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Premnaodorosides D–G: acyclic monoterpenediols iridoid glucoside diesters from leaves of *Premna subscandens*

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Abstract

From leaves of *P. subscandens*, harvested in Ishigaki Island, three iridoid glucoside diesters of 3,7-dimethyloctan-1,8-diol, named premnaodorosides D, E and F, and an iridoid glucoside diester of 3,7-dimethyloctan-2,6-dien-1,8-diol (premnaodoroside G) were isolated together with known congeners, premnaodorosides A, B and C. Their structures were determined by spectroscopic methods. © 1999 Elsevier Science Ltd. All rights reserved.

Keywords: *Premna subscandens*; Verbenaceae; Iridoid glucoside; 3,7-Dimethyloctan-1,8-diol iridoid glucoside diester; 3,7-Dimethyloctan-2,6-dien-1,8-diol iridoid glucoside diester; 10-Hydroxygeraniol; Premnaodorosides D–G

1. Introduction

In previous work on *Premna odorata* Blanco, a plant cultivated in the Phillipines for medicinal use, the acyclic monoterpenediol iridoid glucoside diesters, premnaodorosides A (**1**), B (**2**) and C (**3**), were isolated (Otsuka et al., 1992). Although secoiridoid glucoside diesters to cyclic monoterpenic alcohols (Kitagawa et al., 1988; Muller & Weigend, 1998; Tanahashi et al., 1996) and *p*-hydroxyphenylethanol (Benkrief et al., 1998) have been isolated from some species, so far as we know, iridoid glucoside diesters to an acyclic monoterpenic alcohol have only been isolated from the genus *Premna*. A related species, *P. subscandens* Merr., harvested in Ishigaki Island, Okinawa, Japan, drew our attention and has afforded various 10-*O*-acylated iridoid glucosides (Sudo et al., 1997a, 1998) and phenylethanoids (Sudo et al., 1997b). Further extensive phytochemical work enabled us to isolate four acyclic monoterpene iridoid glucoside diesters, which were named premnaodorosides D–G (**4**–**7**), together known premnaodorosides A–C (**1**–**3**). Their structures were elucidated by spectroscopic methods.

2. Results and discussion

Four new and three known iridoid glucosides of acyclic monoterpene diesters were isolated from the *n*-BuOH-soluble fraction of a MeOH extract of leaves of *P. subscandens* by Diaion HP-20, Si gel and ODS CC, DCCC and prep. HPLC (see Section 3). The structures of known compounds, premnaodorosides A–B (**1**–**3**), were elucidated by comparison of their spectroscopic data with those reported (Sudo et al., 1997a).

Premnaodoroside D (**4**), $[\alpha]_D$ −89.4°, whose elemental composition was determined to be $C_{42}H_{66}O_{20}$ from its HR-FAB mass spectrum was obtained as an amor-

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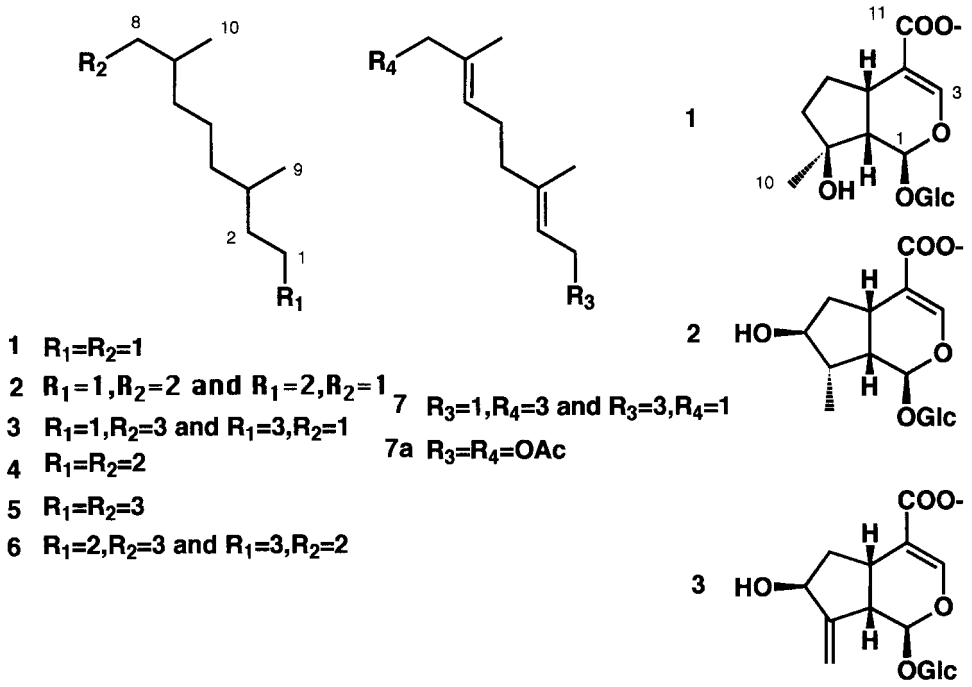

phous powder. The UV absorption maximum at 238 nm and the IR absortion bands at 1680, 1630 and 1280 cm^{-1} suggested that **4** had a conjugated double bond and enol ether, which are characteristic of the chromophore, –OCO–C=CH–O–, in iridoids. The absorption intensity of the UV spectrum ($\log \varepsilon = 4.28$) indicated the presence of two units of iridoid skeleton in **4**. The ^{1}H NMR and ^{13}C NMR spectra showed similar features to those of premnaodorosides A and B (**1** and **2**) from a related Philippine medicinal plant, *P. odorata*, except that in **4**, the two iridoid moieties must be 8-epiloganic acid as shown by the ^{13}C NMR data (Table 1). The central acyclic monoterpene moiety was assumed to be the same as that found in **1**, **2** and **3** due to the close similarity of the NMR signals. Therefore, the structure of **4** was concluded to be the 1,8-diester of the 8-epiloganic acid of 3,7-dimethyloctan-1,8-diol.

Premnaodoroside E (**5**), $[\alpha]_D$ −28.9°, was obtained as an amorphous powder and its elemental composition was determined to be $C_{42}H_{62}O_{20}$, which is four mass units less than **4**. The NMR spectra indicated that **5** is a similar compound to **4** and two triplet sp^2 carbon signals (δ_C 113.2 and 113.2) were observed. Therefore, premnaodoroside E has two gardoside moieties per molecule as shown (**5**).

Premnaodoroside F (**6**) was isolated as an inseparable isomeric mixture, whose elemental composition was determined to be $C_{42}H_{64}O_{20}$ by HR-FAB mass spectrometry. The NMR spectra showed that **6** is analogous to **2** and **3**. The structure of **6** is 3,7-dimethyloctan-1,8-diol esterified with one moiety each of 8-epiloganic acid and gardoside and judging from the integrations of the ^{1}H NMR spectra, two possible positional isomers are present in a nearly 1:1 ratio.

Premnaodoroside G (**7**) was also isolated as an inseparable isomeric mixture, whose elemental composition was determined to be $C_{42}H_{60}O_{20}$. The spectroscopic data for **7** indicated that it is analogous to premnaodoroside C (**3**) with mussaenosidic acid and gardoside as the iridoid glucoside portion. The NMR spectra showed that the acyclic monoterpene contained two double bonds (δ_C 120.5 and 142.7, and 129.5 and 132.1), and upon comparing the NMR spectral data for the central monoterpene moiety of **7a** with those of a synthetic sample of 10-hydroxygeraniol diacetate (**7a**), the signals were found to be essentially superimposable. The structure is as shown (**7**).

3. Experimental

3.1. General

^{1}H NMR and ^{13}C NMR at 400 and 100 MHz, respectively, with TMS as an internal standard; all other instrumentation was the same as reported in the previous paper (Sudo et al., 1997a); the 3,7-dimethylocta-2,6-diene-1,8-diol 1-*O*-acetate was from a previous experiment (Otsuka et al., 1992).

3.2. Plant material

The plant material was collected in Ishigaki Island,

Table 1
^{13}C NMR data for premnaodorosides D–G (**4**–**7**) and the reference compound (**7a**) (CD_3OD, 100 MHz)

Carbon No.	4	5	6	7	7a
1	63.6	63.7	1a, 1b 63.5	61.8	1 62.2
2	36.8	36.8	2a, 2b 36.8	120.5	2 120.3
3	31.1	31.1	3a, 3b 31.0	142.7	3 142.7
4	38.1	38.1	4a, 4b 38.1	39.9	4 40.0
5	25.2	25.2	5a, 5b 25.2	26.9	5 26.9
6	34.8	34.8	6a, 6b 34.7	129.5	6 129.7
7	33.9	34.0	7a, 7b 33.9	132.1	7 132.0
8	70.0	70.1	8a, 8b 70.0	70.7	8 71.1
9	19.9	20.0	9a, 9b 19.9, 20.0	16.5	9 16.4
10	17.5	17.5	10a,10b 17.5, 17.5	14.2	10 14.0
					MeCO 20.9, 20.9
					MeCO 172.8, 172.9
Iridoid aglucone					
1a, 1b	96.2, 96.3	96.7	1a 96.2 1b 96.6 1a' 96.3 1b' 96.7	1a, 1a' } 95.5	1b, 1b' } 96.7
3a, 3b	152.3, 152.4	153.5	3a 152.3 3b }153.4 3a' 152.4 3b'	3a, 3a' }152.0	3b, 3b' }153.6
4a, 4b	114.2, 114.3	111.9	4a }114.2 4b }111.9 4a' 4b'	4a, 4a' }113.7	4b, 4b' }111.9
5a, 5b	31.0, 31.1	31.9, 32.0	5a 31.0 5b 31.9 5a' 31.1 5b' 32.0	5a, 5a' } 32.1	5b, 5b' } 32.0
6a, 6b	41.3, 41.4	40.7, 40.8	6a 41.3 6b 40.7 6a' 41.4 6b' 40.8	6a 31.0 6a' 30.9	6b, 6b' } 40.0
7a, 7b	79.2, 79.3	73.9	7a } 79.2 7b, 7b' } 73.8 7a'	7a, 7a' } 40.8	7b, 7b' } 73.9
8a, 8b	45.2, 45.3	152.8	8a 45.2 8b }152.7 8a' 45.3 8b'	8a, 8a' } 80.6	8b, 8b' }152.8
9a, 9b	43.0	45.0	9a, 9a' } 43.0 9b, 9b' } 44.9	9a, 9a' } 52.4	9b, 9b' } 45.0
10a,10b	14.4	113.2, 113.3	10a, 10a' } 14.4 10b, 10b' }113.2	10a, 10a' } 24.7	10b, 10b' }113.2
11a,11b	169.0, 169.1	168.9	11a 169.0 11b }168.8 11a' 169.1 11b'	11a, 11a' }168.6	11b, 11b' }169.0
Glucose					
1'a,1'b	99.8	99.9	1'a,1'a',1'b,1'b' 99.7, 99.9	99.9, 99.9	
2'a,2'b	74.7	74.8	2'a,2'a',2'b,2'b' 74.7	74.8	
3'a,3'b	78.4	78.5	3'a,3'a',3'b,3'b' 78.4	78.4, 78.5	
4'a,4'b	71.8	71.7	4'a,4'a',4'b,4'b' 71.7, 71.7	71.7, 71.8	
5'a,5'b	78.0	78.0	5'a,5'a',5'b,5'b' 78.0	78.1	
6'a,6'b	63.0	62.9	6'a,6'a',6'b,6'b' 62.9, 63.0	62.9, 63.0	

Okinawa. The plant was identified as *P. subscandens* Merr. by one of the authors (A.T.). A voucher specimen was deposited in the Herbarium of the Institute of Pharmaceutical Sciences, Hiroshima University School of Medicine (PS-92-Okinawa).

3.3. Extraction and isolation

Parts of the extraction and isolation procedures are described in the previous paper (Sudo et al., 1997a).

The residue (10.0 g) of 60%b on Diaion HP-20 CC (Sudo et al., 1997a) was separated by silica gel (500 g) CC with increasing amounts of MeOH in $CHCl_3$ ($CHCl_3$ (2 l) and $CHCl_3$-MeOH [(99:1, 2 l), (49:1, 2 l), (24:1, 4 l), (93:7, 4 l), (9:1, 6 l), (7:1, 6 l), (17:3, 6 l), (4:1, 6 l), (3:1, 1 l) and (7:3, 1 l)], with 500 ml fractions being collected). The residue (220 mg) of fractions 65–72 was purified by DCCC (20 mg, fractions 49–56) and then HPLC [MeOH–H_2O (3:2), R_t 24 min] to give **5** (6 mg).

The residue (5.14 g) of 80%a on Diaion HP-20 CC (Sudo et al., 1997a) was separated by silica gel (400 g) CC with increasing amounts of MeOH in $CHCl_3$ ($CHCl_3$ (2 l) and $CHCl_3$–MeOH [(99:1, 2 l), (49:1, 2 l), (24:1, 4 l), (93:7, 4 l), (9:1, 4 l), (17:3, 4 l), (4:1, 4 l), (3:1, 4 l) and (7:3, 4 l)], with 500 ml fractions being collected). The residue (932 mg) of fractions 44–50 was subjected to RPCC [MeOH–H_2O (1:1, 1 l) → (9:1, 1 l)], DCCC [$CHCl_3$–MeOH–H_2O–*n*-PrOH (9:12:8:2)] and then HPLC (ODS, MeOH–H_2O) to give **1** (201 mg), **2** (103 mg), **3** (68 mg), **4** (17 mg), **6** (30 mg) and **7** (6 mg).

3.4. Known compounds isolated

Premnaodoroside A (**1**), $[\alpha]_D^{22}$ −96.9° (MeOH, *c* 2.07); premnaodoroside B (**2**), $[\alpha]_D^{22}$ −85.7° (MeOH, *c* 1.14); premnaodoroside C (**3**), $[\alpha]_D^{22}$ −56.5° (MeOH, *c* 1.59). All the spectroscopic data for these compounds were essentially the same as the reported values (Otsuka et al., 1992).

3.5. Premnaodoroside D (*4*)

Amorphous powder, $[\alpha]_D^{25}$ −89.4° (MeOH, *c* 1.04), IR ν_{max}^{KBr} cm^{-1} 3350, 2900, 1680, 1630, 1455, 1375,

1280, 1185, 1150, 1070, 900, 765; UV λ_{max}^{MeOH} nm (log ε), 238 (4.28); ^{1}H NMR (CD_3OD): 0.93 and 0.97 [both (3H, d, J = 6 Hz), H_3-9 and 10], 1.04 and 1,05 [both (3H, d, J = 7 Hz), H_3-10a and 10b], 1.83 (2H, m, H-6aa and 6ab), 2.08 (2H, ddd, J = 5, 9 and 18 Hz, H-6ab and 6bb), 2.12 (2H, br sep., J = H8a and 8b), 2,59 (2H, dt, J = 4 and 8 Hz, H-9a and 9b), 3,05 (2H, m, H-5a and H-5b), 3.18 (2H, dd, J = 8 and 9 H, H-2′a and 2′b), 3.24 (2H, t, J = 9 Hz, H-3′a and 3′b), 3.37 (2H, t, J = 9 Hz, H-4′a and 4′b), 3.65 (2H, dd, J = 6 and 12 Hz, H-6′aa and 6′ab), 3.83 (2H, td, J = 5 and 10 Hz, H-7a and 7b), 3.90 (2H, dd, J = 2 and 12 Hz, H-6′ba and 6′bb), 3.93 and 3.99 [(H, dd, J = 7 and 11 Hz) and (H, dd, J = 6 and 11 Hz), H_2-8], 4.16 (2H, t, J = 7 Hz, H_2-1), 4.655 and 4.660 [both (H, d, J = 8 Hz), H-1′a and 1′b], 5.509 and 5.510 [both (H, d, J = 4 Hz), H-1a and 1b], 7.381 and 7.397 [both (H, s), H-3a and 3b]; ^{13}C NMR spectral data: see Table 1; HR-FABMS (negative centroid) m/z: 889.4084 $[M–H]^-$ ($C_{42}H_{65}O_{20}$ requires 889.4070).

3.6. Premnaodoroside E (**5**)

Amorphous powder, $[\alpha]_D^{25}$ −28.9° (MeOH, c 0.28), UV λ_{max}^{MeOH} nm (log ε): 206 (4.07), 236 (4.31); ^{1}H NMR (CD_3OD): 0.93 and 0.97 [both (3H, d, J = 6 Hz, H_3-9 and 10), 2.00 (4H, m, H_2-6a and 6b), 2.90 (2H, m, H-5a and 5b), 3.16 (2H, v br m, H-9a and 9b), 3.20 (2H, dd, J = 8 and 9 Hz, H-2′a and 2′b), 3.25 (2H, t, J = 9 Hz, H-3′a and 3b′), 3.37 (2H, t, J = 9 Hz, H-4′a and 4′b), 3.64 (2H, d, dd, J = 6 and 12 Hz, H-6′aa and 6′ab), 3.89 (2H, dd, J = 2 and 12 Hz, H-6′ba and 6′bb), 3.94 and 4.10 [both (H, dd, J = 6 and 11 Hz, H_2-8], 4.17 (2H, t, J = 6 Hz, H_2-1), 4.37 (2H, v br s, H-7a and 7b), 4.671 and 4.677 [both H, d, J = 8 Hz), H-1′a and 1′b), 5.36 (4H, br s, H_2-10a and 10b), 5,42 (2H, d, J = 5 Hz, H-1a and 1b), 7.43 and 7.45 [both (H, d, J = 1 Hz), H-3a and 3b]; ^{13}C NMR spectral data: see Table 1; HR-FABMS (negative centroid) m/z: 885.3744 $[M–H]^-$ ($C_{42}H_{61}O_{20}$ requires 885.3756).

3.7. Premnaodoroside F (**6**)

Amorphous powder, $[\alpha]_D^{25}$ −69.1° (MeOH, c 1.88), IR ν_{max}^{KBr} cm^{-1} 3350, 2900, 1680, 1630, 1450, 1375, 1280, 1150, 1070, 900, 850, 765; UV λ_{max}^{MeOH} nm (log ε): 236 (4.33); ^{1}H NMR (CD_3OD): δ 0.935 (1/2H_3), 0.936 (1/2H_3) and 0.966 (3H) [all (d, J = 7 Hz), 1/2H_3-9{a}, 9{b}, 10{a} and 10{b}], 1.047 and 1.04 [both (1/2 H_3, d, J = 7 Hz), 1/2 H_3-10a and 10a′), 1.83 (H, m, 1/2Ha-6a and 6a′), 2.00 (2H, m, 1/2Ha-6b and 6b′, and 1/2Hb-6b and 6b′), 2.08 (H, ddd, J = 5, 9 and 12 Hz, Hb-6a and 6a′), 2.12 (H, br sep., J = 6 Hz, 1/2H-8a and 8a′), 2.59 (H, dt, J = 4 and 8 Hz, 1/2H-9a and 9a′), 3.00 (H, m, 1/2H-5b and 5b′), 3,05 (H, m, 1/2H-5a and 5a′), 3.37 and 3.38 [both (H, t, J = 9 Hz), H-4′], 3.64 [2H, dd, J = 6 and 12 Hz, Ha-6′], 3.83 [(H, td, J = 5 and 10 Hz), H-7a and 7a′], 3.89 and 3.90 [both (H, dd, J = 2 and 12 Hz), Hb-6′], 3.92 and 3.94 [both (l/2H, dd, J = 6 and 11 Hz), 1/2H-8{a}a and 8{b}a], 3.99 and 4.10 [both (l/2H, dd, J = 6 and 11 Hz, 1/2H-8{a}b and 8{b}b], 4.15 and 4.17 [both (H, t, J = 6 Hz), 1/2H_2-1{a} and 1/2H_2-1{b}], 4.37 (H, br m, H-7b and 7b′), 4.655, 4.661, 4.672 and 4.677 [all (l/2H, d, J = 8 Hz), H-1′a, 1′a′, 1′b and 1′b′], 5.36 (H, br s, 1/2H-10b and 10b′), 5.43 (H, d, J = 5 Hz, 1/2H-1b and 1b′), 5.507 and 5.510 [both (1/2H, dd, J = 4 Hz), H-1a and 1a′], 7.38, 7,40, 7.43, and 7.45 [(l/2H, s, (1/2H, d, J = 1 Hz), (1/2H, d, J = 1 Hz) and (1/2H, d, J = 1 Hz), H-3a, 3a′, 3b and 3b′]; ^{13}C NMR spectral data: see Table 1; HR-FABMS (negative centroid) m/z: 887.3914 $[M–H]^-$ ($C_{42}H_{63}O_{20}$ requires 887.3913).

3.8. Premnaodoroside G (**7**)

Amorphous powder, $[\alpha]_D^{25}$ −46.6° (MeOH, c 0.34), UV λ_{max}^{MeOH} nm (log ε): 207 (4.03), 236 (4.19); ^{1}H NMR (CD_3OD): δ 1.32 (3H, s, 1/2H_3-10a and 10a′), 1.67 and 1.73 [both (3H, s) 1/2H_3-9{a} and 9{b} and 1/2H_3-10{a} and 10{b}], 1.99 (2H, m, 1/2Ha-6b and 6b′ and 1/2Hb-6b and 6b′), 3.00 (H, m, H-5b and 5b′), 3.65 (2H, dd, J = 6 and 12 Hz, Ha-6′a, 6′a′, 6′b and 6′b′), 3.89 (2H, dd, J = 2 and 12 Hz, 1/2Hb-6′a, 6′a′, 6′b and 6′b′), 4.37 (H v br s, 1/2H-7b and 7b′), 4.68 (2H, d, J = 8 Hz, 1/2H-1′a, 1′a′, 1′b and 1′b′), 5.36 (2H, br s, 1/2Ha-10b and 10b′, and 1/2Hb-10b and 10b′), 5.43 and 5.44 [both (H, d, J = 5 Hz), H-1a, la′, 1b and lb′), 7.39 (H, s, 1/2H-3a and 3a′), 7.45 (H, d, J = 1 Hz, 1/2H-3b and 3b′); ^{13}C NMR spectral data: see Table 1; HR-FABMS (negative centroid) m/z: 883.3606 $[M–H]^-$ ($C_{42}H_{59}O_{20}$ requires 883.3600).

3.9. Acetylation of 3,7-dimethytocta-2E,6E-diene-1,8-diol 1-O-acetate (**7a**)

The monoacetate (50 mg) was acetylated with 200 μl each of acetic andydride and pyridine at 60° for 1 h. The reagents were evaporated off under a stream of N_2 and the residue was purified by prep. TLC [precoated silica gel plate, thickness 0.50 mm, developed with C_6H_6– $(CH_3)_2O$ (9:1), eluted with $CHCl_3$–MeOH (9:1), visualized by spraying with water] to give the diacetate (**7a**) as a colourless liquid. ^{1}H NMR (CD_3OD): δ 1.65 and 1.71 (each 3H, each br s, H_3-3 and 7), 2.01 and 2.04 (each 3H, each s, CH_3CO- × 2), 2.1–2.2 (4H, m, H_2-4 and 5), 4.54 (2H, s, H_2-8), 4.58 (2H, d, J = 7 HZ, H_2-1), 5,34 (1H, qt, J = 1 and 8 Hz, H-6), 5,42 (H, qt, J = 1 and 7 Hz, H-2); ^{13}C NMR spectral data (CD_3OD): see Table 1.

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