Flavonoids from leaves of *Tetracera scandens* L.

Tung Bui Thanh¹, Hai Nguyen Thanh², Huong Duong Thi Ly¹, Huong Le-Thi-Thu³, Loi Vu Duc³ and Tung Nguyen Huu⁴

¹Department of Pharmacology and Clinical Pharmacy, School of Medicine and Pharmacy, Vietnam National University, Hanoi, 144 Xuan Thuy, Cau Giay, Ha Noi, Vietnam
²Department of Pharmaceutics and Pharmaceutical Technology, School of Medicine and Pharmacy, Vietnam National University, Hanoi, 144 Xuan Thuy, Cau Giay, Ha Noi, Vietnam
³Department of Pharmacognosy and Traditional Medicines, School of Medicine and Pharmacy, Vietnam National University, Hanoi, 144 Xuan Thuy, Cau Giay, Ha Noi, Vietnam
⁴Department of Pharmaceutical Chemistry and Drug Quality Control, School of Medicine and Pharmacy, Vietnam National University, Hanoi, 144 Xuan Thuy, Cau Giay, Ha Noi, Vietnam

**ABSTRACT**

*Tetracera scandens* L. (*Dilleniaceae*) is a genus of flowering plants of the Dilleniaceae family. It has been reported that *Tetracera scandens* L. content hight amout of flavonoids. From the ethanol extract of leaves of *Tetracera scandens* L, we have been isolated six known flavonoids compounds. Their structures were identified as quercetin-3-O-L-rhamnoside (1), genistein (2), quercetin (3), quercetin 3-O-β-D-glucuronide (4), kaempferol (5) and quercetin-3-O-α-L-arabinofuranoside (6) on the basis of spectroscopic data and by comparing their physicochemical and spectral data with those published in literatures.

**Keywords:** Flavonoids, Quercetin, *Tetracera scandens* L.

**INTRODUCTION**

*Tetracera scandens* L. (*Dilleniaceae*) is a genus of flowering plants of the Dilleniaceae family. It is widely used in Vietnamese traditional medicine to treat the diseases of hepatitis, gout and inflammation. Therefore, we conducted the investigation about the chemical composition of leaves of *Tetracera scandens* L, which were collected in Nha Trang, Vietnam. From the ethanol extract of leaves of *Tetracera scandens* L, we have been isolated together with six known compounds, which were first reported in this material. Their structures were elucidated on the basis of spectroscopic data and comparing their physicochemical and spectral data with those published in literatures.

**GENERAL AND EXPERIMENTAL PROCEDURES**

UV spectra were recorded in MeOH on a JASCO V-550 UV/vis spectrometer with a 0.5 nm resolution. NMR spectra were obtained on a Varian Unity Inova 500 MHz spectrometer with tetramethylsilane (TMS) as the internal standard. Silica gel (Merck, 63-200 μm particle size), RP-18 (Merck, 40-63 μm particle size), and Sephadex LH-20 were used for column chromatography. TLC was carried out with silica gel 60 F254 and RP-18 F254 plates. HPLC was carried out using a Gilson system with a UV detector and Optima Pak C18 column (10 × 250 mm, 10 μm particle size, RS Tech, Korea). All solvents used for extraction and isolation were of analytical grade.

**PLANT MATERIAL**

The leaves of *Tetracera scandens* L. were collected in October 2013 from Nha Trang province, Vietnam and authenticated by Prof. Nguyen Thanh Hai (School of Medicine and Pharmacy, Vietnam National University, Hanoi). A voucher specimen (No. SMP-2013-0012) was deposited at the Herbarium of SMP, VNU.
EXTRACTION AND ISOLATION

The leaves of Tetracera scandens L. (2.5 kg) were extracted with ethanol (10 L × 3 times) at room temperature for a week. The combined ethanol extract was then concentrated to yield a dry residue (251 g). This crude extract was suspended in water (2.5 L) and partitioned successively with n-hexane (3 × 2 L), EtOAc (3 × 2 L), and BuOH (3 × 2 L). The EtOAc and BuOH soluble fractions were combined (117 g) and chromatographed over a silica gel column (10 × 30 cm; 63–200 μm particle size) eluting with gradient solvent CHCl₃/acetone (19:1, 18:2…1:19, each 2.5 L), to yield six fractions (F₁: 13.2 g; F₂: 22.6 g; F₃: 28.4 g; F₄: 14.5 g; F₅: 23.11 g; F₆: 10.87 g) based on the TLC profile. Fraction F₃ was further applied to an RP-18 column (7 × 30 cm; 40–63 μm particle size) eluting with a stepwise gradient of MeOH/H₂O (1:2 to 10:1) to afford four sub-fractions (F₃.1–F₃.4). Fraction F₃.1 (90 mg) was separated by HPLC (Optima Pak C18 column (10 × 25 mm, 10 μm particle size, RS Tech, Korea); mobile phase MeOH in H₂O containing 0.1% formic acid (0–50 min: 63% MeOH, 50–55 min: 10% MeOH; flow rate 2 mL/min; UV detection at 205 and 254 nm) to give compound 1 (tR = 36.0 min, 16.0 mg) (quercetin-3-O-L-rhamnoside), compound 2 (tR = 39.0 min, 10.0 mg) (genistin). Fraction F₃.2 (80 mg) was purified by HPLC (0–50 min: 75% MeOH, 50–55 min: 100% MeOH) to yield compound 3 (tR = 40.0 min, 5.8 mg) (quercetin).

Quercetin-3-O-L-rhamnoside (1): yellow powder, UV spectrum (MeOH, λmax, nm) 257, 359; PMR spectrum (500 MHz, acetone-d₆, δ, ppm, J/Hz): 6.73 (1H, br, s, H-6), 6.94 (1H, br, s, H-8), 7.97 (1H, d, J = 1.5, H-2'), 7.45 (1H, d, J = 8.4, H-5'), 7.85 (1H, d, J = 8.4, H-6'), 5.99 (1H, m, H-1''), 4.16 (1H, m, H-2''), 3.80 (1H, m, H-3''), 3.61 (1H, m, H-4''). 13C-NMR spectrum (125 MHz, acetone-d₆, δ, ppm): 157.9 (C-2), 135.8 (C-3), 179.2 (C-4), 163.8 (C-5), 99.4 (C-6), 164.7 (C-7), 94.5 (C-8), 158.3 (C-9), 105.7 (C-10), 122.8 (C-1'), 116.1 (C-2'), 145.7 (C-3'), 148.9 (C-4'), 116.7 (C-5'), 122.6 (C-6'), 102.7 (C-1''), 72.0 (C-2''), 72.9 (C-3''), 78.7 (C-4''), 71.3 (C-5''), 17.7 (C-6'). These data are in agreement with those originally reported for quercetin-3-O-a-L-rhamnoside (1).

Genistin (2): yellow powder, UV spectrum (MeOH, λmax, nm) 260, 366; PMR spectrum (500 MHz, acetone-d₆, δ, ppm, J/Hz): 7.87 (1H, s, H-2), 6.32 (1H, d, J = 2.17, H-6), 6.58 (1H, d, J = 2.17, H-8), 7.74 (2H, d, J = 8.73, H-2', H-6'), 7.04 (2H, d, J = 8.72, H-3', H-5'). 13C-NMR spectrum (125 MHz, acetone-d₆, δ, ppm): 148.3 (C-2), 146.7 (C-3), 176.5 (C-4), 162.1 (C-5), 99.1 (C-6), 165.0 (C-7), 94.5 (C-8), 157.8 (C-9), 104.1 (C-10), 121.8 (C-1'), 121.5 (C-2'), 115.9 (C-3'), 146.9 (C-4'), 115.9 (C-5'), 121.5 (C-6'). These data are in agreement with those originally reported for genistin [2].

Quercetin (3): yellow powder, PMR spectrum (500 MHz, acetone-d₆, δ, ppm, J/Hz): 6.27 (1H, d, J = 2.2, H-6), 6.53 (1H, d, J = 2.2, H-8), 7.83 (1H, d, J = 2.2, H-2'), 6.99 (1H, d, J = 9.1, H-5'), 7.69 (1H, d, J = 9.0, H-2'), 6.27 (1H, d, J = 2.2, H-6'); 13C-NMR spectrum (125 MHz, acetone-d₆, δ, ppm): 124.9 (C-2), 136.7 (C-3), 176.5 (C-4), 162.1 (C-5), 99.1 (C-6), 164.9 (C-7), 94.5 (C-8), 157.8 (C-9), 104.1 (C-10), 123.8 (C-1'), 115.8 (C-2'), 145.8 (C-3'), 148.3 (C-4'), 116.2 (C-5'), 121.5 (C-6'). These data are in agreement with those originally reported for quercetin [3].

Quercetin 3-O-β-D-glucuronide (4): yellow powder, UV spectrum (MeOH, λmax, nm) 256, 358; PMR spectrum (500 MHz, acetone-d₆, δ, ppm, J/Hz): 6.23 (1H, d, J = 2.0, H-6), 6.47 (1H, d, J = 2.0, H-8), 7.82 (1H, d, J = 2.4, H-2'), 6.90 (1H, d, J = 8.6, H-5'), 7.69 (1H, d, J = 8.6, H-6'), 5.44 (1H, m, H-1''), 3.52 (1H, m, H-2''), 3.57 (1H, m, H-3''), 3.84 (1H, m, H-4'), 3.87 (1H, m, H-5'), 3.97 (1H, m, H-6'). 13C-NMR spectrum (125 MHz, acetone-d₆, δ, ppm): 157.8 (C-2), 135.1 (C-3), 178.8 (C-4), 162.8 (C-5), 99.6 (C-6), 165.1 (C-7), 94.6 (C-8), 158.1 (C-9), 105.4 (C-10), 123.1 (C-1'), 115.8 (C-2'), 145.3 (C-3'), 149.2 (C-4'), 117.2 (C-5'), 122.5 (C-6'), 104.1 (C-1''), 75.1 (C-2''), 72.3 (C-3''), 72.3 (C-4'), 77.4 (C-5'), 170.1 (C-6'). These data are in agreement with those originally reported for quercetin 3-O-β-D-glucuronide [4].

Kaempferol (5): yellow powder, PMR spectrum (500 MHz, acetone-d₆, δ, ppm, J/Hz): 6.27 (1H, d, J = 2.0, H-6), 6.54 (1H, d, J = 2.0, H-8), 8.15 (2H, d, J = 8.8, H-2', H-6'), 7.01 (2H, d, J = 8.8, H-3', H-5'); 13C-NMR spectrum (125 MHz, acetone-d₆, δ, ppm): 147.0 (C-2), 136.7 (C-3), 176.6 (C-4), 162.1 (C-5), 99.1 (C-6), 165.0 (C-7), 94.6 (C-8), 157.9 (C-9), 104.1 (C-10), 123.4 (C-1'), 130.5 (C-2'), 116.4 (C-3'), 160.1 (C-4'), 116.4 (C-5'), 130.5 (C-6'). These data are in agreement with those originally reported for kaempferol [3].
Quercetin-3-O-α-L-arabinofuranoside (6): yellow powder, UV spectrum (MeOH, λmax, nm) 257, 357; PMR spectrum (500 MHz, acetone-d6, δ, ppm, J/Hz): 6.28 (1H, d, J=2.1, H-6); 6.51 (1H, d, J= 2.1, H-8), 7.73 (1H, d, J=2.1, H-2’); 6.99 (1H, d, J=8.5, H-5’), 7.58 (1H, dd, J=8.5, J= 2.4, H-6’), 5.48 (1H, d, J=0.6, H-1’); 4.10(1H, m, H-2”), 3.61(1H, m, H-3”), 3.97 (1H, m, H-4”); 13C-NMR spectrum (125 MHz, acetone-d6, δ, ppm): 158.1 (C-2), 135.4 (C-3), 179.9 (C-4), 162.8 (C-5), 99.6 (C-6), 165.2 (C-7), 94.7 (C-8), 158.0 (C-9), 105.5 (C-10), 122.9 (C-1’), 116.6 (C-2’), 145.8 (C-3’), 149.2 (C-4’), 116.4 (C-5’), 122.5 (C-6’), 109.2 (C-1”), 82.3 (C-2”), 78.9 (C-3”), 89.5 (C-4”), 62.7 (C-5”). These data are in agreement with those originally reported for quercetin-3-O-α-L-arabinofuranoside [5].

Figure 1. Structure of flavonoids isolated from leaves of *Tetracera scandens* L.
Competing interests
We declare that we have no conflict of interest.

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