Two New Homo-aro-cholestane Glycosides and a New Cholestane Glycoside from the Roots and Rhizomes of *Paris polyphylla* var. *pseudothibetica*

by **Cao-Mao Xiao**[a]), **Jing Huang***[a])[b]), **Xue-Mei Zhong**[a]), **Xiao-Yan Tan**[a]), and **Peng-Chi Deng**[c])

[a]) Department of Chemistry of Medicinal Natural Products, West China School of Pharmacy, Sichuan University, Chengdu 610041, P. R. China (phone and fax: +86-28-85503045; e-mail: huangj_pharm@scu.edu.cn)

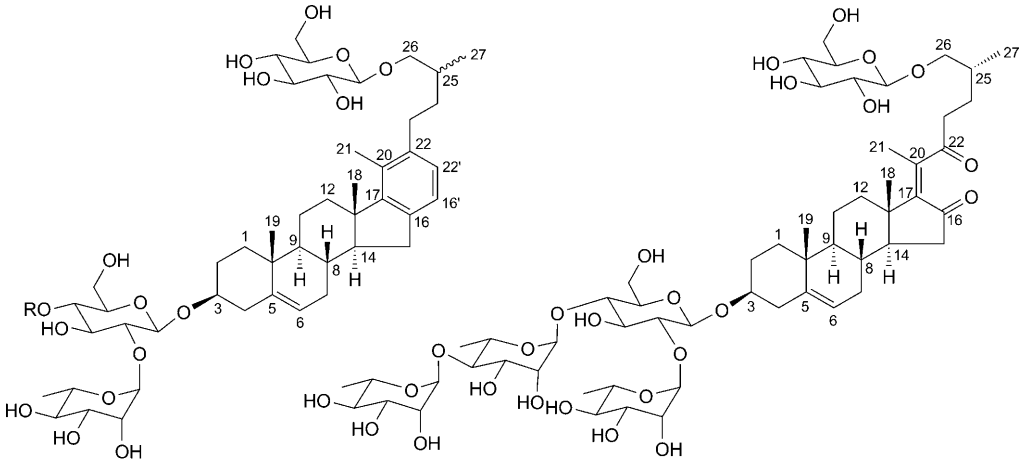
[b]) State Key Laboratory of Oral Diseases, Sichuan University, Chengdu 610041, P. R. China

[c]) Analytical and Testing Center, Sichuan University, Chengdu 610041, P. R. China

Two new homo-aro-cholestane glycosides and a new cholestane glycoside, along with three known saponins, were isolated from the 95% EtOH extract of the roots and rhizomes of *Paris polyphylla* var. *pseudothibetica*. The structures of the new compounds were elucidated as 3β-*O*-{α-L-rhamnopyranosyl-(1 → 4)-α-L-rhamnopyranosyl-(1 → 4)-[α-L-rhamnopyranosyl-(1 → 2)]}-β-D-glucopyranosylhomo-aro-cholest-5-ene-26-*O*-β-D-glucopyranoside (parispseudoside A, **1**), 3β-*O*-α-L-rhamnopyranosyl-(1 → 2)-β-D-glucopyranosylhomo-aro-cholest-5-ene-26-*O*-β-D-glucopyranoside (parispseudoside B, **2**), and (25*R*)-3β-*O*-{α-L-rhamnopyranosyl-(1 → 4)-α-L-rhamnopyranosyl-(1 → 4)-[α-L-rhamnopyranosyl-(1 → 2)]}-β-D-glucopyranosyl-cholesta-5,17(20)-diene-16,22-dione-26-*O*-β-D-glucopyranoside (parispseudoside C, **3**) by spectroscopic methods, including 1D- and 2D-NMR, and MS experiments, as well as chemical evidences.

Introduction. – *Paris polyphylla* SMITH var. *pseudothibetica* H. LI (Trilliaceae), mainly distributed in the Yunnan and Sichuan provinces of China [1], has been used as a traditional medicine to treat parotitis, fractures, intoxication, and for hemostasis [2]. The plant has also been used for the treatment of tumors, immune unbalance, and as an analgesics [3]. The steroidal saponins are regarded as the chief bioactive ingredients, and 50 steroidal saponins have been isolated from 13 *Paris* species [4]. *Liu et al.* detected 10 steroidal saponins from *P. polyphylla* var. *pseudothibetica* by HPLC-ESI-MS method [4]. In our phytochemical investigation, two new homo-aro-cholestane saponins and a new cholestane saponin, named parispseudosides A, B, and C (**1**–**3**, resp.), along with three known compounds, were isolated from the title plant. Here, we describe the isolation and structural elucidation of the three new compounds.

Results and Discussion. –The dried roots and rhizomes of *P. polyphylla* var. *pseudothibetica* were extracted with 95% EtOH. After concentration under reduced pressure, the extract was suspended in H_2O and partitioned with petroleum ether, AcOEt, and BuOH, respectively. The BuOH extract was subjected to repeated column chromatography to afford parispseudosides A (**1**, 32 mg), B (**2**, 4 mg), C (**3**, 21 mg), parisyunnanoside F (8 mg) [5], chonglouoside VII (37 mg) [6], and parispolyside E (18 mg) [7].



1 R = α-L-rhamnosyl-(1→4)-α-L-rhamnosyl
2 R = H

3

Compound **1** was obtained as a colorless amorphous powder, and the positive reactions for the Ac_2O/H_2SO_4 and α-naphthol/H_2SO_4 tests indicated that **1** was a saponin. The IR spectrum revealed the presence of OH (3419 cm^{-1}) and Me groups (2933 cm^{-1}), a C=C bond (1640 cm^{-1}), and an aromatic ring (1568, 1451 cm^{-1}). The molecular formula of **1** was determined to be $C_{59}H_{92}O_{24}$ based on the HR-ESI-MS data (*m*/*z* 1207.5864 ($[M + Na]^+$)). The ^{13}C-NMR spectrum of **1** indicated 59 C-atoms, 29 of which were assigned to the aglycone moiety, while 30 were assigned to the carbohydrate moiety. The ^{1}H- and ^{13}C-NMR spectra (*Tables 1* and *2*) of **1** showed four characteristic Me signals (δ(H) 0.93 (*s*), 1.04 (*d*, *J* = 6.4), 1.11 (*s*), and 2.32 (*s*); δ(C) 14.7, 16.6, 17.4, and 19.4), aromatic ring signals (δ(H) 7.10 (*d*, *J* = 7.6) and 7.04 (*d*, *J* = 7.6); δ(C) 122.9, 127.5, 131.3, 139.8, 140.7, and 151.8), as well as signals for C=C bonds (δ(H) 5.37 (br. *s*); δ(C) 121.8 and 141.1), indicating a steroidal aglycone with a benzene ring and two olefinic C-atoms in **1**. Location of the C=C bond at C(5)/C(6) and fusion of the benzene ring at C(16)/C(17) were determined from the long-distance correlations (Me(19) (δ(H) 1.11)/C(5) (δ(C) 141.1); H−C(6) (δ(H) 5.37)/C(4) (δ(C) 39.0) and C(7) (δ(C) 32.4); Me(18) (δ(H) 0.93)/C(17) (δ(C) 151.8); Me(21) (δ(H) 2.32)/C(17) (δ(C) 151.8), C(20) (δ(C) 131.3), and C(22) (δ(C) 139.8); H−C(22′) (δ(H) 7.04)/C(16) (δ(C) 140.7) and C(20) (δ(C) 131.3); and H−C(16′) (δ(H) 7.10)/C(17) (δ(C) 151.8), C(22) (δ(C) 139.8)) in the HMBC spectrum of **1** (*Fig. 1*). Comparison with literature data showed that compound **1** had the same aglycone as parispolyside E [7], and aethiosides A and B [8], namely homo-aro-cholest-5-ene-3β,26-diol.

TLC Acid hydrolysis of **1** gave glucose and rhamnose, and the absolute configurations of the sugars were assigned to be D-glucose and L-rhamnose according to those commonly found in the steroidal saponins [9]. The ^{1}H- and ^{13}C-NMR data (*Table 2*) of **1** revealed the presence of two D-glucose and three L-rhamnose units (five anomeric H-atom signals at δ(H) 4.87 (*d*, *J* = 6.4), 4.96 (*d*, *J* = 6.8), 5.85 (br. *s*), 6.30 (br. *s*), and 6.41 (br. *s*), two Me H-atom signals at δ(H) 1.60 (*d*, *J* = 5.6) and 1.78 (*d*,

Table 1. *^{1}H- and ^{13}C-NMR Data* (C_5D_5N, 400 and 100 MHz, resp.) *of the Aglycones of Parispseudosides A* (**1**)*, B* (**2**)*, and C* (**3**). δ in ppm, J in Hz.

	Parispseudoside A (**1**)		Parispseudoside B (**2**)		Parispseudoside C (**3**)	
	δ(H)	δ(C)	δ(H)	δ(C)	δ(H)	δ(C)
H_α–C(1)	0.96–1.00 (*m*)	37.4	0.96–1.02 (*m*)	37.3	0.98–1.06 (*m*)	37.0
H_β–C(1)	1.72–1.78 (*m*)		1.70–1.78 (*m*)		1.74–1.80 (*m*)	
H_β–C(2)	1.84–1.90 (*m*)	30.2	1.85–1.90 (*m*)	30.2	1.82–1.88 (*m*)	30.0
H_α–C(2)	2.06–2.12 (*m*)		2.06–2.10 (*m*)		2.04–2.08 (*m*)	
H_α–C(3)	3.84–3.92 (*m*)	78.1	3.86–3.90 (*m*)	78.2	3.85–3.94 (*m*)	77.9
H_β–C(4)	2.70–2.78 (*m*)	39.0	2.70–2.76 (*m*)	39.0	2.71–2.77 (*m*)	38.9
H_α–C(4)	2.82–2.90 (*m*)		2.82–2.90 (*m*)		2.76–2.82 (*m*)	
C(5)	–	141.1	–	141.1	–	140.9
H–C(6)	5.37 (br. *s*)	121.8	5.37 (br. *s*)	121.7	5.27 (*d*, $J = 4.0$)	121.3
H_α–C(7)	2.50–2.54 (*m*)	32.4	2.52–2.56 (*m*)	32.4	1.48–1.56 (*m*)	31.7
H_β–C(7)	2.60–2.64 (*m*)		2.60–2.65 (*m*)		1.72–1.76 (*m*)	
H_β–C(8)	1.66–1.68 (*m*)	30.9	1.66–1.68 (*m*)	30.9	1.50–1.54 (*m*)	30.7
H_α–C(9)	1.00–1.08 (*m*)	50.5	1.00–1.08 (*m*)	50.5	1.00–1.06 (*m*)	49.9
C(10)	–	37.1	–	37.1	–	37.1
H_α–C(11)	1.50–1.56 (*m*)	21.3	1.52–1.58 (*m*)	21.2	1.49–1.55 (*m*)	20.8
H_β–C(11)	1.58–1.62 (*m*)		1.60–1.64 (*m*)		1.52–1.57 (*m*)	
H_α–C(12)	1.60–1.64 (*m*)	37.0	1.58–1.64 (*m*)	36.9	1.52–1.58 (*m*)	36.0
H_β–C(12)	1.68–1.72 (*m*)		1.67–1.73 (*m*)		2.12–2.16 (*m*)	
C(13)	–	47.2	–	47.1	–	43.4
H_α–C(14)	1.50–1.58 (*m*)	57.7	1.50–1.58 (*m*)	57.7	1.38–1.42 (*m*)	50.4
H_α–C(15)	2.56–2.64 (*m*)	31.3	2.56–2.64 (*m*)	31.3	2.68–2.76 (*m*)	38.7
H_β–C(15)	2.68–2.74 (*m*)		2.68–2.74 (*m*)		2.74–2.82 (*m*)	
C(16)	–	140.7	–	140.7	–	210.3
C(17)	–	151.8	–	151.8	–	142.5
Me(18)	0.93 (*s*)	16.6	0.93 (*s*)	16.6	0.94 (*s*)	16.7
Me(19)	1.11 (*s*)	19.4	1.11 (*s*)	19.3	1.05 (*s*)	19.3
C(20)	–	131.3		131.2	–	145.6
Me(21)	2.32 (*s*)	14.7	2.33 (*s*)	14.6	1.97 (*s*)	15.7
H–C(22)	–	139.8	–	139.7	–	205.6
CH_2(23)	1.94–1.98 (*m*), 2.04–2.08 (*m*)	32.0	1.94–1.98 (*m*), 2.02–2.06 (*m*)	32.0	1.98–2.04 (*m*), 2.18–2.22 (*m*)	37.9
CH_2(24)	1.44–1.52 (*m*), 1.80–1.88 (*m*)	35.5	1.42–1.57 (*m*), 1.82–1.87 (*m*)	35.4	1.86–1.92 (*m*), 2.10–2.14 (*m*)	27.9
H–C(25)	1.98–2.04 (*m*)	34.1	1.98–2.04 (*m*)	34.1	2.00–2.04 (*m*)	33.4
CH_2(26)	3.68–3.72 (*m*), 3.96–4.04 (*m*)	75.0	3.66–3.70 (*m*), 3.94–4.02 (*m*)	74.9	3.60–3.68 (*m*), 3.94–4.00 (*m*)	75.0
Me(27)	1.04 (*d*, $J = 6.4$)	17.4	1.05 (*d*, $J = 6.8$)	17.3	1.00 (*d*, $J = 6.4$)	17.4
H–C(16′)	7.10 (*d*, $J = 7.6$)	122.9	7.12 (*d*, $J = 7.6$)	122.9		
H–C(22′)	7.04 (*d*, $J = 7.6$)	127.5	7.04 (*d*, $J = 7.6$)	127.4		

$J = 6.0$), and five anomeric C-atom signals at δ(C) 100.4, 102.2, 102.3, 103.2, and 104.9, as well as three Me C-atom signals at δ(C) 18.4, 18.6, and 18.8). The anomeric configuration of the glucose and rhamnose residues was determined as β and α, respectively, from the J values of their anomeric H-atoms.

Table 2. *^{1}H- and ^{13}C-NMR Data* (C_5D_5N, 400 and 100 MHz, resp.) *of the Sugars of Parispseudosides A* (**1**)*, B* (**2**)*, and C* (**3**). δ in ppm, J in Hz.

	Parispseudoside A (**1**)		Parispseudoside B (**2**)		Parispseudoside C (**3**)	
	δ(H)	δ(C)	δ(H)	δ(C)	δ(H)	δ(C)
3-*O*-Glc I						
H–C(1)	4.96 (*d*, *J* = 6.8)	100.4	5.06 (*d*, *J* = 8.0)	100.3	4.96 (*d*, *J* = 8.0)	100.3
H–C(2)	4.22–4.28 (*m*)	78.0	4.28–4.36 (*m*)	79.6	4.24–4.28 (*m*)	77.9
H–C(3)	3.62–3.72 (*m*)	77.0	3.68–3.78 (*m*)	77.9	3.61–3.68 (*m*)	76.9
H–C(4)	4.36–4.44 (*m*)	77.7[a])	4.40–4.48 (*m*)	74.1	4.38–4.44 (*m*)	77.7[a])
H–C(5)	4.18–4.24 (*m*)	77.7[a])	4.21–4.29 (*m*)	77.9	4.23–4.29 (*m*)	77.7[a])
CH_2(6)	4.02–4.06 (*m*), 4.18–4.28 (*m*)	61.3	4.02–4.06 (*m*), 4.20–4.28 (*m*)	62.7	4.04–4.08 (*m*), 4.22–4.28 (*m*)	61.2
Rha I						
H–C(1)	6.41 (br. *s*)	102.2	6.42 (br. *s*)	102.0	6.41 (br. *s*)	102.1
H–C(2)	4.78–4.86 (*m*)	72.4	4.84–4.92 (*m*)	72.5	4.84–4.90 (*m*)	72.4
H–C(3)	4.58–4.64 (*m*)	72.8[a])	4.65–4.74 (*m*)	72.8	4.63–4.70 (*m*)	72.8[a])
H–C(4)	4.34–4.42 (*m*)	74.1	4.32–4.40 (*m*)	71.8	4.36–4.44 (*m*)	74.1
H–C(5)	4.96–5.04 (*m*)	69.5	5.00–5.06 (*m*)	69.4	4.94–5.02 (*m*)	69.5
Me(6)	1.78 (*d*, *J* = 6.0)	18.6	1.81 (*d*, *J* = 6.0)	18.6	1.78 (*d*, *J* = 6.0)	18.6
Rha II						
H–C(1)	5.85 (br. *s*)	102.3	–	–	5.85 (br. *s*)	102.2
H–C(2)	4.50–4.60 (*m*)	73.2	–	–	4.56–4.62 (*m*)	73.2
H–C(3)	4.44–4.54 (*m*)[a])	72.8 [a])	–	–	4.48–4.56 (*m*)[a])	72.8[a])
H–C(4)	4.40–4.48 (*m*)	80.4	–	–	4.42–4.48 (*m*)	80.3
H–C(5)	4.84–4.92 (*m*)	68.3	–	–	4.90–4.94 (*m*)	68.3
Me(6)	1.60 (*d*, *J* = 5.6)	18.8	–	–	1.60 (*d*, *J* = 5.6)	18.8
Rha III						
H–C(1)	6.30 (br. *s*)	103.2	–	–	6.29 (br. *s*)	103.2
H–C(2)	4.60–4.72 (*m*)	72.6	–	–	4.60–4.68 (*m*)	72.6
H–C(3)	4.48–4.54 (*m*)[a])	72.8[a])	–	–	4.50–4.56 (*m*)[a])	72.8[a])
H–C(4)	4.24–4.36 (*m*)	74.0	–	–	4.26–4.34 (*m*)	74.0
H–C(5)	4.30–4.42 (*m*)	70.4	–	–	4.34–4.42 (*m*)	70.3
Me(6)	1.60 (*d*, *J* = 5.6)	18.4	–	–	1.60 (*d*, *J* = 5.6)	18.4
26-*O*-Glc II						
H–C(1)	4.87 (*d*, *J* = 6.4)	104.9	4.87 (*d*, *J* = 8.0)	104.9	4.82 (*d*, *J* = 8.0)	104.8
H–C(2)	4.00–4.10 (*m*)	75.2	4.02–4.08 (*m*)	75.2	3.96–4.04 (*m*)	75.2
H–C(3)	4.04–4.10 (*m*)	78.4	4.05–4.10 (*m*)	78.4	3.94–4.00 (*m*)[a])	78.4
H–C(4)	4.20–2.32 (*m*)	71.8	4.23–2.30 (*m*)	71.8	4.20–2.26 (*m*)	71.7
H–C(5)	4.24–4.30 (*m*)[a])	78.6	4.22–4.30 (*m*)	78.6	4.22–4.30 (*m*)[a])	78.6
CH_2(6)	4.36–4.46 (*m*), 4.54–4.64 (*m*)	62.9	4.34–4.47 (*m*), 4.54–4.62 (*m*)	62.9	4.38–4.46 (*m*), 4.58–4.64 (*m*)	62.8

[a]) Overlapped signals.

The sequence of oligosaccharide chain was deduced from the long-distance correlations in the HMBC experiment (H–C(1) (δ(H) 4.96) of glucopyranosyl unit I with C(3) (δ(C) 78.1) of the aglycone, H–C(1) (δ(H) 4.87) of the glucopyranosyl unit II with C(26) (δ(C) 75.0) of the aglycone, H–C(1) (δ(H) 6.41) of rhamnopyranosyl unit I with C(2) (δ(C) 78.0) of glucopyranosyl unit I, H–C(1) (δ(H) 5.85) of

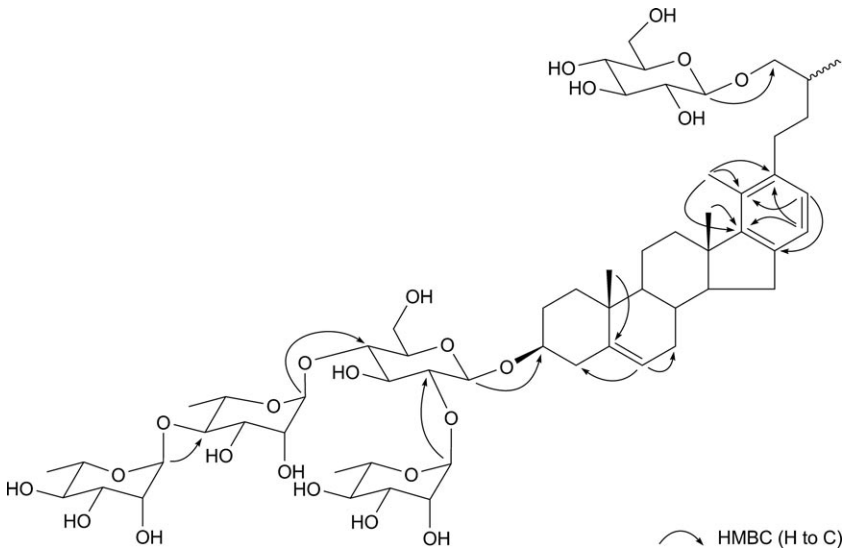


Fig. 1. *Key HMBCs of parispseudoside A* (**1**)

rhamnopyranosyl unit II with C(4) (δ(C) 77.7) of glucopyranosyl unit I, and H–C(1) (δ(H) 6.30) of rhamnopyranosyl unit III with C(4) (δ(C) 80.4) of rhamnopyranosyl unit II). Based on the above findings, the structure of **1** was elucidated as 3β-*O*-{α-L-rhamnopyranosyl-(1 → 4)-α-L-rhamnopyranosyl-(1 → 4)-[α-L-rhamnopyranosyl-(1 → 2)]}-β-D-glucopyranosylhomo-aro-cholest-5-ene-26-*O*-β-D-glucopyranoside[1]), and named parispseudoside A.

Compound **2** was obtained as a colorless amorphous powder and showed positive reactions in the Ac_2O/H_2SO_4 and α-naphthol/H_2SO_4 tests. The molecular formula of **2** was determined to be $C_{47}H_{72}O_{16}$ based on the HR-ESI-MS data (*m*/*z* 915.4708 ([*M* + Na]$^+$)). The ^{1}H- and ^{13}C-NMR data of **2** were very similar to those of **1** (*Tables 1* and *2*), except for the 3-*O*-oligosaccharide chain, indicating the aglycone of **2** was also a homo-aro-cholest-5-ene-3β,26-diol, and that a glucoyranosyl residue was located at C(26) as in **1**.

Regarding the 3-*O*-oligosaccaridic chain, the ^{1}H- and ^{13}C-NMR spectra of **2** (*Table 2*) revealed the presence of only one glucopyranosyl unit and one rhamnopyranosyl unit (δ(H) 5.06 (*d*, *J* = 8.0) and δ(C) 100.3 for the glucopyranosyl unit I, and δ(H) 6.42 (br. *s*), 1.81 (*d*, *J* = 6.0) and δ(C) 102.0, 18.6 for the rhamnopyranosyl unit) in the 3-*O*-oligosaccharide chain. Furthermore, the linkage type of the 3-*O*-oligosaccharide chain in **2** was determined from the long-range correlations between H–C(1) (δ(H) 5.06) of glucopyranosyl unit I and C(3) (δ(C) 78.2) of the aglycone, and H–C(1) (δ(H) 6.42) of the rhamnopyranosyl unit and C(2) (δ(C) 79.6) of glucopyranosyl unit I in the HMBC spectrum. Accordingly, the structure of **2** was elucidated as 3β-*O*-α-L-rhamnopyranosyl-(1 → 2)-β-D-glucopyranosylhomo-aro-cholest-5-ene-26-*O*-β-D-glucopyranoside[1]), and named parispseudoside B.

---

[1]) For systematic names, see *Exper. Part.*

Compound **3** was obtained as a colorless amorphous powder, and showed positive reactions in the Ac_2O/H_2SO_4 and α-naphthol/H_2SO_4 tests. The IR spectrum revealed the presence of OH (3416 cm^{-1}) and Me groups (2933 cm^{-1}), C=O bonds (1710 cm^{-1}), and C=C bonds (1632 cm^{-1}). The molecular formula was established as $C_{57}H_{90}O_{26}$ by the HR-ESI-MS data (*m/z* 1213.5601 ($[M + Na]^+$)). The ^{13}C-NMR data of **3** showed 57 C-atom signals, 27 of which were assigned to the aglycone part, whereas 30 were assigned to the carbohydrate moiety. The ^{1}H- and ^{13}C-NMR spectra (*Tables 1* and *2*) exhibited four characteristic Me signals (δ(H) 0.94 (*s*), 1.00 (*d*, $J = 6.4$), 1.05 (*s*), and 1.97 (*s*); δ(C) 15.7, 16.7, 17.4, and 19.3) and signals attributed to two C=C bonds (δ(H) 5.27 (*d*, $J = 4.0$); δ(C) 121.3, 140.9, 142.5, and 145.6), indicating that the aglycone of **3** should be a cholestan-type steroid. The long-distance correlations (Me(19) (δ(H) 1.05)/C(5) (δ(C) 140.9); H–C(6) (δ(H) 5.27)/C(4) (δ(C) 38.9) and C(7) (δ(C) 31.7); and Me(18) (δ(H) 0.94) and Me(21) (δ(H) 1.97) with C(17) (δ(C) 142.5) and C(20) (δ(C) 145.6)) in the HMBC experiment of **3** (*Fig. 2*) demonstrated that a C=C bond should be placed at C(5)/C(6) and another C=C bond at C(17)/C(20) in **3**. In addition, the ^{13}C-NMR spectrum of **3** showed two CO signals which were assigned to C(16) (δ(C) 210.3) and C(22) (δ(C) 205.6) by the correlations in the HMBC spectrum (H_β–C(15) (δ(H) 2.74–2.82) and Me(21) (δ(H) 1.97) with C(16) (δ(C) 210.3), and of Me(21) (δ(H) 1.97) and H–C(24) (δ(H) 2.10–2.14) with C(22) (δ(C) 205.6)), respectively. The (*R*)-configuration at C(25) in **3** was deduced by the H-atom resonances of CH_2(26) (δ(H) 3.94–4.00 for H_b–C(26) and δ(H) 3.60–3.68 for H_a–C(26)), which showed a Δ(a,b) ($\delta_{Ha} - \delta_{Hb}$) = 0.24–0.40, which is < 0.48 [5][10]. By comparing the NMR data of the aglycone of **3** with those of parisyunnanoside F [5], the aglycone was identified as (25*R*)-3β,26-dihydroxycholesta-5,17(20)-diene-16,22-dione[1]).

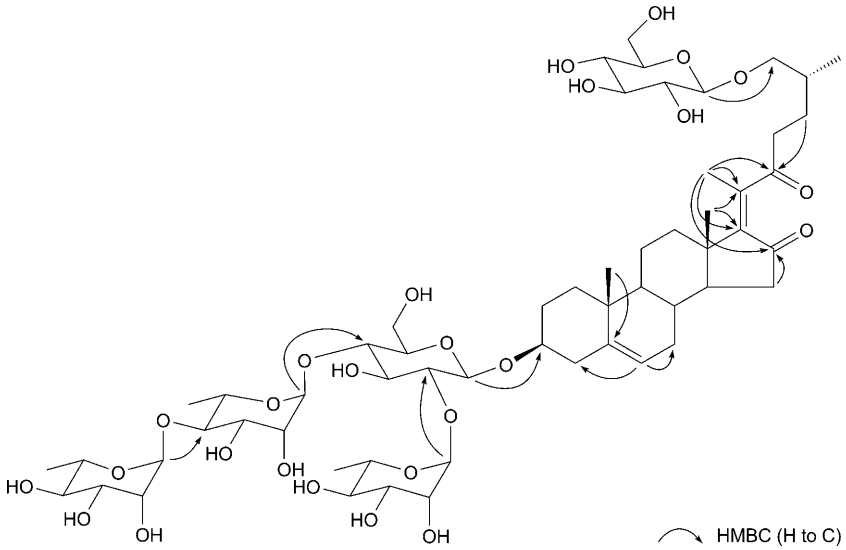


Fig. 2. *Key HMBCs of parispseudoside C* (**3**)

The ^{1}H- and ^{13}C-NMR data of **3** (*Table 2*) revealed the presence of five monosaccharide units (five anomeric signals at δ(H) 4.82 (*d*, $J = 8.0$), 4.96 (*d*, $J = 8.0$), 5.85 (br. *s*), 6.29 (br. *s*), and 6.41 (br. *s*), two Me H-atom signals at δ(H) 1.60 (*d*, $J = 5.6$) and 1.78 (*d*, $J = 6.0$), and five anomeric C-atom signals at δ(C) 100.3, 102.1, 102.2, 103.2, and 104.8, as well as three Me C-atom signals at δ(C) 18.4, 18.6, and 18.8). Based on the acid hydrolysis experiment and NMR techniques, including ^{1}H-NMR, ^{1}H,^{1}H-COSY, HMQC, and HMBC, the five sugar moieties were determined to be two β-D-glucopyranosyl and three α-L-rhamnopyranosyl units, and the connectivity of 3-*O*-oligosaccharide moiety was determined as the same as in **1**. The correlation between H–C(1) (δ(H) 4.82) of glucopyranosyl unit II with C(26) (δ(C) 75.0) of the aglycone in the HMBC experiment indicated that the glucopyranosyl unit II was linked to C(26) of the aglycone of **3**. From the above findings, the structure of **3** was established as (25*R*)-3β-*O*-{α-L-rhamnopyranosyl-(1 → 4)-α-L-rhamnopyranosyl-(1 → 4)-[α-L-rhamnopyranosyl-(1 → 2]}-β-D-glucopyranosylcholesta-5,17(20)-diene-16,22-dione-26-*O*-β-D-glucopyranoside, and named parispseudoside C.

In addition, the three known compounds were identified as parisyunnanoside F (= (25*R*)-26-*O*-β-D-glucopyranosyl-cholesta-5,17(20)-diene-16,22-dione-3β,26-diol-3-*O*-α-L-arabinofuranosyl-(1 → 4)-[α-L-rhamnopyranosyl-(1 → 2)]-β-D-glucopyranoside) [5], chonglouoside VII (= pennogenin 3-*O*-α-L-rhamnopyranosyl-(1 → 4)-α-L-rhamnopyranosyl-(1 → 4)-[α-L-rhamnopyranosyl-(1 → 2)]-β-D-glucopyranoside [6], and parispolyside E (= 3β-*O*-α-L-arabinofuranosyl-(1 → 4)-[α-L-rhamnopyranosyl-(1 → 2)]-β-D-glucopyranosylhomo-aro-cholest-5-ene-26-*O*-β-D-glucopyranoside) [7] by comparison of their ^{1}H- and ^{13}C-NMR, as well as of the ESI-MS data with those reported in the literatures.

The aglycone of **1** and **2** has been already found in parispolyside E, a saponin previously obtained from *P. polyphylla* var. *chinensis* [7]. The compound **3** is reported here for the first time in the genus *Paris*, but the aglycone has been already found in parisyunnanoside F, isolated from *P. polyphylla* var. *yunnanensis* [5]. The aglycone in **3** may be an artifact formed from a furostanol precursor [11]. The known compounds were all found for the first time from *P. polyphylla* var. *pseudothibetica*, but have been already isolated from other species of *Paris* genus [5–7].

Experimental Part

General. All solvents used were of anal. grade (*Tianjin Chemical Plant*, Tianjin, P. R. China). Column chromatography (CC): silica gel (SiO_2; 200–300 mesh; *Qingdao Ocean Chemical Industry Co.*, P. R. China) or *Sephadex LH-20* (*Amersham Biosciences*). Optical rotation: *Perkin-Elmer-241* polarimeter. UV Spectra: *Shimadzu UV-2210-UV/VIS* spectrometer; MeOH solns.; λ_{max} in nm. IR Spectra: *Vector 22-FTIR* spectrometer; in soln. (MeOH) or KBr pellets; in cm^{-1}. NMR Spectra (^{1}H-, ^{13}C-, ^{1}H,^{1}H-COSY, HSQC, and HMBC): *Bruker AV-400* spectrometer, at 400 (^{1}H) or 100 MHz (^{13}C); C_5D_5N solns; δ in ppm *rel*. to $SiMe_4$, *J* in Hz. ESI-MS or HR-ESI-MS: *Bruker micrOTOF-Q* mass spectrometers; in *m*/*z*.

Plant Material. The whole plant of *Paris polyphylla* SMITH var. *pseudothibetica* H. LI was collected from Sichuan province, P. R. China, in June 2006, and was identified by Prof. *Hao Zhang* (West China School of Pharmacy, Sichuan University, Chengdu 610041, P. R. China). A voucher specimen (No. HX.Y060601) was deposited with the West China School of Pharmacy, Sichuan University, Chengdu 610041, P. R. China.

Extraction and Isolation. Dried roots and rhizomes of *P. polyphylla* var. *pseudothibetica* (10 kg) were extracted with 95% EtOH (4 × 10 l) under reflux for 3 h each time, and yielded *ca*. 1300 g of residue after

evaporation of the solvent. The residue was suspended in H_2O (6 l) and partitioned successively with petroleum ether (PE; 3 × 6 l), AcOEt (3 × 6 l), and BuOH (3 × 6 l) to afford a PE extract (103 g), an AcOEt extract (55 g), and a BuOH extract (120 g). The BuOH extract (110 g) was subjected to CC (SiO_2; $CHCl_3$/MeOH/H_2O 10:1:0.1 → 7:3:0.3): *Frs. A–I. Fr. F* (13 g) was subjected to CC (SiO_2; $CHCl_3$/MeOH/H_2O 8:1:0.1 → 3:1:0.1): *Frs. 1–4. Fr. 1* was subjected to CC (SiO_2; $CHCl_3$/MeOH/H_2O 5:1:0.1) to afford chonglouoside VII (37 mg). *Fr. 2* was subjected to CC (SiO_2; $CHCl_3$/MeOH/H_2O 5:1:0.1 → 3:1:0.1; *Sephadex LH-20*; MeOH/H_2O 5:1) to afford **1** (32 mg), **2** (4 mg), and parispolyside E (18 mg). *Fr. 3* was subjected to CC (SiO_2; $CHCl_3$/MeOH/H_2O 5:1:0.1 → 2:1:0.1; *Sephadex LH-20*; MeOH/H_2O 5:1) to afford **3** (21 mg) and parisyunnanoside F (8 mg).

Parispseudoside A (= *3β-*O*-{α-*L*-Rhamnopyranosyl-(1 → 4)-α-*L*-rhamnopyranosyl-(1 → 4)-[α-*L*-rhamnopyranosyl-(1 → 2)-]}-β-*D*-glucopyranosylhomo-aro-cholest-5-ene-26-*O*-β-*D*-glucopyranoside* = *4-[(2*S*,4a*R*,4b*S*,6a*S*,11a*S*,11b*R*)-2-{[6-Deoxy-α-*L*-mannopyranosyl-(1 → 2)-[6-deoxy-α-*L*-mannopyranosyl-(1 → 4)-6-deoxy-α-*L*-mannopyranosyl-(1 → 4)]-β-*D*-glucopyranosyl]oxy}-2,3,4,4a,4b,5,6,6a,11,11a,11b,12-dodecahydro-4a,6a,7-trimethyl-1*H*-indeno[2,1-*a*]phenanthren-8-yl]-2-methylbutyl β-*D*-Glucopyranoside*; **1**). White amorphous powder. $[\alpha]_D^{22} = -7.33$ ($c = 0.15$, MeOH). UV (MeOH): 268 (4.04), 275 (3.95). IR (KBr): 3419, 2933, 1640, 1568, 1451, 1381, 1044, 814. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. HR-ESI-MS: 1207.5864 ($[M + Na]^+$, $C_{59}H_{92}NaO_{24}^+$; calc. 1207.5877).

Parispseudoside B (= *3β-*O*-α-*L*-Rhamnopyranosyl-(1 → 2)-β-*D*-glucopyranosylhomo-aro-cholest-5-ene-26-*O*-β-*D*-glucopyranoside* = *4-[(2*S*,4a*R*,4b*S*,6a*S*,11a*S*,11b*R*)-2-{[2-*O*-(6-Deoxy-α-*L*-mannopyranosyl)-β-*D*-glucopyranosyl]oxy}-2,3,4,4a,4b,5,6,6a,11,11a,11b,12-dodecahydro-4a,6a,7-trimethyl-1*H*-indeno[2,1-*a*]phenanthren-8-yl]-2-methylbutyl β-*D*-Glucopyranoside*; **2**). White amorphous powder. $[\alpha]_D^{22} = -5.20$ ($c = 0.15$, MeOH). UV (MeOH): 268 (4.09), 278 (4.06). IR (MeOH): 3386, 2928, 1637, 1456, 1377, 1047, 814. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. HR-ESI-MS: 915.4708 ($[M + Na]^+$, $C_{47}H_{72}NaO_{16}^+$; calc. 915.4718).

Parispseudoside C (= *(25*R*)-3β-*O*-{α-*L*-Rhamnopyranosyl-(1 → 4)-α-*L*-rhamnopyranosyl-(1 → 4)-[α-*L*-rhamnopyranosyl-(1 → 2)]}-β-*D*-glucopyranosyl-cholesta-5,17(20)-diene-16,22-dione-26-*O*-β-*D*-glucopyranoside* = *(3β,17*Z*,25*R*)-3-{[6-Deoxy-α-*L*-mannopyranosyl-(1 → 2)-[6-deoxy-α-*L*-mannopyranosyl-(1 → 4)-6-deoxy-α-*L*-mannopyranosyl-(1 → 4)]-β-*D*-glucopyranosyl]oxy}-16,22-dioxocholesta-5,17-dien-27-yl β-*D*-Glucopyranoside*; **3**). White amorphous powder. $[\alpha]_D^{22} = -11.0$ ($c = 0.15$, MeOH). UV (MeOH): 223 (4.28), 245 (4.50). IR (KBr): 3416, 2933, 1710, 1632, 1382, 1130, 1043. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. HR-ESI-MS: 1213.5601 ($[M + Na]^+$, $C_{57}H_{90}NaO_{26}^+$; calc. 1213.5618).

Acidic Hydrolysis of **1**–**3**. A soln. of saponins **1**–**3** (1 mg each) in H_2O (1 ml) was treated with 2N aq. CF_3COOH (2 ml) and heated at 90° for 3 h in a sealed tube, resp. After extraction with CH_2Cl_2 (3 × 2 ml), the aq. layer was repeatedly concentrated with MeOH until neutral, and then analyzed by TLC (SiO_2, BuOH/EtOH/H_2O 4:1:2) with authentic samples: glucose (R_f 0.47), and rhamnose (R_f 0.68) for **1**, **2**, and **3**.

This study was supported by the *Self-Study Foundation of State Key Laboratory of Oral Diseases* (Sichuan University). The authors thank the *Analytical and Testing Center of Sichuan University* for the spectral measurements.

REFERENCES

[1] H. Li, 'The Genus Paris (Trilliaceae)', Science Press, Beijing, 1998, p. 193.
[2] H. Li, 'The Genus Paris (Trilliaceae)', Science Press, Beijing, 1998, p. 182.
[3] S. S. Wu, W. Y. Gao, H. Q. Duan, W. Jia, *Chin. Tradit. Herb. Drugs* **2004**, *35*, 344.
[4] H. Liu, Y. Huang, Q. Wang, T. Zhang, Y. Song, *Planta Med.* **2006**, *72*, 835.
[5] Y. Zhao, L.-P. Kang, Y.-X. Liu, Y. Zhao, C.-Q. Xiong, B.-P. Ma, F.-T. Dong, *Magn. Reson. Chem.* **2007**, *45*, 739.
[6] J. Zhang, B.-P. Ma, L.-P. Kang, H.-S. Yu, Y. Yang, X.-Z. Yan, *Chin. J. Magn. Reson.* **2006**, *23*, 31.
[7] Y. Huang, Q. Wang, W. C. Ye, L.-J. Cui, *Chin. J. Nat. Med.* **2005**, *3*, 138.
[8] C. Tagawa, M. Okawa, T. Ikeda, T. Yoshida, T. Nohara, *Tetrahedron Lett.* **2003**, *44*, 4839.

[9] H.-F. Tang, Y.-P. Zhao, Y.-P. Jiang, *Chin. Tradit. Herb. Drugs* **1998**, *29*, 839.
[10] P. K. Agrawal, *Steroids* **2005**, *70*, 715.
[11] T. Nohara, Y. Ogata, M. Aritome, K. Miyahara, T. Kawasaki, *Chem. Pharm. Bull.* **1975**, *23*, 925.

Received November 25, 2008