云 南 植 物 研 究 1995; 17 (4): 468—472
Acta Botanica Yunnanica

法国产夏枯草中的两个新的乌索烷型三萜皂甙

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摘要 从法国产夏枯草（*Prunella vulgaris* L.）的甲醇提取物中分离得到两个新的乌索烷型三萜皂甙：夏枯草甙（pruvuloside）A 和 B；同时还分离到 5 个已知化合物：niga-ichigoside F_2, sericoside, 槲皮素（quercetin），槲皮素-3-O-葡萄糖甙（quercetin 3-O-glucoside），山柰酚-3-O-葡萄糖甙（kaempferol-3-O-glucoside），及 arjunglucoside Ⅰ和 niga-ichigoside F_1 的混合物。它们的结构是通过波谱的方法证明的。

关键词 唇形科，夏枯草，三萜皂甙，夏枯草甙 A，B

TWO NEW URSANE GLYCOSIDES FROM PRUNELLA VULGARIS IN FRANCE

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Abstract Two new ursane glucosides, pruvuloside A and B, together with five known compounds, niga-ichigoside F_2, sericoside, quercetin, quercetin-3-glucoside, kaempferol-3-O-glucoside, and a mixture of niga-ichigoside F_1 and arjunglucoside Ⅰ were isolated from the MeOH extract of *Prunella vulgaris* collected in France. Their structures were determined by means of spectral evidence.

Key words Labiatae, *Prunella vulgaris*, Triterpenoid saponins, Pruvuloside A,B.

Prunella vulgaris L. (Labiatae) as a perennial herb is widely distributed in the temperate zone and tropical mountain area of the whole world. As a wide-disperse species, this plant has been divided as several subspecies and varieties in different places according to their different physiological and ecological characters. It has been used as traditional Chinese medicine for treatment of tuberculosis, pleuritis, bacterial dysentery, icterohepatitis [1]. In Japan only its spike in the flowering period is used as diuretic, and in Europe the whole herb has been used as the same use [2]. Up to now, there were a lot of chemical studies on this plant, and some sterols, β-sitosterol, stigmasterol, stigmast-7-en-3β-ol, α-spinasterol [2—3]; triterpenoids, ursolic acid, oleanolic acid, 2α, 3α, 24-trihydroxyolean-12-en-28-oic acid, methyl 2α, 3α, 23-trihydroxyolean-12-en-28-oate [2], methyl (13S, 14R)-2α, 3α, 24-trihydroxyolean-11-en-28-oate [4], 2α, 3α, 24-trihydroxyolean-11, 13(18)-dien-28-oic acid [5], et al., were isolated. This paper deal with the isolation and structure elucidation of two new ursane glucosides, pruvuloside A **(1)**

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1995-05-03 收稿，1995-06-26 修回

and B **(2)**, together with two known saponins, niga-ichigoside F_2**(3)**, sericoside **(4)**, three known flavonoids, quercetin **(5)**, quercetin-3-O-glucoside **(6)**, kaempferol-3-O-glucoside **(7)** and a mixture **(8)** of niga-ichigoside F_1**(8a)** and arjunglucoside Ⅰ **(8b)** from this plant.

RESULTS AND DISCUSSION

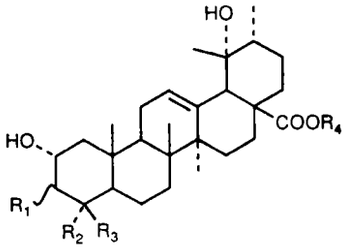
MeOH extract of *P. vulgaris* was suspended in H_2O and then successively extracted with Et_2O, AcOEt and n-Butanol. The n-Butanol extract was repeatedly chromatographed on Sephadex, Rp-8 and silica gel column to give six compounds, 1—7 and a mixture **(8)**. By means of NMR, compounds **3**—**4** were identified as known β-glucosyl ester of A-ring oxygenated 19α-hydroxyursolic and oleanolic acid, niga-ichigoside F_2**(3)** [6] and sericoside **(4)** [7]; compounds **5**—**7** were identified as the known flavonoids, quercetin **(5)**, quercetin-3-O-glucoside **(6)**, kaempferol-3-O-glucoside **(7)** [8], and the mixture **(8)** was determined as niga-ichigoside F **(8a)** [6] and arjunglucoside Ⅰ **(8b)** [9].

Compound 1 showed a quasi molecular ion peak at m / z 811 $[M\ (C_{42}H_{68}O_{15})-H]^-$ and typical fragment ion at m / z 649 $[M-glc]^-$, 487$[M-glc-glc]^-$ in the negative FAB mass spectrum. The ^1H and ^{13}C NMR signals of **1** due to sugar moiety, indicated the presence of two anomeric signals. The ^{13}C signals of **1** in pyridine-d_5[δ177.0 (CO_2-βGlc) and 93.8 (anomeric C of βGlc)], indicated the location of a β-glucopyranosyl group of **1** on COOH group of C-28. Comparison of the ^{13}C NMR data of **1** with that of kaji-ichigoside F_1 [6], showed that their carbon signals for the aglycone moiety were almost superimposable except for sugar moiety. **1** has one more glucose than kaji-ichigoside F_1, and from **1** to kaji-ichigoside F_1, the carbon signals of glucose are shifted by +2.2 (C-1′), -5.6 (C-2′). So, the terminal glucose of **1** was attached at C-2 position of the inner glucose which linked at COOH group of C-28, and this sugar chain were also supported by the comparion of the ^{13}C NMR data with that of β-sophorosyl ester of hederagenin [10]. Thus, the structure of this new triterpenoidal saponin was established as 2α, 3α, 19α-trihydroxyurs-12-en-28-oic acid-28-O-β-D-glucopyranosyl-(1→2)-β-D-glucopyranoside, and named as pruvuloside A.

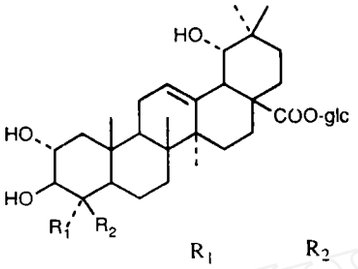
Compound 2 showed a quasi molecular ion peak at m / z 665 $[M(C_{36}H_{58}O_{11})-H]^-$ and typical fragment ion at m / z 503 $[M-glc]^-$ in the negative FAB mass spectrum. The ^1H and ^{13}C NMR signals of **2** due to sugar moiety, indicated the presence of one anomeric signals. The ^{13}C signals of **2** in pyridine-d_5[δ177.0 (CO_2-βGlc) and 95.9 (anomeric C of βGlc)] indicated the location of a β-glucopyranosyl group of **2** on COOH group of C-28. Comparison of the ^{13}C NMR data of **2** with that of niga-ichigoside F_2 [6], showed that all of their carbon signals were almost superimposable except for the signals of C-23 and C-24. Usually, the chemical shift of the C-23 methyl group is at about δ 24.4 if the hydroxyl group is linked at C-24 (δ 65.7) [9] and the chemical shift of C-24 methyl group is at about δ 16.7 if the hydroxyl group is linked at C-23 (δ 71.3) [6]. The chemical shifts of C-23 (δ23.7) and C-24 (δ65.3) in cmopound **2** suggested that the hydroxyl group was linked at C-24. Therefore, the structure of **2** was established as 2α, 3α, 19α, 24-tetrahydroxyurs-12-en-28-oic acid-28-O-β-D-glucopyranoside. This new triterpenoidal saponin named as pruvuloside B.

This is the first report for the isolation of triterpenoid saponins from *P. vulgaris*. Among these compounds we obtained, **6** is the main compound in this plant, and all of the saponins have a hydroxy group at C-19 position and sugar moiety linked at the ester of C-28 of the triterpenoid skeleton, which is different

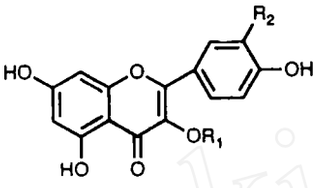
with the previous reports about this plant.



	R_1	R_2	R_3	R_4
1	αOH	CH_3	CH_3	-glc^{2}—glc
2	αOH	CH_3	CH_2OH	glc
3	αOH	CH_2OH	CH_3	glc
8a	βOH	CH_2OH	CH_3	glc



	R_1	R_2
4	CH_3	CH_2OH
8b	CH_2OH	CH_3



	R_1	R_2
5	H	OH
6	glc	OH
7	glc	H

EXPERIMENT

$_1$H and ^{13}C NMR spectra were measured on a Brucker spectrospin AM-400 spectrometer using TMS as internal standards. FAB-MS spectra were measured with VG Autospec mass spectrometer.

Extraction and isolation MeOH extract of *Prunella vulgaris* (46.69 g) was suspended in H_2O and then successively extracted with Et_2O, AcOEt and n-Butanol. The n-Butanol layer was concentrated in vacuo to give a viscous residue (5.09 g), which was repeatedly chromatographed on Sephadex (eluting with 80% MeOH), Rp-8 (eluting with 50% MeOH) and silica gel column (eluting with $CHCl_3$-MeOH-H_2O, 60 : 10 : 1) to give **1** (6 mg), **2** (5 mg), **3** (11 mg), **4** (10 mg), **5** (15 mg), **6** (75 mg), **7** (22 mg) and a mixture **(8)** (25 mg).

Pruvuloside A (1). a white powder. $[\alpha]_D^{24}$-2.6° (MeOH, c 0.39); FAB-MS: m/z 811 [M($C_{42}H_{68}O_{15}$)-H]$^-$, 649 [M-glc]$^-$, 487 [M-glc-glc]$^-$; ^{1}H NMR (C_5D_5N): δ2.70 (1H, s, H-18), 1.61, 1.34, 1.18, 1.11, 0.95, 0.82 (each 3H, s, H-23, 24, 25, 26, 27, 29 respectively), 1.05 (3H, d, J=6.5 Hz, H-30), 5.70 (1H, br s, H-12), 6.22 (1H, d, J=8.2 Hz, inner H_{glu}-1′), 5.70 (1H, d, J=7.8 Hz, terminal H_{glu}-1″); ^{13}C NMR (C_5D_5N): δ43.0 (C-1), 66.2 (C-2), 79.1 (C-3), 38.7 (C-4), 48.7 (C-5), 18.7 (C-6), 33.7 (C-7), 40.9 (C-8), 47.8 (C-9), 38.8 (C-10), 24.2 (C-11), 128.4 (C-12), 139.5(C-13), 42.2 (C-14), 29.8 (C-15), 26.0 (C-16), 48.9 (C-17), 54.6 (C-18), 72.8 (C-19), 42.2 (C-20), 26.8 (C-21), 37.9 (C-22), 29.4 (C-23), 22.3 (C-24), 16.9 (C-25), 17.5 (C-26), 24.7 (C-27), 177.0 (C-28), 26.1 (C-29), 16.8 (C-30), inner glucose: 93.8 (C-1′), 79.4 (C-2′), 78.1 (C-3′), 71.0 (C-4′), 79.1 (C-5′), 62.4 (C-6′), terminal glucose: 104.9 (C-1″), 76.0 (C-2″), 78.1 (C-3″), 72.9 (C-4″), 78.4 (C-5″), 63,9 (C-6″).

Pruvuloside B (2). A white powder. $[\alpha]_D^{24}$-5.1° (MeOH, c 0.20); FAB-MS: m/z 665 [M

($C_{36}H_{58}O_{11}$)−H] , 503 [M−glc] ; ^{1}H NMR (C_5D_5N): δ2.90 (1H, s, H−18), 1.65, 1.61, 1.32, 1.18, 0.92 (each 3H, s, H−23, 25, 26, 27, 29 respecively), 1.05 (3H, d, J = 6.6 Hz, H−30), 6.27 (1H, d, J = 8.0 Hz, H_{glu}−1′); ^{13}C NMR (C_5D_5N): δ43.3 (C−1), 66.3 (C−2), 79.0 (C−3), 42.2 (C−4), 44.2 (C−5), 19.2 (C−6), 33.3 (C−7), 40.9 (C−8), 48.0 (C−9), 38.7 (C−10), 24.5 (C−11), 128.5 (C−12), 139.2 (C−13), 40.9 (C−14), 29.9 (C−15), 26.8 (C−16), 48.7 (C−17), 54.5 (C−18), 72.8 (C−19), 42.2 (C−20), 27.1 (C−21), 37.7 (C−22), 23.7 (C−23), 65.3 (C−24), 17.7 (C−25), 17.2 (C−26), 24.5 (C−27), 177.0 (C−28), 27.1 (C−29), 16.7 (C−30), 95.9 (C_{glu}−1′], 74.1 (C−2′), 79.0 (C−3′), 71.5 (C−4′), 79.2 (C−5′), 62.6 (C−6′).

Niga−ichigoside F_2(3). a white powder. $[\alpha]_D^{24}$+4.0 ° (MeOH, c 0.75); FAB−MS: m / z 665 [M($C_{36}H_{58}O_{11}$)−H] , 503 [M−glc] ; ^{1}H NMR (C_5D_5N): δ3.72, 3.89 (each 1H, d, J = 10.8 Hz, H−23), 2.70 (1H, s, H−18), 1.62, 1.42, 1.32, 1.22, 0.85 (each 3H, s, H−24, 25, 26, 27, 29 respectively), 1.04 (3H, d, J = 8.2 Hz, H−30), 6.30 (1H, d, J = 8.1 Hz, H_{glu}−1′); ^{13}C NMR (C_5D_5N): δ42.9 (C−1), 66.3 (C−2), 79.0 (C−3), 42.3 (C−4), 43.6 (C−5), 18.6 (C−6), 33.3 (C−7), 40.8 (C−8), 47.8 (C−9), 38.5 (C−10), 24.2 (C−11), 128.5 (C−12), 139.4 (C−13), 42.0 (C−14), 29.2 (C−15), 26.2 (C−16), 48.7 (C−17), 54.5 (C−18), 72.7 (C−19), 42.2 (C−20), 26.8 (C−21), 37.8 (C−22), 71.3 (C−23), 16.7 (C−24), 17.8 (C−25), 17.2 (C−26), 24.6 (C−27), 177.0 (C−28), 27.1 (C−29), 16.7 (C−30), 95.9 (C_{glu}−1′), 74.1 (C−2′), 79.0 (C−3′), 71.3 (C−4′), 79.3 (C−5′), 62.5 (C−6′).

Sericoside (4). a white powder $[\alpha]_D^{24}$+4.3 ° (MeOH, c 0.24); FAB−MS: m / z 665 [M($C_{36}H_{58}O_{11}$−H]$^-$, 503 [M−glc]$^-$; ^{1}H NMR (C_5D_5N): δ3.55 (1H, br s, H−18), 1.56, 1.42, 1.24, 1.13, 0.96, 0.89, (each 3H, s, H−23, 25, 26, 27, 29, 30), 6.37 (1H, d, J = 8.0 Hz, H_{glu}−1′); ^{13}C NMR (C_5D_5N): δ47.6 (C−1), 68.8 (C−2), 85.9 (C−3), 44.0 (C−4), 56.7 (C−5), 19.6 (C−6), 33.7 (C−7), 40.4 (C−8), 48.6 (C−9), 38.6 (C−10), 24.6 (C−11), 123.3 (C−12), 144.4 (C−13), 42.2 (C−14), 29.1 (C−15), 28.1 (C−16), 46.6 (C−17), 44.8 (C−18), 81.2 (C−19), 35.6 (C−20), 28.7 (C−21), 33.1 (C−22), 24.2 (C−23), 65.7 (C−24), 17.6 (C−25), 17.3 (C−26), 24.8 (C−27), 177.3 (C−28), 28.7 (C−29), 24.6 (C−30), 95.9 (C_{glu}−1′), 74.2 (C−2′), 79.0 (C−3′), 71.3 (C−4′), 79.2 (C−5′), 62.4 (C−6′).

Quercetin (5). a yellow powder. FAB−MS: m / z 301 [M($C_{15}H_{10}O_7$)−H] ; ^{1}H NMR (DMSO−d_6); δ7.66 (1H, s, H−2′), 7.53 (1H, d, J = 8.4 Hz, H−5′), 6.87 (1H, d, J = 8.4 Hz, H−6′), 6.45 (1H, s, H−6), 6.17 (1H, s, H−8); ^{13}C NMR (DMSO−d_6): 146.8 (C−2), 135.7 (C−3), 175.8 (C−4), 156.1 (C−5), 98.2 (C−6), 164.0 (C−7), 93.4 (C−8), 160.7 (C−9), 102.9 (C−10), 121.9 (C−1′), 115.6 (C−2′), 145.0 (C−3′), 147.7 (C−4′), 115.0 (C−5′), 119.9 (C−6′).

Quercetin−3−O−glucoside (6). A yellow powder. FAB−MS: m / z 463 [M($C_{21}H_{20}O_{12}$)−H] , 300 [M−glc−H]$^-$; ^{1}H NMR (CD_3OD+DMSO−d_6): δ7.72 (1H, s, H−2′), 7.59 (1H, d, J = 8.4 Hz, H−5′), 6.88 (1H, d, J = 8.4 Hz, H−6′), 6.38 (1H, s, H−6), 6.18 (1H, s, H−8), 5.34 (1H, d, J = 7.3 Hz, H_{glu}−1″); ^{13}C NMR (CD_3OD+DMSO−d_6): 158.3 (C−2), 135.6 (C−3), 179.3 (C−4), 162.9 (C−5), 99.9 (C−6), 165.8 (C−7), 94.8 (C−8), 158.8 (C−9), 105.7 (C−10), 123.1 (C−1′), 116.1 (C−2′), 145.9 (C−3′), 149.8 (C−4′), 117.7 (C−5′), 123.1 (C−6′), 104.2 (C−1″), 75.7 (C−2″), 78.1 (C−3″), 71.3(C−4″), 78.4 (C−5″), 62.6 (C−6″).

Kaempferol 3−O−glucoside (7). A yellow powder. FAB−MS: m / z 447 [M($C_{21}H_{20}O_{11}$)−H]$^-$; ^{1}H NMR (DMSO−d_6); δ8.02 (2H, d, J = 8.8 Hz, H−2′ ,6′), 6.87 (2H, d, J = 8.3 Hz, H−3′ 5′), 6.41 (1H, s, H−8), 6.18 (1H, s, H−6), 5.43 (1H, d, J = 7.4 Hz, H_{glu}−1″); ^{13}C NMR (DMSO−d_6): 156.5 (C−2), 133.3 (C−3), 177.4 (C−4), 161.2 (C−5), 98.9 (C−6), 164.8 (C−7), 93.7 (C−8), 156.3 (C−9), 103.8 (C−10), 120.9 (C−1′), 130.8 (C−2′), 115.1 (C−3′), 160.0 (C−4′), 115.1 (C−5′), 130.8 (C−6′), 101.1 (C−1″), 74.3 (C−2″),

76.5 (C-3″), 70.0 (C-4″), 77.4 (C-5″), 60.9 (C-6″).

Mixture (8): a white powder.

Niga-ichigoside F_1(8a). FAB-MS: m / z 665 $[M(C_{36}H_{58}O_{11})-H]^-$, 503 $[M-glc]^-$; ^1H NMR (C_5D_5N): δ2.71 (1H, s, H-18), 6.30 (1H, d, J = 8.1 Hz, H_{glu}-1′); ^{13}C NMR (C_5D_5N): δ48.0 (C-1), 69.0 (C-2), 78.6 (C-3), 44.7 (C-4), 48.5 (C-5), 18.8 (C-6), 33.3 (C-7), 40.8 (C-8), 48.2 (C-9), 38.5 (C-10), 24.3 (C-11), 128.5 (C-12), 139.3 (C-13), 42.3 (C-14), 29.3 (C-15), 26.2 (C-16), 48.7 (C-17), 54.5 (C-18), 72.8 (C-19), 42.2 (C-20), 26.8 (C-21), 37.7 (C-22), 67.0 (C-23), 14.3 (C-24), 17.6 (C-25), 17.5 (C-26), 24.6 (C-27), 177.0 (C-28), 27.1 (C-29), 16.7 (C-30), 95.9 (C_{glu}-1′), 74.1 (C-2′), 79.0 (C-3′), 71.3 (C-4′), 79.2 (C-5′), 62.4 (C-6′).

Arjunglucosde Ⅰ (8b). FAB-MS: m / s 665 $[M(C_{36}H_{58}O_{11})-H]^-$, 503 $[M_{glc}]^-$; ^1H NMR (C_5D_5N): δ2.90 (1H, br s, H-18), 6.38 (1H, d, J = 8.1 Hz, H_{glu}-1′); ^{13}C NMR (C_5D_5N): δ48.0 (C-1), 69.0 (C-2), 78.6 (C-3), 44.7 (C-4), 48.5 (C-5), 18.8 (C-6), 33.3 (C-7), 40.4 (C-8), 48.2 (C-9), 38.7 (C-10), 24.3 (C-11), 123.8 (C-12), 144.4 (C-13), 42.3 (C-14), 29.3 (C-15), 29.1 (C-16), 46.6 (C-17), 43.6 (C-18), 81.2 (C-19), 35.6 (C-20), 29.1 (C-21), 33.1 (C-22), 67.0 (C-23), 14.2 (C-24), 17.8 (C-25), 17.4 (C-26), 24.9 (C-27), 177.3 (C-28), 28.7 (C-29), 24.8 (C-30), 95.9 (C_{glu}-1′), 74.2 (C-2′), 79.0 (C-3′), 71.5 (C-4′), 79. 2 (C-5′), 62.6 (C-6′).

Acknowlegement We are grateful to Dr. A. Meybeck (LVMH Research Institute, France) for providing the MeOH extract of *Prunella vulgaris*, Dr. Bing-Jun Li of our Institute for measuring the $[\alpha]_D$ and the instrument group of our Laboratory for measuring NMR and MS spectra.

References

〔1〕 Jiangsu New Medical College. The Dictionary of Traditional Chinese Medicine. Shanghai: Shanghai People press, 1977. 1827.

〔2〕 Kojima H, Ogura H. Triterpenoids from *Prunella vulgaris*. *Phytochemistry*, 1986, **25**(3): 729—733.

〔3〕 Kojima H, Sato N, Hatano A, *et al*. Sterol glucosides from *Prunella vulgaris*. *Phytochemistry*, 1990, **29**(7): 2351.

〔4〕 Kojima H, Tominaga H, Sato S, *et al*. Two novel hexacyclic triterpenoids from *Prunella vulgaris*. *Phytochemistry*, 1988, **27**(9): 2921—2925.

〔5〕 Kojima H, Tominaga H, Sato S, *et al*. Pentacyclic triterpenoids from *prunella vulgaris*. *Phytochemistry*, 1987, **26**(4): 1107—1111.

〔6〕 Seto T, Tanaka T, Tanaka O. *et al*. β-glucosyl esters of 19α-hydroxyursolic acid derivatives in leaves of *Rubus* species. *Phytochemistry*, 1984, **23**(12): 2829—2934.

〔7〕 Bombardelli E, Bonati A, Gabetta B, *et al*. Triterpenoids of *Terminalia sericea*. *Phytochemistry*, 1974, **13**: 2259.

〔8〕 Markham K R, Ternal B, Stanley R, *et al*. Carbon-13 NMR studies of flavonoids III. Naturally occurring flavonoid glycosides and their acylated derivatives. *Tetrahedron*, 1978, **34**: 1389—1397.

〔9〕 Zhou X H, Kasai R, Ohtai K, *et al*. Oleanane and ursane glucosides from *Rubus* species. *Phytochemistry*, 1992, **31**(10): 3642—3644.

〔10〕 Kimata H, Nakashima T, Kokubun S, *et al*. Saponins of pericarps of *Sapindus mukurossi* Gaerth. and solbiulization of monodesmosides by bisdesmosides. *Chem Pharm Bull*, 1983, **31**: 1998—2005.