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# STUDIES ON THE CHEMICAL CONSTITUENTS OF *Daphne pedunculata*

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The roots and barks of genus *Daphne* plants are used to treat rheumatism, sore throat, toothache, and neuralgic pain as a folk medicine in China. This genus consists of 95 species distributed mainly in China and Europe. It is reported that coumarins of genus *Daphne* are the most important constituents due to their antiinflammation and antitumor activities [1]. The species of *Daphne pedunculata* H. F. Zhou ex C. Y. Chang is an aiophyllus arbuscular plant, and no phytochemical investigation has been reported on this plant yet. Herein, we present the results of a study of the chemical composition of this plant.

The leaves and stems of *D. pedunculata* were collected in Anning, Yunnan Province, China in May, 2006 and was authenticated by Prof. Hanchen Zheng, Department of Pharmacognosy, Second Military Medical University. Air-dried aerial parts of *D. pedunculata* (11.5 kg) were extracted with 70% ethanol (50 L×3) at room temperature. The ethanol extract was dissolved in 2 L of $H_2O$ to form a suspension and extracted successively with petroleum ether, $CHCl_3$, EtOAc, and *n*-BuOH. The petroleum ether-soluble fraction (70 g) and $CHCl_3$-soluble fraction (100 g) were subjected to a series of chromatographic operations, using a silica gel column (mesh 200-300), Sephadex LH-20, and PTLC, yielding compounds **1** (26 mg), **2** (715 mg), **3** (32 mg), **4** (24 mg), **5** (17 mg), **6** (14 mg), **7** (1.2 g), and **8** (120 mg).

Eight compounds were isolated from *D. pedunculata* for the first time.

**Daphnetin (1):** $C_9H_6O_4$, white amorphous powder, mp 262–264°C; EI-MS *m/z*: 178 $[M]^+$; $^1$H NMR (400 MHz, DMSO-$d_6$, δ, ppm, J/Hz): 6.17 (1H, d, J = 9.4, H-3), 6.82 (1H, d, J = 8.4, H-6), 7.00 (1H, d, J = 8.4, H-5), 7.89 (1H, d, J = 9.4, H-4); $^{13}$C NMR (DMSO-$d_6$, δ, ppm): 111.1 (C-3), 112.0 (C-10), 112.4 (C-6), 118.8 (C-5), 132.1 (C-8), 143.7 (C-9), 145.0 (C-4), 149.7 (C-7), 160.3 (C-2) [2].

**Daphnoretin (2):** $C_{19}H_{12}O_7$, gray amorphous powder, mp 243–244°C; ESI-MS *m/z*: 351 $[M–H]^-$; $^1$H NMR (400 MHz, $CD_3OD$, δ, ppm, J/Hz): 3.83 (3H, s, 6-$OCH_3$), 6.37 (1H, d, J = 8.5, H-3′), 6.85 (1H, s, H-8′), 7.11 (1H, dd, J = 8.5, 2.5, H-6), 7.15 (1H, d, J = 2.5, H-8), 7.19 (1H, m, H-5′), 7.68 (1H, d, J = 8.5, H-5), 7.86 (1H, s, H-4′), 8.02 (1H, d, J = 8.5, H-4), 10.22 (1H, s, 7-OH); $^{13}$C NMR ($CD_3OD$, δ, ppm): 56.0 (6-$OCH_3$), 102.7 (C-8), 103.9 (C-8′), 109.4 (C-5′), 110.1 (C-10′), 113.4 (C-6), 113.8 (C-3), 114.4 (C-10), 129.8 (C-5), 130.8 (C-4′), 135.7 (C-3′), 143.9 (C-4), 145.6 (C-6′), 147.4 (C-7′), 150.3 (C-9′), 154.9 (C-9), 156.8 (C-7), 156.9 (C-2′), 159.8 (C-2) [3].

**Daphneticin (3):** $C_{20}H_{18}O_8$, yellow amorphous powder, mp 236–238°C; EI-MS *m/z* 386 $[M]^+$; $^1$H NMR (400 MHz, DMSO-$d_6$, δ, ppm, J/Hz): 3.41 (1H, m, H-3′), 3.67 (1H, m, H-3′), 3.78 (6H, s, 3″, 5″-$OCH_3$), 4.32 (1H, m, H-2′), 4.32 (1H, d, J = 8.0, H-1′), 4.34 (1H, s, 3′-OH), 6.33 (1H, d, J = 10.0, H-3), 6.76 (2H, s, H-2″, 6″), 6.98 (1H, d, J = 9.0, H-6), 7.21 (1H, d, J = 9.0, H-5), 8.00 (1H, d, J = 10.0, H-4), 8.53 (1H, s, 4″-OH); $^{13}$C NMR (DMSO-$d_6$, δ, ppm): 56.3 (3″, 5″-$OCH_3$), 60.5 (C-3′), 77.6 (C-1′), 79.8 (C-2′), 106.2 (C-2″, 6″), 112.9 (C-6), 113.5 (C-3), 113.6 (C-10), 119.7 (C-5), 126.3 (C-1″), 132.4 (C-4″), 138.2 (C-8), 144.1 (C-4), 147.8 (C-7), 149.4 (C-3″, 5″), 149.7 (C-9), 160.5 (C-2) [4].

**Isodaphneticin (4):** $C_{20}H_{18}O_8$, yellow amorphous powder, mp 254–256°C; EI-MS *m/z* 386 $[M]^+$; $^1$H NMR (400 MHz, DMSO-$d_6$, δ, ppm, J/Hz): 3.41 (1H, m, H-3′), 3.67 (1H, m, H-3′), 3.78 (6H, s, 3″, 5″-$OCH_3$), 4.32 (1H, m, H-2′), 4.32 (1H, d, J = 8.0, H-1′), 4.34 (1H, s, 3′-OH), 6.33 (1H, d, J = 10.0, H-3), 6.76 (2H, s, H-2″, 6″), 6.98 (1H, d, J = 9.0, H-6), 7.21 (1H, d, J = 9.0, H-5), 8.00 (1H, d, J = 10.0, H-4), 8.53 (1H, s, 4″-OH); $^{13}$C NMR (DMSO-$d_6$, δ, ppm): 56.1 (3″, 5″-$OCH_3$), 59.8 (C-3′), 76.6 (C-1′), 78.0 (C-2′), 105.5 (C-2″, 6″), 112.7 (C-6), 113.1 (C-3), 113.3 (C-10), 119.7 (C-5), 125.8 (C-1″), 131.0 (C-4″), 136.2 (C-8), 143.0 (C-4), 144.8 (C-7), 146.7 (C-9), 147.9 (C-3″, 5″), 159.8 (C-2) [5].

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**(–)-Pinoresinol (5):** $C_{22}H_{22}O_6$, white amorphous powder, mp 122–123°C, ESI-MS *m/z*: 381 $[M + Na]^+$; $^1$H NMR (500 MHz, $CD_3OD$, δ, ppm, J/Hz): 3.23 (2H, s, H-1, 5), 3.83 (6H, s, 3′, 3″-$OCH_3$), 4.25 (2H, m, H-4a, 8a), 4.73 (2H, d, J = 4.0, H-4b, 8b), 4.85 (2H, s, H-2, 6), 6.76 (2H, d, J = 8.0, H-5′, 5″), 6.80 (2H, dd, J = 8.0, 2.0, H-6′, 6″), 6.92 (2H, d, J = 2.0, H-2′, 2″); $^{13}$C NMR ($CD_3OD$, δ, ppm): 55.4 (C-1, 5), 56.4 (2′, 2″-$OCH_3$), 72.6 (C-2, 6), 87.5 (C-4, 8), 111.1 (C-2′, 2″), 116.1 (C-5′, 5″), 120.1 (C-6′, 6″), 133.8 (C-1′, 1″), 147.3 (C-4′, 4″), 149.1 (C-3′, 3″) [6].

**Daphneolone (6):** $C_{17}H_{18}O_3$, white amorphous powder, mp 118–120°C; ESI-MS *m/z*: 269 $[M–H]^–$; $^1$H NMR (500 MHz, $CD_3OD$, δ, ppm, J/Hz): 1.83 (2H, m, H-4), 2.71 (2H, m, H-5), 3.08 (1H, m, H-2), 3.09 (1H, m, H-2), 4.17 (1H, m, H-3), 6.82 (2H, d, J = 7.0, H-3′, 5′), 7.14 (1H, m, H-4″), 7.18 (4H, m, H-2″, 3″, 5″, 6″), 7.86 (2H, d, J = 7.0, H-2′, 6′); $^{13}$C NMR ($CD_3OD$, δ, ppm): 32.9 (C-5), 40.3 (C-4), 46.6 (C-2), 68.9 (C-3), 112.4 (C-3′, 5′), 126.7 (C-4″), 129.4 (C-3″, 5″), 129.4 (C-2″, 6″), 130.4 (C-1′), 132.0 (C-2′, 6′), 143.4 (C-1″), 163.9 (C-4′), 200.0 (C-1) [7].

**β-Sitosterol (7):** white needles, mp 124–125°C. The physical data of **7** is consistent with that of β-sitosterol and showed the same color and equal $R_f$ value as a standard substance of β-sitosterol while the compounds were applied on TLC. Furthermore, the melting point of the mixture of **7** and β-sitosterol did not decrease. Therefore, this compound was identified as β-sitosterol.

**Daucostero1 (8):** white needles, mp 275–276°C. Compound **8** showed the same color and equal $R_f$ value as a standard substance of daucostero1 when applied on TLC and eluted with different developing solvents. So the compound was characterized as daucostero1.

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## REFERENCES

1. L. Shu Hui, W. Li Jun, and Y. Hong Ying, *China J. Chin. Mater. Med.*, **27**, 402 (2002).
2. W. Ming Shi, L. Wei Gguo, and X. Li Juan, *J. China Pharm. Univ.*, **15**, 2 (1984).
3. W. Wei Wen, Z. Bin Nan, and W. Cheng Rui, *Chin. Tradit. Herb Drugs*, **26**, 566 (1995).
4. Z. Lin Gen, S. Otto, and W. Hildebert, *Phytochemistry*, **22**, 618 (1983).
5. F. Cottiglia, L. Bonsignore, G. Loy, D. Garau, C. Floris, and M. Casu, *Magn. Reson. Chem.*, **40**, 551 (2002).
6. Z. Lin Gen, *Chin. Tradit. Herb Drugs*, **14**, 3 (1983).
7. S. Kogiso, S. Hosozawa, K. Wada, and K. Munakata, *Phytochemistry*, **13**, 2332 (1974).