Phytochemistry 69 (2008) 2336–2340

Contents lists available at ScienceDirect

Phytochemistry

journal homepage: www.elsevier.com/locate/phytochem

18-*nor*-Podocarpanes and podocarpanes from the Bark of *Taiwania cryptomerioides*

Shih-Chang Chien [a], Cheng-Chi Chen [b], Hsi-Lin Chiu [b], Chi-I Chang [c], Mei-Hwei Tseng [d], Yueh-Hsiung Kuo [a,b,e,f,*]

[a] *Tsuzuki Institute for Traditional Medicine, College of Pharmacy, China Medical University, Taichung 404, Taiwan, ROC*
[b] *Department of Chemistry, National Taiwan University, Taipei, Taiwan, ROC*
[c] *Graduate Institute of Biotechnology, National Pingtung University of Science and Technology, Pingtung 912, Taiwan, ROC*
[d] *Department of Science, Taipei Municipal University of Education, Taipei 100, Taiwan, ROC*
[e] *Center for Food and Biomolecules, National Taiwan University, Taipei 106, Taiwan, ROC*
[f] *Agricultural Biotechnology Research Center, Academia Sinica, Taipei 115, Taiwan, ROC*

ARTICLE INFO

Article history:
Received 19 February 2008
Received in revised form 12 May 2008
Available online 27 July 2008

Keywords:
Taiwania cryptomerioides
Taxodiaceae
nor-Podocarpane

ABSTRACT

Seven nor- and podocarpane-type diterpenes were isolated from the bark of *Taiwania cryptomerioides* Hayata, including three 18-*nor*-podocarpanes: 18-*nor*-1β,4α,14-trihydroxy-13-methoxy-8,11,13-podocarpatriene (**1**), 18-*nor*-1β,4α,13,14-tetrahydroxy-8,11,13-podocarpatrien-7-one (**2**), 18-*nor*-1β,4α,14-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**3**), 1β,14,19-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**4**), 1β,13,14,18-tetrahydroxy-8,11,13-podocarpatrien-7-one (**5**), 18-acetoxy-1β,13,14-trihydroxy-8,11,13-podocarpatrien-7-one (**6**), and 1β,14,18-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**7**). Their structures were determined by application of 1D and 2D NMR spectroscopy and other techniques. Podocarpane-type diterpenes do not occur extensively in nature, and the presumed oxidative enzyme in this plant will be of interest to identify.

1. Introduction

Taiwania cryptomerioides Hayata (Taxodiaceae) is a monotypic genus endemic plant in Taiwan which grows at elevations from 1800 to 2600 m in Taiwan's central mountains. It is now a common plantation species and was once an important building material. The heartwood of *T. cryptomerioides* is yellowish-red with distinct purplish-pink streaks. In earlier investigations, various sesquiterpenes, lignans, and abietane-type diterpenes were isolated and identified from its heartwood (Cheng et al., 1967; Kuo et al., 1969; Lin et al., 1968) and bark (Kuo et al., 1979, 1985). Podocarpane-type diterpenes do not occur extensively in nature and are only present in several genera including *Azadirachta* (Ara et al., 1988a,b, 1990; Majumder et al., 1987; Siddiqui et al., 1988), *Humiriantther* (Zoghbi et al., 1981), *Micrandropsis* (Alvarenga et al., 1981), and *Podocarpus* (Cambie and Mander, 1962). Lin et al. (1998) first isolated a podocarpane derivative, 1β,13,14-trihydroxy-8,11,13-podocarpatrien-7-one from the leaves of *T. cryptomerioides*, where they also found many other compounds with unusual skeletons (Lin et al., 1995, 1996, 1997, 1998). As a result, we were encouraged to look further at the plant bark, and found many podocarpane-type trinorditerpenes in *T. cryptomerioides* (Kuo et al., 2000a, 2002a,b; Kuo and Chang, 2000b; Kuo and Chien, 2001). Here, the detailed structures from more polar fractions of the previous extract were identified.

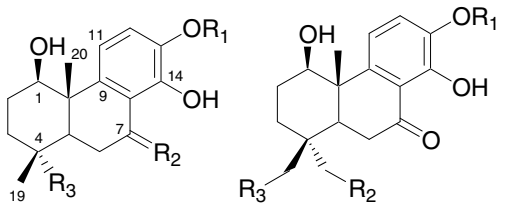
2. Results and discussion

Seven unusual *nor*-podocarpane and podocarpane compounds were isolated from the more polar fractions of the bark of *T. cryptomerioides* Hayata, namely 18-*nor*-1β,4α,14-trihydroxy-13-methoxy-8,11,13-podocarpatriene (**1**), 18-*nor*-1β,4α,13,14-tetrahydroxy-8,11,13-podocarpatrien-7-one (**2**), 18-*nor*-1β,4α,14-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**3**), 1β,14,19-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**4**), 1β,13,14,18-tetrahydroxy-8,11,13-podocarpatrien-7-one (**5**), 18-acetoxy-1β,13,14-trihydroxy-8,11,13-podocarpatrien-7-one (**6**), and 1β,14,18-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**7**) (see Fig. 1).

The first three compounds (**1**, **2**, and **3**) are 18-*nor*-podocarpane-type diterpenes. Compound **1** was assigned the molecular formula of $C_{17}H_{24}O_4$, based on peak matching of the molecular ion and application of ^{13}C NMR spectroscopy. The IR spectrum of **1** displayed a prominent hydroxyl peak group (3400 cm^{-1}). The ^1H NMR (Table 1) spectrum showed singlet methyl groups at δ 1.16, 1.18, and 3.80 (OCH_3) and two *ortho*-coupled phenyl protons at δ 7.75 and 6.65 (*d*, J = 8.9 Hz, H-11, -12). No isopropyl group and no typical H_β-1 resonance (δ 2.00–2.40)

* Corresponding author. Address: Tsuzuki Institute for Traditional Medicine, College of Pharmacy, China Medical University, Taichung 404, Taiwan, ROC. Tel.: +886 2 33661671; fax: +886 2 23636359.
E-mail address: yhkuo@ntu.edu.tw (Y.-H. Kuo).

0031-9422/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.phytochem.2008.06.005



	R_1	R_2	R_3		R_1	R_2	R_3
1	CH_3	H_2	OH	**4**	CH_3	H	OH
2	H	O	OH	**5**	H	OH	H
3	CH_3	O	OH	**6**	H	OAc	H
8	CH_3	H_2	CH_3	**7**	CH_3	OH	H

Fig. 1. Compounds **1**–**8**.

for dehydroabietane and dehydropodocarpane-type derivatives (Lin et al., 1998; Kuo et al., 2000a) were observed. Of the 17 ^{13}C NMR signals (Table 2) noted, six belonged to phenyl carbons. Three of the six phenyl signals (singlet) appeared at lower field, δ 144.2, 146.0, and 144.1, and those were assigned to C-9, C-13, and C-14, respectively. A carbinol resonance at δ 3.73 (*dd*, J = 11.0, 4.7 Hz) was assigned as H_α-1 (axial), as it had NOESY correlation with H-5 (δ 1.49) and H-11 (δ 7.75, *d*, J = 8.9 Hz). Comparisons of the ^1H and ^{13}C NMR spectroscopy data (Table 2) between **1** and **8** (Kuo and Chang, 2000b) showed the only difference was a hydroxyl group instead of a methyl group in compound **1**. The signal at δ 7.75 exhibiting NOESY correlation with H-20 (δ 1.16 was unambiguously consistent with H-11. NOESY also showed a correlation between H-12 (δ 6.66) and OCH_3 (δ 3.80) pinpointing the position of 13-OCH_3. No NOESY correlation was observed between H-5 and H-19 (and H-20) that showed H-5, H-19, and H-20 to all be axially aligned. Therefore, compound **1** was 18-*nor*-1β,4α,14-trihydroxy-13-methoxy-8,11,13-podocarpatriene, the first example of an 18-*nor*-podocarpane skeleton found in nature.

Table 2
^{13}C NMR spectral data for compounds **1**–**7** (100 MHz, **1**, **2** in CD_3OD, **3**–**7** in $CDCl_3$)

No.	1	2	3	4	5	6	7
1	78.4	77.0	75.8	76.5	76.0	75.8	75.9
2	32.0	31.5	31.1	29.6	29.2	29.0	29.1
3	41.5	41.0	40.2	33.0	32.8	33.2	32.7
4	72.8	71.8	71.0	38.0	37.5	36.5	37.5
5	52.2	51.3	49.5	48.7	41.9	42.7	41.4
6	18.5	36.0	34.8	35.7	35.4	35.4	35.5
7	25.4	207.9	205.6	205.6	205.8	205.2	205.9
8	124.4	116.9	115.5	115.3	115.3	115.2	115.5
9	144.2	148.3	146.1	146.8	146.7	146.4	146.8
10	44.4	44.8	43.4	43.7	43.3	43.4	43.2
11	119.2	117.8	115.7	115.9	116.6	116.6	115.8
12	109.9	122.9	117.7	117.9	121.0	121.0	117.8
13	146.0	145.1	146.8	146.8	143.1	143.2	146.5
14	144.1	152.0	153.1	153.0	149.4	149.4	152.8
18				25.9	70.4	71.0	70.2
19	22.7	22.5	22.6	64.6	17.0	17.1	17.0
20	18.9	17.1	16.3	17.6	17.4	17.5	17.3
OCH_3	56.6		56.1	56.2			56.1
$OCOCH_3$						20.9	
$OCOCH_3$						170.9	

The molecular formula ($C_{16}H_{20}O_5$), UV (λ_{max} 218.0, 274.0, and 363.0 nm) and IR (3395, 1636, and 1510 cm^{-1}) spectrum of compound **2** suggested that it contains conjugated ketone, aromatic, and hydroxyl groups. A comparison of the ^1H and ^{13}C NMR spectra of compounds **1** and **2** suggests that **2** was also a *nor*-podocarpane-type diterpene. The UV maximum absorption bands and strong hydrogen bonding absorption (2500–3300 cm^{-1}) indicated a carbonyl group located at the C-7 position. The ^1H NMR signals at δ 1.19 and 1.24 (3H each, *s*) were assigned as H-20 and H-19, respectively, and the resonances at δ 7.70 and 6.95 (1H each, *d*, J = 8.5 Hz) were assigned as *ortho*-phenyl protons. The former phenyl proton displayed in the lower field was attributed to deshielding by the receiving 1_β-hydroxyl group, as in compounds **1** and **8** (Kuo and Chang, 2000b). The signal at δ 3.91 was assigned as H_α-1 from NOESY correlation with H-5 (δ 2.03) and H-11. The absence of any NOESY correlation between H-19 and H-5 indicated that H-19 was in a β-axial orientation. The HMBC spectrum exhibited

Table 1
^1H NMR spectroscopic data for compounds **1**–**7** (400 MHz, **1**, **2** in CD_3OD, **3**–**7** in $CDCl_3$)

No.	1	2	3	4	5	6	7
1	3.73 *dd* (11.0, 4.7)	3.91 *dd* (11.2, 4.5)	4.05 *dd* (11.1, 4.5)	4.03 *dd* (10.8, 5.1)	3.95 *dd* (9.2, 6.2)	3.99 *dd* (9.9, 5.8)	3.90 *dd* (9.7, 6.2)
2	1.75 *m*	1.85, 1.72 *m*	1.92, 1.70 *m*	1.78 *m*	1.83 *m*	1.84 *m*	1.80 *m*
3	1.55, 1.76 *m*	1.59, 1.78 *m*	1.60 *m*	1.90 *m*	1.69 *m*	1.63 *m*	1.67 *m*
4			1.86 *dt* (12.6, 3.4)		1.35 *br d* (13.2)	1.44 *dt* (13.6, 3.4)	1.31 *dt* (13.3, 3.2)
5	1.49 *dd* (12.6, 2.1)	2.03 *dd* (12.8, 4.7)	2.07 *dd* (13.8, 3.8)	1.92 *dd* (14.0, 3.6)	2.20 *dd* (13.6, 4.0)	2.12 *dd* (13.7, 4.0)	2.14 *dd* (13.5, 4.3)
6	1.69, 2.12 *m*	2.87 *dd* (18.6, 4.7)	2.98 *dd* (18.6, 3.8)	2.76 *dd* (18.6, 3.6)	2.63 *dd* (18.7, 4.0	2.63 *dd* (18.7, 4.0)	2.61 *dd* (18.7, 4.3)
		2.81 *dd* (18.6, 12.8)	2.75 *dd* (18.6, 13.8)	2.89 *dd* (18.6, 14.0)	2.75 *dd* (18.7, 13.6	2.79 *dd* (18.7, 13.7)	2.73 *dd* (18.7, 13.5)
7	2.59 *ddd* (17.8, 11.3, 8.3)						
	2.86 *dd* (17.8, 5.9)						
8							
9							
10							
11	7.75 *d* (8.9)	7.70 *d* (8.5)	7.68 *d* (8.6)	7.67 *d* (8.6)	7.63 *d* (8.6)	7.65 *d* (8.6)	7.62 *d* (8.7)
12	6.66 *d* (8.9)	6.95 *d* (8.5)	6.98 *d* (8.6)	6.98 *d* (8.6)	7.02 *d* (8.6)	7.05 *d* (8.6)	6.95 *d* (8.7)
13							
14							
18				1.01 *s*	3.15 *d* (10.9)	3.70 *d* (11.4)	3.10 *d* (11.0)
					3.40 *d* (10.9)	3.79 *d* (11.4)	3.37 *d* (11.0)
19	1.18 *s*	1.24 *s*	1.28 *s*	3.62 *d* (10.8)	0.92 *s*	0.99 *s*	0.87 *s*
				3.82 *d* (10.8)			
20	1.16 *s*	1.19 *s*	1.19 *s*	1.22 *s*	1.23 *s*	1.24 *s*	1.20 *s*
OCH_3	3.80 *s*		3.86 *s*	3.85 *s*			3.83 *s*
13-OH			13.03 *s*	12.94 *s*	12.74 *s*	12.72 *s*	12.88 *s*
14-OH					5.60 *br s*	5.57 *s*	
$OCOCH_3$						2.00 *s*	

the following correlations: H-19/C-3, C-4, C-5; H-20/C-1, C-5, C-9, C-10, confirming **2** to be 18-*nor*-1β,4α,13,14-tetrahydroxy-8,11,13-podocarpatrien-7-one.

The molecular formula for compound **3** is $C_{17}H_{22}O_5$ based on its HREIMS and ^{13}C NMR data. A comparison of the ^1H and ^{13}C NMR spectra of **3** and **2** showed the only difference to be a methoxy group at C-13 in **3** replacing a hydroxyl group in **2**. The UV absorptions (λ_{max} 216.5, 271.0, and 361.5 nm) and the signal at δ 13.03 (exchangeable with D_2O) confirmed the presence of the C-7 carbonyl and C-14 hydroxyl groups. The lower field singlet methyl group at H-19 (δ 1.28) indicated an adjacent hydroxyl group. Two *ortho*-phenyl protons were observed at δ 7.68 (1H, *d*, *J* = 8.6 Hz) and 6.98 (1H, *d*, *J* = 8.6 Hz). The former proton was assigned as H-11 due to its NOESY correlation with H-1 (δ 4.05); the latter phenyl proton had a NOESY correlation with a phenolic methyl (δ 3.86) group. Based on the chemical shifts and coupling patterns, the ABX system signals at δ 2.07 (1H, *dd*, *J* = 13.8, 3.8 Hz), 2.98 (1H, *dd*, *J* = 18.6, 3.8 Hz), and 2.75 (1H, *dd*, *J* = 18.6, 13.8 Hz) were assigned as H-5, H_α-6 and H_β-6, respectively. Taken together with the HMBC, NOESY, and COSY spectra, these data confirmed **3** to be 18-*nor*-1β,4α,14-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one.

Eighteen ^{13}C NMR signals and the exact mass spectrum data confirmed the molecular formula of **4** to be $C_{18}H_{24}O_5$. Three kinds of functional absorption bands (aromatic, conjugated carbonyl, and hydroxyl) are present in its IR spectrum. Two singlet methyl groups at δ 1.01 (H-18) and 3.85 (OCH_3) and two *ortho*-coupling phenyl protons at δ 7.67 and 6.98 (*d*, *J* = 8.6 Hz, H-11, -12) (Table 1) were observed in the ^1H NMR spectrum. Three of six phenyl signals (singlet) appearing at δ 146.8, 146.8, and 153.0 were assigned as C-9, C-13, and C-14, respectively. The methoxy group with a NOESY correlation with resonance δ 6.98 (H-12) suggested methoxy and hydroxyl groups located at C-13 and -14, respectively. A signal at δ 4.03 was assigned as H-1 as it had a NOESY correlation with H-11. Based on the chemical shift and coupling pattern, the ABX system signals at δ 1.92 (1H, *dd*, *J* = 14.0, 3.6 Hz), 2.76 (1H, *dd*, *J* = 18.6, 3.6 Hz), and 2.89 (1H, *dd*, *J* = 18.6, 14.0 Hz) were assigned as H-5, H_α-6, and H_β-6, respectively. The presence of an hydroxymethyl groups was established from the following data: δ 3.62 and 3.82 (1H each, *d*, *J* = 10.8 Hz) and δc 64.6. The protons at δ 3.62 and 3.82 showed NOESY correlations with H-20 (δ 1.22) confirming the position of the primary hydroxyl group at C-19. Therefore, **4** was identified as 1β,14,19-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one.

The molecular formula ($C_{17}H_{22}O_5$), UV (λ_{max} 224.0, 279.0, and 354.0 nm) and IR (3412, 1634, 1608, and 1510 cm^{-1}) data of compound **5** suggested that it contained conjugated ketone, aromatic, and hydroxyl groups. Two singlet methyl groups at δ 0.92 (H-19), 1.23 (H-20), and two *ortho*-coupling phenyl protons at δ 7.63 and 7.02 (*d*, *J* = 8.6 Hz, H-11, -12) (Table 1) were observed in its ^1H NMR spectrum. The typical ABX system signals at δ 2.20 (1H, *dd*, *J* = 13.6, 4.0 Hz), 2.63 (1H, *dd*, *J* = 18.7, 4.0 Hz), and 2.75 (1H, *dd*, *J* = 18.7, 13.6 Hz) were assigned as H-5, H_α-6, and H_β-6, respectively. Two exchangeable phenolic and hydroxyl protons were present at δ 12.74 (1H, *s*) and 5.60 (1H, *br s*), respectively. The H-1 (δ 3.95) resonance exhibited NOESY correlation with H-11 (δ 7.63), which also showed a NOESY correlation with resonance at δ 1.23 (3H, *s*). Therefore, this methyl group was assigned as H-20. The signals at δ 3.40, 3.15 (1H each, *d*, *J* = 10.9 Hz), and δc 70.4 confirmed the presence of an hydroxymethyl group. The hydroxyl group located at C-18 was attributable to NOESY correlation between H-20 and H-19 (δ 0.92, 3H, *s*). Therefore, structure **5** was 1β,13,14,18-tetrahydroxy-8,11,13-podocarpatrien-7-one.

Compound **6** has the molecular formula $C_{19}H_{24}O_6$ based on HREIMS and ^{13}C NMR spectroscopic data. Two singlet methyl groups at δ 0.99 (H-19) and 1.24 (H-20), one acetoxy group at δ 2.00 and two *ortho*-coupling phenyl protons at δ 7.65 and 7.05 (*d*, *J* = 8.6 Hz, H-11, -12) (Table 1) were observed in its ^1H NMR spectrum. Typical ABX system signals at δ 2.12 (1H, *dd*, *J* = 13.7, 4.0 Hz), 2.63 (1H, *dd*, *J* = 18.7, 4.0 Hz), and 2.79 (1H, *dd*, *J* = 18.7, 13.7 Hz) were assigned as H-5, H_α-6, and H_β-6, respectively. Two exchangeable phenolic protons were present at δ 5.57 (1H, s) and δ 12.72 (1H, *s*), the latter datum confirmed the presence of 7-oxo and C-14 -OH substituents. The H-1 (δ 3.99) resonance exhibited a NOESY correlation with H-11 (δ 7.65). The resonance at δ 1.24 (s, 3H) was assigned as H-20 as it had a NOESY correlation with H-11. A comparison of the ^1H and ^{13}C NMR spectra of **6** and **5** showed that the only difference was an acetoxy group at C-18 in **6** replacing a hydroxyl group in **5**. Therefore, **6** was 18-acetoxy-1β,13,14-trihydroxy-8,11,13-podocarpatrien-7-one.

A comparison of the ^1H and ^{13}C NMR spectra of **7** and **5** shows almost all of the data to be similar. The difference between these two compounds is that **7** had an additional methyl group (δ 3.83) attached to the phenolic position. The UV absorptions (λ_{max} 218.5, 271.5, and 359.0 nm) and the signal at δ 12.88 (exchangeable with D_2O) confirmed the presence of the C-7 carbonyl and C-14 hydroxyl group. Three singlet methyl groups at δ 0.87 (H-19), 1.20 (H-20), and 3.83 (OCH_3) and two *ortho*-coupling phenyl protons at δ 7.62 and 6.95 (*d*, *J* = 8.7 Hz, H-11, -12) (Table 1) were observed in its ^1H NMR spectrum. Typical ABX system signals at δ 2.14 (1H, *dd*, *J* = 13.5, 4.3 Hz), 2.61 (1H, *dd*, *J* = 18.7, 4.3 Hz), and 2.73 (1H, *dd*, *J* = 18.7, 13.5 Hz) were assigned as H-5, H_α-6, and H_β-6, respectively. An exchangeable phenolic proton was present at δ 12.88 (1H, *s*). The H-1 (δ 3.90) resonance exhibited NOESY correlation with H-11 (δ 7.62). H-11 had NOESY correlation with resonance at δ 1.20 (*s*), so this methyl group was assigned as H-20. The hydroxyl group located at C-18 was attributable to NOESY correlation between H-20 and H-19. Therefore, structure **7** was identified as 1β,14,18-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one.

2.1. Conclusions

Podocarpane-type diterpenes do not occur extensively in nature. No podocarpane diterpenes have been discovered in parts of *T. cryptomerioides* other than the bark with the exception of one (1β,13,14-trihydroxy-8,11,13-podocarpatrien-7-one) found in the leaf. The first three compounds **1**, **2**, and **3** are 18-*nor*-podocarpane type diterpenes, with this skeleton being first reported in this study. In our previous studies of this plant, we found 21 new podocarpane derivatives including seven 1β-hydroxydehydropodocapanes in the bark. This study extends the total number of known 1β-hydroxydehydropodocarpane (including 18-*nor*-podocarpane), derivatives to 14.

Oxidation at C-1 in tricycloditerpenes is very rare. The pressured oxidative enzyme in this plant is of considerable interest because it selectively oxidizes at the C-1β position. The oxidization produced 13, 14-dioxygenation more than 12,13-dioxygenation; there was correspondingly less oxidation at C-19 than at C-18. The 1β-hydroxyl group causes the H-11 downshift to δ 7.6-7.8. It is very easy to recognize the location of a 1_β-hydroxyl group from this H-11 shift.

3. Experimental section

3.1. General experimental procedures

Melting points were determined with a Yanagimoto micromelting point apparatus and are uncorrected. IR spectra were recorded on a Perkin–Elmer 983G spectrophotometer. ^1H, ^{13}C, and DEPT spectra were acquired on a Bruker DMX-400 spectrometer, and two-dimensional NMR spectra were obtained using a Bruker

DMX-500 spectrometer. EIMS, UV, and specific rotations were determined using a JEOL JMS-HX 300, Hitachi S-3200 spectrometer, and JASCO DIP-180 digital polarimeter, respectively. Extracts were initially fractionated on silica gel (Merck 70–230 mesh, 230–400 mesh, ASTM) and then purified with a semi-preparative normal-phase HPLC column (250 × 10 mm, 7 μm, LiChrosorb Si 60) on an LDC Analytical-III system.

3.2. Plant material

Bark samples of *T. cryptomerioides* were collected in Taichung County, Taiwan, in 1996. The identity of the plant material was confirmed by Mr. Muh-Tsuen Gun, formerly of the Department of Botany, National Taiwan University. A voucher specimen (No. 013542) has been deposited at the Herbarium of the Department of Botany, National Taiwan University, Taipei, Taiwan.

3.3. Extraction and isolation

Air dried pieces of the bark of *T. cryptomerioides* (12 kg) were extracted with acetone (3 × 60 L) at room temperature (7 days for each time). The acetone extract was evaporated *in vacuo* to leave a black residue, which was suspended in H_2O (8 L), and then partitioned with EtOAc (3 × 1 L). The EtOAc fraction was subjected to silica gel cc using a hexane-EtOAc gradient solvent system and purified by repeated HPLC (normal phase on LiChrosorb Si 60) using isocratic solvent.

18-*nor*-1β,4α,14-trihydroxy-13-methoxy-8,11,13-podocarpatriene (**1**) (18.2 mg), 18-*nor*-1β,4α,13,14-tetrahydroxy-8,11,13-podocarpatrien-7-one (**2**) (60.3 mg), 18-*nor*-1β,4α,14-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**3**) (4.4 mg), 1β,14,19-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**4**) (14.6 mg), 1β,13,14,18-tetrahydroxy-8,11,13-podocarpatrien-7-one (**5**) (39.8 mg), 18-acetoxy-1β,13,14-trihydroxy-8,11,13-podocarpatrien-7-one (**6**) (18.0 mg), and 1β,14,18-trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (**7**) (63.4 mg), were eluted with 100% EtOAc.

3.4. Compound characterization

3.4.1. 18-nor-1β,4α,14-Trihydroxy-13-methoxy-8,11,13-podocarpatriene (*1*)

Colorless needle; m.p. 137–139 °C; $[\alpha]_D^{23}$ +15.1 (*c* 0.59, MeOH); UV λ_{max}^{MeOH} nm (log ε): 218.5 (3.96, *sh*), 278.5 (3.34), 363.0 (3.42); IR (film) ν_{max} 3400, 1604, 1493, 1283, 1232, 1080 cm^{-1}; for ^1H and ^{13}C NMR (CD_3OD) spectroscopic data, see Tables 1 and 2; EIMS 70 eV, *m/z* (rel. int.): 292 $[M]^+$ (43), 274 $[M-H_2O]^+$ (100), 259 $[M-H_2O-Me]^+$ (26), 233 $[M-59]^+$ (67), 230 $[M-62]^+$ (43), 175 $[M-117]^+$ (29); HREIMS *m/z* 292.1664 (calcd. for $C_{17}H_{24}O_4$, 292.1668).

3.4.2. 18-nor-1β,4α,13,14-Tetrahydroxy-8,11,13-podocarpatrien-7-one (*2*)

Pale yellow crystal; m.p. 247–249 °C; $[\alpha]_D^{23}$ −36.7 (*c* 1.85, MeOH); UV λ_{max}^{MeOH} nm (log ε): 218.0 (4.05, *sh*), 274.0 (3.90), 363.0 (3.42); IR (film) ν_{max} 3395, 1636, 1602, 1510, 1247, 1032 cm^{-1}; for ^1H and ^{13}C NMR (CD_3OD) spectroscopic data, see Tables 1 and 2; EIMS 70 eV, *m/z* (rel. int.): 292 $[M]^+$ (100), 274 $[M-H_2O]^+$ (79), 241 $[M-51]^+$ (56), 191 $[M-101]^+$ (37), 173 $[M-119]^+$ (41), 101 $[M-191]^+$ (37), 59 $[M-233]^+$ (73); HREIMS *m/z* 292.1307 (calcd. for $C_{16}H_{20}O_5$, 292.1305).

3.4.3. 18-nor-1β,4α,14-Trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (*3*)

Amorphous solid; $[\alpha]_D^{23}$ −24.8 (*c* 0.14, $CHCl_3$); UV λ_{max}^{MeOH} nm (log ε): 216.5 (4.08), 271.0 (3.64), 361.5 (3.22); IR (film) ν_{max} 3422, 1635, 1605, 1490, 1249, 1049, 757 cm^{-1}; for ^1H and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2; EIMS 70 eV, *m/z* (rel. int.): 306 $[M]^+$ (100), 231 $[M-75]^+$ (38), 205 $[M-101]^+$ (23), 190 $[M-116]^+$ (21), 173 $[M-133]^+$ (23); HREIMS *m/z* 306.1462 (calcd. for $C_{17}H_{22}O_5$, 306.1461).

3.4.4. 1β,14,19-Trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (*4*)

White powder; m.p. 130–132 °C; $[\alpha]_D^{23}$ −30.8 (*c* 0.45, $CHCl_3$); UV λ_{max}^{MeOH} nm (log ε): 223.5 (4.05), 272.0 (3.95), 358.0 (3.36); IR (film) ν_{max} 3395, 1635, 1590, 1495, 1465, 1437, 1263, 1248, 1021, 755 cm^{-1}; for ^1H and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2; EIMS 70 eV, *m/z* (rel. int.): 320 $[M]^+$ (100), 205 $[M-115]^+$ (28), 189 $[M-131]^+$ (9), 173 $[M-147]^+$ (18); HREIMS *m/z* 320.1610 (calcd. for $C_{18}H_{24}O_5$, 320.1617).

3.4.5. 1β,13,14,18-Tetrahydroxy-8,11,13-podocarpatrien-7-one (*5*)

Light yellow crystal; m.p. 206–208 °C; $[\alpha]_D^{23}$ −6.1 (*c* 1.21, $CHCl_3$); UV λ_{max}^{MeOH} nm (log ε): 224.0 (4.06, *sh*), 279.0 (3.96), 354.0 (3.30); IR (film) ν_{max} 3412, 1634, 1608, 1510, 1384, 1273, 1179, 1051, 759 cm^{-1}; for ^1H and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2; EIMS 70 eV, *m/z* (rel. int.): 306 $[M]^+$ (100), 191 $[M-115]^+$ (25), 173 $[M-133]^+$ (15), 161 $[M-145]^+$ (11); HREIMS *m/z* 306.1466 (calcd. for $C_{17}H_{22}O_5$, 306.1461).

3.4.6. 18-Acetoxy-1β,13,14-trihydroxy-8,11,13-podocarpatrien-7-one (*6*)

Amorphous solid; $[\alpha]_D^{23}$ −9.2 (*c* 0.55, $CHCl_3$); UV λ_{max}^{MeOH} nm (log ε): 220.0 (4.08), 274.5 (3.94), 361.0 (3.40); IR (film) ν_{max} 3449, 1734, 1636, 1595, 1490, 1249, 1039, 762 cm^{-1}; for ^1H and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2; EIMS 70 eV, *m/z* (rel. int.): 348 $[M]^+$ (100), 255 $[M-93]^+$ (15), 191 $[M-157]^+$ (22), 190 $[M-158]^+$ (21); HREIMS *m/z* 348.1561 (calcd. for $C_{19}H_{24}O_6$, 348.1566).

3.4.7. 1β,14,18-Trihydroxy-13-methoxy-8,11,13-podocarpatrien-7-one (*7*)

Amorphous solid; $[\alpha]_D^{23}$ −35.1 (*c* 1.93, $CHCl_3$); UV λ_{max}^{MeOH} nm (log ε): 218.5 (4.07), 271.5 (3.82), 359.0 (3.40); IR (film) ν_{max} 3427, 1632, 1578, 1485, 1457, 1252, 1052, 1028, 825, 754 cm^{-1}; for ^1H and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2; EIMS 70 eV, *m/z* (rel. int.): 320 $[M]^+$ (100), 205 $[M-115]^+$ (25), 189 $[M-131]^+$ (11), 173 $[M-147]^+$ (17), 58 $[M-262]^+$ (36); HREIMS *m/z* 320.1616 (calcd. for $C_{18}H_{24}O_5$, 320.1617).

Acknowledgements

This research was supported by the National Science Council of the Republic of China. We thank Dr. Harry Wilson of Academia Sinica for editing the final manuscript.

References

Alvarenga, M.A.D., Silva, J.D., Gottlieb, H.E., Gottlieb, O.R., 1981. Diterpenoids from *Micrandropsis scleroxylon*. Phytochemistry 20, 1159–1163.

Ara, I., Siddiqui, B.S., Faigi, S., Siddiqui, S., 1988a. Terpenoids from the stem bark of *Azadirachta indica*. Phytochemistry 27, 1801–1804.

Ara, I., Siddiqui, B.S., Faigi, S., Siddiqui, S., 1988b. Tricyclic diterpenoids from the stem bark of *Azadirachta indica*. J. Nat. Prod. 51, 1054–1061.

Ara, I., Siddiqui, B.S., Faigi, S., Siddiqui, S., 1990. Three new diterpenoids from the stem bark of *Azadirachta indica*. J. Nat. Prod. 53, 816–820.

Cambie, R.C., Mander, L.M., 1962. Chemistry of the podocarpaceae-VI: constituents of the heartwood of *Podocarpus totara* G. Benn. Tetrahedron 18, 465–475.

Cheng, Y.S., Kuo, Y.H., Lin, Y.T., 1967. Extractive components from the wood of *Taiwania cryptomerioides* Hayata: the structure of T-cadinol and T-muurolol. J. Chem. Soc. Chem. Commun., 565–566.

Kuo, Y.H., Cheng, Y.S., Lin, Y.T., 1969. Extractive components from the wood of *Taiwania cryptomerioides* Hayata: three new sequiterpene alcohols, muurolane-3-ene-9 α -ol-2-one, muurolane-2α,9 β -diol-3-ene and muurolane-2β,9 β -diol-3-ene. Tetrahedron Lett. 10, 2375–2377.

Kuo, Y.H., Shih, J.S., Lin, Y.T., Lin, Y.T., 1979. 6β-Acetoxy-7α- hydroxyroyleanone, a new compound from *Taiwania cryptomerioides* Hayata. J. Chin. Chem. Soc. 26, 71–73.

Kuo, Y.H., Lin, Y.T., Lin, Y.T., 1985. Taiwanin H, a new lignan from the bark of *Taiwania cryptomerioides* Hayata. J. Chin. Chem. Soc. 32, 381–383.

Kuo, Y.H., Chang, C.I., Lee, C.K., 2000a. Six podocarpane-type trinorditerpenes from the bark of *Taiwania cryptomerioides*. Chem. Pharm. Bull. 48, 597–599.

Kuo, Y.H., Chang, C.I., 2000b. Podocarpane-type trinorditerpenes from the bark of *Taiwania cryptomerioides*. J. Nat. Prod. 63, 650–652.

Kuo, Y.H., Chien, S.C., 2001. Quinone-type podocarpanes from the bark of *Taiwania cryptomerioides*. Chem. Pharm. Bull. 49, 1033–1035.

Kuo, Y.H., Chien, S.C., Huang, S.L., 2002a. Four new podocarpane-type trinorditerpenes from the bark of *Taiwania cryptomerioides*. Chem. Pharm. Bull. 50, 544–546.

Kuo, Y.H., Chien, S.C., Kuo, C.C., 2002b. New and antioxidative 7-oxodehydropodocarpane-type trinorditerpenes from the bark of *Taiwania cryptmerioides*. Planta Med. 68, 1020–1023.

Lin, Y.T., Cheng, Y.S., Kuo, Y.H., 1968. Extractive components from the wood of *Taiwania cryptomerioides* Hayata: a new sesquiterpene Keto-alcohol, cadinane-3-ene-9(-ol-2-one. Tetrahedron Lett. 9, 3881–3882.

Lin, W.H., Fang, J.M., Cheng, Y.S., 1995. Uncommon diterpenes with the skeleton of six-five-six fused-rings from *Taiwania cryptomerioides*. Phytochemistry 40, 871–873.

Lin, W.H., Fang, J.M., Cheng, Y.S., 1996. Diterpenes and related cycloadducts from *Taiwania cryptomerioides*. Phytochemistry 42, 1657–1663.

Lin, W.H., Fang, J.M., Cheng, Y.S., 1997. Cycloadducts of terpene quinones from *Taiwania cryptomerioides*. Phytochemistry 46, 169–173.

Lin, W.H., Fang, J.M., Cheng, Y.S., 1998. Diterpenoids and steroids from *Taiwania cryptomerioides*. Phytochemistry 48, 1391–1397.

Majumder, P.L., Maiti, D.C., Kraus, W., Bokel, M., 1987. Nimbidiol, a modified diterpenoid of the root-bark of *Azadirachta indica*. Phytochemistry 26, 3021–3023.

Siddiqui, S., Ara, I., Faigi, S., Mahmood, T., Siddiqui, B.S., 1988. Phenolic tricyclic diterpenoids from the bark of *Azadirachta indica*. Phytochemistry 27, 3903–3907.

Zoghbi, M.D.G.B., Roque, N.F., Gottlieb, H.E., 1981. Humirianthenolides, new degraded diterpenoids from *Humirianthera rupestris*. Phytochemistry 20, 1669–1673.