by a phase-sensitive NOESY spectrum to be compatible with that of **2**, except for C-1 (Figure 3). A strong NOE correlation between H-1 and H-5 suggested that the configuration of the hydroxyl group at C-1 was β , and from the coupling constants between H-1 and H-2 (8.4 Hz), H-2 and H-3 α ($J_{2,3\alpha} = J_{3\alpha,3\beta} = 12.1$ Hz) and H-2 and H-3 β (5.1 Hz), the H-1 and H-2 protons were deduced to be α and β , respectively. The structure of perforaquassin C was determined to be **3**.



1 : R=H
quassin : R=OCH₃

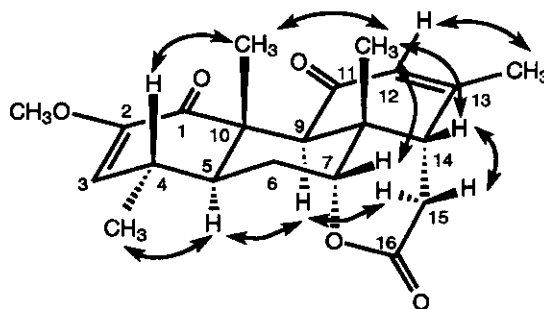
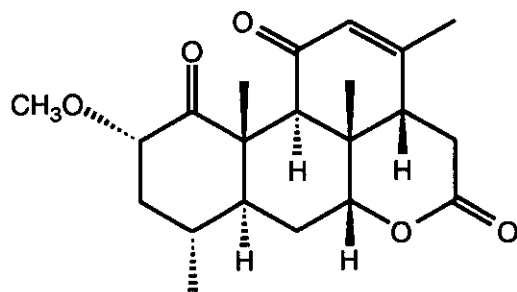


Figure 1. NOESY correlations of **1**



2

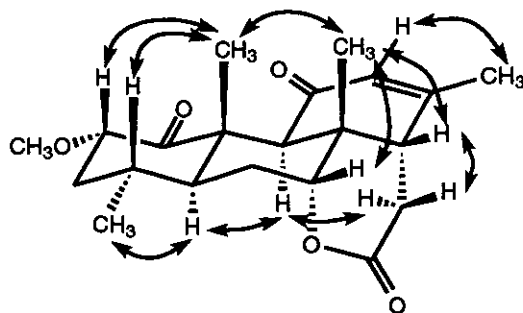


Figure 2. NOESY correlations of **2**

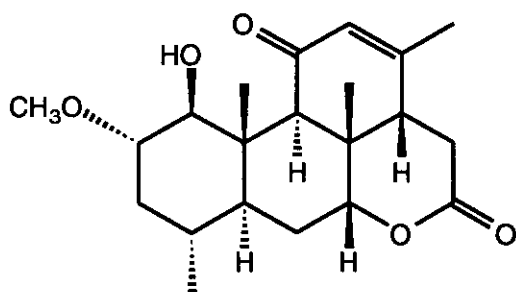
Table 1. ^1H Nmr spectral data for Compounds (1-3) and quassin in CDCl_3

H	1	2	3	quassin
1			3.17 d (8.4)	
2		4.62 dd (12.7, 7.3)	3.26 ddd (12.1, 8.4, 5.1)	
3 α	5.30 d (2.6)	1.31-1.41 m	1.03 q (12.1)	5.31 d (2.9)
β		2.46 m	2.07 ddd (12.1, 5.1, 3.7)	
4	2.48 m	2.12 m	1.49 m	2.49 m
5	1.84-1.89 m	1.49-1.54 m	1.29 ddd (13.2, 10.6, 2.6)	1.78-1.84 m
6 α	2.09 m	2.06-2.10 m	2.02 dt (14.7, 2.6)	2.09 m
β	1.92-1.97 m	1.92 dt (12.8, 2.2)	1.75 ddd (14.7, 13.2, 2.6)	1.85-1.92 m
7	4.31 m	4.43 t (2.2)	4.31 t (2.6)	4.28 m
9	2.95 s	3.33 s	2.61 s	2.99 s
12	5.84 d (1.5)	5.92 d (1.1)	5.88 d (1.5)	
14	2.36 dd (12.1, 7.0)	2.52 dd (12.5, 7.0)	2.35 dd (12.8, 6.2)	2.39 dd (11.7, 7.0)
15 α	2.60 dd (18.7, 12.1)	2.71 dd (18.7, 12.5)	2.51 dd (18.3, 12.8)	2.61 dd (18.7, 11.7)
β	2.96 dd (18.7, 7.0)	3.06 dd (18.7, 7.0)	2.93 dd (18.3, 6.2)	3.00 dd (18.7, 7.0)
18	1.12 d (6.6)	1.04 d (6.2)	0.93 d (6.2)	1.12 d (7.0)
19	1.53 s	1.54 s	1.17 s	1.57 s
20	1.19 s	1.28 s	1.19 s	1.21 s
21	1.93 d (1.5)	2.07 d (1.1)	1.97 d (1.5)	1.88 s
OMe-2	3.58 s	3.50 s	3.51 s	3.59 s
OMe-12				3.67 s
OH			6.15 s	

Coupling constants (J in Hz) are given in parentheses.

Table 2. ^{13}C Nmr spectral data for Compounds (1-3) and quassin in CDCl_3

C	1	2	3	quassin
1	197.9	210.4	82.8	197.8
2	148.0	78.9	81.0	148.1
3	116.1	44.1	38.6	116.3
4	31.3	28.6	28.0	31.2
5	43.5	47.2	43.0	43.2
6	25.8	26.3	25.4	25.9
7	82.2	82.2	83.2	82.1
8	37.2	36.9	38.8	37.1
9	45.3	46.2	55.1	46.3
10	47.2	48.5	43.5	45.9
11	194.9	194.9	202.2	191.0
12	127.0	126.3	127.2	148.4
13	155.2	157.9	158.8	137.4
14	45.6	47.6	48.1	46.7
15	31.1	31.0	30.9	31.7
16	169.0	169.0	168.6	169.0
18	19.5	18.4	20.0	19.5
19	12.8	14.9	12.1	12.7
20	22.4	23.0	23.0	22.4
21	22.0	22.3	21.9	15.3
OMe-2	55.0	57.7	57.4	55.0
OMe-12				59.3



3

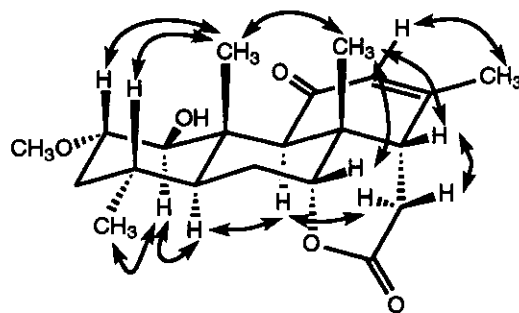


Figure 3. NOESY correlations of 3

Perforin A (**4**) was obtained as a white powder, $[\alpha]_D +97.3^\circ$ (MeOH). The molecular formula, $C_{34}H_{40}O_{14}$, was determined by EI-ms (m/z 672 $[M]^+$) and from the ^{13}C nmr spectrum. The ir spectrum of **4** indicated the presence of δ -lactone (ν_{max} 1751 cm^{-1}) and α,β -unsaturated lactone (ν_{max} 1714 cm^{-1}) groups.

Table 3. 1H Nmr spectral data for Compounds (**4**) and (**5**) in $CDCl_3$

H	4	5
1	7.01 d (11.7)	6.52 d (11.7)
2	5.82 d (11.7)	5.96 d (11.7)
5	2.67 d (12.1)	2.60 dd (13.9, 5.1)
6	5.15 dd (12.1, 2.6)	2.29 dd (13.9, 5.1)
		2.99 t (13.9)
7	4.89 d (2.6)	
9	3.01 d (12.5)	2.15 dd (8.2, 4.0)
11	5.53 d (12.5)	—*
12	4.69 s	—*
15	3.64 s	3.66 s
17	5.90 s	5.46 s
18	1.45 s	1.13 s
19	1.52 s	1.50 s
21	7.33 m	7.42 br s
22	6.23 m	6.37 m
23	7.43 t (1.8)	7.40 t (1.5)
28	1.52 s	1.46 s
29	1.44 s	1.50 s
30	1.39 s	1.25 s
OCOMe-6	2.01 s	
OCOMe-7	2.18 s	
OCOMe-11	2.03 s	
OCOMe-12	2.22 s	

Coupling constants (J in Hz) are given in parentheses.

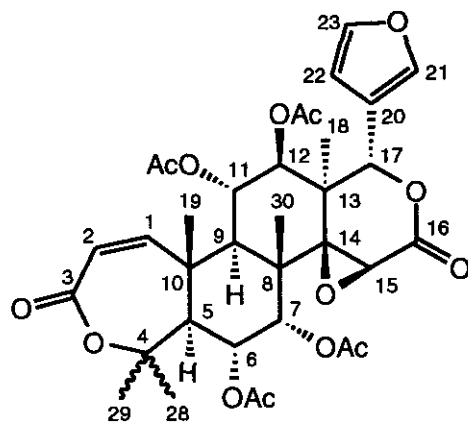
*Could not be assigned.

The 1H nmr spectrum of **4** showed signals due to five tertiary methyls at δ 1.39 (Me-8), 1.44 (Me-4), 1.45 (Me-13) and 1.52 (Me-4 and Me-10), a disubstituted double bond at δ 5.82 and 7.01, and a β -substituted furan at δ 6.23, 7.33 and 7.43. The 1H and ^{13}C nmr spectra of **4** were closely related to those of obacunone (**5**), except that four extra signals due to acetoxyl groups were observed in **4**. These acetoxyl groups were deduced to located at C-6, 7, 11 and 12, respectively by the HMBC spectrum in that

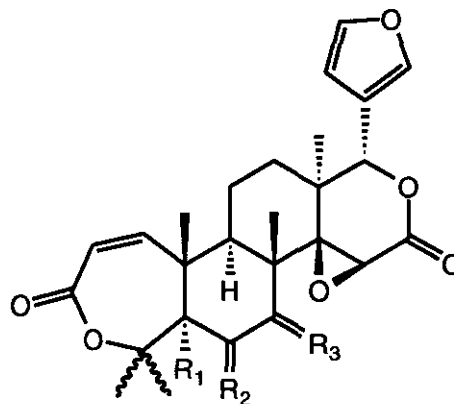
correlations, between the methine protons and the corresponding carbonyl carbons of the acetoxyl groups were observed, respectively. The complete assignments of the nmr signals (Tables 3 and 4) were made based on the ^{13}C - ^1H COSY and HMBC spectra.

Table 4. ^{13}C Nmr spectral data for Compounds (4) and (5) in CDCl_3

C	4	5	C	4	5
1	160.2	156.8	18	16.1	21.1
2	120.3	122.9	19	16.9	16.4
3	167.3	166.9	20	118.9	120.1
4	84.2	84.0	21	141.7	141.0
5	52.4	57.3	22	109.3	109.8
6	68.9	39.9	23	143.9	143.2
7	71.6	207.4	28	35.1	32.0
8	41.9	52.9	29	27.0	26.8
9	45.3	49.2	30	17.7	19.4
10	42.3	43.1	OCOMe-6	20.9	
11	73.3	17.0	OCOMe-6	169.8	
12	78.1	32.7	OCOMe-7	21.0	
13	41.6	37.4	OCOMe-7	169.7	
14	67.7	65.1	OCOMe-11	21.2	
15	54.3	53.3	OCOMe-11	167.7	
16	166.1	166.7	OCOMe-12	21.2	
17	74.3	78.0	OCOMe-12	168.5	



4



R_1 R_2 R_3
 5: H H_2 O
 6: OH O $\alpha\text{-OCH}_3$, $\beta\text{-OH}$

The relative stereochemistry was established by a phase-sensitive NOESY spectrum and the coupling constants of the ^1H nmr spectrum. In spite of the vicinal relation, spin-spin coupling between H-11 at δ 5.53 and H-12 at δ 4.69 was not observed in the ^1H nmr spectrum of **4**, so Discover-cff91 force field molecular dynamics (MD) calculations⁹ of **4** were performed to clarify the conformation. The conformational search suggested the most stable conformation to be in Figure 4. The coupling constant for the $J_{\text{H11, H12}}$ was calculated based on the selected dihedral angle, which was monitored during the MD calculations. The calculated coupling constant¹⁰ for $J_{\text{H11, H12}}$ was 0.2 Hz (dihedral angle of H(11)-C(11)-C(12)-H(12) = -101.0°), which was in accord with the corresponding coupling constant in the experiment (*ca.* 0 Hz). The absolute stereochemistry of **4** was assigned based on the cd spectrum. The cd spectrum of **4** showed a distinct negative split Cotton effect, due to the ene lactone and the epoxy lactone, indicating the absolute stereochemistry of **4** to be the same as that of obacunone (**5**). The structure of perforin A was determined to be **4**.

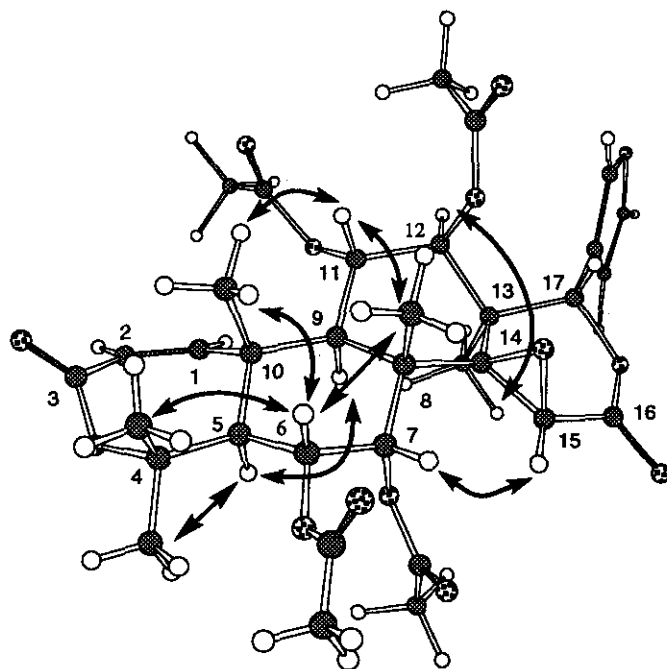


Figure 4. NOESY correlations of **4** as shown in an energy minimized conformation.

EXPERIMENTAL

General experimental procedures. Melting points were determined on a Yanagimoto micromelting point apparatus and are uncorrected. Ir spectra were recorded as KBr pellets on a JASCO 300 FT-ir spectrophotometer. Uv spectra were recorded on a Hitachi 340 uv-vis spectrophotometer in MeOH. Optical rotations were determined on a JASCO DIP-370 digital polarimeter. Cd spectra were recorded on a JASCO J-720W spectrometer in MeOH. EI-ms were measured on a JEOL D-300 and HR-ms and FAB-ms, on a JEOL DX-303 mass spectrometer. ^1H , ^{13}C and 2D nmr spectra were recorded on a JEOL EX-400 (400 MHz for ^1H and 100 MHz for ^{13}C) spectrometer, using TMS as int. standard.

Extraction and isolation. Dried bark (2.8 kg) of *H. perforata* collected in Haicao, Hainan, China, in January 1992, was extracted with CH_2Cl_2 (35 l) and MeOH (35 l) under reflux for 3 h. The CH_2Cl_2 and MeOH extracts were concentrated under reduced pressure to give residues of 60 and 110 g, respectively. The CH_2Cl_2 extract was chromatographed on silica gel (500 g) with CH_2Cl_2 as eluent containing increasing amounts of MeOH (1, 5, 10, 15, 30, 50 and 100%). Fraction obtained by eluting with CH_2Cl_2 and CH_2Cl_2 -MeOH (95:5) was repeatedly chromatographed on silica gel to give ptaerochromenol (15.0 mg), umtatin (4.2 mg), physcion (4.0 mg) and alloptaeroxylin-5-methyl ether (perforatin A) (10.0 mg). The MeOH extract was chromatographed on Diaion HP-20 (1.5 kg, Mitsubishi Kasei) with H_2O as eluent containing increasing amounts of MeOH (10, 20, 30, 50 and 100%). Fraction obtained by eluting with H_2O -MeOH (1:1) and MeOH was repeatedly chromatographed on silica gel and ODS to give **1** (15.3 mg), **2** (77.7 mg), **3** (15.4 mg), **4** (7.0 mg), **5** (25.4 mg), **6** (12.0 mg), 2-hydroxymethylalloptaeroxylin-5-methyl ether (16.4 mg) and scopoletin (10.0 mg).

Perforaquassin A (1). White powder, mp 125-127°C (dec.), $[\alpha]_{\text{D}}^{25} +38.2^\circ$ (c =1.0, MeOH). Uv $\lambda_{\text{max}}^{\text{MeOH}}$ nm (log ϵ): 236 (4.18). Ir $\nu_{\text{max}}^{\text{KBr}}$ cm^{-1} : 1734, 1675, 1637, 1458, 1381, 1250, 1223 and 1041. Cd (c = 5.6×10^{-5} mol/l, MeOH): $[\theta]_{332} -39369$, $[\theta]_{254} +84117$, $[\theta]_{236} -14850$. HR-ms m/z : 358.1772 $[\text{M}]^+$ (calcd for $\text{C}_{21}\text{H}_{26}\text{O}_5$: 358.1773). EI-ms m/z (rel. int.): 358 $[\text{M}]^+$ (53), 343 (6), 325 (7), 297 (14), 283 (7), 237 (7), 213 (6), 159 (14), 127 (26), 105 (26), 91 (48), 85 (18), 77 (35), 69 (89), 53 (31) and 41 (100); ^1H Nmr: Table 1; ^{13}C nmr: Table 2.

Perforaquassin B (2). White powder, mp 103-105°C (dec.), $[\alpha]_D^{25} -31.4^\circ$ ($c=1.0$, MeOH). Uv $\lambda_{\max}^{\text{MeOH}}$ nm (log ϵ): 236 (4.04). Ir $\nu_{\max}^{\text{KBr}}$ cm^{-1} : 1724, 1672, 1439, 1381, 1228, 1111 and 1041. Cd ($c=5.6 \times 10^{-5}$ mol/l, MeOH): $[\theta]_{328} -41415$, $[\theta]_{241} +93159$, $[\theta]_{224} -32538$. HR-ms m/z : 360.1951 $[M]^+$ (calcd for $\text{C}_{21}\text{H}_{28}\text{O}_5$: 360.1929). EI-ms m/z (rel. int.): 360 $[M]^+$ (17), 345 (5), 328 (43), 300 (4), 285 (16), 271 (11), 259 (25), 243 (51), 219 (20), 199 (24), 185 (53), 173 (17), 133 (22), 105 (28), 91 (49), 85 (100), 67 (35), 55 (39) and 41 (61). ^1H Nmr: Table 1; ^{13}C nmr: Table 2.

Perforaquassin C (3). Colorless needles (MeOH), mp 202-204°C, $[\alpha]_D^{25} +25.4^\circ$ ($c=1.0$, MeOH). Uv $\lambda_{\max}^{\text{MeOH}}$ nm (log ϵ): 243 (4.09). Ir $\nu_{\max}^{\text{KBr}}$ cm^{-1} : 3325, 1736, 1649, 1444, 1365, 1319, 1273, 1232, 1153, 1097, 1045 and 1009. Cd ($c=5.5 \times 10^{-5}$ mol/l, MeOH): $[\theta]_{322} -82401$, $[\theta]_{240} +182556$, $[\theta]_{217} +185031$. HR-ms m/z : 362.2092 $[M]^+$ (calcd for $\text{C}_{21}\text{H}_{30}\text{O}_5$: 362.2085). EI-ms m/z (rel. int.): 362 $[M]^+$ (2), 347 (1), 330 (100), 314 (35), 297 (9), 269 (15), 219 (9), 187 (15), 149 (30), 135 (15), 122 (32), 107 (14), 91 (21), 85 (78), 77 (13), 67 (19), 55 (30) and 41 (41). ^1H Nmr: Table 1; ^{13}C nmr: Table 2.

Acetylation of 3. To a solution of **3** (3 mg) in dry pyridine (1 ml), Ac_2O (1 ml) was added and the mixture was allowed to stand at room temperature for 24 h. The reaction mixture was diluted with cold water. The solvent was evaporated under reduced pressure and the residue was purified by preparative tlc [solvent system 5% MeOH in CHCl_3 -EtOAc (1:1)] and preparative hplc [PEGASIL ODS, 10 mm i.d. x 250 mm, SSC; solvent system H_2O -MeOH (6:4), uv detection at 254 nm, flow rate 2 ml min^{-1}] to afford monoacetate (**3a**, 1 mg). Compound (**3a**). White powder, mp 38-40°C (dec.), ^1H Nmr (CDCl_3): δ 0.93 (3H, d, $J=6.6$ Hz, H-18), 1.06 (1H, q, $J=12.4$ Hz, H-3 α), 1.13 (3H, s, H-19), 1.25 (3H, s, H-20), 1.32 (1H, m, H-5), 1.53-1.62 (1H, m, H-4), 1.66 (1H, ddd, $J=14.7, 12.8, 2.2$ Hz, H-6 β), 1.84 (3H, d, $J=1.5$ Hz, H-21), 1.97 (1H, dt, $J=14.7, 3.3$ Hz, H-6 α), 2.01 (3H, s, OCOCH_3 -1), 2.13 (1H, ddd, $J=12.4, 5.5, 3.8$ Hz, H-3 β), 2.24 (1H, dd, $J=12.8, 6.2$ Hz, H-14), 2.62 (1H, dd, $J=18.3, 12.8$ Hz, H-15 α), 2.81 (1H, s, H-9), 2.89 (1H, dd, $J=18.3, 6.2$ Hz, H-15 β), 3.34 (3H, s, OCH_3 -2), 3.37 (1H, ddd, $J=12.4, 9.5, 5.5$ Hz, H-2), 4.27 (1H, m, H-7), 4.56 (1H, d, $J=9.6$ Hz, H-1) and 5.63 (1H, d, $J=1.5$ Hz, H-12).

Perforin A (4). White powder, mp 156-158°C (dec.), $[\alpha]_D^{20} +97.3^\circ$ ($c=0.3$, MeOH). Ir $\nu_{\max}^{\text{KBr}}$ cm^{-1} : 1751, 1714, 1631, 1515, 1466, 1373, 1227, 1134, 1030. Cd ($c=3.0 \times 10^{-5}$ mol/l, MeOH): $[\theta]_{251} -138402$,

$[\theta]_{218} +360426$. EI-*ms m/z* (rel. int.): 672 $[M]^+$ (11), 269 (8), 111 (20), 95 (38), 71 (32) and 43 (100).

1H and ^{13}C Nmr: Table 3.

Conformational calculations of 4. The geometries chosen for the starting conformations were constructed for compound (4) and submitted to energy minimization by using the Discover-cff91 force field program.⁹ The local minima found for them were taken as the starting structures for molecular dynamics (MD) calculations in vacuo at 300k and at a time step of 1 fs. The equilibration time was 10 ps and the total simulation time 100 ps. Trajectory frames were saved every 0.01 ps. The trajectories were then examined with the Analysis module of INSIGHT II.⁹ Calculations were performed on an IRIS Indigo Elan R-4000 computer.

Obacunone (5). Colorless prisms (MeOH), mp 235-237°C (lit.,² mp 226-228°C), $[\alpha]_D^{25} -49.6^\circ$ ($c = 1.0$, MeOH). Ir $\nu_{max}^{KBr} \text{ cm}^{-1}$: 1737, 1709, 1389, 1282, 1030 and 876. FAB-*ms m/z* 455 $[M+H]^+$.

1H and ^{13}C Nmr: Table 3.

Harrisonin (6). Colorless needles (MeOH), mp 161-163°C (lit.,² mp 155-156°C), $[\alpha]_D^{25} +103.4^\circ$ ($c = 1.0$, $CHCl_3$). Ir $\nu_{max}^{KBr} \text{ cm}^{-1}$: 3481, 1745, 1718, 1626, 1439, 1207, 1109, 1020 and 876; EI-*ms m/z* (rel. int.): 517 $[M+H]^+$ (4).

Ptaerochromenol. Colorless prisms (acetone), mp 184-186°C (lit.,³ mp 176°C). Ir $\nu_{max}^{KBr} \text{ cm}^{-1}$: 3323, 1625, 1577, 1460, 1410, 1286, 1128, 1088 and 1034. EI-*ms m/z* (rel. int.): 274 $[M]^+$ (15).

Umtatin. Colorless needles (acetone), mp 179-181°C (lit.,³ mp 178°C), $[\alpha]_D^{20} -38.1^\circ$ ($c = 0.42$, MeOH). Ir $\nu_{max}^{KBr} \text{ cm}^{-1}$: 3278, 1662, 1593, 1464, 1331, 1304, 1248, 1182, 1142, 1105 and 1082. EI-*ms m/z* (rel. int.): 274 $[M]^+$ (47).

Physcion. Orange needles (CH_2Cl_2), mp 203-205°C (lit.,⁴ mp 206°C). Ir $\nu_{max}^{KBr} \text{ cm}^{-1}$: 1676, 1630, 1477, 1387, 1367, 1325, 1273, 1228, 1163 and 1136. EI-*ms m/z* (rel. int.): 284 $[M]^+$ (100).

The spectral data for **alloptaeroxylin-5-methyl ether (perforatin A)**, **2-hydroxymethylalloptaeroxylin-5-methyl ether** and **scopoletin** were referenced.⁵⁻⁷

ACKNOWLEDGMENT

We are grateful to Mr. Gi Ouyang (Hainan People's Hospital) and Mr. Ze Hai Fu (Pharmaceutical Society of Hainan) for plant collection and help in the identification and investigation of folk medicinal plants and Dr. M. Takayama (the Analytical Laboratory of this school) for measuring *ms*.

REFERENCES AND NOTES

1. a) K. Mitsunaga, K. Koike, and T. Ohmoto, *Phytochemistry*, 1994, **37**, 1443; b) T. Tanaka, K. Koike, K. Mitsunaga, K. Narita, S. Takano, A. Kamioka, E. Sase, Y. Ouyang, and T. Ohmoto, *Phytochemistry*, 1995 (in press).
2. I. Kubo, S. P. Tanis, Y.-W. Lee, I. Miura, and K. Nakanishi, *Heterocycles*, 1976, **5**, 485.
3. F. M. Dean, B. Porton, A. W. Price, N. Somvichien, and D. A. H. Talor, *Tetrahedron Lett.*, 1967, 2737.
4. M. Takido, *Chem. Pharm. Bull.*, 1958, **6**, 397.
5. F. M. Dean and M. L. Robinson, *Phytochemistry*, 1971, **10**, 3221.
6. A. M. Balde, M. Vanhaelen, and R. Ottinger, *Phytochemistry*, 1987, **26**, 2415.
7. Y. Saiki, K. Morinaga, O. Okegawa, S. Sakai, A. Ueno, and S. Fukushima, *Yakugaku Zasshi*, 1971, **91**, 1313.
8. K. Koike, K. Mitsunaga, and T. Ohmoto, *Chem. Pharm. Bull.*, 1990, **38**, 2746.
9. Discover 2.9.5 program; Insight II 3.5.0 program, Biosym Technol. Inc., San Diego, CA, USA.
10. a) R. U. Lemieux and J. W. Lown, *Can. J. Chem.*, 1964, **42**, 893. b) R. A. Wohl, *Chimia*, 1964, **18**, 219.

Received, 8th November, 1995