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茎花葱臭木化学成分的研究

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摘 要: 从茎花葱臭木种子中分离得到 5 个化合物,经理化与波谱分析鉴定为 β-谷甾醇(1)、没食子酸乙酯(2)、胡萝卜苷(3)、1-*O*-β-D-吡喃葡萄糖基-(2*S*,3*S*,4*R*,8*Z*)-2-*N*-(2′-羟基二十四烷酰氨基)十八二氢鞘氨-8-烯(4)和 2,3,2″,3″-四氢穗花杉双黄酮(5)。这 5 个化合物均首次从该植物中分离得到。其中化合物 5 进行细胞毒活性测试,没有显示抑制活性。

关键词: 茎花葱臭木; 楝科; 2,3,2″,3″-四氢穗花杉双黄酮; 细胞毒活性

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Chemical Constituents from *Dysoxylum cauliflorum* (Meliaceae)

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Abstract: Five compounds were isolated from the seeds of *Dysoxylum cauliflorum* and their structures were elucidated as β-sitosterol (**1**), ethyl gallate (**2**), daucosterol (3), 1-*O*-β-D-glucopyranosyl-(2*S*,3*S*,4*R*,8*Z*)-2-*N*-(2′-hydroxytetracosanoyl) octadecasphinga-8-ene (**4**), and 2,3,2″,3″-tetrahydroamentoflavone (**5**). All of these compounds were isolated from this plant for the first time. Compound **5** was evaluated for cytotoxicity against five human cancer cell lines but showed no inhibitory activities.

Key words: *Dysoxylum cauliflorum*; Meliaceae; 2,3,2″,3″-tetrahydroamentoflavone; cytotoxicity

Introduction

Dysoxylum cauliflorum Hiern (Meliaceae) is a plant from the genus *Dysoxylum* originated in Fiji, Papua New Guinea, and New Zealand. Many species of the genus have been used as medicines to relieve fever, rigid limbs, convulsions, haemorrhage, and facial distortion in children [1-3]. Various bioactive compounds have been obtained by previous studies on the chemical constituents of the *Dysoxylum* genus, such as cytotoxic tirucallane-type alkaloids [4], antifeeding limonoids [5], cytotoxic diterpenes [6], antileukemic triterpene glucosides [7], and antibacterial triterpenoids [8,9]. The phytochemical investigation of *D. cauliflorum* has only afforded several dammarane triterpenoids and one sesquiterpene [10,11]. Thus, this plant was selected cultured in China for further investigation looking forward to get more bioactive compounds. The present study led to the discovery of five known compounds isolated from this plant for the first time.

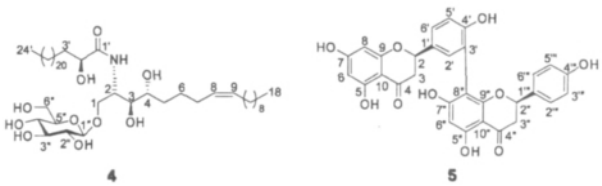

Fig. 1 The chemical structures of compounds 4 and 5

Experimental

General

^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker AVANCE III-600, Bruker DRX-500 and Bruker

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AM-400 spectrometer using TMS as internal standard. EIMS spectra were measured on a AutoSpec Premier P776 spectrometer. Column chromatography was performed on silica gel (200-300 mesh, Qingdao Meigao Chemical Co. Ltd., Qingdao, China). All solvents were distilled prior to use.

Plant material

The seeds of *D. cauliflorum* were collected at Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences, Yunnan Province, in November 2008, and identified by Xiao Chunfen of the Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences. A voucher specimen (No. 02059) was deposited in the Herbarium of the Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences.

Extraction and isolation

The air-dried powder of the seeds (8.1 kg) were extracted with 95% EtOH three times (15 L × 3)) at room temperature. The crude extracts (1.6 kg) were suspended in water, and then partitioned with petroleum ether and EtOAc, respectively. The petroleum ether extract (130 g) was subjected to silica gel column chromatography eluted with petroleum ether /Me_2CO (from 98 ∶ 2 to 0 ∶ 100) to give eight fractions (1-8) and afford compounds **1** (42.4 mg), **3** (1.23 g), and **4** (5 mg) purified by crystallization and recrystallization with Me_2CO and CH_3OH, respectively. Fraction **6** (3.3 g) was subjected to silica gel column eluted with $CHCl_3$/Me_2CO (from 100 ∶ 0 to 85 ∶ 15) to afford **2** (1.21 g). Fraction **7** (8.8 g) was further purified on silica gel column ($CHCl_3$/Me_2CO from 100 ∶ 0 to 85 ∶ 15) to yield **5** (1.20 g).

Identification

***β*-sitosterol (1)** C_{29} HO, white needle crystal (Me_2CO). It was confirmed by comparing it with the standard sample.

Ethyl gallate (2) $C_9H_{10}O_5$, white powder (Me_2CO), ^{1}H NMR (pyridine-d_5, 500 MHz) *δ*: 7.57 (2H, s, H-2, H-6), 4.26 (2H, q, *J* = 7.0 Hz, H-8), 1.16 (3H, t, *J* = 7.2 Hz, H-9); ^{13}C NMR (pyridine-d_5, 100 MHz) *δ*: 167.1 (s, C-7), 147.7 (s, C-3, C-5), 141.0 (s, C-4), 121.4 (s, C-1), 110.2 (d, C-2, C-6), 60.5 (t, C-8), 14.4 (q, C-9). The NMR data were identical to those of literature [12].

Daucosterol (3) $C_{35}H_{60}O_6$, white powder (Me_2CO), ^{1}H NMR (pyridine-d_5, 500 MHz) *δ*: 5.34 (1H, br. d, *J* = 4.5 Hz, H-6), 3.99 (1H, m, H-3), 2.47 (1H, m, H-17), 0.65 (3H, s, H-18), 0.92 (3H, s, H-19), 0.88 (3H, d, *J* = 6.5 Hz, H-21), 0.98, 0.86 (each 3H, d, *J* = 6.5 Hz, H-26, H-27), 0.90 (3H, t, *J* = 7.5 Hz, H-29), 5.06 (1H, d, *J* = 8.0 Hz, Glu-H-1), 4.07 (1H, t, *J* = 8.0 Hz, Glu-H-2), 4.30 (2H, m, Glu-H-3, Glu-H-4), 3.98 (1H, m, Glu-H-5), 4.57 (1H, d, *J* = 11.5 Hz, Glu-H-6a), 4.43 (1H, br. d, *J* = 13.0 Hz, Glu-H-6b). The 1H NMR date were accorded with the literature [13].

1-*O*-*β*-D-glucopyranosyl-(2*S*, 3*S*, 4*R*, 8*Z*)-2-*N*-(2′-hydroxytetracosanoyl) octadecasphinga-8-ene (4) $C_{48}H_{93}NO_{10}$, white powder (Me_2CO), ^{1}H NMR (pyridine-d_5, 500 MHz) *δ*: 8.56 (1H, d, *J* = 9.0 Hz, NH), 5.58-5.39 (2H, m, H-8, H-9), 5.28 (1H, m, H-2), 4.95 (1H, br. s, H-1″), 4.70 (1H, m, H-1b), 4.57 (1H, br. s, H-2′), 4.50 (1H, m, H-1a), 4.47 (1H, m, H-6″b), 4.34 (1H, m, H-6″a), 4.28 (1H, m, H-3), 4.19 (3H, m, H-4, H-3″, H-4″), 4.01 (1H, t, *J* = 7.5 Hz, H-2″), 3.86 (1H, br. s, H-5″), 2.23 (2H, m, H-7), 2.06 (2H, m, H-10), 1.95 (2H, m, H-5), 1.23-1.29 (54H, br. d, *J* = 32.0 Hz, 27 × CH_2), 0.85 (6H, t, *J* = 6.5 Hz, H-18, H-24′); ^{13}C NMR (pyridine-d_5, 150 MHz) *δ*: 176.1 (s, C-1′), 130.9 (d, C-9), 130.7 (d, C-8), 106.1 (d, C-1″), 79.1 (d, C-3″), 78.9 (d, C-5″), 76.3 (d, C-3), 75.7 (d, C-2″), 72.9 (d, C-2′), 72.8 (d, C-4), 71.8 (d, C-4″), 71.0 (t, C-1), 62.9 (t, C-6″), 52.1 (d, C-2), 36.0 (t, C-3′), 34.4 (t, C-5), 32.6 (t, C-10), 32.1 (t, C-7), 30.6-30.1 (t, n × CH_2), 27.3 (t, C-4′), 23.4 (t, C-17, C-23′), 14.7 (q, C-18, C-24′). The NMR date were consistent with the literature [14].

2, 3, 2″, 3″-tetrahydroamentoflavone (5) $C_{30}H_{22}O_{10}$, yellow powder ($CHCl_3$), EI-MS *m/z*: 542 $[M]^+$. ^{1}H NMR (DMSO-d_6, 400 MHz) *δ*: 7.20-7.18 (4H, m, H-2′, H-6′, H-2″′, H-6″′), 6.82 (1H, d, *J* = 8.0 Hz, H-5′), 6.68 (2H, d, *J* = 8.4 Hz, H-3″′, H-5″′), 6.04 (1H, s, H-6″), 5.87 (2H, s, H-6, H-8), 5.39 (2H, d, *J* = 12.8 Hz, H-2, H-2″), 2.57-3.23

(4H, m, H-3, H-3″); ^{13}C NMR (DMSO-d_6, 100 MHz) δ: 197.2 (s, C-4″), 196.7 (s, C-4), 166.9 (s, C-7), 164.8 (s, C-7″), 163.7 (s, C-5), 163.4 (s, C-5″), 162.5 (s, C-9), 160.3 (s, C-9″), 157.6 (s, C-4‴), 156.3 (s, C-4′), 131.6 (s, C-1′), 129.2 (s, C-1‴), 128.4 (d, C-6′), 128.3 (d, C-2‴, C-6‴), 127.4 (d, C-2′), 120.3 (s, C-3′), 115.3 (d, C-5′, C-3‴, C-5‴), 106.2 (s, C-8″), 102.0 (s, C-10, C-10″), 96.1 (d, C-6, C-6″), 95.2 (d, C-8), 78.9 (d, C-2), 78.1 (d, C-2″), 42.5 (t, C-3), 41.6 (t, C-3″). The NMR data were equal to those of literature [15].

Compound **5** was evaluated for its cytotoxicity against five human cancer cell lines using the MTT method [16], which included human myeloid leukemia HL-60, hepatocellular carcinoma SMMC-7721, lung cancer A-549 cells, breast cancer MCF-7, and colon cancer SW480. Cisplatin (Sigma, USA) was used as the positive control and the IC_{50} value was calculated by Reed and Muench's method [17]. The result showed compound **5** was inactive with its IC_{50} values >40 μM.

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