

Phytochemical Constituents of *Schizonepeta tenuifolia* Briquet

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Abstract – Column chromatographic separation of the MeOH extract from the aerial parts of *Schizonepeta tenuifolia* Briquet led to the isolation of twelve terpenes (**1** - **11** and **17**), four phenolics (**13** - **16**) and a hexenyl glucoside (**12**). Their structures were determined by spectroscopic means to be (–)-pulegone (**1**), piperitenone (**2**), *p*-cymene-3,8-diol (**3**), schizonepetoside A (**4**), schizonepetoside C (**5**), (+)-spatulenol (**6**), ursolic acid (**7**), 2 α ,3 α ,24 α -trihydroxyolean-12-en-28oic acid (**8**), 5 α ,8 α -epidioxyergosta-6,22-diol-3 β -ol (**9**), stigmast-4-en-3-one (**10**), β -sitosterol (**11**), (*Z*)-3-hexenyl-1-*O*- β -D-glucopyranoside (**12**), rosmarinic acid (**13**), apigenin-7-*O*- β -D-glucopyranoside (**14**), luteolin-7-*O*- β -D-glucuronopyranoside (**15**), hesperidin (**16**) and *trans*-phytol (**17**). Compounds **2**, **3**, **8**, **9** and **12** were for the first time isolated from *S. tenuifolia* Briq.

Key word – *Schizonepeta tenuifolia* Briquet, Terpenoids, Phenolics

Introduction

Schizonepeta tenuifolia Briq. has been used as a Korean traditional medicine for anti-inflammatory (Yamahara *et al.*, 1980), anti-diabetic (Kim *et al.*, 1996), anti-puritic (Tohda *et al.*, 2000) and fumigant (Park *et al.*, 2006). Monoterpenes (Masayoshi *et al.*, 1986), triterpenes and flavonoids (Hu *et al.*, 2006) were reported from *S. tenuifolia* Briq. As part of our systematic study on the genus *Schizonepeta* of Korea Labiatae plants, we have investigated the constituents of *S. tenuifolia* Briq.

The column chromatographic separation of the MeOH extract (180 g) of this plant resulted in the isolation of twelve terpenes (**1** - **11** and **17**), four phenolics (**13** - **16**) and one hexenyl glucoside (**12**). Their structures were identified by spectroscopic methods. Compounds **2**, **3**, **8**, **9** and **12** were for the first time isolated from the *S. tenuifolia* Briq.

Experimental

General – Melting points were determined on Gallenkamp melting point apparatus and uncorrected. Optical rotations were measured on a JASCO P-1020 Polarimeter. NMR spectra were recorded on Varian UNITY INOVA 500 NMR spectrometer. FAB-MS data

were obtained on a Agilent 1100 mass spectrometer. GC-MS data were obtained on a Agilent 6890N. Preparative HPLC used a Wellchrom K1001 A pump with Knauer Dual Detector and Apollo Silica 5 μ m column (250 \times 22 mm) or Econosil[®] RP-18 10 μ m column (250 \times 22 mm). Silica gel 60 (Merck, 70 ~ 230 mesh and 230 ~ 400 mesh) was used for column chromatography. TLC used Merck precoated Silica gel F₂₅₄ plates and RP-18 F_{254s} plates. Packing material of molecular sieve column chromatography was Sephadex LH-20 (Pharmacia Co.). Low pressure liquid chromatography was carried out over Merck LiChrorep Lobar[®]-A Si 60 (240 \times 10 mm) or LiChrorep Lobar[®]-A RP-18 (240 \times 10 mm) column with FMI QSY-0 pump (ISCO).

Plant materials – The aerial parts of *S. tenuifolia* Briq. (2 kg) were collected at Yeongcheon, Gyeongbuk Province, Korea in August, 2006. A voucher specimen (SKKU-2007-1) was deposited at the College of Pharmacy in Sungkyunkwan University, Korea.

Extraction and Isolation – The half dried and chopped aerial parts of *S. tenuifolia* Briq. (2 kg) were extracted with 80% MeOH at room temperature and evaporated under reduced pressure to give residue (180 g), which was dissolved in water (800 ml) and solvent partitioned to give *n*-hexane fraction (8 g), EtOAc fraction (18 g), and *n*-BuOH fraction (20 g), respectively.

The *n*-hexane fraction (8 g) was chromatographed over a silica gel column with *n*-hexane : EtOAc (10 : 1) as the

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eluent to give nine fractions (STH1-STH9). The fraction STH2 (100 mg) was subjected to a Sephadex LH-20 (methylene chloride : methanol = 1 : 1) and purified with silica gel prep. HPLC (Apollo Silica 5 μ column, 250 \times 22 mm; *n*-hexane : EtOAc = 7 : 1) to yield compound **1** (23 mg). The fraction STH4 (470 mg) was also subjected to a RP-C₁₈ silica gel column with 95% MeOH as the eluent and purified with silica gel prep. HPLC (*n*-hexane : EtOAc = 7 : 1) to yield compounds **2** (5 mg) and **5** (12 mg). The fraction STH5 (700 mg) was also subjected to a RP-C₁₈ silica gel column with 95% MeOH as the eluent and purified with silica gel prep. HPLC (*n*-hexane : EtOAc = 7 : 1) to yield compounds **17** (45 mg) and **3** (6 mg). The fraction STH7 (700 mg) was also subjected to a RP-C₁₈ silica gel column with 90% MeOH as the eluent and purified with silica gel prep. HPLC (*n*-hexane : EtOAc = 2.5 : 1) to yield compound **9** (11 mg). The fraction STH8 (220 mg) was also subjected to a RP-C₁₈ silica gel column with 80% MeOH as the eluent and purified with silica gel prep. HPLC (*n*-hexane : EtOAc = 7.5 : 1) to yield compound **10** (7 mg). The fraction STH9 (1.5 g) was also subjected to a Sephadex LH-20 (methylene chloride : methanol = 1 : 1), silica Lobar A[®]-column (*n*-hexane : EtOAc = 4 : 1) as the eluent and purified with silica gel prep. HPLC (*n*-hexane : Chloroform : EtOAc = 6 : 2 : 1) to yield compound **11** (15 mg).

The EtOAc fraction (18 g) was chromatographed over a silica gel column with *n*-hexane : chloroform : EtOAc (6 : 2 : 5) as the eluent to give nine fractions (STE1-STE7). The fraction STE2 (2.9 g) was also subjected to a RP-C₁₈ silica gel column with 70% MeCN as the eluent and purified by RP-C₁₈ prep. HPLC (Econosil[®] RP-18 10 μ column, 250 \times 22 mm; 65% MeOH), silica gel prep. HPLC (*n*-hexane : chloroform : EtOAc = 4 : 7 : 0.3) to yield compound **7** (20 mg). The fraction STE5 (1.0 g) was also subjected to a RP-C₁₈ column with 45% MeCN as the eluent and purified by silica gel prep. HPLC (*n*-hexane : methylene chloride : methanol = 3 : 10 : 1) to yield compound **8** (22 mg). The fraction STE6 (1.9 g) was subjected to a Sephadex LH-20 (methylene chloride : methanol = 1 : 1) and purified by RP C-18 Lobar A[®]-column (40% MeOH) to yield compound **14** (13 mg). The fraction STE7 (220 mg) was also subjected to a RP C-18 column with 30% MeOH as the eluent and purified by silica gel prep. HPLC (*n*-hexane : methylene chloride : methanol = 5 : 7 : 5) to furnish compound **13** (520 mg). The *n*-BuOH fraction (20 g) was chromatographed over a HP-20 column with solvent system of 100% MeOH and 100% H₂O as the eluent to give two fractions (STB1 ~ STB2). The fraction STB2 (12 g) was subfractionated

with silica gel column with (chloroform : methanol : water = 13 : 4 : 0.3) as the eluent to give nine fractions (STB21-STB29). The fraction STB22 (480 mg) was also subjected to a RP C-18 column with 20% MeCN as the eluent and purified by RP-C₁₈ prep. HPLC (40% MeOH), silica gel prep. HPLC (chloroform : methanol = 10 : 1) to give compounds **5** (30 mg), **12** (13 mg) and **4** (20 mg). The fraction STB26 (700 mg) was also subjected to a RP C-18 column with 50% MeOH as the eluent and purified by RP-C₁₈ prep. HPLC (25% MeCN) to give compound **16** (430 mg). The fraction STB27 (4.2 g) was also subjected to a RP C-18 column with 30% MeOH as the eluent and purified by RP-C18 prep. HPLC (25% MeOH) to yield compound **15** (230 mg).

(-)-Pulegone (1) – Yellowish oil. $[\alpha]_D -25.7^\circ$ (*c* 1.05, CHCl₃). IR ν_{max} cm⁻¹: 1741, 1059, 1019, 678. GC-MS *m/z*: 152 [M]⁺. ¹H-NMR (CDCl₃, 500 MHz): δ 0.97 (3H, d, *J* = 6.5 Hz, H-7), 1.76 (3H, s, H-9), 1.96 (3H, s, H-10). ¹³C-NMR (CDCl₃, 125 MHz): δ 22.8 (C-10), 22.2 (C-9), 23.1 (C-7), 28.7 (C-5), 31.6 (C-1), 32.9 (C-6), 50.9 (C-2), 131.3 (C-4), 141.9 (C-8), 204.3 (C-3).

(+)-Piperitenone (2) – Colorless oil. UV λ_{max} (CHCl₃, log ϵ): 274 (4.76) nm. IR ν_{max} cm⁻¹: 1741, 1059, 1019, 678. GC-MS *m/z*: 150 [M]⁺. ¹H-NMR (CDCl₃, 500 MHz): δ 1.88 (3H, s, H-10), 1.95 (3H, s, H-9), 2.12 (3H, s, H-8), 2.26 (2H, dd, *J* = 12.5, 6.5 Hz, H-5), 2.68 (2H, dd, *J* = 12.5, 6.5 Hz, H-6), 5.91 (1H, br.s, H-2). ¹³C-NMR (CDCl₃, 125 MHz): δ 22.7 (C-10), 22.9 (C-9), 24.0 (C-7), 28.2 (C-6), 32.1 (C-5), 129.0 (C-2), 129.6 (C-8), 142.7 (C-1), 159.8 (C-4), 191.9 (C-3).

***p*-Cymene-3,8-diol (3)** – White powder. m.p 60–62 °C. $[\alpha]_D +1.6^\circ$ (*c* 0.19, CHCl₃). UV λ_{max} (CHCl₃, log ϵ): 277 (4.40) nm. GC-MS *m/z*: 166 [M]⁺. ¹H-NMR (CD₃OD, 500 MHz): δ 1.68 (6H, s, H-9, H-10), 2.29 (3H, s, H-7), 6.65 (1H, d, *J* = 8.0 Hz, H-6), 6.72 (1H, s, H-2), 6.97 (1H, d, *J* = 8.0 Hz, H-5). ¹³C-NMR (CD₃OD, 125 MHz): δ 21.1 (C-7), 30.6 (C-10), 30.6 (C-9), 76.2 (C-8), 118.3 (C-2), 120.5 (C-6), 125.4 (C-5), 128.2 (C-1), 139.3 (C-4), 155.7 (C-3).

Schizonepetoside A (4) – Colorless needle. $[\alpha]_D -13.5^\circ$ (*c* 0.10, MeOH). IR ν_{max} cm⁻¹: 3286, 1569, 1479, 1124, 1034. FAB-MS *m/z*: 329 [M – H]⁻. ¹H-NMR (Pyridine-*d*₅, 500 MHz): δ 0.85 (3H, d, *J* = 6.5 Hz, H-7), 1.86 (3H, br.s, H-10), 5.14 (1H, d, *J* = 7.5 Hz, H-1'), 6.55 (1H, br.s, H-9). ¹³C-NMR (Pyridine-*d*₅, 125 MHz): δ 12.1 (C-10), 22.3 (C-7), 31.4 (C-5), 33.9 (C-6), 35.0 (C-1), 50.5 (C-2), 54.2 (C-4), 62.5 (C-6'), 71.3 (C-4''), 74.9 (C-2'), 78.4 (C-5'), 79.0 (C-3'), 104.9 (C-1'), 113.6 (C-8), 142.0 (C-9), 209.6 (C-3).

Schizonepetoside C (5) – Colorless needle. $[\alpha]_D -24.4^\circ$

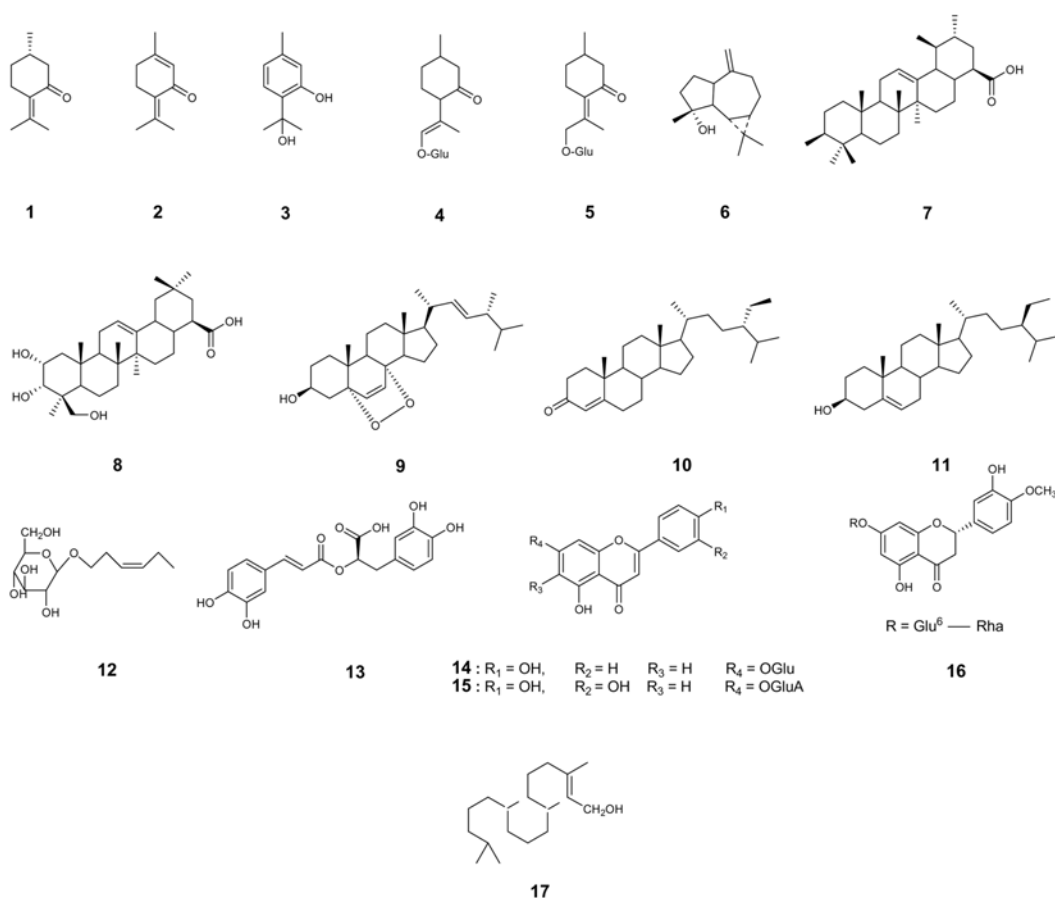


Fig. 1. The Structures of 1 - 17 from of *S. tenuifolia* Briq.

(*c* 0.18, MeOH). UV λ_{\max} (CHCl₃, log ϵ): 353 (4.37) nm. IR ν_{\max} cm⁻¹: 3336, 1567, 1480, 1054, 1016. FAB-MS m/z : 329 [M + Na]⁺. ¹H-NMR (Pyridine-*d*₅, 500 MHz): δ 1.03 (3H, d, *J* = 6.5 Hz, H-7), 1.94 (3H, br.s, H-10), 4.80 (1H, d, *J* = 7.5 Hz, H-1). ¹³C-NMR (Pyridine-*d*₅, 125 MHz): δ 19.0 (C-10), 22.2 (C-7), 29.3 (C-5), 32.8 (C-1), 34.0 (C-6), 52.0 (C-2), 63.3 (C-6'), 69.1 (C-9), 72.1 (C-4'), 75.7 (C-2'), 79.1 (C-5'), 79.2 (C-3'), 103.8 (C-1'), 137.0 (C-4), 138.3 (C-8), 204.6 (C-3).

(+)-**Spatulenol (6)** – Colorless oil. $[\alpha]_D^{25} +2.8^\circ$ (*c* 0.27, CHCl₃). UV λ_{\max} (CHCl₃, log ϵ): 274 (4.76) nm. GC-MS m/z : 220 [M]⁺. ¹H-NMR (CDCl₃, 500 MHz): δ 0.45 (1H, d, *J* = 9.0 Hz, H-7b), 0.47 (1H, d, *J* = 9.0 Hz, H-1a), 1.06 (3H, s, H-8), 1.07 (3H, s, H-9), 1.30 (3H, s, H-11), 4.68 (1H, s, H-10a), 4.71 (1H, s, H-10b). ¹³C-NMR (CDCl₃, 125 MHz): δ 16.6 (C-8), 20.5 (C-1), 24.9 (C-2), 26.3 (C-11), 26.9 (C-5), 27.7 (C-1a), 28.9 (C-9), 30.1 (C-7b), 39.0 (C-3), 41.9 (C-6), 53.6 (C-4a), 54.6 (C-7a), 81.2 (C-7), 106.5 (C-10), 153.8 (C-4).

Ursolic acid (7) – White powder. m.p 238 - 240 °C. $[\alpha]_D^{25} +78.3^\circ$ (*c* 0.36, EtOH). FAB-MS m/z : 455 [M + H]⁺.

¹H-NMR (Pyridine-*d*₅, 500 MHz): δ 0.91 (3H, s, H-25), 0.97 (3H, d, *J* = 6.0 Hz, H-30), 1.02 (3H, d, *J* = 7.0 Hz, H-29), 1.03 (3H, s, H-24), 1.07 (3H, s, H-26), 1.25 (3H, s, H-27), 1.26 (3H, s, H-23), 2.65 (1H, d, *J* = 11.0 Hz, H-18), 3.47 (1H, dd, *J* = 10.0, 6.0 Hz, H-3), 5.51 (1H, m, H-12). ¹³C-NMR (Pyridine-*d*₅, 125 MHz): 15.6 (C-25), 16.5 (C-24), 17.4 (C-26), 17.5 (C-26), 18.8 (C-6), 21.4 (C-30), 23.6 (C-27), 23.9 (C-29), 24.9 (C-16), 28.1 (C-2), 28.7 (C-15), 28.8 (C-23), 31.1 (C-21), 33.5 (C-7), 37.3 (C-22), 37.4 (C-10), 39.1 (C-1), 39.3 (C-20), 39.4 (C-4), 39.5 (C-19), 39.9 (C-8), 42.5 (C-14), 48.0 (C-9), 48.1 (C-17), 53.6 (C-18), 55.8 (C-5), 78.1 (C-3), 125.6 (C-12), 139.2 (C-13), 179.9 (C-28)

2 α ,3 α ,24-Trihydroxyolean-12en-28oic acid (8) – White powder. m.p 266 - 258 °C. $[\alpha]_D^{25} +108.4^\circ$ (*c* 0.16, MeOH). IR ν_{\max} cm⁻¹: 3357, 1629, 1125, 1052. FAB-MS m/z : 487 [M - H]⁻. ¹H-NMR (DMSO-*d*₆, 500 MHz): δ 0.91 (3H, s, H-30), 0.99 (3H, s, H-26), 1.01 (3H, s, H-25), 1.02 (3H, s, H-29), 1.12 (3H, s, H-27), 1.67 (3H, s, H-23), 3.31 (1H, dd, *J* = 14.0, 4.0 Hz, H-18), 3.84 (1H, d, *J* = 11.0 Hz, H-24b), 4.13 (1H, d, *J* = 11.0 Hz, H-24a), 4.47 (1H, m, H-

2), 4.60 (1H, d, $J=3.0$ Hz, H-3), 5.48 (1H, br.s, H-12). $^{13}\text{C-NMR}$ (DMSO- d_6 , 125 MHz): 17.4 (C-25), 17.7 (C-26), 19.2 (C-6), 24.0 (C-23), 24.1 (C-16), 24.2 (C-11), 24.4 (C-30), 26.6 (C-27), 28.6 (C-15), 31.2 (C-20), 33.5 (C-22), 33.5 (C-7), 34.0 (C-29), 34.5 (C-21), 39.0 (C-10), 40.3 (C-8), 42.3 (C-18), 42.5 (C-1), 43.3 (C-4), 45.5 (C-14), 46.7 (C-19), 46.9 (C-17), 48.5 (C-9), 49.8 (C-5), 65.5 (C-24), 66.5 (C-2), 74.6 (C-3), 122.8 (C-12), 145.1 (C-13), 180.5 (C-28).

5 α ,8 α -Epidioxergosta-6,22-diol-3 β -ol (9) – White powder. m.p 176 - 178 °C. $[\alpha]_D -20.0^\circ$ (c 0.22, CHCl_3). FAB-MS m/z : 427 $[\text{M} - \text{H}]^-$. $^1\text{H-NMR}$ (CDCl_3 , 500 MHz): δ 0.83 (3H, d, $J=7.0$ Hz, H-28), 0.84 (3H, s, H-18), 0.86 (3H, d, $J=7.0$ Hz, H-21), 0.90 (3H, s, H-19), 0.92 (3H, d, $J=7.0$ Hz, H-16), 1.01 (3H, d, $J=6.3$ Hz, H-17), 3.99 (1H, m, H-3), 5.20 (2H, m, H-22, H-23), 6.25 (1H, d, $J=8.6$ Hz, H-7), 6.52 (1H, d, $J=8.6$ Hz, H-6). $^{13}\text{C-NMR}$ (CDCl_3 , 125 MHz): δ 13.1 (C-19), 17.8 (C-28), 18.4 (C-19), 19.9 (C-21), 20.2 (C-26), 20.8 (C-27), 21.1 (C-11), 23.6 (C-16), 28.9 (C-15), 30.3 (C-2), 33.3 (C-25), 34.9 (C-9), 37.1 (C-10), 37.1 (C-1), 37.5 (C-12), 39.9 (C-20), 42.9 (C-24), 44.8 (C-13), 51.3 (C-4), 51.9 (C-14), 56.4 (C-17), 66.7 (C-3), 79.6 (C-5), 82.4 (C-8), 130.9 (C-7), 132.5 (C-23), 135.4 (C-22), 135.6 (C-6).

Stigmast-4-en-3-one (10) – White powder. $[\alpha]_D +39.8^\circ$ (c 0.17, CHCl_3). IR ν_{max} cm^{-1} : 1563, 1484, 1129, 1048. FAB-MS m/z : 413 $[\text{M} + \text{H}]^+$. $^1\text{H-NMR}$ (CDCl_3 , 500 MHz): δ 0.73 (3H, s, H-18), 0.83 (3H, d, $J=6.5$ Hz, H-27), 0.84 (3H, d, $J=6.5$ Hz, H-26), 0.85 (3H, t, $J=7.5$ Hz, H-29), 0.93 (3H, d, $J=6.5$ Hz, H-21), 1.20 (3H, s, H-19), 5.74 (1H, br.s, H-4). $^{13}\text{C-NMR}$ (CDCl_3 , 125 MHz): δ 12.1 (C-29), 12.2 (C-18), 17.6 (C-19), 18.9 (C-21), 19.2 (C-27), 20.0 (C-26), 21.2 (C-11), 23.3 (C-28), 24.4 (C-15), 26.3 (C-23), 28.4 (C-16), 29.4 (C-25), 32.3 (C-7), 33.2 (C-6), 34.1 (C-2), 34.2 (C-22), 35.8 (C-8), 35.9 (C-1), 36.3 (C-20), 38.8 (C-10), 39.8 (C-12), 42.6 (C-13), 46.0 (C-24), 54.0 (C-9), 56.1 (C-14), 56.2 (C-17), 124.0 (C-4), 171.9 (C-5), 199.9 (C-3).

β -Sitosterol (11) – White powder. m.p 283 - 285 °C. FAB-MS m/z : 437 $[\text{M} + \text{Na}]^+$. $^1\text{H-NMR}$ (CDCl_3 , 500 MHz): δ 0.70 (3H, s, H-18), 0.84 (3H, t, $J=6.5$ Hz, H-29), 0.92 (3H, d, $J=6.5$ Hz, H-26), 0.95 (3H, d, $J=6.5$ Hz, H-21), 1.03 (3H, s, H-19), 3.54 (1H, m, H-3), 5.37 (1H, d, $J=5.0$ Hz, H-6). $^{13}\text{C-NMR}$ (CDCl_3 , 125 MHz): δ 12.0 (C-29), 12.1 (C-18), 19.0 (C-19), 19.6 (C-21), 19.7 (C-26), 20.0 (C-27), 20.4 (C-11), 23.4 (C-28), 24.5 (C-15), 26.4 (C-23), 28.5 (C-16), 29.4 (C-25), 32.1 (C-8), 32.1 (C-2), 34.2 (C-22), 36.4 (C-20), 36.4 (C-20), 36.8 (C-10), 37.5 (C-1), 40.0 (C-12), 42.5 (C-13), 46.0 (C-4), 46.1 (C-24), 50.4 (C-9), 56.3 (C-17), 56.9 (C-14), 72.0 (C-3), 121.9

(C-6), 141.0 (C-5).

(Z)-3-Hexenyl-1-O- β -D-glucopyranoside (12) – Colorless needle. $[\alpha]_D -31.0^\circ$ (c 0.17, MeOH). AB-MS m/z : 285 $[\text{M} + \text{Na}]^+$. $^1\text{H-NMR}$ (Pyridine- d_5 , 500 MHz): δ 0.98 (3H, t, $J=7.5$ Hz, H-6), 2.09 (2H, quint, $J=7.5$ Hz, H-5), 2.40 (2H, q, $J=7.0$ Hz, H-2), 3.29 (2H, d, $J=8.4$ Hz, H-1), 3.32 (1H, quint, $J=1.7$ Hz, H-4'), 3.36 (1H, t, $J=9.0$ Hz, H-2'), 3.56 (1H, m, H-5'), 3.68 (1H, dd, $J=11.5, 5.5$ Hz, H-3'), 3.88 (1H, dd, $J=11.5, 1.5$ Hz, H-6'b), 4.55 (1H, dd, $J=11.5, 4.5$ Hz, H-6'a), 4.86 (1H, d, $J=8.0$ Hz, H-1'), 5.39 (1H, dddd, $J=17.0, 12.0, 7.0, 2.0$ Hz, H-4), 5.47 (1H, dddd, $J=17.0, 12.0, 7.0, 2.0$ Hz, H-3). $^{13}\text{C-NMR}$ (Pyridine- d_5 , 125 MHz): δ 14.9 (C-6), 21.3 (C-5), 28.9 (C-2), 63.3 (C-6'), 69.9 (C-1), 72.2 (C-4'), 75.7 (C-2'), 79.0 (C-5'), 79.1 (C-3'), 105.2 (C-1'), 126.0 (C-4), 134.0 (C-3).

Rosmarinic acid (13) – Yellow powder. m.p 207 °C. UV λ_{max} (CHCl_3 , $\log \epsilon$): 327 (5.19), 290 (5.11) nm. IR ν_{max} cm^{-1} : 3286, 1576, 1121, 1054, 1033. FAB-MS m/z : 359 $[\text{M} - \text{H}]^-$. $^1\text{H-NMR}$ (CD_3OD , 500 MHz): δ 7.51 (1H, d, $J=16.0$ Hz, H-7), 7.03 (1H, d, $J=2.0$ Hz, H-2), 6.91 (1H, dd, $J=8.0, 2.0$ Hz, H-6), 6.77 (1H, d, $J=8.0$ Hz, H-5), 6.76 (1H, d, $J=8.0$ Hz, H-2), 6.68 (1H, d, $J=8.0$ Hz, H-5), 6.63 (1H, dd, $J=8.0, 2.0$ Hz, H-6), 6.27 (1H, d, $J=16.0$ Hz, H-8), 5.09 (1H, dd, $J=10.0, 4.0$ Hz, H-8), 3.10 (1H, dd, $J=14.0, 4.0$ Hz, H-7a), 2.94 (1H, dd, $J=14.0, 10.0$ Hz, H-7b). $^{13}\text{C-NMR}$ (CD_3OD , 125 MHz): δ 38.8 (C-7'), 77.8 (C-8'), 115.1 (C-2), 115.6 (C-8), 116.2 (C-5'), 116.5 (C-5), 117.5 (C-2'), 121.7 (C-6'), 122.9 (C-6), 127.9 (C-1), 131.2 (C-1'), 144.7 (C-4'), 145.9 (C-3'), 146.6 (C-7), 146.7 (C-3), 149.4 (C-4), 169.2 (C-9), 177.8 (C-9').

Apigenin-7-O- β -D-glucopyranoside (14) – Yellow powder. m.p 236 - 238 °C. UV λ_{max} (MeOH, $\log \epsilon$): 333 (5.08), 286 (5.03) nm. IR ν_{max} cm^{-1} : 3332, 1569, 1481, 11245. FAB-MS m/z : 433 $[\text{M} + \text{H}]^+$. $^1\text{H-NMR}$ (DMSO- d_6 , 500 MHz): δ 3.51 (1H, m, H-6''), 3.73 (1H, m, H-6''), 3.20 - 3.40 (3H, m, H-2'', 3'', 4''), 5.08 (1H, d, $J=7.5$ Hz, H-1''), 6.45 (1H, d, $J=2.0$ Hz, H-6), 6.83 (1H, d, $J=2.0$ Hz, H-8), 6.87 (1H, s, H-3), 6.94 (2H, d, $J=8.5$ Hz, H-3', H-5'), 7.95 (2H, d, $J=8.5$ Hz, H-2', H-6'), 12.97 (1H, br.s, 5-OH). $^{13}\text{C-NMR}$ (DMSO- d_6 , 125 MHz): δ 56.4 (–OCH₃), 92.0 (C-8), 104.9 (C-3), 107.0 (C-10), 115.4 (C-3'), 124.3 (C-1'), 129.0 (C-6'), 129.0 (C-2'), 132.7 (C-6), 148.2 (C-5), 150.9 (C-9), 155.7 (C-7), 163.4 (C-4'), 164.5 (C-2), 183.7 (C-4).

Luteolin-7-O- β -D-glucuronopyranoside (15) – Yellow powder. m.p 187 - 189 °C. $[\alpha]_D -48.0^\circ$ (c 0.08, MeOH). UV λ_{max} (MeOH, $\log \epsilon$): 339 (5.31), 269 (5.24), 239 (5.08) nm. FAB-MS m/z : 461 $[\text{M} - \text{H}]^-$. $^1\text{H-NMR}$ (DMSO-

d_6 , 500 MHz): δ 3.61 (1H, d, $J = 9.0$ Hz, H-5''), 4.89 (1H, d, $J = 6.5$ Hz, H-2''), 4.91 (2H, d, $J = 7.0$ Hz, H-1''), 6.15 (1H, d, $J = 1.0$ Hz, H-7), 6.50 (1H, br.s, H-3), 6.72 (1H, br.s, H-9), 6.90 (1H, d, $J = 8.0$ Hz, H-5'), 7.43 (1H, d, $J = 1.5$ Hz, H-2'), 7.59 (1H, dd, $J = 8.0, 1.5$ Hz, H-6'). $^{13}\text{C-NMR}$ (DMSO- d_6 , 125 MHz): δ 72.1 (C-4''), 73.3 (C-2''), 74.1 (C-5''), 75.9 (C-3''), 94.1 (C-8), 98.9 (C-6), 102.7 (C-1''), 102.8 (C-3), 103.4 (C-10), 115.9 (C-2'), 116.9 (C-5'), 121.0 (C-1'), 122.2 (C-6'), 146.0 (C-3'), 152.3 (C-4'), 157.2 (C-9), 161.3 (C-5), 163.3 (C-7), 164.7 (C-2), 172.5 (C-6''), 181.6 (C-4).

Hesperidine (16) – Yellow powder. m.p 269 - 270 °C. UV λ_{max} (CHCl₃, log ϵ): 327 (4.50), 284 (5.24) nm. IR ν_{max} cm⁻¹: 3455, 1740, 1642, 1368, 1215, 1061. FAB-MS m/z : 611 [M + H]⁺. $^1\text{H-NMR}$ (DMSO- d_6 , 500 MHz): δ 0.87 (3H, d, $J = 6.0$ Hz, H-6''), 2.79 (2H, dd, $J = 17.0, 3.0$ Hz, H-3), 4.53 (1H, s, H-5''), 4.97 (2H, d, $J = 7.5$ Hz, H-6''), 5.50 (1H, dd, $J = 12.0, 3.0$ Hz, H-2), 6.11 (1H, d, $J = 2.0$ Hz, H-7), 6.15 (1H, d, $J = 2.0$ Hz, H-9), 6.87 (1H, dd, $J = 8.5, 2.0$ Hz, H-6'), 6.92 (1H, d, $J = 2.0$ Hz, H-2'), 6.94 (1H, d, $J = 8.5$ Hz, H-5'), 9.06 (1H, br.s, 3'-OH), 12.01 (1H, br.s, 5-OH). $^{13}\text{C-NMR}$ (DMSO- d_6 , 125 MHz): δ 72.1 (C-4''), 73.3 (C-2''), 74.1 (C-5''), 75.9 (C-3''), 94.1 (C-8), 98.9 (C-6), 102.7 (C-1''), 102.8 (C-3), 103.4 (C-10), 115.9 (C-2'), 116.9 (C-5'), 121.0 (C-1'), 122.2 (C-6'), 146.0 (C-3'), 152.3 (C-4'), 157.2 (C-9), 161.3 (C-5), 163.3 (C-7), 164.7 (C-2), 172.5 (C-6''), 181.6 (C-4).

trans-Phytol (17) – Colorless oil. $[\alpha]_{\text{D}}^{20} +4.8^\circ$ (c 0.2, CHCl₃). GC-MS m/z : 296 [M]⁺. $^1\text{H-NMR}$ (CDCl₃, 500 MHz): δ 0.90 (12H, d, $J = 7.0$ Hz, H-7a, 11a, 15a, 16), 1.10 - 1.60 (20H, m), 1.70 (3H, s, H-3a), 2.06 (2H, m), 4.49 (2H, d, $J = 6.5$ Hz, H-1), 5.78 (1H, t, $J = 6.5$ Hz, H-2). $^{13}\text{C-NMR}$ (CDCl₃, 125 MHz): δ 16.7 (C-3a), 20.4 (C-7a), 20.4 (C-11a), 23.2 (C-15a), 23.3 (C-16), 25.3 (C-9), 25.6 (C-13), 26.0 (C-5), 28.7 (C-15), 33.4 (C-7), 33.5 (C-11), 37.4 (C-6), 38.0 (C-12), 38.1 (C-10), 38.2 (C-8), 40.1 (C-14), 40.7 (C-4), 59.4 (C-1), 126.8 (C-2), 137.4 (C-3).

Results and Discussion

The column chromatographic separation of the MeOH extract of *S. tenuifolia* Briq. led to the isolation of twelve terpenoids (**1 - 11** and **17**), hexenyl glucoside (**12**) and four phenolic compounds (**13 - 16**).

The structures of **1**, **4 - 7**, **10 - 11** and **13 - 17** were identified to be (-)-pulegone (**1**) (Madyastha *et al.*, 1999), schizonepetoside A (**4**) (Kubo *et al.*, 1986), schizonepetoside C (**5**) (Kubo *et al.*, 1986), spatulenol (**6**) (Ulubelen *et al.*, 1994), ursolic acid (**7**) (Kim *et al.*, 1998), stigmast-4-en-3-one (**10**) (Kolak *et al.*, 2005), β -sitosterol (**11**)

(Chang *et al.*, 1981), rosmarinic acid (**13**) (Claire *et al.*, 2005), apigenin-7-O- β -D-glucopyranoside (**14**) (Kim *et al.*, 2004), luteolin-7-O- β -D-glucuronopyranoside (**15**) (Beninger *et al.*, 2005), hesperidine (**16**) (Panadda *et al.*, 2001) and *trans*-phytol (**17**) (James *et al.*, 1976) by comparison of ^1H -, ^{13}C -NMR and MS data with those reported in the literatures. Compounds **2**, **3**, **8**, **9** and **12** were for the first time isolated from *S. tenuifolia* Briq.

Compound **2** was obtained as yellowish oil. The IR spectrum showed absorption band for the presence of an C=O functional group at 1741 cm⁻¹. The MS spectrum of **2** showed a molecular ion peak at m/z 150 [M]⁺. The $^1\text{H-NMR}$ spectrum appeared three methyl signals at δ 1.88 (3H, s, H-10), 1.95 (3H, s, H-9) and 2.12 (3H, s, H-7). Two methylene signals appeared at δ 2.26 (2H, dd, $J = 12.5, 6.5$ Hz, H-5) and 2.68 (2H, dd, $J = 12.5, 6.5$ Hz, H-6). The $^{13}\text{C-NMR}$ spectrum showed three methyl signals at δ 24.0 (C-7), 22.9 (C-9) and 22.7 (C-10), two methylene signals at δ 32.1 (C-5) and 28.2 (C-6), three quaternary carbon signals at δ 159.8 (C-4), 142.7 (C-1) and 129.6 (C-8) and a carbonyl signal at 191.9 (C-3). Based on the above consideration and the comparison of the ^1H -, ^{13}C -NMR, IR and MS spectral data with those reported in the previous paper (Manenzhe *et al.*, 2004), the structure of **2** was identified as piperitenone (**2**). Compound **3** was obtained as white powder. The MS spectrum of **3** showed a molecular ion peak at m/z 166 [M]⁺. The $^1\text{H-NMR}$ spectrum showed two methyl signals at δ 1.68 (6H, s, H-9, H-10) and 2.29 (3H, s, H-7). The $^{13}\text{C-NMR}$ spectrum showed the presence of two methyl signals at δ 21.1 (C-10) and 30.6 (C-9, C-10) and aromatic carbon signals at δ 118.3 (C-2), 120.5 (C-6), 125.4 (C-5), 128.2 (C-1), 139.3 (C-4) and 155.7 (C-3), two oxygenated carbon signal at δ 76.2 (C-8) and 155.7 (C-3). Based on the above consideration and the comparison of the ^1H -, ^{13}C -NMR, IR and MS spectral data with those reported in the previous paper (Kobayashi *et al.*, 2006), the structure of **3** was identified as *p*-cymene-3,8-diol (**3**).

Compound **8** was obtained as white powder. The MS spectrum of **8** showed a molecular ion peak at m/z 487 [M + H]⁺. Compound **8** showed IR absorption bands for hydroxyl and carboxyl groups at 3357 cm⁻¹ and 1629 cm⁻¹, respectively. The $^1\text{H-NMR}$ spectrum showed six tertiary methyl signals at δ 0.91 (3H, s, H-30), 0.99 (3H, s, H-26), 1.01 (3H, s, H-25), 1.02 (3H, s, H-29), 1.12 (3H, s, H-27) and 1.67 (3H, s, H-23) and olefinic proton signal at δ 5.48 (1H, br.s, H-2). Beside oxygenated proton signals showed at δ 4.47 (1H, m, H-2) and 4.60 (1H, d, $J = 3.0$ Hz, H-3). The $^{13}\text{C-NMR}$ spectrum showed thirty carbon signals, including of carbonyl carbon signal at δ 180.5 (C-28) and

olefinic carbon peak at δ 122.8 (C-12) and 145.1 (C-13). Beside oxygenated carbon peaks at δ 65.5 (C-24), 66.5 (C-2) and 74.6 (C-3), six methyl group carbons signal showed at δ 17.4 (C-25), 17.7 (C-26), 24.0 (C-23), 24.4 (C-30), 26.6 (C-27), 34.0 (C-29). Based on the above consideration and the comparison of the ^1H -, ^{13}C -NMR, IR and MS spectral data with those reported in the previous paper (Hisanshi *et al.*, 1987), the structure of **8** was identified as $2\alpha,3\alpha,24$ -trihydroxyolean-12-en-28-oic acid (**8**).

Compound **9** was obtained as white powder. The MS spectrum of **9** showed a molecular ion peak at m/z 429 $[\text{M} + \text{H}]^+$. The ^1H -NMR spectrum showed six methyl signals δ 0.83 (3H, d, $J = 7.0$ Hz, H-28), 0.84 (3H, s, H-18), 0.86 (3H, d, $J = 7.0$ Hz, H-21), 0.90 (3H, s, H-19), 0.92 (3H, d, $J = 7.0$ Hz, H-26) and 1.01 (3H, d, $J = 7.0$ Hz, H-27). The AB coupling system appeared at δ 6.25 (1H, d, $J = 8.5$ Hz, H-7) and 6.52 (1H, d, $J = 8.5$ Hz, H-6). The ^{13}C -NMR spectrum showed 28 carbon peaks, including six methyl carbons at δ 13.1 (C-18), 17.8 (C-28), 18.4 (C-19), 19.9 (C-21), 20.2 (C-26) and 20.8 (C-27), two double bond signals at δ 130.9 (C-7), 132.5 (C-23), 135.4 (C-22) and 135.6 (C-6), and oxygenated quaternary carbon signal at δ 66.7 (C-3). Based on the above consideration and the comparison of the ^1H -, ^{13}C -NMR and MS spectral data with those reported in the previous paper (Xu *et al.*, 2007), the structure of **9** was identified as $5\alpha,8\alpha$ -epidioxyergosta-6,22-diol-3 β -ol (**9**).

Compound **12** was obtained as colorless gum. The MS spectrum of **12** showed a molecular ion peak at m/z 485 $[\text{M} + \text{Na}]^+$. The ^1H -NMR spectrum showed olefinic proton signals at δ 5.39 (1H, dddd, $J = 17.0, 12.0, 7.0, 2.0$ Hz, H-4) and 5.47 (1H, dddd, $J = 17.0, 12.0, 7.0, 2.0$ Hz, H-5). The signals at δ 3.32 (1H, quint, $J = 1.7$ Hz, H-4'), 3.36 (1H, t, $J = 9.0$ Hz, H-2'), 3.56 (1H, m, H-5'), 3.68 (1H, dd, $J = 11.5, 5.5$ Hz, H-3'), 3.88 (1H, dd, $J = 11.5, 1.5$ Hz, H-6'b) and 4.55 (1H, dd, $J = 11.5, 4.5$ Hz, H-6'a) indicated the presence of glucose. The ^{13}C -NMR spectrum showed two olefinic carbon signals at δ 126.0 (C-4) and 134.0 (C-3). Six oxygenated carbon signals for glucose at δ 63.3 (C-6'), 72.2 (C-4'), 75.7 (C-2'), 79.0 (C-5'), 79.1 (C-3') and 105.2 (C-1') were also shown. Based on the above consideration and the comparison of the ^1H -, ^{13}C -NMR and MS spectral data with those reported in the previous paper (Lee *et al.*, 2005) the compound **12** was identified as (Z)-3-hexenyl- β -D-glucopyranoside.

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