Available online at www.sciencedirect.com
ScienceDirect
Phytochemistry 69 (2008) 518–526
PHYTOCHEMISTRY
www.elsevier.com/locate/phytochem

Diterpenoids from the pericarp of *Platycladus orientalis*

Ya-Zhou Wang, Chun-Ping Tang [a], Chang-Qiang Ke [a], Hans-Christoph Weiss [b], Ernst-Rudolf Gesing [c], Yang Ye [a,*]

[a] *State Key Laboratory of Drug Research, Shanghai Institute of Materia Medica, Chinese Academy of Sciences, 555 Zu-Chong-Zhi Road, Zhangjiang Hi-Tech Park, Shanghai 201203, PR China*
[b] *Bayer Industry Services GmbH and Company OHG, SUA-SPA, 51368 Leverkusen, Germany*
[c] *Bayer CropScience AG, R&D – Research Insecticides – Chemistry Insecticides, Agricultural Center Monheim, D-40789 Monheim, Germany*

Received 1 November 2006; received in revised form 8 May 2007
Available online 12 September 2007

Abstract

Eight labdane-type diterpenes, 7β,13*S*-dihydroxylabda-8(17),14-dien-19-oic acid (**1**), 12*R*,15-dihydroxylabda-8(17),13*E*-dien-19-oic acid (**3c**), 12*R*,15-dihydroxylabda-8(17),13*Z*-dien-19-oic acid (**3d**), 12*R*,13*R*,14*S*-trihydroxylabda-12,15-epoxy-8(17)-en-19-oic acid (**4a**), 12*S*,13*S*,14*R*-trihydroxylabda-12,15-epoxy-8(17)-en-19-oic acid (**4b**), 15-hydroxy-12-oxolabda-8(17),13*E*-dien-19-oic acid (**5**), 14*R*,15-dihydroxylabda-8(17),12*Z*-dien-19-oic acid (**7a**) and 14*S*,15-dihydroxylabda-8(17),12*Z*-dien-19-oic acid (**7b**), along with 20 known diterpenoids, were isolated from the pericarp of *Platycladus orientalis*. Their structures were unambiguously elucidated by NMR spectroscopic and single crystal X-ray diffraction analyses, as well as via chemical correlation conversion. NMR spectroscopic data of known isomers **8c** and **8d** were reported as a supplement to existing data.

Keywords: *Platycladus orientalis*; Cupressaceae; Labdane-type; Diterpenes

1. Introduction

Platycladus orientalis (L.) Franco (syn. *Biota orientalis* Endl.), which is widespread in China, belongs to the monotypic genus *Platycladus* of the Cupressaceae family. The seeds of this plant, prescribed in the Chinese Pharmacopeia as 'Bo-zi-ren', have long been used as a traditional Chinese medicine to cure haemorrhaging, abundant expectoration, coughs, palpitations, insomnia, ephidrosis, asthma and bronchitis. Pharmacological studies have also shown that a *P. orientalis* extract has inhibitory activity in platelet-activating factor (PAF) receptor binding (Han et al., 1998), antiplasmodial activity (Asili et al., 2004) and in improving impairment of memory acquisition (Nishiyama et al., 1995). Through many previous phytochemical investigations, various components were isolated, including sesquiterpenoids and diterpenoids from heartwood (Erdtman and Pelchowicz, 1956; Chetty and dev, 1964; Tomita et al., 1968), flavonoids from leaves (Pelter et al., 1970; Khabir et al., 1985), diterpenoids from seeds (Inoue et al., 1985; Ren and Ye, 2006), leaves and branches (Asili et al., 2004) and the pericarp (Kuo and Chen, 1990,1999; Kuo et al., 2000), as well as monolignol derivatives from pollen (Ohmoto and Yamaguchi, 1988). Further investigation on the chemical components of the extract from the pericarp of this plant resulted in the isolation and structural elucidation of diterpenoid components, involving eight new labdane-type diterpenes **1**, **3c**, **3d**, **4a**, **4b**, **5**, **7a** and **7b**, as well as 20 known diterpenoid compounds. The NMR spectroscopic data of **8c** and **8d** were also listed herein as a supplement to the reported data (Fang et al., 1993).

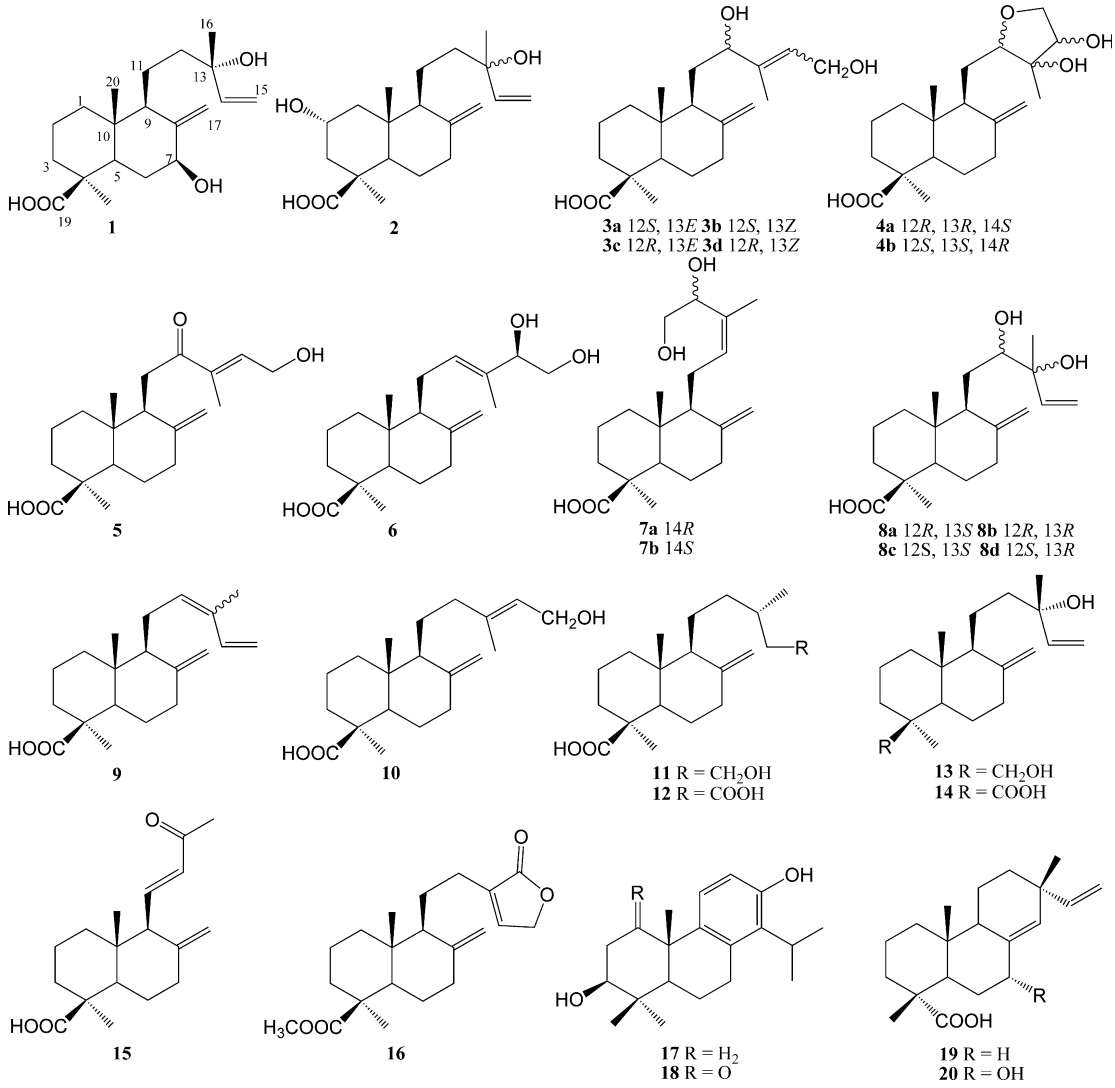
2. Results and discussion

Air dried pericarp tissue of *P. orientalis* was percolated at room temperature with petroleum ether and 95%

* Corresponding author. Tel.: +86 21 50806600x3326); fax: +86 21 50807088.
E-mail address: yye@mail.shcnc.ac.cn (Y. Ye).

0031-9422/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.phytochem.2007.07.023

ethanol, successively, to afford petroleum ether and ethanol extracts. The ethanol residue was then extracted with CH_2Cl_2 and EtOAc successively. The EtOAc extract was separated by repeated column chromatography on silica gel. The known diterpenes were isolated and identified as: labda-8(17),14-diene-2α,13-diol-19-oic acid (**2**) (Lin and Rosazza, 1998); isomers of 12,15-dihydroxylabda-8(17),13-dien-19-oic acid (**3a** and **3b**) (Fang et al., 1993; Fujimoto et al., 1990); 14(*R*),15-dihydroxylabda-8(17),12(*E*)-dien-19-oic acid (**6**) (Ren and Ye, 2006); two pairs of isomers of 12,13-dihydroxylabda-8(17),14-dien-19-oic acid (**8a** and **8b**, **8c** and **8d**) (Inoue et al., 1985; Fang et al., 1993); a mixture of *cis*- and *trans*- communic acids (**9**) (Kuo and Chen, 1999); isocupressic acid (**10**) (Chiang et al., 2003; Fang et al., 1996); imbricatolic acid (**11**) (Su et al., 1994); enantio-oliveric acid (**12**) (Calderon et al., 1987); 13-epitorulosol (**13**) (Su et al., 1994); 13-epicupressic acid (**14**) (Su et al., 1994); 14,15-bisnor-13-oxo-8(17),11*E*-labdadien-19-oic acid (**15**) (Inoue et al., 1985; Kuo and Chen, 1999); pinusolide (**16**) (Kuo and Chen, 1999; Yang et al, 1995); 3β-hydroxytotarol (**17**) (Campello et al., 1975); 1-oxo-3β-hydroxytotarol (**18**) (Kuo and Chen, 1994); sandaracopimaric acid (**19**) (Kitajima et al., 1982) and 7α-hydroxysandaracopimaric acid (**20**) (Esquivel et al., 1989) by analyses of their physical and spectroscopic properties.



The molecular formula of unknown compound **1** was assigned as $C_{20}H_{32}O_4$ by its HR-EIMS and NMR spectroscopic analyses. IR absorptions at 3423 and 1646 cm^{-1} showed the presence of a carboxylic group. The analyses of ^1H NMR spectrum (Table 1) indicated the existence of mono-substituted olefinic proton resonances at δ 5.90 (1H, *dd*, J = 17.2, 11.0), 5.18 (1H, *dd*, J = 17.2, 1.3) and 5.02 (1H, *dd*, J = 11.0, 1.3), exocyclic olefinic proton signals

Table 1
^{1}H NMR spectroscopic data of compounds **1**, **3c**, **3d**, **4a**, **4b**, **5**, **7a** and **7b** (δ values in ppm, J values in Hz)

No.	1	**3c**	**3d**	**4a**	**4b**	**5**	**7a**	**7b**
1	1.16 *m*	1.15 *m*	1.22 *m*	1.14 *m*	1.18 *m*	1.18 *m*	1.18 *m*	1.18 *m*
	1.86 *m*	1.80 *m*	1.80 *m*	1.79 *m*	1.86 *m*	1.70 *m*	1.88 *m*	1.90 *m*
2	1.48 *m*	1.48 *m*	1.52 *m*	1.52 *m*	1.54 *m*	1.44 *m*	1.50 *m*	1.50 *m*
	1.88 *m*	1.90 *m*	1.92 *m*	1.86 *m*	1.86 *m*	1.90 *m*	1.88 *m*	1.90 *m*
3	1.16 *m*	1.06 *m*	1.08 *m*	1.04 *m*	1.08 *m*	1.10 *m*	1.04 *ddd* (17.2, 13.3, 4.0)	1.06 *ddd* (17.2, 13.5, 4.4)
	2.16 *m*	2.12 *m*	2.14 *m*	2.14 *m*	2.16 *m*	2.18 *m*		
							2.14 *m*	2.14 *m*
5	1.82 *m*	1.44 *m*	1.40 *m*	1.37 *m*	1.40 *m*	1.46 *m*	1.36 br *d* (11.1)	1.36 *dd* (12.7, 3.0)
6	2.08 *m*	1.90 *m*	1.90 *m*	1.86 *m*	1.86 *m*	1.90 *m*	1.86 *m*	1.86 *m*
		1.98 *m*	2.02 *m*	1.98 *m*	2.00 *m*	2.08 *m*	1.98 *m*	1.98 *m*
7	4.28 *t*(1.6)	1.90 *m*	1.98 *m*	1.94 *m*	1.93 *m*	1.98 *m*	1.89 *m*	1.92 *m*
		2.38 *m*	2.42 *m*	2.39 *m*	2.40 *m*	2.38 *m*	2.38 *m*	2.38 *m*
9	2.12 *m*	2.12 *m*	2.05 *m*	1.92 *m*	2.00 *m*	2.52 *m*	1.70 br *d* (11.3)	1.72 br *d* (10.7)
11	1.28 *m*	1.47 *m*	1.36 *m*	1.46 *m*	1.72 *m*	2.64 *dd* (17.0, 10.0)	2.14 *m*	2.01 *m*
	1.66 *m*	1.62 *m*	1.80 *m*	1.62 *m*		3.06 *dd* (17.0, 3.2)	2.36 *m*	2.45 *m*
12	1.28 *m*	3.92 br *d* (9.9)	4.48 br *d* (10.0)	3.63 br *d* (11.1)	3.71 *t* (6.6)		5.23 *t* (5.7)	5.25 *t* (6.2)
	1.70 *m*							
14	5.90 *dd* (17.2, 11.0)	5.52 *t* (6.3)	5.40 *dd* (7.3, 6.3)	3.80 *dd* (5.4, 3.0)	3.79 *dd* (5.4, 2.3)	6.80 *dd* (5.7, 1.3)	4.60 *dd* (7.6, 5.2)	4.60 *dd* (7.8, 5.4)
15	5.02 *dd* (11.0, 1.3)	4.08 br *d* (6.3)	4.05 *dd* (12.7, 6.3)	3.59 *dd* (10.5, 3.0)	3.63 *dd* (10.5, 2.3)	4.35 br *d* (5.7)	3.46 *dd* (11.1, 5.2)	3.45 *dd* (11.2, 5.4)
	5.18 *dd* (17.2, 1.3)		4.15 *dd* (12.7, 7.3)	4.10 *dd* (10.5, 5.4)	4.12 *dd* (10.5, 5.4)		3.55 *dd* (11.1, 7.6)	3.54 *dd* (11.2, 7.8)
16	1.22 *s*	1.61 *s*	1.75 *s*	1.16 *s*	1.23 *s*	1.74 *s*	1.63 *s*	1.63 *s*
17	4.60 *s*	4.50 *s*	4.45 *s*	4.50 *s*	4.67 *s*	4.30 *s*	4.52 *s*	4.52 *s*
	5.02 *s*	4.85 *s*	4.80 *s*	4.84 *s*	4.89 *s*	4.70 *s*	4.84 *s*	4.86 *s*
18	1.18 *s*	1.20 *s*	1.20 *s*	1.22 *s*	1.25 *s*	1.22 *s*	1.18 *s*	1.18 *s*
20	0.59 *s*	0.55 *s*	0.64 *s*	0.60 *s*	0.62 *s*	0.74 *s*	0.63 *s*	0.63 *s*

3c in CD_3COCD_3 (400 MHz), **4a** and **4b** in $CDCl_3$ (600 MHz), others in CD_3OD (300 MHz).

Table 2
^{13}C NMR spectroscopic data of compounds **1**, **3c**, **3d**, **4a**, **4b**, **5**, **7a** and **7b** (δ values in ppm)

C	**1**	**3c**	**3d**	**4a**	**4b**	**5**	**7a**	**7b**
1	40.8*t*	40.7*t*	40.7*t*	39.0*t*	39.0*t*	41.1*t*	41.1*t*	41.2*t*
2	21.7*t*	21.6*t*	21.6*t*	19.8*t*	19.7*t*	21.6*t*	21.8*t*	21.7*t*
3	39.9*t*	39.9*t*	39.9*t*	37.9*t*	37.7*t*	39.8*t*	40.0*t*	39.9*t*
4	45.3*s*	45.7*s*	45.7*s*	44.1*s*	44.0*s*	45.7*s*	45.8*s*	45.8*s*
5	50.4*d*	59.7*d*	58.0*d*	56.1*d*	55.9*d*	57.8*d*	58.0*d*	58.0*d*
6	34.4*t*	28.1*t*	28.1*t*	26.0*t*	25.9*t*	27.6*t*	27.9*t*	27.9*t*
7	75.3*d*	40.4*t*	40.4*t*	38.6*t*	38.4*t*	39.7*t*	40.3*t*	40.3*t*
8	151.5*s*	150.8*s*	150.9*s*	147.9*s*	147.8*s*	151.1*s*	150.0*s*	150.2*s*
9	52.0*d*	53.7*d*	53.6*d*	52.1*d*	50.7*d*	53.2*d*	58.8*d*	58.9*d*
10	42.2*s*	41.7*s*	41.7*s*	40.1*s*	40.4*s*	41.3*s*	42.0*s*	41.9*s*
11	19.2*t*	32.0*t*	31.5*t*	23.5*t*	23.5*t*	34.7*t*	23.8*t*	23.7*t*
12	42.6*t*	76.4*d*	69.0*d*	81.0*d*	80.4*d*	203.9*s*	130.7*d*	130.8*d*
13	74.8*s*	143.0*s*	142.8*s*	77.6*s*	77.7*s*	138.4*s*	135.5*s*	135.8*s*
14	146.9*d*	125.2*d*	126.5*d*	76.6*d*	77.1*d*	142.6*d*	72.4*d*	72.0*d*
15	112.5*t*	59.7*t*	59.1*t*	72.7*t*	72.2*t*	60.6*t*	66.0*t*	65.8*t*
16	28.4*q*	14.0*q*	18.6*q*	19.4*q*	19.1*q*	12.3*q*	18.8*q*	18.8*q*
17	110.0*t*	107.5*t*	107.3*t*	107.0*t*	106.9*t*	107.2*t*	108.6*t*	108.3*t*
18	29.9*q*	28.1*q*	30.1*q*	29.0*q*	28.8*q*	30.1*q*	30.2*q*	30.1*q*
19	182.1*s*	181.8*s*	181.8*s*	182.6*s*	182.3*s*	181.8*s*	182.5*s*	182.3*s*
20	12.9 *q*	12.6*q*	14.0*q*	12.7*q*	12.1*q*	14.2*q*	14.0*q*	13.9*q*

4a and **4b** in $CDCl_3$ (150 MHz), others in CD_3OD (100 MHz).

at δ 4.60 (*s*) and 5.02 (*s*), one secondary alcohol proton resonance at δ 4.28 (1H, *t*, *J* = 1.6), and three singlet methyls at δ 0.59, 1.18 and 1.22. In its ^{13}C NMR spectrum (Table 2), 20 resonances were subclassified by DEPT experiments into three methyls, eight methylenes (two sp^2 methylenes), four methines (one oxygenated methine and one sp^2 methine), and five quaternary carbons (one oxygenated and one sp^2 carbons). All signals in the above spectra showed that compound **1** had a bicyclic-labdane diterpenoid skeleton with two allylic hydroxyl groups. Similarity comparison of the ^{13}C NMR spectrum of **1** with that of 7α,13-dihydroxy-8(17),14-labdadien-19-oic acid (Rodrigues-Filho et al., 2002) showed that both compounds shared the same skeleton. This was further confirmed by HMQC and HMBC experiments (Fig. 1). In the ROESY spectrum of **1**, signal correlations between H-5 and H-7, H-9 and H-7 suggested a α-configuration of H-7. The 13*S*-configuration was further established by oxidation of compound **14** (Su et al., 1994) into **1** by selenium dioxide in MeOH–CH_2Cl_2 at room temperature. The oxidation product of **14** was identified as **1** by NMR comparison. Thus **1** was designated as 7β,13*S*-dihydroxylabda-8(17),14-dien-19-oic acid.

The molecular formula $C_{20}H_{32}O_4$ of compounds **3c** and **3d** was assigned by HR-EIMS and NMR spectroscopic analyses. The ^{1}H NMR spectrum of **3c** (Table 1) displayed characteristic signals of a bicyclic labdane-type diterpene, particularly an allylic alcohol moiety $R_2C=CHCH_2OH$ as characterized by the olefinic proton signal at δ 5.52 (1H, *t*, *J* = 6.3), the secondary alcohol resonances appearing at δ 4.08 (2H, br *d*, *J* = 6.3), an exocyclic double bond at δ 4.50 (*s*) and 4.85 (*s*), and three singlet methyl groups at δ 0.55, 1.20 and 1.61. By comparing the ^{13}C NMR spectroscopic data of **3c** (Table 2) and **3a** (Fang et al., 1993), the skeletal structure of **3c** was indicated as 12,15-dihydroxylabda-8(17),13-dien-19-oic acid, which was further confirmed by HMQC and HMBC experiments. The signal correlations observed between H-12 and H-14, H-15 and CH_3-16 in the ROESY spectrum was indicative of a 13*E*-configuration for **3c**. The ^{1}H and ^{13}C NMR resonances of **3d** (Tables 1 and 2) were very close to those of **3b** (Fang et al., 1993), thus the skeletal structure of **3d** was deduced as 12,15-dihydroxylabda-8(17),13*Z*-dien-19-oic acid, the same as **3b**. The 13*Z*-configuration of **3d** was confirmed by the signal correlations between H-12 and H-15, H-14 and CH_3-16 in its ROESY spectrum. The chirality of C-12 of compounds **3c** and **3d** could be elucidated from the chemical shifts of the vinyl protons at C-17 (Bell et al., 1975; Hasegawa and Hirose, 1985; Fang et al., 1993). Due to the deshielding effect of the hydroxyl group at C-12, H-17 protons in a 12*S*-isomer appeared at lower field (near δ 4.70 and 4.80) than those protons (near δ 4.40 and 4.80) in a 12*R*-isomer. Thus the H-17 proton signals of **3c** (at δ 4.50 and 4.85) and **3d** (at δ 4.45 and 4.80) suggested that they both had a12*R*-configuration.

The molecular formula of compounds **4a** and **4b** was determined as $C_{20}H_{32}O_5$ by their HR-ESIMS and NMR spectroscopic analyses. The IR spectrum of **4a** showed the absorptions of a carboxylic group at 3425 and 1643 cm^{-1}. The ^{1}H NMR spectrum of **4a** (Table 1) indicated three singlet methyl groups (δ 0.60, 1.16 and 1.22) and an exocyclic methylene group (δ 4.50 and 4.84). The ^{13}C NMR spectrum of **4a** displayed 20 resonances, attributable to three methyls, eight methylenes (one oxygenated methylene and one sp^2 methylene), four methines (one oxygenated methine and one sp^2 methine), and five quaternary carbons (one oxygenated and one sp^2 carbons) through a DEPT experiment. Thus **4a** was assigned as a labdane-type diterpene with three rings by further analyses of both its degree of unsaturation and its NMR spectra. The skeletal structure of **4a** was confirmed on the basis of its HMQC and HMBC (Fig. 1) spectra. The proton signals appearing at δ 4.50 and 4.84 (H-17) suggested a 12*R*-configuration in **4a**. The relative configurations of CH_3-16 and H-14 in **4a** were determined to be both β-oriented by the correlations between H-12α and H-15α, H-15β and H-14, H-15β and CH_3-16 in its ROESY spectrum (Fig. 2). The structure of **4a** was therefore established as 12*R*,13*R*,14*S*-trihydroxylabda-12,15-epoxy-8(17)-en-19-oic acid. The ^{1}H

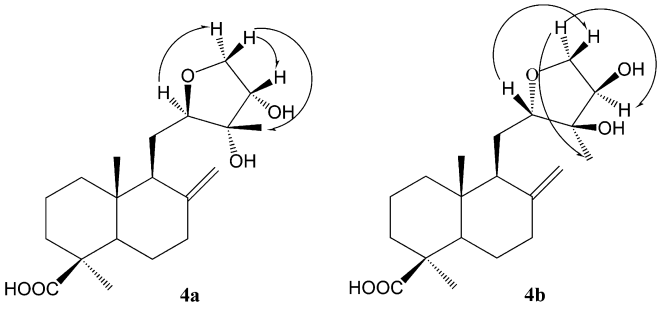


Fig. 2. Selected correlations (⌒) in the ROESY spectra of **4a** and **4b**.

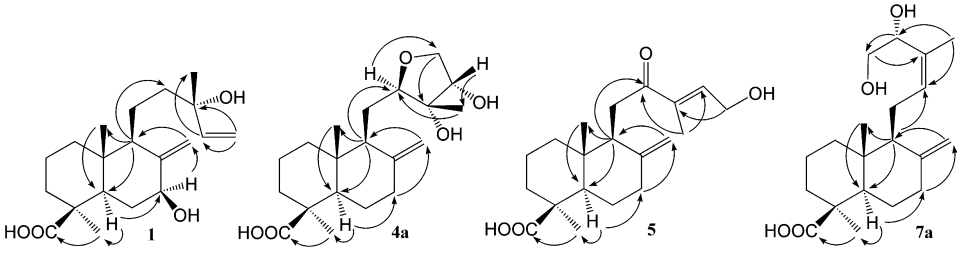


Fig. 1. Selected ^{1}H -^{13}C correlations (⌒) in the HMBC spectra of **1**, **4a**, **5** and **7a**.

and ^{13}C NMR resonances of **4b** were very close to those of **4a** (Tables 1 and 2), except for resonances of H-17 protons observed at δ 4.67 and 4.89, which suggested a 12*S*-configuration in **4b**. By analysis of the signal correlations between H-12β and H-15β, H-15α and H-14, H-15α and CH_3-16 in the ROESY spectrum of **4b** (Fig. 2), the chiralities of C-13 and C-14 were considered as 13*S* and 14*R*. Compound **4b** was therefore assigned as 12*S*,13*S*,14*R*-trihydroxylabda-12,15-epoxy-8(17)-en-19-oic acid.

Compound **5** had an exact mass at *m*/*z* 334.2146 attributable to the molecular formula $C_{20}H_{30}O_4$. The IR spectrum showed characteristic absorptions of a carboxylic group. The UV absorption at 229 nm indicated the presence of an α, β-unsaturated ketone. The ^{13}C NMR signals (Table 2) at δ 203.9 (*s*), 138.4 (*s*), 142.6 (*d*), 60.6 (*t*) and 12.3 (*q*), together with the ^1H NMR resonances at δ 6.80 (1H, *dd*, *J* = 5.7, 1.3), 4.35 (2H br *d*, *J* = 5.7) and 1.74 (3H, *s*) indicated existence of a –CO–C(CH_3)=CH–CH_2OH moiety. By analyses of the ^1H and ^{13}C NMR spectra, **5** was characterized as 15-hydroxy-12-oxolabda-8(17),13-dien-19-oic acid, which was confirmed by HMQC and HMBC experiments (Fig. 1). The signal correlations between H-15 and CH_3-16 in the ROESY spectrum suggested a 13*Z*-configuration. Thus **5** was assigned as 15-hydroxy-12-oxolabda-8(17),13*E*-dien-19-oic acid.

Compounds **7a** and **7b** were assigned the molecular formula $C_{20}H_{32}O_4$ by their HR-ESIMS and NMR analyses. **7a** showed IR absorptions at 3421 and 1693 cm^{-1} attributable to a carboxylic group. In the ^1H NMR spectrum (Table 1) of **7a,** signals appearing at δ 4.60 (1H, *dd*, *J* = 7.6, 5.2), 3.46 (1H, *dd*, *J* = 11.1, 5.2) and 3.55 (1H, *dd*, *J* = 11.1, 7.6) suggested the existence of a R_3C–CH(OH)–CH_2OH moiety. By comparing the ^1H and ^{13}C NMR spectra of **7a** with those of **6** (Ren and Ye, 2006), it was deduced to contain the same skeletal construction as 14,15-dihydroxylabda-8(17),12-dien-19-oic acid, which was further confirmed by HMQC and HMBC experiments (Fig. 1). The signal correlations between H-12 and CH_3-16, H-11 and H-14 in the ROESY spectrum of **7a** indicated a 12*Z*-configuration. The relative configuration at C-14 was determined as *R* by a single crystal X-ray diffraction experiment (Fig. 3). The experiment further confirmed the relative configuration of the whole compound. Thus **7a** was designated as 14*R*,15-dihydroxylabda-8(17),12*Z*-dien-19-oic acid. A thorough analysis of the 1D and 2D NMR spectra of **7b** suggested that it shared the same skeletal structure as **7a**. The ROESY correlations between H-12 and CH_3-16 also indicated a 12*Z*-configuration of **7b**. Moreover, the ^1H and ^{13}C NMR spectra of **7b** (Tables 1 and 2) were very similar to those of **7a**, except for H-11 proton signals of **7b** appearing at δ 2.01 and 2.45 while those of **7a** at δ 2.14 and 2.36, which could be deduced by the stereochemistry influence of the hydroxyl at C-14. The absolute configuration of C-14 of **7b** was determined by the modified Mosher method. Selective silylation of C-15 hydroxyl of **7b** gave **7B**, the (*R*)-α- and (*S*)-α-2-methoxy-2-phenyl-2-(trifluoromethyl)-acetic (MTPA) esters of **7B** (**7BR** and **7BS**) were prepared by the experiments in NMR tubes (Su et al., 2002). The observed chemical shift differences ($\Delta\delta_{S-R}$, Fig. 4) unambiguously indicated the absolute configurations of C-14 of **7B** to be *S*. Thus **7b** was assigned as 14*S*,15-dihydroxylabda-8(17),12*Z*-dien-19-oic acid.

Bo-Zi-Ren, the seeds of *P. orientalis*, has been used as an often prescribed material in traditional Chinese medicine. From a modern pharmacological point of view, the medicinal material might have blood circulation activation and memory enhancing activities. Although most of the affordable compounds have been screened in an anti-PAF cell model as well as a scopolamine-induced memory deficits animal model, tested compounds showed very weak or no activities in both tests.

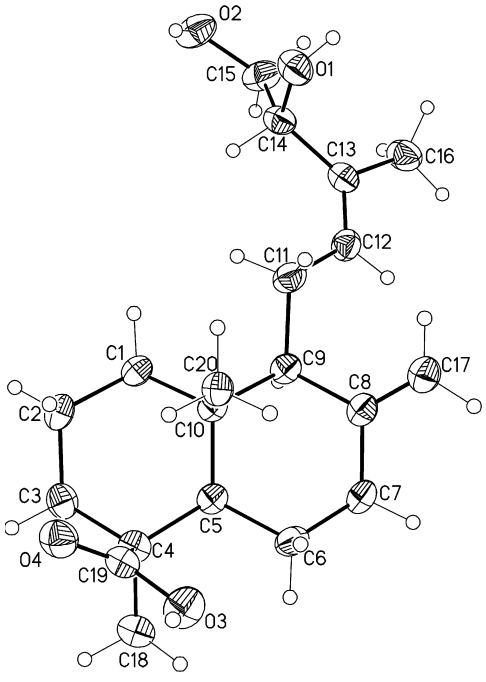


Fig. 3. Perspective ORTEP drawing for compound **7a**.

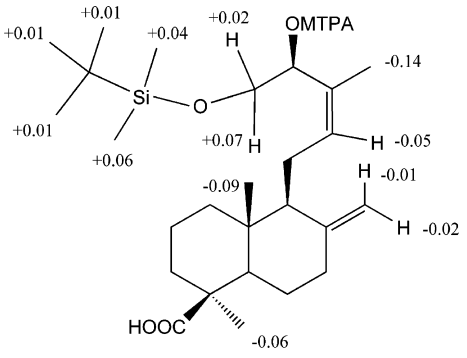


Fig. 4. ^1H Chemical-shift differences ($\Delta\delta_{S-R}$) between the (*S*)- and (*R*)-MTPA esters of 7B. $\Delta\delta_{S-R}$ values are expressed in Hz (600 Hz).

3. Experimental

3.1. General

Optical rotations were measured with either a Perkin–Elmer 241MC polarimeter or a Perkin-Elmer 341 polarimeter, whereas UV spectra were recorded with a Beckman DU-7 spectrometer. IR spectra were acquired using a Perkin–Elmer 577 Spectrometer. LR-ESIMS were measured using a Finnigan LCQ-DECA mass spectrometer, LR-EIMS were obtained with a MAT-95 spectrometer, HR-EIMS with a Kratos 1H spectrometer, and HR-ESIMS with a Q-TOF Micro LC-MS-MS spectrometer. NMR spectra were acquired on either Bruker AM-400 or Bruker AM-600 spectrometers with TMS as internal standard. Column chromatographic (CC) separations were carried out using silica gel H60 (300–400 mesh, Qingdao Haiyang Chemical Group Corporation, People's Republic of China), MCI GEL CHP20P (75–150 μm, Mitsubishi Chemical Industries) and Sephadex LH-20 (Pharmcia Biotech AB, Uppsala, Sweden) as packing materials, respectively. HSGF254 silica gel TLC plates (Yantai Chemical Industrial Institute, People's Republic of China) were used for analytical TLC.

3.2. Plant material

Pericarp of *P. orientalis* were collected from the Wenshan County in Yunnan province, China, and authenticated by Professor Jin-Gui Shen of the Shanghai Institute of Materia Medica. A voucher specimen (SIMM20031017) had been deposited in the herbarium of the Shanghai Institute of Materia Medica, Chinese Academy of Sciences.

3.3. Extraction and isolation

Powdered air-dried pericarp of *Platycladus orientalis* (10.2 kg) were percolated at room temperature with petroleum ether and EtOH–H_2O (95:5, v/v) (both 20 L × 3) successively, to afford petroleum ether extract (1.3 kg) and ethanol extracts (650 g). The EtOH residue was suspended in H_2O (1.0 L) and then extracted with CH_2Cl_2 and EtOAc (both 500 ml × 3) successively, affording CH_2Cl_2 (150 g) and EtOAc extracts (105 g). The CH_2Cl_2 extracts was subjected to silica gel CC, eluted with a gradient of petroleum ether-EtOAc (3:1 ~ 0:1) to give Fr. C1 (43.87 g), Fr. C2 (12.80 g), Fr. C3 (12.20 g), Fr. C4 (10.44 g), Fr. C5 (6.67 g), Fr. C6 (14.6 g) and Fr. C7 (8.94 g). These fractions were then, respectively subjected to repeated silica gel CC using $CHCl_3$-MeOH as eluent. Compounds **9** (1.84 g), **16** (80 mg) and **19** (425 mg) were obtained from Fr. C2, **14** (2.54 g) and **12** (56 mg) from Fr. C3, **10** (20 mg), **11** (113 mg), **13** (99 mg), **17** (88 mg), **18** (63 mg), **15** (710 mg) and **20** (16 mg) from Fr. C4, **8a** (74 mg), **8b** (56 mg), **8c** (11 mg) and **8d** (19 mg) from Fr. C5, **3d** (11 mg), **6** (12 mg), **1** (12 mg) and **5** (25 mg) from Fr. C6 and **3a** (4 mg), **3b** (3 mg), **3c** (85 mg), **7a** (3 mg), **7b** (2 mg) from Fr. C7. The EtOAc fraction was subjected to silica gel CC, eluted with a gradient of EtOAc-MeOH (20:1, 10:1, 5:1, 1:1) to give Fr. C1′ (23.1 g), Fr. C2′ (21.7 g), Fr. C3′ (31.3 g) and Fr. C4′ (12.8 g). Fr. C1′ was then separated by a MCI column, eluted with EtOH–H_2O (4:6, 5:5, 6:4, 7:3, 8:2, 1:0), to give Fr. C1′a (2.61 g), Fr. C1′b (450 mg), Fr. C1′c (1.5 g), Fr. C1′d (3.34 g), Fr. C1′e (5.15 g), Fr. C1′f (7.8 g). The Fr. C1′d was subjected to silica gel CC repeatedly, using $CHCl_3$-MeOH as eluent, to afford **3a** (36 mg), **3c** (124 mg), **3b** (137 mg), **7a** (6 mg), **7b** (5 mg), **6** (213 mg), **4a** (5 mg), **4b** (4 mg), **2** (73 mg) and **5** (45 mg).

3.4. 7β,13S-dihydroxylabda-8(17),14-dien-19-oic acid (1)

White amorphous powder, $[\alpha]_D^{20}$ +3 (*c* 0.16, MeOH); IR, ν_{max}^{KBr} cm^{-1}: 3423, 2937, 1693, 1646, 1465, 1384, 1203, 1035, 902. For ^1H and ^{13}C NMR spectroscopic data, see Tables 1 and 2. ESI-MS, *m/z*: 359.3 $[M + Na]^+$; EI-MS, *m/z* (rel. int.): 318 $[M - H_2O]^+$ (13), 300 (26), 285 (22), 272 (24), 255 (21), 239 (30), 173 (43), 149 (82), 123 (100), 109 (80). HR-EIMS, *m/z*: 318.2173 $[M - H_2O]^+$ (Calcd. for $C_{20}H_{30}O_3$, 318.2196).

3.5. 12R,15-dihydroxylabda-8(17),13E-dien-19-oic acid (3c)

White amorphous powder, $[\alpha]_D^{20}$ + 60 (*c* 0.14, MeOH); IR, ν_{max}^{KBr} cm^{-1}: 3419, 2941, 1695, 1643, 1467, 1444, 1387, 1232, 1175, 1003, 889, 758, 563. For ^1H and ^{13}C NMR spectroscopic data, see Tables 1 and 2. EI-MS, *m/z* (rel. int.): 336 $[M]^+$ (9), 318 $[M - H_2O]^+$ (14), 305 (43), 285 (22), 235 (42), 222 (30), 189 (82), 121 (100), 81 (75); HR-EIMS, *m/z*: 336.2291 $[M]^+$ (Calcd. for $C_{20}H_{32}O_4$, 336.2300).

3.6. 12R,15-dihydroxylabda-8(17),13Z-dien-19-oic acid (3d)

White amorphous powder, $[\alpha]_D^{20}$ + 49.2 (*c* 0.59, MeOH); IR, ν_{max}^{KBr} cm^{-1}: 3415, 2939, 1697, 1643, 1467, 1448, 1385, 1234, 1175, 1029, 889, 570. For ^1H and ^{13}C NMR spectroscopic data, see Tables 1 and 2. EI-MS, *m/z* (rel. int.): 318 $[M - H_2O]^+$ (21), 303 (26), 285 (23), 235 (24), 221 (66), 189 (63), 121 (100), 83 (95); HR-EIMS, *m/z*: 318.2189 $[M - H_2O]^+$ (Calcd. for $C_{20}H_{30}O_3$, 318.2196).

3.7. 12R,13R,14S-trihydroxylabda-12,15-epoxy-8(17)-en-19-oic acid (4a)

White amorphous powder, $[\alpha]_D^{20}$ + 18.6 (*c* 0.70, MeOH); IR, ν_{max}^{KBr} cm^{-1}: 3425, 2935, 1697, 1643, 1384, 1176, 1043, 756. For ^1H and ^{13}C NMR spectroscopic data, see Tables 1 and 2. ESI-MS (*m/z*): 375.2 $[M + Na]^+$; HR-ESIMS, *m/z*: 375.2130 $[M + Na]^+$ (Calcd. for $C_{20}H_{32}O_5Na$, 375.2147).

3.8. 12S,13S,14R-trihydroxylabda-12,15-epoxy-8(17)-en-19-oic acid (**4b**)

White amorphous powder, $[\alpha]_D^{20}$ + 7.5 (*c* 0.56, MeOH); IR, ν_{max}^{KBr} cm^{-1}: 3425, 2935, 1695, 1645, 1448, 1384, 1176, 1057, 756. For ^1H and ^{13}C NMR spectroscopic data, see Tables 1 and 2. ESI-MS (*m*/*z*): 375.2 $[M + Na]^+$; HR-ESIMS, *m*/*z*: 375.2146 $[M + Na]^+$ (Calcd. for $C_{20}H_{32}O_5Na$, 375.2147).

3.9. 15-hydroxy-12-oxolabda-8(17),13E-dien-19-oic acid (5)

White amorphous powder, $[\alpha]_D^{20}$ + 11 (*c* 0.24, MeOH); UV λ_{max} (MeOH) nm (log ε): 229 (3.89); IR, ν_{max}^{KBr} cm^{-1}: 3406, 2943, 1714, 1662, 1645, 1450, 1388, 1217, 1163, 1014, 891, 611. For ^1H and ^{13}C NMR spectroscopic data, see Tables 1 and 2. EI-MS, *m*/*z* (rel. int.): 334 $[M]^+$ (4), 316 $[M - H_2O]^+$ (20), 285 (12), 257 (20), 221 (38), 189 (32), 149 (100), 121(75), 99 (90); HR-EIMS, *m*/*z*: 334.2146 $[M]^+$ (Calcd. for $C_{20}H_{30}O_4$, 334.2144).

3.10. 14R,15-dihydroxylabda-8(17),12Z-dien-19-oic acid (7**a**)

White amorphous powder, $[\alpha]_{mhboxD}^{20}$ + 24 (*c* 0.10, MeOH), IR, ν_{max}^{KBr} cm^{-1}: 3421, 2935, 1693, 1647, 1450, 1385, 1261, 1176, 1072, 891, 561. For ^1H and ^{13}C NMR spectroscopic data, see Tables 1 and 2. ESI-MS (*m*/*z*): 359.2 $[M + Na]^+$; EI-MS, *m*/*z* (rel. int.): 318 $[M - H_2O]^+$ (10), 300 (78), 287(100), 239 (18), 201 (22), 175 (52), 121 (76), 93 (52); HR-EIMS, *m*/*z*: 318.2186 $[M - H_2O]^+$ (Calcd. for $C_{20}H_{30}O_3$, 318.2196).

3.11. 14S,15-dihydroxylabda-8(17),12Z-dien-19-oic acid (7**b**)

White amorphous powder, $[\alpha]_D^{20}$ + 41 (*c* 0.31, MeOH), IR, ν_{max}^{KBr} cm^{-1}: 3408, 2931, 1693, 1647, 1450, 1385, 1263, 1178, 1072, 889. For ^1H and ^{13}C NMR spectroscopic data, see Tables 1 and 2. ESI-MS (*m*/*z*): 359.2 $[M + Na]^+$; EI-MS, *m*/*z* (rel. int.): 318 $[M - H_2O]^+$ (13), 300 (65), 287(100), 175 (55), 133 (51), 121 (72), 93 (53), 81 (60); HR-EIMS, *m*/*z*: 318.2189 $[M - H_2O]^+$ (Calcd. for $C_{20}H_{30}O_3$, 318.2196).

3.12. 12S,13S-dihydroxylabda-8(17),14-dien-19-oic acid (8**c**)

White amorphous powder. ^1H NMR (300 MHz, CD_3COCD_3): 0.58 (3H, *s*, CH_3-20), 1.16 (3H, *s*, CH_3-18), 1.20 (3H, *s*, CH_3-16), 3.32 (1H, *t*, *J* = 5.5 Hz, H-12), 4.73 (1H, *s*, H-17), 4.80 (1H, *s*, H-17), 5.03 (1H, *dd*, *J* = 10.8, 1.8 Hz, H-15), 5.25 (1H, *dd*, *J* = 17.5, 1.8 Hz, H-15), 5.98 (1H, *dd*, *J* = 17.5, 10.8 Hz, H-14). ^{13}C NMR (100 MHz, $CDCl_3$): δ 12.6 (*q*, C-20), 19.8 (*q*, C-16), 24.4 (*t*, C-2), 25.7 (*t*, C-11), 26.0 (*t*, C-6), 28.9 (*q*, C-18), 37.9 (*t*, C-3), 38.7 (*t*, C-7), 39.0 (*t*, C-1), 41.1 (*s*, C-10), 44.2 (*s*, C-4), 54.4 (*d*, C-9), 56.3 (*d*, C-5), 75.7 (*s*, C-13), 79.4 (*d*, C-12), 107.1 (*t*, C-17), 114.4 (*t*, C-15), 140.7 (*d*, C-14), 151.0 (*s*, C-8), 182.9 (*s*, C-19). ESI-MS (*m*/*z*): 359.3 $[M + Na]^+$.

3.13. 12S,13R-dihydroxylabda-8(17),14-dien-19-oic acid (8**d**)

White amorphous powder. ^1H NMR (300 MHz, CD_3COCD_3): 0.57 (3H, *s*, CH_3-20), 1.15 (3H, *s*, CH_3-18), 1.22 (3H, *s*, CH_3-16), 3.33 (1H, *t*, *J* = 5.6 Hz, H-12), 4.70 (1H, *s*, H-17), 4.79 (1H, *s*, H-17), 5.10 (1H, *dd*, *J* = 10.7, 2.0 Hz, H-15), 5.27 (1H, *dd*, *J* = 17.6, 2.0 Hz, H-15), 5.98 (1H, *dd*, *J* = 17.6, 10.7 Hz, H-14).

3.14. Allylic hydroxylation of 13-epicupressic acid (14) with SeO_2 in MeOH-CH_2Cl_2

13-Epicupressic acid (**14**) (150 mg), SeO_2 (40 mg) and *N*-methylmorpholine *N*-oxide (215 mg) were dissolved in MeOH–CH_2Cl_2 (10 ml) (2:1, v/v) and stirred at room temperature for 5 h. After removal of the solvent, H_2O (20 ml) was added to the residue, and then extracted with EtOAc (30 ml × 3). The organic phase was purified by silica gel CC eluted with $CHCl_3$/MeOH (60:1) to afford compound 1 (42 mg).

3.15. X-Ray Crystallographic data for 7**a**

$C_{20}H_{32}O_4$, mol. wt. = 336.46, orthorhombic space group $P2_12_12_1$, *a* = 7.6793(1) Å, *b* = 13.1780(2) Å, *c* = 18.7288(3) Å, *V* = 1895.31(5) $Å^3$, *Z* = 4, *d* = 1.179 g/cm^3. *F*(000) = 736, μ = 0.641 mm^{-1}. A single crystal of dimensions 0.15 × 0.08 × 0.02 mm was used for X-ray measurements. The data collection was performed on a Gemini R Ultra diffractometer using Cu-Kα-radiation. Data were collected up to θ = 65.55° at 100 K. A total of 11435, thereof 3189 independent reflections were measured giving a Rint of 0.0292. Programs used: Data collection and reduction Crysalis Version 1.171.35 (Oxford Diffraction, 2006). Crystal structure solution and refinement was achieved using direct methods as implemented in SHELXTL Version 6.12 (Sheldrick, Universtitat Gottingen (Germany), 2000) and visualized using XP program. 240 Parameters were refined using 3067 reflections with $F_0 > 4\sigma$ (F_0) giving R1 = 0.0281, wR2 = 0.0735, Goodness of Fit 1.061, remaining electron density 0.15 e_ $Å^{-3}$. The absolute structure could be determined properly giving a Flack × Parameter of 0.0168(0.1623). CCDC 624276 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge via http://www.ccdc.cam.ac.uk/conts/retrieving.html (or from the CCDC, 12 Union Road; Cambridge CB2 1EZ, UK; fax: +44 1223 336 033; email: deposit@ccdc.cam.ac.uk).

3.16. Selective silylation of C-15 hydroxyl of 7b

Compound **7b** (7 mg) and chloro-*tert*-butyldimethylsilane (20 mg) were resolved in anhydrous CH_2Cl_2 solution (2 ml) under N_2 with stirring at 0 °C for 15 min. Et_3N (0.3 ml) was dropwise added to the reaction flask. The reaction solution was then stirred at room temperature for 5 h, with H_2O (10 ml) subsequently added to the reaction mixture. The aqueous layer was next extracted with CH_2Cl_2 (5 ml × 3). The combined organic layers were concentrated under reduced pressure. The residue so obtained was next purified by silica gel CC eluted with $CHCl_3$/MeOH (70:1) to afford compound **7B** (3.0 mg). A colorless oil. ^{1}H NMR (600 MHz, C_5D_5N) : 0.18 (*s*, 6H, Si-CH_3), 0.93 (*s*, H-20), 0.97 (*s*, 3H, Si–C–CH_3), 0.98 (*s*, 6H, Si–C–CH_3), 1.36 (*s*, 3H, H-18), 2.02 (*s*, 3H, H-16), 3.96 (*dd*, *J* = 5.6, 10 Hz, H-15), 4.08 (*dd*, *J* = 7.0, 10 Hz, H-15), 4.72 (*s*, H-17), 5.05 (*s*, H-17), 5.14 (*t*, *J* = 6.0 Hz, H-14), 5.56 (*t*, *J* = 5.8 Hz, H-12). ESI-MS (*m/z*): 473.2 $[M + Na]^+$, 449.3 $[M - H]^-$.

3.17. Preparation of the (S)- and (R)- MTPA ester derivatives of 7B

Compound **7B** (1.5 mg) was transferred to a clean and completely dry NMR tube. Deuterated pyridine (0.7 ml) and (*R*)-MTPA chloride (10 μl) were added to the NMR tube immediately, and then the NMR tube was shaken carefully to evenly mix the sample and MTPA chloride. The reaction mixture was then permitted to stand at room temperature and monitored every 2 h by ^{1}H NMR spectroscopic analyses. The reaction was found to be complete after 4 h. The ^{1}H NMR spectroscopic data of a (*S*)-MTPA ester derivative **7BS** of **7B** (600 MHz, C_5D_5N; obtained from the reaction NMR tube directly): 0.16 (*s*, 6H, Si–CH_3), 0.58 (*s*, 3H, H-20), 0.95 (*s*, 9H, Si–C–CH_3), 1.18 (*s*, 3H, H-18), 1.61 (*s*, 3H, H-16), 3.91 (*dd*, *J* = 3.5, 11.0 Hz, H-15), 4.12 (*dd*, *J* = 8.0, 11.0 Hz, H-15), 4.63 (*s*, H-17), 4.96 (*s*, H-17), 5.50 (*t*, *J* = 6.0 Hz, H-12), 6.29 (*dd*, *J* = 8, 3.5 Hz, H-14). In the manner described for **7BS**, another portion of compound **7B** (1.5 mg) was reacted in a second NMR tube with (*S*)-MTPA chloride (10 μl) at room temperature for 4 h using deuterated pyridine (0.7 ml) as solvent, to afford the (*R*)-MTPA ester derivative **7BR** of **7B**. ^{1}H NMR data of **7BR** (600 MHz, C_5D_5N): 0.10 (*s*, 3H, Si–CH_3), 0.12 (*s*, 3H, Si-CH_3), 0.67 (*s*, 3H, H-20), 0.94 (*s*, 9H, Si–C–CH_3), 1.24 (*s*, 3H, H-18), 1.75 (*s*, 3H, H-16), 3.89 (*dd*, *J* = 4.0, 10 Hz, H-15), 4.05 (*dd*, *J* = 8.0, 10 Hz, H-15), 4.66 (*s*, H-17), 4.97 (*s*, H-17), 5.55 (*t*, *J* = 6.0 Hz), 6.37 (*dd*, *J* = 8.0, 4.0 Hz, H-14). To further confirm the above procedure, the reaction mixtures were transferred from the NMR tubes and purified over a small sephdex LH-20 column, respectively eluted with $CHCl_3$/MeOH (1:1), to afford **7BS** and **7BR**. ESI-MS (*m/z*): 659.3 $[M + Na]^+$ and 665.3 $[M - H]^-$ (**7BS**); 659.3 $[M + Na]^+$ and 665.4 $[M - H]^-$ (**7BR**).

References

Asili, J., Lambert, M., Ziegler, H.L., Stærk, D., Sairafianpour, M., Witt, M., Asghari, G., Ibrahimi, I.S., Jaroszewski, J.W., 2004. Labdanes and isopimaranes from *Platycladus orientalis* and their effects on erythrocyte membrane and on *Plasmodium falciparum* growth in the erythrocyte host cells. J. Nat. Prod. 67, 631–637.

Bell, R.A., Gravestock, M.B., Taguchi, V.Y., 1975. Synthesis of methyl 12S- and 12R-hydroxylabd-8(17)-en-19-oates. Can. J. Chem. 53, 2869–2873.

Calderon, J.S., Quijano, L., Gomez-Garibay, F., Moran, M., Rios, T., 1987. Labdane diterpenes from *Brickellia glomerata*. Phytochemistry 26, 2639–2641.

Campello, J.D.P., Fonseca, S.F., Chang, C.J., Wenkert, E., 1975. Terpenes of *Podocarpus lambertius*. Phytochemistry 14, 243–248.

Chetty, G.L., Dev, S., 1964. Ketones from "mayur pankhi": some new cuparene-based sesquiterpenoids. Tetrahedron Lett. 5, 73–77.

Chiang, Y.M., Liu, H.K., Lo, J.M., Chien, S.C., Chan, Y.F., Lee, T.H., Su, J.K., Kuo, Y.H., 2003. Cytotoxic constituents of the leaves of *Calocedrus formosana*. J. Chin. Chem. Soc. (Taipei) 50, 161–166.

Erdtman, H., Pelchowicz, Z., 1956. Die chemie der ordnung cupressales, XII. Mitteil.: inhaltsstoffe des kernholzes von *Biota orientalis*. Endl. Chem. Ber. 89, 341–343.

Esquivel, B., Martínez, N.d.S., Cárdenas, J., Ramamoorthy, T.P., Rodríguez-Hahn, L., 1989. The pimarane-type diterpenoids of *Salvia microphylla* var *neurepia*. Planta Med. 55, 62–63.

Fang, J.M., Chen, Y.C., Wang, B.W., Cheng, Y.S., 1996. Terpenes from heartwood of *Juniperus chinensis*. Phytochemistry 41, 1361–1365.

Fang, J.M., Sou, Y.C., Chiu, Y.H., Cheng, Y.S., 1993. Diterpenes from the bark of *Juniperus chinensis*. Phytochemistry 34, 1581–1584.

Fujimoto, Y., Kakinuma, K., Eguchi, T., Ikekawa, N., Hirayama, N., Mbarushimana, A., Ntahomvukiye, D., 1990. 12,15-Dihydroxylabda-8(17),13-dien-19-oic acid from *Guizotia scabra*. Phytochemistry 29, 319–321.

Hasegawa, S., Hirose, Y., 1985. Diterpenes from the seed of *Sciadopitys verticillata*. Phytochemistry 24, 2041–2046.

Han, B.H., Yang, H.O., Kang, Y.H., Suh, D.Y., Go, H.J., Song, W.J., 1998. In vitro platelet-activating factor receptor binding inhibitory activity of pinusolide derivatives: a structure-activity study. J. Med. Chem. 41, 2626–2630.

Inoue, M., Hasegawa, S., Hirose, Y., 1985. Terpenoids from the seed of *Platycladus orientalis*. Phytochemistry 24, 1602–1604.

Khabir, M., Khatoon, F., Ansari, W.H., 1985. Phenolic constituents of *Thujia orientalis*. Current Science 54, 1180–1185.

Kitajima, J., Komori, T., Kawasaki, T., 1982. Studies on the constituents of the crude drug "Fritillariae bulbus" III. On the diterpenoid constituents of fresh bulbs of *Fritillaria thunbergii* Miq. Chem. Pharm. Bull. 30, 3912–3921.

Kuo, Y.H., Chen, W.C., 1990. 15-Hydroxypinusolidic acid, a new diterpene from the pericarp of *Platycladus orientalis* Franco. Heterocycles 31, 1705–1709.

Kuo, Y.H., Chen, W.C., 1994. Three new diterpenes, 1,3-dioxototarol, isototarolenone, and 1-oxo-3β-hydroxytotarol, from the roots of *Juniperus chinensis* Linn. Chem. Pharm. Bull. 42, 1774–1776.

Kuo, Y.H., Chen, W.C., 1999. Chemical constituents of the pericarp of *Platycladus orientalis*. J. Chin. Chem. Soc. (Taipei). 46, 819–824.

Kuo, Y.H., Chen, W.C., Lee, C.K., 2000. Four new terpenes from the pericarp of *Platycladus orientalis*. Chem. Pharm. Bull. 48, 766–768.

Lin, S.J., Rosazza, J.P.N., 1998. Microbial transformations of isocupressic acid. J. Nat. Prod. 61, 922–926.

Nishiyama, N., Chu, P.J., Saito, H., 1995. Beneficial effects of Biota, a traditional Chinese herbal medicine, on learning impairment induced by basal forebrain-lesion in mice. Biol. Pharm. Bull. 18, 1513–1517.

Ohmoto, T., Yamaguchi, K., 1988. Constituents of pollen XV. Constituents of *Biota orientalis* (L.) Endl. (1). Chem. Pharm. Bull. 36, 807–809.

Pelter, A., Warren, R., Hameed, N., Khan, N.U., Ilyas, M., Rahman, W., 1970. Biflavonyl pigments from *Thuja orientalis* (Cupressaceae). Phytochemistry 9, 1897–1898.

Ren, X.-Y., Ye, Y., 2006. Labdane diterpenes from the seeds of *Platycladus orientalis*. J. Asian Nat. Prod. Res. 8, 677–682.

Rodrigues-Filho, E., Magnani, R.F., Xie, W.P., Mirocha, C.J., Pathre, S.V., 2002. Hydroxylation of the labdane diterpene cupressic acid by *Fusarium graminearum*. J. Braz. Chem. Soc. 13, 266–269.

Su, B.N., Park, E.J., Mbwambo, Z.H., Santarsiero, B.D., Mesecar, A.D., Fong, H.H.S., Pezzuto, J.M., KingHorn, A.D., 2002. New chemical constituents of *Euphorbia quinquecostata* and absolute configuration assignment by a convenient Mosher ester procedure carried out in NMR tubes. J. Nat. Prod. 65, 1278–1282.

Su, W.C., Fang, J.M., Cheng, Y.S., 1994. Labdanes from *Cryptomeria japonica*. Phytochemistry 37, 1109–1114.

Tomita, B., Hirose, Y., Nakatsuka, T., 1968. Terpenoids. XVI. New constituents of *Biota orientalis*. Tetrahedron Lett. 9, 843–848.

Yang, H.O., Suh, D.Y., Han, B.H., 1995. Isolation and characterization of platelet-activating factorreceptor binding antagonists from *Biota orientalis*. Planta Med. 61, 37–40.