

CHEMICAL CONSTITUENTS FROM *Ficus tikoua*

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Ficus tikoua Bur., a herbaceous plant, is widely distributed in south China, India, Vietnam, and Laos. It has long been used in traditional folk medicine to treat diseases such as chronic bronchitis, diarrhea, dysentery, mastadenitis, rheumatism, edema, impetigo, and so on. Previous studies show that extracts of stems of *F. tikoua* exhibited excellent antifungal activity; however, only a few studies on the chemical constituents of *F. tikoua* have been reported [1, 2]. In order to find the bioactive secondary metabolites from the plant, the stems of *F. tikoua* were investigated, which led to the isolation of seven known compounds. All these compounds were isolated from *F. tikoua* for the first time.

The stem of *F. tikoua* was collected in Hongya County, Sichuan Province, P. R. China, in September 2009, and authenticated by Prof. Hua Yi of the College of Life Sciences, Northwest Agricultural & Forestry University. Voucher specimens (samples No. NWAU2009-FT15) were deposited at the College of Life Sciences, Northwest Agricultural & Forestry University.

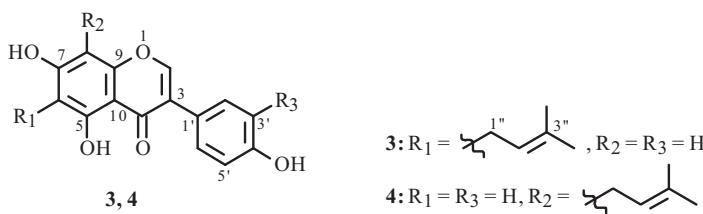
The dried and pulverized stem of *F. tikoua* (5.0 kg) was extracted three times (4 h each time) under reflux with analytical grade methanol. After solvent removal under vacuum, the viscous extract was partitioned with EtOAc. The EtOAc fraction was purified by column chromatography with silica gel, RP-C₁₈, Sephadex LH-20, and pre-HPLC to yield compounds **1–7**.

All compounds were identified by spectroscopic methods, including NMR and mass spectrometry. The spectroscopic data of all compounds were in good agreement with the literature data. The antifungal activities against *Phytophthora infestans* were tested *in vitro* for these compounds.

Genistein (1), colorless needles, C₁₅H₁₀O₅, mp 290–292°C. UV/Vis (MeOH, λ_{max} , nm): 291. ESI-MS: *m/z* 269 ([M – H][–], 85), 539([2M – H][–], 100) [3].

Myrsininone A (2), pale yellow oil, C₂₅H₂₆O₅. UV/Vis (λ_{max} , MeOH, nm): 265. ESI-MS: *m/z* 405 ([M – H][–], 100), 811 ([2M – H][–], 30), ESI-MS/MS (negative): *m/z* 405 (100), 336 (100), 319 (20), 293 (50), 281 (50), 151 (5) [4].

Wighteone (Erythrinin B) (3), light yellow needles, C₂₀H₁₈O₅, mp 218–219°C. UV/Vis (MeOH, λ_{max} , nm): 268. ESI-MS: *m/z* 337([M – H][–], 100), 339 ([M + H]⁺, 100), ESI-MS/MS (negative): *m/z* 337 (85), 282 (100). ¹H NMR (500 MHz, CD₃OD, δ , ppm, J/Hz): 7.95 (1H, s, H-2), 6.20 (1H, s, H-8), 7.34 (2H, d, J = 8.0, H-2', 6'), 6.85 (2H, d, J = 8.0, H-3', 5'), 3.32 (2H, d, J = 7.5, H₂-1''), 5.24 (1H, t, J = 7.5, H-2''), 1.66 (3H, s, CH₃-4''), 1.78 (3H, s, CH₃-5''). ¹³C NMR (125 MHz, CD₃OD, δ , ppm): 156.14 (C-2), 122.14 (C-3), 180.87 (C-4), 159.09 (C-5), 111.78 (C-6), 162.29 (C-7), 92.65 (C-8), 152.96 (C-9), 104.81 (C-10), 123.24 (C-1''), 130.05 (C-2', C-6''), 114.96 (C-3', C-5''), 157.25 (C-4''), 21.00 (C-1''), 122.06 (C-2''), 130.80 (C-3''), 24.78 (C-4''), 16.74 (C-5'') [5, 6].



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Lupiwighteone (4), yellow solid, $C_{20}H_{18}O_5$, mp 132–133°C. UV/Vis (MeOH, λ_{\max} , nm): 270. ESI-MS: m/z 337 ([M – H]⁻, 100), 339 ([M + H]⁺, 100), ESI-MS/MS (negative): m/z 337 (75), 282 (100). ¹H NMR (500 MHz, CD₃OD, δ, ppm, J/Hz): 8.13 (1H, s, H-2), 6.28 (1H, s, H-6), 7.39 (2H, d, J = 9.0, H-2', 6'), 6.85 (2H, d, J = 9.0, H-3', 5'), 3.43 (2H, d, J = 7.0, H₂-1''), 5.22 (1H, t, J = 7.0, H-2''), 1.69 (3H, s, CH₃-4''), 1.82 (3H, s, CH₃-5''). ¹³C NMR (125 MHz, CD₃OD, δ, ppm): 155.44 (C-2), 122.02 (C-3), 181.19 (C-4), 156.01 (C-5), 98.17 (C-6), 161.99 (C-7), 106.54 (C-8), 153.42 (C-9), 104.82 (C-10), 122.95 (C-1'), 130.01 (C-2', C-6'), 114.85 (C-3', C-5'), 157.37 (C-4'), 20.87 (C-1''), 122.02 (C-2''), 131.01 (C-3''), 24.50 (C-4''), 16.50 (C-5'') [7].

Naringenin (5), fine needles, $C_{15}H_{12}O_5$, mp 258–260°C. UV/Vis (MeOH, λ_{\max} , nm): 289. ESI-MS: m/z 271 ([M – H]⁻, 65), 543 ([2M – H]⁻, 100); ESI-MS/MS (negative): m/z 271 (30), 177 (30), 165 (10), 151 (100) [8–10].

6-Prenylnaringenin (6), yellow powder, $C_{20}H_{20}O_5$, mp 210–212°C. UV/Vis (MeOH, λ_{\max} , nm): 290. ESI-MS: m/z 339 ([M – H]⁻, 100) [11–13].

8-Prenylnaringenin (7), yellow powder, $C_{20}H_{20}O_5$, mp 195–197°C. UV/Vis (MeOH, λ_{\max} , nm): 291. ESI-MS: m/z 339 ([M – H]⁻, 100), ESI-MS/MS (negative): m/z 339 (10), 245 (10), 220 (20), 219 (100) [14–17].

Antifungal Activity of compounds **1–7** against *P. infestans* was investigated by spore germination assay. The results showed that all the compounds had antifungal activity against *P. infestans* except myrsininone A. Naringenin, 6-prenylnaringenin, and 8-prenylnaringenin had the strongest antifungal activity, and the IC₅₀ were 10.447 $\mu\text{g}\cdot\text{mL}^{-1}$, 16.828 $\mu\text{g}\cdot\text{mL}^{-1}$, and 10.864 $\mu\text{g}\cdot\text{mL}^{-1}$, respectively. Meanwhile, wighteone and lupiwighteone had moderate antifungal activities, and the IC₅₀ were 198.153 $\mu\text{g}\cdot\text{mL}^{-1}$ and 90.365 $\mu\text{g}\cdot\text{mL}^{-1}$. Genistein showed much weaker activity than others, and the IC₅₀ was 706.318 $\mu\text{g}\cdot\text{mL}^{-1}$. This fact appears to indicate that naringenin, 6-prenylnaringenin, and 8-prenylnaringenin may be responsible for the antifungal effect of *F. tikoua*.

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