A New Triterpenoid Isolated from *Lagerstronemia speciosa* (L.) PERS.

Yoshihito OKADA, Ayako OMAE, and Toru OKUYAMA*

Department of Natural Medicine and Phytochemistry, Meiji Pharmaceutical University; 2–522–1 Noshio, Kiyose, Tokyo 204–8588, Japan. Received November 28, 2002; accepted January 6, 2003

A new triterpenoid along with four known compounds were isolated from the leaves of *Lagerstronemia speciosa* (L.) PERS. (Lythraceae). On the basis of chemical and spectroscopic evidence, the new compound was established as 3β,23-dihydroxy-1-oxo-olean-12-en-28-oic acid.

Key words 3β,23-dihydroxy-1-oxo-olean-12-en-28-oic acid; *Lagerstronemia speciosa*; Lythraceae

Lagerstronemia speciosa (L.) PERS. (Lythraceae) is a deciduous tree native to regions from India and southern China to tropical Australia. Murakami *et al.*[1] isolated two triterpenoids, colosolic acid and maslinic acid, from the leaves of this plant and colosolic acid was shown to be a glucose transport activator. Studies on tannin,[2,3] acetal,[4] ellagic acid derivatives,[5] alkaloids,[6] and sterol[2] have also been reported so far. Its leaves, locally called "banaba," are well-known as a popular home remedy for diabetes and other conditions. We began studies on the plant in order to isolate the active components and confirm their usefulness for treating diabetic complications.

This paper describes the isolation of a new triterpene along with known compounds from the leaves of the plant.

The leaves of *L. speciosa* were extracted with ethyl acetate, 50% acetone and then water, successively, to yield the corresponding extract. The ethyl acetate extract was separated and purified by using column chromatography and HPLC with silica gel and/or ODS to yield compounds **1**, **2**, **3**, **4**, and **5**.

Compound **1**, a white powder, mp 245—247 °C, demonstrated a fragment peak due to typical retro Diels–Alder cleavege at *m*/*z* 248 in its mass spectrum, suggesting the presence of a 12-en olean or ursan type triterpenoid. The high resolution-mass spectrum (HR-MS) gave an M^+ ion peak at *m*/*z* 486.3341 to confirm the molecular formula for $C_{30}H_{46}O_5$. All of the ^{13}C-NMR signals of **1** are shown in Table 1, which reveals the presence of six methyl, a carboxyl, and a carbonyl carbon. The ^{1}H-NMR (Table 2) gave six tertiary methyl signals, and a trisubstituted olefinic proton at δ 5.50 ppm due to a C-12 double bond and a hydroxy methyl proton signal. The hydroxy methyl proton was suggested to be at C-23 or 24 by the analysis of ^{1}H-, ^{13}C-NMR, and heteronuclear multiple bond connectivity (HMBC) spectra. The location of the hydroxy methyl was confirmed by the different nuclear Overhauser effect (NOE) experiments. A different NOE was observed between the methyl proton at C-25 and the methyl proton at C-23 or C-24, suggesting that a methyl group was attached to the β position at C-4. The location of the β methyl group at C-4 assigned in the ^{13}C-NMR chemical shift value appeared at δ 13.7 ppm.[7,8] Therefore, the hydroxy methyl group at C-4 was determined to be α, while the signal of C-24 was a β methyl group.

The proton signal at δ 4.55 ppm (1H, dd, J=11.9, 4.9 Hz) had a correlation with the C-24 methyl carbon in the HMBC spectrum and had correlations with two protons at C-**2** in ^{1}H–^{1}H correlation spectroscopy (COSY). It was therefore proved to be the C-3 proton and from the coupling constant and different NOE, the C-3 proton was determined to be α and the hydroxyl group was a β orientation, respectively. In the HMBC spectrum of **1**, a carbon signal at δ 213.1 ppm correlated with the C-25 methyl proton and with two proton signals at C-2. The C-2 and C-10 carbon signals shifted to a lower field for 18 ppm and 15 ppm, respectively, compared with oleanolic acid. Therefore, the carbonyl group was found to be located at the C-1 position.

On the basis of the above evidence, **1** was established to be 3β,23-dihydroxy-1-oxo-olean-12-en-28-oic acid.

Compound **2** was determined to be $C_{30}H_{46}O_4$ from the HR-EI-MS. As shown in Table 1, seven tertiary methyls and a carboxyl group at δ 180.1 ppm and a carbonyl group at δ 213.0 ppm in the ^{13}C-NMR signals were observed. Therefore, it was also deduced to be a similar skelton, except for

Table 1. ^{13}C-NMR Data for Compounds **1** and **2**

Carbon	**1**	**2**
1	213.1 s	213.0 s
2	45.1 t	45.4 t
3	72.8 d	78.4 d
4	43.8 s	40.0 s
5	47.4 d	54.7 d
6	17.9 t	18.2 t
7	32.9 t	33.2 t
8	39.7 s	39.7 s
9	39.6 d	39.7 d
10	52.4 s	52.7 s
11	25.7 t	25.7 t
12	123.2 d	123.2 d
13	144.1 s	144.1 s
14	42.2 s	42.5 s
15	28.3 t	28.3 t
16	23.7 t	23.7 t
17	46.8 s	46.8 s
18	42.4 d	42.3 d
19	46.1 t	46.2 t
20	30.9 s	31.0 s
21	34.3 t	34.3 t
22	33.1 t	33.2 t
23	66.0 t	29.0 q
24	13.7 q	16.8 q
25	15.6 q	15.0 q
26	18.1 q	18.1 q
27	26.0 q	26.0 q
28	180.0 s	180.1 s
29	33.3 q	33.3 q
30	23.8 q	23.8 q

ppm from TMS, in pyridine-d_5, 125 MHz, room temperature.

* To whom correspondence should be addressed. e-mail: okuyama@my-pharm.ac.jp

Table 2. ^{1}H-NMR Data for Compounds **1** and **2**

Proton	1	2
2α	2.79 1H dd (J=11.6, 4.9)	2.69 1H dd (J=12.1, 4.9)
2β	3.44 1H t-like (J=11.9)	3.37 1H t-like (J=11.9)
3	4.55 1H dd (J=11.9, 4.9)	3.74 1H dd (J=11.9, 4.9)
5	1.91 1H br d-like (J=10.7)	1.09 1H br d-like (J=11.6)
9	2.65 1H dd (J=11.0, 5.8)	2.56 1H dd (J=11.1, 4.5)
7	1.29 1H dd-like (J=9.5, 3.1)	
11α	2.72 1H dt-like (J=18.1, 5.5, 4.6)	2.73 1H dt-like (J=18.0, 5.2, 4.6)
11β	1.99 1H ddd (J=18.3, 11.3, 3.1)	
12	5.50 1H t-like	5.52 1H t-like (J=3.7)
15	1.12 1H dd-like (J=13.1)	
18β	3.26 1H dd (J=13.7, 4.0)	3.29 1H dd (J=13.7, 4.9)
19α	1.73 1H d-like (J=13.7)	1.78 1H d (J=13.7)
19β	1.21 1H d-like (J=4.6)	
21	1.41 1H td (J=13.6, 3.7)	
23	3.71 1H d (J=10.7) 4.12 1H d (J=10.7)	1.21 3H s
24	1.17 3H s	1.20 3H s
25	1.36 3H s	1.27 3H s
26	1.08 3H s	1.07 3H s
27	1.23 3H s	1.31 3H s
29	0.93 3H s	0.96 3H s
30	0.99 3H s	1.01 3H s

ppm from TMS, J=Hz, in pyridine-d_5, 500 Mz, room temperature. Only signals assigned clearly are shown in this Table.

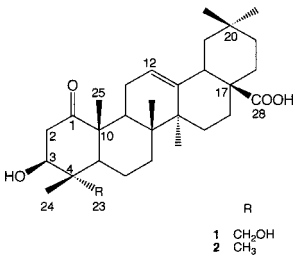

Fig. 1. Structures of **1** and **2**

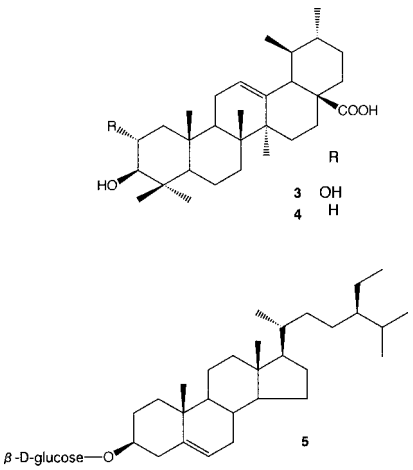

Fig. 2. Structures of **3**, **4** and **5**

one hydroxy methyl, to that of **1**. From various spectral data, **2** was established to be 3β-hydroxy-1-oxo-olean-12-en-28-oic acid, which was previously reported by Ulubelen as virgatic acid.[9]

Icli[10] showed the ^{13}C-NMR assignments of virgatic acid. However, the assignments for some signals are different from

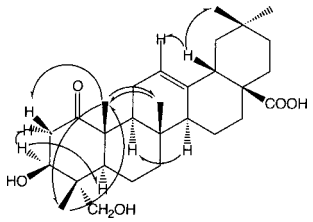

Fig. 3. NOE Correlations for **1**

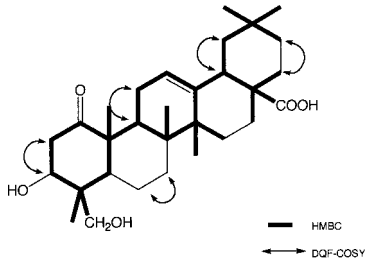

Fig. 4. HMBC and DQF-COSY of **1**

ours. Their structure elucidation or ^{13}C-NMR assignments may not have been completely adequate.

Compounds **3**, **4**, and **5** were identified with colosolic acid,[1] which has been already isolated from the title plant by Murakami *et al.*, ursolic acid, and β-sitosterol glucoside, respectively, on the basis of various spectral and chemical data.

Experimental

General Procedure Melting points were determined on a Yanagimoto micromelting point apparatus and were uncorrected. Mass spectra were obtained with a JEOL JMS DX-302 mass spectrometer. ^{1}H- and ^{13}C-NMR spectra were taken on a JEOL A-500 spectrometer using tetramethylsilane as an internal standard. Optical rotations were measured with a JASCO DIP-370.

Plant Material Leaves of *Lagerstronemia speciosa* (Lythraceae) were provided from Tonan-Shokubutsu-Rakuen in Okinawa Prefecture. The voucher specimen is deposited in the Department of Natural Medicine & Phytochemistry, Meiji Pharmaceutical University.

Extraction and Isolation The air dried leaves (600 g) of *L. speciosa* were extracted with AcOEt (2 d, 3 times, room temperature), 50% aqueous acetone (2 d, 3 times, room temperature), and water (5 h, 2 times, hot), respectively, to yield the corresponding extract (16 g, 120 g, 38 g). The AcOEt extract was separated and purified with a combination of column chromatography and HPLC to yield **1** (29 mg), **2** (28 mg), **3** (31 mg), **4** (3 mg) and **5** (7 mg).

Compound **1**: A white powder, mp 245—247, $[\alpha]_D$ +112.1° (c=0.58, MeOH), $C_{30}H_{46}O_5$, EI-MS m/z: 486 (M^+,19), 468 (12), 248 (100), 203 (92), HR-EI-MS m/z: obsd. 486.3341, calcd. for 486.3345. ^{13}C-, ^{1}H-NMR are shown in Tables 1 and 2, respectively.

Compound **2**: A white powder, mp 255—257, $[\alpha]_D$ +120.1° (c=0.37, MeOH), $C_{30}H_{46}O_4$, EI-MS m/z: 470 (M^+, 25), 452 (19), 248 (100), 203 (88), HR-EI-MS m/z: obsd. 470.3398, calcd. for 470.3396. ^{13}C-, ^{1}H-NMR as shown in Tables 1 and 2, respectively.

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