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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**), 2α,3α,24α,-trihydroxyolean-12en-28oic acid (**8**), 5α,8α-epidioxyergosta-6,22-diol-3β-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 – *Schzonepeta 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 μm column (250 × 22 mm) or Econosil® RP-18 10 μm column (250 × 22 mm). Silica gel 60 (Merck, 70 ~ 230 mesh and 230 ~ 400 mesh) was used for column chromatography. TLC used Merck precoated Silica gel F_{254} 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 LiChroprep Lobar®-A Si 60 (240 × 10 mm) or LiChroprep Lobar®-A RP-18 (240 × 10 mm) column with FMI QSY-0 pump (ISCO).

Plant materials – The arial 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 × 22 mm; *n*-hexane : EtOAc = 7 : 1) to yield compound **1** (23 mg). The fraction STH4 (470 mg) was also subjected to a RP-C_{18} 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-C_{18} 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-C_{18} 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-C_{18} 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-C_{18} silica gel column with 70% MeCN as the eluent and purified by RP-C_{18} prep. HPLC (Econosil® RP-18 10 μ column, 250 × 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-C_{18} 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% H_2O 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-C_{18} 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-C_{18} 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° (*c* 1.05, $CHCl_3$). IR ν_{max} cm^{-1} : 1741, 1059, 1019, 678. GC-MS *m/z* : 152 $[M]^+$. ^{1}H-NMR ($CDCl_3$, 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). ^{13}C-NMR ($CDCl_3$, 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} ($CHCl_3$, log ε) : 274 (4.76) nm. IR ν_{max} cm^{-1} : 1741, 1059, 1019, 678. GC-MS *m/z* : 150 $[M]^+$. ^{1}H-NMR ($CDCl_3$, 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). ^{13}C-NMR ($CDCl_3$, 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° (*c* 0.19, $CHCl_3$). UV λ_{max} ($CHCl_3$, log ε) : 277 (4.40) nm. GC-MS *m/z* : 166 $[M]^+$. ^{1}H-NMR (CD_3OD, 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). ^{13}C-NMR (CD_3OD, 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° (*c* 0.10, MeOH). IR ν_{max} cm^{-1} : 3286, 1569, 1479, 1124, 1034. FAB-MS *m/z* : 329 $[M - H]^-$. ^{1}H-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). ^{13}C-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°

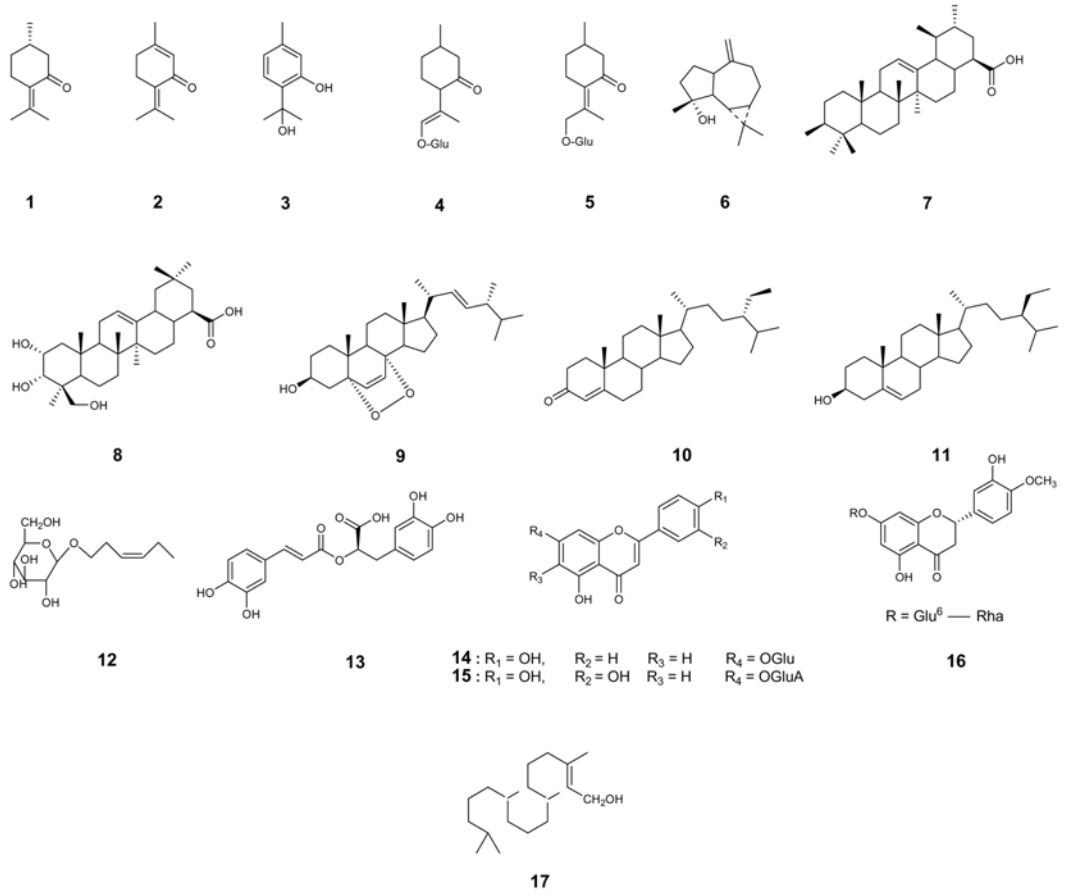

Fig. 1. The Structures of **1** - **17** from of *S. tenuifolia* Briq..

(*c* 0.18, MeOH). UV λ_{max} ($CHCl_3$, log ε) : 353 (4.37) nm. IR ν_{max} cm^{-1} : 3336, 1567, 1480, 1054, 1016. FAB-MS *m/z* : 329 $[M + Na]^+$. ^{1}H-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 H-1'). ^{13}C-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. $[\alpha]_D$ +2.8° (*c* 0.27, $CHCl_3$). UV λ_{max} ($CHCl_3$, log ε) : 274 (4.76) nm. GC-MS *m/z* : 220 $[M]^+$. ^{1}H-NMR ($CDCl_3$, 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). ^{13}C-NMR ($CDCl_3$, 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. $[\alpha]_D$ +78.3° (*c* 0.36, EtOH). FAB-MS *m/z* : 455 $[M + H]^+$. ^{1}H-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). ^{13}C-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)

2α,3α,24-Trihydroxyolean-12en-28oic acid (**8**) – White powder. m.p 266 - 258 °C. $[\alpha]_D$ +108.4° (*c* 0.16, MeOH). IR ν_{max} cm^{-1} : 3357, 1629, 1125, 1052. FAB-MS *m/z* : 487 $[M - H]^-$. ^{1}H-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}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).

5α,8α-Epidioxyergosta-6,22-diol-3β-ol (**9**) – White powder. m.p 176 - 178 °C. $[\alpha]_D$ –20.0° (c 0.22, $CHCl_3$). FAB-MS m/z : 427 $[M-H]^-$. ^{1}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}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° (c 0.17, $CHCl_3$). IR ν_{max} cm^{-1} : 1563, 1484, 1129, 1048. FAB-MS m/z : 413 $[M+H]^+$. ^{1}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}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 $[M+Na]^+$. ^{1}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}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° (c 0.17, MeOH). AB-MS m/z : 285 $[M+Na]^+$. ^{1}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}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 ε) : 327 (5.19), 290 (5.11) nm. IR ν_{max} cm^{-1} : 3286, 1576, 1121, 1054, 1033. FAB-MS m/z : 359 $[M-H]^-$. ^{1}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}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 ε) : 333 (5.08), 286 (5.03) nm. IR ν_{max} cm^{-1} : 3332, 1569, 1481, 11245. FAB-MS m/z : 433 $[M+H]^+$. ^{1}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}C-NMR (DMSO-d_6, 125 MHz) : δ 56.4 (–OCH_3), 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° (c 0.08, MeOH). UV λ_{max} (MeOH, log e) : 339 (5.31), 269 (5.24), 239 (5.08) nm. FAB-MS m/z : 461 $[M-H]^-$. ^{1}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}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} ($CHCl_3$, 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}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, 3.0Hz, 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}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. $[\alpha]_D$ +4.8° (*c* 0.2, $CHCl_3$). GC-MS *m/z* : 296 $[M]^+$. ^{1}H-NMR ($CDCl_3$, 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}C-NMR ($CDCl_3$, 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 ^{1}H-, ^{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}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}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 ^{1}H-, ^{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}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}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 ^{1}H-, ^{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}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, brs. 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}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 ^{1}H-, ^{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 2α,3α,24-trihydroxyolean-12en-28oic acid (**8**).

Compound **9** was obtained as white powder. The MS spectrum of **9** showed a molecular ion peak at *m/z* 429 $[M + H]^{+}$. The ^{1}H-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 ^{1}H-, ^{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 5α,8α-epidioxyergosta-6,22-diol-3β-ol (**9**).

Compound **12** was obtained as colorless gum. The MS spectrum of **12** showed a molecular ion peak at *m/z* 485 $[M + Na]^{+}$. The ^{1}H-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 ^{1}H-, ^{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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