Medicinal Foodstuffs. XXIII.[1] Structures of New Oleanane-Type Triterpene Oligoglycosides, Basellasaponins A, B, C, and D, from the Fresh Aerial Parts of *Basella rubra* L.

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Received January 22, 2001; accepted February 22, 2001

Basellasaponins A, B, C, and D, oleanane-type triterpene oligoglycosides having the dioxolane-type substituent, were isolated from the fresh aerial parts of *Basella rubra* L. together with betavulgaroside I, spinacoside C, and momordins IIb and IIc. The chemical structures of basellasaponins A, B, C, and D were determined from chemical and physicochemical evidence.

Key words basellasaponin; *Basella rubra*; medicinal foodstuff; spinacoside C; dioxolane-type substituent; Basellaceae

The Basellaceae plant, *Basella* (*B.*) *rubra* L. (Indian spinach, Japanese name "Tsurumurasaki") is extensively cultivated as an ornamental and the aerial parts such as the leaves, stems, and young shoot with buds are consumed as a vegetable and health food. In Chinese traditional medicine, the leaves or the aerial parts of *B. rubra* (Chinese name "落葵") have been used for the treatment of constipation and also as a diuretic, a toxicide, and an anti-inflammatory. As chemical constituents of this plants, betacyanins, carotenoids, and organic acids have been isolated from *B. rubra.*[2]

During the course of our studies on bioactive saponin constituents in medicinal foodstuffs,[1,3] we have characterized betavulgarosides with a novel dioxolane-type or acetal-type substituent. Both of these substituents were presumed to be biosynthesized through an oxidative degradation process of terminal monosaccharide moiety in saponin, from the roots and leaves of *Beta vulgaris* L. (sugar beet), and we have reported their hypoglycemic activity.[4] As a continuing study, we have isolated new saponins named basellasaponins A (**1**), B (**2**), C (**3**), and D (**4**), having a dioxolane-type substituent, along with betavulgaroside I (**5**),[4] spinacoside C (**6**),[5] momordins IIb (**7**)[6] and IIc (**8**)[6] from the aerial parts of *B. rubra*. In this paper, we present the isolation and structure elucidation of basellasaponins A (**1**), B (**2**), C (**3**), and D (**4**) from the aerial parts of *B. rubra*.

The methanolic extract from the fresh aerial parts of *B. rubra* cultivated in Kyoto Prefecture was subjected to Diaion HP-20 ($H_2O \rightarrow MeOH \rightarrow CHCl_3$) and silica gel ($CHCl_3$–$MeOH$–$H_2O$) column chromatography and finally HPLC [YMC-Pack ODS-A, CH_3CN–1% aqueous acetic acid (AcOH), CH_3CN–1% aqueous trifluoroacetic acid (TFA)] to give basellasaponins A (**1**, 0.0003%), B (**2**, 0.0002%), C (**3**, 0.0004%), and D (**4**, 0.0001%) together with betavulgaroside I[4] (**5**, 0.0004%), spinacoside C[5] (**6**, 0.0007%), and momordins IIb[6] (**7**, 0.0001%) and IIc[6] (**8**, 0.0004%).

Basellasaponin A (**1**) was obtained as colorless fine crystals from aqueous methanol. The IR spectrum of **1** showed an absorption band at 1736 cm^{-1} ascribable to carboxyl function and strong absorption bands at 3403, 1076, and 1036 cm^{-1}, suggestive of an oligoglycosidic structure. In the negative- and positive-ion FAB-MS of **1**, quasimolecular ion peaks were observed at *m/z* 969 $(M-H)^-$ and *m/z* 993 $(M+Na)^+$, whose elemental composition was determined to be $C_{47}H_{70}O_{21}$ by high-resolution MS measurement. Fragment ion peaks were observed at *m/z* 809 $(M-C_5H_5O_6)^-$ and *m/z* 807 $(M-C_6H_{11}O_5)^-$ in the negative-ion FAB-MS of **1**. Acid

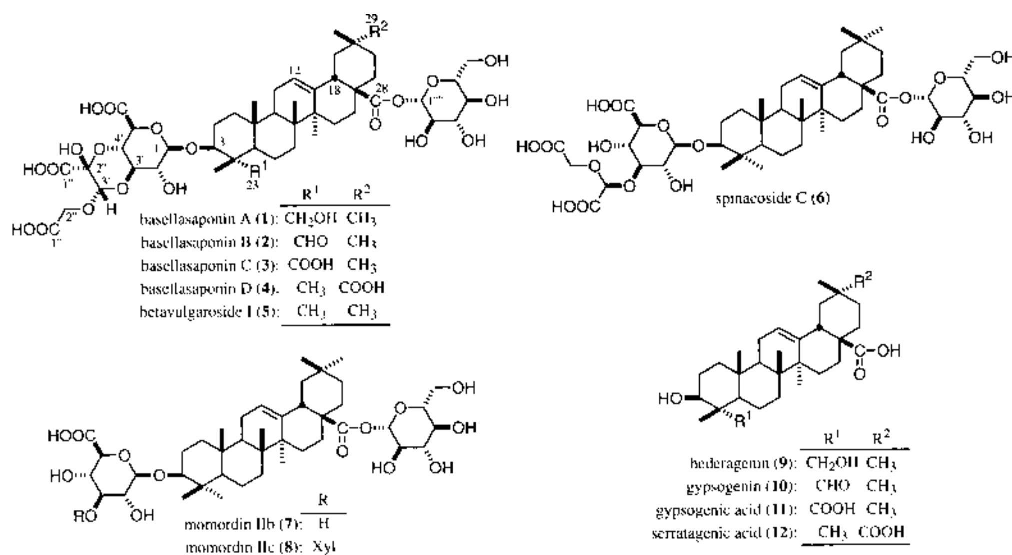

Chart 1

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hydrolysis of **1** with 5% aqueous sulfuric acid (H_2SO_4)–1,4-dioxane (1 : 1, v/v) liberated hederagenin (**9**),[4b,7)] D-glucose, and D-glucuronic acid. These component monosaccharides were identified by GLC analysis of their trimethylsilyl thiazolidine derivatives.[8)] The ^{1}H-NMR (pyridine-d_5) and ^{13}C-NMR (Table 1) spectra of **1**, which were assigned by various NMR experiments,[9)] showed the presence of a glucopyranosiduronic acid moiety having the dioxolane-type substituent composed of 3-oxopyruvic acid and glycolic acid [δ 4.75 (s, 2‴-H_2), 5.17 (d, J=7.6 Hz, 1′-H), 5.91 (s, 3″-H)], together with a hederagenin moiety [δ 4.22 (m, 3-H), 5.40, (dd-like, 12-H)] and a 28-*O*-β-D-glucopyranoside moiety [δ 6.27 (d, J=7.6 Hz, 1⁗-H)]. The planar structure of the dioxolane part at the 3′- and 4′-positions of the 3-*O*-glucopyranosiduronic acid moiety in **1** was confirmed by the HMBC experiment. Namely, long-range correlations were observed between the following protons and carbons of **1**: 1′-H and 3-C; 3′-H and 3″-C; 3″-H and 3′-, 2‴-C; 4′-H and 2″-C; 2‴-H_2 and 3″-, 1‴-C (Fig. 1). The carbon signals in the ^{13}C-NMR (Table 1) spectrum of **1** were very similar to those of betavulgaroside I (**5**), except for the signals due to the 23-position of the aglycone moiety in **1**. This evidence allowed us to presume that the stereostructure of the dioxolane substituent in **1** was the same as **5**. Consequently, the structure of basellasaponin A (**1**) was determined as shown.

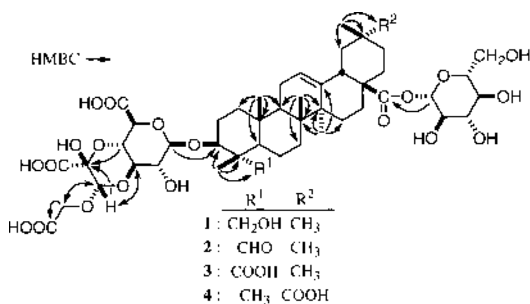

Fig. 1

Basellasaponin B (**2**), also isolated as colorless fine crystals of mp 226—228 °C from aqueous methanol, liberated gypsogenin (**10**),[10)] D-glucose, and D-glucuronic acid on acid hydrolysis with 5% aqueous H_2SO_4–1,4-dioxane.[8)] The molecular formula $C_{47}H_{68}O_{21}$ of **2** was clarified from quasimolecular ion peaks [*m/z* 967 $(M-H)^-$ and *m/z* 991 $(M+Na)^+$] and by high-resolution MS analysis observed in the negative- and positive-ion FAB-MS. Furthermore, fragment ion peaks were observed at *m/z* 807 $(M-C_5H_5O_6)^-$ and *m/z* 805 $(M-C_6H_{11}O_5)^-$ in the negative-ion FAB-MS of **2**. The ^{1}H-NMR (pyridine-d_5) and ^{13}C-NMR (Table 1) spectra[9)] of **2** showed the presence of a β-D-glucopyranosiduronic acid moiety having the dioxolane-type substituent [δ 4.79 (s, 2‴-H_2), 4.88 (d, J=7.6 Hz, 1′-H), 5.96 (s, 3″-H)], a β-D-glucopyranosyl moiety [δ 6.30 (J=8.0 Hz, 1⁗-H)], and a gypsogenin moiety [δ 3.18 (dd-like, 18-H), 4.10 (m, 3-H), 5.40 (dd-like, 12-H)]. The glycosidic structure of **2** was characterized by the HMBC experiment, in which long-range correlations were observed between the following protons and carbons: 1′-H and 3-C; 3′-H and 3″-C; 3″-H and 3′-, 2‴-C; 4′-H and 2″-C; 2‴-H_2 and 3″-, 1‴-C; 1⁗-H and 28-C. The carbon signals in the ^{13}C-NMR data of **2** closely resembled those of **1** and **5**, except for the signals due to the 23-position of the aglycone moiety in **2**. On the basis of this evidence, the structure of basellasaponin B (**2**) was characterized as shown.

Basellasaponins C (**3**) and D (**4**) were obtained as colorless fine crystals of mp 230—232 °C and mp 215—217 °C from aqueous methanol, respectively. Saponins **3** and **4** were found to have the same molecular formula, $C_{47}H_{68}O_{22}$, which was determined from the quasimolecular ion peaks [*m/z* 983 $(M-H)^-$, *m/z* 1007 $(M+Na)^+$] in the negative- and positive-ion FAB-MS and by high-resolution MS measurement. Acid

Table 1. ^{13}C-NMR Data for Basellasaponins A (**1**), B (**2**), C (**3**), and D (**4**) and Betavulgaroside I (**5**)

	1[a)]	2[a)]	3[a)]	4[b)]	5[a)]		1[a)]	2[a)]	3[a)]	4[b)]	5[a)]
C-1	38.6	38.0	38.6	38.5	38.4	25	16.1	15.6	15.9	17.4	15.3
2	25.9	25.1	26.1	26.5	26.4	26	17.6	17.4	17.3	17.4	17.3
3	82.4	82.6	85.8	89.3	89.2	27	26.1	26.1	26.1	26.1	26.0
4	43.4	55.4	53.2	39.5	39.3	28	176.4	176.4	176.4	176.2	176.3
5	47.6	47.6	52.1	55.6	55.5	29	33.1	33.1	33.1	180.8	33.0
6	18.2	20.4	21.2	18.5	18.3	30	23.7	23.6	23.6	19.9	23.5
7	32.9	32.5	32.8	33.0	33.0	C-1′	106.5	105.7	106.5	107.5	107.4
8	40.0	40.2	40.2	39.9	39.7	2′	72.0	71.6	71.8	72.1	71.9
9	48.1	47.9	48.0	47.9	47.8	3′	72.6	72.5	72.3	72.6	72.4
10	36.9	36.0	36.6	36.9	36.7	4′	70.0	70.1	70.1	70.2	70.0
11	23.5	23.5	23.3	23.5	23.4	5′	75.2	75.2	75.2	75.2	75.1
12	122.7	122.7	122.8	123.1	122.7	6′	171.6	171.9	171.9	171.9	171.4
13	144.1	144.2	144.1	143.6	144.0	C-1″	171.2	171.3	171.4	171.4	171.0
14	42.2	42.2	42.1	42.1	42.0	2″	94.0	94.1	94.1	94.1	93.8
15	28.3	28.2	28.2	28.2	28.1	3″	98.2	98.1	97.9	98.1	97.9
16	23.9	23.6	23.6	46.9	23.6	C-1‴	172.3	172.5	172.4	172.4	172.2
17	47.0	47.0	46.9	40.8	46.8	2‴	65.1	65.0	64.7	65.0	64.7
18	41.8	41.7	41.7	40.8	41.6	C-1⁗	95.7	95.8	95.8	95.8	95.7
19	46.2	46.1	46.1	42.4	46.0	2⁗	74.1	74.1	74.1	74.2	74.0
20	30.8	30.8	30.8	29.1	30.6	3⁗	78.9	78.9	78.9	78.9	78.7
21	34.1	34.0	34.0	31.7	33.8	4⁗	71.3	71.1	71.1	71.1	70.9
22	32.6	32.4	32.5	28.1	32.4	5⁗	79.3	79.4	79.4	79.4	79.1
23	64.5	206.6	180.1	16.9	27.9	6⁗	62.4	62.2	62.2	62.3	62.0
24	13.5	10.3	12.6	15.5	16.7						

Pyridine-d_5, *a*) 68 MHz, *b*) 125 MHz.

hydrolysis of **3** furnished gypsogenic acid (**11**),[10] D-glucose, and D-glucuronic acid, while serratagenic acid (**12**),[11] D-glucose, and D-glucuronic acid were obtained by acid hydrolysis of **4**.[8] The proton and carbon signals in the ^{1}H-NMR (pyridine-d_5) and ^{13}C-NMR (Table 1) spectra[9] of **3** and **4** were superimposable on those of **5**, except for signals due to the 23 or 29-position of the aglycone moiety. The ^{1}H-NMR (pyridine-d_5) and ^{13}C-NMR (Table 1) spectra[9] of **3** and **4** showed the signals assignable to an aglycone moiety [**3**: δ 3.17 (dd-like, 18-H), 4.62 (m, 3-H), 5.40, (dd-like, 12-H); **4**: δ 3.26 (dd, J=4.0, 11.6 Hz, 3-H), 3.32 (dd-like, 18-H), 5.46, (dd-like, 12-H)] together with a 28-*O*-β-D-glucopyranosyl moiety [**3**: δ 6.31 (d, J=7.9 Hz, 1′′′′-H); **4**: δ 6.35 (J=8.0 Hz, 1′′′′-H)] and a 3-*O*-β-D-glucopyranosiduronic acid moiety having the dioxolane-type substituent [**3**: δ 4.79 (s, 2′′′-H$_2$), 5.19 (d, J=7.6 Hz, 1′-H), 5.97 (s, 3″-H); **4**: δ 4.81 (s, 2′′′-H$_2$), 4.99 (d, J=7.6 Hz, 1′-H), 5.98 (s, 3″-H)]. In the HMBC experiments of **3** and **4**, long-range correlations were observed between the following protons and carbons: 1′-H and 3-C; 3′-H and 3″-C; 3″-H and 3′-, 2′′′-C; 4′-H and 2″-C; 2′′′-H$_2$ and 3″-, 1′′′-C; 1′′′′-H and 28-C. These findings led us to elucidate the structures of basellasaponins C (**3**) and D (**4**) as shown.

Experimental

The following instruments were used to obtain physical data: specific rotations, Horiba SEPA-300 digital polarimeter (l=5 cm); UV spectra, Shimadzu UV-1200 spectrometer; IR spectra, Shimadzu FTIR-8100 spectrometer; CD spectra, J-720WI; ^{1}H-NMR spectra, JEOL EX-CALIBUR (270 MHz) and JNM-LA500 (500 MHz) spectrometers; ^{13}C-NMR spectra, JEOL EX-CALIBUR (68 MHz) and JNM-LA500 (125 MHz) spectrometers with tetramethylsilane as an internal standard; MS and high-resolution MS, JEOL JMS-SX 102A mass spectrometer and JMS-GCMATE; HPLC, Shimadzu LC-10AS chromatograph.

The following experimental conditions were used for chromatography: normal-phase column chromatography; Silica gel BW-200 (Fuji Silysia Chemical, Ltd., 150—350 mesh), reversed-phase column chromatography; Chromatorex ODS DM1020T (Fuji Silysia Chemical, Ltd., 100—200 mesh): TLC, pre-coated TLC plates with Silica gel 60F$_{254}$ (Merck, 0.25 mm) (normal-phase) and Silica gel RP-18 F$_{254S}$ (Merck, 0.25 mm) (reversed-phase); HPTLC, pre-coated TLC plates with Silica gel RP-18 WF$_{254S}$ (Merck, 0.25 mm) (reversed-phase). Detection was performed by spraying with 1% aqueous $Ce(SO_4)_2$–10% aqueous H_2SO_4, followed by heating.

Isolation of Basellasaponins A (1), B (2), C (3), and D (4) and Known Saponins (5—8) from the Fresh Aerial Parts of *B. rubra* L. The fresh aerial parts (13.8 kg) of *B. rubra* L. was cut and extracted three times with MeOH under reflux. Evaporation of the solvent under reduced pressure provided the MeOH extract (247.2 g, 1.79%), and this extract (241.0 g) was subjected to Diaion HP-20 column chromatography [Nippon Rensou, Co., Ltd. (3.0 kg), H_2O→MeOH→$CHCl_3$] to give the H_2O-eluate (170.0 g, 1.23%), the MeOH-eluate (47.3 g, 0.34%), and the $CHCl_3$ eluate (21.5 g, 0.16%). Normal-phase silica gel column chromatography [BW-200 (Fuji Silysia, Ltd., 480 g), $CHCl_3$–MeOH–H_2O (7 : 3 : 0.5→6 : 4 : 1→5 : 5 : 1, v/v)] of the MeOH eluate (45.0 g) gave seven fractions [fr. 1 (5.4 g), fr. 2 (4.2 g), fr. 3 (7.8 g), fr. 4 (9.8 g), fr. 5 (2.4 g), fr. 6 (13.1 g), fr. 7 (3.8 g)]. Fraction 3 (526 mg) was purified by HPLC [YMC-Pack ODS-A (250×20 mm i.d., YMC Co., Ltd.), MeOH–1% aq. AcOH (40 : 60, v/v)] to give momordins IIb[6] (**7**, 13 mg, 0.0001%) and IIc[6] (**8**, 59 mg, 0.0004%). Fraction 5 (100 mg) was purified by repeated HPLC [CH_3CN–1% aq. TFA (40 : 60, v/v)] to give spinacoside C[5] (**6**, 90 mg, 0.0007%). Fraction 6 (100 mg) was purified by HPLC [CH_3CN–1% aq. TFA (40 : 60, v/v)] to give betavulgaroside I[4] (**5**, 50 mg, 0.0004%). Fraction 7 (1.5 g) was purified by repeated HPLC [1) CH_3CN–1% aq. TFA (40 : 60, v/v); 2) CH_3CN–1% aq. TFA (20 : 80, v/v); 3) CH_3CN–1% aq. TFA (35 : 65, v/v)] to give basellasaponins A (**1**, 42 mg, 0.0003%), B (**2**, 22 mg, 0.0002%), C (**3**, 21 mg, 0.0002%), and D (**4**, 16 mg, 0.0001%). The known compounds (**5**—**8**) were identified by comparison of their physical date ([α]$_D$, IR, ^{1}H-NMR, ^{13}C-NMR) with reported values.[4—6]

Basellasaponin A (**1**): Colorless fine crystals from MeOH–H_2O, mp 228—230 °C, $[\alpha]_D^{24}$ +30.1° (c=0.1, MeOH). High-resolution positive-ion FAB-MS: Calcd for $C_{47}H_{70}O_{21}Na$ (M+Na)$^+$: 993.4307. Found: 993.4343. IR (KBr): 3403, 1736, 1076, 1036 cm^{-1}. ^{1}H-NMR (270 MHz, pyridine-d_5) δ: 0.88 (12H, s, 24, 25, 29, 30-H$_3$), 1.09, 1.22 (3H each, both s, 26, 27-H$_3$), 3.16 (1H, dd-like, 18-H), 3.65 (1H, d, J=11.2 Hz), 4.20 (1H, m) (23-H$_2$), 4.22 (1H, m, 3-H), 4.75 (2H, s, 2′′′-H$_2$), 5.17 (1H, d, J=7.6 Hz, 1′-H), 5.40 (1H, dd-like, 12-H), 5.91 (1H, s, 3″-H), 6.27 (1H, d, J=7.6 Hz, 1′′′′-H). ^{13}C-NMR (68 MHz, pyridine-d_5) δ_C: given in Table 1. Negative-ion FAB-MS: m/z 969 (M−H)$^-$, 809 (M−$C_5H_5O_6$)$^-$, 807 (M−$C_6H_{11}O_5$)$^-$. Positive-ion FAB-MS: m/z 993 (M+Na)$^+$.

Basellasaponin B (**2**): Colorless fine crystals from MeOH–H_2O, mp 226—228 °C, $[\alpha]_D^{26}$ +57.4° (c=0.1, MeOH). High-resolution positive-ion FAB-MS: Calcd for $C_{47}H_{68}O_{21}Na$ (M+Na)$^+$: 991.4150. Found: 991.4125. IR (KBr): 3415, 1728, 1076, 1036 cm^{-1}. ^{1}H-NMR (270 MHz, pyridine-d_5) δ: 0.81, 0.89, 0.92, 1.04 (3H each, all s, 25, 30, 29, 26-H$_3$), 1.26 (6H, s, 24, 27-H$_3$), 3.18 (1H, dd-like, 18-H), 4.10 (1H, m, 3-H), 4.79 (2H, s, 2′′′-H$_2$), 4.88 (1H, d, J=7.6 Hz, 1′-H), 5.40 (1H, dd-like, 12-H), 5.96 (1H, s, 3″-H), 6.30 (1H, J=8.0 Hz, 1′′′′-H), 9.69 (1H, s, 23-H). ^{13}C-NMR (68 MHz, pyridine-d_5) δ_C: given in Table 1. Negative-ion FAB-MS: m/z 967 (M−H)$^-$, 807 (M−$C_5H_5O_6$)$^-$, 805 (M−$C_6H_{11}O_5$)$^-$. Positive-ion FAB-MS: m/z 991 (M+Na)$^+$.

Basellasaponin C (**3**): Colorless fine crystals from MeOH–H_2O, mp 230—232 °C, $[\alpha]_D^{25}$ +42.1° (c=0.1, MeOH). High-resolution positive-ion FAB-MS: Calcd for $C_{47}H_{68}O_{22}Na$ (M+Na)$^+$: 1007.4100. Found: 1007.4132. IR (KBr): 3426, 1737, 1076, 1036 cm^{-1}. ^{1}H-NMR (270 MHz, pyridine-d_5) δ: 0.87 (6H, s, 25, 30-H$_3$), 0.89, 1.07, 1.23, 1.51 (3H each, all s, 29, 26, 27, 24-H$_3$), 3.17 (1H, dd-like, 18-H), 4.62 (1H, m, 3-H), 4.73 (2H, m, 2′′′-H2), 4.79 (2H, s, 2′′′-H$_2$), 5.19 (1H, d, J=7.6 Hz, 1′-H), 5.40 (1H, dd-like, 12-H), 5.97 (1H, s, 3″-H), 6.31 (1H, d, J=7.9 Hz, 1′′′′-H). ^{13}C-NMR (68 MHz, pyridine-d_5) δ_C: given in Table 1. Negative-ion FAB-MS: m/z 983 (M−H)$^-$, 821 (M−$C_6H_{11}O_5$)$^-$. Positive-ion FAB-MS: m/z 1007 (M+Na)$^+$.

Basellasaponin D (**4**): Colorless fine crystals from MeOH–H_2O, mp 215—217 °C, $[\alpha]_D^{27}$ +24.0° (c=0.1, MeOH). High-resolution positive-ion FAB-MS: Calcd for $C_{47}H_{68}O_{22}Na$ (M+Na)$^+$: 1007.4100. Found: 1007.4112. IR (KBr): 3426, 1732, 1076, 1036 cm^{-1}. ^{1}H-NMR (500 MHz, pyridine-d_5) δ: 0.80, 0.93, 1.08, 1.24, 1.28, 1.45 (3H each, all s, 25, 24, 26, 23, 27, 30-H$_3$), 3.26 (1H, dd, J=4.0, 11.6 Hz, 3-H), 3.32 (1H, dd-like, 18-H), 4.81 (2H, s, 2′′′-H$_2$), 4.99 (1H, d, J=7.6 Hz, 1′-H), 5.46 (1H, dd-like, 12-H), 5.98 (1H, s, 3″-H), 6.35 (1H, J=8.0 Hz, 1′′′′-H). ^{13}C-NMR (125 MHz, pyridine-d_5) δ_C: given in Table 1. Negative-ion FAB-MS: m/z 983 (M−H)$^-$. Positive-ion FAB-MS: m/z 1007 (M+Na)$^+$.

Acid Hydrolysis of Basellasaponins (1—4) A solution of **1**—**4** (2 mg each) in 5% aq. H_2SO_4–1,4-dioxane (0.5 ml, 1 : 1, v/v) was heated under reflux for 1 h. After cooling, the reaction mixture was neutralized with Amberlite IRA-400 (OH^- form) and the residue was removed by filtration. After removal of the solvent from the filtrate *in vacuo*, the residue was transferred to a Sep-Pak C18 cartridge with H_2O and MeOH. The MeOH eluate from **1**—**4** was concentrated and the residue was purified by normal-phase silica gel column chromatography [1.0 g, $CHCl_3$–MeOH–H_2O (10 : 3 : 1, v/v)→MeOH] to give sapogenols [hederagenin (**9**, 0.8 mg, 82% from **1**), gypsogenin (**10**, 0.6 mg, 62% from **2**), gypsogenic acid (**11**, 0.8 mg, 81% from **3**), and serratagenic acid (**12**, 0.8 mg, 81% from **4**)], which were identified by physical data comparison with reported values.[4b,7,10,11] The H_2O eluate was concentrated and the residue was treated with L-cysteine methyl ester hydrochloride (4 mg) in pyridine (0.5 ml) at 60 °C for 1 h. After reaction, the solution was treated with *N*,*O*-bis(trimethylsilyl)trifluoroacetamide (0.2 ml) at 60 °C for 1 h. The supernatant was then subjected to GLC analysis to identify the derivatives of D-glucuronic acid (i) and D-glucose (ii); GLC conditions: column: Supeluco STB^{TM}-1, 30 m×0.25 mm (i.d.) capillary column, column temperature: 230 °C, He flow rate: 15 ml/min, t_R: i: 24.4 min, ii: 24.2 min.

References and Notes

1) Part XXII: Murakami T., Kohno K., Matsuda H., Yoshikawa M., *Chem. Pharm. Bull.*, **49**, 73—77 (2001).
2) *a*) Takami A., *Oita Daigaku Kyoikugakubu Kenkyo Kiyo, Shizenkagaku*, **3**, 53—62 (1969) [*Chem. Abstr.*, **74**, 108135*q* (1971)]; *b*) Banerjee A. K., Jain M., Dubey V., *Fitoterapia*, **63**, 377 (1992) [*Chem. Abstr.*, **118**, 109476*r* (1993)]; *c*) Glassgen W. E., Metzger J. W., Heuer S., Strack D., *Phytochemistry*, **33**, 1525—1527 (1993).
3) *a*) Matsuda H., Li Y., Yoshikawa M., Life Sci., **66**, PL41—46 (2000); *b*) *Idem*, *ibid.*, **66**, 2233—2238 (2000); *c*) *Idem*, *ibid.*, **67**, 2921—2927 (2000); *d*) Li Y., Wen S., Yamahara J., Yoshikawa M., *Eur. J. Pharmacol.*, **387**, 339—344 (2000); *e*) Li Y., Matsuda H., Yamahara J., Yoshikawa M., *ibid.*, **392**, 71—77 (2000); *f*) Murakami T., Kishi A., Matsuda H., Yoshikawa M., *Chem. Pharm. Bull.*, **48**, 994—1000

(2000); *g*) Murakami T., Kohno K., Kishi A., Matsuda H., Yoshikawa M., *ibid.*, **48**, 1673—1680 (2000); *h*) Murakami T., Nakamura J., Kageura T., Matsuda H., Yoshikawa M., *ibid.*, **48**, 1720—1725 (2000); *i*) Murakami T., Emoto A., Matsuda H., Yoshikawa M., *ibid.*, **49**, 54—63 (2001).

4) *a*) Yoshikawa M., Murakami T., Kadoya M., Matsuda H., Yamahara J., Muraoka O., Murakami N., *Heterocycles*, **41**, 1621—1626 (1995); *b*) Yoshikawa M., Murakami T., Kadoya M., Matsuda H., Muraoka O., Yamahara J., Murakami N., *Chem. Pharm. Bull.*, **44**, 1212—1217 (1996); *c*) Yoshikawa M., Murakami T., Inaduki M., Hirano K., Yamahara J., Matsuda H., *ibid.*, **45**, 561—563 (1997); *d*) Yoshikawa M., Murakami T., Kadoya M., Yamahara J., Matsuda H., *ibid.*, **46**, 1758—1763 (1998); *e*) Murakami T., Matsuda H., Inadzuki M., Hirano K., Yoshikawa M., *ibid.*, **47**, 1717—1724 (1999).

5) Yoshikawa M., Murakami T., Hirano K, Matsuda H., Yamahara J., Ohtani K, Kasai R., Yamasaki K., *Heterocycles*, **49**, 93—96 (1998).

6) Iwamoto M., Okabe M., Yamauchi T., *Chem. Pharm. Bull.*, **33**, 1—7 (1985).

7) Yoshikawa M., Yoshizumi S., Ueno T., Matsuda H., Murakami T., Yamahara J., Murakami N., *Chem. Pharm. Bull.*, **43**, 1878—1882 (1995).

8) *a*) Hara S., Okabe H., Mihashi K., *Chem. Pharm. Bull.*, **34**, 1843—1845 (1986); *b*) Zhang D., Miyase T., Kuroyanagi M., Umehara K., Ueno A., *ibid.*, **43**, 966—970 (1995).

9) The ^{1}H- and ^{13}C-NMR spectra of **1**, **2**, **3**, and **4** were assigned on the basis of homo- and hetero-correlation spectroscopy (^{1}H–^{1}H, ^{1}H–^{13}C COSY), homo- and heteronuclear Hartmann-Hahn spectroscopy (^{1}H–^{1}H, ^{1}H–^{13}C HOHAHA) and heteronuclear multiple bond correlation (HMBC) experiments.

10) Frechet D., Christ B., Sorbier B. M., Fischer H., Vuilhorgne M., *Phytochemistry*, **30**, 927—931 (1991).

11) *a*) Rangaswami S., Sarangan S., *Tetrahedron*, **25**, 3701—3705 (1969); *b*) Kasai R., Oinaka T., Yang C R., Zhou J., Tanaka O., *Chem. Pharm. Bull.*, **35**, 1486—1490 (1987).