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Short Communication

Polyphenolic Constituents from the Aerial Parts of *Thymus quinquecostatus* var. *japonica* Collected on Ulleung Island

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Phytochemical investigation on the polar constituents in the aerial parts of *Thymus quinquecostatus* var. *japonica* resulted in the isolation of three flavonoids along with four known phenolic components. The isolated compounds were characterized as 2(*S*)-5,7,3',5'-tetrahydroxyflavanone (1), (+)-taxifolin (2), (+)-aromadendrin (3), rosmarinic acid (4), caffeic acid (5), protocatechuic acid (6), and protocatechuic aldehyde (7) from comparisons of their physicochemical and spectroscopic data (^{1}H-, ^{13}C-NMR, two-dimentioal NMR, and mass spectrometery) with those of authentic samples and reference data. This is the first report on the isolation of compounds 1-7 from *T. quinquecostatus* var. *japonica*. In addition, compounds 2 and 4 exhibited the most potent pancreatic lipase inhibitory activities in a concentration-dependent manner with IC_{50} values of 15.4±0.8 and 62.8±2.7 μM, respectively.

Key words: obesity, pancreatic lipase, phenolic compounds, (+)-taxifolin, *Thymus quinquecostatus* var. *japonica*

Plants belonging to the genus *Thymus* (Lamiaceae) consist of about 350 species, mainly distributed throughout Europe, Asia, and North Africa. One of these, *Thymus quinquecostatus* var. *japonica*, is native to Korea, and the aerial parts of the plant have been used as ingredients in folk remedies for cough, diaphoresis, and parasiticides [Kim *et al*., 1997]. Crude extracts of *T. quinquecostatus* var. *japonica* have displayed a range of biological effects, such as antitumor and antibacterial activities [Shin and Kim, 2004; Sun *et al*., 2005]. Recently, several monoterpenes in this plant were found to exhibit antibacterial activity against *Propionibacterium* species [Oh *et al*., 2009]. However, the polar constituents of *T. quinquecostatus* var. *japonica* of Ulleung Island origin have not been investigated yet. As part of our continuing search for novel bioactive natural products, an ethyl acetate (EtOAc) extract of aerial parts of *T. quinquecostatus* var. *japonica* collected on Ulleung Island was investigated and three flavonoids, together with four phenolic metabolites were isolated. This paper describes a procedure for the isolation and structure elucidation of compounds **1-7**, as well as their inhibitory activity against porcine pancreatic lipase. This is the first report on the isolation of the phenolic constituents **1-7** from *T. quinquecostatus* var. *japonica.*

Optical rotations were measured with a JASCO DIP-4 digital polarimeter (Tokyo, Japan). The UV spectra were obtained with a Hitachi U-2000 spectrophotometer (Boston, MA), and the CD spectra were run on a JASCO J-720W spectrometer (Tokyo, Japan). Both ^{1}H and ^{13}C NMR spectra were measured on a Varian VNS600 instrument (Palo Alto, CA) operating at 600 and 150 MHz, respectively. The chemical shifts were given in δ (ppm) values relative to that of the solvent CD_3OD (δ_H 3.35; δ_C 49.0) on a tetramethylsilane (TMS) scale. The

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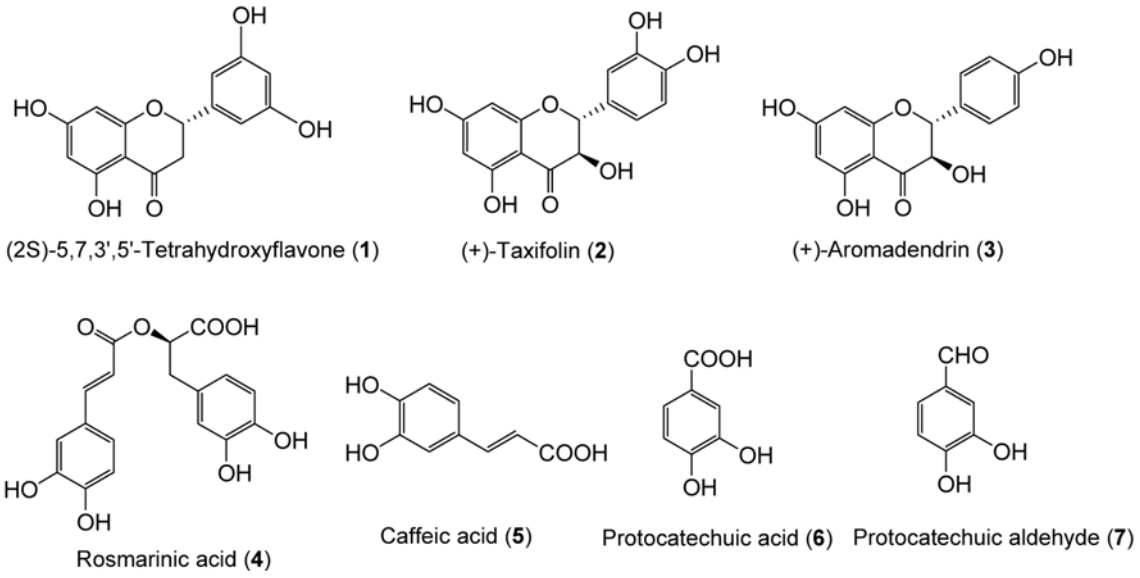

Fig. 1. Structures of isolated compounds 1-7 from the aerial parts of *T. quinquecostatus* var. *japonica*.

standard pulse sequences programmed into the instruments were used for each 2D measurement. The J_{CH} value was set at 8 Hz in the heteronuclear multiple bond connectivity (HMBC) spectra. Fast atom bombardment-mass spectrometer (FAB-MS) using 3-nitrobenzyl alcohol as the matrix agent was performed on a Micro Mass Auto Spec OA-TOF spectrometer (Tokyo, Japan). High-performance liquid chromatography (HPLC) analysis was carried out on a YMC-Pack ODS A-302 column (4.6 mm i.d.×150 mm; YMC Co., Kyoto, Japan) and developed at 40°C with 1% HCOOH-MeCN (85:15) (RP 1), 1% HCOOH-MeCN (8 : 2) (RP 2), and 1% HCOOH-MeCN (7 : 3) (RP 3). The detection wavelength was 280 nm. Column chromatography was carried out on Toyopearl HW-40 (coarse grade; Tosoh Co., Tokyo, Japan), YMC GEL ODS AQ 120-50S (YMC Co., Kyoto, Japan), and Sephadex LH-20 (Pharmacia Fine Chemicals Co., Uppsala, Sweden). Thin-layer chromatography (TLC) was performed on Kieselgel 60 F254 plates (0.2 mm layer thickness, Merck, Darmstadt, Germany), and the spots were detected by ultraviolet irradiation (254, 365 nm) and by spraying with 10% H_2SO_4 reagent.

The aerial parts of *T. quinquecostatus* var. *japonica* were collected from Ulleung Island, Korea in June 2010, and identified by prof. Tae Hoon Kim. A voucher specimen (KAJ-0058) was deposited at the Natural Product Chemistry Laboratory, College of Herbal Bio-industry, Daegu Hanny University.

Fresh milled *T. quinquecostatus* var. *japonica* (500 g) was extracted with 80% EtOH (18 L×3) at room temperature, and the solvent was evaporated *in vacuo*. The combined crude EtOH extract (42.0 g) was suspended in 10% MeOH (1.6 L), and then partitioned in turn with *n*-hexane (1.5 L×3), EtOAc (1.5 L×3), and *n*-BuOH (1.5 L×3) to yield dried *n*-hexane- (2.6 g), EtOAc- (4.4 g), *n*-BuOH (7.2 g) and H_2O-soluble (28.7 g) residues. A portion (4.0 g) of the EtOAc extract was chromatographed on a Toyopearl HW-40 column (coarse grade; 3.0 cm i.d. ×55 cm; Tosoh Co., Tokyo, Japan) with H_2O containing increasing amounts of MeOH in a stepwise gradient mode. The 30% MeOH eluate was subjected to column chromatography over a YMC GEL ODS AQ 120-50S column (1.1 cm i.d.×37 cm) with aqueous MeOH to yield pure compounds **6** (4.7 mg; RP 1: t_R=2.9 min) and **7** (4.0 mg; Rp 2: t_R=4.0 min). The 50% MeOH eluate was subjected to a combination of chromatography over Sephadex LH-20 (1.1 cm i.d.×41 cm) (with EtOH), and YMC GEL ODS AQ 120-50S (1.1 cm i.d.×38 cm) (with aqueous MeOH) to yield pure compounds **4** (21.4 mg; RP 2: t_R=8.5 min) and **5** (7.5 mg; RP 2:t_R=3.2 min). Similarly, the 70% MeOH eluate was chromatographed over YMC GEL ODS AQ 120-50S (1.1 cm i.d.×37 cm) with aqueous MeOH to yield pure compounds **1** (5.2 mg; RP 3: t_R=5.0 min), **2** (2.0 mg; RP 3: t_R=3.8 min), and **3** (5.4 mg; RP 3: t_R=2.9 min).

Compound **1** (**2(*S*)-5,7,3',5'-tetrahydroxyflavanone**): Yellow amorphous powder; $[\alpha]_D^{20}$ +8.4° (*c,* 1.0, MeOH); UV λ max MeOH nm (log ε): 226 (3.57), 283 (3.52), 325 (2.73); CD (MeOH, Δε) 327 (+0.28), 286 (-1.41) nm; FAB-MS *m/z* 289 $[M+H]^+$; ^1H and ^{13}C NMR data, see Table 1.

Compound **2** (**(+)-Taxifolin**): Yellow amorphous powder; $[\alpha]_D^{20}$ +17.6° (*c*, 1.0, MeOH); CD (MeOH, Δε) 330 (+0.11), 297 (-2.57) nm; FAB-MS *m/z* 305 $[M+H]^+$;

^{1}H- and ^{13}C-NMR data, see Table 1.

Compound **3** (**(+)-Aromadendrin**): Yellow amorphous powder; $[\alpha]_D^{20}$ +19.3° (*c*, 1.0, MeOH); CD (MeOH, Δε) 327 (+0.31), 295 (-3.05) nm; FAB-MS *m/z* 289 $[M+H]^+$; ^{1}H- and ^{13}C-NMR data, see Table 1.

Compound **4** (**Rosmarinic acid**): Yellow amorphous powder; $[\alpha]_D^{20}$ +85.3° (*c*, 1.0, MeOH); FAB-MS *m/z* 361 $[M+H]^+$; ^{1}H-NMR (600 MHz, CD_3OD) δ 7.54 (1H, d, *J*= 15.8 Hz, H-7), 7.03 (1H, d, *J*=1.2 Hz, H-2'), 6.94 (1H, dd, *J*=7.8, 1.8 Hz, H-6), 6.77 (1H, dd, *J*=7.8 Hz, H-5), 6.74 (1H, d, *J*=1.8 Hz, H-2), 6.69 (1H, d, *J*=7.8 Hz, H-5'), 6.60 (1H, dd, *J*=7.8, 1.8 Hz, H-6'), 6.25 (1H, d, *J*=15.8 Hz, H-8), 5.18 (1H, dd, *J*=8.4, 4.2 Hz, H-8'), 3.09 (1H, dd, *J*=14.4, 4.2 Hz, H-7'), 3.00 (1H, dd, *J*=14.4, 8.4 Hz, H-7'). ^{13}C-NMR (150 MHz, CD_3OD) δ 173.6 (C-9'), 168.5 (C-9), 149.7 (C-4), 147.7 (C-7), 146.8 (C-3'), 146.1 (C-3'), 145.2 (C-4'), 129.3 (C-1'), 127.7 (C-1), 123.1 (C-6), 121.8 (C-6'), 117.6 (C-2'), 116.5 (C-5), 116.3 (C-5'), 115.2 (C-8), 114.4 (C-2), 74.7 (C-8'), 37.9 (C-7').

Compound **5** (**Caffeic acid**): White amorphous powder; ^{1}H-NMR (600 MHz, CD_3OD) δ 7.41 (1H, d, *J*=15.5 Hz, H-8), 7.08 (1H, d, *J*=2.0 Hz, H-2), 6.91 (1H, dd, *J*=8.0, 2.0 Hz, H-5), 6.79 (1H, d, *J*=8.0 Hz, H-6), 6.23 (1H, d, *J*=15.5 Hz, H-8).

Compound **6** (**Protocatechuic acid**): White amorphous powder; ^{1}H-NMR (600 MHz, CD_3OD) δ 7.43 (1H, *d*, *J*=1.8 Hz, H-2), 7.15 (1H, d, *J*=2.4 Hz, H-2), 7.40 (1H, dd, *J*=7.8, 1.8 Hz, H-6), 6.78 (1H, d, *J*=7.8 Hz, H-5); ^{13}C-NMR (150 MHz, CD_3OD) δ 170.5 (C-7), 151.4 (C-4), 146.3 (C-3), 123.8 (C-1), 123.5 (C-2), 117.7 (C-6), 115.7 (C-5).

Compound **7** (**Protocatechuic aldehyde**): White amorphous powder; ^{1}H-NMR (600 MHz, CD_3OD) δ 9.68 (1H, s, CHO), 7.30 (1H, *d*, *J*=1.8 Hz, H-2), 7.29 (1H, dd, *J*=8.0, 1.8 Hz, H-6), 6.90 (1H, d, *J*=7.8 Hz, H-6); ^{13}C-NMR (150 MHz, CD_3OD) δ 193.0 (C-7), 153.7 (C-3), 147.2 (C-4), 130.8 (C-1), 126.3 (C-2), 116.2 (C-6), 115.7 (C-5).

Assay of Pancreatic Lipase Activity: The ability of the compounds to inhibit porcine pancreatic lipase was evaluated using the previously reported method with a minor modification [Kim *et al*., 2007]. Briefly, an enzyme buffer was prepared by adding 30 μL (10 U) porcine pancreatic lipase solution (Sigma, St. Louis, MO) in 10 mM 4-morpholinepropanesulphonic acid (MOPS) and 1 mM EDTA, pH 6.8 to 850 μL of Tris buffer (100 mM Tris-HC1 and 5 mM $CaCl_2$, pH 7.0). Subsequently, 100 μL of the compounds and orlistat (Roche, Basel, Switzerland) at the test concentration were mixed with 880 μL of the enzyme-buffer, and incubated for 15 min at 37°C, with the addition of 20 μL substrate solution [10 mM *p*-nitrophenylbutyrate (*p*-NPB) in dimethyl formamide], and the enzymatic reactions were allowed to proceed for 15 min at 37°C. The pancreatic lipase activity was determined by measuring the hydrolysis of *p*-NPB into *p*-nitrophenol at 405 nm using an ELISA reader (Tecan,

Table 1. ^{1}H and ^{13}C NMR spectral data of compounds 1-3

Position	1		2		3	
	δ_H (*J* in Hz)[b]	δ_C	δ_H (*J* in Hz)[b]	δ_C	δ_H (*J* in Hz)[b]	δ_C
2	5.27 (dd, 12.6, 3.0)	80.5	4.90 (d, 11.4)	85.1	4.97 (d, 11.4)	85.0
3β	2.69 (dd, 17.4, 3.0)	44.1	4.49 (d, 11.4)	73.7	4.55 (d, 11.4)	73.6
3α	3.06 (dd, 17.4, 12.6)					
4		197.8		198.4		198.5
5		165.5		165.3		168.9
6	5.88 (d, 2.4)	97.0	5.91 (d, 2.4)	97.3	5.92 (d, 2.4)	97.4
7		168.4		168.8		165.3
8	5.87 (d, 2.4)	96.2	5.88 (d, 2.4)	96.3	5.88 (d, 2.4)	96.3
9		164.9		164.5		164.6
10		103.4		101.8		101.8
1'		131.8		129.9		129.3
2'	6.78 (s)	116.3	6.84 (dd, 8.4, 1.8)	116.1	7.35 (d, 8.4)	130.4
3'		146.9		146.3	6.83 (d, 8.4)	116.1
4'	6.91 (s)	119.3		147.2		159.2
5'		146.5	6.79 (d, 8.4)	115.9	6.83 (d, 8.4)	116.1
6'	6.77 (s)	114.7	6.95 (d, 1.8)	120.9	7.35 (d, 8.4)	130.4

[a] ^{1}H-NMR measured at 600 MHz, ^{13}C-NMR measured at 150 MHz; obtained in CD_3OD with TMS as internal standard. Assignments based on HMQC and HMBC NMR spectra.

[b] *J* values (Hz) are given in parentheses.

Infinite F200, Groedig, Austria). Inhibition of the lipase activity was expressed as the percentage decrease in the OD when porcine pancreatic lipase was incubated with the test compounds.

Successive column chromatographic purification of the EtOAc-soluble fraction of the aqueous ethanolic extract of *T. quinquecostatus* var. *japonica* led to the isolation and characterization of three flavonoids **1-3** along with four known phenolic constituents **4-7**. The isolated compounds were identified as 2(*S*)-5,7,3',5'-tetrahydroxy-flavanone (**1**), (+)-taxifolin (**2**), (+)-aromadendrin (**3**), rosmarinic acid (**4**), caffeic acid (**5**), protocatechuic acid (**6**), and protocatechuic aldehyde (**7**) from comparisons of their physicochemical and spectroscopic data (^{1}H-, ^{13}C-NMR, 2D NMR, and MS) with those of authentic samples and reference data. Compounds **4** and **5** were found to be the major metabolites of the EtOAc-soluble portion of the aerial parts of *T. quinquecostatus* var. *japonica.* All compounds **1-7** were isolated for the first time from *T. quinquecostatus* var. *japonica* of Ulleung Island origin, and compounds **2** and **4** showed significant inhibitory activities against porcine pancreatic lipase.

Compound **1** was obtained as a yellow amorphous powder, and the molecular formula ($C_{21}H_{21}O_9$) was deduced from the molecular ion peak at *m/z* 289 $[M+H]^+$ by FAB-MS and NMR data. The absorption maxima at 225, 283, and 325 nm in the UV spectrum were attributed to a flavanone nucleus [Mabry *et al.*, 1970]. The presence of the flavanone skeleton was further suggested by ^{1}H-NMR spectrum of **1** (Table 1) for ABX systems at δ_H 2.69 (1H, dd, *J*=17.4, 3.0 Hz, H-3β), 3.06 (1H, dd, *J*=17.4, 12.6 Hz, H-3α), 5.27 (1H, dd, *J*=12.6, 3.0 Hz, H-2). In addition to the characteristic aliphatic signals, AB_2 aromatic signals were observed at δ_H 6.91 (1H, s, H-4'), 6.78 (1H, s, H-2') , 6.77 (1H, s, H-6'), along with two aromatic protons in the A ring at δ_H 5.88 (1H, d, *J*=2.4 Hz, H-6) and 5.87 (1H, d, *J*=2.4 Hz, H-8). Consistent with the above ^{1}H-NMR analysis, the ^{13}C-NMR spectrum (Table 1) of compound **1** displayed the signals for an aliphatic methylene at δ_C 44.1 (C-3), an oxygenated methine at δ_C 80.5 (C-2), two aromatic rings at δ_C 168.4 (C-7), 165.5 (C-5), 164.9 (C-9), 146.9 (C-3'), 146.5 (C-5'), 131.8 (C-1'), 119.3 (C-4'), 116.3 (C-2'), 114.7 (C-6'), 103.4 (C-10), 97.0 (C-6), 96.2 (C-8), and a conjugated ketone at δ_C 197.8 (C-4). These one-dimensional (1D) NMR data, in combination with the observed 2D NMR [^{1}H-^{1}H, ^{1}H-^{1}H correlated spectroscopy (COSY); heteronuclear multiple quantum coherence (HMQC); heteronuclear multiple bond correlation (HMBC)] correlations, suggeted that compound **1** is a flavanone possessing a trisubstituted B ring. The *trans* relative stereostructures of the H-2 and H-3 positions in the flavanone B-ring were characterized by large coupling constant (*J*=12.6 Hz) [Carrcache-Blanco *et al.*, 2003]. The absolute configuration at C-2 was determined as *S* on the basis of a negative Cotton effect at 286 nm in the circular dichroism (CD) spectral comparison with the authentic analogs [Gaffield, 1970]. Thus compound **1** was identified as 2(*S*)-5,7,3',5'-tetrahydroxyflavanone through comparison of the physicochemical and spectroscopic data with those of reference data [Sun *et al*., 2007; Zheng *et al*., 2008]. A molecular formula of $C_{15}H_{12}O_7$ was assigned to compound **2** on the basis of FAB-MS (*m/z* 305 $[M+H]^+$). In a manner similar to **1**, the ^{1}H-NMR spectrum of **2** (Table 1) showed signals for two oxygenated methine doublets at δ_H 4.90 (1H, d, *J*=11.4 Hz, H-2), 4.49 (1H, d, *J*=11.4 Hz, H-3), two aromatic doublets at δ_H 5.91 (1H, d, *J*=2.4 Hz, H-6), 5.88 (1H, d, *J*=2.4 Hz, H-8), and ABX-type aromatic signals at δ_H 6.95 (1H, d, *J*=1.8 Hz, H-6'), 6.84 (1H, dd, *J*=8.4, 1.8 Hz, H-2'), 6.79 (1H, d, *J*=8.4 Hz, H-3'). The ^{13}C-NMR resonances of the oxygenated methines (δ_C 85.1, 73.7) and conjugated ketone (δ_C 198.4) observed for compound **2** were very close to those of **1**. This results indicated that compound **2** was flavanonol metabolite [Shen *et al.*, 1993]. The large coupling constant (*J*=11.4 Hz) between H-2 and H-3 indicated a *trans* relationship. The CD spectrum of **2** showed a negative Cotton effect at 297 nm (Δε +0.11), indicating that the absolute configuration of **2** was the 2*R*,3*R*-configuration [Nonaka *et al*., 1987]. Thus, compound **2** was identified as (+)-taxifolin through comparison of the physicochemical and spectroscopic data with those of reference data [Nonaka *et al.*, 1987].

Compound **3** was isolated as a yellow amorphous powder, $[\alpha]_D^{20}$ +19.3° (MeOH). The FAB-MS of **3** showed a pseudomolecular ion peak at *m/z* 289 $[M+H]^+$, which was 16 mass units larger than **2**. The ^{1}H- and ^{13}C-NMR spectra of **3** were very similar to those of **2**, except for the presence of a magnetically equivalent 2H-singlet (δ_H 6.83) instead of the *meta*-coupled aromatic signals in **2**. Compound **3** was regarded as the 3-hydroxyl congener of **2**. The 2*R*,3*R*-configuration of **3** was also evidenced by the similar CD spectrum of its a negative Cotton effect at 295 nm (Δε -3.05). Thus compound **3** was identified as (+)-aromadendrin through comparison of the physicochemical and spectroscopic data with those of reference data [Kasai *et al.*, 1988].

Compound **4** was isolated as a yellow amorphous powder, $[\alpha]_D^{20}$ +85.3° (MeOH) and exhibited a pseudo-molecular ion peak $[M+H]^+$ at *m/z* 361 in a positive FAB-MS spectrum. The ^{1}H-NMR spectrum of **4** showed signals for an oxygenated methine proton at δ_H 5.18 (1H, dd, *J*=8.4, 4.2 Hz, H-8'), *trans*-olefinic protons at δ_H 7.54 (1H, d, *J*=15.8 Hz, H-7), 6.25 (1H, d, *J*=15.8 Hz, H-8), a

methylene group at δ_H 3.09 (1H, dd, *J*=14.4, 4.2 Hz, H-7'), 3.00 (1H, dd, *J*=14.4, 8.4 Hz, H-7'), and two sets of ABX-type signals at δ_H 7.03 (1H, d, *J*=1.2 Hz, H-2'), 6.94 (1H, dd, *J*=7.8, 1.8 Hz, H-6), 6.77 (1H, dd, *J*=7.8 Hz, H-5), 6.74 (1H, d, *J*=1.8 Hz, H-2), 6.69 (1H, d, *J*=7.8 Hz, H-5'), 6.60 (1H, dd, *J*=7.8, 1.8 Hz, H-6'). Consistent with the above ^{1}H-NMR analysis, the ^{13}C-NMR spectrum of compound **4** displayed signals for an aliphatic methylene at δ_C 37.9 (C-7'), an oxygenated methine at δ_C 74.7 (C-8'), one carbonyl carbon at δ_C 173.6 (C-9'), two olefinic carbons at δ_C 147.7 (C-7), 115.2 (C-8), and two sets of aromatic carbons at δ_C 149.7 (C-4), 146.8 (C-3'), 146.1 (C-3'), 145.2 (C-4'), 129.3 (C-1'), 127.7 (C-1), 123.1 (C-6), 121.8 (C-6'), 117.6 (C-2'), 116.5 (C-5), 116.3 (C-5'), 114.4 (C-2). These 1D NMR data, in combination with the observed 2D NMR (^{1}H-^{1}H COSY, HMQC, HMBC) correlations, suggeted that compound **4** was a caffeic acid dimer. Thus, compound **4** was identified as rosmarinic acid by comparing the spectral data with those of reported data [Satake *et a.l*, 1999].

Compounds **5**, **6**, and **7** were identified as caffeic acid, protocatechuic acid, and protocatechuic aldehyde, respectively, by comparison with the spectral data in the literature [Cui *et al.*, 1990; Yamanaka *et al.*, 1995; Xu *et al.*, 2007]. Identification of **5**, **6**, and **7** was further confirmed by direct comparison of HPLC analysis with authentic samples. To the best of our knowledge, phenolic constituents **1-7** from the aerial parts of *T. quinquecostatus* var. *japonica* of Ulleung Island origin were isolated for the first time in the present study.

Obesity is caused by an imbalance between energy intake and expenditure and is widely recognized as a major public health problem. Obesity could result in hypertension, hyperlipidemia, arteriosclerosis, and type II diabetes [Cooke and Bloom, 2006]. Pancreatic lipase is a key enzyme for triglyceride absorption in the small intestine. This enzyme is secreted from the pancreas and hydrolyzes triglycerides into glycerol and fatty acids [Lowe *et al.*, 1994]. Therefore, pancreatic lipase inhibitors are considered to be a valuable therapeutic reagent for treating diet-induced obesity in humans. The success of orlistat [Hill *et al.*, 1999], which is a specific pancreatic lipase inhibitor, has prompted research to identify new enzyme inhibitors derived from various medicinal plants [Birari and Bhutani, 2007; Yun, 2010]. The compounds isolated from *T. quinquecostatus* var. *japonica* were evaluated for their ability to inhibit pancreatic lipase activity [Kim *et al.*, 2007] using orlistat as a positive control (Table 2). Among the tested compounds, compound **2** showed significantly higher pancreatic lipase inhibitory activity (IC_{50}, 15.4±0.8 μM) than those of the other tested isolates. On the other hand, the flavanone derivatives (**1**) and (**3**) displayed lower inhibitory effects than that of **2**. These results suggested that the degree of hydroxylation at the B- and C-ring on the flavanone skeleton may have influenced pancreatic lipase inhibition. Additionally, rosmarinic acid (**4**), a caffeic acid dimer, exhibited improved inhibitory activity against pancreatic lipase with IC_{50} value of 62.8±2.7 μM, when compared to caffeic acid (**5**).

Table 2. Pancreatic lipase inhibitory activities of compounds 1-7

Compound	IC_{50}[a] (μM)
2(*S*)-5,7,3',5'-Tetrahydroxyflavanone (**1**)	>200
(+)-Taxifolin (**2**)	15.4±0.8
(+)-Aromadendrin (**3**)	>200
Rosmarinic acid (**4**),	62.8±2.7
Caffeic acid (**5**)	>200
Protocatechuic acid (**6**)	>200
Protocatechuic aldehyde (**7**)	>200
Orlistat[b]	0.6±0.2

[a]All compounds were examined in duplicate experiments; IC_{50} values of compounds represent the concentration that caused 50% reduction in enzyme activity.
[b]Orlistat was used as a positive control.

In summary, phytochemical and biological investigations of *T. quinquecostatus* var. *japonica* collected from Ulleung Island were carried out in the present study for the first time. The seven constituents isolated from the aerial parts of this biomass were first identified from the present study. In addition, some of the isolated compounds showed weak to strong inhibitory activities against porcine pancreatic lipase. Therefore, these findings suggested that the alcoholic extracts containing some phenolics from the aerial parts of *T. quinquecostatus* var. *japonica* may useful materials for suppressing lipid uptake.

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