Stomachic Principles in Ginger. III.[1] An Anti-ulcer Principle, 6-Gingesulfonic Acid, and Three Monoacyldigalactosylglycerols, Gingerglycolipids A, B, and C, from Zingiberis Rhizoma Originating in Taiwan

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An anti-ulcer constituent, 6-gingesulfonic acid, and three monoacyldigalactosylglycerols, gingerglycolipids A, B, and C, were isolated from Zingiberis Rhizoma, the dried rhizome of *Zingiber officinale* ROSCOE which was cultivated in Taiwan, together with (+)-angelicoidenol-2-*O*-β-D-glucopyranoside. Based on chemical reactions and physicochemical evidence, the structures of 6-gingesulfonic acid, gingerglycolipids A, B, and C have been determined. In addition, the absolute stereostructure of (+)-angelicoidenol-2-*O*-β-D-glucopyranoside was clarified on the basis of its synthesis from *d*-borneol. 6-Gingesulfonic acid showed weaker pungency and more potent anti-ulcer activity than 6-gingerol and 6-shogaol.

Keywords Zingiberis Rhizoma; *Zingiber officinale*; 6-gingesulfonic acid; gingerglycolipid; anti-ulcer principle; pungent effect

Zingiberis Rhizoma (ginger), which is prepared from the rhizome of *Zingiber officinale* ROSCOE (Zingiberaceae), is one of the best known crude drugs and it is prescribed in Chinese and Japanese traditional preparations used for treatment of headache, nausea, stomach-ache, and colds. Zingiberis Rhizoma is used in both processed and unprocessed forms known in Japanese as Kankyo (steamed rhizome) and Shoukyo (dried rhizome). On the other hand, Zingiberis Rhizoma is classified in China into two types as 乾姜 (gan jiang, dried rhizome) and 生姜 (sheng jiang, fresh rhizome), which have quite distinct applications in Chinese traditional medicine.[2] In recent years, due to the poor supply of Japanese Zingiberis Rhizoma, imported Zingiberis Rhizoma originating in China and Taiwan have been widely used for medicinal preparations in Japan.[1,3]

In regard to chemical studies on the constituents of Zingiberis Rhizoma, many chemical investigations of lipophilic constituents have been carried out and large numbers of terpenoids and pungent principles have been identified.[4]

During the course of our screening to find biologically active constituents contained in crude drugs, we have so far found several bioactive compounds, showing anti-cathartic, antiserotonergic, and gastrointestinal motility-enhancing effects, from Zingiberis Rhizoma.[5] As a part of these studies, by monitoring for anti-ulcer effects in rats, we have isolated β-sesquiphellandrene, β-bisabolene, *ar*-curcumene, α-zingiberene, 6-gingerol (**6**), and 6-shogaol (**7**) as active principles in stomachic medication from the lipophilic portion of Zingiberis Rhizoma originating in Taiwan.[1,6] Furthermore, we have developed quantitative and qualitative analytical methods by means of HPLC and gas liquid chromatography (GLC) for bioactive constituents contained in various kinds of Zingiberis Rhizoma and the fresh rhizome of *Zingiber officinale*, either domestic or foreign.[7] As an application of those quantitative analytical methods, changes of the constituents during the processing of Zingiberis Rhizoma were characterized.[7,8] In continuing studies, we have examined the water-soluble portion of Zingiberis Rhizoma originating in Taiwan and have isolated an anti-ulcer principle named 6-gingesulfonic acid (**1**), together with three monoacyldigalactosylglycerols designated gingerglycolipids A (**2**), B (**3**), and C (**4**) and (+)-angelicoidenol-2-*O*-β-D-glucopyranoside (**5**). Here we present a full account of the structure elucidation of **1**, **2**, **3**, and **4**,[9] as well as a synthesis of **5** to clarify the absolute stereostructure.

The methanol extract of Zingiberis Rhizoma, the dried rhizome of *Zingiber officinale* ROSCOE cultivated in Taiwan, was partitioned into ethyl acetate and water to furnish the ethyl acetate-soluble portion and the water-soluble portion as described in a previous paper.[1] The water-soluble portion was partitioned into a mixture of 1-butanol and water and the 1-butanol fraction was subjected to ordinary and reversed-phase column chromatography to furnish 6-gingesulfonic acid (**1**, 0.0013% from the crude drug), gingerglycolipids A (**2**, 0.0013%), B (**3**, 0.0014%), and C (**4**, 0.0014%), (+)-angelicoidenol-2-*O*-β-D-glucopyranoside (**5**, 0.0014%) and uridine (0.0011%).

6-Gingesulfonic Acid (1) 6-Gingesulfonic acid (**1**) was obtained as a white powder. The infrared (IR) spectrum of **1** showed absorption bands ascribable to hydroxyl (3590 cm^{-1}), carbonyl (1710 cm^{-1}), and sulfonic acid (1209, 1171, 1046 cm^{-1}) functions. Typical benzenoid absorption was seen at 281.5 nm in the ultraviolet (UV) spectrum of **1**. The negative fast atom bombardment MS (negative FAB-MS) of **1** showed the quasimolecular ion peak at *m/z* 357 $(M-H)^-$, while the positive FAB-MS showed the quasimolecular ion peak at *m/z* 403 $(M-H+2Na)^+$ (glycerol matrix containing NaCl) and at *m/z* 371 $(M-H+2Li)^+$ (glycerol matrix containing LiCl).

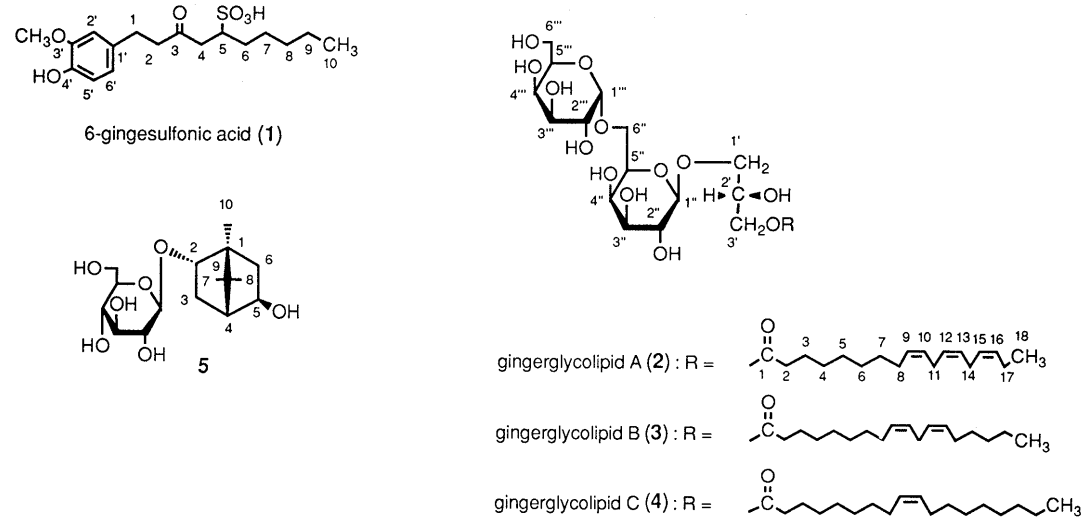

Chart 1

The high-resolution MS measurement of **1** revealed the molecular formula to be $C_{17}H_{26}O_6S$. The proton nuclear magnetic resonance (^{1}H-NMR) spectrum of **1** showed a sulfonic acid-bearing methine proton at δ 3.32 (m, 5-H) together with many other signals closely resembling those of 6-gingerol (**6**). Acetylation of **1** with acetic anhydride in pyridine yielded the monoacetate (**1a**). Comparison of the ^{1}H-NMR and ^{13}C-NMR (Table I)[10] spectra for **1** and **1a** with those for 6-gingerol (**6**) led us to presume the structure **1** which has a sulfonic acid function at the 5-position. Furthermore, the structure **1** was substantiated by chemical correlation with **6**. Namely, sulfonation of 6-shogaol (**7**), which was derived from **6** by acid treatment, with sodium hydrogen sulfite ($NaHSO_3$) in the presence of *tert*-butyl perbenzoate in refluxing methanol furnished (±)-**1** in 90% yield. Thus, the structure of 6-gingesulfonic acid (**1**)[11] was determined to be as shown.

Gingerglycolipid A (2) Gingerglycolipid A (**2**) was also obtained as a white powder. The molecular formula $C_{33}H_{56}O_{14}$ of **2** was confirmed from the quasimolecular ion peaks at m/z 699 $(M+Na)^+$ and m/z 683 $(M+Li)^+$ in the positive FAB-MS and by high-resolution MS measurement. The IR spectrum of **2** showed absorption bands due to hydroxyl and ester groups at 3570 and 1734 cm^{-1}, while the ^{1}H-NMR spectrum of **2** showed signals which were characteristically attributable to a glyceroglycolipid. Treatment of **2** with sodium methoxide in methanol furnished a glycerol digalactoside and methyl linolenate, which was identified by GLC. The glycerol digalactoside, $[\alpha]_D$ +80.0° (H_2O), was shown to be identical with (2′*R*)-glyceryl 6-*O*-(α-D-galactopyranosyl)-β-D-galactopyranoside (**8**), which was obtained by alkaline treatment of a digalactosyl diacylglycerol isolated from *Medicago sative*.[12] Detailed comparisons of the ^{1}H-NMR and ^{13}C-NMR data for **2** with those for **8** and various known digalactosyl acylglycerols[13] showed that the fatty acid residue is attached to 3′-OH in the glycerol moiety of **2**. Consequently, the chemical structure of gingerglycolipid A was determined to be (2′*S*)-3′-*O*-linolenoylglyceryl 6-*O*-(α-D-galactopyranosyl)-β-D-galactopyranoside (**2**).

Gingerglycolipids B (3) and C (4) Gingerglycolipid B

Table I. ^{13}C-NMR Data for **1**, **1a** and **6**

	1	**1a**	**6**
C-1	30.4	30.5	33.7
C-2	45.9	45.6	47.2
C-3	210.6	210.2	212.8
C-4	44.7	44.6	52.1
C-5	57.0	57.2	69.7
C-6	32.9	33.0	39.1
C-7	27.8$^{a)}$	28.0$^{a)}$	27.1$^{a)}$
C-8	31.9$^{a)}$	32.0$^{a)}$	31.0$^{a)}$
C-9	23.5	23.6	24.4
C-10	14.4	14.4	15.2
C-1′	133.9	139.4	134.8
C-2′	113.0	113.9	113.9
C-3′	148.8	152.4	149.6
C-4′	145.6	141.9	146.5
C-5′	116.0	123.5	116.9
C-6′	121.6	121.4	122.5
OMe	56.3	56.4	57.1

δ_C at 67.5 MHz, in CD_3OD. *a*) The assignments may be interchangeable within the same column.

Table II. ^{13}C-NMR Data for **2**, **3**, and **4**

	2	**3**	**4**
1′	72.9	72.9	72.9
2′	70.4	70.4	70.4
3′	67.4	67.4	67.3
1″	106.1	106.1	106.1
2″	73.4$^{a)}$	73.3	73.3
3″	75.4	75.3	75.3
4″	70.9$^{b)}$	70.8$^{a)}$	70.8$^{a)}$
5″	75.4	75.3	75.3
6″	68.6	68.5	68.5
1‴	101.3	101.3	101.3
2‴	71.0$^{b)}$	71.0$^{a)}$	71.0$^{a)}$
3‴	72.2	72.2	72.2
4‴	71.8	71.8	71.8
5‴	73.3$^{a)}$	73.3	73.3
6‴	63.5	63.5	63.5
1	176.2	176.2	176.3
18	15.4	15.3	15.3

δ_C at 67.5 MHz, in CD_3OD. *a*, *b*) The assignments may be interchangeable within the same column.

(**3**) was obtained as a white powder. The IR spectrum of **3** showed the presence of hydroxyl groups and an ester function. The positive FAB-MS of **3** showed the quasimolecular ion peaks at *m*/*z* 701 $(M+Na)^+$ and *m*/*z* 685 $(M+Li)^+$ and the molecular formula $C_{33}H_{58}O_{14}$ was confirmed by high-resolution MS measurement. The ^{1}H-NMR and ^{13}C-NMR spectra of **3** closely resembled those of gingerglycolipid A (**2**) except for the signals due to the fatty acid moiety. Alkaline treatment of **3** as carried out for the solvolysis of **2** furnished the same glyceryl digalactoside (**8**) together with methyl linoleate. Furthermore, the ^{1}H-NMR and ^{13}C-NMR analysis of **3** in comparison with **2** indicated that the linoleic acid residue in **3** was attached to 3′-OH in the glycerol moiety instead of linolenic acid in **2**. Based on the above mentioned evidence, the chemical structure of gingerglycolipid B (**3**) was determined to be as shown.

Gingerglycolipid C (**4**), obtained as a white powder, gave quasimolecular ion peaks at *m*/*z* 703 $(M+Na)^+$ and *m*/*z* 687 $(M+Li)^+$ in the positive FAB-MS and the molecular formula $C_{33}H_{60}O_{14}$ was determined by high-resolution MS measurement. Alkaline treatment of **4** provided **8** and methyl oleate. The ^{1}H-NMR and ^{13}C-NMR spectra of **4** closely resembled those of gingerglycolipids A (**2**) and B (**3**) and showed signals which were characteristic of a 3′-acyl-1′-digalactosylglycerol. Thus, the structure of gingerglycolipid C (**4**) was determined to be as shown.

Synthesis of (+)-Angelicoidenol-2-*O*-β-D-glucopyranoside (5) (+)-Angelicoidenol-2-*O*-β-D-glucopyranoside (**5**) has been isolated from *Berchemia racemosa* together with (−)-angelicoidenol-2-*O*-β-D-glucopyranoside and their absolute configuration was clarified by application of the ^{13}C-NMR glucosidation shift rule.[14] In order to confirm the identification of **5** isolated from Zingiberis Rhizoma and also to provide chemical evidence of its absolute stereostructure, **5** was synthesized from (+)-borneol (**9**) and D-glucose. The starting material, (+)-5-ketoborneol (**10**), was prepared from **9** using a literature procedure.[15] Namely, acetylation of **9** followed by oxidation with chromic trioxide (CrO_3) in acetic acid gave (+)-2-acetyl-5-ketoborneol, which was subjected to deacetylation to furnish **10** in 31.8% yield from **9**. Glycosidation of **10** with 1-bromo-2,3,4,6-tetra-*O*-acetyl-D-glucopyranose in the presence of silver carbonate in dry benzene under reflux furnished **11** in 81% yield. Reduction of the carbonyl function and elimination of acetyl groups in **11** with sodium borohydride ($NaBH_4$) in methanol gave (+)-angelicoidenol-2-*O*-β-D-glucopyranoside, which was found to be identical with **5** obtained by us from Zingiberis Rhizoma.

Anti-ulcer Activity and Pungent Effect of 6-Gingesulfonic Acid (1) By monitoring the effect on hydrochloric

TABLE III. Effect of 6-Gingesulfonic Acid (**1**), 6-Gingerol (**6**), 6-Shogaol (**7**), and Cetraxate on HCl–Ethanol Induced Gastric Ulcer in Rats

Treatment	Dose (mg/kg)	*N*	Total length (mm) (Mean ± S.E.)	Inhibition (%)
Control		5	101.5 ± 19.5	—
6-Gingerol (**6**)	150	5	43.1 ± 9.2[a)]	57.5
6-Shogaol (**7**)	150	5	30.2 ± 5.5[a)]	70.2
6-Gingesulfonic acid (**1**)	150	5	7.4 ± 1.6[b)]	92.7
6-Gingesulfonic acid (**1**)	300	6	0.5 ± 0.3[b)]	99.6
Cetraxate	300	5	1.5 ± 1.1[b)]	98.5

a) $p<0.05$, *b)* $p<0.01$.

TABLE IV. Pungent Effect of 6-Gingesulfonic Acid (**1**), 6-Gingerol (**6**), and 6-Shogaol (**7**)

	Pungent effect (μmol/l)				
	0.1	1.0	10	100	1000
6-Shogaol (**7**)	−	+	+	++	++
6-Gingerol (**6**)	−	−	−	+	+
6-Gingesulfonic acid (**1**)	−	−	−	−	+

Quality of tests: ++, conspicuous; +, evident; −, absent.

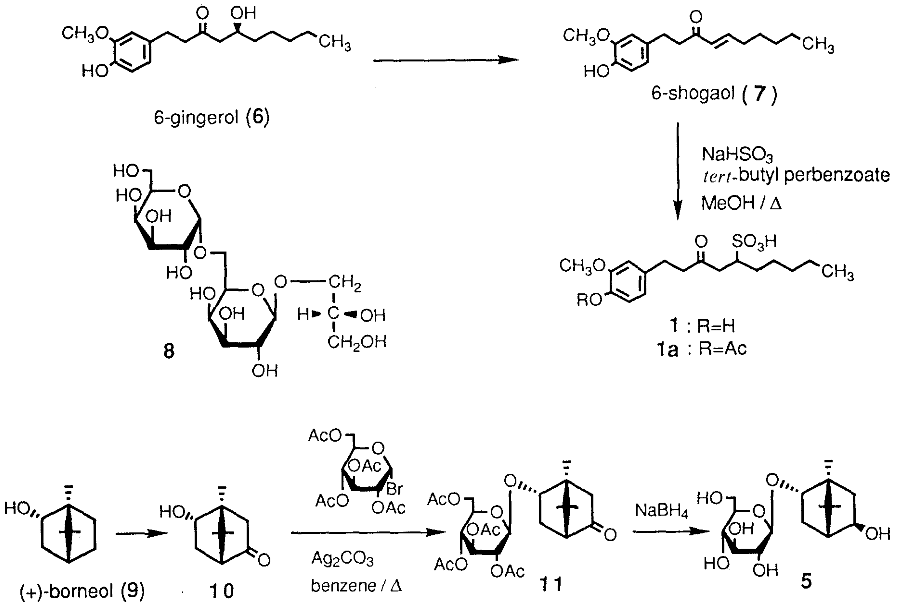

Chart 2

acid/ethanol-induced gastric lesions in rats, we have isolated various anti-ulcer principles such as 6-gingerol (**6**), 6-shogaol (**7**), and four bisabolane-type sesquiterpenes from the lipophilic fraction of Zingiberis Rhizoma.[1] We next examined the anti-ulcer activity of 6-gingesulfonic acid (**1**) in comparison with **6** and **7**. As shown in Table III, **1** significantly inhibited gastric lesions by 92.7% at the dose of 150 mg/kg (*p.o.*), while **6** and **7** showed inhibition of 57.5% and 70.2%, respectively, at the same dose. At the dose of 300 mg/kg (*p.o.*), **1** completely inhibited gastric lesions, as did a positive control, Cetraxate (a synthetic anti-ulcerative).

6-Gingesulfonic acid (**1**) exhibited weaker pungency as shown in Table IV than 6-gingerol (**6**) and 6-shogaol (**7**). Furthermore, **1** was obtained as a stable white powder and showed higher water solubility than the oily compounds **6** and **7**. These properties may be important for its use as a stomachic.

Experimental

The instruments used to obtain physical data and experimental conditions for chromatography were the same as described in our previous paper.[16]

Isolation of 6-Gingesulfonic Acid (1), Gingerglycolipids A, B, and C (2, 3, 4) and (+)-Angelicoidenol-2-*O*-*β*-D-glucopyranoside (5) The water-soluble portion[1] was partitioned into a mixture of 1-BuOH and water. After removal of the solvent from the 1-BuOH layer, the residue was subjected to silica gel column chromatography [$CHCl_3$–MeOH–H_2O (65 : 35 : 10, lower layer)] to give three fractions (frs. 1, 2 and 3). Silica gel column [$CHCl_3$–MeOH–H_2O (10 : 3 : 1, lower layer) followed by reversed-phase silica gel column [MeOH–H_2O (1 : 2)] chromatography of fraction 1 furnished 6-gingesulfonic acid (**1**, 195 mg), gingerglycolipids A (**2**, 195 mg) and B (**3**, 210 mg) and (+)-angelicoidenol-2-*O*-*β*-D-glucopyranoside (**5**, 210 mg). Fraction 2 was subjected to silica gel column [$CHCl_3$–MeOH–H_2O (65 : 35 : 10, lower layer)] and reversed-phase silica gel column [MeOH–H_2O (1 : 2)] chromatography to give gingerglycolipid C (**4**, 210 mg) and uridine (178 mg), which was identified by TLC, ^{1}H-NMR, and ^{13}C-NMR.

6-Gingesulfonic Acid (**1**): A white powder $[\alpha]_D^{20}$ −1.0° (c = 10.8, MeOH). High-resolution FAB-MS: Found: 371.1697; Calcd for $C_{17}H_{25}Li_2O_6S$ $(M-H+2Li)^+$: 371.1693. UV λ_{max}^{MeOH} nm (ε): 221.5 (1110), 281.5 (1000). IR ν_{max}^{KBr} cm^{-1}: 3590, 1710, 1520, 1209, 1171, 1046. ^{1}H-NMR (270 MHz, CD_3OD) δ: 0.78 (3H, t, J = 7 Hz, 10-H_3), 0.97—1.26 (6H, m, 7, 8, 9-H_2), 1.34 (1H, m), 1.80 (1H, m) (6-H_2), 2.41 (1H, dd, J = 6, 18 Hz), 2.94 (1H, dd, J = 6, 18 Hz) (4-H_2), 2.70 (4H, m, 1, 2-H_2), 3.32 (m, 5-H), 3.73 (3H, s, OCH_3), 6.51 (1H, dd, J = 2, 8 Hz, 6′-H), 6.59 (1H, d, J = 8 Hz, 5′-H), 6.68 (1H, d, J = 2 Hz, 2′-H). ^{13}C-NMR (67.5 MHz, CD_3OD) δ_C: see Table I. Positive FAB-MS: m/z 371 $(M-H+2Li)^+$ (glycerol matrix + LiCl), 403 $(M-H+2Na)^+$ (glycerol matrix + NaCl). Negative FAB-MS m/z: 357 $(M-H)^-$.

Gingerglycolipid A (**2**): A white powder, $[\alpha]_D^{20}$ +37.7° (c = 10.0, MeOH). High-resolution FAB-MS: Found: 699.3577; Calcd for $C_{33}H_{56}NaO_{14}$ $(M+Na)^+$: 699.3587. IR ν_{max}^{KBr} cm^{-1}: 3570, 1734. ^{1}H-NMR (270 MHz, CD_3OD) δ: 0.89 (3H, t, J = 7 Hz, 18-H_3), 1.33 (10H, m, 3, 4, 5, 6, 7-H_2), 2.08 (4H, m, 8, 17-H_2), 2.35 (2H, t, J = 8 Hz, 2-H_2), 2.80 (4H, dd, J = 6, 6 Hz, 11, 14-H_2), 4.13 (2H, d, J = 6 Hz, 3′-H_2), 4.24 (1H, d, J = 7 Hz, 1″-H), 5.53 (6H, m, 9, 10, 12, 13, 15, 16-H). ^{13}C-NMR (67.5 MHz, CD_3OD) δ_C: 22.2 (1C), 26.8 (1C), 27.2 (1C), 27.3 (1C), 28.9 (1C), 31.0 (2C), 31.1 (1C), 31.5 (1C) (C-3, 4, 5, 6, 7, 8, 11, 14, 17), 35.7 (C-2), 129.0 (1C), 129.6 (1C), 129.9 (2C), 131.6 (1C), 133.5 (1C) (C-9, 10, 12, 13, 15, 16), and the other signals given in Table II. Positive FAB-MS: m/z 683 $(M+Li)^+$ (glycerol matrix + LiCl), 699 $(M+Na)^+$ (glycerol matrix + NaCl).

Gingerglycolipid B (**3**): A white powder, $[\alpha]_D^{20}$ +50.9° (c = 7.5, MeOH), High-resolution FAB-MS: Found: 701.3737; Calcd for $C_{33}H_{58}NaO_{14}$ $(M+Na)^+$: 701.3724. IR ν_{max}^{KBr} cm^{-1}: 3590, 3569, 1735. ^{1}H-NMR (270 MHz, CD_3OD) δ: 0.89 (3H, t, J = 7 Hz, 18-H_3), 1.34—1.80 (16H, m, 3, 4, 5, 6, 7, 15, 16, 17-H_2), 2.06 (4H, m, 8, 14-H_2), 2.36 (2H, t, J = 7 Hz, 2-H_2), 2.78 (2H, m, 11-H_2), 4.15 (2H, d, J = 6 Hz, 3′-H_2), 4.25 (1H, d, J = 7 Hz, 1″-H), 5.31—5.37 (4H, m, 9, 10, 12, 13-H). ^{13}C-NMR (67.5 MHz, CD_3OD) δ_C: 24.4 (1C), 26.8 (1C), 27.3 (1C), 29.0 (2C), 31.0 (2C), 31.1 (1C), 31.3 (1C), 31.5 (1C), 33.5 (1C) (C-3, 4, 5, 6, 7, 8, 11, 14, 15, 16, 17), 35.7 (C-2), 129.9 (2C), 131.7 (2C) (C-9, 10, 12, 13), and the other signals given in Table II. Positive FAB-MS: m/z 685 $(M+Li)^+$ (glycerol matrix + LiCl), 701 $(M+Na)^+$ (glycerol matrix + NaCl).

Gingerglycolpid C (**4**): A white powder, $[\alpha]_D^{20}$ +26.9° (c = 10.0, MeOH). High-resolution FAB-MS: Found: 703.3851; Calcd for $C_{33}H_{60}NaO_{14}$ $(M+Na)^+$: 703.3881. IR ν_{max}^{KBr} cm^{-1}: 3571, 1736. ^{1}H-NMR (270 MHz, CD_3OD) δ: 0.90 (3H, t, J = 8 Hz, 18-H_3), 1.30—1.62 (22H, m, 3, 4, 5, 6, 7, 12, 13, 14, 15, 16, 17-H_2), 2.03 (4H, m, 8, 11-H_2), 2.36 (2H, t, J = 7 Hz, 2-H_2), 4.15 (2H, d, J = 5 Hz, 3′-H_2), 4.25 (1H, d, J = 7 Hz, 1″-H), 5.29 (2H, ddd, J = 5, 11, 16 Hz, 9, 10-H). ^{13}C-NMR (67.5 MHz, CD_3OD) δ_C: 24.6 (1C), 27.0 (1C), 29.0 (2C), 31.0 (2C), 31.1 (2C), 31.3 (1C), 31.4 (1C), 31.6 (2C), 33.8 (1C) (C-3, 4, 5, 6, 7, 8, 11, 12, 13, 14, 15, 16, 17), 35.7 (C-2), 131.7 (2C), (C-9, 10) and the other signals given in Table II. Positive FAB-MS: m/z 687 $(M+Li)^+$ (glycerol matrix + LiCl), 703 $(M+Na)^+$ (glycerol matrix + NaCl).

(+)-Angelicoidenol-2-*O*-*β*-D-glucopyranoside (**5**): A white powder, $[\alpha]_D^{20}$ −10.3° (c = 10.0, MeOH). High-resolution FAB-MS: Found: 333.1908; Calcd for $C_{16}H_{29}O_7$ $(M+H)^+$: 333.1913. IR ν_{max}^{KBr} cm^{-1}: 3417, 1078, 1032. ^{1}H-NMR (270 MHz, CD_3OD) δ: 0.81, 0.91, 1.02 (3H each, all s, 8, 9, 10-H_3), 1.08 (1H, dd, J = 3, 15 Hz, 3-H), 1.28 (1H, br d, J = 15 Hz, 6-H), 1.61 (1H, br d, J = 5 Hz, 4-H), 2.20 (1H, ddd, J = 5, 10, 15 Hz, 3-H), 2.45 (1H, dd, J = 8, 15 Hz, 6-H), 3.14 (1H, br d, J = 8 Hz, 5-H), 3.77 (1H, m, 2-H), 4.21 (1H, d, J = 8 Hz, 1′-H). ^{13}C-NMR (67.5 MHz, CD_3OD), δ_C: 14.4 (C-10), 21.1 (C-9), 22.0 (C-8), 36.7 (C-3), 40.4 (C-6), 50.8 (C-7), 52.2 (C-1), 54.3 (C-4), 63.5 (C-6′), 72.2 (C-4′), 76.1 (C-5), 76.6 (C-2′), 78.4 (C-3′), 78.8 (C-5′), 86.8 (C-2), 106.6 (C-1′). Positive FAB-MS: m/z 333 $(M+H)^+$, (glycerol matrix), 339 $(M+Li)^+$ (glycerol matrix + LiCl).

Acetylation of 6-Gingesulfonic Acid (1) Giving the Monoacetate (1a) A solution of **1** (40 mg) in pyridine (0.5 ml) was treated with Ac_2O (0.5 ml) and the whole was stirred at room temperature under an N_2 atmosphere for 12 h. The reaction mixture was poured into ice-water and the whole was extracted with AcOEt. The AcOEt extract was washed with diluted aqueous HCl, aqueous saturated $NaHCO_3$, and brine, and then dried over $MgSO_4$. After removal of the solvent from the AcOEt extract under reduced pressure, the product was purified by silica gel column chromatography [$CHCl_3$–MeOH–H_2O (65 : 35 : 10, lower layer)] to furnish **1a** (38 mg).

6-Gingesulfonic Acid Monoacetate (**1a**): A white powder, $[\alpha]_D^{20}$ −0.5° (c = 10.0, MeOH). IR ν_{max}^{KBr} cm^{-1}: 1770, 1720, 1512, 1219, 1200, 1050. ^{1}H-NMR (270 MHz, CD_3OD) δ: 0.87 (3H, t, J = 7 Hz, 10-H_3), 1.11—1.59 (6H, m, 7, 8, 9-H_2), 1.48 (1H, m), 1.92 (1H, m) (6-H_2), 2.22 (3H, s, OAc), 2.52 (1H, dd, J = 6, 17 Hz, 4-H), 2.86 (4H, m, 1, 2-H_2), 3.04 (1H, dd, J = 6, 17 Hz, 4-H), 3.30 (1H, m, 5-H), 3.77 (3H, s, –OCH_3), 6.77 (1H, dd, J = 2, 8 Hz, 6′-H), 6.88 (1H, d, J = 8 Hz, 5′-H), 6.93 (1H, d, J = 2 Hz, 2′-H). ^{13}C-NMR (67.5 MHz, CD_3OD) δ_C: 20.5, 171.0 (acetyl-C) and the other signals given in Table I.

Sulfonation of 6-Shogaol (7) Giving (±)-1 A solution of **7** (70 mg, 0.25 mmol) in methanol (3 ml) was treated with $NaHSO_3$ (520 mg, 5 mmol) and *tert*-butyl perbenzoate (200 μl, 1.05 mmol) and the whole was stirred at 80 °C for 36 h. After removal of the solvent from the reaction mixture, the product was purified by silica gel column chromatography [$CHCl_3$–MeOH–H_2O (10 : 3 : 1, lower layer)] to furnish (±)-**1** (82 mg, 90.3%). (±)-**1**, thus obtained, was identical, by comparisons of TLC behavior [$CHCl_3$–MeOH–H_2O (65 : 35 : 10, lower layer)] and ^{1}H-NMR (CD_3OD) and ^{13}C-NMR (CD_3OD) spectra, with authentic 6-gingesulfonic acid (**1**) except for the specific rotation: (±)-**1**, $[\alpha]_D^{20}$ 0.0° (c = 8.0, MeOH).

Alkaline Treatment of Gingerglycolipid A (2) A solution of **2** (15 mg) in 2% NaOMe–MeOH (1 ml) was heated under reflux for 30 min under an N_2 atmosphere. After cooling, the reaction mixture was neutralized with Dowex 50W × 8 (H^+ form) and filtered to remove the resin. After removal of the organic solvent from the filtrate under reduced pressure, the residue was partitioned into AcOEt–H_2O. Removal of the solvent from the AcOEt-soluble portion gave methyl linolenate, which was identical with an authentic sample on GLC. GLC conditions: column, CBJWAX-M30-025; 0.25 mm (i.d.) × 30 m capillary column; column temperature 140 °C; He flow rate 15 ml/min, t_R: 41 min 47 s. After removal of the solvent from the water-soluble portion, the residue was purified by silica gel column chromatography [$CHCl_3$–MeOH–H_2O (6 : 4 : 1)] to give **8**, which was identified by comparison of $[\alpha]_D$ and ^{13}C-NMR (D_2O) data with reported values.[12]

Alkaline Treatment of Gingerglycolipid B (3) Compound **3** (13 mg) was treated with 2% NaOMe–MeOH (1 ml) under the same conditions as described for the alkaline treatment of **2**. The resulting AcOEt-soluble portion was analyzed by GLC to identify methyl linoleate (t_R: 32 min 46 s the same GLC conditions as described for methyl linolenate). The resulting water-soluble portion was purified by silica gel column chromatography [$CHCl_3$–MeOH–H_2O (6 : 4 : 1)] to give **8**, which was identified in the same way as described above.

Alkaline Treatment of Gingerglycolipid C (4) Compound **4** (20 mg) was treated with 2% NaOMe–MeOH (1 ml) under the same conditions as described above. From the resulting AcOEt-soluble portion and water-soluble portion, methyl oleate (t_R: 27 min 47 s, under the same GLC conditions as above) and **8** (the same conditions as above) were identified, respectively.

(+)-5-Ketoborneol (10) A solution of (+)-borneol (**9**, 10.0 g) in pyridine (10 ml) was treated with Ac_2O (8 ml) and the whole mixture was stirred at room temperature under an N_2 atmosphere for 12 h. The reaction mixture was poured into ice-water and the whole was extracted with AcOEt. After work-up of the AcOEt extract, a residue was purified by silica gel column chromatography [*n*-hexane–AcOEt (30 : 1)] to give (+)-2-*O*-acetylborneol (10.5 g). A solution of (+)-2-*O*-acetylborneol (9.0 g) in AcOH (150 ml) was treated with CrO_3 (22.9 g) and the whole mixture was heated under reflux for 2 h. After cooling, the reaction mixture was diluted with water and then extracted with ether. The ether extract was washed with 5% NaOH and aqueous saturated $NaHCO_3$, and then dried over Na_2SO_4 powder. After removal of the solvent from the ether extract under reduced pressure, the residue was purified by silica gel column chromatography [*n*-hexane–AcOEt (4 : 1)] to furnish (+)-2-*O*-acetyl-5-ketoborneol[15] (3.71 g), colorless oil. IR ν_{max}^{KBr} cm^{-1}: 1748, 1242. ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 0.96, 1.02, 1.03 (3H each, all s, $CH_3 \times 3$), 1.32 (1H, dd, J=4, 15 Hz, 3-H), 2.00 (1H, dd, J=2, 19 Hz, 6-H), 2.19 (1H, d, J=5 Hz, 4-H), 2.55 (1H, d, J=19 Hz, 6-H), 2.64 (1H, ddd, J=5, 10, 15 Hz, 3-H), 5.08 (1H, ddd, J=2, 4, 10 Hz, 2-H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ_C: 12.7 (C-10), 17.5 (C-9), 20.0 (C-8), 21.0 (OAc), 31.8 (C-3), 41.9 (C-6), 47.0 (C-7), 49.4 (C-1), 59.7 (C-4), 77.5 (C-2), 170.9 (OAc), 216.0 (C-5).

A solution of (+)-2-*O*-acetyl-5-ketoborneol (1.0 g) in 1% NaOMe–MeOH (14.5 ml) was stirred at room temperature for 1 h. The reaction mixture was neutralized with Dowex 50W × 8 (H^+ form) and then filtered. Removal of the solvent from the filtrate furnished crude **10**, which was crystallized from *n*-hexane–AcOEt to yield pure crystals of **10** (470 mg). **10** was identified by comparison of its melting point, and IR (KBr), and ^{1}H-NMR ($CDCl_3$) data with reported values.[15]

Glycosidation of (+)-5-Ketoborneol (10) Giving 11 In the presence of Ag_2CO_3 (689 mg, 2.5 mmol), a solution of **10** (105 mg, 0.63 mmol) in dry benzene (5 ml) was treated with 1-bromo-2,3,4,6-tetra-*O*-acetyl-D-glucopyranose (1.00 g, 2.5 mmol) and the whole mixture was heated under reflux for 6 h. The reaction mixture was then passed through a Celite column to remove inorganic material. The eluate was evaporated under reduced pressure to give a residue, which was purified on a silica gel column [benzene–acetone (6 : 1)] to furnish **11** (201 mg, 80.6%).

11: A white powder. IR ν_{max}^{KBr} cm^{-1}: 1755, 1735, 1225, 1038. ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 0.93, 0.97, 0.99 (3H each, all s, $CH_3 \times 3$), 1.60 (1H, dd, J=4, 15 Hz, 3-H), 1.85 (1H, dd, J=2, 19 Hz, 6-H), 2.01, 2.03, 2.04, 2.09 (3H each, all s, OAc × 4), 2.10 (1H, d, J=6 Hz, 4-H), 2.40 (1H, ddd, J=6, 10, 15 Hz, 3-H), 2.50 (1H, d, J=19 Hz, 6-H), 3.69 (1H, ddd, J=2, 4, 10 Hz, 2-H), 3.95 (1H, ddd, J=2, 5, 10 Hz, 5'-H), 4.12 (1H, dd, J=2, 12 Hz, 6'-H), 4.22 (1H, dd, J=5, 12 Hz, 6'-H), 4.56 (1H, d, J=8 Hz, 1'-H), 4.99 (1H, dd, J=8, 10 Hz, 2'-H), 5.06 (1H, t-like, 4'-H), 5.20 (1H, dd, J=10, 10 Hz, 3'-H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ_C: 12.7 (C-10), 17.4 (C-9), 19.9, 20.5, 20.6 (acetyl methyl), 20.7 (C-8), 32.2 (C-3), 41.5 (C-6), 46.4 (C-7), 50.1 (C-1), 59.8 (C-4), 61.8 (C-6'), 68.3 (C-4'), 71.3 (C-2'), 71.6 (C-5'), 72.5 (C-3'), 85.0 (C-2), 102.0 (C-1'), 169.2, 169.3, 170.2, 170.5 (OAc × 4), 216.0 (C-5).

Treatment of 11 with $NaBH_4$ Giving 5 A solution of **11** (860 mg) in MeOH (10 ml) was treated with $NaBH_4$ (1.31 g) and the whole mixture was stirred at room temperature for 30 min. The reaction mixture was neutralized with Dowex 50W × 8 (H^+ form) and then filtered. After removal of the solvent from the filtrate, the residue was purified by reversed-phase silica gel column chromatography [MeOH–H_2O (1 : 2)] to furnish **5** (352 mg), which was identical, on the basis of $[\alpha]_D$ (MeOH), TLC [$CHCl_3$–MeOH–H_2O, 65 : 35 : 10 (lower layer), ^{1}H-NMR (CD_3OD), and ^{13}C-NMR (CD_3OD) comparisons, with the natural **5** isolated from Zingiberis Rhizoma.

Inhibitory Effect of 6-Gingesulfonic Acid (1) on HCl/Ethanol-Induced Gastric Lesions The effect on HCl/ethanol-induced gastric lesions was examined as follows: male Wistar rats (weighing approximately 250 g, Kitayama Labs) were divided into groups of 5—7 rats. After 24 h of fasting, they were given the test drug orally. One hour thereafter, 1 ml of HCl/ethanol solution (150 mM HCl in 60% ethanol) was administered orally to each rat. Rats were then killed with ether 1 h after the administration of the necrotizing agent, and the stomachs were excised. Followed by treatment of the stomach in 2% formalin solution, the lesion index was calculated as the sum of the lengths (cm) of lesions in the mucous membrane of the fundus region. Cetraxate (Daiichi Pharmaceutical) was used as the reference drug. The results for 6-gingesulfonic acid (**1**), 6-gingerol (**6**), and 6-shogaol (**7**) are summarized in Table III.

Sensory Evaluation for Pungent Effect of 6-Gingesulfonic Acid (1) The pungent effect of **1** was evaluated by a human sensory panel. All samples (0.03 mmol) was dissolved in ethanol (1 ml)–water (2 ml) and then diluted with water to prepare graduated concentrations from 1000 μmol/l to 0.1 μmol/l. All samples were evaluated by six panelists by the same procedure as described in the previous paper.[1] The result for **1** in comparison with **6** and **7** is given in Table IV.

References and Notes

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9) This work was partly reported in our preliminary communication: M. Yoshikawa, S. Hatakeyama, K. Taniguchi, H. Matsuda, J. Yamahara, *Chem. Pharm. Bull.*, **40**, 2239 (1992).
10) The assignments for the ^{13}C-NMR data of **1**, **1a**, and **6** were confirmed by using ^{1}H–^{1}H COSY and ^{1}H–^{13}C COSY, except for the assignments of C-7 and C-8. The assignments for C-2 and C-4 in the previous communication[9] were exchanged.
11) Although **1**, obtained from Zingiberis Rhizoma, shows a little optical activity ($[\alpha]_D$ $-1.0°$), the absolute stereostructure at C-5 or the optical purity of **1** have not yet been elucidated.
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