STEROIDAL SAPONINS FROM *Paris mairei*

Xiao-xiao Liu,[1] Lei Wang,[2]
Jie Yang,[1] and Qiang Wang[1*]

UDC 547.918

The genus *Paris* (Liliaceae) comprises 24 species in the world, among which 19 species are distributed in China, particularly in the southwestern part of the country [1, 2]. *Rhizoma paridis* and its preparations have been widely used in traditional Chinese medicine for the treatment of injuries, fractures, hemorrhage, and parotitis [2]. To the best of our knowledge, more than 50 steroidal saponins have been isolated from thirteen *Paris* species; however, no phytochemical study on *P. mairei* has previously been reported . As part of a systematic examination of the Chinese species, we have investigated the chemical components of the title plant.

The rhizomes of *P. mairei* were collected in May 2005, in Yunnan province of China, and were authenticated by Pr. Qiang Wang (Department of Chinese Materia Medica Analysis, China Pharmaceutical University, China). A voucher specimen was deposited in the Herbarium of the Department of Pharmacognosy, China Pharmaceutical University, China (MAL020).

Dry raw material (15 kg) was extracted three times with ethanol (80%). The extract was concentrated and suspended in H_2O, then successively extracted with petroleum ether, chloroform, and *n*-butanol.

The *n*-butanol extract was applied to an HPD-100 macroreticular resin column and eluted with water and aqueous alcohol (20% and 60%). The 60% alcohol fraction was rechromatographed on silica gel eluted with chloroform–methanol gradient. The subfractions were purified on Sephadex LH-20 and ODS to afford compounds **1**–**7**. These compounds were elucidated by IR, ^{1}H NMR, ^{13}C NMR, HSQC, HMBC, and MS analysis. All these results were in good agreement with the literature data.

Compound **1**: colorless needles (MeOH), mp 270–272°C. Negative ESI-MS: *m/z* 915 $[M–H]^-$.

^{1}H NMR (500 MHz, Py-d_5, δ, ppm, J/Hz): 6.38 (1H, d, J = 1.1, H-1″), 5.66 (1H, d, J = 5.0, H-7), 5.07 (1H, d, J = 7.5, H-1‴), 4.85 (1H, d, J = 8.0, H-1′), 4.82 (1H, H-3), 4.52 (1H, H-16), 4.25(1H, d, H-6), 3.51 (1H, H-26b), 3.40 (1H, H-26a), 1.75 (3H, d, J = 6.0, CH_3-6″), 1.49 (3H, s, CH_3-19), 1.08 (3H, d, J = 6.5, CH_3-21), 0.75 (3H, s, CH_3-18), 0.65 (3H, d, J = 6.0, CH_3-27).

^{13}C NMR (125 MHz, Py-d_5, δ, ppm): 34.3 (C-1), 30.8 (C-2), 76.6 (C-3), 38.7 (C-4), 76.7 (C-5), 75.4 (C-6), 121.3 (C-7), 141.9 (C-8), 44.2 (C-9), 39.0 (C-10), 23.2 (C-11), 40.7 (C-12), 42.7 (C-13), 55.9 (C-14), 32.6 (C-15), 81.7 (C-16), 63.8 (C-17), 17.5 (C-18), 19.4 (C-19), 43.4 (C-20), 15.8 (C-21), 110.2 (C-22), 32.7 (C-23), 30.2 (C-24), 31.5 (C-25), 67.8 (C-26), 18.2 (C-27), 101.3 (C-1′), 77.8 (C-2′), 90.5 (C-3′), 70.3 (C-4′), 79.5 (C-5′), 63.4 (C-6′), 102.9 (C-1″), 73.6 (C-2″), 73.4 (C-3″), 75.2 (C-4″), 70.4 (C-5″), 19.6 (C-6″), 105.3 (C-1‴), 75.9 (C-2‴), 79.7 (C-3‴), 72.4 (C-4‴), 78.6 (C-5‴), 63.3 (C-6‴).

These assignments were completed on the basis of the results of HSQC and HMBC experiments. This compound is characterized as 3β,5α,6α-trihydroxy-7(8)-en-isospirostanol-3-*O*-β-D-glucopyranosyl(1→3)-[α-L-rhamnopyranosyl(1→2)]-β-D-glucopyranoside [3].

Compound **2**: colorless needles (MeOH), mp 263–265°C. IR spectrum (KBr, ν_{max}, cm^{-1}): 3600–3200 (OH), 980, 920, 900, 890. Nagative ESI-MS: *m/z* 1029 $[M–H]^-$.

^{1}H NMR (500 MHz, Py-d_5, δ, ppm, J/Hz): 6.37 (1H, d, J = 1.0, H-1″), 6.25 (1H, d, J = 1.3, H-1⁗), 5.80 (1H, s, H-1‴), 5.29 (1H, d, J = 5.2, H-6), 3.83 (1H, m, H-3), 3.50 (2H, m, H-26), 1.75 (3H, d, J = 6.2, CH_3-6″), 1.59 (3H, d, J = 5.4, CH_3-6‴), 1.58 (3H, d, J = 5.9, CH_3-6⁗), 1.21 (3H, d, J = 7.2, CH_3-21), 1.07 (3H, s, CH_3-19), 0.94 (3H, s, CH_3-18), 0.67 (3H, d, J = 5.8, CH_3-27).

1) Key Laboratory of Modern Chinese Medicines, Department of Chinese Materia Medica Analysis, China Pharmaceutical University, Nanjing 210038, P. R. China, e-mail: qwang_49@hotmail.com; 2) Department of Phytochemistry, China Pharmaceutical University, Nanjing 210038, P. R. China. Published in Khimiya Prirodnykh Soedinenii, No. 6, pp. 661-662, November-December, 2008. Original article submitted May 1, 2007.

0009-3130/08/4406-0821

^{13}C NMR (125 MHz, Py-d_5, δ, ppm): 37.3 (C-1), 30.0 (C-2), 77.5 (C-3), 38.8 (C-4), 140.6 (C-5), 121.6 (C-6), 32.2 (C-7), 31.6 (C-8), 50.0 (C-9), 37.0 (C-10), 20.8 (C-11), 31.9 (C-12), 44.9 (C-13), 52.8 (C-14), 32.1 (C-15), 89.9 (C-16), 89.8 (C-17), 16.9 (C-18), 19.2 (C-19), 44.6 (C-20), 9.5 (C-21), 109.6 (C-22), 31.9 (C-23), 28.6 (C-24), 30.2 (C-25), 66.5 (C-26), 17.1 (C-27), 100.1 (C-1′), 77.9 (C-2′), 76.8 (C-3′), 77.8 (C-4′), 77.6 (C-5′), 61.0 (C-6′), 102.0 (C-1″), 72.3 (C-2″), 72.6 (C-3″), 73.9 (C-4″), 69.3 (C-5″), 18.4 (C-6″), 102.0 (C-1‴), 73.0 (C-2‴), 72.6 (C-3‴), 80.2 (C-4‴), 68.1 (C-5‴), 18.6 (C-6‴), 103.0 (C-1⁗), 72.4 (C-2⁗), 72.6 (C-3⁗), 73.8 (C-4⁗), 70.2 (C-5⁗), 18.2 (C-6⁗).

The compound was identified as 25(*R*)-pennogenin-3-*O*-α-L-rhamnopyranosyl(1→4)-α-L-rhamnopyranosyl(1→4)-[α-L-rhamnopyranosyl(1→2)]-β-D-glucopyranoside [4].

Compound **3**: colorless needles (MeOH), mp 276–278°C. IR spectrum (KBr, ν_{max}, cm^{-1}): 3600–3200 (OH), 980, 920, 900, 890. Nagative ESI-MS: *m/z* 869 $[M–H]^-$.

^{1}H NMR (500 MHz, Py-d_5, δ, ppm, J/Hz): 6.25 (1H, d, J = 1.2, H-1″), 5.89 (1H, d, J = 1.7, H-1‴), 5.28 (1H, d, J = 5.1, H-6), 3.83 (1H, m, H-3), 3.50 (2H, m, H-26), 1.74 (3H, d, J = 6.2, CH_3-6″), 1.21 (3H, d, J = 7.2, CH_3-21), 1.07 (3H, s, CH_3-19), 0.95 (3H, s, CH_3-18), 0.68 (3H, d, J = 5.8, CH_3-27).

^{13}C NMR (125 MHz, Py-d_5, δ, ppm): 37.6(C-1), 30.2 (C-2), 77.7 (C-3), 39.0 (C-4), 140.8 (C-5), 121.8 (C-6), 32.5 (C-7), 31.8 (C-8), 50.3 (C-9), 37.2 (C-10), 21.0 (C-11), 32.1 (C-12), 45.1 (C-13), 53.0 (C-14), 32.4 (C-15), 90.2 (C-16), 90.1 (C-17), 17.2 (C-18), 19.5 (C-19), 44.8 (C-20), 9.7 (C-21), 109.9 (C-22), 32.1 (C-23), 28.8 (C-24), 30.4 (C-25), 66.7 (C-26), 17.3 (C-27), 100.2 (C-1′), 78.2 (C-2′), 77.4 (C-3′), 76.7 (C-4′), 77.1 (C-5′), 62.5 (C-6′), 101.9 (C-1″), 72.5 (C-2″), 72.8 (C-3″), 74.2 (C-4″), 69.5 (C-5″), 18.7 (C-6″), 109.7 (C-1‴), 82.7 (C-2‴), 77.9 (C-3‴), 86.7 (C-4‴, 61.4 (C-5‴).

These spectral data led to the structure of 25(*R*)-pennogenin-3-*O*-α-L-arabinofuranosyl (1→4)-[α-L-rhamnopyranosyl (1→2)]-β-D-glucopyranoside [4].

Compound **4**: colorless needles (MeOH), mp 261–263°C. IR spectrum (KBr, ν_{max}, cm^{-1}): 3600–3200 (OH), 980, 920, 900, 890. Nagative ESI-MS: *m/z* 737 $[M – H]^-$.

^{1}H NMR (500 MHz, Py-d_5, δ, ppm, J/Hz): 6.20 (1H, d, J = 1.2, H-1″), 5.27 (1H, d, J = 5.1, H-6), 3.83 (1H, m, H-3), 3.51 (2H, m, H-26), 1.74 (3H, d, J = 6.2, CH_3-6″), 1.20 (3H, d, J = 7.2, CH_3-21), 1.07 (3H, s, CH_3-19), 0.95 (3H, s, CH_3-18), 0.68 (3H, d, J = 5.8, CH_3-27).

^{13}C NMR (125MHz, Py-d_5, δ, ppm): 37.6 (C-1), 30.0 (C-2), 78.2 (C-3), 39.0 (C-4), 140.9 (C-5), 121.8 (C-6), 32.5 (C-7), 31.8 (C-8), 50.3 (C-9), 37.2 (C-10), 21.0 (C-11), 32.1 (C-12), 45.1 (C-13), 53.1 (C-14), 32.4 (C-15), 90.2 (C-16), 90.1 (C-17), 17.3 (C-18), 19.5 (C-19), 44.8 (C-20), 9.7 (C-21), 109.8 (C-22), 32.1 (C-23), 28.8 (C-24), 30.4 (C-25), 66.7 (C-26), 17.2 (C-27), 100.4 (C-1′), 79.7 (C-2′), 77.8 (C-3′), 71.9 (C-4′), 77.9 (C-5′), 62.7 (C-6′), 102.1 (C-1″), 72.6 (C-2″), 72.9 (C-3″), 74.2 (C-4″), 69.5 (C-5″), 18.7 (C-6″).

These spectral data led to the structure of 25(*R*)-pennogenin-3-*O*-α-L-rhamnopyranosyl(1→2)-β-D-glucopyranoside [4].

Compound **5**: colorless needles (MeOH), mp 295–297°C. IR spectrum (KBr, ν_{max}, cm^{-1}): 3500–3400 (OH), 980, 917, 898, 880. Nagative ESI-MS: *m/z* 883 $[M–H]^-$.

^{1}H NMR (500 MHz, Py-d_5, δ, ppm, J/Hz): 6.37 (1H, d, J = 1.1, H-1″), 5.32 (1H, d, J = 5.2, H-6), 5.01 (1H, d, J = 8.0, H-1‴), 3.82 (1H, m, H-3), 3.59 (2H, m, H-26), 1.75 (3H, d, J = 6.2, CH_3-6″), 1.13 (3H, d, J = 6.9, CH_3-21), 1.05 (3H, s, CH_3-19), 0.82 (3H, s, CH_3-18), 0.68 (3H, d, J = 5.7, CH_3-27).

^{13}C NMR (125 MHz, Py-d_5, δ, ppm): 37.3(C-1), 29.9 (C-2), 78.3 (C-3), 38.5 (C-4), 140.6 (C-5), 121.7 (C-6), 32.1 (C-7), 31.6 (C-8), 50.0 (C-9), 36.9 (C-10), 20.9 (C-11), 39.7 (C-12), 40.2 (C-13), 56.4 (C-14), 32.0 (C-15), 80.9 (C-16), 62.7 (C-17), 16.1 (C-18), 19.2 (C-19), 41.8 (C-20), 14.8 (C-21), 109.0 (C-22), 31.5 (C-23), 29.0 (C-24), 30.4 (C-25), 66.6 (C-26), 17.1 (C-27), 99.8 (C-1′), 78.5 (C-2′), 89.3 (C-3′), 71.3 (C-4′), 77.7 (C-5′), 62.2 (C-6′), 102.0 (C-1″), 72.3 (C-2″), 72.6 (C-3″), 73.9 (C-4″), 69.4 (C-5″), 18.4 (C-6″), 104.3 (C-1‴), 74.8 (C-2‴), 76.8 (C-3‴), 69.4 (C-4‴), 77.5 (C-5‴), 62.2 (C-6‴).

This compound is characterized as 25 (*R*) diosgenin-3-*O*-β-D-glucopyranosyl (1→3)-[α-L-rhamnopyranosyl (1→2)]-β-D-glucopyranoside [5].

Compound **6**: colorless needles (MeOH), mp 274–276°C. IR spectrum (KBr, ν_{max}, cm^{-1}): 3500–3300 (OH), 982, 920, 898, 866. Nagative ESI-MS: *m/z* 853 $[M-H]^-$.

^{1}H NMR (500 MHz, Py-d_5, δ, ppm, J/Hz): 6.21 (1H, d, J = 1.1, H-1″), 5.85 (1H, d, J = 1.7, H-1‴), 5.25 (1H, d, J = 5.2, H-6), 3.83 (1H, m, H-3), 3.54 (2H, m, H-26), 1.70 (3H, d, J = 6.7, CH_3-6″), 1.08 (3H, d, J = 6.9, CH_3-21), 1.01 (3H, s, CH_3-19), 0.78 (3H, s, CH_3-18), 0.64 (3H, d, J = 5.7, CH_3-27).

^{13}C NMR (125 MHz, Py-d_5, δ, ppm): 38.1(C-1), 30.7 (C-2), 77.7 (C-3), 39.6 (C-4), 141.4 (C-5), 122.4 (C-6), 32.9 (C-7), 32.3 (C-8), 50.9 (C-9), 37.7 (C-10), 21.7 (C-11), 40.5 (C-12), 41.1 (C-13), 57.2 (C-14), 32.8 (C-15), 81.7 (C-16), 63.5 (C-17), 16.9 (C-18), 20.0 (C-19), 42.6 (C-20), 15.9 (C-21), 109.9 (C-22), 32.4 (C-23), 29.9 (C-24), 31.2 (C-25), 67.5 (C-26), 17.9 (C-27), 100.8 (C-1′), 78.5 (C-2′), 78.3 (C-3′), 77.3 (C-4′), 78.0 (C-5′), 63.1 (C-6′), 102.5 (C-1″), 73.4 (C-2″), 73.0 (C-3″), 74.7 (C-4″), 70.0 (C-5″), 19.2 (C-6″), 110.2 (C-1‴), 83.2(C-2‴), 78.7 (C-3‴), 87.3 (C-4‴), 62.0 (C-5‴).

This compound is characterized as 25(*R*)-diosgenin-3-*O*-α-L-arabinofuranosyl (1→4)-[α-L-rhamnopyranosyl (1→2)]-β-D-glucopyranoside [6, 7].

Compound **7**: colorless needles (MeOH), mp 240–242°C. IR spectrum (KBr, ν_{max}, cm^{-1}): 3600–3200 (OH), 980, 918, 900, 865. Nagative ESI-MS: *m/z* 721 $[M - H]^-$.

^{1}H NMR (500 MHz, Py-d_5, δ, ppm, J/Hz): 6.36 (1H, d, J = 1.2, H-1″), 5.31 (1H, d, J = 5.2, H-6), 3.94 (1H, m, H-3), 3.49 (2H, m, H-26), 1.77 (3H, d, J = 6.2, CH_3-6″), 1.13 (3H, d, J = 7.4, CH_3-21), 1.05 (3H, s, CH_3-19), 0.82 (3H, s, CH_3-18), 0.68 (3H, d, J = 5.7, CH_3-27).

^{13}C NMR (125 MHz, Py-d_5, δ, ppm): 37.3(C-1), 30.0 (C-2), 78.1 (C-3), 38.8 (C-4), 140.7 (C-5), 121.5 (C-6), 32.1 (C-7), 31.6 (C-8), 50.1 (C-9), 37.0 (C-10), 21.0 (C-11), 39.7 (C-12), 40.3 (C-13), 56.5 (C-14), 32.0 (C-15), 80.9 (C-16), 62.7 (C-17), 16.1 (C-18), 19.2 (C-19), 41.8 (C-20), 14.8 (C-21), 109.0 (C-22), 31.5 (C-23), 29.1 (C-24), 30.4 (C-25), 66.7 (C-26), 17.1 (C-27), 100.2 (C-1′), 79.5 (C-2′), 77.8 (C-3′), 71.7 (C-4′), 77.7 (C-5′), 62.5 (C-6′), 101.9 (C-1″), 72.4 (C-2″), 72.7 (C-3″), 74.0 (C-4″), 69.3 (C-5″), 18.5 (C-6″).

Identified as 25(*R*)-diosgenin-3-*O*-α-L-rhamnopyranosyl (1→2)-β-D-glucopyranoside [8].

All these steroidal saponins were isolated for the first time from *P. mairei.*

REFERENCES

1. *Flora Reipublicae Popularis Sinicae*, Institute of Botany, Chinese Academy of Sciences, Science Press, Beijing, 1978, Vol. **15**, 86 p.
2. H. Li, *The genus Paris*, Science Press, Beijing, 1998.
3. Y. Huang, L. J. Cui, Q. Wang, and W. C. Ye, *Acta Pharm. Sinica*, **41**, 361 (2006).
4. L. P. Kang, B. P. Ma, J. Zhang, C. Q. Xiong, D. W. Tan, and Y. W. Cong, *Chin. J. Med. Chem.* (chinese), **15**, 25 (2005).
5. C. X. Chen and J. Zhou, *Acta Botanica Yunnanica* (chinese), **6**, 111 (1984).
6. C. X. Chen, J. Zhou, Y. T. Zhang, Ch. And K. Gao, *Acta Botanica Yunnanica* (chinese), **5**, 219 (1983).
7. T. Nohara, H. Yabuta, M. Suenobu, R. Hida, K. Miyahara, and T. Kawasaki, *Chem. Pharm. Bull.*, **21**, 1240 (1973).
8. Y. Watanabe, S. Sanada, Y. Ida, and J. Shoji, *Chem. Pharm. Bull.*, **31**, 3486 (1983).