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Terpenoids and steroids isolated from Calophyllum polyanthum

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ABSTRACT

Three triterpenoids, one diterpenoid and two steroids were isolated from the aerial parts of Calophyllum polyathum. The structures of these compounds were determined by 1D NMR and mass spectroscopic data. Among them, compounds 1, 3, and 4 was obtained for the first time from Calophyllum polyathum.

Keywords: Calophyllum polyathum; Guttiferae; Terpenoid; Steroid; Structure elucidation

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Introduction

The genus Calophyllum (Guttiferae) comprises about 200 species, mainly distributed in tropical areas [1]. Many plants of the Calophyllum are used as traditional herbal medicines to treat peptic ulcer, malaria, high blood pressure, inflammations, trauma infection, et al. [2-4]. Previous phytochemical studies have revealed that the genus Calophyllum is rich in active natural products, such as coumarins, xanthones, flavonoids, terpenoids [5-7]. There are fours species in China, and *Calophyllum polyanthum* is one of them. In our search for bioactive contituents from this plant, an investigation of the chemical constituents of *Calophyllum polyanthum* Wall. ex Choisy, which was collected in Yunnan Province Longchuan County, led to the isolation of six compounds, cassipourol (**1**) [8], friedelin (**2**) [9], glut-5-en-3*β*-ol (**3**) [10], putranjivic acid (**4**) [11], a 1:1 mixture of *β*-sitosterol (**5**) and stigmasterol (**6**) [12]. In this manuscript, the isolation, 1D NMR and mass spectroscopic data of these compounds are presented.

Results and discussion

1D NMR and mass spectroscopic data

Cassipourol (**1**). colorless liquid. FAB-MS(m/z): 295 ([M+H]$^+$, $C_{20}H_{38}O$). ^{1}H-NMR (Chloroform-d, 600 MHz) δ_H 5.40 (1H, tq, J = 6.9, 1.4 Hz, H-14), 4.15 (2H, d, J = 7.0 Hz, H-15), 1.66 (3H, s, H-20), 0.87 (3H, s, H-16), 0.86 (3H, s, H-17). ^{13}C-NMR (Chloroform-d, 151 MHz) δ_C 140.5 (C-13), 123.3 (C-14), 59.6 (C-15), 40.1 (C-12), 39.6 (C-2), 37.6 (C-10), 37.6 (C-8), 37.5 (C-4), 36.9 (C-1), 33.0 (C-6), 32.9 (C-9), 28.2 (C-5), 25.4 (C-11), 25.0 (C-3), 24.7 (C-7), 22.9 (C-16), 22.8 (C-17), 20.0 (C-18), 19.9 (C-19), 16.4 (C-20).

Friedelin (2). colorless needle crystal. ESI-MS(m/z): 465 ([M+K]$^+$, $C_{30}H_{50}O$); ^{1}H-NMR (Chloroform-d, 600 MHz) δ_H 1.17 (3H, s, H-28), 1.04 (3H, s, H-27), 1.00 (3H, s, H-26), 0.99 (3H, s, H-30), 0.95 (3H, s, H-29), 0.88 (3H, d, J = 6.7 Hz, H-23), 0.86 (3H, s, H-25), 0.72 (3H, s, H-24); ^{13}C-NMR (Chloroform-d, 151 MHz) δ_C 213.5 (C-3), 59.7 (C-10), 58.4 (C-4), 53.3 (C-8), 43.0 (C-18), 42.4 (C-5), 41.7 (C-2), 41.5 (C-6), 39.9 (C-13), 39.5 (C-22), 38.5 (C-10), 37.6 (C-9), 36.2 (C-16), 35.8 (C-11), 35.5 (C-19), 35.2 (C-29), 33.0 (C-21), 32.6 (C-28), 32.3 (C-15), 32.0 (C-30), 30.7 (C-12), 30.2 (C-17), 28.4 (C-20), 22.5 (C-1), 20.5 (C-26), 18.9 (C-27), 18.4 (C-7), 18.2 (C-24), 14.9 (C-25), 7.1 (C-23).

Glut-5-en-3*β*-ol (3). colorless needle. EI-MS (m/z): 426 ([M]$^+$, $C_{30}H_{50}O$); ^{1}H-NMR (Chloroform-d, 600 MHz) δ_H 5.63 (1H, d, J = 5.9 Hz, H-6), 3.47 (1H, s, H-3), 1.16 (3H, s, H-27), 1.14 (3H, s, H-26), 1.09 (3H, s, H-25), 1.04 (3H, s, H-29), 1.00 (3H, s, H-30), 0.99 (3H, s, H-23), 0.95 (3H, s, H-28), 0.85 (3H, s, H-24). ^{13}C-NMR (Chloroform-d, 151 MHz) δ_C 141.8 (C-5), 122.3 (C-6), 76.5 (C-3), 49.9 (C-10), 47.6 (C-18), 43.3 (C-8), 41.0 (C-14), 39.5 (C-4), 39.2 (C-22), 38.0 (C-13), 36.2 (C-16), 35.3 (C-19), 35.0 (C-9), 34.8 (C-11), 34.7 (C-29), 33.3 (C-21), 32.6 (C-28), 32.3 (C-15), 32.3 (C-30), 30.6 (C-12), 30.3 (C-17), 29.2 (C-23), 28.5 (C-20), 28.0 (C-7), 25.7 (C-24), 23.8 (C-1), 19.8 (C-27), 18.6 (C-26), 18.4 (C-2), 16.4 (C-25).

Putranjivic acid (4). colourless needles. ESI MS(m/z): 441([M-H]$^-$, $C_{30}H_{50}O_2$). ^{1}H-NMR (Chloroform-d, 600 MHz) δ_H 5.61 (1H, dd, J = 17.4, 10.7 Hz, H-4), 4.91 (2H, m, H-23), 2.33 (2H, m, H_2-2), 1.17 (3H, s, H_3-5), 1.02 (3H, s, CH_3), 0.99 (6H, s, 2×CH_3), 0.94 (3H, s, CH_3), 0.88 (3H, s, CH_3). ^{13}C-NMR (Chloroform-d, 151 MHz) δ_C 179.2 (C-3), 151.1 (C-4), 111.2 (C-23), 58.4 (C-10), 53.2 (C-8), 43.0 (C-18), 42.3 (C-5), 41.7 (C-2), 39.8 (C-13), 39.5 (C-22), 38.8 (C-14), 38.5 (C-9), 37.2 (C-6), 36.2 (C-16), 35.5 (C-19), 35.4 (C-11), 35.2 (C-30), 33.0 (C-21), 32.5 (C-15), 32.3 (C-29), 32.0 (C-28), 30.4 (C-12),

30.2 (C-17), 28.4 (C-20), 21.4 (C-1), 20.4 (C-27), 19.0 (C-26), 18.3 (C-25), 18.2 (C-24), 18.2 (C-7) .

***β*-Sitosterol (5) and Stigmasterol (6)**. colourless needles. EI-MS(m/z): 400([M]$^+$, $C_{28}H_{48}O$), 412([M] $^+$, $C_{29}H_{48}O$). ^{1}H-NMR (Chloroform-d, 600 MHz) δ_H 5.35 (2H, d, J = 5.2 Hz, H-6), 5.15 (1H, dd, J = 15.2, 8.7 Hz, H-22), 5.01 (1H, dd, J = 15.2, 8.6 Hz, H-23), 3.52 (2H, m, H-3), 1.02 (3H, d, J = 7.0 Hz, 21-CH_3), 1.01 (3H, s, 19-CH_3), 0.85 (3H, d, J = 6.24 Hz, 26-CH_3), 0.82 (3H, d, J = 7.6 Hz, 29-CH_3), 0.80 (3H, d, J = 5.82 Hz, 27-CH_3), 0.68 (3H, s, 18-CH_3). β-Sitosterol (5): ^{13}C-NMR (Chloroform-d, 151 MHz) δ_C 141.0 (C-5), 121.9 (C-6), 72.0 (C-3), 57.0 (C-14), 56.2 (C-17), 50.4 (C-9), 46.0 (C-24), 42.5 (C-13), 42.4 (C-4), 39.9 (C-12), 37.5 (C-1), 36.7 (C-10), 36.4 (C-20), 34.1 (C-22), 32.1 (C-8), 32.1 (C-7), 31.9 (C-2), 29.3 (C-25), 28.5 (C-16), 26.3 (C-23), 24.5 (C-15), 23.3 (C-28), 21.4 (C-11), 20.0 (C-26), 19.6 (C-19), 19.2 (C-27), 19.0 (C-21), 12.1 (C-18). Stigmasterol (6): ^{13}C-NMR (Chloroform-d, 151 MHz) δ_C 141.0 (C-5), 138.5 (C-22), 129.5 (C-23), 121.9 (C-6), 72.0 (C-3), 57.1 (C-14), 56.3 (C-17), 51.5 (C-24), 50.4 (C-9), 42.5 (C-13), 42.4 (C-4), 40.7 (C-20), 40.0 (C-12), 37.5 (C-1), 36.7 (C-10), 32.1 (C-8), 32.1 (C-7), 32.1 (C-25), 31.9 (C-2), 29.1 (C-16), 25.6 (C-28), 24.6 (C-15), 21.4 (C-11), 21.3 (C-26), 21.3 (C-21), 19.6 (C-19), 19.2 (C-27), 12.3 (C-29), 12.2 (C-18).

Experimental Section

General

NMR spectra were recorded on a Bruker Avance III-600 spectrometer in $CCID_3$ with TMS as internal standard. Mass spectra were taken on a VG Auto spec-3000 spectrometer or on a Finnigan MAT 90 instrument. Materials for column chromatography were silica gel (200–300 mesh; Qingdao Marine Chemical Inc.), Sephadex LH-20 (40–70 μm; Merk Co., Ltd.), and RP-18 (40–60 μm; YMC, Milford, MA). Spots were visualized under UV light (254 nm) or by spraying with 10% H_2SO_4 in 95% EtOH (v/v) followed by heating.

Plant material

The aerial parts of *C. polyathum polyanthum* Wall. ex Choisy were collected in Longchuan County, Dehong Prefecture, Yunnan Province, P. R. China, in April 2019. Identification of the plant was performed by Prof. Tang Li-ping of Kunming Medical University. A voucher specimen is deposited in Kunming Medical University.

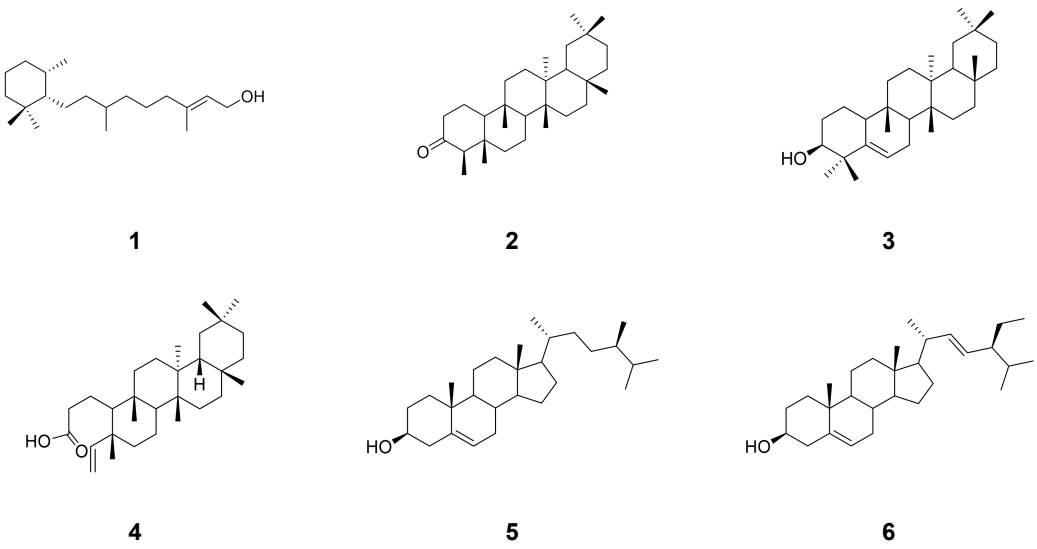
Extraction and isolation

Powder (20.0 kg) of the dried aerial parts was extracted with 95% EtOH (60 L×3) at room temperature. Filtration and condensation of the solution to dryness under vacuum yielded an extract (1.40 kg). The extract was fractionated over MCI gel (CHP20p) column chromatograph with MeOH-H_2O (0%, 10%, 30%, 50%, 70%, 85%, 95%) to obtain five fractions (A–F). Decolorized fraction F (100.8 g) and E (85.0 g) were combined and subjected to silica gel column chromatograph (PE/EA, 400:1 to 0:100 (v/v)) to give 26 fractions (EF_1−EF_{26}).

Fraction EF_8 (438.0 mg) was chromatographed on silica gel column chromatograph (200−300 mesh) eluting with PE/EA (100:1 to 1:1 (v/v)) and then purified by Sephadex LH-20 column chromatograph with CH_2Cl_2-MeOH (3:1 (v/v)) to give compound **2** (14.0 mg). Fraction EF_9 (10.0 g) was subjected to silica gel column chromatograph eluted with PE/EA (100:1 to 1:1 (v/v)) to get compound **3** (8.0 mg). Fraction EF_{11} (2.3 g) was chromatography over silica gel column chromatograph eluted with PE/EA (100:1 to 1:1 (v/v)) to obtain 13 fractions (EF_{11-1}~EF_{11-13}). Fraction EF_{11-9} (200.0 mg) was separated on Sephadex LH-20 column chromatograph with

CH_2Cl_2-MeOH (3:1 (v/v)) and then purified by RP silica gel (RP-18) with MeOH-H_2O (70:30 to 0:100 (v/v)) to give compound **1** (14.0 mg).

Fraction EF_{14} (5.0 g) was subjected to silica gel CC eluted with PE/EA (100:1 to 1:1 (v/v)) to give 33 fractions (Fr. $EF_{14\text{-}1}$~ $EF_{14\text{-}33}$). Fraction $EF_{14\text{-}12}$ (258.0 mg) was separated by Sephadex LH-20 column chromatograph with CH_2Cl_2-MeOH (3:1 (v/v)) to acquire 12 fractions (Fr. $EF_{14\text{-}12\text{-}1}$~Fr. $EF_{14\text{-}12\text{-}12}$). Fraction $EF_{14\text{-}12\text{-}4}$ (32.7 mg) was separated by preparative TLC with petroleum ether-dichloromethane (3:1 (v/v)) to yield compound **5** and **6** (10.4 mg). Fraction $EF_{14\text{-}9}$ (416.0 mg) was isolated over Sephadex LH-20 column chromatograph with CH_2Cl_2-MeOH (3:1 (v/v)) to give 15 fractions (Fr. $EF_{14\text{-}9\text{-}1}$~Fr. $EF_{14\text{-}9\text{-}15}$). Fraction $EF_{14\text{-}9\text{-}12}$ (170.0 mg) was separated by RP-18 silica gel column chromatograph with MeOH-H_2O (70:30 to 0:100 (v/v)) and then purified by preparative TLC with petroleum ether-dichloromethane (3:1 (v/v)) to yield compound **4** (5.0 mg).



1 **2** **3**

4 **5** **6**

Figure 1. Structures of compounds **1**–**6**

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References

[1] Zhong, H., Ruan, J. L., Yao, Q. Q. (2010). Two new 4-arylcoumarins from the seeds of *Calophyllum polyanthum*. J Asian Nat Prod Res, 12(7): 562-568.

[2] Ito, C., Matsui, T., Kobayashi, T., Tokuda, H., Shanmugam, S., Itoigawa, M. (2018). Cancer chemopreventive activity of xanthones from *Calophyllum elatum*. Nat Prod Commun. 13(4): 447-449.

[3] Xiao, Q., Zeng, Y. B., Mei, W. L., Zhao, Y. X., Deng, Y. Y., Dai, H. F. (2008). Cytotoxic prenylated xanthones from *Calophyllum inophyllum*. J Asian Nat Prod Res 10(10): 993-997.

[4] Cuesta-Rubio, O., Oubada, A., Bello, A., Maes, L., Cos, P., Monzote, L. (2015). Antimicrobial assessment of resins from *Calophyllum antillanum* and *Calophyllum inophyllum*. Phytother Res, 29 (12): 1991-1994.

[5] Ee, G. C. L., Jong, V. Y. M., Sukari, M. A., Rahmani, M., & Kua, A. S. M. (2009). Xanthones from *Calophyllum inophyllum*. Pertanika J Sci Technol, 17 (2): 307-312.

[6] Daud, S. B., Cheng, G., Malek, E. A., Ahmad, Z., Hashim, N. M., See, I. (2016). A new pyrano-

xanthone from the stem bark of *Calophyllum buxifolium*. Chem Nat Compd, 52(5): 807-809.

[7] Bernabé-Antonio, A., Álvarez-Berber, L. P., Cruz-Sosa, F. (2014). Biological importance of phytochemicals from *Calophyllum brasiliense* Cambess. Annu Res. Rev Biol, 4 (10): 1502-1517.

[8] Chaturvedula, V. S., Norris, A., Miller, J. S., Ratovoson, F., Andriantsiferana, R., Rasamison, V. E., Kingston, D. G. (2006). Cytotoxic diterpenes from *Cassipourea madagascarien-sis* from the Madagascar rainforest. J Nat Prod, 69(2): 287–289.

[9] Li, X. H., Yang, W. Y., Wang, J., (2019). Study on chemical constituents of Dai traditional medicine "Ya-Haiba" (*Phyllanthus niruri* L.). Asia-Pac Trad Med, 15(02): 59-63.

[10] El-Seedi H. R. (2005). Antimicrobial triterpenes from *Poulsenia armata* Miq. Standl. Nat Prod Res, 19(2): 197–202.

[11] Leong, Y. W., Harrison, L. J. (1999). (20*R*, 23*E*)-Eupha-8,23-diene-3*β*,25-diol from *Tripe-talum cymosum*. Phytochemistry, 50 (5): 849-857.

[12] Chundattu, S. J., Agrawal, V. K., Ganesh, N. (2016). Phytochemical investigation of *Calotro-pis procera*. Arab J Chem, 9: S230-S234.